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President of the American Society of Sugar Beet Technologists for the biennium 1962-63 is Guy Rorabaugh. Mr. Rorabaugh is General Chemist, Holly Sugar Corporation with headquarters at Colorado Springs, Colorado.

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Silver Jubilee of the Society

DEWEY STEWART[2]

According to custom, the President of our Society addresses the members at the opening session of the General Meeting. This is a privilege for which I ask your indulgence.

Consideration has been given to the presentation of some highlights of accomplishments in sugar beet research since my initiation to this fascinating endeavor in 1925. After reviewing the program with its sessions of technical reports in several disciplines and the symposia on topics pertaining to production and processing, it was felt that my remarks would not serve as a proper prelude to the information that will be ably presented during the Twelfth General Meeting of our Society. I have elected to discuss the history of the Society (1937-1962) and its accomplishments rather than accomplishments in sugar beet research.

The American Society of Sugar Beet Technologists was organized 25 years ago the 7th of January. It seems fitting that we take notice of this silver anniversary. Our Society had an humble beginning in what amounted to local meetings of a small group of investigators. It has grown strong through activity and has attained prominence through service to one of our principal agricultural crops and to one of our most progressive industries. Our Society has served as a useful and effective forum for the exchange of ideas and the presentation of research on problems pertaining to sugar beet production and processing technology.

Some of the early history of our Society can be obtained from the Proceedings of the first meeting, but a richer source of information concerning the background and events leading up to the organization of the Society is the store of memories of those who participated in its establishment. Dr. Harvey E. Brewbaker gave an account of the early development of our Society in his Presidential Address at the Fifth General Meeting in 1948.

The American Society of Sugar Beet Technologists grew out of conferences on sugar beet research which were held at Fort Collins, Colorado, in 1935, 1936, and 1937. These conferences, designated "Round Tables", were sponsored and organized by the Agricultural Extension Service of Colorado State University

[1] Presidential Address, American Society of Sugar Beet Technologists, Twelfth General Meeting, Denver, Colorado, February 5, 1962.

[2] Leader, Sugar Beet Investigations, Tobacco and Sugar Crops Research Branch, Crops Research Division, Agricultural Research Service, U.S. Department of Agriculture, Beltsville, Maryland.

in cooperation with the local office of Sugar Beet Investigations, U.S. Department of Agriculture, and conducted with the encouragement of the beet sugar companies.

At the banquet of the Third Round Table, unanimous opinion was expressed in favor of enlarging the Conferences to include all sugar beet research activity in North America. Thus the concept of a Society of sugar beet technologists had its inception in the festive atmosphere of the "banquet table"—probably an outstanding feature of the Round Tables.

Action was taken at the closing session of the Third Round Table, January 7, 1937, to organize the American Society of Sugar Beet Technologists. A President, a Vice President, and a Secretary-Treasurer were elected; and Salt Lake City was chosen as the first convention city. A Committee was charged with the duty of drafting a constitution and bylaws.

The Constitution was adopted at the first meeting, January 11-13, 1938, in Salt Lake City. It was indicated that "The object of the Society shall be to foster all phases of sugar beet and beet sugar research and act as clearing house for the exchange of ideas resulting from such work."

Membership was open to any individual or business engaged in or interested in the object of the Society. Dues for individual membership were $1.50. Company dues were based on number of factories—which apparently was taken as an indication of their ability to pay.

Article 3 of the Bylaws pertained to "Collection of Dues" and read as follows: "The Secretary-Treasurer of the American Society of Sugar Beet Technologists shall distribute to each member a program listing papers to be presented." The program, apparently, was in some manner expected to stimulate payment of dues. The present Secretary-Treasurer continues to send out programs, but (as you know) he employs a more positive approach to collection of dues! The Society has just adopted extensive revisions in the Constitution.

The Proceedings of the first meeting of the Society, January 11-13, 1938, records 140 members present from 15 States and 2 Canadian Provinces; 4 members were absent. In the original organization there were 5 Sections of the Society. Only 1 has been added since the first meeting.

Our Vice President, J. C. Keane, was in attendance at the first meeting in 1938 and served as Chairman of Section E, Chemistry. There was only one session of this Section with only two

papers—one by the Section Chairman himself and one by Dr. F. R. Bachler. There is no record of the attendance at this session; but it was a significant event! Previous meetings of the Round Tables pertained only to breeding, agronomy, and other phases of production research. The union of chemists and factory technologists with investigators in biological disciplines was new for a society concerned with sugar beet research.

The participation of factory technologists in our Society has been an outstanding development. At this meeting of our Society, Section E, Chemistry and Factory Operations, will meet for six sessions—four comprising 31 technical papers, one symposium on chemistry, and two symposia on factory operations. This is a remarkable growth for Section E which had only one session consisting of two papers in 1938.

At the first meeting of our Society in 1938, greetings were sent to the Institut International de Recherches Betteravieres (I.I.R.B.) stating that our Society was in session and wished success for their Congress, which was scheduled at that time. At our 1960 meeting, 2 years ago in Salt Lake City, a committee report was accepted that expressed favor for some form of joint meeting with I.I.R.B. and suggested that the first meeting be held in England in 1961, the second in United States and Canada in 1964, and similar meetings on a 3- to 5-year rotating basis. It was specified that the joint meetings would not constitute any form of integration of the two Societies. Our Secreary was instructed by the President to transmit our wishes to the Secretary of I.I.R.B. for consideration.

The proposal for the joint meeting was accepted, with the first meeting to be held in London, England, May 19, 1961, following the Summer Congress of I.I.R.B. The meeting was held in the Conference Rooms of the International Sugar Council, with arrangements made through the courtesy of the British Sugar Corporation. The Report of the First Joint Meeting has been issued.

Five members of our Society attended: Mr. B. E. Easton, Canada and Dominion Sugar Company; Dr. F. H. Peto, British Columbia Sugar Refining Company Ltd.; Mr. Bion Tolman, Utah-Idaho Sugar Company; and Dr. C. W. Doxtator and Dr. R. E. Finkner of American Crystal Sugar Company. As President of the Society I made reservations for attendance, but illness prevented travel. My remarks were presented by Mr. Tolman and were included in the Report of the Meeting.

Thus, our Society—which received only casual notice at the time ot birth—has grown in strength, and its influence now reaches beyond the shores of this continent. We can point with pride to our Society's past and recognize its present importance as a forum for the exchange of ideas and results of sugar beet research. At present we have a membership of 633 residing in 35 states and 20 countries. Our excellent Journal serves as a messenger to 59 countries!

The recognition of 1962 as the Silver Jubilee of our Society should be accepted as a milestone along our journey to a greater future of service to sugar beet research.

Sugar Beet Research and the Sugar Act

ROBERT H. SHIELDS[1]

It is a pleasure and an honor and a privilege to be here with you today, to be associated with you distinguished men of science as you embark on this, the Twelfth General Meeting of the American Society of Sugar Beet Technologists.

In looking over your full schedule of sessions for the next three and one-half days, I am indeed impressed—as anyone must be—by the broad scope of your work and the high goals you have established for yourselves. Your program is evidence that you are continuing your relentless search for ways to add still more to the tremendous contribution you and your colleagues have already made to the revolutionary progress of this dynamic American industry.

We hear and read much about the modern revolution in American agriculture, the sweeping changes that enable a farmer in one hour of work today to produce four times as much food and fiber as the farmer produced in one hour of work forty years ago. Sometimes overlooked is the basis for this great revolution —research. The keynote of our progress in agriculture, as in other fields, is research, coupled with the practical application of the new scientific discoveries which research develops. And behind that research, the thing the makes it fruitful, is the never-ending drive of people like you to learn more and more of nature's mysteries and even to improve upon that very nature when it is possible.

More than a century ago, Abraham Lincoln described the stimulation that agricultural research gives to the mind, and suggested the limitless scope of such research. He did this so effectively that his words, spoken in Milwaukee on September 30, 1859, we may fittingly use today to set the tone and suggest the breadth of your meeting here.

"I know nothing so pleasant to the mind"—Lincoln said—"as the discovery of anything that is at once new and valuable—nothing that so lightens and sweetens toil, as the hopeful pursuit of such discovery. And how vast, and how varied a field is agriculture, for such discovery. . . . Every blade of grass is a study; and to produce two, where there was but one, is both a profit and a pleasure. And no grass alone; but soils, seeds, and seasons —hedges, ditches, and fences, draining, droughts, and irrigation —plowing, hoeing, and harrowing—reaping, mowing, and thresh-

[1] President and General Counsel, United States Beet Sugar Association, Washington, D.C., Prepared for delivery at the Twelfth General Meeting of American Society of Sugar Beet Technologists, Denver, Colorado, February 5, 1962, as the Keynote Address.

ing—saving crops, pests of crops, diseases of crops, and, what will prevent or cure them—implements, utensils, and machines, their relative merits and to improve them . . . the thousand things of which these are specimens—each a world, of study within itself."

In thus finding, in Lincoln's words of more than a hundred years ago, a theme which is indeed appropriate for your meeting today, we are reminded that your work, to be appropriately evaluated, must be viewed for the long range. The vagaries of wind and rain and heat and cold may cause sharp variations, in some years, from your otherwise steady advancements—there may be occasional disappointments and departures from your long-range rate of progress.

Last year's sugar beet crop, for example, was a disappointment in a great many parts of the producing area, and the total crop was a disappointment to everyone.

Although acreage planted in 1961 was nearly 15 percent greater than the acreage in 1960, sugar production from the crop will be about the same as the 2,475,000 tons produced in 1960; last summer there were reasonably-based estimates as high as 2,800,000 tons of sugar. The yield of beets per acre in 1961 was only 16.5 tons, the lowest yield since 1955. To compound the felony, the sugar content in many areas was low. The average sugar content looks as if it will turn out to be the lowest in 25 years. The combination of low yields and low sugar content was completely contrary to the normal relationship between per-acre yields and sugar content.

The 1961 crop does not mean our technology has failed. The poor crop resulted from a combination of factors that could not be controlled even by you who have unveiled and harnessed the mysteries of genes, male steriles and hybrids—an unusual combination of natural adversities covering much of the beet area the like of which this industry has seldom experienced, on such a widespread scale, all in a single crop year.

The spring was unduly wet in some areas and unduly dry in others. Abandonment of planted acreage was nearly double the rate of the year before, and in one state more than 13 percent of the planted acres were abandoned. Heavy, washing rains caused thin stands in many areas. Hail damaged the crop in at least four states. Water supplies for irrigation were short in many parts of the mountain and central states. In the largest producing state, beets planted the previous fall were good but the spring-planted beets seemed to attract a host of insects and insect-borne

diseases. Early wet snows and winter storms in many other states caused additional losses of beets late in the harvest period.

All in all, it was a pretty rough year. But in spite of the natural adversities, an all-time record tonnage of sugar beets was produced—17,966,000 tons, 9 percent greater than the year before and 35 percent larger than the 10-year, 1950-59 average. With anything like normal sugar content, the industry would have also hit a spectacular new high in sugar production.

We may therefore consider the experience of 1961 as a combination of adverse conditions which is, we hope, wholly unlikely to occur again. That is not to say that we should ignore the experience, for perhaps it does point to some areas in which intensified effort is desirable. Perhaps research should be stepped up in the development of still hardier varieties, still better quality, still more effective disease and insect control. Perhaps it will be possible in the future to keep even a year of unusual adversity, such as 1961, from causing so great a dip in your chart of upward progress. Also, perhaps you may see if the techniques now used in forecasting crop yields may be improved. I am prompted to make this suggestion by the fact that the industry did not realize what was happening this year until the eleventh hour.

For the long run, however, we must look at averages and trends, not at a single year. And in looking at those long-range averages, anyone can see that you have done a terrific job.

For the basic advancements the industry has made in this century—or the last fifteen, or ten, or five years—have been technological advancements, the results of your research, gains in the field and in the factory that have made the American beet sugar industry the efficient industry that it is today.

Of course I know there are still problems and there always will be. For your work is never done. Your achievements of today are merely the starting points for your work of tomorrow. There is always the challenge of "How can we do it better?" Without meeting this challenge we die.

Let us see how you have been meeting and answering this challenge during the 15 years from 1946 to 1960, the longest recent period for which complete statistics are available.

In 1946, the industry produced 10,863,000 tons of sugar beets. In 1960, production amounted to 16,530,000 tons—an increase of more than 5 and one-half million tons or 52 percent. This was achieved by an increase of only 17 percent in harvested acres—818,000 acres in 1946 compared with 957,000 harvested acres in 1960.

Obviously, this means that you increased the yield of beets per acre, and you did—from 13.28 tons per acre in 1946 to 17.26 tons in 1960, an increase of 30 percent.

Sugar production increased even more than beet production or beet yields—from 1,568,000 tons of beet sugar in 1946 to about 2,475,000 tons of beet sugar in 1960, an increase of 58 percent.

Mind, you, this increase of 58 percent in sugar production took place when there was an increase of only 17 percent in the number of harvested acres.

Obviously again, technological advancement was the reason, this time expressed in yield of sugar per harvested acre. In 1946, an acre yielded 1.92 tons of sugar, while in 1960, the average acre yielded 2.58 tons of sugar, an increase of 34 percent in those 15 years.

Truly, these are remarkable achievements, the concrete results of your combined efforts in agriculture, factory operations, and chemistry. Your achievements have been a primary factor in keeping the industry alive and progressive, in the face of a continuing cost-price squeeze, both in the factory and on the farm.

These achievements also have a direct bearing on sugar legislation—on the kind of law which the industry needs and must have in order to continue its parade of progress—for legislation must reflect and even forecast the achievements of science, or there is trouble ahead.

To put this another way: Unless the quota provisions of the Sugar Act permit a growth in the beet sugar quota which at least keeps pace with the technological advancements of the industry, your progress is nullified and pressures build up which could cause explosions having far-flung repercussions.

The truth of this statement is demonstrated by our experience of the past.

You will recall that the Sugar Act of 1948, the first revision to be enacted after World War II, imposed a fixed ceiling on the beet sugar industry and other segments of the domestic sugar producing industry for a temporary period Those fixed quotas may have seemed generous at the time, but the progress of the industry was such that we were, before long, bumping our heads against the ceiling. When domestic producers again were permitted to share in the growth of our continuously growing sugar market, in the amendments passed in 1956, the beet sugar industry's share in that growth was set at about 22 percent. It was anticipated then that 22 percent would provide sufficient quota to allow for the industry's technological advancements and in addition to permit a modest growth in the industry.

But you have proved to be better than the Congress thought you would be. Your research has developed the new miraculous, high-yielding hybrid sugar beet seeds. You have developed the long-sought-for monogerm seed, and by patient plant breeding worked into that seed the desirable characters you earlier had worked into the multigerm seed. You have greatly improved all sugar beet cultural practices. As a result of your practical research, the technological advancements you have made, the increase in yield per acre has far outstripped the increase in beet sugar quotas provided by the "growth formula" written in the 1956 law. On the basis of about 150,000 tons average annual increase in United States sugar consumption, the 22 percent accruing to the beet sugar industry amounts to about 33,000 tons. But the technological advancements of the industry result in an average increase in production each year of between 40,000 and 50,000 tons of sugar at a constant acreage figure.

To keep production within the quota levels of the present law, we would have had to reduce sugar beet acreage sharply in recent years—if misfortunes had not come to the offshore domestic producing areas of Hawaii, and Puerto Rico. A series of catastrophes—hurricanes, droughts and strikes—has plagued those areas, resulting in production well below their quota levels. Substantial amounts of the deficits in those quotas were allocated to the beet sugar area. From 1957 through 1961, the beet area received allocations of nearly one and one-half million tons of Hawaiian and Puerto Rican deficits. Without these allocations, the only alternative to reducing acreage would have been to pile up burdensome inventories of beet sugar.

Now we know that dependence on uncertain deficits from other domestic areas—dependence, in short, on someone else's misfortune or inability—is not the best way to maintain a stable climate conducive to a healthy beet sugar industry. So the industry's legislative committee has sought to develop[a legislative program for the future which would at least minimize that dependence, and put the beet sugar quota on a sounder basis.

A program has been developed which has the support of all the domestic sugar producing and refining groups—the beet sugar industry, the cane sugar refiners, and the cane industries of Louisiana, Florida, Hawaii and Puerto Rico.

The program would establish a new basic beet sugar quota which would recognize the industry's recent achievements in production and marketing, a quota of 2,665,000 tons at the current level of sugar consumption. For the future, the industry's pro-

gram would reserve for the beet sugar industry a sufficiently larger share of the normal increase in consumption to accommodate your accomplishments, as fully as we can anticipate their trend, and to permit some expansion of the industry.

The program would include a growth formula which, on the average, would provide for an increase in the beet sugar quota, above the proposed new base, at the rate of about 75,000 tons of sugar a year. We hope that this will achieve the purposes we seek. We hope that this time we have more accurately estimated your ability to increase the per-acre yields of sugar.

In this connection, I recall that in a talk prepared for your meeting exactly ten years ago I raised the question as to whether it would be safe to predict, then, that in the next 25 years you would double the average production of sugar per acre. Experience has shown that such a prediction may have been on the optimistic side, but you have made considerable progress toward that achievement, and the industry's current legislative program has been developed in line with your demonstrated long-term rate of progress.

Along with increases in the basic beet sugar quota and in the share of future growth for the beet industry, the legislative program also envisions increases in the basic quota and growth percentage allocated to the mainland cane sugar producing industry. These two continental producing areas—the beet sugar area and the mainland cane sugar area—have both demonstrated a willingness and an ability to provide a larger share of sugar for the American market than they have been permitted to supply in the past.

Our experience with Cuba shows how quickly a supposedly reliable and friendly foreign source of sugar for American consumers can become unreliable and unfriendly. Yet our dependence on foreign sugar is still as great as it was before the Cuban supplies were cut off. Not a single ounce of the former Cuban quota has been allocated to domestic producers—it has all been allocated to foreign countries. Under the present law, we still are obliged to depend upon foreign nations for nearly half—over 45 percent—of our annual sugar supplies.

Repeatedly, the Congress has stressed the importance of a domestic sugar-producing industry for national defense and strategic reasons. As recently as June 6, 1960, the House Committee on Agriculture said in a report that a primary purpose of the Sugar Program is to "make it possible, *as a matter of na-*

tional security, to produce a substantial part of our sugar requirements within *continental* United States . . ." (Italics supplied) .

Surely in these troubled times it is in the national interest to increase the percentage of sugar we obtain from the sugar industry of the continental United States.

Of course, in legislation as in research, the high hopes we have at the beginning of a project may not always be fully realized. When the cauldron of Congress boils, vapors as strange as the vapors in your laboratories occasionally ensue. Mutations as unexpected as those you encounter in your greenhouses and test plots frequently occur in legislation between the time a bill is dropped in the hopper in the House and the time it reaches the President's desk for his signature.

Whatever may take place on the legislative front, however, cannot diminish the importance of your work. The industry will continue to rely upon you—scientists, technologists—to maintain industry advancement, to continue and improve present high rates of efficiency, to intensify your unceasing efforts to reduce production costs both on the farm and in the factory, to keep the beet sugar industry among the most progressive industries in America.

And the nation will continue to rely upon this industry for a large share of its sugar with the assurance that this is *one* source of supply that is not and cannot be dominated by *the* Communist world—that American-produced beet sugar is available here and now, in the continental United States, and is not subject to the uncertainties of unstable foreign governments.

To the extent that you contribute to the dependabilitv of the beet sugar industry—and your contribution on this score is indeed large—you contribute to the stability of America.

This is a thought which I hope will give vou heart and inspiration as you conduct vour discussions and vour studies this week on your myriad subjects—each subject "a world of study within itself."

Current Events in Sugar

LAWRENCE MYERS[1]

The kind invitation of Dr. Stewart to address you made me both thankful and fearful. You people who understand what goes on in a test tube and who know about genes and moments of inertia always fill me with awe. Nevertheless, those of you attending this meeting are never satisfied until your scientific advances are brought into practical application so as to increase the efficiency of sugar production and to improve the sugar economy as a whole. Therefore, you may be interested in a review of a few of the recent developments in other aspects of the sugar economy.

Ever since the end of World War II those interested in the world sugar economy have been hopeful that a method could be developed for preventing a repetition of the depression in world sugar such as the one that started in the late 1920's and that reached bottom in the early 1930's. It was for this purpose that the major sugar exporting and importing countries of the world entered into the International Sugar Agreement. That Agreement went into effect on January 1, 1954, with 24 member countries. Todav the membership has increased to 43 countries. The member countries now account for roughly 85 percent of the world's production of sugar.

The bulk of the world's sugar export trade, therefore, is supposed to be carried on in an orderly manner under quotas designed to achieve a reasonable degree of stabilization in the world market. The fact is, however, that world sugar prices have been irregular for the past three years and they started on a major downward trend last spring.

With the corning into power of the Castro Revolutionary Government in Cuba in January 1959, the huge sugar industry of Cuba was thrown quickly and inexorably into the Communist orbit. Price pronouncements ranging all the way from promises of stabilization to threats of price wars poured out of Cuba with utter irresponsibility and immaturity. In July of 1960 the United States Government had to recognize that a Communistic Cuba was not a dependable source of sugar supply.

In negotiations of the International Sugar Agreement which extended with a short interruption from early last September until mid-December it was finally recognized that Cuba would

[1] Director, Sugar Division, Agricultural Stabilization and Conservation Service. U. S. Department of Agriculture, Washington, D.C., before the American Society of Sugar Beet Technologists, Denver, Colorado, Wednesday, February 7, 1962.

not agree to any quota that other countries could accept and that to agree to the quota provisions that Cuba and the other Communist Countries demanded would nullify the effectiveness of the Agreement and in large measure turn the sugar industry of the world over to the Communist Bloc while the free world would be restricted.

As a result no world sugar quotas will be in effect for 1962 or 1963, the two remaining years of the present International Sugar Agreement. It is hoped that a new agreement can be negotiated next year.

The dropping of quotas for the world market has caused some to have tremendous fears for the future of the world sugar market. I do not wish to forecast the future of world sugar prices or to give assurances of when world prices will stabilize or improve. However, I do not believe that the absence of quotas under the International Sugar Agreement will be a major determinant of prices during 1962 or 1963.

When the United States stopped buying Cuban sugar in 1960 the Soviet Union undertook to increase its purchase from Cuba by a like amount. In 1961 the USSR and Red China imported huge quantities of Cuban sugar. Obviously these purchases by Communist Bloc Countries made a home for large quantities of Cuban sugar. However, it soon became evident that Russia was not taking normal quantities of sugar from its older satellites, Poland, Czechoslovakia and Hungary. Accordingly, these three satellite countries had to sell additional quantities of sugar in the world market. At times last year European white beet sugar sold at lower prices than raw cane sugar. That is one of the reasons that world sugar prices have been uncertain for the past six months.

During the negotiations last fall Cuba made a great point of its sale of 4,860,000 tons of sugar for each of the ensuing five years to Russia, Poland, Czechoslovakia, Hungary and Red China. It became evident immediately, however, that large quantities of this sugar would become available for re-export sale by these countries. Therefore, the sugar will not be entirely removed from the world market.

After the Conference recessed in October it was learned that Cuba had exported in excess of its 1961 quota and was continuing to export. Cuban officials frankly admitted this and stated that their 1961 exports would exceed their quota by 1,100,000 tons. Recent trade reports indicate that their exports exceeded their quota by 1,400,000 tons. Nevertheless, when the Conference resumed negotiations in December the Cuban delegate offered no

apology for his country's violation of the Agreement and he offered no guarantee that his country would refrain from violating the Agreement in the future. On the contrary he demanded a quota that probably would have exceeded Cuba's ability to export in 1962. Worse yet, the methods of computing the quota would have been inconsistent with those used in computing quotas for non-Communist exporters and the computation would have involved a retroactive and ficticious determination of a condition of force majeure in 1960. Such a determination of force majeure would have been used to excuse a part of Cuba's overshipment *in* 1961.

Clearly, such a quota would have been of no value in stabilizing the world market. Moreover, acceptance of it would have made every member country a party to the establishment of a double standard of statistical treatment which would have given a Communist country preferred treatment over Capitalistic countries. Also, acceptance of the phoney claim of force majeure would have made other countries moral partners to Cuba's violation of the Agreement. Under such conditions a continuation of quotas would have been less than useless.

At the negotiating conference Cuba blamed the United States for most of the real and imaginary ills of the sugar market. It was not difficult to disprove these false charges. Tn fact most of them fell of their own weight. There is one criticism that is being made against our sugar policy, however, that will become progressively more valid if our program remains unchanged. The argument is made that the United States, by paying foreign producers twice the world price for sugar under its quota system, is tending to stimulate foreign production. Fortunately, we could show that up to last year the great expansion in world sugar production came in the Communist Countries and not in the countries supplying the United States. This may not be the case in the future.

The newest development in our sugar program is the undertaking of a barter-like operation. Under this program a part of our sugar will be obtained from countries that agree to make specific reciprocal purchases of American surplus farm crops. The countries agreeing to purchase the largest dollar volume of such crops per ton of sugar will receive quota reallocations to sell sugar in the United States. This program will not apply to the bulk of our sugar imports which must be supplied in accordance with formulae contained in the Sugar Act. It will, however, apply to quantities that present quota countries are unable to fill.

Our present sugar legislation lasts only through June 30 of this year. Therefore, Congress soon will be at work on new legislation to extend the program. The President in his budget message stated that the Act would be extended with substantial revisions to bring it into line with the greatly changed world sugar situation and to provide for the recapture by the United States Government of the premium at which domestic prices are held above world prices. This is of great importance to our foreign suppliers and it will necessitate some revisions in the methods and procedures followed by importers of foreign sugar. I do not see, however, why it need be of major concern to purely domestic producers. No proposal has been made for reducing or changing the amount of protection afforded domestic sugar producers. Domestic growers can be protected as adequately and as certainly by the proposed method as by the present one.

One of the problems that will confront the sugar industry and the government in developing new legislation is the extent to which basic marketing quotas will be increased and the nature and extent of provisions for meeting the demands of new producers and new producing areas. To appreciate the nature of these problems, it is necessary to recognize the effects of three very different factors.

1. In recent years Puerto Rico and Hawaii have failed to fill their basic marketing quotas. Hawaii's failure resulted from the prolonged and disastrous strike of 1958 and its aftereffects. Gradually these effects are wearing off and Hawaiian production is recovering. Puerto Rico's failure to fill its quota was the result of adverse weather conditions and the low sucrose content of recent crops. So far as I am aware, the low sucrose content has not been explained. However, Puerto Rico has been harvesting peak tonnages of cane and its production has recovered considerably from the recent low point. To the extent that production in the offshore areas improves there will be smaller deficits to reallocate to the mainland areas.

Because of the large offshore deficits in recent years stocks in the mainland areas have been greatly reduced and neither mainland area was able to fill its quota in 1960. Low production of beet sugar in 1961 further reduced stocks in that area. No acreage restrictions were in effect in 1961 and none will be in effect for the 1962 crop. It is anticipated that 1962 production in the mainland cane and beet areas will be sufficient to permit these areas to fill their marketing quotas and to have larger carryovers at the beginning of 1963. In other words, the current acreages

of mainland sugarcane and beets are in excess of the acreages required to fill the basic quotas of these two areas. If present quotas for the domestic areas remain in effect, therefore, or even if moderately increased quotas should be established, it is probable that some cutback in acreage will be necessary in 1963.

2. Domestic sugar prices in the post-war period have been relatively stable, whereas prices of other farm crops rose sharply during the Korean fighting and then declined. As a result present returns from sugar beets are attractive compared with the returns from other farm crops. This has made established sugar beet growers wish to increase their production and has caused farmers in many parts of the country to want to start production. The demand for acreage now greatly exceeds factory capacity in nearly every part of the country. There is strong grower pressure for the erection of new factories. These pressures exist all the way from Maine and New York State in the northeast, to Arizona in the southwest and to Washington in the northwest.

If present price relationships could be guaranteed and if marketing opportunities were guaranteed, this country could go far in the direction of domestic self-sufficiency in sugar. However, the comparatively favorable returns from sugar crops is not the result of greatly increased sugar prices but of lower prices of competing crops. Many farmers who are now clamoring to raise sugar beets would turn to the proved and established crops for their respective communities if the prices of such crops were to recover. Sugar beet processors have learned to their sorrow that they cannot operate plants profitably in areas in which farmers wish to plant sugar beets only in years when sugar is high in price or when other crops are low in price.

3. The agricultural revolution that has had such tremendous effect on our agricultural production as a whole has also affected sugar beet production. This is resulting in a desire for larger sugar beet acreages per farm and has made sugar beets a more attractive crop to many farmers. Also the development of irrigation, private as well as public, has made it possible to grow sugar beets successfully in many areas that could not do so a few years ago.

For the above reasons the pressures to produce sugar beets are now greater than ever before and this pressure comes at a time when the industry is already operating at factory capacity as a result of temporary conditions.

Great sympathy and wisdom will be needed in dealing with this situation over the next few years. Certainly the demands of

new producers and new producing areas must be met to the maximum extent feasible. This is a field, however, where we need careful analysis and hard-headed business judgment as well as sympathy. The beet sugar industry has had more than its share of sad examples of misplaced factories. Now that production requires such large capital investments farmers as well as processors need to use caution and make certain that new production projects are wisely located for long-time efficient production.

You beet sugar technicians can perform a great service for your industry and for the country in developing criteria for examining prospective sugar beet enterprises. The Department of Agriculture is literally deluged with proposals involving new plants and new producing areas. Frequently we are told that representatives of one or more of the existing companies have visited the area and have indicated an interest in obtaining acreage or in constructing a factory in the area. In some cases these are the same general areas that have gone out of sugar beet production in the past. In none of these cases has there been the long and careful experimental work or study of comparative costs and profits necessary to determine the long-time interest of farmers in producing sugar beets in the particular locality. Neither have the proposals indicated any adequate analysis of the marketing problems that would confront the new factory. Since World War II, 25 sugar beet factories have gone out of existence, while others have prospered. Many of those that have failed were built in a promotional atmosphere in areas that were not suited culturally or economically to produce beet sugar on a competitive basis.

Suitable areas have a right to look forward to the erection of factories and the undertaking of sugar beet production. I am glad to see the beet sugar industry making specific provision in its legislative proposals for meeting, in some degree, the aspirations of new areas. I hope the existing industry will go farther, however, and outline the basic information needed to determine whether or not an area is suited to produce sugar competitively. It will be disappointing indeed if the end result of today's relatively favorable prices for sugar beets is to be the erection of plants destined to wither and die because they are not located where they can survive in today's competitive struggle.

I now want to turn to a development that must be attributed in no small part to the work of you technologists. Sugar beet production has been rather thoroughly revolutionized since World War II. Virtually all of the crop is now harvested by machines and over 40 percent of the crop is thinned by machines. The

acreage of sugar beets per farm for the country as a whole has increased significantly. We have not yet seen the end results of the monumental development of monogerm seed nor have we come to the end of the road in the application of herbicides and other chemicals to increase production and improve efficiency in the growing of sugar beets.

Since the war there has been a reduction of 44 percent in the man hours of field labor required to produce a ton of beet sugar. Even though there has been a simultaneous increase of 44 percent in the hourly earnings of field workers, the total cost for field labor has been reduced. I mention these developments in farm practices because they are well known. There have been corresponding improvements in processing and in marketing. The beet sugar industry is to be commended for the increases it has made in the efficiency of production and marketing throughout its ramified system. The industry must be encouraged to continue these improvements. Again I ask your help.

The domestic sugar industry is not only highly protected, it is also highly regulated. It is the only agricultural industry in which the Department of Agriculture has the responsibility for determining fair wages and fair prices. Farmers and processors accept these regulations and appear to take pride in them. Well they should, for one of the end purposes of protecting an agricultural enterprise must be to improve the standard of living of farm people, including farm laborers.

In administering these regulatory provisions of the Sugar Act, however, we must keep in mind some of the fundamental economic prerequisites for increasing efficiencies. Production and marketing efficiencies, in both fields and factories, involve large capital investments. We must make certain that we give the efficient farmer and the efficient; processor an opportunity to make a profit from these additional capital investments if we expect our industry to continue to improve.

Today it is vital that there be complete understanding and confidence between processors and growers on projects that affect the grower's returns from his beets. Governmental determinations of fair prices and fair wages can afford a degree of protection to growers and laborers and may instill a certain measure of confidence. However, such determinations cannot be a substitute for understanding and negotiation. When growers and processors have full, frank and timely discussions of their mutual problems and projects there can be little doubt of their ability to reach a solution that will foster progress.

On that note I wish to close. If we keep in mind that our sugar industry is highly competitive and that its various segments have many divergent interests, it seems to me that it shows a commendable degree of tolerance, understanding and mutual respect. By continuing the drive for sound, objective solutions to production, processing and marketing problems, you men with your associates on farms and in factories and distribution centers can assure the continued success of the domestic beet sugar industry.

The Influence of Research on Efficiency of Sugar Beet Production

M. W. PARKER[1]

It is a pleasure for me to attend the Twelfth General Meeting of the American Society of Sugar Beet Technologists and discuss the influence of research on your industry. Too frequently we are inclined to forget research accomplishments that contribute to developing, maintaining, and assuring the future of a sound industry. Some of these high lights will be briefly reviewed in order to bring some of our current problems into sharper focus. In addition, I am certain that you will be interested in a few examples of our current basic research programs which benefit sugar beet production as well as other crops.

The sugar beet industry, including production, has grown in importance in our agricultural economy at a pace commensurate with the increase in sugar quota and acreage allotments. These yearly manifestations of vigor and responsiveness to increase production demands may be attributed to several factors such as improved economic environment, new developments in technology, and more efficient management in industry and on the farm. But agricultural research can justly claim credit for the remarkable improvement in acreable yields of roots and sugar; increased efficiency in sugar production, including a reduction of labor requirements; and, above all, for protective measures against certain disease hazards that once seriously threatened continuance of growing sugar beets in several major districts.

The sugar beet, as other crops, has been through periods of discouragement. Many of you can recall the low yields and erratic productions of the twenties and early thirties when recurrent epidemics of diseases, such as curly top in the West and leaf spot and black root in the eastern sugar-beet regions, resulted in low quality of roots for the processor and in unsatisfactory returns to the grower. These diseases adversely influenced the economy of beet sugar production for several years.

Relief from these disease hazards was not the result of some benevolent act of Mother Nature or a change in the weather. Actually the diseases are still present, but protection has been accomplished through the development of resistant varieties and the application of improved field practices.

These advances have been the product of well-organized research programs conducted by groups of devoted scientists em-

[1] Director, Crops Research Division, Agricultural Research Service, U. S. Department of of Agriculture, Washington, D.C., at the Twelfth General Meeting, American Societv of Sugar Beet Technologists, Denver, Colorado, February 7, 1962.

ployed by the beet sugar industry and by Federal and State agencies. The financial support, as well as cooperative assistance, received from the beet sugar industry for our joint research activities has been remarkable. It stands as one of the best examples of government and industry working together toward a common objective.

An early significant accomplishment in sugar-beet breeding was the development of curly-top-resistant varieties that gave new life to the beet sugar industry in most of the districts west of the Rocky Mountains. The havoc caused by curly top in the varieties available in the twenties was an appalling sight. Probably no research assignment appeared more difficult than the control of curly top through breeding of resistant varieties. For this reason, the accomplishments have been most gratifying.

The level of curly-top resistance that has been attained in commercial varieties such as US 22 and its improved releases is remarkable and may have given rise to a feeling of unconcern for the disease. Not only is the disease still brought into irrigated districts from rangelands each spring by the leafhopper, as in the past, but new and more virulent strains of the virus have been found and other new ones may be expected to occur. These new strains of curly top are capable of causing severe damage to US 22 and other varieties that gave protection in the past. Therefore, research on curly top is still important and must be included in our over-all program of sugar beet research.

Breeding for resistance to leaf spot and black root lor the districts east of the Rocky Mountains has resulted in benefits comparable to those derived from curly-top-resistant varieties for the western region. With the introduction of American varieties in the Great Lakes region, the acreable yields of roots have shown a steady increase, and some districts in this region are now well above the national average in productivity.

Sugar beet crops are now threatened by virus yellows or a complex of viruses that bring about yellowing of foliage and strikingly influence the yield and quality of the sugar beet. No doubt this disease will be a factor of increasing concern in the economy of beet sugar production in this country, as has been true for Europe where the disease has been under investigation since the thirties. Virus yellows was first identified in the United States in 1951 from plants collected in Michigan. Since that time, the disease has been found in all major sugar beet districts where surveys have been conducted. The disease has reached epidemic proportions in California and in areas where sugar beets or other susceptible plants are growing most of the year. The damage

depends upon the age of the plants when infected and on the virulence of the strains of the virus involved. Damage appraisal tests conducted in California have indicated reduction in root yields from 2 to 47 percent and in sucrose content ranging up to 3 percentage units.

The immediate relief from damage caused by virus yellows must come from measures directed at the vectors or at field practices and cropping systems. The ultimate goal is protection through the development of resistant varieties. Progress has been made in the breeding of basic lines that do not react to the virus by the yellowing of the foliage, while the development of productive varieties that are more tolerant or immune to virus yellows is a goal of the future.

Nematodes have long plagued the sugar-beet producer. As research advances are made, the complexity of the nematode problem is revealed through a multiplicity of alternate host relationships and an increasing knowledge in the number of different kinds of nematodes. Some empirical control has been obtained with soil fumigants at a high cost, and by more practical control through cropping practices including fallow or crop rotations without alternate hosts. For ultimate control of the sugar beet nematode, the most promising project is breeding for resistance or tolerance by using the wild *Beta* species. In the meantime, information on the biology of the sugar beet nematode and its relation to alternate hosts, as the tomato, gives some basis for guidance in modified cropping practices until more suitable varieties are developed. Several species of the root-knot nematodes well known in other crops, particularly in California, contribute to production hazards and most economical and efficient production of sugar beets. Also, gall-forming nematodes, sometimes confused with the root-knot nematode, add to the complexity in Colorado, Wyoming, Montana, Kansas and Nebraska. These gall-forming nematodes are known to have a definite effect on the efficiency of beet production along with a nematode complex involving root lesion nematodes, spiral nematodes, and pin nematodes, commonly found in the association with sugar beets. The significance of each will not be clear until the whole biological relationship can be established with one another and with the crop.

The establishment of a sugar-beet-seed production enterprise in the United States was a direct result of the accomplishments of our sugar-beet geneticists and breeders. Various segments of the industry joined forces in this endeavor to insure a dependable

source of seed with maximum disease resistance, and agronomic characteristics suitable to regional environments. This industry has in turn provided a wealth of material for the plant breeders to continue their work on improving the crop. Our current sugar-beet economy would be quite different today if industry had not provided wise management of seed stocks, maintenance of reserves, and facilities to permit an orderly and rapid change-over to new varieties.

Hybrid sugar-beet varieties have shown roughly 15 percent increase in yield over the open-pollinated varieties. The discovery of cytoplasmic male sterility in the sugar beet and the utilization of this character as a tool in the production of hybrid seed have had a measurable influence on the economy of beet-sugar production. Of special significance is the recent discovery that certain combinations of inbred lines show heterosis for sucrose percentage as well as for root yield. If future combining ability tests with inbred lines should reveal the general occurrence of this phenomenon in sugar-beet-breeding material, one should be able to push forward with higher root yields and with improved quality through one breeding procedure. Obviously such a development would have a profound effect on the efficiency of beet-sugar production.

One of the most elusive factors that we have to deal with in all crops research is quality of the finished product. This is due to the fact that our agriculture products must meet the requirements and standards of diverse end-use. Sugar beets having only one principal end-use simplify the problem to a degree, however, quality is conditioned by several factors, such as disease, nutrition, environment, and genetic components. No doubt the processor, as well as the grower, has the impression that quality is a temperamental condition that can be upset by many factors, Actually the physiologist must admit that he does not have all the answers. Certainly nutritional and climatic environments are known to be associated with low quality, but it is also true that these same factors favor disease. Therefore, it is difficult to separate the causes into their component parts. It has been clearly established that imbalance of nutrients, especiallv heavy and untimely applications of nitrogen, results in low sucrose percentage without bringing about an increase in root yield. There are several papers on this subject in the technical sessions that should help in developing the proper fertilizer practices.

The ability to completely mechanize all field operations in sugar-beet production must be attained in order to insure the

grower of maximum production potential. Monogerm varieties of sugar beets that are now available or in advanced stages of development for all regions, will have a far-reaching effect on this goal when we learn to use them in proper field practices.

Weeds are among the last remaining obstacles to complete mechanization of many crops, and excellent progress has been made in the development of herbicides for weed control in sugar beets. Complete mechanization of sugar-beet production can only be accomplished when more effective and selective herbicides become available to the sugar beet growers in all areas of production.

Progress in the development of herbicides for weed control in sugar beets through the combined effort of federal, state, and sugar company employees has been most significant during the past decade. Within this period, trichloroacetic acid (TCA) ; 3,6-endoxohexahydrophthalic acid (endothal) ; isoproply N-phenylcarbamate (IPC); 2,2-dichloropropionic acid (dalapon); ethyl N,N-di-/i-propylthiolcarbamate (EPTC) ; propyl ethyl w-butylthiolcarbamate (PEBC) ; 4-chloro-2-butynyl N- (3-chlorophenyl) carbamate (barban) ; and 2,3-dichloroallyl diisopropylthiolcarbamate (DATC) have been developed for the control of broadleaved weeds and grasses in sugar beets.

Even with all of these advances more effective herbicides for the control of broadleaved weeds in sugar beets are needed. The recent development of pre-planting soil-incorporated treatments with EPTC and PEBC for the control of both grasses and broadleaved weeds represents a significant improvement in chemical methods of controlling weeds in this crop.

Barban and dalapon have proved highly useful for the control of wild oats after they emerge in sugar beets. The development of these two chemicals for wild oat control in sugar beets represents a significant accomplishment because TCA, endothal, and other herbicides used as pre-emergence treatments are not effective in controlling wild oats in this crop.

Progress has also been made in fundamental research on the selective action of herbicides. Basic research on the differential effects of dalapon on sugar beets and weed grasses has yielded valuable information. Fundamental research on the mechanisms of action of herbicides, the basis for their selective action, the behavior of herbicides in soils, and the effects of environmental factors on their efficiency has been of great value in the synthesis and development of new herbicides for weed control in sugar beets. Basic research has also provided guidelines for more

effective use of herbicides presently available for weed control in this crop.

New herbicides are being developed at a rapid rate and each new one must be evaluated to determine its usefulness for weed control in sugar beets. Promising new herbicides, including such additives as surfactants, must be thoroughly evaluated as pre-planting soil-incorporated, pre-emergence, and post-emergence treatments, including their behavior in soils, effects on crops grown in the rotation, and the effects of environmental factors on their efficiency in controlling weeds without injury to the crop.

The Agricultural Research Service has established 16 pioneering research laboratories since 1957. These laboratories operate on broad charters with the primary objective of exploring the unknown in order to discover basic principles that will be useful to agriculture in future years. The Crops Research Division has two such laboratories. One is concerned with plant physiology and the other with plant virology. Actually our research in the physiology laboratory started in 1936 with work on photoperiodism. In 1957, the charter for this group as a Pioneering Laboratory was broadened to cover effect of light on plant growth and development.

Very important advances in our understanding of the physiology of plants have resulted from studies of their responses to light. Flowering and growth responses of plants to different lengths of day indicate an internal regulation for measuring time. The controlling factor is found to be the length of the uninterrupted night. Sugar beets flower when days are long and nights are short. Interruption of the nights by dim incandescent lights to make two short nights out of each long one promotes flowering of beets and other Ions- day plants, thus showing that duration of darkness, not light, is the controlling factor.

Detailed experiments have shown that red light is more effective in controlling flowering than any other color. The light energy required to induce flowering is extremely low. The effectiveness of light applied in the night results from absorption, and since red light is the most effective in controlling flowering, the responsible pigment must be blue. The concentration of this pigment is too low for visual detection.

The minimum light energy for a particular response as a function of color or wave length has been identified for flowering of short and long day plants; stem and leaf growth; germination of many seeds; and pigment formation. Responses to light of different colors, or action spectra, are remarkably similar, sug-

gesting that all of these different phenomena are regulated by the same receiver in the plant. Maximum effectiveness was at about 650 m_u, which is in the center of the red region of the spectrum.

Seed germination can be either promoted or inhibited by red light, and the process is reversible. Radiation at 730 m_u or far red which is at the visual limit is inhibitory; while that at 650 m_u is promotive. The level of germination is independent of the number of alternations between 650 m_u, (promotion) and 730 m_u (inhibition) . It is completely dependent upon the wave length of the last radiation given in a series.

Re-examination of the flowering and stem elongation responses previously known proved that they were also reversible by exactly the same wave lengths that control germination of seeds.

These various responses led to the conclusion that they are all controlled by a single photoreversible reaction and implies that the photoreceptor must undergo reversible changes from one form to another—one form absorbs red light (650 m_u) and the other far-red (730 m_u) . The length of the night is measured by the change in darkness of the far red to the red absorbing form.

Physiological experimentation developed these facts. Now biochemists have isolated phytochrome from 20 or 30 plant species including sugar beet leaves. The action compound is a protein that denatures at temperatures about 50°C when isolated from the leaf and permanently loses its reversibility.

The far red absorbing form of phytochrome is an enzyme, but the reaction it controls in the plant is unknown. That this reaction is a very basic one of plants is shown by its control of numerous widely different plant responses. Its point of control is evidently a very primitive one in the reaction sequences that lead to display of these various responses.

This has been a very abbreviated summary of the development of knowledge in this field in the last 10 or 15 years, initiated 40 years ago by Garner and Allard. A study designed to investigate the light control of flowering led step-by-step to eventual awareness that the controlling mechanism is not peculiar to flowering but is exhibited in innumerable phenomena of plant development. A few decades ago most of us looked upon photoperiodism as a biological curiosity of casual interest but no immediate general concern. Today, we look upon it as a key response of plants to a fundamental reaction that has most diverse and far-reaching consequences.

Where will these investigations lead in the future? Prediction is unsafe. Ten years ago we surely could not have predicted that studies of photoperiodic response of soybeans might lead to understanding why tomatoes in a grocery store are often pinkish instead of orange-red, why one should not cultivate after applying a pre-emergence herbicide, or why pieces of green apple skin floating on sugar quickly smell like stored apples in darkness but not in light. These are only a few of the phenomena one finds himself contemplating with at least partial understanding as a result of logical step-by-step study of the influence of light on flowering.

In our Pioneering Research Laboratory in Plant Virology the study of developmental forms of plant viruses may be possible as a result of recent research findings. An infectious material distinct from tobacco mosaic virus has been isolated from infected tobacco leaves. This infectious material can be broken down by the plant enzyme ribonuclease which indicates that the material is ribonucleic acid (RNA), the chemical building blocks in living cells.

Tobacco mosaic virus particles are rods consisting of a core of RNA surrounded by protein. It is believed that during tobacco mosaic virus multiplication in infected leaves, nucleic acid exists free as an early form of the virus. Until now this free RNA could not be isolated from infected leaves (except by methods which also extract the RNA from complete virus), because the ribonuclease in the plant material destroyed the RNA before isolation could be accomplished.

It has been found possible to purify and separate viruses by a system involving diffusion—filtration through glass columns packed with a buffered suspension of agar gel chips. Spherical virus particles diffuse into the agar chips and particles of different sizes move down through the column at different speeds depending upon their diffusion coefficients. Long particles such as those of tobacco mosaic virus, cannot move into the gel and pass through the column quite rapidly. This method is useful for the separation of very small contaminants from virus suspensions and for the sorting of viruses which differ by as little as 3 or 4 $m\mu$ in particle diameter. Preliminary work with enzymes and other large protein molecules suggest this method of purification and separation will be extremely useful for the purification and separation of many biological components in addition to the virus work for which it is currently being used.

Experiments are under way to purify the curly-top virus of sugar beets and to determine its size by diffusion-filtration. In

February 1956, Dr. Steere reported at the San Francisco meeting of the American Society of Sugar Beet Technologists, the isolation of an infectious component from curly-top infected sugar beets which had a particle diameter of 16 m_u but was not willing to publish a paper on the infectious particles he isolated, because his final product was unstable and he feared that the 16 m_u particles might be a breakdown product of the virus resulting from the purification procedure employed. The diffusion-filtration procedure is both rapid and extremely gentle on the virus particles and we expect to have some interesting results with curly-top virus in the near future.

Basic research on plant growth regulators provides a background for more applied research on many of our crops. Some rather recent examples have a direct application or at least contribute toward a better understanding for the development of practices in sugar-beet production or applied research contributing to advances through production research. Three chemicals—Ammo 1618, phosphon, and *CCC*—found to retard plant stem growth in our laboratories and later adapted to limited commercial use, have been found to prevent salt damage to soybean plants growing in highly saline soils. Soybean plants growing in pots with a fertilizer application equivalent to 7,800 lb. per acre, with plants treated with 38 milligrams of the chemical growth retardant, grew to maturity and produced viable seed. Untreated plants in this high fertilizer concentration wilted within 24 hours and died within 3 weeks. While this specific finding cannot be applied directly to sugar beets in a field practice, it offers a very significant lead which should be investigated for crops like sugar beets often grown on soils of high salinity.

A new antibiotic, phleomycin, previously known to be effective against organisms causing human and livestock diseases, has been found to be effective in preventing or curing rust disease of snap beans under greenhouse conditions. Our scientists at Beltsville have demonstrated that an exceotionallv low concentration of phleomycin—one part of the antibiotic per million of water—sprayed on the leaf surfaces, will control bean rust. This lead has opened the way to further experiments to determine the effectiveness of phleomycin against other rusts and against downy mildew and anthracnose diseases.

Another chemical known as PAC (penacridane chloride), developed originally for medical purposes, is promising as a foliar and seed treatment fungicide. In laboratory tests at Beltsville, PAC killed both fungal and bacterial disease organisms carried on seed surfaces and it did not seem to slow seed germ-

ination. It has been applied to seeds as a soak, dip, or spray with equal success, and is bound to the surface even better than some chemicals accepted for commercial seed treatments. Even repeated washings of treated tomato seeds left enough of the PAC on the surface to prevent the growth of bacteria. The material may have an added advantage for practical use in its apparent absence of toxic properties to humans and animals. This development for a new use of a chemical to control seed-borne diseases is promising for any crop propagated by seed, and should not be overlooked for its possibilities in sugar-beet disease control.

In another area of work, our Federal-State scientists have recently found concrete evidence of substances in plants that make them physiologically resistant or susceptible to disease. A protein of the globulin type found in a particular race of flax rust fungus, was found to occur also in flax plants susceptible to the same race of the fungus. Plants resistant to the particular race do not contain the protein. This discovery is a basic one in plant science, and may prove especially important to plant breeders searching for disease-resistant plant materials. In principle, it offers a new tool to our scientists for almost all crops including sugar beets. This principle of physiological disease resistance in plants serves as an example of the results and need for the close working relationship between our scientists in the different disciplines—— specifically the geneticists and physiologists in this case. Here specific information on the globulins from each of four lines of flax and four races of rust of the fungus Me*lampsora linii* were used in serological analysis, which tests show a clear basic relationship between susceptibility to particular fungus races and plant varieties, thereby opening the door to a new approach in disease control.

These are only a few examples of our current research program. If time permitted I would like to tell you about our work on the Biological Control of Root Disease where we are attempting to develop "bugs to fight bugs"; of our work on translocation of large molecules from leaves to roots; of our plant exploration work to provide new germ plasm——etc.

This year we are commemorating: the 100th Anniversary of the U. S. Department of Agriculture and the approval of the Morrill Act, which created the national system of land-grant universities and colleges. In all of these institutions and in the U.S. Department of Agriculture, dedicated scientists have provided the knowledge that has enabled American agriculture to be the most productive that the World has ever known. We must

keep our research program strong to insure a constant flow of new knowledge that will be required in the future. Continued progress demands dynamic action. The graduate students of today must be convinced that Biological sciences have as many challenges and rewards as the Physical sciences, otherwise the next generation will not have the trained manpower to cope with problems ahead. The general public should be better informed as to our aims and objectives in agricultural research. We should never take our minds from the primary objective—to provide the most wholesome food supply in the most efficient and economical manner possible.

A Review of Recent Developments in the Chemistry of Sugar Beet

A. CARRUTHERS[1]

Mr. President, I was deeply honored when I received the invitation, from Mr. G. Rorabaugh, to speak before your general assembly and I count it a special privilege to be able to do so here in Denver, where many distinguished members of your Society have lived and worked. The honor which has been afforded to me is, I believe, a tribute to the work of our research group at Nottingham and in this connection T should like to mention especially Mr. T. F. T. Oldfield who has been actively concerned in all of the work which our group has carried out.

It can justifiably be stated that our knowledge of the composition of sugar beet juice and of the chemical changes which occur during processing: of the beet lias advanced very considerably during; the past two decades. Prior to this time quantitative data on juice analysis referred to oroups of components classifying them as sugars, nitrogen-containing; organic substances, non-nitrogenous organic substances and ash. Certainly many of the individual substances within the groups had been recognized for a long time, but now that thev can be separated and their concentrations can be measured bv methods which are specific and nrecise we are in a mudi better nosition to evaluate their significance in relation to factory performance.

With these advances in knowledge of the chemical composition of beet, and of the chemk al reactions in the factory process, it is possible to define the extent to which the technologist is irrevocably limited by the composition of his beet material and the extent to which he is ultimately capable of modifying the juice composition for maximum operating efficiency.

The composition of raw juice is basically determined by the composition of beet juice but it has been clearly demonstrated that the conditions operating in the diffuser can have a profound effect en the final composition. For instance the content of the pectin cornplex may be increased at least tenfold if the water used in diffusion is even mildly alkaline, as it may be if ammonlacal condensates are used for make-up. The extraction of excessive amounts of pectin not only represents a loss of valuable feeding stuff but it may also be detrimental to the process. The polygalacturonide fraction of the pectin is removed in clarifica-

[1] Director of Research, British Sugar Corporation, Limited, The Grange, Bramcote, Nottingham, England. Presented at the Twelfth General Meeting, American Society of Sugar Beet Technologists, Denver, Colorado, February 7, 1962.

tion but some of the araban portion is liberated during the liming stage and remains in the clarified liquors. When unfavorable diffusion conditions prevailed, amounts of araban of the order of 300 mg/lOOS were found in molasses and the pectin extracted was equivalent to a loss of as much as 7% of the beet marc.

It is now realized that even when temperatures throughout the diffusion system are adequate to suppress the activity of mesophilic bacteria, nevertheless, marked effects on juice composition can arise through the activity of thermophilic organisms. Strains of *Bacillus stearothermophihis* which can flourish at. temperatures up to 80°C have been found in factory juices and if the diffusion conditions allow these organisms to attain the logarithmic phase they will rapidly produce lactic acid and corresponding losses of sucrose will ensue. Following the demonstration that beet juice contains negligible amounts of lactic acid and that raw juice might at times contain as much as 0.6 g/lOOS, factories in greneral have adopted more stringent measures, either by maintaining a high level of temperature throughout the diffusion system or by a greater use of bactericidal agents, to suppress bacterial activity.

The thermophilic bacteria also attack nitrate which is derived from the beet and convert it to nitrite. At a later stage in the process sulphur dioxide is introduced into the juice and, by a complex reaction with the nitrite, yields imidodisulphonic acid, the potassium salt of which is sparingly soluble. If the concentration in the final syrups exceeds the saturation level, the imidodisulphonate may crystallize out with the sugar. Even where this does not occur it is still important to note that, to the extent that nitrite reacts with sulphur dioxide, this reduces the value of the latter since the real purpose of adding it is to minimize color formation.

When raw juice is produced under more or less sterile conditions its pH is about 6.3 and the aim of the clarification process is to prepare second carbonatation juice, containing less calcium than the raw juice, but with a pH of about 9. To bring about this change in pH without adding any bases, it is necessary that the acidic radicals, principally phosphate, oxalate, citrate, and to a less extent sulphate and malate, removed during clarification should appreciably exceed the removal of the basic magnesium and calcium ions. Fortunately the acid removal normally amounts to some 25 - 40 meq. per 100 sugar while the base removal only ranges from about 10-15 meq. per 100 sugar. The situation is still finely balanced, however, because some 4 - 6 meq. of excess base remaining in the juice is associated at the higher pH with

the amino acids and residual citrate which buffer the juice, while the acids produced by fermentation during diffusion, and by degradation of invert during clarification, neutralize a further 5-°15 meq. of excess base. Only the remaining fraction of the base excess is then available to permit absorption of carbon dioxide to the point in the carbonate : bicarbonate equilibrium corresponding to minimum residual lime salts.

Now that analytical techniques have made it feasible to measure the changes which occur during clarification it is possible to avoid empirical assessments such as 'effective' or 'natural alkalinity' and, in terms of precise chemical constituents, to ascribe unfavorable lime salts and lack of juice stability either to deficiencies in juice composition or to inadequate operating technique.

It is not always practicable or desirable to attempt to measure all of the constituents of juice but considerable insight into the processing features of juice, or of beet, can be obtained by determination of four main components which together are responsible for some 80 - 90% of the refractometric nonsugars in second carbonatation juice.

These four main components, potassium, sodium, amino acid nitrogen and betaine, are present in the same proportions relative to sucrose in raw[r] juice, or aqueous extracts of brei and they are not significantly eliminated in preparation of second carbonatation juice.

Potassium and sodium can be readily measured by flame-photometry and together these ions are responsible for virtually all of the ash components in second carbonatation juice. The associated anions in second carbonatation juice differ from those in raw juice but the average equivalent weight is known so that it is possible to calculate the weight contribution of these components.

In the past, the Stanek Pavlas copper reagent has been used extensively to estimate amino acid nitrogen in beet and it has also been applied to process juices. It has, however, been demonstrated that this value gives only a rough indication and is generally much higher than the true content of amino acid nitrogen. A direct determination using the Moore and Stein nmhydrin-hydrindantin reagent has now been developed to give a value which is far more closely correlated with the sum of the individual amino acids than is the Stanek Pavlas value.

About half of the nitrogen in clarified juice originates as amino acid in beet and most of this nitrogen is present in beet as glutamine which contains both an amide and an amino nitrogen group. The conversion of this amino acid to pyrrolidone car-

boxylic acid and ammonia causes particular problems during processing as the ammonia is volatilized in the evaporators, leaving the acid residue to contribute to juice instability.

The other main component, betaine, is a particularly stable compound and does not apparently associate in the specially undesirable processing difficulties but, since it is quantitatively the most prominent single organic nonsugar in beet juice, it obviously has a considerable influence on purity.

The main difficulty in estimating betaine has been that no specific reagent is known and hence the removal of interfering ions has hitherto been tedious. The discovery that betaine is not absorbed by mixtures of strong anion and weak cation exchangers, while all known interfering ions can be absorbed on the same resin mixture, now permits the simple but precise colorimetric determination of betaine after precipitation as betaine reineckate.

Particular emphasis has been given to these four determinations because they are part of our aim to give the factory technologist and the seed breeder simple methods of analysis which will yield results capable of precise interpretation. In this respect, individual measures of potassium and sodium provide more information than conductivity or ash, Moore and Stein nitrogen replaces noxious nitrogen, and we can also include betaine as the principal remaining nitrogen compound.

If the quality of beet is to be assessed by determination of individual nonsugars it is essential to have some method of compounding the individual results so that, for example, we can discriminate between a sample having a high potassium and a low amino acid content and another sample low in potassium but high in amino acid content.

The mean equivalent weight of the anions in second carbonatation juice is about 58 so that the potassium and sodium salts are respectively equal to 2 1/2 and 3 1/2 times the weight of potassium and sodium per 100 sugar. The effective weight of the amino acids and their degradation products in second carbonatation juice is assumed to be 10 times the Moore and Stein nitrogen per 100 sugar in raw juice or brei extract. The factor of 10 is slightly larger than the average factor required to convert amino acid nitrogen to weight of amino acid, but the ultimate contribution of the amino acids to the nonsugars will be greater than their actual weight if the juice stability is sufficiently reduced to make addition of soda ash necessary. Betaine passes unchanged through the factory process and the weight contribution of this

compound to the second carbonatation juice nonsugars is equal to the concentration per 100 sugar in raw juice or brei extract.

The contributions of the principal nonsugars in terms of values measured per 100 sugar in raw juice or brei extract are therefore summed to give an impurity value of 2.5 potassium + 3.5 sodium + 10 amino acid nitrogen + betaine. By this summation it is possible not only to obtain a measure of the total nonsugars which should be present in second carbonatation juice but also to assess the relative importance of each of the principal constituent groups.

The seed breeder may choose to concentrate his selection on one particular component but, since these components may vary independently, the ultimate criterion of quality is the purity of second carbonatation juice. Although second carbonatation juice can be prepared in the laboratory for such an assessment, the procedure is not generally suitable for treatment of large numbers of samples, particularly when the quantities of beet material are small.

At the 8th Meeting of the American Society of Sugar Beet Technologists, Brown and Serro reported a new method for clarifying pressed juice from individual beets to yield a clear juice for purity determination. The pressed juice was treated with lime and clarified in two stages with saturated oxalic acid solution. Data were presented to show that the purities obtained by the new method, called oxalation, and by standard carbonatation were essentially identical and the procedure was recommended for the assessment of beet quality. Subsequent analysis of oxalated juice, however, showed that the inorganic constituents were present in rather different proportions from those in carbonatated juice and the residual calcium level was some 10 to 20 times greater than normal. Since the solubility of calcium oxalate in water is very low, it is surprising that oxalic acid is not a more efficient agent in the juice system, but we also know that about 3% of the oxalate in raw juice is not precipitated in the factory clarification process, even though the residual calcium and oxalate far exceed the aqueous solubility product.

The oxalate treatment also eliminates about one-fifth to one-quarter of the potassium and sodium ions and, though these two effects are to some extent compensating in effect on purity, it is obviously desirable that the clarified extract should be as similar as possible to real second carbonatation juice. We have therefore used an adaption of the Brown and Serro method using 3 M phosphoric acid instead of saturated oxalic acid for deliming. The residual calcium, potassium and sodium in the phosphated juice

are very similar to the factory levels and the phosphate treatment is also superior in that less dilution is caused by 3 M phosphoric acid than by saturated oxalic acid, which is only 0.8 m. The purity of the phosphated juice is not distinguishable from that of standard second carbonatation juice and if further information on composition is required, the brilliantly clear juice can be employed for determination of potassium, sodium and betaine.

Amino acid nitrogen can also be assessed since the slight cyclisation of glutamine under the clarification conditions is largely balanced by the liberated ammonia which is determined at equivalent color yield by the Moore and Stein reagent. It is probably preferable, however, to measure the amino acid nitrogen directly in the pressed juice or in the lead extract used for determination of sugar in beet, since the dilution required is such that the color of the initial juice is unimportant.

An automated process has been installed at the Central Laboratory of the British Sugar Corporation to prepare phosphated juice from all of the thousands of samples of beet from the seed variety, fertilizer and other trials organized by the Corporation. Electronically controlled apparatus dilutes the pressed juice to a standard brix, adds the milk of lime and titrates the mixture with pH controlled burettes to the two end points. The heating and cooling stages are thermostatically controlled and the samples are processed at a rate of 12 per hour. The entire process, including the polarization of the clarified juice and estimation of solids with a fifth place dipping refractometer, is operated by one person.

In addition to sucrose, invert sugar and raffinose, de Whalicy and Gross showed chromatographically that beet syrups contained kestose, one of a series of trisaccharides composed of two molecules of fructose and one of glucose. Three such fructosyl-sucrose compounds can be formed by the transfructosylase activity of yeast or mold invertase and the detection of the trisaccharide as in sugar beet products was originally attributed to an action of yeasts or other micoorganisms. However, it has since been confirmed that the trisaccharide occurs naturally in beet and apparently may be present in greater amounts in beets which have been grown under drought conditions. Two of the trisaccharides can be produced from sucrose by an enzyme preparation from the leaves of sugar beet and these are apparently similar to those formed by mold invertase while yeast invertase additionally produces a preponderance of the third trisaccharide which does not generally occur in significant concentration in beet products.

It is fitting to recall that Brown and Serro revealed that myo-inositol and galactinol are normal constituents of beet and in some areas the inositol content may equal that of raffinose. The detection of these oligosaccharides and glycosides illustrates the superiority of the chromatographic over the chemical or enzymatic methods for the determination of raffinose.

Although white sugar is produced to extraordinary standards of purity, the improved analytical techniques now permit the estimation of some of the minute traces of impurities still remaining in the sugar and, from examination of the amounts of these constituents, it is apparent that some components are present in relatively higher proportions in sugar than in the standard liquor from which the sugar was crystallized. It is therefore clear that the impurities did not arise simply from the presence of a film of mother liquor on the crystals. This finding was to be expected if co-crystallization of raffinose and sucrose occur, or if the mother liquor becomes saturated with any compound such as potassium imidodisulphonate, but apparently this phenomena also arises with floc constituents which may be present in sugar at a concentration of more than 10 times that which could be attributed to a mother liquor film.

Floc and foaming present related, but not identical, problems in the production of high quality sugar. About 10 years ago, Eis and his collaborators showed that raw juice floc was largely composed of oleanolic acid and its glycosides. This group of compounds is commonly called saponin and Walker and Owens later demonstrated that white sugar floc contained many other constituents. They considered that the acid insoluble saponin was the prime cause of floc and that, as the saponin coagulated, it scavenged other impurities from the solution. Since floe is manifested in acidified beverages, methods have been evolved for measuring the floe produced in acidified white sugar solutions either visually or gravinietrically. The principal disadvantage of these methods is that the floe coagulates only slowly so that there is necessarily a considerable delay between production of the sugar and the determination of the floc characteristics. Moreover the gravimetric method normally measures only the methanol soluble portion, or alternatively the saponin fraction of the floe.

This latter was found to represent only about 20% or less of the total floe in British beet sugars and consequently a more general estimate of surface-active trace impurities seemed desirable as an assessment of the suitability of sugar for bottling purposes. The depressive effect of surface-active impurities on the polarographic oygen maxima of sugar solutions proved to be

a suitable basis for a general estimation of this type. The pioneer work of Vavruch in evaluating sugars on the basis of polarographic behavior had led to a simple technique and, by using a recording polarograph to determine the polarographic peak height from sugar samples collected at the commencement of each strike, it is possible to assess the quality of the sugar sufficiently rapidly to decide whether or not the sugar is suitable for bagging as bottlers' sugar.

That it has been possible to resolve many of the problems associated with sugar beet chemistry is due to the development of new analytical techniques such as paper chromatography, high voltage electrophoresis and ion exchange fractionation. These methods have permitted the detection and determination of constituents at concentrations far below the limits of the classical approach. Even with very precise methods of analysis, however, it is tedious, and sometimes impossible, to elucidate the order of events in a dynamic chemical or biological process but, with the advent of the atomic pile, cheap radioactive isotopes have become available so that it is now commonplace to employ radioactive labelling of specific components in order to detect any reaction products unequivocably. Moreover since the labelling is quantitative the ratio of each product to the precursor can readily be determined.

Carbon-14, one of the radio isotopes most suited for sugar investigations, has a very long half life so that the diminution in activity is of no significance in the period of any normal experiment and, as the emission is pure *(3* radiation of fairly low energy, only relatively simple screening and health precautions are required in the laboratory. The low energy of the emission presents some counting difficulties but, with very thin end-window counters, it is feasible to make direct G.M. counts of labelled sugars very rapidly and with a precision, as measurer by the count-rate : background ratio, similar to that obtained by the more time-consuming scintillation count techniques.

As an illustration, the detection and determination of residual oxalate in second carbonatation juice was achieved by clarification of juice containing a negligible weight concentration of radioactive oxalic acid. Traces of oxalic acid in clarified juice were identified chemically but, if calcium oxalate precipitation during defecation and carbonatation were incomplete, the measurement of residual oxalate in clarified juice by conventional calcium precipitation would require considerable verification because this latter precipitation might also be incomplete. In contrast only a few hours work were required to demonstrate that 3% of the

radioactivity, and hence 3% of the total oxalate in raw juice, was not removed by clarification. Experiments involving alkaline degradation of fructose-C14 also showed that the quantities of oxalate which could be produced by destruction of monosaccharides in evaporation were very small in comparison with the residual oxalate in thin juice and it was therefore possible to conclude that the prime cause of oxalate scaling in evaporators was incomplete oxalate elimination in clarification and not decomposition of oxalogenic substances.

The range of products which may be produced by the alkaline degradation of reducing sugars under different conditions is so vast that it would be a formidable task to attempt chemical separations for determination of the yield of any but the major degradation products in clarified beet juice. In this field also the load of work is significantly lessened by labelling the monosaccarides before clarification. In one typical experiment raw juice containing added glucose-14 was defecated with lime and carbonatated in the laboratory. Of the fructose 95% was degraded and, of the degraded material, none was absorbed on a strong cation exchanger while 93% was separated and recovered by absorption on a strong anion exchanger and elution with ammonium carbonate. This material was concentrated and separated into nine bands by high voltage electrophoresis. The bands were detected by autoradiography and the relative amounts of each were determined by elution and direct G.M. counting. About 5% and 40% of the activity was present in the two most mobile bands corresponding to glycollic and lactic acid respectively.

The four principal remaining bands contained saccharinic acids of increasing chain length as the mobility decreased. Alone responsible for one band was 2-4-Dihydroxybutyric acid, but as the chain length increased each band contained an increasing number of the isomeric saccharinic acids so that on lactonization of the hexosaccharinic acid band it was possible to identify five isomeric glucosaccharinc-lactones. At this stage the proportion of individual saccharinic acids in each band has not been established but it is known that the relative proportions of the acidic products can be varied by changing the clarification conditions and a range of saccharinic acids can be separated from molasses by ion exchange fractionation.

As an example of the more complex radiochemical applications in the field of photosynthesis, the work of Calvin and his collaborators is well known. On exposing photosynthetic materials to carbon-14 dioxide these workers found that, although

phosphates of glucose and fructose were rapidly produced, the first detectable free carbohydrate was sucrose and not a monosaccharide. Detailed examination of the changes in concentration of the radioactive compounds demonstrated that UDPG was labelled at an early stage of the process. UDPG was also being investigated by Leloir and Cardini and the characterization of this compound as a possible intermediary provided an important link in the chain of events leading to the present knowledge of the vital role of UDPG as a co-enzyme in the synthesis of sucrose.

Buchanan employed carbon-14 dioxide in photosynthetic experiments and demonstrated the formation of sucrose phosphate in the leaves of sugar beet as well as of other plants. He postulated that the sucrose phosphate was formed according to:

$$\text{UDPG} + \text{fructose 1 phosphate} \rightleftarrows \text{sucrose phosphate} + \text{UDP} \quad (1)$$

Leloir and his associates later demonstrated that an enzyme could be obtained from certain plant materials which would synthesise sucrose according to the following reaction:

$$\text{UDPG} + \text{fructose} \rightleftarrows \text{sucrose} + \text{UDP} \quad (2)$$

Extracts of sugar beet leaves or roots gave negative or non-reproducible results. However, workers in the Western Regional Laboratories of the USDA have recently demonstrated the presence of enzymes in young sugar beet leaves which will accomplish both of the reactions 1 and 2. In our laboratories at Bramcote it has been established that the root of the sugar beet contains an enzyme which will effect the synthesis of sucrose according to reaction 2. It was also shown that the enzymes and substrates necessary to provide a supply of the co-enzyme UDPG are present in the root and it is, therefore, not axiomatic that all of the sucrose is synthesized in the leaf system.

It is unnecessary to stress the importance of polarimetry, both financially and in process control in the sugar factory, and it has long been realized that automatic polarization is desirable, not only to minimize human errors, but also because the operation may be combined with automatic recording.

Many of the earliest attempts to avoid visual balancing of the polarimeter employed a single-field polarizer with a single photocell, and the 'crossed' position of the analyser was detected by the minimum in the photocell output. While the accuracy was similar to that of visual instruments, the photoelectric measurement was much more time consuming.

Photoelectric polarimeters were later designed using the conventional double-field polarizer and the two half-fields were fed separately to two photocells, the outputs of which were balanced

by rotating the analyzer. To eliminate differences in characteristics between the two photocells an additional blank balancing operation was required.

This additional balancing stage was avoided in an automatic polarimeter developed by the Spreckels Sugar Company. The two half-fields were fed to a single photocell but the light was interrupted by a rotating semicircular shutter so that, when the fields were unbalanced, the light intensity varied between a maximum and a minimum during each rotation of the shutter. The resulting alternating current output from the photocell was amplified to operate a balancing motor to equalize the field automatically; the phase difference between the mainline alternating current and the photocell output was employed to drive the quartz compensator in the correct direction to the balance point at which the alternating current from the amplifier fell to zero. As far as is known this instrument, which was described to the ASSBT in 1948, represented the first successful fully-automatic polarimeter. The polarization of the sample was printed directly from the quartz compensator and the results, estimated to 0.01%, could be obtained and printed at the rate of 400 samples per hour.

The principle of scanning the fields from a double-field polarizer to produce alternating current from a single photocell, in conjunction with various forms of time-base to indicate the correct direction of adjustment to the balance point, has been used in several subsequent automatic saccliarimeters and polarimeters and some of these instruments have been employed commercially.

A major advance in practical automatic polarimetry has however occurred more recently in the development at the National Physical Laboratory of an instrument having no moving parts. Both the cyclic modulation of the incident polarized light and the balancing of the optical rotation of the sample are accomplished by means of the magneto-optic or Faraday effect, that is the use of a controlled electromagnetic field to render a glass block optically active. An adaptation of this instrument, the ETL-NPL automatic polarimeter, has been installed in control laboratories and in the tare laboratories at many sugar factories. Interference and Polaroid filters are used to produce a narrow waveband of plane polarized light which is passed through a glass rod forming the core of an electromagnet carrying a 60 cycle a-c supply. The alternating magnetic field induces alternating optical activity in the glass so that the plane of polarization is modulated over an angle of 3° either side of the unmodulated direction. The mod-

ulated beam then passes successively through the sugar solution, through a second Faraday cell to a Polaroid analyzer set in the crossed position relative to the polarizer and thence to a photomultiplier. The photomultiplier output is rectified to provide negative feedback inducing optical activity in the second Faraday cell equal and opposite to the rotation of the sample. The amplifier gain is so arranged that the instrument remains balanced automatically and the current flowing in the second Faraday cell is proportional to the optical rotation of the sample. This current can be used to operate a precision indicator or to display the polarization on an illuminated digital converter and the polarization can also be recorded automatically on a printing unit or punch card machine. The basic range of the instrument is only ± 0.5 angular degrees and the tube length employed is much less than in visual polarimetry. High relative precision at low rotation is advantageous because the absorption of the sample usually decreases exponentially while the rotation decreases linearly with decreasing cell length. It is therefore possible to measure optical activity in solutions which are far too dark for precise visual polarimetry. The polarizer can also be offset to examine angular rotations within its range of ± 0.5° anywhere in the total range of — 90° to + 90°.

For solutions containing more than 1.3% sucrose the ultimate precision of the polarimeter is about twice that of visual instruments while for solutions of low optical activity the automatic polarimeter is considerably the more precise since, with no offset, it is possible to obtain a full-scale reading for a 1.6% solution of sucrose in a 4 cm cell. The polarization of this dilute solution can be determined to 1 part in 2,500.

Various other automatic polarimeters have been developed using the Faraday effect either for modulation, compensation or both and these instruments have raised interesting problems in connection with the International Sugar Scale. There is a growing tendency to employ green light sources because their photoelectric characteristics and ease of reproduction are superior and photoelectric instruments also permit the use of shorter tube lengths. The International Sugar Scale at the moment is strictly only applicable to the dichromate filtered white light source and the 20 cm tube length and consequently, I.C.IJ.M.S.A. is endeavoring to define a sugar scale for modern instruments. We hope to reach some measure of agreement on the new scale at the 1962 Session.

Chemical Control of Cercospora Leaf Spot in Sugar Beets

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Leaf spot of sugar beets caused by the fungus parasite *(Cercospora beticola Sacc.)* is commonly found on sugar beets in the humid areas of the United States and in areas where temperatures are favorable and irrigation is used. Severe epidemics frequently occur in Minnesota, Ohio, Michigan, Nebraska, Colorado and several other states (1 I)[2]. The lesions on the leaf result from the invasion of the germ tube from the fungus spore through the leaf stomato and progressive growth of mycelium within the leaf. Older leaves are affected, and when enough spots occur they coalesce causing a complete breakdown of leaf surface. As these leaves die the beets produce new leaves, using stored reserves of sugar. The end results of a severe epidemic are a reduction of sucrose percent in the beet along with slower beet growth, and consequently a lower tonnage yield.

In 1899 and again in 1909 Duggar (6, 7) suggested Bordeaux mixture for the control of Cerospora leaf spot in the United States. In 1914 Townsend (11) reported satisfactory control by this method. Vestal (12) reported that European workers gave much more attention to the control of leaf spot by use of fungicides than investigators in the United States. Nearly all of these European workers reported encouraering results from the use of Bordeaux and that the increase in yield more than paid for the material and labor.

Coons et al., (5) conducting spray and dust tests over a 3-year period, showed satisfactory results when leaf spot was a factor. Five applications of copper sulphate-lime dust in 1925 srave the best control of leaf spot that year. Tests in 1926 and 1927, years in which the incidence of leaf spot damage was slight, showed no significant gains from either dusting or spraying. In 1928 copper sulphate-lime dust applied three or more times at the rate of 35 pounds per acre per application, gave on the average, increased tonnage, sugrar percent, purity, and estimated sugar production which was more than enough to offset the cost of treatment.

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[2] Numbers in parentheses refer to literature cited.

Experiments were carried out by Vestal (12) in the Mason City-Britt, Iowa, areas in 1929 and 1980 using Bordeaux mixture, copper hydroxide paste, copper-lime dust as fungicides. A very slight increase in the percentage of sugar was reported, but the over-all sugar per acre was not increased.

LeClerg (8) reported results of two seasons' experiments (1931 and 1932) on the efficiency of copper sulphate-lime dust and Bordeaux mixture for the control of Cercospora leaf spot in the vicinity of Chaska, Minnesota. In three tests conducted under epidemic conditions of the disease, spraying significantly increased yields. The increase from dusting was significant in only two tests with the third test approaching significance. LeClerg also observed that the dusted plots appeared to have a somewhat lower measure of leaf spot control than the sprayed plots, but a comparison of only statistically significant data from dusting- and spraying tests revealed that the percentage increase in yield due to dusting and spraying was nearly identical. He concluded that four to five treatments were necessary for control.

Brown (3) reported that gain in sugar production from dusting and spraying of plots in Canada was quite marked in years when Cercospora occurred. His results showed dusting was as good or better than spraying.

In 1941 the American Crystal Sugar Company (1) conducted tests at Rocky Ford on 12 varieties using yellow cuprocide sprayed twice. Significant increases in yield and sucrose were obtained, but only on those varieties whch were leaf-spot susceptible. Tests with dusts were conducted at Chaska, Minnesota, in 1941 and at Mason City in 1942. There was practically no leaf spot in the 1941 tests, and no differences were obtained from four copper dusts used. In 1942 at Mason City, Iowa, yellow copper oxide treatment gave increased yield and sugar percent.

Young (13) of the Ohio Agricultural Experiment Station conducted spray and dust treatments in 1939 and 1940 using tribasic copper and obtained much higher root yield and sugar percent than from non-treated plots and strips.

In 1947 Stewart (10) conducted a test on the Plant Industry Station at Beltsville, Maryland, to evaluate susceptible and resistant varieties of sugar beets under extreme conditions of leaf-spot exposure, with and without fungicidal treatment. He used Bordeaux mixture as the fungicide and sprayed eight times. His results showed a gross sugar increase, for the fungicide treated plots over the untreated, of 225 percent for the susceptible variety and 122 percent increase for the resistant variety.

In 1957, 1958 and 1959 (unpublished data), leaf-spot epidemics were noted in the Mason City, Iowa, and Grand Island, Nebraska, areas. Spraying and dusting programs using Nabam and Maneb were used with spraying showing the greater promise. All this work indicated that chemical control of this disease was possible.

In 1960 several replicated spray tests were designed to check the following hypotheses: (1) Spraying with fungicides will control leaf spot and increase gross sugar yield; (2) addition of soil nitrogen will help control leaf spot; (3) fungicides contribute needed nutrients (leaf feeding) to the plants as well as protecting them from leaf spot; (4) fungicidal sprays have a greater effect on susceptible varieties than resistant varieties; (5) and Maneb gave better protection than Nabam as measured by gross sugar yield.

Methods and Materials

Three replicated tests were involved in this study, two at Mason City, Iowa, and one near East Grand Forks, Minnesota. Each test was replicated 12 times with split plots. The main plots consisted of four treatments: (1) Check or no treatment; (2) 200 units of nitrogen sidedressed after thinning; (3) spraying with Nabam at two quarts plus 3/4 pound of zinc sulphate per acre; (4) and spraying with Maneb at 1 1/2 pounds per acre. The subplots were varieties American #3S and American #3N. The variety American #3S was considered as leaf-spot resistant. The subplots were planted in strips and the analysis of variance used was similar to an example given by Cochran and Cox (4) page 232.

One test at Mason City was conducted in the leaf spot nursery, i.e., an area where the incidence of disease is favored. The climatic conditions, which are usually hot and humid during- the summer, were supplemented by sprinkling each morning. The nursery had grown a crop of infected sugar beets the previous season and the infected tops were left on the ground. In addition the sugar beet plants were inoculated with a suspension of spores obtained from washing; over-winter leaves on which the pathogen was profusely sporulatins;. The spore suspension was sprayed over the entire test area on June 27.

Leaf-spot symptoms could be found throughout the test on July 15 and the first of the fungicide sprayings was started on that date. Other spraying applications were made on July 29, August 17 and August 26. The last spraying was 21 days prior to harvest.

Plots were eight rows wide (22-inch rows) and 40 feet long. 1 he three outside rows on each side of the plots were planted

to a susceptible variety, therefore, the resistant variety American #3S was surrounded with at. least six rows of susceptible plants. The two center rows, one American #3S and the other American #3N, were harvested separately for yield. Two ten-beet samples were taken from each subplot for sucrose and nitrogen determinations.

The other test at Mason City was similar to the one described above except the test was not inoculated with leaf spot and it did not have beets as a preceding crop. Also, it was not under a sprinkling system. This test was relatively free from leaf spot throughout the season, although it was within 1/2 mile of the leaf spot nursery test, and on similar soil type.

The spray and nitrogen treatments also were the same as previously described. The leaf-spot nursery test was harvested September 27 while the field test was harvested September 26, 1960.

The third test near East Grand Forks, Minnesota, was verisimilar to the leaf-spot nursery test at Mason City, Iowa, except plots were 6-rows wide and 50-feet long. This test was inoculated with a spore suspension and the preceding crop was sugar beets, however it did not have a sprinkler system to supplement the humid conditions. Leaf spot reached epidemic proportions in the check plots later than in the Mason City leaf-spot nursery. Spraying dates were July 20, July 30, August 11 and August 23. Plots were harvested October 6, 1960.

The total nitrogen content was determined by a micro-Kjeldahl digestion process as described by Payne et al., (9), and is reported as percent on dry substance.

Results and Discussion

The results of these three tests are shown in Tables 1, 2 and 3. In comparing gross sugar yield of the treatments in Tables I and 3 (the tests under heavy leaf-spot epidemic), both spray treatments were above the one percent level of significance when compared with the check and Maneb was significantly higher than Nabam. The yields after nitrogen treatment in both tests were lower but not significantly different than the check for yross sugar per acre. The Maneb treatment was significantly higher than the check for tonnage, percent sugar, purity and lower in total nitrogen content. The Nabam treatment was significantly higher than the check in tonnage and percent sugar in Table 1, and significantly higher in tonnage in Table 3.

Comparing the nitrogen treatment and the check under leaf-spot conditions, there was no significant increase in tonnage,

however, the trend was in favor of the nitrogen treatment. For percent sugar and purity there was definite reduction, which was highly significant when the nitrogen treatment was compared with the check. The nitrogen treatment also increased the total nitrogen content of the beets which is an extremely harmful constituent in sugar beet processing.

Table 1.—Stand, yield and chemical results for treatments and varieties from a test at Mason City, Iowa, in the leaf-spot nursery.

Treatments	L.S.[1] reading	Lbs sugar per acre	Tons per acre	Percent sucrose	Percent purity	Total nitrogen	No. roots per 40 ft
Maneb	1.21	4245	19.80	10.78	86.8	1.09	30.8
Nabam	2.12	3522	17.50	10.11	85.7	1.16	29.3
Check	4.42	2992	16.39	9.15	84.1	1.23	29.1
200 Units N.	3.42	2744	16.61	8.22	79.8	1.63	28.9
LSD (0.05)		317	1.10	.65	1.9	.16	NS
LSD (0.01)		426	1.48	.88	2.6	.22	NS
Varieties							
American #9N	3.14	3407	18.26	9.27	83.6	1.22	32.1
American #9S	2.58	3343	16.88	9.85	84.5	1.33	26.9
LSD (0.05)		NS	1.28	.15	NS	.08	2.3
LSD (0.01)		NS	.....	.21	NS	.....	3.3
Treatment x variety		NS	NS	NS	NS	NS	NS

[1] 1 = Very good resistance
2 = Good resistance
3 = Average resistance
4 = Susceptible
5 = Very susceptible

Visual leaf-spot readings were taken on the Mason City nursery test and are given in Table 1. Maneb rated better than Nabam and both of these fungicides ranked better than the nitrogen treatment or the check. These observations were in accord with the yield data. The nitrogen treatment, however, was rated better than the check which is not in accord with the yield data. Evidently the plants in the nitrogen treated plots were somewhat greener and perhaps recovered somewhat quicker which made them look more resistant. Although the leaf-spot ratings were better for the nitrogen treatment, the data in Table 1 shows that the tonnage was not significantly increased over the check and that sugar percent and purity were significantly decreased.

From the results of these two tests under leaf-spot conditions we can conclude: (1) that spraying of fungicides for the control ot leaf spot was effective; (2) Maneb was a better fungicide than Nabam; (3) and the use of additional nitrogen did not protect the plants against leaf spot but caused a decrease in gross yield and purity and an increase in total nitrogen. Therefore the use

of nitrogen was detrimental rather than helpful for leaf-spot control.

The data shown in Table 2 were from the plot field at Mason City, Iowa, which was relatively free of leaf spot. This test was within a half mile of test results shown in Table 1. The objective of this test (Table 2) was to determine if spraying with Nabam or Maneb was beneficial in the absence of leaf spot, i.e., were we leaf feeding the plants with these sprays?

The results in Table 2 show that the check and the two spray treatments are not significantly different for any of the characteristics studied. There was no indication of leaf feeding resulting from spraying Maneb or Nabam on the plots.

The nitrogen treatment again significantly lowered the gross yield of sugar, sugar percent and purity below all the other treatments.

Because the two tests at Mason City were not within the same test area, they cannot be considered as crucial tests. However, they do support the hypothesis that the main effect obtained by spraying was to protect the plants against the leaf-spot fungus, and that the increased yield was not due to the correcting of an element deficiency.

Table 2.—Stand, yield and chemical results for treatments and varieties from a test at Mason City, Iowa, which was relatively free of leaf spot.

Treatments	Lbs sugar per acre	Tons per acre	Percent sucrose	Percent purity	Total nitrogen	No. roots per 50 ft
Maneb	6815	23.52	14.57	88.1	.99	40.7
Nabam	6760	23.62	14.39	87.4	1.04	40.1
Check	6700	23.40	14.40	87.8	1.07	40.5
200 Units of N.	6168	24.19	12.79	84.1	1.39	41.4
LSD (0.05)	457	NS	0.39	1.2	.09	NS
LSD (0.01)	...	NS	0.53	1.6	.12	NS
Varieties						
American #3N	7045	25.93	13.60	86.6	1.09	43.2
American #3S	6174	21.42	14.47	87.0	1.14	38.2
LSD (0.05)	NS	1.10	0.40	NS	NS	1.9
LSD (0.01)	NS	1.56	0.56	NS	NS	2.6
Treatment x variety	NS	NS	*	NS	NS	NS

Significant interaction for percent sugar

Treatment \ Varieties	Am #3N	Am #3S
Maneb	14.22	14.92
Nabam	14.00	14.77
Check	13.77	15.02
200 Units of N.	12.41	13.17

LSD (0.05) For the difference between two variety means for the same treatment = 0.38
LSD (0.01) For the difference between two treatment means for the same variety = 0.56

Table 3.—Stand, yield and chemical results for treatments and varieties from a test near East Grand Forks, Minnesota, in the leaf-spot nursery.

Treatments	Lbs sugar per acre	Tons per acre	Percent Sucrose	Percent Purity	Total Nitrogen	No. roots per 40 ft
Maneb	4161	15.46	14.52	84.4	1.38	34.7
Nabam	4126	14.82	13.95	83.6	1.55	36.1
Check	3497	12.84	13.65	83.0	1.57	34.5
200 Units of N.	3409	13.23	12.88	81.3	1.75	35.8
LSD (0.05)	227	0.86	0.36	1.2	.12	NS
LSD (0.01)	304	1.15	0.48	1.6	.17	NS
Varieties						
American #3N	3614	13.06	13.80	83.8	1.41	35.7
American #3S	4131	15.10	13.69	82.3	1.72	34.9
LSD (0.05)	226	0.86	NS	0.8	.09	NS
LSD (0.01)	318	1.21	NS	1.1	.13	NS
Treatment x variety	NS	NS	**	NS	NS	NS

Significant interaction for percent sugar

Varieties / Treatment	Am#3N	Am #3S
Maneb	14.86	14.17
Nabam	14.24	13.66
Check	13.57	13.73
200 Units of N.	12.55	13.21

LSD (0.05) For the difference between two variety means for the same treatment = 0.29
LSD (0.01) For the difference between two treatment means for the same variety = 0.31

The varieties reactions in these tests are also recorded in Tables 1, 2 and 3. In Tables 1 and 2 the varieties were not significantly different for sugar per acre, however, American #3N was the top ranking variety in both tests. American #3N also was significantly higher in tonnage but lower in percent sugar than American #3S. This tonnage increase could be due to the significantly better stand of American #3N in the Mason City tests. Table 3 shows that American #3S yielded significantly more sugar per acre than American #3N. This significance was mainly due to a highly significant increase in tonnage in favor of American #3S. American #3N however was significantly higher in purity and lower in nitrogen content. The stands of the two varieties at East Grand Forks were nearly equal, therefore, the tonnage differences cannot be contributed to stand in this test.

Two significant treatment X variety interactions for percent sugar were detected. They are shown in Tables 2 and 3. The interaction in Table 2 just reached the significant point and may be due to chance. The varieties did respond slightly differently to the various treatments but no significant trends were found. The three treatments, Maneb, Nabam and Check were not significantly different from each other in either variety. The

magnitude and the percentage differences between the check plots and the nitrogen plots were different for the two varieties. American #3N showed a drop of 1.36 percent sugar or a percentage decrease of 9.9 while American #3S showed 1.85 percent sugar drop or a 12.3 percent decrease. These slight differences in trends probably were the main factors for this significant interaction and it probably has little or no real biological meaning.

The treatment X variety interaction for percent sucrose in Table 3 was highly significant indicating the varieties were not reacting the same to the various treatments in this leaf-spot exposure test. The interaction data given in Table 3 shows that the sugar percent of American #3N was significantly increased when sprayed with Nabam or Maneb. Based on the significantly better sugar content, Maneb produced significantly better protection when compared with Nabam. The nitrogen treatment significantly reduced the sugar content one full percent below the check.

When American #3S was treated with Nabam the sugar percent was not significantly increased as it was with American #3N. When American #3S was treated with Maneb the sugar content was significantly raised over the check, but was significantly lower than the American #3N Maneb treatment.

When American #3S was treated with 200 units of nitrogen it reduced the sugar content by 0.5 percent below the check treatment, while American #3N was reduced a full percent.

In this test the sugar content of the susceptible variety was increased to a greater degree by spraying than was the American #3S resistant variety. However, under other conditions such as excess soil nitrogen, the sugar content of American #3N was decreased to a greater degree than was American #3S.

The data, shown in Table 1 did not reveal any treatment times variety interaction. The difference in results obtained in these two tests could be explained by location and soil difference. However, a more reasonable explanation would come from the observation that the Mason City test reached its peak about three weeks sooner than the East Grand Forks test, therefore, the beets in the Mason City test had more time to recover.

Another interesting factor concerning the two varieties was the total nitrogen content. In all tests American #3S had a higher nitrogen content than the American #3N, and in two of the tests this difference was significant. These data indicate that American #3S was a heavier nitrogen feeder than American #3N. This might have been expected as American #3S inherently produced more and taller leaves than American #3N, however, in

most tests American #3S has produced as good, if not better, sugar content than American #3N. The nitrogen assimilation by American #3S certainly affected the purity because this variety was equal to American #3N in sucrose percent, but was highly significantly lower in purity.

Andersen (2) postulated that dalapon caused plant protein degradation in sugar beet seedlings and the degraded proteins were translocated to the roots causing the young seedlings to turn yellow and chlorotic. Perhaps leaf spot causes a similar phenomena, i.e., as the leaves become more infected, more nitrogen compounds are translocated to the roots. This hypothesis would explain why beets sprayed with Maneb have less nitrogen in their roots than the check. The same would be true for Nabam sprayed plots.

In susceptible plants like American #3N the leaf spot may "burn the plants down" rather quickly, allowing them time to recover before harvest. Whereas in the resistant varieties like American #3S the plants may withstand the disease epidemic for several weeks, but later in the season the resistant varieties start to lose their leaves. As the resistant varieties become infected, protein degradation occurs in the leaves and nitrogenous compounds are translocated to the roots. If this happens within a week prior to harvest or under frost conditions which slow up growth, it would be possible for the resistant plants to have more nitrogen in the roots than a susceptible plant. This extra amount of nitrogen could cause a substantial reduction in beet juice purity.

Additional research will be needed to test the above hypothesis.

Summary

Three replicated tests with the same treatments and design were involved in this study, two tests were under leaf-spot epidemic and one test was relatively free from leaf spot. Different treatments were compared for the control of leaf spot under nursery epidemic conditions and these were compared to a test which had identical treatments but was relatively free from leaf spot.

From the data submitted in this report the following conclusions were drawn:

1. Cerospora leaf spot of sugar beets can be controlled by spraying with Maneb or **Nabam.**

2. The Maneb spray treatment gave better yields and better leaf-spot control in the disease tests than Nabam.

3. Additional soil nitrogen caused a reduction in sugar percent and purity in all tests and did not help the plants to resist the leaf-spot disease.

4. No other physiological effects from the use of these compounds were detected. The main effect of Maneb and Nabam was for plant protection.

5. Two treatment X variety interactions for sucrose were found. In general, however, the spray treatments helped the resistant variety as well as the susceptible variety.

6. An hypothesis was postulated that Cerospora leaf spot causes protein degradation in the sugar beet leaves and some of the degraded proteins were translocated to the roots.

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Sugar Beet Mechanization in the U.S.S.R.

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Sugar beets are grown principally in the Ukraine area on collective farms and in Central Siberia on state farms. In the New Lands area in Central Siberia where 80 million acres have been brought under cultivation in 5 years, spring wheat is now the main crop. Thirty five percent of all cropland is in wheat; 1.1 percent in sugar beets. However, the plan is to raise more sugar beets and less wheat in the future. In 1958 there were 5,750,000 acres of sugar beets in the Soviet Union; in the next five years the plan is to reach 8,500,000 acres. The land devoted to sugar beets has been increasing steadily since 1945.

Slightly over 50 percent of the population lives on the land. There are 6500 state farms and 53,400 collective farms in the U.S.S.R. The average size of a state farm is 22,000 acres and of a collective farm 7000 acres.

Plowing is done to a depth of 13 to 14 inches for sugar beets. Plans call for going to 16 inches depth in the near future. The following fertilizers per acre are recommended in the Ukraine: phosphate, 150 lb (20% P); ammonia, 40 lb (30% N); and potassium, 40 lb (30-60% K). Where available, 4 to 7 tons of manure per acre are used for sugar beets. Sugar beet and dairy enterprises often are on the same farm.

A spacing for mature sugar beet plants of 6 to 9 inches after thinning is desired. A 12-row sugar beet planter was being tested which placed rows 18 inches apart. Another unit was seen with three separate six-row units mounted on a three-point hitch—one behind and one to each side of a crawler tractor. Each of die units could be operated independently with the hydraulic system.

On one state farm visited, 250 acres were used for seed stock. A large trencher made a ditch two feet wide and six feet deep. The sugar beet roots were placed in the trench at a depth of more than 2 1/2 feet. A one-inch layer of soil was placed over each layer of roots in the trench and the top 2 1/2 feet filled with soil to prevent freezing. The root stocks are used for seed production the following spring.

Whole seed was used exclusively; no segmented seed was being used. Monogerm seed was being tried on an experimental basis and it was anticipated that 250,000 acres would be planted in

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1959. With a plant population of 28,000 per acre, approximately 15 percent higher yield was claimed as compared with a plant population of 40,000.

Normally about 40 man-hours are required per acre to block and thin the sugar beets. When a moving covered platform was used to transport the workers, the labor requirement for thinning was 20 man-hours per acre. Mechanical thinners were being used on an experimental basis. Cross cultivation with conventional small duck-foot tools followed by manual thinning was being used on a field scale basis which required 30 man-hours per acre. Ninety percent of all labor before harvesting was spent on manual thinning and weeding.

The most popular harvester is a three-row unit which grabs the beet, and carries it to the rear of the machine where the top is removed. The tops are placed in one container and the topped beets in another. Each is accumulated and then dropped in separate piles on the field. Women clean the piles of sugar beets—usually four women to a group—which requires about 40 man-hours per acre. Eighty five percent of all labor in harvesting sugar beets is used in cleaning and loading. The tops are used for silage and animal feed.

About four to six times as much labor is spent on production of sugar beets in the U.S.S.R. as compared to the U.S.A. In 1956, the U.S.A. required 0.23 man-hours per hundredweight of yield; in the U.S.S.R. on state farms, 0.95 man-hours per hundredweight; and on collective farms 1.41 man-hours per hundredweight were required for sugar beet production (Volin, 1958).

The beets weighed about 2 pounds each. The sugar content is from 17 to 18 percent. The director of one farm indicated that 80 percent of the Ukraine is now machine-harvested, 15 percent mechanically lifted and the remainder manually harvested. There are 30,000 sugar beet harvesters in the Soviet Union.

In the Altai region in the New Lands area, approximately 6 to 7 tons per acre yields are obtained; in the Ukraine 8 to 9 tons per acre. The average yield for the U.S.S.R. has been 7 tons per acre and the U.S.A. 16.8 tons per acre during the last four years. A well-managed state farm visited by the group in the Ukraine area near Kiev produced about 16 tons per acre. Another state farm in the New Lands area produced 11 tons per acre.

The Soviet Union is dependent on sugar from beets exclusively for its source of sucrose. The acres in sugar beets and the yield of sugar beets have been increasing steadily since World War II. The production of centrifugal raw beet sugar in 1959 was 7,160,000 tons (of 2000 lb), which is double the 1951 production.

About 450 acres are cultivated for each tractor and 650 acres per combine. In the U.S.A. each tractor and combine would cover about 100 acres per machine. In the Central Siberia area there are 35 acres of tillable land per farm worker For the U.S.S.R there are 4.2 acres of cropland per farm inhabitant; U.S.A., 20 acres per farm inhabitant.

An experimental three-row sugar beet harvester was seen equipped with a hydraulic row positioning device that would automatically stay on the rows. Machines did not have power take-off shields for safety. In the Ukraine harvested sugar beets are left on the ground from 1 to 5 days. The harvest is usually completed by the end of October.

Small and large mechanical loaders are available ana just beginning to be used for moving piles of beets to a truck or wagon. Two different sugar beet loaders were demonstrated. One was an individual unit mounted on the back of a tractor which backed into the pile and elevated the beets onto a truck. The other was mounted on the rear of the truck with a long boom-type hook which reached out and dragged the beets up onto the loader.

The beets are sold to a government-owned sugar plant. Forced ventilation of the sugar beet piles is practiced. Prices are set by the government. To encourage production, a higher price is paid for beets produced over the government plan.

All sugar sold in the Soviet Union is beet sugar which is more coarse in grain size than sugar in the U.S.A. A crystalline brown sugar is available in restaurants for coffee and tea. Sugar cubes are also available. The people on the collective farms must work one hour for enough money to buy a quart of pasteurized milk, two and one half hours for five pounds of sugar, and two months for a suit.

Table 1.—Sugar beet and sugar production in U.S.S.R. and U.S.A., 1935-59.

	1935-39		1945-49		1950-56		1958	
	U.S.S.R.	U.S.A.	U.S.S.R.	U.S.A.	U.S.S.R.	U.S.A.	U.S.S.R.	U.S.A.
Acres harvested (thousands)	5100	950	2500	870	3800	880	5750	950
Yields (tons/acre)	6.5	12	6	13.5	6.5	16	7	17
Production roots (million tons)	20	9.5	12	10.5	23	13	39.5	15
Production sugar			2.3	1.5	4.0	1.9	6.2	2.3

Summary

The planting operations are well mechanized. Thinning still requires considerable manual labor. The harvesting of sugar beets is being mechanized rapidly with three-row harvesters.

These units are followed by crews of four women each to clean and finish topping the piled beets. Financial incentives are being used to encourage greater production. All sugar sold in the Soviet Union is from sugar beets.

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Thin Layer Chromatography of Sugar Beet Saponins

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Introduction

Van der Haar (3,4)- first recognized oleanolic acid and glucuronic acid as components of sugar beet saponin. He isolated three saponins with different solubility properties. In as much as saponins easily form emulsions and addition compounds with many substances (fats, fatty acids, phosphatids, e.g.) they are very difficult to purify and have not yet been obtained in crystallized form. The sapogenins on the other hand crystallize readily and can be obtained in a relatively pure state. It is, however, very difficult to determine the purity of a sapogenin preparation as the molecular weight is high (oleanolic acid = 456), the melting-point, accordingly, is very high and there are several substances in the same group with only slight differences in composition and structure.

Paper chromatography seemed to offer a good way to analyze these substances. Walker and Owens (9) investigated floe components of beet sugar by this method and by paper electrophoresis. They did not get a satisfactory separation; with several solvents the spots either failed to move or moved with the solvent front. Finally a solvent was found that separated oleanolic acid and saponin with R_F values of 0.2 and 0.8 respectively. The resolution of the spots, however, depended on the concentration of the mixture. Paper electrophoresis was not satisfactory either, because of poor resolution of different sapogenins. There was an indication that saponin from floe consisted of two substances.

Bauserman and Hanzas (1) found that on paperchromatograms, purified beet saponin behaved differently from saponins in beet juice. They ascribed the differences in behavior to saponins in the beet juice being in the salt form; with purified saponin salts they too obtained as R_F values either 0.0 or 1.0 and in some cases variable R_F values; R_F of oleanolic acid was 1.0 or 0.0. Mg, Ba and Ca were found associated with the saponin spots. With water as solvent, two spots were found for saponin.

Thin layer chromatography was first developed by Kirchner, Miller and Keller (5); they used glass strips, covered with a thin layer of absorbent (chromatostrips). Silica gel appeared to be the best absorbent for terpenes. Other workers used glass plates (chromatoplates) covered with absorbent (6,7,8,10).

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[2] Numbers in parentheses refer to literature cited.

This new method, in contrast to paper partition chromatography, is essentially absorption chromatography, and as such is suitable for substances insoluble in water. Therefore, it seemed a good means of separating sapogenins and possible also saponins.

Methods

The chromatograms were made on glass plates (15x15 cm) covered with a thin layer (~ 0.3 mm) of silicagel (Kieselsaüregel G, Merck, a special preparation of silicagel, mixed with a small amount of gypsum). The plates were dried in the air at room temperature. The spots were placed 2 cm from one side. The plates stood vertically in glass tanks (18x18x12.5 cm), the bottom of which was covered with 100 ml solvent. The time required for development is very short, about 20 to 40 minutes.

A saturated solution of antimony trichloride in chloroform was used as a spray reagent. After heating for 15 to 30 minutes

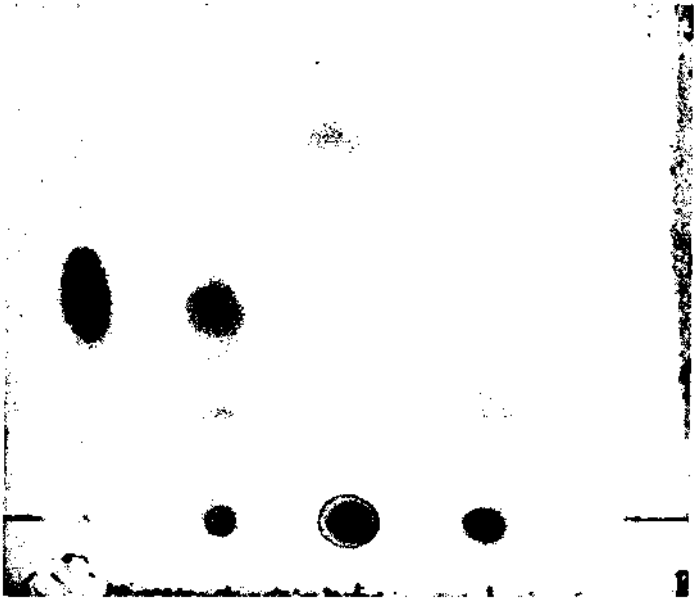

Figure 1.—1 = beet sapogenin b, 2 = beet sapogenin a, 3 = quillaic acid, 4 = glycyrrhetic acid. Solvent: (benzene 90 ml, ethanol 96% 10 ml).

at 100°C, blue-violet and sometimes yellow spots are visible. The blue-violet spots have a light orange fluorescence in ultraviolet light.

The procedure used for the isolation of saponin from sugar beet was as follows:

Acidify raw juice to pH 1 and heat at 90°C for 1 hour, let cool overnight and collect the precipitate by decantation and centrifugation and wash with slightly acidified water. Extract the wet precipitate with 96% ethanol. Evaporate the ethanol carefully in a vacuum desiccator at room temperature.

The saponin preparation can be separated in two fractions as follows: to obtain fraction a, extract the dry matter with warm acetone and evaporate the acetone at room temperature in a vacuum. The residue, fraction b, is soluble in dilute ammonia and may be precipitated by acidification.

The preparation of sapogenin was the following: hydrolyze by boiling the saponin for 6 to 7 hours in a solution, containing, 45 to 50% ethanol and 5% HCl. After cooling, dilute with water and collect the precipitate. For comparison with beet sugar saponins we isolated some closely related saponins from the same chemical group (β- amyrin group, Figure 2).

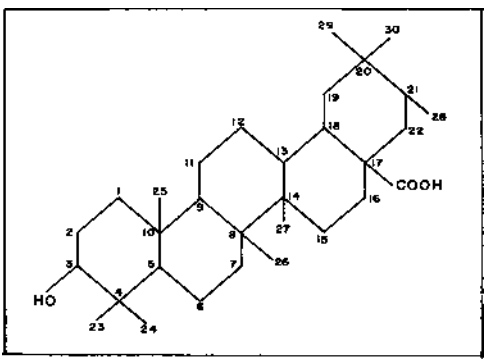

Figure 2.—Sapogenin formula.

β- amyrin group

oleanolic acid : 23,24.25,26,27,29 and 30 CHs - groups; 28 = OH group
hederagenin : 28 = OH - group 24,25,26,27,29 CHs - groups; 30 = CH_2OH group
quillaic acid : 23 = CHO - group 24,25,26,27,29 and 30 CHs - groups to C- atom 16 is an OH group attached
Clycyrrhetic acid : To C 11 is an = O group attached; between C 12 and C 13 is double binding; 24,25,26,27,29 and 30 : CHs- groups.

The saponin from soapbark *(Quillaja saponaria)* was isolated in the same manner. Glycyrrhizic acid was extracted from licorice *(Radix liquiritiae)* with ethanol and precipitated with ether. Hederagenin was also extracted with ethanol, from soapnuts *(Sapindus rarak* from Indonesia).

The unpurified preparations were used for chromatography.

Discussion and Results

After some experimentation we found the following solvents suitable for the separation of beet and closely related sapogenins:

benzene- ethanol 90:10 (see Table 1 and Figure 2)

A mixture of hexane-ethylacetate was also found to be suitable for the separation of the sapogenins (see Table 2).

With the benzene-ethanol solvent the saponins remained practically at the starting point, while with the hexane-ethylacetate solvent a good separation was obtained (see Table 3).

A fairly good separation of saponins was also obtained, using one of the solvents in use in paper chromatography (butanol-acetic acid- water 4:1:1, see Table 4).

It appears that the major components of beet sapogenin fractions a and b are probably the same: they have about the same R_F value in two solvents. The major components of the other sapogenins are distinctly different from the major component of beet sapogenin.

It is of course usual in chromatographic research to compare the unknowns with the pure substances that are supposed to be in it. However, in this case we did not succeed in obtaining the pure substances for comparison of the components of the sapogenins tested. The sapogenins of the β-amyrin group (see Figure 2), oleanolic acid, hederagenin, quillaic acid and glycyrrhetic acids, are known to be the main components of the saponins of respectively sugar beet (3,4), sapindus varieties (2), soap bark and licorice.

From the chromatograms of the sapogenins it is clearly seen which are the major components. So it is highly probable that these substances are indeed the above mentioned sapogenins. This has to be confirmed of course by experiments with the pure substances.

Figure 2 gives the probable structual formula of the four sapogenins. The differences are relatively small. The difference is slightest between oleanolic acid and hederagenin, as is also the case on the chromatograms.

Table 1.—Chromatographic behavior of sugar btct sapogenins and sapogenins from other sources Solvent: benzene-ethanol 90.10.

Beet sapogenin fraction a			Beet sapogenin fraction b			Sapogenin from *Sapindus rarak*			Quillaic acid sapogenin			Glycyrrhetic acid sapogenin		
R_F x 100	Intensity of spot	Color	R_F x 100	Intensity of spot	Color	R_F x 100	Intensity of spot	Color	R_F x 100	Intensity of spot	Color	R_F x 100	Intensity of spot	Color
21.5	w	*S*				19	w	*S*	9	vw	bg	17	+	y
26	+	*rv*				37	+ + +	bp	18	vw	bg	21	+ +	y
47	+ + +	bp	48	+ + +	bp	61	+	b	27	vw	bg	44	+	b
64	w	b	64		b									
73.5	w	b	75	+	b	71	w	p	65	++	bg	65	vw	b
92	+	p	91.5	+	p	92	w	bp	85	w	bg	74	vw	b

Legend for table 1,2,3 and 4: b = blue, p - purple, g = green, y = yellow, r = red, w = weak, vw = very weak, 4- = moderate, ++ = strong, + + + = very strong.

Table 2.—Chromatographic behavior of sugar beet sapogenins and sapogenins from other sources. Solvent: hexane-ethylacetate 50:50.

Beet sapogenin fraction a			Beet sapogenin fraction b			Sapogenin from *Sapitidus rarak*			Quillaic acid sapogenin		
RF X 100	Intensity of spot	Color	RF X 100	Intensity of spot	Color	RF X 100	Intensity of spot	Color	RF X 100	Intensity of spot	Color
15	vw	p	18	+	b	8	+	bg	24	vw	b
22	+	1)							28	vw	b
34	+++	b	32	+ + +	b	32	+++	b	35	vw	b
60	+	b	60	+	b						
68	vw	bp	68	+	bp	68	+	b			
75	4-	b	76	+	b	77	+	bp	73	+++	b
91	++	p	89	+	p	92	+	b	82	+	b

Table 3.—Chromatographic behavior of sugar beet saponin and saponin from *Sapindus rarak.* Solvent: hexane-ethylacetate 50:50.

Beet saponin fraction a			Beet saponin fraction b			Saponin from *Sapindus rarak*		
Rf × 100	Intensity of spot	Color	Rf × 100	Intensity of spot	Color	Rf × 100	Intensity of spot	Color
22	w	b	18	vw	b	0	+++	bl
35	w	b	34	vw	b			
64	++	b	53	+	b			
81	w	b						
91	+++	b	89	+	b			

Table 4.—Chromatographic behavior of sugar beet saponin and saponin from *Sapindus rarak.* Solvent: butanol-acctic acid-water 4:1:1.

Beet saponit fraction a			Beet saponin fraction b			Saponin from *Sapindus rarak*		
Rf × 100	Intensity of spot	Color	Rf × 100	Intensity of spot	Color	Rf × 100	Intensity of spot	Color
61.5	w	p	61.5	w	p	28.5	+++	p
75	++	b	75[1]	+	b			
78.5	++	b	79[1]	++	b			
81	+	g						
90.5	++	p	93	w	p			

[1] The spots RF 75 and 79 were horizontally elongated and very clearly separated.

It appears from the chromatograms that there are probably at least six different sapogenins in sugar beet saponin, probably also closely related substances. Possibly quillaic acid is one of them.

There seems to be little difference between fraction a and b sapogenin, only in relative amounts.

It may be thought that the six sapogenin spots represent partial hydrolysis products of the sugar beet saponin.

As however oleanolic acid and related substances only contain one hydroxyl group to attach a carbohydrate moiety, it is extremely unlikely that one molecule of oleanolic acid binds more than one carbohydrate molecule. Van der Haar (3) indeed found from molecular weight determinations one molecule glucuronic acid in one molecule of beet saponine. So the presence of partial hydrolysis-products is unlikely.

The saponins, fraction a and b, too are much the same, except in relative amounts. The difference in solubility, therefore, has to be ascribed to the presence of complexes or absorption compounds.

It is remarkable that *Sapindus* saponin gives only one spot, but contains as many sapogenins as the beet saponin. Probably the carbohydrate moiety determines this property to a high degree.

It is possible that in beet saponins other carbohydrates occur apart from glucuronic acid. This has not yet been investigated by modern methods as far as we know. Even the presence of glucuronic acid is not without doubt, as this has been confirmed only by color reactions.

If other carbohydrates also occur in beet saponins the number of possible compounds is high.

Summary

With thin-layer chromatography it is possible to separate closely related sapogenins and saponins. From chromatographic evidence it seems likely that beet saponin contains at least six different sapogenins, four of which are as yet unknown. Fractions of saponins obtained by solubility differences contain the same sapogenins.

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A Survey of Sugar Beet Nematode in Beet Growing Areas of the Utah-Idaho Sugar Company

RONALD C. JOHNSON AND JOSEPH NEMAZI[1]

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The sugar beet nematode *(Heterodera schachtii)* has long been recognized as one of the most serious problems of the sugar beet industry. Visual symptoms are readily recognizable in the field but by this time the infestation is generally high enough that yields have been greatly reduced. Yields are also frequently reduced even after rotation with nonhost crops. Obviously, infestations are being recognized too late and ideas as to degree of infestation are often erroneous.

A survey as to distribution and degree of infestation can be an essential part of a system of control by crop rotation. Such a system can work satisfactorily if the fields are found in the early stage of infestation (3)[2]

In view of these facts, in 1957, the Utah-Idaho Sugar Company initiated an extensive survey program to determine the extent and severity of infestation of fields where beets had been or were to be grown. Laboratories were established in Utah, Idaho and Washington for this purpose.

Method of Collecting Soil Samples

Two methods of collecting soil samples were used for this survey, field sampling and tare sampling.

Field Sampling

Samples were taken from the field with a soil probe or tube to a depth of 4 to 8 inches with at least five tube samples per acre of land. If a previous crop showed a spot where nemotode was suspected, a separate sample was taken from this area. The samples were thoroughly mixed and approximately 500 grams of soil sent to the laboratory.

Tare Sampling

A sample of soil was taken from the tare dirt at the receiving stations by holding a sample catcher (Figure 1) about a foot below the Reinks screens as a load of beets was being delivered. The container was held in the center of the area that the cone of dirt forms, sometimes being necessary to shake it to remove vegetative matter. Frequently this procedure would have to be repeated for a second load from the same field to obtain 500 grams of soil for the laboratory analyses.

[1] Assistant Research Director and Research Agronomist, respectively, Utah-Idaho Sugar Company.

[2] Numbers in parentheses refer to literature cited.

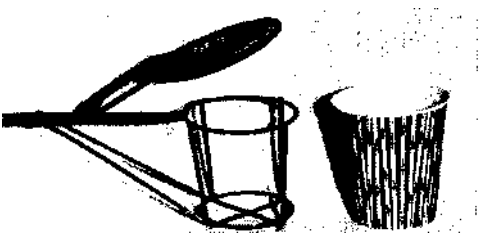

Figure 1.—A sample catcher which is used to take a sample of soil from the tare dirt. The catcher is held about a foot below the Reinks screens as a load of beets is being delivered.

The tare sampling method has proven to be cheaper and more convenient; however, the following precautions must be exercised in catching the sample by this method:

1. Obtain samples only during the last half of the load to eliminate the possibility of contaminating the sample from the previous load.
2. Obtain samples only during dry weather conditions as wet dirt sticks to belts and increases the possibility of contamination.
3. Observe the necessary caution in working around moving equipment. Holes have been cut in the sides of most of the dirt hoppers to allow the sample catcher to be placed under the Reinks screens. This eliminates much of the danger.

The soil samples taken by either method were carefully labeled as to date, grower, contract number, field location, and a note made if nematodes were suspected. The samples were sent to the laboratory where they were allowed to become dry or nearly dry before being analyzed.

Methods of Separating Cysts From Soil

The specific gravity of cysts in dry or nearly dry soil is less than water. The principle of separation used was to float the cysts to the surface and remove them by passing the sample through a series of screens.

Flotation: A 500-gram soil sample was thoroughly mixed with water in a 12-inch pan and allowed to settle momentarily. The water was then passed through a U.S. 10 series sieve to a second

Figure 2.—Equipment needed to separate the cysts from the soil.

pan. The residue collected on the screen was gently washed, and the sediment in the pan and the residue on the screen were discarded.

Sieving: The muddy water in the second pan was passed through U. S. series 30 and 60 screens. After rinsing the 30-mesh screen gently, the residue on the screen and the sediment in the pan were discarded. The residue on the 60-mesh screen is composed of nematode cysts along with some organic matter. This residue is washed into a 250 cc beaker by directing a small stream of water from a wash bottle on the back of the screen.

Filtering: The cysts and organic matter floating in the beaker were poured onto a marked filter paper in a Buchner funnel. Filtering was accomplished by using a suction pump or an aspirator connected by rubber tubing to a filtering flask.

The filter paper was placed on a plastic plate where it was examined and a cyst count made using a 15x binocular with wide-angle magnification.

Cyst Report: A simple cyst count of the total number of cysts in 500 grams of soil can be misleading in advocating a program to the grower. Hijner (2) has shown that an average of 40% of the cysts become nonviable each year whereas only 13% of the cysts decay. Therefore, a more accurate population estimate can be made by determining the number of viable cysts. In this survey, the total number of cysts was determined and, in addition, a representative number of cysts was ruptured and examined If any eggs or larvae were present in the cysts examined, they

were considered viable and the percent of viable cysts was also determined. As no standard meaning of viable cysts has been agreed upon by workers in this field, this method of determining viability is used to help in advisory work. The total number of cysts will give information as to how heavily infested the field is or has been, and the number of viable cysts along with crop history and soil type will give information as to the effectiveness of the rotation.

Classification of Infestation

An arbitraary scale of nemotode cyst counts has been established for making recommendations to the growers. It is difficult to closely associate a cyst count with the damage done by the nematodes. However, field trials conducted by the Utah-Idaho Sugar Company support the following classifications:

1. Noninfested—no cysts present.
2. Slightly-infested—1 to 10 viable cysts.
3. Moderately-infested—11 to 50 viable cysts.
5. Heavily-infested—more than 50 viable cysts.

Results

In the four years from 1958 to 1961 inclusive, 4,375 samples were collected and analyzed. The results of these tests are shown in Tables 1 through 4. Tables 1 and 2 show the data for each year separately and Table 4 shows a summary of the results for the four years. Table 3 shows a relationship between total and viable cyst counts as determined by samples taken in Utah.

The degree of infestation may seem to have changed from year to year, however, this may only be a reflection of the information desired by the fieldman. Some years samples may have been collected at random and without regard to known or suspected infestation. Other years many of the samples may have been collected to ascertain the infestation of a suspected field. This would result in a higher percentage having nematodes. Other years many of the samples may have been collected to determine the degree of infestation in fields known to have nematode, which would result in an even higher percentage of infestation. Some years the fieldman may have selected many of the samples to determine the possible spread of nematode into areas thought to be free of infestation. This would result in a lower percentage of infestation than any of the others.

These data show that some areas are much more heavily infested than are others. The tables only show the difference between districts but similar differences also occur between areas within districts.

growing beets this does not necessarily mean that 40 percent of the beet land is infested with nematode. However, it clearly indicates that a fairly high percentage of the land is infested and that a strong program will have to be pursued to keep the nematode under control.

For all of the samples, the fieldmen indicated whether or not a nematode infestation was suspected. In practically all cases where nematode was suspected, the infestation was high and in a large percentage of fields where it was not suspected the infestation varied from low to high. These data indicate that nonsuspected fields are frequently infested and that infestation can be determined long before there are visual symptoms in the field. It also indicated that when visual symptoms appear, the cyst count is always above 50 but that the cyst count may be above 50 without visual symptoms. This is generally because the damage is masked by good growing conditions.

Discussion

Recommendations based on the cyst count.

1. No viable cysts—Permissible for sugar beet production, however it is suggested that beets be planted only two years out of six. Beets probably can be produced indefinitely on this program providing the rotation does not include host crops other than beets.
2. 1-10 viable cysts—One crop of beets can be grown after which the field should be put into at least a four-year rotation with nonhost crops.
3. 11-50 viable cysts—Land should be rotated for four years with nonhost crops before beets are planted unless the soil is fumigated. A four-year rotation will probably reduce populations enough to permit one crop of beets; however, if fumigation is practiced the soil should be tested again before planting beets.
4. Above 50 viable cysts—Land should definitely go into a minimum of a four-year rotation of nonhost crops or be fumigated before planting beets. Soil should be tested again before planting beets without fumigation.

Many fumigation trials have been conducted with the above cyst counts as the measure of infestation. The response to fumigation supports these recommendations.

The following additional practices are recommended for all beet-growing areas. These practices have resulted from known and proven facts frequently reported.

1. Have a good crop rotation for each field.
2. Keep field free of weeds.
3. Have soil tested for nematode infestation and plant beets only if cyst count is favorable.
4. Plant early—most of the nematode egg hatching and thus, nematode damage is done when the soil temperature is 60 degrees F or higher (1). Sugar beets grow fairly well in temperatures less than 60 degrees. If sufficient early growth is made the beets will be able to produce fairly good yields despite the presence of nematodes.
5. Apply adequate fertilizer and water. The earliest symptom of nematodes is the appearance of drought areas in the field. Adequate fertilizer and water help the beet make rapid growth and thereby help it resist and outgrow the damage caused by the nematode.
6. Never return tare dirt to the farm. There is always the possibility that infested beets have gone over the receiving station and that the tare dirt is contaminated. The Utah-Idaho Sugar Company has made areas available for the tare dirt so that it is not returned to the farm.
7. Fumigate to get one more beet crop in the rotation. This will further increase the nematode population and though satisfactory yields can generally be obtained it is recommended only as an emergency program. Continuous fumigation is discouraged as other pests and diseases will become troublesome.
8. Adopt strict regulations concerning moving equipment and machinery from an infested area to a noninfested area. Frequently nematodes are introduced into new areas on machinery of a grower using the same equipment in infested and noninfested areas.

The above programs can control nematode infestations so as to minimize the damage and permit profitable production of sugar beets.

Summary

During the years from 1958 to 1961 inclusive, 4,375 soil samples were collected and analyzed for sugar beet nematode. Some districts had a much higher infestation than did others.

This varied from a low of only one percent in the relatively new Columbia Basin area to a high of 79 percent in the West Jordan District.

The data indicate that a soil sampling program will ascertain the degree of infestation and a control program can then be effected long before there are visual symptoms in the field. It further shows that in many infested fields neither the grower nor the fieldman were aware of the presence of nematode.

Arbitrary standards of infestations were established and the following control program suggested depending upon the degree of infestation.

No viable cysts—Permissible to grow beets. Suggest planting beets only two years of six.

11-50 viable cysts—Have a four-year crop rotation of nonhost crops or fumigate before planting beets. Test soil again before planting beets if soil is fumigated.

Above 50 viable cysts—Have rotation of nonhost crops for a minimum of four years—preferably longer. Field can be fumigated for an extra crop of beets, however, this will further increase nematode population and make a longer rotation necessary. Test soil again before growing beets without fumigation.

A program of testing the soil and then initiating a control program depending upon the degree of infestation will minimize the damage from the sugar beet nematode.

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The Major Considerations in the Problem of Package Weight Control

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Received for publication February 14, 1962

Since the passing of the 1958 Amendment to the Federal Food, Drug, and Cosmetic Act there has been increased activity in all areas coming under the jurisdiction of the Food and Drug Administration. One of these areas is concerned with the proper labeling of food packages which includes the specific question as to whether or not the package contains the weight or measure as shown on the label. This is of particular interest to the beet sugar industry. It should be, for almost 27 million bags (cwt) of beet sugar were sold in packages in 1960, which was 67 percent of the total beet sugar sales (l)[2].

The increased activity in this field by the FDA has been paralleled by the regulatory agencies of most states and some of the larger municipalities.

All this is not the result of pressure from a suspicious public, but rather an honest attempt on the part of those charged with the protection of the consumer to meet the increasing complexities of the job. As reputable manufacturers, we must welcome this emphasis on proper package weight control as an opportunity to prove to the consumer that he is getting full value when he purchases our products.

Regulating Agencies

The role of the FDA in connection with package weights has already been mentioned. It is of interest to examine the authority of this agency which has jurisdiction over weights of food packages moving in interstate commerce.

The Federal Food, Drug, and Cosmetic Act lists two general categories of acts which are prohibited; namely, adulteration and misbranding. Discrepancies between actual and labeled weight come under the latter category, and the enforcement of the Act is carried out by the FDA.

The Federal Trade Commission also concerns itself with the subject of misbranding, but through agreement, exercises jurisdiction over advertising, leaving the field of labeling to be covered by the FDA (2, 3).

All states have laws governing the proper labeling of com modities in commerce within its borders. In some cases, these are supplemented by ordinances of large municipalities. The department responsible for administering these laws varies among the

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[2]numbers in parentheses refer to literature cited.

states, although in most cases the Department of Agriculture is assigned the responsibility.

In the opinion of those responsible for carrying out the laws, the problem of packages that are short of the declared weight receives too little attention. This is apparently due to a lack of funds in most cases. To improve this situation, it has been suggested by Mr. George P. Larrick, Commissioner of Food and Drugs, U. S. Department of Health, Education and Welfare, that the FDA commission State officials already engaged in weights and measures handle enforcement work (4). This would be a cooperative effort to gain greater coverage of the problem. In making this suggestion Commissioner Larrick stated, "We would like to see a concerted nationwide effort by the State officials and the Food and Drug Administration to stamp out the shipment of short-weight merchandise."

As an industry involved in selling packaged food items, we can certainly expect to have our products checked more frequently in the future. The results of such checks and our reaction to these results may have considerable influence on consumer confidence in our products.

The Governing Laws

As previously mentioned, the Federal Food, Drug, and Cosmetic Act covers the subject of package weights under the heading of misbranding. The Act clearly specifies that:

a. The label will state the minimum quantity or the average quantity contained, that the term "minimum" must be stated or the label amount shall be considered to express an average quantity.

b. Where the average weight is expressed, which applies to our own case, variations from the stated weight are permitted provided that the variations are unavoidable, and remain within the limits of good packaging practice. Variations will not be permitted, however, to such extent that the average of the quantities of the packages comprising a shipment is below the quantity stated, and no unreasonable shortage in any package shall be permitted even though overages in other packages in the same shipment compensate for such shortage.

The important point is that the FDA allows packages to be filled to an average weight and recognizes the necessity for allowing reasonable variations in the weight of packages. To some this may appear as a loophole, but to properly comply in meeting the average weight without unreasonable shortage closes the door on such a possibility.

The laws of the individual states covering package weights vary widely in detail as would be expected. It is significant to note, however, that at least 47 states, the District of Columbia, and Puerto Rico recognize reasonable variation (5). In this detail, then, there is almost unanimous agreement between the State and Federal regulations.

The 46th National Conference on Weights and Measures (1961) approved a model state law covering labeling, advertising and packaging (6). The conference, composed of representatives from Federal and State agencies as well as trade and industry, took an important step forward in endorsing such uniformity. This model law provides for the declaration of an average net weight and recognizes reasonable variations from the labeled weight. In these details, the model law accurately parallels the existing Federal regulations.

Economic Compliance

Producing packages of proper net weight is a quality control problem, for this is as important a specification to the consumer of our products as are the other quality factors such as color or sediment. Controlling package weights should, therefore, be a function of those normally responsible for quality control. Accepting this principle provides the use of a ready-made technical organization in the company and plant to apply modern techniques for efficient control.

This leads to the crux of the situation—what are the most efficient and economic control procedures available to meet the problem?

Remembering that the legal requirement in packaging is to have the average w^eight of each shipment equal to or in excess of the label weight with no unreasonable shortage in any package, it is obvious that the target weight at the packaging station must exceed the label weight to some degree in order to be safe. Depending on the degree of security desired, it is also expensive.

Using the concepts of Statistical Quality Control (SQC) the degree of safety can be evaluated against the product giveaway and both controlled within the most acceptable limits according to the judgement of management. SQC is the application of the mathematics of probability to the numerical results of any process, operation, experiment, etc., for the purpose of expressing the true meaning of the results. These techniques are widely used today wherever processing or manufacturing results must be controlled to specifications.

1 he scope of SQC is tremendous and the subject is well covered from fundamentals to applications in texts and periodicals.

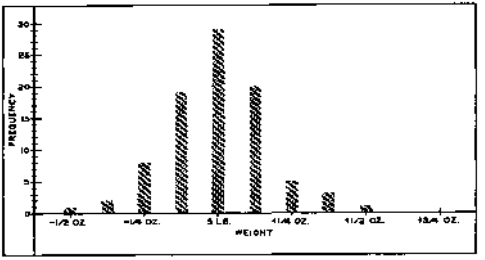

Figure 1.—Weight distribution histogram.

It is not the purpose here to describe detailed techniques of SQC applicable to package weights. However, for those unfamiliar with the subject a brief description of the principles relative to our problem follows:

Suppose a shipment of packages produced at a target weight of 5 pounds net is randomly sampled and checkweighed to the nearest 1/8ounce. A plot of the individual weights obtained for frequency will present a weight distribution histogram such as shown in Figure 1. This describes mathematically the normal distribution curve covering the variations in weights in the shipment shown in Figure 2. Note that weights center about the target weight but that half of the packages will be less than label weight. Obviously a safety factor should have been included to provide assurance that the shipment would comply with the law.

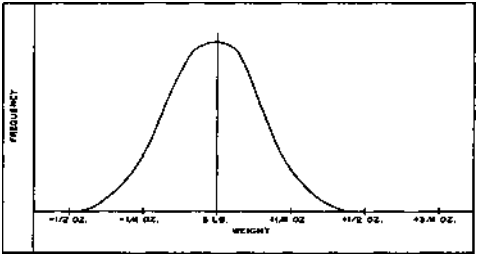

Figure 2.—Normal distribution curve.

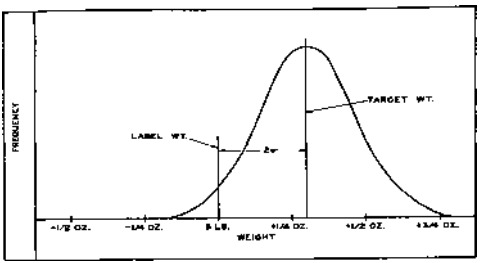

Figure 3.—Assuring compliance.

The standard deviation (designated by the Greek letter sigma) of the sample weights provides a useful guide in adjusting the target weight to gain the required assurance. If 95% assurance is desired that all packages will be at least label weight, then the target weight will be set 2 standard deviations above the label weight. This is depicted in Figure 3. If 99% assurance is desired, the target weight is set 3 standard deviations above the label weight. From these figures it can be seen that SQC provides the necessary assurance that the average net weight of the lot or shipment will be not less than the label weight. It is also obvious that the amount of giveaway product necessary for such insurance will be less by this control than if all scales are kept adjusted to allow nothing less than label weight.

These statistical techniques serve another important purpose in establishing the magnitude of package weight variation. Excess variation requires excess giveaway product to assure the proper net weight average, and increases the danger of shortages at the unreasonable level. When such is indicated, the cause must be located and eliminated or minimized. This may require improvements in weighing equipment, maintenance and, or operation. Figure 4 demonstrates an improved condition.

These statistical concepts along with others have been converted to the tools of SQC. The methods of sampling, recording, calculating, and evaluating have been simplified to the point that the ordinary station operator or foreman can be trained to carry out the entire analysis and take action according to the results. The use of statistical methods will not only insure the most economic compliance with the law, but the resulting records should provide good evidence of intended conformance.

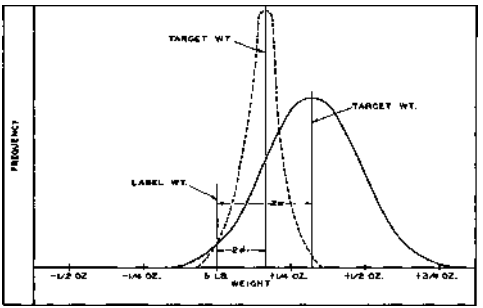

Figure 4.—Compliance at reduced giveaway.

New Trends

Equipment manufacturers now offer a variety of machines designed to aid in the task of package weight control. Among the features offered are 100% checkweighing, automatic rejecting of packages not meeting specifications, and a continuous and permanent record of results. The economic advantages of such equipment lies in the automation of the process and the narrowing of the net weight variation allowing a minimum amount of giveaway product. The latter feature, again, is accomplished through the use of statistical procedures and controls.

Viewing the entire picture of package weight control, it may be said that by proper understanding of the regulations and their enforcement along with an effective weight control program, the increased scrutiny of the enforcement agencies can be successfully and efficiently met. This view is slightly marred by a recent incident relevant to this subject (7).

Early in 1961, the State of California adopted a uniform procedure for its inspectors to follow in checking package weights. This code is based on statistical methods and is designed to cover the average net weight and unreasonable shortage features of the law. At a hearing prior to the adoption of the code, a representative of the California Office of Consumer Counsel objected to the adoption of the code on the basis that it allows reasonable tolerances below the labeled weight. It would seem that in this case the objection should be to the regulation, not to the method of enforcement.

The point to be made on this incident is that here we have a consumer representative opposing the average net weight and reasonable variation concepts and supporting the minimum net weight concept for all food packages. This could be accomplished only by a change in the present Federal and State regulations. However, this offers little consolation in view of the fact that the Federal Government is considering establishing a Department of Consumers (8) which might well support the same view.

A minimum net weight requirement for packages would require excesses of product to be included in packages well above that normally required under the present law for compliance. The beet sugar industry can ill afford to give away a greater amount of sugar.

It is imperative, then, that this industry as well as all food industries whose products are sold in packages not only comply with the existing package weight regulations but also support and defend them against changes and interpretations which do not consider the importance of reasonable variation.

Summary

The increased activities of the FDA in recent years have increased attention to package weights. The regulating agencies involved are the FDA and its counterpart in the states and larger cities.

The Federal Food, Drug, and Cosmetic Act covers the subject of package weights under the heading of misbranding. The Act specifically allows variations from the labeled weight provided that these variations are reasonable and unavoidable, and that the average weight of a lot or shipment is not less than the labeled weight. This concept is supported by the laws of practically all states and the 46th National Conference on Weights and Measures.

Economic compliance with the regulations can best be achieved by Statistical Quality Control. The techniques provide for establishing the safe limits of package weights for the minimum amount of giveaway product. New automatic equipment is now available to assist in reducing labor and product loss.

The possibility exists that consumer representative groups may oppose the accepted average net weight concept in favor of a regulation based on the minimum net weight concept. Such a change would require an increased amount of excess product in the package to assure compliance. It behooves the beet sugar industry to comply and support the present regulations.

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Minutes of the Twelfth General Meeting of the American Society of Sugar Beet Technologists

The business meeting of the Twelfth General Meeting of the American Society of Sugar Beet Technologists was called to order by Mr. Dewey Stewart, President of the Society, at 11:00 a.m. on Wednesday, February 7, 1962, in the Silver Glade Room of the Cosmopolitan Hotel, Denver, Colorado.

Mr. Stewart, serving as chairman of the meeting, announced that the Twelfth Biennial Business Meeting of the Society was called for the purpose of hearing committee reports and to transact such business as may be appropriate.

The chairman called for the reading of the minutes of the Eleventh General Meeting Business Session held in Salt Lake City, Utah, February 4, 1960. Upon motion made, seconded and unanimously carried, reading of the minutes of such meeting was dispensed with.

The chairman then asked for the Report of the Secretary. The Secretary briefly reported on highlights of the Society's activities during the period since the last biennial meeting. Upon motion made, seconded and unanimously carried, the Report of the Secretary was accepted, ordered placed on file and made a part of the minutes. Highlights of the report are herewith included:

Report of the Secretary

Membership in the Society at the close of the 1960-61 biennium was 633 individuals and companies. Members reside in 34 states of the United States and the District of Columbia and 20 foreign countries. Membership for the biennium ending December 31, 1961, showed an increase over the previous biennium by 55 members. A list of states and countries showing the number residing in each is made a part of this report as follows:

Membership by States and Countries—1960-1961

Washington, D. C.	3
Arizona	8
California	141
Colorado	109
Connecticut	1
Delaware	4
Florida	1
Georgia	2
Hawaii	1
Idaho	36
Illinois	14
Iowa	5
Kansas	2
Louisiana	1
Maryland	5
Massachusetts	2
Michigan	28
Minnesota	11
Missouri	2
Montana	20
Nebraska	14
New Jersey	1
New Mexico	1
New York	12
North Carolina	1
Ohio	14
Oklahoma	1
Oregon	6
Pennsylvania	3
South Dakota	6
Texas	9
Utah	60
Washington	12
Wisconsin	7
Wyoming	13

FOREIGN COUNTRIES

Canada	38
England	5
Sweden	3
Denmark	7
Belgium	1
Ireland	2
Spain	2
Netherlands	3
Japan	3
Iran	2
Chile	1
Syria	1
Israel	1
Iraq	2
West Pakistan	1
Uruguay	1
Australia	1
Nigeria	1
Italy	2
Germany	3
TOTAL	626
Company Members	7
	633

Approximately 925 copies of the *Journal of the American Society of Sugar Beet Technologists* are mailed each printing to some 40 states and to more than 50 foreign countries. During the biennium of this report, restrictions with respect to mailing of Journals to "Iron Curtain Countries" were removed. This in part accounts for the increase in foreign subscriptions, some of which are still in process of arranging for payment and currency exchange through reliable agents.

The Secretary wishes to again express the Society's thanks to the Beet Sugar Development Foundation for providing office space and staff, without cost to the Society. In addition thereto, the Foundation has provided an annual grant of $1,000 to the Society to help defray the cost of publishing the Journal.

The chairman then requested that the Report of the Treasurer be read. The Treasurer briefly reviewed the balance sheet showing receipts and disbursements for the biennium January 1, 1960 through December 31, 1961. Upon motion made, seconded and unanimously carried, the Report of the Treasurer was accepted, ordered placed on file and made a part of these minutes.

Report of the Treasurer

Balance Sheet
December 31, 1961

Cash Balance, January 1, 1960	$ 3,681.87
Savings Account Balance, January 1, 1960	5,359.10
1960 Interest Earned on Savings Account	230.18
1960 Cash Receipts	12,372.74
1961 Interest Earned on Savings Account	240.06
1961 Cash Receipts	4,135.42
	$26,019.37
Less: Transfer from Savings to Checking Account	— 500.00
Interest on Short-Term Loan	— 1.25
	$25,518.12
1960 Cash Disbursements	$12,874.01
1961 Cash Disbursements	6,420.81
Savings Account Balance, December 31, 1961	5,328.09
Cash Balance, December 31, 1961	895.21
	$25,518.12

The Chairman then called for a report from Mr. Bion Tolman on the First Joint IIRB-ASSBT meeting. Mr. Tolman reported that the First Joint Meeting of the Institut Inernational De Recherches Betteravieres - American Society of Sugar Beet Technologists was held at the Conference Rooms of The International Sugar Council in London, England, on Friday, May 19, 1961. This meeting followed demonstrations and tours.

Representatives present from the North American continent included: Bion Tolman, F. H. Peto, Ralph E. Finkner, B. E. Faston, and C. W. Doxtator. Our Society President, Dewey Stewart, planned to attend but was forced to remain at home because of untimely illness.

The North American delegates to the joint meeting express their praise of the plans, organization and hospitality by their many European friends. They further express regrets over the fact that so few from this continent were able to attend.

It was then reported by the Chairman that a Tally Committee had been appointed to determine the elected officers for the biennium 1962-63. Although the results of the tally were not made known until the banquet the evening of the day of this business meeting, the results are herewith recorded:

Meritorious Service
Award Presented
to

HARVEY P. H. JOHNSON

Harvey P. H. Johnson was born in Hawley, Minnesota, where he spent his boyhood days on his father's farm. He attended Northwest School of Agriculture at Crookston, Minnesota, and Concordia College at Moorhead, Minnesota, from which he graduated with a major in economics. Following graduation, he taught in high school at Stanton, North Dakota, and became an instructor in the Moorhead, Minnesota Public Schools. While at Moorhead, he attended night classes at North Dakota State University at Fargo, North Dakota, and later attended the University where in 1939 he earned a master of science degree in agricultural economics. For a year and a half he was employed with the Federal Land Bank at the National Farm Loan Association office in Long Prairie, Minnesota. He had the dubious honor of being the first person drafted into the armed service from Todd County and spent five years in the service from which he was separated with the rank of Captain. In 1946, he was employed by the Beet Sugar Development Foundation at Fort Collins, Colorado, as statistician-agronomist. In 1950, he was appointed manager of the Foundation and later that year moved to Denver to accept the position of assistant general agriculturist with the American Crystal Sugar Company. In 1953, he became general agriculturist and was elected vice president of his company in 1960. Mr. Johnson has been a member of the Society since 1946, was its secretary-treasurer in 1948-49, and president in 1958-59. He has twice served on the constitution and bylaws committee, the nominating committee and the advisory council. He has served on the resolutions committee, awards committee, local arrangements committee and served as general program chairman for the Society's Eighth General Meeting.

VOL. 1-2, No. 1, APRIL 1962

Meritorious Service
Award Presented
to
BION TOLMAN

Bion Tolman was born at Murtaugh, Idaho, in 1907. He graduated from Twin Falls, Idaho High School in 1925 and attended Utah State University from 1925 to 1927. Two years were spent on a mission for his church, after which, he returned to Utah State University, graduating in 1932 with a B.S. degree in agriculture and earning a master's degree in plant breeding in 1933. He started with the Utah State Department of Agriculture in plant quarantine enforcement and later joined the Utah State University Extension Service as county agricultural agent. The next year he earned an appointment with the Division of Sugar Plant Investigations of the USDA where he worked for the next nine years. His work under Dr. F. V. Owen at the Salt Lake Station included sugar beet improvement studies and agronomic experiments on sugar beet seed production in southern Utah and the Willamette Valley of Oregon. In 1945, he was employed by the Utah-Idaho Sugar Company and was charged with the responsibilities of setting up and directing an agricultural research program. In 1948, he was promoted to general agricultural superintendent and director of agricultural research. In 1959, he became vice president in charge of agricultural operations including the agricultural research department.

Mr. Tolman has been a member of the American Society of Sugar Beet Technologists since its beginning, was elected vice president for two bienniums, has been elected to the advisory council for four bienniums, served two terms on the committee on standardization of experimental methods, and was appointed general program chairman for the Sixth General Meeting.

Meritorious Service
Award Presented

HEWITT M. TYSDAL

Dr. H. M. Tysdal experienced his first contacts with sugar beets while enjoying the honor of being a Spragg Memorial Lecturer in 1942 at Michigan State University. The alfalfa research in which he was engaged expanded to western Nebraska where he became interested in the use of alfalfa with sugar beets in rotation. In 1948, he became directly associated with sugar beet research as head of the U. S. Agricultural Research Station at Salinas, California. In 1954, he became chief of the Sugar Plant Section, USDA, whereupon, it became his responsibility to develop an effective working relationship with industry and growers. His primary objective as head of the USDA program was to hold and recruit the very best men for sugar beet research. Dr. Tysdal continues to be proud of the dedicated group of outstanding research people who worked under his direction while he was chief of the branch. While Dr. Tysdal's personal fame came from research in alfalfa, he does not hesitate to acknowledge the outstanding research of the sugar beet workers of the U. S. Department of Agriculture. It was quite rewarding to Dr. Tysdal to see the completion of a fine new research laboratory and greenhouses at Utah State University which he envisioned as a stimulation to sugar beet research and an aid to the solution of the multitude of problems affecting improved production. He retired from Government service in 1961.

Dr. Tysdal became a member of the American Society of Sugar Beet Technologists in 1954. During his membership he was elected to the advisory council for two terms. He has been pleased with the prominence that each member of his staff has gained in the Society and has been foremost in projecting the objectives of the Society.

Forty Year Veteran Awards

ELJGKNK C. ANDKRSON, Franklin County Division ol The Amalgamated Sugar Company

JOSKPH H. BINGGHAM, The Amalgamated Sugar Company

ALBERT L,. COOPKR, Holly Sugar Corporation

IRA D. CROGIIAN, Holly Sugar Corporation

W. S. HALLAM, Holly Sugar Corporation

E. CLARK JONKS, Utah-Idaho Sugar Company

ARTHUR C. JOOST, Northern Ohio Sugar Company

CHARLES A. LAVIS, Holly Sugar Corporation

Louis F. OSWALD, Utah-Idaho Sugar Company

CHARLKS PRICE, U. S. Department of Agriculture

W. H. PUNCHARD, Canada & Dominion Sugar Company, Limited

J. F. RASMUSSKN, The National Sugar Manufacturing Company

JAMES W. SILVER, Ogden Iron Works Company

E. L. TEWKS, Holly Sugar Corporation

GERALD THORNE, University of Wisconsin

J. E. TRINNAMAN, Utah-Idaho Sugar Company

W. D. WARNER, Utah-Idaho Sugar Company

In Memoriam

J. D. ALGIER, Hardin, Montana
QUINDARO S. BALE, Solana Beach, California
HENRY W. DAHLBERG, Denver, Colorado
J. DEDEK, Tirlemont, Belgium
EDWARD ELIOX, Washington, D. C.
H. J. KLIXGE, Preston, Idaho
E. C. KUNDTNGER, Sebewaing, Michigan
T. H. LACY, Santa Ana, California
WILLIAM J. MCGREGOR, Chatham, Ontario, Canada
CHASE OFTEDAL, Missoula, Montana
E. G. UTLEY, Spreckels, California
H. E. ZITKOWSKI, Denver, Colorado

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Control of Cercospora Leaf Spot of Sugar Beets With Protective Fungicides

F. R. FORSYTH AND C. E. BROADWELL[1]

Received for publication January ly, 1962

Cercospora leaf spot is one of the major problems of beet cultivation in central and southern Europe, and at times, in certain parts of North America. The disease is caused by the fungus *Cercospora beticola* Sacc. and is spread by airborne spores produced on the leaf lesions. The fungus thrives at high temperatures, but does not become epidemic unless temperatures (20°C or more) are combined with high humidity. The spots are isolated at first but in severe attacks, they coalesce and the leaf shrivels. Consequently, badly attacked plants are surrounded by a ring of dead, brown leaves, still attached to the crown but lying on the ground. The destruction of the foliage reduces yield and produces a lower sugar content.

In 1959, and again in 1961, the incidence of leaf spotting on sugar beet leaves reached amounts considerably above normal in southwestern Ontario and led to renewed interest in the use of fungicides to control this disease. Spraying crops periodically with 4-4-50 Bordeaux mixture has been resorted to in some countries to avoid epidemics. However, in Europe, the trend is now toward the use of the dithiocarbamate and organo-tin fungicides for the control of leaf spotting diseases of sugar beets. Consequently, in the tests reported here, examples of the above-mentioned chemicals were used.

The plots were located at the Pesticide Research Institute, London, Ontario. The sugar beet seed was of the monogerm type supplied by the Canada and Dominion Sugar Company Limited, Chatham, Ontario. Each plot consisted of 4-rod rows of sugar beets. These rows were separated from the next plot by alleys 3-feet wide. Four replicate plots were used for the treatments which occurred once, randomized, in each of four blocks of plots.

The chemicals[2] listed in Table 1 were sprayed on the plants in an equivalent of 66 gallons of water per acre but applied as

[1] Senior Plant Pathologist, Pesticide Research Institute, London, Ontario, and Research Supervisor, Canada and Dominion Sugar Company Limited, Chatham, Ontario, respectively.

[2] Generic, chemical, trade name and source (in parentheses) of the fungicides used in this paper: maneb-manganese ethylene bisdithiocarbamate-Dithane M-22 (80%), zinc-b-zinc ethylene bisdithiocarbamate-Dithane Z-78 (65%), nabam-disodium ethylene bisdithiocarbamate-Dithane A-40 (100%), 9/4527-composition not reported, (Rohm and Haas Co.); Bordeaux 12.5% copper-Bordo powder (Niagara Brand Chemicals); dodine-n-dodecylguanidine acetate-Cyprex 65W (American Cyanamid Co.); triphenyltinacetate (Pesticide Research Institute, London, Canada).

500 ml per 4-rod rows. The spreader-sticker Triton X-114 was used with all fungicides except triphenyltinacetate. Triton X-100 was used with the latter. Seven weekly applications were made of all fungicides except triphenyltinacetate which was applied six times. Applications began on July 26 although the first trace of Cercospora spotting was noted on June 21, at which time the beets were in the 5- to 7-leaf stage. A one gallon knapsack sprayer was operated at 40 psi in applying the fungicides.

The area used for sugar beets in this study had been used for the same purpose the previous year. Spotting of leaves began in one corner of the field and the disease spread slowly to the entire plot area by fall. Whenever the Cercospora leaf spots began to appear in 1961 (approximately June 21) they were somewhat more numerous at first on leaves in the originally contaminated corner of the field than elsewhere. Within three weeks after the first spots were noted the beets of the entire area were showing traces of leaf spotting. It is assumed that inoculum from the 1960 plants infested the entire area giving rise to a uniform source of inoculum for the 1961 season.

Table 1.- The effect on disease rating and sucrose content of spraying sugar beet plants with fungicides

Fungicide	Rate in grams in 500 ml per four rod rows	Klein wanzlebener[1] *Cercospora* Chart	Avg. % Sucrose
Ma neb	T3(7	2.5	15.4**
Zineb	1.50	3.0	14.9**
Bordeaux	3.75	3.0	14.9**
Dodine	1.50	5.0	13.5
Nabam plus zinc sulphate	0.75	3.0	14.7**
Triphenyltinacetate	0.50	1.5	16.0**
0/4527	1.50	2.5	15.6**
Untreated		5-0	14.2
			L.S.D. 1% 0.44

[1] Kleinwanzlebener Chart Reading 1.5 = some plants with spots on outer leaves only—somt on both inner and outer leaves; 2.5 = spots on inner leaves—some spots joining together; 3.0 = spots joining to form large areas of dead leaf; 5.0 = outer leaves dead, inner leaves severely damaged, fresh foliage begins to grow.

Table 1 reports the fungicides used, their source and rate of application, estimation of the amount of leaf spotting and the percentage of sucrose present in the roots. The date of applications for all the fungicides except triphenyltinacetate were July 26, August 3, 10, 16, 23, 30 and September 6. In the case of the latter chemical the August 10 application was missed. The estimation of leaf spotting was made on October 6 using a Klein wanzlebener *Cercospora* chart (2)[3]. By that time, the outer leaves

[3] Numbers in parentheses refer to literature cited.

of the untreated plants had been killed by the *Cercospora* and new inner leaves were being produced. This regrowth tends to mask the observable damage but the effect of the early loss of the outer leaves is noted in reduced sucrose levels.

The data for sugar percentage were obtained for each treatment from eight 5-beet samples collected at random, two from each plot, and analysed in the laboratory of the Canada and Dominion Sugar Company, Chatham. The reduction in average sucrose percentage caused by the disease is evident in Table 1. Whereas the percentage sucrose was 14.2 in unsprayed plots, it was as high as 16.0 in treated ones. Under the conditions of this experiment, all treatments except dodine gave increased levels of sucrose significant at the 1% level when compared with the levels of untreated samples.

The results presented here are merely indications of which fungicides might be used economically to control this disease. In Germany and Italy, (1, 3, 4) sprays of copper, organo-tin or dithiocarbaniate fungicides are applied from nvo to four times at two-week intervals depending on the date of beginning of the natural infection. Economical control under North American conditions will probably be possible only if the number of applications can be kept as low as those in Europe. More testing is required to determine the best fungicide for the control of this disease.

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Experimental and Commercial Results With Tillam[1] for Weed Control in Sugar Beets

J. ANTOGMNI[2, 3]

Received for publication February 8, 1962

Introduction

Tillam (propyl ethyl-n-butylthiolcarbamate), a soil - incorporated selective herbicide closely related to Eptam[1] (ethyl di-n-propyl-thiolcarbamate), has been used extensively in experimental and commercial applications in California during the past two years. Eptam was originally tested for weed control in beets but under certain commercial practices it was found that the margin between weed control and beet injury was too narrow. The findings with Eptam led to a program designed to find a compound similar in activity but with a wider safety margin. This program resulted in R-2061 (Code number used for Tillam) being tested as a preplant soil-incorporated herbicide in small-scale field trials at various locations in the United States in 1959. Results from these trials were sufficiently encouraging to warrant an expanded program in 1960.

1960 California Program and Results

An extensive field testing program was undertaken in California with the majority of the trials applied in the Sacramento Valley. Trials were, however, applied in all other beet-growing areas of the state. The trials were located so that various soil types, moisture situations and irrigation practices were involved. At most locations Eptam and Tillam were each applied at 2, 4, and 8 lbs per 50 gallons of water per acre. These rates were on an over-all solid coverage basis and with band treatments, the rate per crop acre was reduced according to the width of the band treated and the row spacing. All applications, spraying and incorporation, were accomplished with commercial equipment. In all trials, each treatment was a minimum of one-half acre in size. Incorporation of the chemicals into the soil was accomplished within minutes of application using various types of equipment. Solid coverage treatments on flat ground were incorporated by discing or by spike-tooth harrowing. Where band applications were made on both flat and bed planted beets, incorporation was

[1] Tillam is Stauffer's trademark for an herbicide.

[2] Research Representative. Stauffer Chemical Company, Mt. View, California.

[3] The author wishes to express his thanks to personnel of Holly Sugar Corporation. Union Sugar Division of Consolidated Foods Corporation. Spreckels Sugar Company and American Crvstal Sugar Company who cooperated in trials. Special thanks are extended to Mr. W. Reed of American Crvstal Sugar Company at Clarksburg. California, who obtained all of the yield data in addition to aiding with application of a number of experiments.

[4] Eptam is Stauffer's registered trademark for an herbicide.

accomplished with Bye-Hoes, Cultros, Chattins or ground-driven rotary hoes.

Results showed that beets were significantly more tolerant to Tillam than to Eptam and that good weed control could be obtained with 4 lbs per acre of Tillam under a wide variety of conditions, providing thorough soil incorporation immediately followed application. Data on weed control, stand of beets and growth of beets were obtained early in the season (just prior to thinning) and additional data were obtained throughout the season which included yield data and weed control at harvest time. Data on early weed control, beet stand and beet growth from two trials are presented in Tables 1 and 2.

Table 1.—Weed control and beet growth with Tillam and Eptam five application and seeding.[1]

Chemical	Lbs/A	Beet Stand2	Visual Rating of Stand	Growth[3]	% Control	
					Water grass	Red-root Pigweed
Tillam	2	28	Excellent	10	80	69
Tillam	4	26	Excellent	10	100	90
Tillam	8	24	Excellent	7	100	100
Eptam	2	28	Excellent	9	95	80
Eptam	4	17	Good	5	100	95
Eptam	8	14	Poor	2	100	100
Check	—	25	Excellent	10		

[1] The soil type was clay loam and incorporation was done with a Bye-Hoe.
[2] Stand per 3 ft of row (avg of eight random counts).
[3] Growth rated on a scale of 0 to 10 with 10 being normal compared to the check and 0 being death.

Table 2.—Weed control and beet growth with Tillam and Eptam 4 weeks after application and seeding.[1]

Chemical	Lbs/A	Beet Stands	Visual Rating of Stand	Growth[3]	Control of	
					Water grass	Lambsquarters
Tillam	2	25	Excellent	10	Good	Fair
Tillam	4	26	Excellent	10	Excellent	Excellent
1 illam	8	23	Excellent	6	Excellent	Excellent
Eptam	2	23	Excellent	10	Excellent	Fair
Eptam	4	22	Excellent	7	Excellent	Excellent
Eptam	8	16	Poor	3	Excellent	Excellent
Check		24	Excellent	10		

[1] The soil type was sandy loam and incorporation wa: done with a spike-tooth harrow.
[2] Stand per 3 ft of row (avg of eight random counts).
[3] Growth rated on a scale of 0 to 10 with 10 being normal compared to the check and 0 being death.

The data show control of water grass, red-root pigweed and lambsquarters but other trials showed that a number of annual grasses and broadleaves were controlled with the 4-lbs per acre late. In addition, excellent control of yellow nutgrass was obtained with the same rate. Over a variety of conditions, the best weed control was obtained with a Bye-Hoe where band treatments

were made and with cross discing where solid treatments were made. Similar results were obtained with commercial applications in 1961 and are discussed in detail below.

Beet stands were not affected by any rate of Tillam at any location. At most locations, Eptam severely reduced the stands at 8 lbs per acre with slight to moderate stand reduction at the 4-lb rate. Early growth of beets with Tillam was not affected at the 2- and 4-lbs per acre rates, and at the 8-lb rate, only slight to moderate early stunting occurred. Depending upon moisture and other growing conditions, this stunting was not visible 3 to 6 weeks after application and seeding. Eptam severely reduced early growth in most trials at the 8-lb rate and at 4 lbs per acre, early growth reduction was usually moderate.

A summary of yield data obtained from four trials is presented in Table 3. Only the Tillam 4-lb per acre treatments and the checks were sampled for beet tonnage and sugar content. At all four locations, the yield of sugar was greater with the Tillam treatment.

Table 3.—Effect of Tillam at 4 lbs per acre on beet tonnage and sugar content.

Trial No.	Soil Type	Tons of beets/A		% Suci		Lbs of sugar/A	
		Tillam	Check	Tillam	Check	Tillam	Check
7	Sandv loam	10.91	9.57	14.4	13.6	3,412.1	2,603.0
9	Clav loam	24.11	17.85	16.4	16.5	7.908.0	5,890.5
12	Light peat (15.1% O.M.)	215.90	21.26	13.9	13.0	6.644.2	5,527.6
16	Light peat (16.1% P.M.)	16.12	14.98	14.0	14.0	4.594.8	4.194.4
Totals		75.33	63.66	58.7	57.1	22.289.1	18.215.5
Averages		18.83	15.91	14.7	14.3	5,575.6	4.562.2

Soil residual of Tillam at 4 lbs per acre was determined by sampling a number of the treated fields. Soil samples were obtained at intervals throughout the season and immediately bioassayed. The bioassaying was done in a greenhouse using cultivated oats, a highly Tillam-susceptible crop, as the test plant. A total of ten locations were sampled to cover conditions varying from sprinkler irrigation to furrow irrigation and from sand to clay loam to light peat soils. The oats germinated and grew normally at least by the sixteenth week after treatment at all locations. Other data, experimental and commercial, show that recommended rates of both Eptam and Tillam pose no problem to subsequent susceptible crops, providing adequate moisture has been available for the treated crop to produce satisfactorily.

1961 California Program and Results

During the 1961 season, field experiments were continued in some areas in addition to an experimental sales program in all areas. Under the experimental sales program, a total of 6,000 acres of beets was treated with the majority of the acreage in the Sacramento Valley. Commercial applications were made, however, in all beet-growing areas which cover both spring and fall plantings. The results are discussed below under appropriate headings:

A. *Rates of Application:* The rate of application used was 4 lbs on all mineral soils and 6 and 8 lbs per acre on peat soils. These rates were used on an over-all solid coverage basis and with band treatments, the rate per crop acre was reduced according to the width of the band treated and the row spacing. In many fields, 8 lbs per acre were applied to a small portion as a check on beet tolerance. On mineral soils there was no injury wherever the 8-lb rate was used but in two cases where only the 4-lb rate was used, slight reduction in early growth occurred. On peat soils there was no injury from either the 6- or 8-lb rates.

B. *Incorporation:* Excellent results were obtained wherever Tillam was immediately and thoroughly incorporated into the soil. Delayed, and/or poor incorporation resulted in varying degrees of weed control ranging from no control to near commercial control.

The majority of applications were band treatments using a Bye-Hoe for incorporation The Bye-Hoe was found to be the best tool under a wide variety of conditions as discussed below under "Soil Factors." The Cultro and Chattin were found to be effective only on peat soils and light, sandy soils. With solid treatment for flat planted beets, cross discing was uniformly effective. Spike-tooth harrowing three to four times with each harrowing at a right angle to the previous one was effective if the soil were quite loose and friable.

C. *Bed Shaping and Planting:* The main causes of poor control on bedded beets were: failure to form the beds prior to application, removing treated soil from the bed-top with sled planters and moving untreated soil from the furrows onto the treated bed during the planting operation.

D. *Soil Factors:* Soil moisture and mineral soil types did not affect results except indirectly as they influenced incorporation. This was true with all equipment other than the Bye-Hoe. With equipment other than the Bye-Hoe, adequate incorporation becomes more difficult the heavier and wetter the soil. One trial on clay loam soil was applied where the soil had free water on the

surface and was above field capacity in the top four inches. Even under these conditions, excellent weed control was obtained when incorporation with a Bye-Hoe was done immediately after spraying. Mineral soil types ranged from light sands to the heavy clay-high salt soils of the Imperial Valley.

Organic soils did not affect the results until the organic matter content exceeded 20%. The 4-lb per acre rate gave good results at organic matter contents up to 20% and in the 20 to 30% organic matter content range, 6 lbs per acre were required for good weed control. At organic matter contents above 30%, a rate of 8 lbs per acre was required for satisfactory weed control.

Following application, all types of soil moisture conditions prevailed. Following application to dry, moist and wet soils, conditions ranged from immediate rainfall, sprinkler irrigation or furrow irrigation to no additional moisture for a period of two weeks or longer. In all cases, excellent results were obtained where incorporation was done immediately and properly.

E. *Weeds Controlled:* A number of annual grasses were controlled with the major ones being water grass *(Echinochloa* spp.) and wild oats *(Avena fatua).* Of the annual broadleaf weeds controlled by the 4-lb rate of Tillam wherever the rhizomes were *(Chenopodium album),* nettle-leaf goosefoot *(Chenopodium murale),* red-root pigweed *(Amaranthus retroflexus)* and purslane *(Portulaca oleracea).*

In addition to control of annual weeds, yellow nutgrass continued to be controlled in all cases with the recommended rate of 4 lbs per acre. It was observed in a limited number of fields that Bermuda grass and Johnson grass from rhizomes were controlled by the 4-lb. rate of Tillam wherever the rhizomes were thoroughly chopped up prior to, or during the application.

F. *Length of Weed Control:* Most applications resulted in weed control well beyond thinning time and where solid treatments were made on flat planted beets, weed control extended to harvest time in a number of fields. The length of control in a given field was dependent upon numerous factors such as band or solid treatment, rainfall and irrigation, weeds involved, and type and frequency of cultivation.

Summary and Conclusions

Two years of extensive field testing and one year of commercial use have proven Tillam to be a selective preplant soil-incorporated herbicide effective in controlling many of the major annual grassy and broadleaved weeds and some of the perennial weeds which occur in sugar beets. Weeds controlled include watergrass, wild oats, red-root pigweed, lambsquarters, purslane

and nutgrass. Rates of 4 lbs per over-all acre have been effective on all mineral soils and organic soils containing up to 20% organic matter. On soils containing above 20% organic matter, rates of 6 to 8 lbs per acre over-all are required depending upon the organic matter content. Under most conditions rates at least double those required for weed control caused no appreciable injury to sugar beets.

Results have been highly uniform over a wide range of soil and climatic conditions wliere incorporation of the "Tillam into the soil has been done immediately and properly. Under commercial conditions the most satisfactory method of application has been to spray and incorporate (with a power-driven rotary tiller) on the front tool bar of the tractor and to seed with the same tractor by having the seeders mounted on the rear tool bar.

Electrostatic Separation of Cysts of the Sugar Beet Nematode

D. R. VIGLIERCHIO AND J. R. GOSS[1]

Received for publication February 14, 1962

The separation of a large number of nematode cysts cannot be accomplished by repeated use of any one highly-refined separation process. Since there can be considerable overlap in the properties of cysts and accompanying debris, a higher degree of enrichment can be achieved by utilizing a series of less sensitive procedures, each based on different properties (2, 3)[2]; hence, the investigation of the electrostatic separation of cysts from debris.

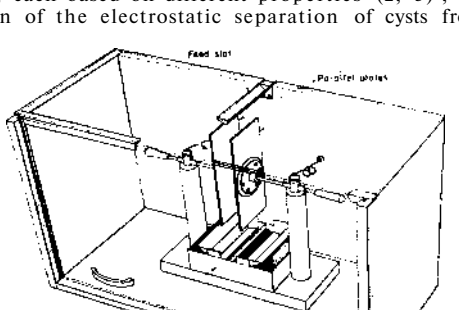

Figure 1.—Schematic diagram of the electrostatic separator.

Apparatus and Technique

The device used for this purpose (Figure 1) consisted of brass parallel plates (100 X 250 X 3 mm) attached to brass sliding rods mounted on bakelite posts which in turn were mounted on a bakelite floor plate. The fraction collector consisted of a beta pan ($\pm\beta$ fraction collector, 205 X 155 X 62 mm with a floor sloping away from either side of a central knife edge) and 2 alpha pans ($\pm a$ fraction collectors, 55 mm wide) constructed as shown so that the positions of the knife edges were adjustable. In practice the three fraction-collector knife edges were fixed so that the plate gap was divided into four equal sections for each gap setting, $+a$ representing that section nearest the positive plate, $+\beta$ the

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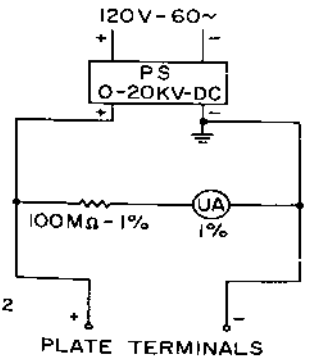
[2] Numbers in parentheses refer to literature cited.

adjacent section on the positive side of the central knife edge, $-\beta$ the section on the negative side of the central knife edge, and $-a$ the section nearest the negative plate. When the plates were in contact and vertically aligned with the feed slot and the beta pan knife edge, they were 90 mm above the beta pan and 10 mm below the feed slot. The plate assembly was positioned so that the plates extended 10 mm beyond both ends of the feed slot in order to reduce the edge effects of the electrostatic field. In practice only the middle portion of the slot was used since the pour time for the small samples was about 20-30 seconds.

The enclosure (330 X 750 X 480 mm) constructed of plyboard and plexiglass reduced cyst movement by air currents and served as a shield from the high voltages applied to the plates.

The variable high voltage supply was provided by a Pt. No. HV200-102 from Plastic Capacitors Inc., Chicago, Illinois. The resistors and microammeter were precision 1% tolerance devices (Figure 2). The applied plate voltage was calculated from the 100 megohm series resistance and the measured current flow. The plates were sprayed with acrylic resin to reduce particle jumping from plate to plate at high potentials. Grounding of the feed slot improved the reproducibility of the separation.

Three samples were run at each set of conditions, cysts and debris in each fraction were hand separated for weighing and average weights then represented the results for a given set of conditions.



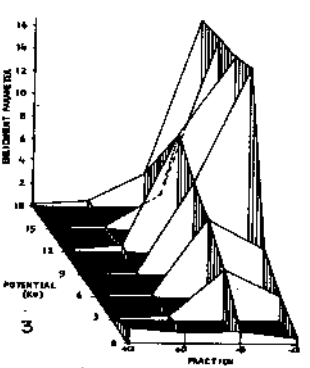


Figure 2.—Schematic wiring diagram for the separator.

Figure 3.—Enrichment parameter variation with increasing potential each fraction at constant plate gap (12 cm) and humidity (25% RH).

Results

A good separation pattern is shown in Figure 3. The fractions were arranged in sequence from positive to negative plate. The measure of degree of cyst separation and yield has been expressed as a composite relaton, EP (enrichment parameter) where

$$EP = \left(\frac{\text{Avg. wt. of cysts in a fraction}}{\text{Avg. wt. of debris in a fraction}}\right)\left(\frac{\text{Avg. wt. of fraction}}{\text{Avg. wt. of sample}}\right)$$

EP was selected in this manner as a compromise between purity and overall cyst yield. The bulk of the material was usually collected about the center of the gap, i.e., $+\beta$ and $-\beta$. The greatest purity was obtained near the plates, i.e., $+a$ and $-a$. When $-\beta$ and $-a$ had similar EP values, the $-\beta$ value resulted from a greater proportion of material at low purity whereas the $-a$ value resulted from a low proportion of material at much higher purity. When EP values were very high >10, both purity and yield were relatively high resulting in the greatest number of pure cysts. For example at 25% RH, a plate gap of 12 cm and from 9-18 KV, the best fraction, $-a$, contained about 35% of the cysts and 1.9% of the debris from a sample, 75% cysts and 25% debris. Cysts in an electrostatic field therefore migrated preferentially toward the negative plate.

The electric field intensity (E) between parallel plates was essentially a function of plate potential (V) and distance between plates (d), i. e.,

$$E = \frac{V}{d}$$

when d was small with respect to the dimensions of the plates. The EP curves in Figure 4 show that enrichment was proportional to electric field intensity at the smaller plate gaps where the d assumption was valid. At larger plate gaps fringe field effects could no longer be neglected and EP was no longer proportional to $\frac{V}{d}$. Higher potentials than those shown for each plate gap were impractical since charge transfer caused the particles to jump from plate to plate and rendered the biodata unreliable.

In recycling the enrichment parameter for an efficient system could be expected to decrease with the removal of the more pure fraction. In Figure 5 it is evident that separation was essentially completed in two passes when about 75% of the cysts were obtained. It became increasingly more difficult to separate the remaining cysts from the more concentrated debris

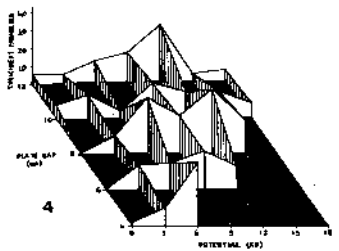


Figure 4.—Enrichment parameter variation with increasing plate gap and potential at constant humidity (45% RH) for the $-a$ fraction.

Figure 5.—The enrichment parameter of successive $-a$ fractions obtained by re-running the combined $-\beta$, $+\beta$ and $+a$ fractions from the previous run. D is the final recombination.

Moisture, troublesome in sustaining useful static charge distributions because of increased leakage currents or charge migrations, was a factor in electrostatic separation (Figure 6). The cyst concentrate was stored at 43% RH until just before processing through the apparatus which was maintained at the RH indicated in Figure 6. With cyst concentrate stored at 90% and

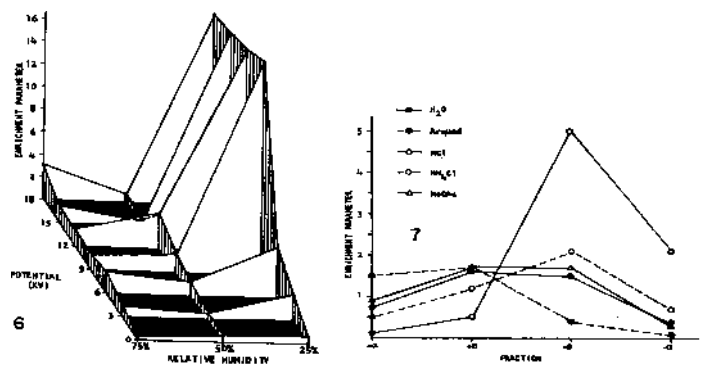


Figure 6.—Enrichment parameter variation with increasing potential and relative humidity at constant plate gap (12 cm).

Figure 7—Enrichment parameter variation of separator fractions with different treatment of lots of cysts from a common source.

53% RH and then processed through the separator at 45% RH the EP values were of the same order observed in Figure 6 for 75% and 50% RH. Cyst separation could be achieved at a higher RH by using electric fields of greater intensity though the enrichment was never as great as that achieved in the drier atmospheres.

The effect upon cyst separation of pretreatment with dilute solutions of ionic substances is shown in Figure 7. Dilute HC1 improved the normal cyst-debris separation. Ammonium chloride solution also improved the cyst separation but less markedly. Sodium acetate appeared to have no effect. Arquad, a cationic surface active agent (1), tended to reverse the normal cathode drift, i.e., the cysts migrated preferentially to the positive plate.

In view of the results (Figure 3) with a negatively grounded feed slot it might be expected that positively grounding the slot would affect the sample distribution. The polarity of the grounded feed slot was determined by the polarity of the grounded side of the high voltage supply, Figure 2. Positive grounding of the feed slot in processing impure cyst material did not necessarily improve the separation. Direct comparison of all four fractions showed that a negatively grounded feed slot was more effective as is also shown by an alternate comparison (Figure 5).

It is of interest to note that when cysts were collected on greased paper to preserve orientation, about 80% of the cysts were aligned with their major axis parallel with the electric: field irrespective of feed slot grounding polarity.

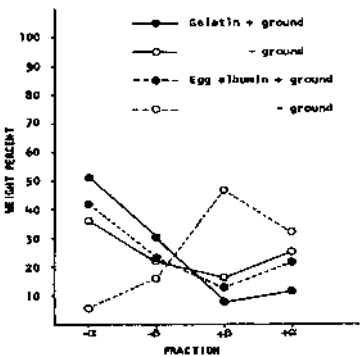


Figure 8.—Relative weight distribution in fractionation of gelatin and egg albumin with negatively and positively grounded feed slots.

If two purified proteins were treated in the same manner the resulting distributions were altogether different. Gelatin particles migrated preferentially toward the cathode plate; the migration was increased by using a positively grounded feed slot (Figure 8). Egg albumin particles migrated toward the cathode plate with a positively grounded feed slot but toward the anode plate with a negatively grounded feed slot.

Discussion

The orientation of the cysts with the major axis parallel to the electric field indicated that there was induced axial polarization of the cysts much as would occur with any elipsoidal particle. Presumably the polarization was superimposed upon the net charge of each particle. The cyst reaction was not indigenous to proteins. Gelatin and ovalbumin with similar iso-electric points differed markedly in their reactions with the electrostatic separator used for cysts. The charge on the ovalbumin particle was determined largely by the polarity of the grounded feed slot where the charge on the gelatin particle was only slightly modified by the polarity of the feed slot. It would be difficult to predict the successful electrostatic separation of other cyst-forming nematodes. The outcome would need to be empirically determined in view of the electrostatic response of the two purified proteins and the effects of mineral and surface active solutions on *Heterodera schachtii* cysts. The mechanism of separation is uncertain; further investigation is essential for a better understanding of the underlying principles.

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The Sphere Photometer

A New Instrument for the Measurement of Color and Turbidity in Solutions of White Granulated Sugars.

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Introduction

Solutions of pure sucrose appear to the human eye as water-white and brilliantly-clear fluids. In the manufacture of white granulated sugars, the industry aims at making a product which is essentially pure sucrose. The appearance of slight coloration and haze, common to solutions of all granulated sugars, is indicative of minute traces of impurities in the product. The accurate measurement of color and turbidity in sugar solutions has been of great concern to the industry, and a variety of methods and instruments were developed and evaluated over the years. The 'International Commission for Uniform Methods of Sugar Analysis" (ICUMSA) maintains standing committees on the subject (Subject 13).

Measurements of color and turbidity have a two-fold purpose: a) in process control, color and turbidity are considered indicative of the degree of decolorization and filtration achieved; b) in marketing, color and turbidity are considered to be expressive of the visual appearance of the product.

Color

Measurements of light absorption by the solution are the basis for the characterization of sugar color. The term 'color" is used here loosely, since it is used in the industry to express a) the quantity of impurities and b) the visual appearance to the human eye. Color is determined customarily by transmittancy measurements in the blue region of the spectrum. Most methods, recognized by ICUMSA and used in the industry, specify a wavelength of 420 mμ. The "color" is then expressed by an absorption index.

The colorants in sugar solutions absorb radiant energy more strongly in the blue region of the spectrum than in the red region, as shown in Figures 1 and 2. The shapes of the absorbancy curves vary significantly and it may be assumed that the variations are due to differences in raw material, processing, and pH of the solutions.

While the area under each of the curves may be a measure of the quantity of colorant in the solution, the absorbancy at a given wavelength is not necessarily a precise measure of this

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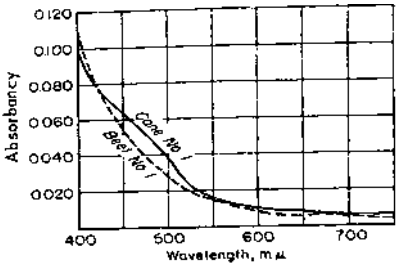

Figure 1 •

Figure 2.

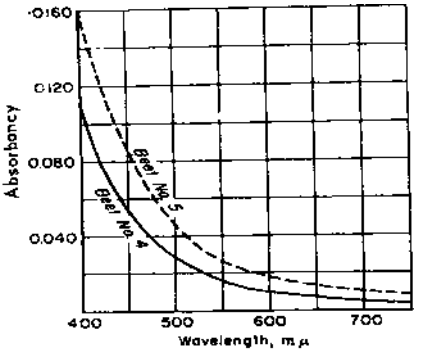

Figure 1 and Figure 2.—Absorbancies of granulated sugar solutions, filtered, from 400 to 750 mμ. Reference standard: Clear, colorless sugar solution, 50 rds. solutions, 5cm cell.

quantity. The absorbancy has, however, great utility in the evaluation of the decolorization processes.

The absorption index is also regarded as being a measure of "color" in terms of visual appearance. This is erroneous since visual color perception is a psychophysical process and may be expressed quantitatively only on a three-dimensional scale, such as the CIE tristimulus coordinates [1][2] and related systems. It has been shown, however, that the absorbancy of sugar solutions at 420 mμ correlates well with visual rankings [2].

[2] Numbers in brackets refer to literature cited.

Turbidity

In science the word turbidity has a very definite optical meaning. The general term for the phenomenon is light scattering, which the human eye perceives as a haze. It is the result of refractive index gradiants in the sugar solution. While insoluble material in sugar solutions contributes the larger part of optical turbidity, the use of the word turbidity to denote a quantity of insoluble material had led to confusion. It is erroneous to assume that the colorants in sugar solutions only absorb light and that the suspensoids only scatter light. It is now considered most likely that dissolved absorbing molecules also scatter light and that scattering suspensoids also absorb light. Color and turbidity, when evaluated by optical means, are optical quantities which cannot be accurately separated by mechanical means, such as filtration or centrifuging.

Rieger and Carpenter [3] investigated the scattering of light by sugar solutions and give the following definitions:

"Turbidity is the amount of light scattered per unit path length as defined by the equation:

$$-\ln T_s = \tau\, b, \qquad (1)$$

wherein T_8 is the transmittancy of the solution, b is the path length in cm, and $_T$ is the turbidity in cm^{-1}. The equation applies only to systems which scatter light without absorption.

"In systems which absorb light without scattering:

$$\frac{-\log T_s}{bc} = a \qquad (2)$$

wherein a is the absorption index, and c is the sugar concentration in grams per milliliter.

"In systems which absorb and scatter light:

$$\frac{-\log T_s}{bc} = a^* \qquad (3)$$

wherein a* is the attenuation index.

"Equation (1) for scattering without absorption may be written in the same form as equations (2) and (3), to define the scattering index s, as follows:

$$\frac{-\log T_s}{bc} = \frac{\tau}{2.303c} = s \qquad (4)$$

"In sugar solutions, which both absorb and scatter light, the attenuation is assumed to equal the sum of absorption and scattering. In terms of the indices, one writes:

$$a^* = a + s.$$

(The indices of white sugar solutions are small. To eliminate fractions, we write: 1,000 a = Color Index, and 1,000 s = Turbidity Index.)

"In all of the preceding equations, the turbidity is expressed in terms of light lost from the transmitted beam. However, turbidity may also be evaluated by a direct measurement of all light scattered in all directions:

$$\tau = 2\pi \int_0^{\pi} R_\Theta \sin \Theta \, d\Theta, \quad (6)$$

wherein Θ is the angle of observation and RΘ is the Rayleigh ratio, expressed as:

$$R_\Theta = \frac{i_\Theta r^2}{I_o V} \quad (7)$$

wherein r is the distance between the scattering volume, V, and the observer, iΘ is the intensity of the scattered light, and I_0 is the irradiance of the incident light."

Measurements of iΘ can be made on special photometers [4] but the complete procedure is too time-consuming for routine use. An abbreviated procedure was developed by Rieger and Carpenter [3], but the necessary instruments are quite expensive

Photometers and colorimeters using transmission measurements in the blue and red regions of the spectrum for the estimation of color and turbidity are not completely satisfactory. The use and calibration of these instruments are predicated on the erroneous assumption that a mechanical removal of suspensoids by filtration is suitable for the estimation of turbidity.

Since turbidity can be accurately evaluated only through measurements of light scattering, Spreckels Sugar Company undertook the development of a new photoelectric instrument for the evaluation of sugar solution color and turbidity through measurements of light transmission and scattering.

Description of apparatus optics

The optical train of the new apparatus is shown schematically in Figure 3.

Light from the filament of the lamp LS is collimated by lens LI and passes through filter F to lens L2, which focuses the image of the filament into aperture Al in the wall of the integrating sphere. Lens L3 again collimates the light and lens L4 brings the image of aperture Al to focus in aperture A2. When the exit shutter is open, essentially all light entering the sphere through aperture Al leaves through aperture A2. The interior of the sphere, the exit shutter, and the baffle are covered with a white paint of high diffuse reflectance. When the exit shutter is closed, the light entering the sphere through Al is directed by lenses L3 and L4 to the surface of the exit shutter and reflected

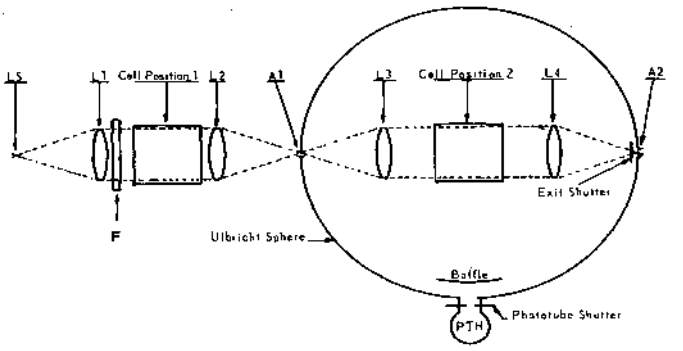

Figure 3.—Optical train, schematic.

Legend:		
	LS	**Lamp, GE No. 1493**
	L1 to L4	**Lenses, achromatic, 63 mm FL, 40 mm dia.**
	F	**Interference filter, 420 mμ, B & L Cat. No. 33-78-42**
	A1	**Entrance aperture, 1.5 mm dia.**
	A2	**Exit aperture, 3.0 mm dia.**
	PTH	**Housing for 1P21 phototube**
	Cell	**Aminco DX-5-1005, glass**
	Sphere	**10 in. diameter**

to illuminate the interior of the sphere. After repeated reflections within the sphere, a fraction of the light illuminating the interior of the sphere enters the phototube housing PTH, where it strikes the light-sensitive cathode of the tube. The phototube is connected, through suitable electronic circuits, to calibrated potentiometers on which the light flux striking the cathode is read.

Solutions to be analyzed are placed in a cylindrical sample cell made from clear glass, having a length and diameter of 5 cm. The cell is placed into the instrument so that the collimated light beam passes axially through the cell. When the cell is in position 1, the exit shutter in the sphere is closed and the light flux reaching the phototube after reflection from the exit shutter and sphere wall is a measure of the transmittancy of the solution.

When the cell is placed in position 2, the exit shutter is opened and the light transmitted by the cell contents passes out of the sphere through aperture A2. The light scattered by the suspensoids in the sample is attenuated by the absorbers in the sample and, after reflection from the interior of the sphere.

reaches the phototube. The light flux to the phototube is a measure of the turbidity of the solution.

Since photoelectric detectors respond to radiant flux, rather than intensity, the equations presented below are in terms of flux. Designating the flux incident on the first surface of the sample as F_o, and the flux leaving the second surface of the sample as F_t, the transmittance of the sample,

$$T = F_t/F_o, \tag{8}$$

and the transmitted flux,

$$F_t = TF_o. \tag{9}$$

In the sugar industry it is customary to express the transmittancy of a sample solution relative to the transmittance of a "standard," a clear, colorless sugar solution having a sugar concentration equal to that of the sample solution. This reference solution is regarded as the solvent. The transmittance of the sample solution,

$$T_s = T_{soln}/T_{solv}. \tag{10}$$

Expressed in terms of flux, the transmittancy of the sample solution,

$$T_s = F_{t_{soln}}/F_{t_{solv}}. \tag{11}$$

In practical applications, it is customary to standardize the photometer by placing the sample cell containing the standard solution into the instrument and adjusting it to indicate a transmittancy of unity.

Then the transmittance of the sample solution,

$$T_s = F_t. \tag{12}$$

The incident flux F_o, illuminating the colorants and turbidants in the sample solution, then equals unity, and the transmitted flux F_t expresses the transmittancy of the impurities in the sample solution. Designating the transmittancy of the absorbers of radiant energy in the samples as T_a, and the transmittancy of the scatterers of radiant energy in the samples as T_x, the following notation is applicable:

When the sample contains only absorbers, the transmitted flux

$$F_t = T_a. \tag{13}$$

When the sample contains only scatterers, the transmitted flux

$$F_t = T_x. \tag{14}$$

When both are present in the sample, the transmitted flux

$$F_t = T_aT_x. \tag{15}$$

When the sample contains only absorbers, the absorbed flux

$$F_a = 1\text{-}T_a. \tag{16}$$

When the sample contains only scatterers, the scattered flux

$$F_x = 1\text{-}T_x. \tag{17}$$

When both impurities are present in the sample and the transmitted and scattered beams pass through equal distances within the solution, the scattered flux

$$F_x = (1-T_x)\ T_u. \qquad (18)$$

Theoretical considerations in design

1. Light scattered by the suspensoids in solutions of white granulated sugars travels in a predominantly forward direction, i.e., in the direction of the transmitted beam. It is inevitable that a fraction of the scattered flux enters the detector together with the transmitted flux. The fraction k of the scattered flux entering the receiver in the transmittancy measurement is a function of the angular distribution of the scattered energy and of the angle of acceptance of the detector. The true transmittancy of a sample containing only scatterers,
$$T_x = F_t. \qquad (14)$$
The measured transmittancy of a sample containing only scatterers,
$$T_M = F_t + kF_x. \qquad (19)$$
In most currently used colorimeters, no attempt was made to exclude forward scattered light from the transmittancy measurement and the measured transmittancies of sugar solutions are significantly higher than the true transmittancies. The new photometer reduces the transmittancy error to a negligible value through the use of small apertures in the optical system.
2. In the scattering measurement according to equation (18), the length of the path traversed by the scattered beam must equal that traversed by the transmitted beam. This condition may be entirely met only in Angular Scattering Photometers [4]. In the concept of the sphere photometer it is partly met when working with solutions of white granulated sugars, since most of the scattered flux travels in a forwardly direction. The use of a clear glass cell, having equal diameter and length, facilitates inclusion of essentially all scattered energy in the measurement of scattered flux, but the condition of equal path lengths cannot be entirely fulfilled. For the measurement of scattered flux, we conceived placing the sample cell into an Ulbricht sphere (integrating sphere), and making provision for the entry of the incident flux and the exit of the transmitted flux.
3. When the measured fluxes F_t and F_x represent the functions of equations (15) and (18), transmittancies T_u and T_x may be calculated.

$$F_t = T_s T_x \quad (15)$$

$$F_x = T_s - T_s T_x \quad (18)$$

$$F_t + F_x = T_s \quad (20)$$

$$F_t / T_s = T_x \quad (21)$$

4. While considering the use of the integrating sphere in the measurement of scattered flux, it was anticipated that light reflected from the lenses and sample cell windows would augment the flux reaching the phototube. The light reflected from lens L3 and the entrance window of the sample cell is a constant fraction of the incident flux, while the light reflected from lens L4 and the exit window of the cell varies with the transmittancy of the cell contents. Designating the energy reflected by lens L4 and the exit window as m, and that reflected by lens L3 and the entrance window as n, the flux to the phototube is defined by the equation

$$F_x = (1 - T_x) T_s + m T_s + n. \quad (22)$$

The evaluation of quantities m and n is discussed under "Experimental procedures."

5. Equation (22) may be written:

$$F_x = (1 + m) T_s + n - T_s T_x \quad (23)$$

Since $F_t = T_s T_x$, eq. (15):

$$T_s = \frac{F_t + F_x - n}{1 + m} \quad (24)$$

Practical considerations in design

1. Since the two separate measurements required cannot be made simultaneously, the incident flux must be held constant during the time required for the measurements. For a maximum measurement error of ± 0.1%, the voltage supplied to the light source, an incandescent filament lamp, is held stable within ± 0.03%.
2. The low light levels involved in the measurements require the use of a beam multiplier phototube. This tube has a voltage-dependent amplification. To maintain a stability of ± 0.1%, the applied voltage is held constant within ± 0.015%. The tube passes a small current when not illuminated, the dark current. Means for neutralizing this dark current are provided.
3. The two required measurements should be made with identical light sources, filters, and detectors. To meet this requirement, a single instrument with provisions for both measurement functions and using a single light source, filter, and detector is used.

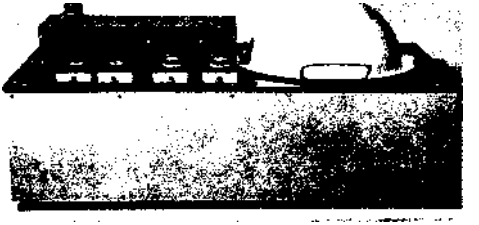

Figure 4. — General appearance of the photometer.

4. Since mechanical filtration is not suitable for the separation and identification of absorbers and scatterers of radiant energy, the apparatus is adaptable to calibration by other fundamental means.
5. It is not practical to use routinely the primary standard, a 50 rds clear, colorless sugar solution, in the standardization of the instrument. Secondary standard glass plates, as used in a number of colorimeters, are subject to gradual deterioration and breakage. Manual adjustment of the transmittancy dial to the transmittancy of the secondary standard adds to manipulations required and tends to increase transmittancy errors. The new instrument uses the air path through the optics as the secondary standard and the manual standardization is automated.
6. To permit determination of flux levels to 0.1%, the apparatus employs potentiometers capable of 0.1% resolution and a null indicator of corresponding sensitivity.
7. To insure adequate photometric sensitivity, the apparatus uses a wave length of 420 m_u The optical filter used is a Bausch and Lomb interference filter, Catalog No. 38-78-42.

Description of apparatus

The apparatus was designed to meet the theoretical and practical requirements outlined in the preceding sections, and to make the operation of the instrument reasonably simple and foolproof.

Figure 4 shows the general appearance of the photometer. The elevated structure on the left contains the light source, lenses LI and L2, the filter, and supports for the sample cell in the measurement of transmitted flux (Position 1). In front of this structure are located the operating controls. The hemisphere on the right is the supper half of the integrating sphere.

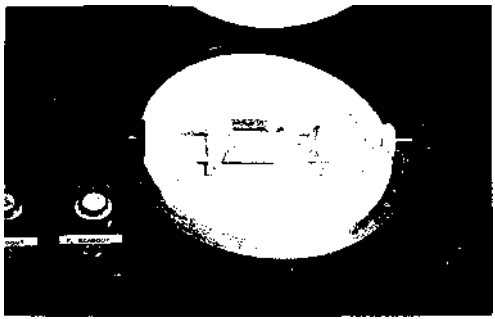

Figure 5.—Sphere opened.

Figure 5 shows the sphere opened, with the sample cell in position for the measurement of scattered flux (Position 2). Visible at the right is the sphere exit shutter.

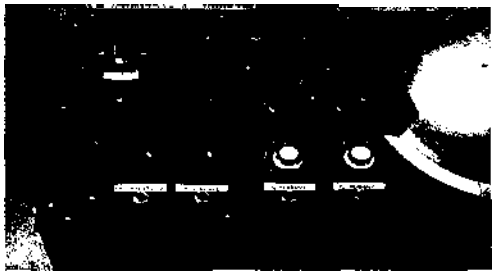

Figure 6 — Close-up of the cell compartment and controls.

Figure 6 is a close-up of the cell compartment, operating controls, and null-indicating galvanometer. Located in front of each operating control is a pushbutton. When a pushbutton is depressed, internal circuits are switched and the shutters are actuated as needed to perform the selected function. The controls shown, from left to right, are: 1) dark current compensation, 2) standardization, 3) readout of transmitted flux, F_t, and 4) readout of scattered flux, F_x. The null indicator indicates the direction in which the selected control must be rotated to balance the corresponding circuit. When the null indicator returns to the zero position, the control function is completed.

Experimental procedures and data

The instrument is connected to the 117 volt, 60 cycle lines through a three-wire cable, and turned on by operation of the line switch, located on the right side of the cabinet. After a warm-up period of about 5 minutes, dark-current compensation is made by manipulation of the dark-current controls. The instrument is then ready for use.

1. *Standardization*

The purpose of the initial standardization procedure is the adjustment of the "Secondary Standard" to provide a reference potential related to the transmittancy of the primary standard. The reference potential permits routine standardization of the photometer, using the air in the optical path through the instrument as a secondary standard.

The primary standard, a clear, colorless 50 rds sugar solution, is prepared according to prescribed procedures. The clean sample cell, filled with this solution, is placed in position 1 in the instrument and the dark-current compensation checked. The F_t dial is set to indicate 1.000. While depressing the F_t button, the standardization control on the control panel is rotated to balance the null indicator. The sample cell is removed from the instrument and a "Secondary Standard" potentiometer inside the instrument case is adjusted, while depressing the "Standardize" button, to rebalance the null indicator. Hereafter the instrument may be standardized while the sample cell is outside the instrument, by operation of the standardizing button and control to balance the null indicator.

2. *Measurement of* F_t *and* F_x.

After adjustment of the dark current and standardizing controls, the cell containing the sample is placed in position 1 in the instrument. The F_t button is depressed and the F_t control is rotated to balance the null indicator. The transmitted flux of the sample is now indicated on the digital display of the F_t control.

The sample cell is next placed in position 2 inside the sphere and the sphere is closed. The F_x button is depressed and the F_x control is rotated to balance the null indicator. The scattered flux is now indicated on the digital display of the F_x control. Flux values are indicated to three decimal places.

F_t and F_x values were determined on the primary standard and on distilled water which had been filtered through a thin layer of Darco on a .45 *m^* Millipore membrane for the removal of suspended solids. The values obtained are listed in Table 1. The data indicate that the distilled water is suitable for use as a primary standard in the photometer at a wave length of 420 m^.

Table 1.—Evaluation of Standards.

Sample	Ft	F_x
Primary Std.	1.000	.257
Dist. Water	1.000	.257

The scattered flux indicated is due to radiant energy reflected by the optical components in the sphere, since the primary standard is considered turbidity-free. Since the distilled water and the primary standard display identical optical properties at 420 m_u, the distilled water may be conveniently used in the preparation of artificial absorbing solutions and scattering suspensions necessary for the evaluation of the photometer.

3. *Photometer performance with turbid suspensions.*

Suspensions of Dicalite fines in water scatter light in a manner which closely approximates the scattering of light by the natural suspensoids in solutions of white granulated sugars. For the investigation of the relationship of transmitted and scattered flux in the photometer, such suspensions are well suited. The suspensions are prepared by dispersion of about 2 tablespoons of Dicalite in a one-liter graduate filled with distilled water. The suspension is left to settle about 24 hours. After removal of the upper 100 ml by syphonina:, the following 100 ml are transferred to an Erlenmeyer flask and diluted to approximately 1 liter. The dilute suspension can be used for several days when mildly agitated by a magnetic stirrer.

Transmitted and scattered flux were measured on the photometer, on the distilled water and five suspensions of Dicalite fines are listed in Table 2.

Table 2.—Dicalite Suspensions.

Sample	F_t	F_x (y)	1-F_t (x)
Water	1.000	.257	0
Susp. 1	.986	.275	.014
Susp. 2	.968	.304	.032
Susp. 3	.895	.390	.105
Susp. 4	.753	.571	.247
Susp. 5	.548	.851	.452

From the data, we may calculate the relationship of F_t and F_x:

$N = 6 \qquad 5x = .855 \qquad Sy = 2.628$

$Sx^2 = .2779 \qquad 2xy = .5727$

$y_c — .257 + 1\text{-}270\ x$

Thus the scattered flux $F_v = .257 + 1270\ (1\text{-}F_t)$. A plot of the data, Figure 7, shows that the scattered flux is a linear function of the transmitted flux. The constant .257 is the flux contributed by reflections from the optical components in the sphere.

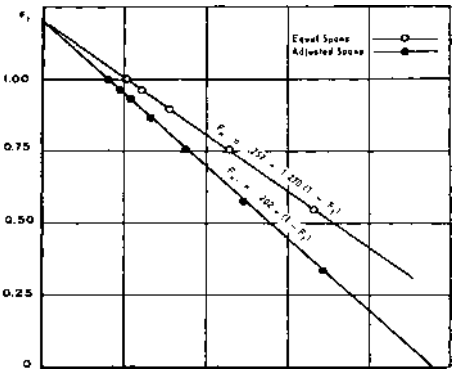

Figure 7.—Dicalite suspensions.

The factor 1.270 indicates that the flux incident on the sample in position 2 is 1.270 times the flux incident on the sample in position 1. While it is not convenient to adjust the two incident fluxes to equality, the factor 1.270 may be readily eliminated by making the span of the F_x potentiometer 1.270 times the span of the F_t potentiometer.

Then: $F_x = .202 + (1\text{-}F_t)$ (25)

The ratio adjustment was checked with a series of Dicalite suspensions, noting the F_t and F_x, values for each dilution. The data are plotted on Figure 7, showing that the scattered flux, $F_x = .202 + (1\text{-}F_t)$, as calculated.

4. ***Photometer performance with dye solutions.***

The effect of absorbers on the measured flux F_x was investigated, using dilutions of a dye which does not contribute measurable scattering. A stock solution of a red dye was prepared in distilled water and filtered through a 0.45 mμ Millipore membrane. Transmitted and scattered flux were measured on a series of dilutions as presented in Table 3.

The data were plotted and are presented in Figure 8. It is evident that for the transmittancy range of 1.000 to 0.575, flux F_x is a linear function of the transmittancy, and that the relationship may be expressed by the equation:

$$F_x = .038 + 0.162\ F_t \qquad (26)$$

Since a transmittancy of .575 is indicative of a color index of 78 when using a 50 rds solution in a 5 cm cell, the equation is usable

Table 3.—Dye Solutions.

Sample		
Water	1.000	.200
1)\e 1	.928	188
Dye 2	.863	
Dve 3	.751	
Dye 4	.575	131
Dve 5	.346	103
Dve 6	.133	
Dve 7	.027	

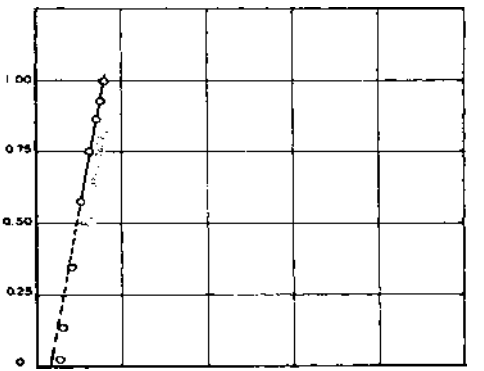

Figure 8. — Dye solutions.

for white granulated and darker sugars. It may be assumed that the constant .038 represents the radiant energy reflected by lens L3 and the entrance window of the sample cell, and the factor 0.162 represents the radiant energy reflected by lens L4. An examination of the sphere optics, Figure 3, indicates that this assumption is reasonable. The flux incident on lens 1,3 and the entrance window of the sample cell in position 2 is constant. When the cell contents absorb radiant energy, the flux incident on L4 is attenuated and the fraction reflected by the lens is attenuated to the same degree.

5. *Development of working eqziation.*

From the data and equations (25) and (26), derived independently for turbid solutions and absorbing solutions, the general equation for the flux F_x, incident on the phototube when the cell is filled with a solution which scatters and absorbs radiant

energy, may be developed. Assuming that the radiant flux scattered by the suspensoids in the solution traverses a path length through the solution which is substantially equal to that traversed by the transmitted beam, the flux incident on the phototube,

$$F_x = (1-T_x)T_a + 0.162\ T_a + 0.038 \quad (27)$$

$$F_x = 1.162\ T_a + 0.038 - T_aT_x \quad (28)$$

We may now determine the transmittancy of the absorbers, T_a, by combining equations (15) and (28):

$$T_a = \frac{F_t + F_x - .038}{1.162} \quad (29)$$

The transmittancy of the scatterers:

$$T_x = F_t / T_a \quad (21)$$

To test the practical validity of equation (29), samples of different transmittancies were prepared from the dye stock solution. To these were added varying amounts of the Dicalite suspension while holding the dye concentration constant. The values of T_a, calculated from the measured fluxes F_t and F_x with the aid of equation (25), are listed in the body of Table 4.

Table 4.—Transmittancies of Dye Solutions with Added Dicalite.

	ml Dicalite Suspension Added			
Sample	**0**	**10**	**20**	**40**
	Transmittancy, T_a			
Water	1.000	1.000	.997	1.000
Dye 1	.964	.962	.963	.962
Dye 2	.929	.928	.928	.927
Dye 3	.866	.865	.863	.861
Dye 4	.752	.752	.751	.748

The data indicate that there is a slight reduction in transmittancy at the higher levels of added turbidity. This is not necessarily due to erroneous assumptions in the development of equation (25). It is quite possible that some interaction occurs between the dye and the Dicalite fines. The practical effect of the observed variations is best illustrated when the transmittancies are converted to indices. The "Color Index" = 1,000 (-log T_a) /bc, and the "Turbidity Index" = 1,000 (-log T_x) /bc, wherein b is the cell length in centimeters and c is the sugar concentration of the sample in grams per milliliter. The color indices calculated from the data are shown in Table 5.

The turbidity levels produced by addition of varying amounts of Dicalite are, in turbidity indices, approximately 5 for 10 ml, 10 for 20 ml, and 20 for 40 ml. The color and turbidity index range covered by the solutions exceeds the range found in solutions of white granulated sugars. The variations in the tabulated

Table 5.—Color Indices of Dye Solutions With Added Dicalite.

Sample	ml Dicalite Suspension Added			
	0	10	20	40
	Color Indices, 1,000 (-log T_∞)/bc			
Water	0	0	0.4	0
Dye 1	5.2	5.5	5.3	5.5
Dye 2	10.5	10.6	10.6	10.8
Dye 3	20.3	20.5	20.8	21.1
Dye 4	40.2	40.2	40.4	41.0

color indices are partly due to the uncertainty (+ .001) in each of the two flux measurements, which would produce an error in the indices of 0 to ± 0.3 index units. The data from dye 3 show the largest relative variation, ± 0.4 units on a mean of 20.7. This variation is ± 2% of the measured quantity and is considered acceptable.

6. *Reproducibility of analyses.*

The overall performance of the instrument is a criterion of the stability of the electronic circuits and of the light source, as well as the sensitivity and resolution of the readout devices. To evaluate the performance of the instrument, about 2 liters of a 50 rds solution of a granulated sugar were prepared, and ten sets of readings were taken on the instrument. For each set of readings the sample cell was filled with fresh solution and the previous sample discarded. Data are given in Table 6.
The Δ values listed in the table are deviations from the corresponding averages. Standard deviations were calculated from the data. For the color index, $\sigma = 0.103$ and for the turbidity index, $\sigma = 0.145$.

7. *Effect of sugar concentration on indices.*

The absorbers of radiant energy in sugar solutions substantially follow Beer's law. Therefore the absorbancy index, (-log T) /bc, is constant for any concentration of sugar and independent of the refractive index of the solution. Sugar solutions were analyzed at different concentrations in the sphere photometer to determine the attenuation, absorption, and scattering indices. The data are presented in Figure 9, which shows that the absorption index, a, is constant, while the attenuation index, a*, and the scattering index, s, increase with decreasing sugar concentration. It is therefore necessary to report the sugar concentration at which the scattering index of a sugar solution was determined.

Table 6.—Performance Check With Sugar Solution

Test No.	F_x	F_y	Color Index	Turb. Index	ΔF_x	ΔF_y	A Color Index	A Turb. Index
1	.851	.164	18.04	4.75	-.0023	.0015	0.13	0.17
2	.852	.163	18.04	4.58	-.0013	.0005	0.13	0.06
3	.854	.162	17.88	4.45	.0007	-.0005	-0.03	-0.07
4	.853	.163	17.88	4.58	-.0003	.0005	-0.03	0.06
5	.854	.162	17.88	4.45	.0007	-.0005	-0.03	-0.07
6	.853	.162	18.04	4.45	-.0003	-.0005	0.13	-0.07
7	.852	.164	17.88	4.75	.0017	.0015	-.03	0.17
8	.855	.162	17.71	4.45	.0017	-.0005	-0.20	-0.07
9	.855	.161	17.88	4.29	.0017	-.0015	-0.03	-0.23
10	.854	.162	17.88	4.45	.0007	-.0005	-0.03	-0.07
Avg.	.8533	.1625	17.91	4.52				

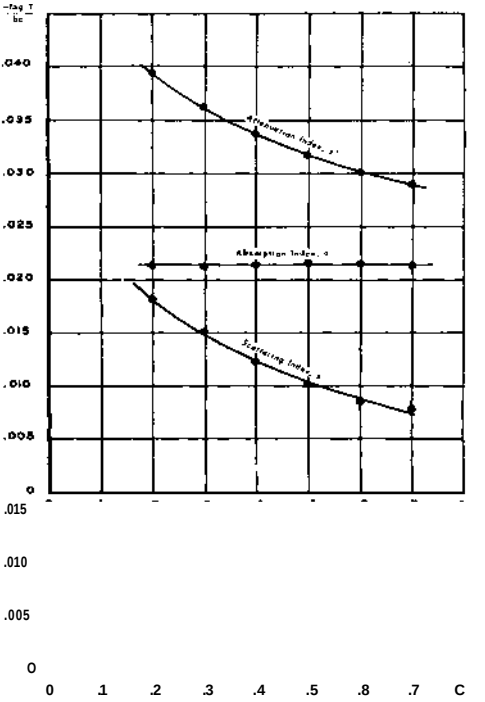

Figure 9.—Effect of sugar concentrations on indices.

Summary

Color and turbidity in solutions of white sugars may be accurately characterized by indices of absorption and scattering. To facilitate the evaluation of the indices, a new photoelectric instrument—the Sphere Photometer—was developed for the rapid measurement of light scattering and absorption. Test data are presented which show that the measured indices are reasonably accurate estimations of the optical properties of the sample solutions.

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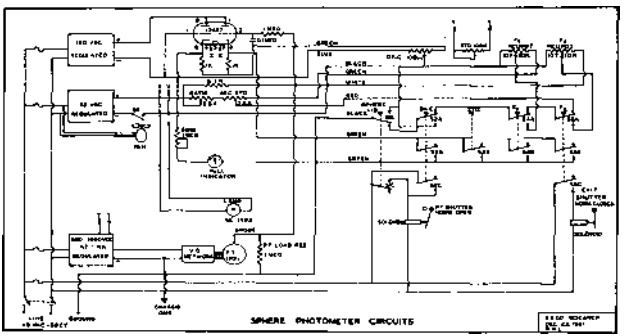
1 . **Appendix**

Electronic Circuitry.

A schematic diagram of the sphere photometer circuits is given in Figure 10, which shows the power supplies, light source, photo-
f tube, readout circuits, function switches, shutter solenoids and potentiometers necessary for the operation of the instrument.



The 150 VDC power supply furnishes plate voltage for the 12AT7 vacuum tube, employed as a bridge-type voltmeter. Regulation for this power supply is provided by an 0A2 gas regulator tube connected across the output.

The 6.3 VDC power supply furnishes power for the light source of the photometer, a GE No. 1493 lamp, for the filament of the 12AT7, and for the potentiometers used in calibration of the photometer and in the measurement of transmitted and scattered light intensity. This power supply is completely transistorized and has a regulation of ± 0.05% against line voltage changes of ± 10%. The power supply requires cooling, which is provided by a small fan blowing air over the terminal plate. With this power supply it is necessary to apply line voltage a few seconds before the load is connected. To provide this time delay, switch S6 — a vane-actuated Microswitch — was installed in the load circuit. When the instrument is turned on by closing of the line switch, line voltage is applied to the power supply and to the cooling fan. The air blast from the fan moves the vane on switch S6 and closes the load circuit. Since the fan motor requires a few seconds to reach operating speed, the time delay necessary for the operation of the power supply is assured.

The high voltage supply furnishes operating potentials for the 1P21 beam-multiplier phototube. The highly regulated out-

put voltage may be adjusted over a range of 100 volts by a variable resistor, the standardizing control on the instrument panel.

The phototube and its associated load resistor comprise the transducer which converts luminous flux to a proportional electrical potential. The amplification of the phototube, i.e., the ratio of output voltage to luminous flux, may be controlled by adjustment of the voltage applied to the voltage divider network connected to the tube. The standardizing control on the control panel on the instrument is connected to the high-voltage supply and serves in this function. The anode end of the phototube load resistor is connected to grid 2 of the 12AT7 vacuum tube through a filter network consisting of a 1 megohm resistor and a 0.1 mfd. capacitor. The filter protects the grid circuit against alternating currents which may be picked up by the wiring. The ground end of the load resistor is connected to the negative (-) lead of the 150-volt power supply of the 12AT7 tube through one of four switch-selected potentiometers. When the potential across the selected potentiometer equals that across the phototube load resistor and is of opposite polarity, grid 2 is at the potential of the negative lead. Since grid 7 is directly connected to this lead, it is at the same potential. As a result, the currents flowing through the two cathode resistors of the 12AT7 tube are equal and the two cathodes (3 and 8) are at the same potential. Thus no current flows through the null indicator and its needle rests in the zero position, indicating balance.

The push-button actuators are connected to multi-section switches which are electrically interlocked to prevent damage to the circuitry through operator error. On the circuit diagram the switches are labeled SI through S5. Switch sections with the suffix A select the potentiometer required for each of the functions. Switch sections with suffix B close the null-indicator circuit. Switch sections with suffix C actuate light-shutter solenoids, as required. When none of the push buttons are depressed, as during warm up or stand by of the photometer between analyses, the sample cell is outside the instrument and the light reaching the phototube is at its maximum. The potential across the load resistor is also at its maximum and, due to the position of switch section A, is opposed by the potential across the "Secondary Standard" potentiometer. The null indicator is inoperative, since switch sections B are open. When switch S3B is closed by depressing of the standardizing pushbutton, the null indicator will indicate any unbalance. Rotation of the standardization control, which adjusts the output of the high voltage supply, restores balance by matching of the potential across the load resistor to the potential across the "Secondary Standard" potentiometer.

Switch S2 performs the functions necessary for dark current compensation. When the push button is depressed, section A connects the dark current compensating potentiometer into the measuring circuit, section B closes the null indicator circuit, and section C actuates a solenoid which closes the shutter in the phototube housing. Rotation of the dark current control restores balances by adjustment of the voltage across the potentiometer.

Switch SI, connected to the lid of the photometer sphere, performs a similar function. When the lid is opened, section C energizes the solenoid and closes the shutter in the phototube housing, while section A connects the dark current potentiometer into the circuit. This interlocked function protects the phototube against excessive illumination and possible damage.

The "Dark Current" compensating potentiometer requires only infrequent adjustment because the dark current changes little during the life of the phototube.

The "Secondary Standard" potentiometer furnishes a reference potential which permits the standardization of the photometer in routine use without recourse to the primary standard.

The "Ratio" potentiometer permits the adjustment of the ratio of the potentials applied to the F_t and F_x potentiometers, i.e., the ratio of their effective spans.

The "Secondary Standard" and "Ratio" potentiometers are located inside the instrument case and are equipped with shaft locks to prevent accidental disturbance of the adjustments.

Transmittancies of sugar solutions are measured by manipulation of the F_t controls. When the push button is depressed, switch S4A connects the 10-turn Helipot potentiometer F_t to the measuring circuit and switch S4B closes the null indicator circuit. Balance is restored by rotation of the F_t control. At balance, the flux transmitted by the solution, F_t, is displayed in digital form on the control.

Scattering intensities of sugar solutions are measured by operation of the F_x controls. When the push button is depressed, switch S5A connects the 10-turn Helipot potentiometer F_x to the circuit and switch S5B closes the null indicator circuit. Switch S5C energizes a solenoid which opens the exit shutter in the sphere. When balance is restored by rotation of the F_x control, the magnitude of the scattered flux, F_x, is displayed in digital form.

A bank of four fuses for the protection of components against accidental overloads is located inside the instrument case. The instrument is connected to the electric lines through a three-conductor cable and plug, which provides grounding of the instrument case. A double-pole switch., located on the right side of the instrument, is used to turn the instrument on and off.

Effect of Incorporation Methods and Carrier Type of Endothal (TD-66) On Control of Weeds In Sugar Beets[1]

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Chemical control of weeds in sugar beets, particularly those weeds in the adjacent three to four inches of the row, has received considerable attention the last few years. However, the final results are not always easily predicted at the time the chemicals are applied. Part of this variability probably is due to the incorporation method and the type of carrier used for the herbicide. It is the purpose of this paper to report results of research on the above factors.

Studies of the mixing characteristics of various incorporation devices in 1960 (3)[3] indicated that the rototiller mixed the granular carrier the most uniformly into the soil. The finger weeder gave fairly uniform lateral distribution with a higher concentration of carrier near the surface of the ground than at the bottom of the operating depth of the fingers.

The distribution pattern of the rotary hoe showed heavier concentrations of carrier near the surface than at operating depth and in the vicinity of penetration of the tooth. The Sinner weeder (Figure 4), which consists of a row-crop ditcher shovel 6 inches in width with a spray nozzle or a granular distributor and covering blades mounted behind, caused the carrier to be concentrated on a strip 6 inches wide at the operating depth of the shovel (1 to 1 *1/2* inches). This strip is then covered with soil by the covering blades. The no-incorporation-front method resulted in some incorporation of the carrier by the furrow opener, the covering chains, and the press wheel. The carrier is applied in a band behind the press wheel for the no-incorporation-rear method.

Experimental Procedure

Methods of Incorporation

The effect of incorporation methods on control of weeds in sugar beets by the Endothal (TD-66) herbicide was studied at

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[3] Numbers in parentheses refer to references.

two locations in Wyoming during 1961—at Torrington, where the experiment was begun April 18, and at Powell, where the experiment was begun April 21.

The experiments were of the randomized-block design, with treatments replicated four times. There were 26 plots, 2 rows wide and 75 feet long, in each replication. One of the rows in each plot was treated and the other was not treated. The treatments were made up of 1. two herbicide formulations—spray and granular (30/60 RVM attapulgite, 2i/£% active ingredients); 2. two rates of application—1 lb and 2 lbs of active ingredient per acre, band basis; and 3. six methods of incorporation—rototiller (RT), rotary hoe (RH), finger weeder behind the planter (FWR), Sinner weeder (SW), no incorporation ahead of the planter furrow opener (NIF), and no incorporation, carrier applied behind the press wheel (NIR). Each replicate had two check plots.

The finger-weeder-rear (Figure 1) device was developed for testing during 1961 because the results secured during 1960 (4) suggested a need to study a method which would give shallow incorporation of the herbicide behind the planter unit to reduce the concentration of the herbicide around the sugar beet seed.

Figure 1.—Bottom view of the finger-weeder-rear incorporation unit. Figure 2 shows the unit mounted on the experimental planter.

Figure 2.—Equipment used for planting the two-row plots. The finger-weeder-rear incorporating unit is shown.

The various incorporation devices and planters (Figure 2) were mounted on a tool bar. One of the planters was used to provide for an untreated row between each treated row. The drive wheels of the two planters were connected by a flexible shaft to insure equal plate speeds for each planter. The planters were set to space the seed approximately 3 inches in the row. A University of Wyoming distributor (Figure 3) was used to distribute the granules, and spray nozzles were placed to give a 6-inch band of spray. Details of this distributor have been reported earlier. (2)

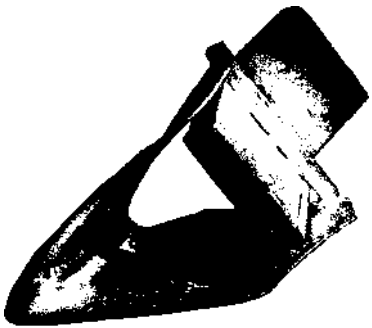

Figure 3.—The University of Wyoming distributor mounted behind the shovel of the Sinner weeder incorporation unit. See Figure 4 for a picture of the covering blades.

Figure 4.—The Sinner weeder incorporation unit. Figure 3 is the bottom view of the distributor and shovel.

Weed and beet population counts were taken when the sugar beets were in the 2- to 4-leaf stage of growth. The weed counts were taken from an area 20 feet in length and 6 inches wide, 3 inches on either side of the beet row. The plant population was classified as to (A) sugar beets, (B) broadleaved weeds, and (C) grass weeds. Grasses most commonly found growing in the sugar beet plot were green foxtail, *(Setaria viridis* Beauv.), barnyardgrass, *(Echinochloa crusgalli* (L.) Beauv.), and witchgrass *(Panicum capillare* L.). Broadleaved weeds consisted mainly of rough pigweed, *(Amaranthus retroflexus* L.), prostrate pigweed, *(Amaranthus graecizans* L.), lambsquarters *(Chenopodium album* L.), kochia, *(Kochia scoparia* L.), smartweed, *(Polygonum pennsylvanicum* L.), and wild buckwheat, *(Polygo?ium convolvulus).* Yield was determined by harvesting 10 feet of each treated row.

Granule Size and Type

The effect of granule size and type on the control of weeds in sugar beets by the Endothal (TD-66) herbicide was studied at the same two locations, Torrington and Powell.

The experiments were of the randomized-block design with treatments replicated four times. There were 22 plots, 2 rows in width and 75 feet long, in each replication. The treatments were made up of (A) five herbicide formulations—spray, 16/30 LVM (calcined attapulgite granules), 16/30 RVM (non-calcined granules), 24/48 LVM granules, and 24/48 RVM granules; and (B) two rates of application—1 pound and 2 pounds of active ingredient per acre, band basis. At the 1-pound rate, it is estimated that there would be one granule per .091 cubic inch of soil for 16/30 size granules and one per .0135 cubic inch of soil for the 24/48 size.

All formulations were incorporated to an approximate 1½-incli depth by a 6-inch width rototiller incorporation device. The equipment used for planting the sugar beet seed and metering the spray and granules was the same as described in the previous section. Weed and beet population counts and beet yields were taken with the same procedure described earlier in the section on the effects of incorporation methods.

The cultivation effect of the incorporation devices on weed control was not separated from the chemical effect in these studies.

Results and Discussion

Incorporation Methods

The results of the weed counts, beet-stand counts, and yield data for the various incorporation methods are shown in Figures 5 and 6[4]. The percent control was determined from the counts in the treated row compared with the untreated row in each **plot.**

The treatment effects for broadleaf-weed control, grass-weed control, and sugar beet seedling stand were statistically significant. In each case, a large portion of the treatment differences was accounted for by the treatment versus check and by incorporation versus no-incorporation effects. The 2-pound application rate did not result in better weed control or reduce the beet

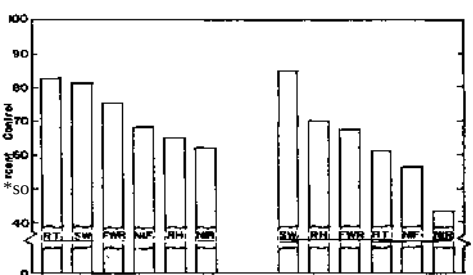

Figure 5.—Xhe percent control of weeds in sugar beets by Endothal for various methods in incorporation, Torrington and Powell experiments combined. See the footnote for an explanation of the statistical inferences.

* With reference to Figures 5, 6. 7, and 8, the values are placed in descending order from left to right. A break in the underline denotes statistically significant differences between the component underlined parts at the 5 percent level. For example, the SW, RH, FWR, RT method of incorporation resulted in significantly better broadleaved-weed control than the NIR method and the SW method was significantly better than the RT, NIF. and NIR methods.

SW stands for Sinner weeder, RH for rotary hoe, FWR for finger-wteder-rear, RT for rototiller, NIF for no incorporation, carrier applied in front of planter, NIR for no incorporation, carrier applied behind the planter press wheel, and CH for check plot.

stand more than the 1-pound rate. Over all, there were *no* differences between the granular and liquid carriers.

Analysis of the weed-control results indicated a distinct advantage for certain methods of incorporation of the chemical. The Sinner weeder (SW), which placed the herbicide in a layer 1 to H/4 inches below the surface, appeared to be the most effective method of placing the carrier of the chemical. Incorporation by the finger-weeder-rear method was effective for weed control and ranked well *on* the basis of the beet-seedling stand. This method appears to have promise where chemicals are used that have relatively close tolerances on the basis of toxicity to the crop. The above results are attributed to the fact that the shallow incorporation above the beet seed resulted in less toxicity to the beets and at the same time gave relatively good weed control.

On the basis of the results secured for the Sinner-weeder method of incorporation, it would appear that placing the herbicide in a layer, 1 to 1 $\frac{1}{2}$ inches below the surface of the ground, is an effective way of placing the carrier (either liquid or granular) of Endothal (TD-66).

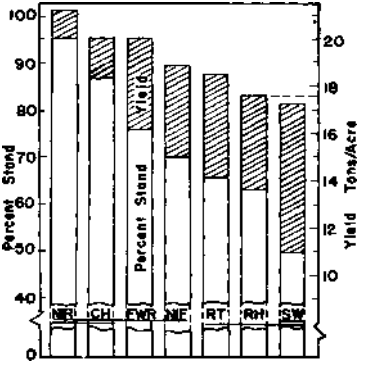

Figure 6.—The yield of sugar beets and percent sugar beet seedling stand for various methods of incorporating Endothal, Torrington and Powell experiments combined. The statistical data refer to the sugar beet seedling data. None of the yields for treated plots were significantly different than the yield of the check plots. See footnote for an explanation of the statistical inferences.

A complex statistical analysis on sugar beet yield data for Torrington and Powell combined (Figure 6) indicated that none of the treatment yields was significantly different than the yield of the check plots. However, the yield lor the Sinner-weeder treatment was significantly less than the yield of the check plots at Powell. Figure 6 suggests a decrease in yield with the lower sugar beet seedling stands even though the yield differences were not significantly different.

Granule Size and Type

The results of the percent control grass and broadleaved weed for various types of carriers for Endothal (TD-66) are shown in Figure 7 and the results of the beet-seedling counts and the sugar beet yields are shown in Figure 8.

The percentage weed control was not significantly different between the 1-pound and 2-pound rates, although the counts showed fewer weeds for the 2-pound rate.

The treatment effects for broadleaved and grass-weed control and sugar beet seedling stand were statistically significant; however, most of the treatment effect was attributed to the treatment versus no-treatment comparison.

The differences between treatment due to granule sizes and type, and granules versus spray, were not statistically significant on the basis of sugar beet yield, sugar beet seedling stand, or weed control.

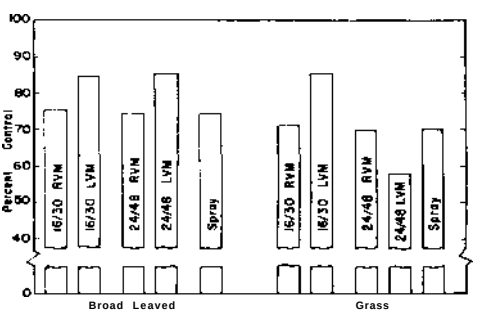

Figure 7.—The percent control of weeds for different sizes and types of granular formulations of Endothal and for spray formulations of Endothal, Torrington and Powell combined. The treatment differences were not statistically significant.

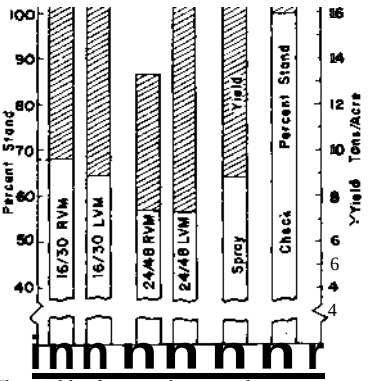

Figure 8.—The yield of sugar beets and percent sugar beet seedling stand for different sizes and types of granular formulations of Endothal and for spray formulation of Endothal, Torrington and Powell experiments combined. The treatment differences were not statistically different.

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Bulk Sugar Storage - Weibull Silo

ALLAN WOODS[1]

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Introduction

Storage of large quantities of bulk sugar involves a great many problems both with respect to the handling of sugar in the plant and until it finally reaches the consumer. This paper has been prepared to show the methods and some of the results obtained by Union Sugar utilizing a Swedish design Weibull silo.

Being located near the coast and subject to frequent fogs and winds, the climate at Betteravia is extremely variable. The relative humidity will vary from 40 to 100% within a 24-hour period and frequently will change this much in six hours. Storage and handling of sugar under such varying conditions has dictated a search for means of storing sugar in a more favorable atmosphere. The Weibull silo was chosen as a means of storing sugar under constant temperature and humidity conditions without influence of ambient air changes.

Silo Construction and Design

The silo was designed to hold 20,000 tons (400,000 cwt.) of refined sugar. The main structure is a steel shell 116 feet in diameter by 82 feet high to the eaves, resting on a flat concrete base and containing a central tower 1 1 feet in diameter. The installation is insulated and waterproofed externally, thus providing a virtually airtight enclosure. The maximum height of the sugar below the reclaiming mechanism is 74 feet.

Filling and emptying the silo is completely automatic in the sense that the operator has only to set and adjust the controls periodically. Sugar enters and leaves the silo through the bottom conveyor which is reversible. Sufficient interlocks are provided so the handling mechanism must be started and operated in the correct order. Panel board lights are provided to assist the operation.

Complete erection including foundations, steelwork, insulation and equipment placement was contracted to the Chicago Bridge & Iron Company. Foundations and steelwork were redesigned by them to conform to American erection methods and all materials, excluding the sugar reclaiming mechanism and air conditioning machinery, were obtained on the west coast.

Description of Silo Internals

The center of the roof is supported by a central tubular steel tower 11 feet in diameter extending from below the floor through the apex of the silo. The tower contains **a** bucket elevator, a

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manlift, central air ducts and electrical panels. Radial trusses from the tower serve to stabilize the upper part of the shell, like spokes of a wheel.

The main floor of the silo consists of two layers of concrete 5 inches and 7 inches thick separated by special-shaped corrugated, galvanized iron sheeting to allow passage of air underneath the floor. The steel sides are covered with the same type of corrugated, galvanized iron sheeting to form air ducts up the outer wall. Three inches of fiberglass insulation with aluminum sheeting on the outside serve to insulate the sides. Two-inch holes in the top and bottom of the steel plate sides allow the passage of air from underneath the floor, up the outside walls and into the 15-inch space between the aluminum sheathing beneath the radial beams in the roof and underneath the $^{1}/_{8}$-inch steel roof base plating. The outer surface of the roof is insulated with three inches of stiff fiberglass board and covered with asphalt roofing-paper.

The Silo Bridge

Supported on a rail around the top of the shell and the central tower is a large radial bridge which continuously rotates while the silo is being filled or emptied and provides the means whereby the sugar surface is kept level. The bridge carries two circular spreading mechanisms for sugar distribution and a winch for raising and lowering the reclaiming screw which is suspended below it. Based on two welded plate girders, 52 feet in span and suitably cross braced, the bridge is mounted at the inner end on single flanged wheels set parallel with its radial axis on either side of the central tower. The outer end runs on four double flanged wheels. The wheel axles at opposite ends of the bridge, being at right angles to one another, correct for errors in concentricity of the tower and the shell rail track.

The electrical supply to the bridge is through protected bare copper sliding contactors.

Platforms running the full length of the bridge on either side and around the tower give full access to the distribution mechanism and the electrical panel containing the various motor starters.

Silo Temperature and Humidity

Constant temperature and humidity within the silo are attained by constantly circulating warm or cold air in the shell around the sugar in storage and by removing the moisture from the air above the stored sugar. Either warm or cold air, depending on the temperature above the sugar, flows underneath the floor, up the side ducts and through the air space in the roof

of the silo at all times. The direction of the flow is reversed every twenty minutes by an automatic timing device. The volume of air, approximately 10,000 C.F.M., is heated or cooled by circulating water radiators located in the ductwork at the base of the silo.

The relative humidity is regulated by means of a dehumidifier located in the top of the central tower. A constant stream of air is moved from the space above the sugar through a dust collector and refrigeration coils and recirculated back into the silo. A hygrostat located within the silo chamber regulates the operation of the two-ton coil unit.

The air above the sugar may be changed at any time by opening vents in the silo roof and allowing air from the circulation system to be released directly into the space above the sugar. Simultaneously, vents are opened at the apex of the roof to allow the internal air within the silo to be exhausted into the atmosphere. The fresh air intake for the circulatory air system is located above the highest point of the silo roof.

Sugar Handling to and from Storage

Sugar from production is screened, the minus 60-mesh sugar separated out and sacked each day. The balance of the sugar is conveyed to the silo by means of a screw conveyor underneath the temporary storage bins to a 20-inch, white-rubber belt conveyor. From the belt conveyor the sugar is elevated by a bucket elevator and discharged onto a circular turntable where it is ploughed off into chutes leading to two revolving distributors. The purpose of the distributors is to evenly distribute the sugar within the central tower as the bridge slowly revolves. Most of the dust is carried downward by the thin conical stream as it descends.

Filling the silo by sprinkling in a manner resembling falling snow, as distinct from normal pouring method, produces two important effects: The in-going sugar is cooled in air by spreading it thinly over a wide surface; compacting is reduced by allowing each crystal to rest where it falls without sliding.

Sugar is removed from the silo through twelve 6-inch X 18-inch openings located at the base of the central tower. The rate of discharge through these openings is regulated by sliding gates into chutes extending down to the face of the lower revolving turntable in the base of the central tower.

Sugar discharged from the lower turntable may be either recirculated or withdrawn for shipment by reversing the 20-inch belt conveyor used to transport the sugar into the silo. From the belt conveyor the sugar is discharged to an 18-inch screw

conveyor where it may be sent to either the rotex screens or shipped direct. The reclaiming screw within the silo is used only to keep sufficient sugar in supply next to the central tower or to level the sugar in the silo during filling.

Silo Operation and Sugar Quality

With the removal of the minus 60-mesh sugar from the silo feed, the dust problem is considerably diminished. The remaining dust is removed continuously. Other than during the period of filling, the air above the sugar is clear. The small volume of dust removed is transported from the collector base by vacuum lines to the main floor of the packing room where it is sacked periodically for remelt.

As a minimum of equipment is required to handle the incoming and outgoing sugar, breaking of crystals is negligible (as shown in Figure 1). Differences observed are not significant at 19:1 odds (l)[2].

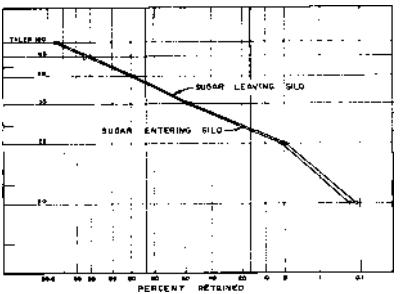

Figure 1.—Composite screen analysis of sugar entering and leaving silo.

The loss by attrition due to the reclaiming has not been significant and to date, all sugar flowing out of the base of the silo has been very free-flowing. Only occasional poking or probing of the outlets has been required. All handling screws are double flight screws to insure an even flow at all capacities and are of ample capacity.

During the first season of operation, sugar was recirculated once every eight hours to keep the sugar next to the center column in a free-flowing condition. This practice has been discontinued and we now depend on normal shipment withdrawals to keep free-flowing sugar in this area. No sugar is circulated over the weekends during interseason.

[2] Numbers in parentheses refer to references.

Color

During any given period, no significant difference can be noted between the color of the incoming sugar and the sugar stored in the silo. Table 1 shows a small difference in the average color analysis of the air slide cars in comparison to the average of the campaign analysis of each strike. This is due largely to the fact that more than ten times as many analyses were run during the year on the campaign samples as on the shipment samples and any variation in the shipment color would be magnified to a greater extent.

Table 1.—Comparison *oi* sugar colors, sugar produced and sugar shipped.

	1961 Imperial	1961 Coastal
Sugar to Silo	91.9	92.8
Sugar Shipped	**91.4**	**91.3**

Sugar color expressed as % T of 50% solution, 50 mm light path at a wave length of 425 inu; instrument standardized against distilled water.
Color of sugar to silo is average value of all sugar introduced to silo.
Color of sugar shipped is the average of sugar colors from air slide shipments.

Sugar Temperature and Moisture

The temperatures of the incoming and outgoing sugar are shown in Figure 2. The incoming sugar temperature is governed by temperature of the sugar leaving the granulators. No special cooling is provided. During the filling of the lower half of the silo, the sugar loses about 15° F by falling through the air and being distributed over the large sugar surface in the silo. This temperature difference is gradually reduced as the silo fills.

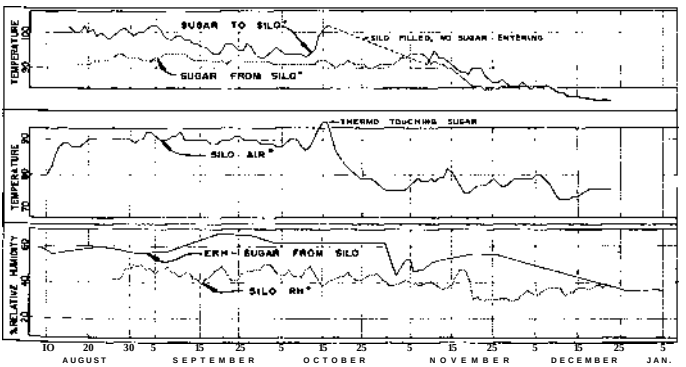

Figure 2.—Temperature of incoming and outgoing sugar.

At the close of the season, the sugar temperature drops slowly as the air temperature in the air space above the sugar is lowered to between 60° and 70° F. The upper limit of the hygrostat controlling the relative humidity of the air above the silo sugar is set at 53%. The average relative humidity of the air in the space ranges between 40 and 50%. This low relative humidity has a slow drying effect on the sugar in storage as evidenced by the difference in moisture between the sugar entering and as shipped. The average moisture content of the entering sugar during 1961 was 0.016% and the average moisture content of the shipped sugar was 0.009%.

During the past season, equipment was purchased and the "Equilibrium Relative Humidity Values" of the stored sugar as described by McGimpsey and Mead (3), were taken periodically.

These values did not change greatly during season when the silo was constantly being filled. A sharp drop was shown during the period when no sugar entered the silo and the sugar was held in storage. Further results are being taken to continue these studies in hopes of finding out more about what actually takes place during sugar storage.

Conclusion

The storage of sugar through the use of a Weibull silo has provided an economical means of handling bulk sugar with the minimum of equipment and labor. The sugar held in this manner has been completely free-flowing at all times, sparkling in appearance, with a very small amount of crystal breakage.

While we do not feel this to be the final answer to the handling of sugar in bulk, we do feel this method of storage presents a significant advance in the field of bulk sugar storage from the standpoint of maintaining sugar quality at low handling costs.

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Trends in Sugar Beet Planter Design in Colorado[1]

R. D. BARMI.NGTON[2]

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Introduction

Progress in the development of planting equipment continues to help improve the competitive position of the sugar beet orower in the world sugar market. Monogerm seed is now playing an important part in the rapid reduction of thinning and labor costs, previously required to produce this crop.

Problems still exist, however, in the use of monogerm seed. The major problems are to correctly space the seed in the row and to place it in the soil so that it will germinate, thus producing only evenly spaced, vigorous single plants.

The evolution of sugar beet planter design is a slow process and does not always keep pace with other scientific achievements. When a new advancement such as monogerm seed is suddenly presented, planting equipment may not be capable of making the best use of potential benefits contributed through plant breeding techniques.

Two General Patterns of Development

In general, improvements in sugar beet planters have come about in two ways. First, through refinements in existing designs and manufacturing procedures and second, through the development of new ideas and new principles.

In recent years very few completely new ideas have reached the manufacturing stage and eventually, the beet grower. However, modern planters are doing a much better job in the field than they did 10 years ago. Much of this improvement is due to careful seed polishing and seed sizing to narrow size limits for a given seed lot. Accurate seed sizing is practiced by most sugar companies and is giving the engineer something specific to work with. The combined efforts have resulted in a remarkable improvement in field results.

The engineers' contribution to improved field results using polished and sized seed has been in the areas of planter design and manufacturing refinements. These refinements include machined hopper bottoms, carefully fitted plates, improved seed cutoff and knockout mechanisms, elimination of substandard parts, careful assembly, and matching plate thickness and cell size to fit a specific size of seed.

[1] Colorado Agricultural Experiment Station Scientific Series Paper No. 762.

[2] Associate Agricultural Engineer, Colorado State University. Fort Collins, Colorado.

Field Emergence Is Still a Serious Problem

With all the improvements and refinements in present design and manufacture, the vast problem area of seedling emergence remains. If all the viable seeds planted would produce a healthy vigorous plant, truly great strides could be made in eliminating spring labor costs. It appears that a solution to this problem will require a departure from standard planter design and accepted field planting practices. In an attempt to find a solution to this problem, engineers at Colorado State University have re-examined a design first built and tested at this station in 1948 (l)[3].

This planting unit design consisted of a solid steel wheel having a "V" shaped rim which pressed a furrow into the seedbed one and one-eighth inches deep ahead of the seed drop. A small shoe made to fit the shape of the furrow followed immediately behind the wheel. The shoe, which was set slightly deeper than the furrow, trowelled the bottom of the furrow and at the same time held the loose soil out while the seed was deposited at the rear of the shoe on a compacted furrow bottom. Field tests at Colorado State University and some other beet-growing areas (2) have consistently shown outstanding improvement in emergence from this system of depositing seed in the soil.

Field tests in the Rocky Mountain area have shown that some type of device to fill the seed furrow (covering device) followed by high-unit-pressure surface packing is desirable (3). Properly adjusted cover blades are more effective than cover chains for filling the seed furrow- and narrow, relatively firm tires are more effective than wide, soft tires for packing the soil. Although surface packing of the soil by the planter presswheel has proven to be highly beneficial in the Rocky Mountain area, it is not always true in other beet-growing areas. Tests reported by Stout, Buchele, and Snyder (4) in Michigan, showed that in the laboratory, seedling emergence was impeded by high surface pressure when there was adequate soil moisture for germination or where water was sprayed on the surface to simulate rain after packing.

Renewed Interest In An Old Idea

Because field tests in Colorado have produced overwhelming evidence that packing the soil below the seed improves seedling emergence and vigor, this principle (developed in 1948 at Colorado State University) has been re-examined. Two machines using the "V" shaped wheel previously described have been built. One machine which will be referred to as the CSU planter was designed and a field model fabricated in the CSU shop. The other machine will be referred to as the CSU-J.D. and was a re-

[3] Numbers in parentheses refer to literature cited.

design of the John Deere #70 flexi-planter incorporating the "V" shaped wheel for sub-surface soil firming.

The Experimental CSU Planter

The CSU planter was a unit type machine designed with three objectives in mind, namely: (1) gentle handling of the seed for minimum seed damage, (2) firming the soil below the seed, and (3) positive covering of the seed with moderately high unit pressure applied by the presswheel. Figure 1 shows the complete planting unit as it was used in the 1961 field test.

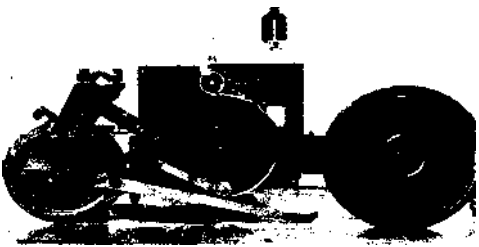

Figure 1.—Experimental CSU planting unit showing the tool bar mounting linkage, wheel for pressing the seed furrow into the soil, seed shoe, cover knives, presswheel, and seed metering wheel with rotating seed cutoff.

To accomplish these objectives, a 14-inch diameter vertical seed plate was used with an auxiliary seed chamber and rotating seed cutoff. This seed metering system eliminated practically all seed damage. Firming of the soil below the seed was done with a solid cast iron wheel fourteen inches in diameter with a "V" shaped rim. Integral depth bands on the wheel permitted the "V" to be pressed into the soil one and one-eighth inches leaving a compressed groove. A steel seed shoe which fits the shape of this groove was run behind the wheel and slightly deeper than the groove, to hold loose soil out while the seed was deposited at the rear of the shoe. After the seed was covered with cover blades, the soil was packed by a standard A.S.A.E. 2 x 20-inch presswheel with a zero pressure rubber tire on the rim. The presswheel was also used to drive the planting mechanism.

The CSU-John Deere Planter

The CSU-J.D. planter was designed to incorporate the soil firming principle into the basic John Deere #70 flexi-planter unit. The mounting linkage, seed hopper, and presswheel were essentially unchanged. The problem was to adapt to the #70

series planter, the furrow forming wheel and shoe which Deere & Co. had, at one time, made available as optional equipment for the #64 and #66 series planters. Figure 2 shows the completely converted planter unit. The conversion involved several operations which were rather difficult in a research shop but would be comparatively simple for a manufacturer starting with such a design in mind.

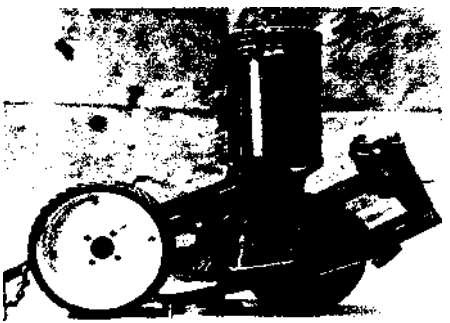

Figure 2.—Completely converted CSU-J.D. unit showing the method of converting the John Deere Furrow Forming system to a unit planter similar to the J.D. #70. The basic wheel and seed shoe was supplied as optional equipment for the John Deere #64 and #66 beet planters.

Other Planter Designs Worthy of Mention

New ideas are frequently presented by inventors and most of these new designs offer principles worthy of careful consideration. A machine using a vacuum seed pickup principle built by the Silver Engineering Company has been used. Another machine tested had a rotating seed hopper in which seeds were mechanically pushed through cells in a rubber membrane and was built by G. E. Ferguson of Forsyth, Montana. A machine built by Elmer Bergh of Harlem, Montana, incorporated a rotating seed plate with a valve mechanism to trigger an air ejection device for positive placement of seed in the furrow. A planting mechanism built by George Walters of The Great Western Sugar Company has an inclined rubber belt for metering seed. This arrangement requires no seed cutoff in the hopper which eliminates seed damage at this point. A planter constructed by J. P. Freitus of Longmont, Colorado, placed a soluble tape in the ground in which the seed had previously been placed.

Although laboratory and field tests have indicated that these machines are not ready for commercial use, they all offer principles of real value to the sugar beet industry if the design, construction, and cost can be worked out to make them competitive.

Foreign Planters

More planter designs are offered to the European beet grower than to his American counterpart. From time to time European planters have been tested at Colorado State University. The Stanhay planter from England, Taxigraine from France, and Sernora from Switzerland have been included in these tests. These planters all have some interesting features but are not currently being used in the United States.

Discussion of Field Results

Some of the planters producing the earliest and most uniform stands of vigorous plants in the 1961 tests, did not show the highest percentages when the stand counts were made just prior to thinning. This was due to extensive weather damage in the form of hail, flood, and freezing which occurred three weeks after planting. One field in the Windsor district was particularly outstanding with respect to differences between planters, before the severe storm of May 12 and 13. The Great Western Sugar Company fieldman in this district reported outstanding results from the experimental CSU planter. Where this planter was used the seeds germinated quickly producing uniform stands of vigorous plants before seeds from other planters had germinated at all. This was also true in other fields where the CSU and the CSU-J.D. planters equipped with wheels for packing the soil below the seed, showed spectacular improvement in rapid seed germination. Most of the fields were planted April 19 to 22, 1961. The next three weeks were dry and windy followed by exceptionally heavy precipitation in the form of hail, rain, and snow with freezing temperatures. Germination from conventional planters was spotty or none at all during the first three weeks after planting. However, when stand counts were made at thinning time some of the conventional planters produced higher plant population because the plants were not so severely damaged by the bad weather.

The practice of counting stands just prior to thinning was not entirely satisfactory as a measure of planter performance because it measured plant survival rather than emergence. A better method would be to make several stand counts in the same location *in* the row starting when the first plants were visible and continuing until the beets were thinned. Limited time and labor

made such a procedure impossible so the data shown in Tables 1 and 2 were taken just before the beets were thinned.

Even with the stand reduction caused by weather, Table 1 shows the CSU planter to be relatively high among the planters in number of plants at thinning time and in percent singles.

Table 1.—Results from 1961 sugar beet planter field tests using Great Western No. 2 bare polished monogerm seed.

Planter	Percent pre-thin stand	Percent singles	Percent cell fill
Planet Jr.	57.02	84.94	107.68
C S U	56.18	92.53	110.86
I H C	51.67	92.52	102.36
C S U - J D	51.96	91.49	101.46
J D #70	50.28	93.63	104.38
Milton	45.13	90.37	119.45
Avg.	52.04	90.91	107.69
L. S. D. 5% level	5.53	2.26	3.93
1% level	7.16	3.04	5.28

Table 2.—Season averages for three commercial planters* reflecting influence of polishing and sizing seed.

Year	Seed Used	Percent pre-thin stand	Percent singles	Percent cell fill
1959	Screened 8- 10/64"	61.02	83.70	111.70
1960	Polished 7- 8/64"	62.62	91.87	104.25
L. S. D. 5% level		7.37	3.12	8.50
1% level		10.00	4.24	11.53

*Planters used were: John Deere #70, International #185 and Milton.

Table 2 shows the effect of polishing and sizing the seed between narrow limits in 1960 compared to broader limits in 1959. As would be expected, polishing and sizing did not materially affect germination but did improve the percentage of single plants in the row. Statistically, the cell fill was border line. However, experience with the machines in the laboratory and the field indicated that vast improvements in accuracy and reduction in seed damage were achieved by seed processing and careful sizing.

Conclusions

Field tests conducted at CSU from 1959 through 1961 have shown the value of three things. First, careful cleaning and adjusting of any planter is essential for realizing the greatest potential benefits which have been designed into the machine. Second, seed preparation and narrow size limits combined with matching planter hopper parts has contributed more to the high percentage

of single plants in the row than anything in recent years. Third, packing the soil below the seed with good soil coverage over the seed and firm presswheel action has shown the greatest benefits from the standpoint of vigorous seedling emergence.

With only 50 to 60 percent of the planted seeds producing plants at thinning time, it is evident that more developments are necessary in the area of emergence before a major breakthrough can be made permitting growers to plant to a final stand.

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The Sugar Beet Nematode, *Heterodera schachtii* Schmidt, in Southern Alberta[1]

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The sugar beet nematode, *Heterodera schachtii* Schmidt, is a serious pest of sugar beets in Europe (2)[4] and is present in 15 beet-producing states of the U. S. A. (1). The plant parasite was first discovered in Canada in 1931 near St. Catharines, Ontario (4). In 1939 it was found on sugar beets near Sarnia, Ontario (3).

In June 1961, an unthrifty stand of beets 13 acres in size was found near Taber, Alberta. The plants in approximately one-quarter of the field were severely stunted, the leaves were badly wilted, and there was considerable root proliferation (Figures 1 and 2). Numerous white cysts were found on the roots of the stunted plants as well as on the roots of other sugar beets throughout the field (Figures 3 and 4). Cysts were also found on flixweed, *Descurainia sophia* (L.) Webb, and on oak-leaved goosefoot, *Chenopodium glaucum.* L., in the same field. The cysts were identified as *H. schachtii* by Dr. A. D. Baker and R. H. Mulvey of the Nematology Section, Entomology Research Institute, Ottawa, Ontario.

Although beets were first grown commercially in Alberta in 1903 and have been grown since 1925 in the district where the infested farm is located, this was the first time the sugar beet nematode had been found in western Canada.

Until 1950 the infested field was flood irrigated but since that time it has been sprinkler irrigated. The area where damage was evident in the field was previously a knoll that had been levelled. The farmer had noted stunting of the beets in this area in 1957, which suggests that the infestation may have been present for at least 4 years.

Although the average yield on this farm was generally higher than that of the surrounding area, the farmer used a very short cropping sequence, in which he grew beets in 10 of the last 17 years. This sequence would be expected to favor a rapid increase in the numbers of nematodes once the field became infested.

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[4] Numbers in parentheses refer to literature cited.

Figure 5.—(lower left) Photomicrograph of a cyst of *H. schachtii* opened to show the eggs (X30).

Figure 6.—(lower right) Enlargement of the nematode eggs shown in Figure 5 (XI30).

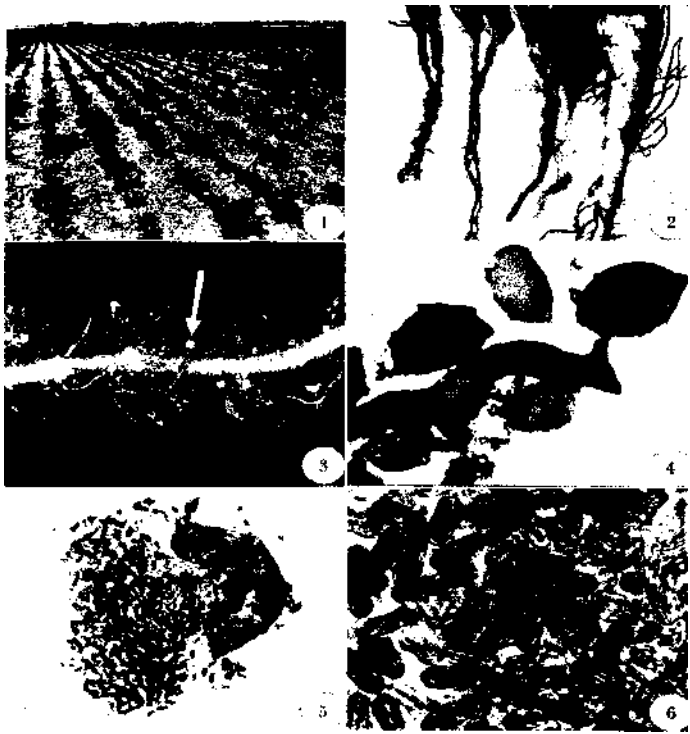

Figure 1.—(upper left) Sugar beet field near Taber, Alberta, severely infested with *Heterodera schachtii* Schmidt. The beets were wilted, stunted, and chlorotic.

Figure 2.—(upper right) Comparison of a normal beet (right) with three beets severely stunted by *H. schachtii.* "Hairiness", exhibited by the beet at the left, is often indicative of the presence of the sugar-beet nematode.

Figure 3.—(center left) A portion of a heavily infested beet. Arrow points to one of the cysts (X10).

Figure 4.—(center right) Photomicrograph of one of the secondary roots of a beet with adhering lemon-shaped cysts of *H. schachtii* (X30). Cysts ranged in color from white to brown.

Survey

In early July, sugar beets and soil from the most unthrifty areas of 721 sugar beet fields throughout southern Alberta were examined by the authors in the laboratory for cysts of *H. schachtii.* No other infestations were discovered, [ones (2) found, in England, that with a population of cysts under one million per acre there were no crop symptoms and the infestation was not detectable. It is possible, therefore, that there may be light, undetectable infestations of the nematode in southern Alberta.

Some beets examined during the survey had an abnormally large number of lateral rootlets. In most cases this abnormal growth appeared to result from damage by the sugar-beet root aphid, *Pemphigus betae* Doane, the sugar-beet root maggot, *Tetanops rnyopaeformis* (Roder), or the wireworms *(Ctenicera destructor* (Brown) and *Hypolithus bicolor* Esch.

In the infested field, soil samples taken from around beets contained 135 cysts per 200 grams of soil. Both old and young cysts were present in July, the latter full of eggs and second-stage larvae (Figures 5 and 6). The presence of old cysts and the degree of infestation indicated that this pest had probably been present in the field for more than one year.

On July 14 several small beets from the infested field were lifted with adjacent soil and planted in 6-inch pots in a greenhouse. Approximately 170 days later one 100-gram sample of soil was taken from an area immediately adjacent to the beet in each of 8 pots. The average number of cysts obtained from the soil samples was 1,192.

Control measures

The ability of *H. schachtii* to increase rapidly and spread made it desirable to reduce this infestation as quickly as possible. The field was plowed and fumigated on August 14 by applying approximately 25 gallons per acre of the nematocide Shell DD at a depth of 6 to 8 inches. Forty-five days later beets were planted in 8 pots containing soil from the fumigated field. Approximately 95 days after planting, the pots contained an average of 170 nematodes per 100 grams of soil. Although the nematocide appeared to greatly reduce the number of nematodes in the field the residual population could still cause serious damage to beets.

It was recommended that alfalfa, which is not a host of *H. schachtii,* should be grown on this land for at least 6 years and that on the remainder of the farm sugar beets or other susceptible crops should not be grown oftener than once every 4 years. To prevent serious infestations of this pest from developing in sugar-

beet-growing areas of Alberta, officials of the sugar beet growers' association and the Canadian Sugar Factories Limited have agreed to adhere to these recommendations and also to a general recommendation that susceptible crops should not be grown oftener than once every 4 years.

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Selection for Low and High Aspartic Acid and Glutamine in Sugar Beets

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In recent years there has been an increasing widespread and copious use of nitrogen fertilizer in sugar beet production. In most cases this has resulted in an increase in beet yield along with low sucrose content. Impurities "or nonsugars" have increased, causing a low extraction of sugar per ton of beets and an increased production of molasses. The increases of amino acids and of other nitrogenous compounds in the beet are the major factors contributing to the reduction of the quality of beet juice for sugar extraction.

The objectives of the present investigations were: To determine if certain amino acids could be increased or decreased by ordinary mass selection; to ascertain if these selections reacted the same under different nitrogen levels of fertility; and to determine how these selections affected other chemical compounds in the beets.

The classical protein and oil selection experiments on corn conducted at Illinois have demonstrated that chemical composition of plants was in part under genetic control (10)[2]. Selections in sugar beets for high and low quantities of chemicals such as sucrose, sodium, potassium, galactinol, raffinose, and purity have been successful as determined by progeny tests. Many investigators (1, 2, 3, 4, 17, 19) have applied selection pressure for low sodium content of individual roots. All have shown that the sodium content was significantly correlated with sucrose but in a negative relationship. All have shown by progeny tests that significant reductions or increases in sodium content could be accomplished by mass selection. Wood (17) reported on progeny tests of roots selected for high and low ramnose content. He concluded that the ramnose content of beets could be significantly reduced by mass selection. Later Wood et al. (18) studied the inheritance of raffinose production in sugar beets and reported that the number of effective factor pairs involved in the oroduction of raffinose between the two parents used was about five; at least one was isodirectional and all were equal in magnitude. In the crosses studied neither dominance, nor heterosis, nor linkage appeared to be involved. Quantitatively, the factors for ramnose production in the two parents followed an arithmetic

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[2] Numbers in parentheses refer to literature cited.

scale and consequently were additive. Finkner et al. (4, 6) also studied beets selected for high and low raffinose content. They found through progeny tests that these selections bred true for high or low raffinose content. They also studied these selections under different harvest dates and storage conditions. Again it was found that the high selections remained high and the low selections stayed low.

Powers et al. (14) summarized much of the recent data concerning selection for high and low sodium and raffinose contents. They also reported on selections for thin juice purity and sucrose content. The data for thin juice purity showed that selection for greater purity has resulted in an increase in this character.

Agricultural scientists are aware that the soil has long been recognized as the basis of agricultural production. In the elucidation of the chemodynamics of soil plant complex it has been shown by several investigators on several crops that the addition of fertilizer to the soil can change the chemical composition of plants. Hac et al. (7) and Walker et al. (15) studied the effect of nitrogen fertilizer on the glutamic acid content in sugar beets. They found that the application of nitrogen fertilizer caused an increase in glutamic acid of beets. Walker and Hac (16) observed that as the soil moisture increased under both furrow and sprinkler irrigation, nitrogen fertilizer increasingly stimulated yield and glutamic acid content of beets. Haddock et al. (8) showed that several nitrogen constituents, especially glutamine, increased with nitrogen fertilizer applications. Finkner et al. (5) presented data using three different levels of nitrogen applications and found that the amino acid content of the beets increased as the rates of nitrogen increased. In a study of nine different amino acids and the total amino acid content Finkner et al. (5) showed a significant linear increase response to nitrogen application.

Payne et al. (11, 12) made population genetic studies pertaining to nitrogen compounds in sugar beets and concluded that varieties of beets could be bred which would contain lower amounts of nitrogen constituents even when grown on high fertility soils.

Materials and Methods

The variety used in this experiment was SLC 24, a self-sterile monogerm. A total of 2,272 beets was selected in 1958 by the unit block method similar to that developed by Powers (13). The selection unit block was 35 feet long and 11 feet wide. It differed from Powers' method in that no inbreds or F, hybrids were planted to measure the environmental variation.

Each individual beet was weighed, sampled and analyzed for sucrose, aspartic acid and glutamine. The number of roots selected from each unit was recorded and the range and mean calculated. The standard deviation of each unit block was estimated by utilizing the formula, Range/s = mean ratio, a short-cut method described in Snedecor's "Statistical Methods", 5th Edition, Table 2.2.2, page 38.

Based on these estimated standard deviations, selections were made within the unit block for beets which were higher or lower than the block mean for aspartic acid and glutamine. Beets selected for high aspartic acid were at least twice the standard deviation higher than the block mean and the beets selected for low aspartic acid were at least 1.3 times the standard deviation below the block mean. The selection deviation values used for glutamine were 2.3 times the standard deviation for the high selection and 1.1 times the standard deviation below the block means for the low selection. A random selection of approximately every 25th root from the total population was saved and considered the check.

Although sugar and weight were recorded for the individual roots, selections were based on the amount of the two amino acids. The amino acids were determined by a paper chromatographic procedure reported by Hanzas (9). All paper chromatographic determinations are reported as percent on dry substance.

The number of roots selected for each group, the pedigree numbers, the general means for each character studied for each group, and for the entire population, are shown in Table 1.

Table 1.—Means of weight, chemical characteristics and number of roots for five amino acid selections.

Pedigree number	Character	No. beets selected	Individual root data			
			Wt. Lbs.	% Suc.	% Asp. A.	% Gluta.
59-407	Low Aspartic Acid	59	2.7	11.0	.10	.14
59-408	High Aspartic Acid	81	2.7	11.0	.44	.87
59-409	Low Glutamine	68	2.7	11.4	.15	.14
59-410	High Glutamine	81	2.6	11.3	.39	1.54
59-411	Random Selection	84	2.4	11.1	.22	.45
Entire Population (2,272 Beets)			2.5	10.8	.22	.44

From the above table it will be seen that the high and low selections for aspartic acid and glutamine were greatly different, but in weight and sucrose percents they were similar.

Roots of each of the five groups were space isolated in the spring of 1959 and produced seed that fall. Tn 1960 the five seed lots were planted in a split plot replicated test at Rocky Ford, Colorado, and at East Grand Forks, Minnesota. Tn these

tests the three main plots were the levels of nitrogen, and the selections were the subplots. The plots were single rows with a commercial variety planted on each side to give uniform competition for each selection. The selections were replicated six times in each test.

Plots were harvested for weight, sucrose and other juice quality characters. Paper chromatography was used to determine amino acids, total amino acid, galactinol[4], and raffinose. Total nitrogen was determined by a modified micro-Kjeldahl nesslerization (11)- Sodium and potassium were determined by the flame spectrophotometer. Sugar and purity were analyzed by standard sugar analysis procedures.

Experimental Results

Remarkable differences were obtained in the progeny tests of the five amino acid selections. Very reliable differences between the high and low amino acid selections were obtained for all characters tested at one or the other, or both locations, except for sucrose percent. The results of these progeny tests under different nitrogen levels are shown in Table 2 for the Rocky Ford test and Table 3 for the East Grand Forks test. It should be noted that the levels of nitrogen fertilizer used were different for each test as the soils at East Grand Forks contained much more organic matter than the Rocky Ford soils.

There was a total of four significant variety X nitrogen interactions in both tests. Only one of these was highly significant; the others were possibly chance deviations. The results indicate that the varieties, in general, reacted similarly in the three different soil nitrogen environments. In the Rocky Ford test the addition of nitrogen had only minor effects on the cliaracters studied, as significant differences between rates were detected for only seven characters from a total of twenty. These differences also were significant only at the five percent level. In the East Grand Forks test the addition of nitrogen had a greater effect on these characters than the test at Rocky Ford as ten significant differences were detected and many of these were highly significant. The addition of nitrogen significantly decreased the percent sucrose and increased the total amino acid content in each test.

Selections for high and low aspartic acid and glutamine contents significantly separated the original populations into distinct groups. In sugar per acre, tonnage, sucrose and purity, the selections gave varying results. The low aspartic acid selection (59-407) increased root yield in both tests, but the stands also

[3] Recent improvements in technique indicate that the galactinol values may be too high.

Table 2.—Stand, yield and chemical results of live amino acid selections planted at Rocky Ford, Colorado, at three different nitrogen fertility levels.

Selection No.	Character	No. Roots per 35'	Lbs. Sugar per acre	Tons Beets per acre	Percent on beet			Pct. purity	Percent on dry substance												
					sucrose	% K	% Na.		Raffinose	Galactinol	Asp. A.[1]	Glut. A.	Aspara	Gluta.	Glycine	G.A. B.A.	Alanine	Valine	Leucines	Total amino acids	Total Nitrogen
59-407	Low asp. A.	54.9	4030	17.02	11.80	.212	.243	82.6	.466	.443	.130	.067	.097	.545	.077	.200	.050	.058	.066	1.29	0.01
59-408	Hi Asp. A.	46.1	3583	14.40	12.53	.206	.190	83.9	.488	.388	.153	.065	.139	.733	.099	.220	.074	.062	.075	1.64	1.06
59-409	Low gluta.	52.0	3814	15.11	12.60	.191	.203	85.7	.422	.395	.115	.055	.073	.488	.072	.198	.047	.049	.055	1.16	0.95
59-410	Hi gluta.	45.2	3811	15.16	12.56	.220	.196	85.6	.488	.410	.165	.063	.152	.838	.114	.245	.082	.076	.097	1.84	1.11
59-411	Random sel.	46.7	3468	14.72	11.83	.229	.223	82.5	.453	.429	.143	.070	.117	.647	.084	.221	.065	.063	.074	1.49	1.03
Sign. diff. (19:1)		5.7	578	1.38	NS	.014	.018	1.5	NS	.039	.016	NS	.017	.095	.011	.023	.011	.009	.009	.14	.06
Sign. diff. (99:1)		7.5	NS	1.84	NS	.018	.024	2.0	NS	NS	.022	NS	.023	.113	.017	.031	.015	.013	.012	.18	.08
Nitrogen Rates																					
0		49.8	3861	15.26	12.64	.199	.196	84.20	.425	.379	.132	.058	.106	.588	.079	.210	.055	.056	.066	1.36	1.00
75		48.6	3797	15.38	12.37	.212	.206	84.28	.474	.424	.137	.060	.113	.643	.093	.215	.066	.060	.070	1.47	1.00
150		48.6	3575	15.20	11.78	.223	.232	82.44	.444	.436	.154	.074	.127	.719	.095	.225	.069	.069	.084	1.62	1.09
Sign. diff. (19:1)		NS	NS	NS	0.50	NS	.019	NS	NS	.042	NS	.011	NS	.097	NS	NS	NS	NS	.012	.17	NS
Sign. diff (99:1)		NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
Variety x nitrogen interaction		NS	NS	NS	NS	NS	*	NS	NS	**	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS

[1] Amino acids listed in order:
Aspartic acid
Glutamic acid
Asparagine
Glutamine
Glycine
Gamma amino butyric acid
Alanine
Valine
Leucines

Table 3.—Stand, yield and chemical results of five amino acid selections planted at East Grand Forks, Minnesota, at three different nitrogen fertility levels.

Selection No.	Character	No. Roots per 35'	Lbs. Sugar per acre	Tons per acre	Percent on beet			Pct. purity	Percent on dry substance												
					sucrose	% K	% Na.		Raffinose	Galactinol	Asp. A.[1]	Glut. A.	Aspara.	Gluta.	Glycine	G.A. B.A.	Alanine	Valine	Leucine.	Total amino acids	Total Nitrogen
59-407	Low asp. A.	29.4	5428	17.29	15.86	.134	.073	87.4	.234	.241	.162	.041	.137	0.72	.094	.168	.044	.073	.115	1.54	1.34
59-408	Hi asp. A.	26.4	4620	14.45	15.98	.137	.057	86.9	.205	.198	.187	.042	.200	1.00	.151	.185	.066	.097	.151	2.06	1.51
59-409	Low gluta.	28.4	5118	15.88	16.18	.128	.064	87.2	.220	.223	.174	.037	.152	0.65	.110	.171	.048	.075	.112	1.48	1.33
59-410	Hi gluta.	28.2	4805	15.09	15.90	.139	.063	85.5	.193	.209	.189	.042	.212	1.04	.189	.197	.086	.122	.168	2.22	1.72
59-411	Random sel.	26.6	4647	14.66	15.82	.144	.066	85.8	.230	.207	.161	.053	.188	0.94	.138	.182	.059	.093	.142	1.93	1.49
Sign. diff. (19:1)		NS	467	1.48	NS	.007	.007	1.3	.024	.026	NS	.010	.027	.16	.021	.015	.009	.014	.017	.21	.12
Sign. diff. (99:1)		NS	621	1.98	NS	.009	.009	NS	.032	NS	NS	NS	.036	.22	.028	.020	.012	.019	.022	.28	.16
Nitrogen rates																					
0		27.0	4825	14.86	16.20	.130	.060	87.8	.211	.222	.158	.035	.149	0.87	.115	.176	.054	.077	.126	1.74	1.37
50		28.7	4990	15.51	16.11	.134	.066	86.8	.208	.217	.169	.041	.169	0.80	.131	.177	.056	.088	.132	1.76	1.46
100		27.7	4955	15.96	15.54	.145	.068	85.1	.230	.208	.197	.052	.202	0.94	.163	.188	.071	.108	.154	2.06	1.61
Sign. diff. (19:1)		NS	NS	NS	.54	NS	NS	1.1	NS	NS	.016	.011	.021	NS	.022	NS	.006	.013	.014	.21	NS
Sign. diff. (99:1)		NS	NS	NS	NS	NS	NS	1.5	NS	NS	.022	NS	.030	NS	.031	NS	.009	.019	.019	NS	NS
Variety x nitrogen interaction		NS	NS	*	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	*	NS	NS	NS	NS	NS

[1] Amino acids listed in order:
Aspartic acid
Glutamic acid
Asparagine
Glutamine
Glycine
Gamma amino butyric acid
Alanine
Valine
Leucines

were at least ten percent higher than the check (59-411) in both tests. This increase in stand may have helped to increase the yield. The low glutamine selection (59-409) with a slight increase in stand, showed only slight increases in yield and sugar per acre, and was not significantly above the check for any of these characters. The low selections were higher in all of these desirable characters than the high amino acid selections. Selecting for high or low aspartic acid or glutamine did not affect the percent sucrose. Purity of juice was significantly improved by low glutamine selection in both tests, and to a lesser degree by the aspartic acid selection.

The raffinose and galactinol contents were changed by the selection for low and high amino acids. In the East Grand Forks test, the high selection of both amino acids significantly decreased the raffinose content. In the Rocky Ford test the selections were not significantly different for raffinose content. The galactinol content was significantly increased in the East Grand Forks test by selecting for low aspartic acid, however in the Rocky Ford test the high selection significantly lowered the galactinol amount. Therefore it appears that selections for high and low aspartic acid decreased and increased the galactinol content, respectively, in these varieties. The glutamine selections were not significantly different from each other for galactinol content but the results for the East Grand Forks test were similar to the trend mentioned above for the aspartic acid selections. In the Rocky Ford test the glutamine selections were not significantly different for galactinol content but the general trend was reversed.

The sodium and potassium results were different for the two amino acid selections. The low glutamine selection reduced the potassium content significantly in both tests, but did not affect the sodium content. The low aspartic acid selection increased the sodium content significantly while the high aspartic acid decreased the sodium content significantly, but did not affect potassium. These results were obtained in both tests.

There were nine amino acids evaluated plus total amino acids and total nitrogen in the progeny tests. The selections for low and high aspactic acid contents shifted all nine amino acids in their respective directions of selection, i.e., all amino acids had a tendency to increase in the high selection and to decrease in the low selection. The same was true for the glutamine selection except that the separations were greater in magnitude. There were slight variations from these results but none reached the significant level. It can be concluded therefore, that selection for a reduction or an increase of these two amino acids, reduced

or increased all amino acids. The glutamine selections were more effective than the aspartic acid selections in shitting the populations. Furthermore, total nitrogen in the beet juices of these low selections was significantly lower than the check, while the total nitrogen in the high glutamine selection was significantly higher than the check.

Discussion of Results

It is evident from the data that the aspartic acid and/or glutamine content can be increased or decreased in the root by selection pressure. Such pressure affected the nitrogen metabolism of the plants as there was a general increase or decrease of the total amino acid content and the total nitrogen content, depending on the direction of the selection pressure.

The effects of the selection pressure applied in these tests were not completely confined to the nitrogen-containing compounds. Selection of aspartic acid significantly affected the sodium content while the selection applied to glutamine significantly changed the potassium content. These were the only two mineral elements studied. Shifts in amounts of other elements also probably occurred.

Changes were noted in the carbohydrates studied. The high amino acid selections significantly decreased the raflinose content and a similar trend was apparent for galactinol. Sucrose content remained unchanged.

The general trend was for purities to increase with the lowering of amino acid content. This Avas to be expected, as a large part of the nonsugars are nitrogenous compounds. Considering all the characters studied in these tests, the selection for low amino acid content was beneficial by increasing sugar yield and juice quality, but not beneficial by causing an increase in raffinose, galactinol and sodium. These three latter constituents have a tendency to lower beet juice quality. This was overbalanced however, by the beneficial effects of the lower amino acid content as reflected in the purity data.

Considering the aspartic acid selections and the glutamine selections, it appears that either one could be used satisfactorily to improve beet varieties, although the glutamine selection was slightly more effective in spreading the populations into separate groups. In a breeding program the selection pressure applied against glutamine would be just as effective as selecting against both aspartic acid and glutamine at the same time. The low glutamine selection also had lesser detrimental effects as it did not significantly change the sodium, raffinose or galactinol content

of the beets. On the other hand increases in tonnage and sugar per acre were definitely associated with low aspartic acid selections.

It had been postulated that a decrease in one amino acid might cause an increase in some of the other amino acids of the beet. In these tests, there was no striking evidence of this occurring. If one amino acid was reduced or increased by selection, then all amino acids had a tendency to be reduced or increased.

It would be interesting to know more about the physiology of the sugar beet plant, and the chemical pathways ot the many metabolic systems. In this investigation, amino acid selections caused changes in the concentration of some carbohydrates and also of the mineral elements studied. Why and how, in the various metabolic systems, does selection for high or low amino acids affect the carbohydrate physiology or the utilization of minerals? The data presented show that it does happen but additional basic physiological studies are needed to elucidate these interlocking metabolic systems.

The varieties responded to the fertilizer treatments as was expected, i.e., when more nitrogen was applied the increase in amino acids and nitrogenous compounds was greater. If the plant breeder develops varieties which are low in amino acids under moderate levels of nitrogen, these plant breeding advances can be offset by applying excessive amounts of nitrogen. Therefore good agricultural practices must be adhered to for improved varieties to work most efficiently.

Summary

(1) Selection for high and low aspartic acid and glutamine contents of sugar beets caused an increase or a decrease respectively in all nine amino acids, as well as total amino acid content and total nitrogen content. Purity of juice was significantly improved by the low glutamine selection in both tests, and to a lesser degree by the aspartic acid selection.

(2) Low glutamine selection reduced the potassium content significantly in both tests but did not greatly affect the sodium content. The low aspartic acid selection increased the sodium content significantly while the high aspartic acid selection decreased the sodium content, but the different aspartic acid selections had little effect upon potassium. Low aspartic acid selections also increased yield but percent sugar was not significantly affected by an amino acid selection.

(3) Differences were obtained between different nitrogen applications but selection X nitrogen interactions were not important.

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The Development of Control Charts for Package Weights

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Weight control on a package line has always been subject to question. The question of how often should samples be taken, when is a scale adjustment required, what constitutes a light weight, how much overweight is required, how to tabulate and evaluate check weighings and many more come up whenever the subject of weights arises. Most of the questions can be answered through the construction and use of control charts.

Control chart technique has proven to be a valuable tool as evidenced by rapidly expanding use in industry and governmental agencies over the past several years. The literature contains a great deal of information on this subject and on related statistical problems. A list of what we believe to be excellent references will appear at the end of this paper.

Each weighing system presents a somewhat different problem. Also each piece of equipment yields a weight distribution pattern around some mean or average weight produced. This paper will outline and briefly discuss the steps necessary to set up Weight Control Charts for multiple scale equipment used in the production of five- and ten-pound packages of granulated sugar. The same general procedure can be applied to other size packages.

Unfortunately, it is a rare occasion when the same control chart can be applied to two pieces of equipment apparently identical in every respect. Furthermore, from time to time the performance of the equipment changes due to mechanical wear, difference in product and other causes. Consequently, it is necessary to construct a Control Chart for each multiple head unit and to re-evaluate the performance of any unit from time to time or after a major overhaul.

A Control Chart program can be developed and put into operation by following the steps presented herein. For more detail and information relative to the derivation of the mathematical relationships the reader is referred to the literature list on this subject.

Calibration of Check Scales

If the scales used for check-weighing show only over or under and exact weight, it is necessary to inscribe calibration marks on the scale dial. Our company uses Toledo check scales for the five- and ten-pound packages. These scales can be calibrated in five-gram units and can be read to two and one-half grams. The

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sensitivity of the scales is such that a one-gram weight will deflect the pointer slightly with a ten-pound load on each side of the scale. The five-gram division has been found to be more satisfactory than a one-quarter ounce division. It is necessary to select a division small enough to yield a satisfactory distribution curve for the package being produced from the equipment. The calibration and subsequent use of the scale divisions are easier to handle if they are referred to as units rather than their actual value. In this case one unit is equal to five grams but the data are recorded in units and half units and not as grams.

Determination of Tare Weight

In practice the sewn bag is used for check-weighing rather than the unsewn bag. This procedure eliminates the chance of spilling sugar or otherwise spoiling the sample. It also makes it easier to sample the production line.

During the sewing operation a small amount of the bag top is clipped off and the tape plus thread is added. The net change in the weight of the empty bag caused by this step should be determined. It usually will not exceed 1.2 grams.

A random sample of 15 to 20 bags must be withdrawn from each of several lots of empty bags to determine the average weight of the empty container. A tare weight equal to this average minus the sewing loss is then made from an old weight or some other suitable material. The weight for the empty bag should be determined for each shipment of empty bags. This weight is used with the appropriate five- or ten-pound weight used on the check scales.

The practice of using an empty bag for a tare weight is satisfactory only if it is adjusted for clip-off and is changed each day or so. Paper is subject to weight change caused from moisture changes and dust.

The check scales and weights must be kept clean, free from vibration and checked for zero balance at all times.

Evaluation of Packaging Equipment Variability

In most instances, packaging equipment for five- and ten-pound units consists of four or six scale buckets, filled and emptied in sequence. Variability in delivered weights is subject to the total of several effects. Sugar condition, cleanliness of the linkages, vibration, sensitivity of mercoid switches or sensing devices, mechanical sequencing and gearing, all have an effect on the uniformity of the delivered weight. To estimate the variation it is necessary to collect and check-weigh thirty to fifty sets of packages produced over a period of three or four hours. A set consists of six bags (one from each bucket in se-

quence) for a 6-bucket scale unit. The results of the first five sets are examined first to determine whether or not the six buckets are all the same in delivered weights. If any are found to be consistently underweight or overweight, that bucket is adjusted to be more nearly equal to the correct amount or in line with the others. Sampling is then continued, without further scale adjustment until the thirty or more sets are obtained and the results tabulated as shown in Table 1.

Table 1.—Typical package machine data for control chart development. Nampa Factory 5 lb. Machine—6 Buckets, 1 Unit = 5 Grams

Test no.	1	2	3	4	5	6		Avg.(1)	R(1)
1	1.0	—.5	0	1.5	3.5	2.5	8.0	1.3	4.0
2	0.5	1.0	0.5	2.0	2.5	1.5	8.0	1.3	2.0
3	1.5	2.5	1.5	1.0	—1.5	1.5	6.5	1.1	4.0
4	2.0	4.0	2.0	3.5	3.5	2.5	17.5	2.9	2.0
5	0.5	—1.0	1.0	1.0	0	2.0	1.5	0.3	5.0
6	1.5	0.5	1.5	3.5	1.5	—1.0	7.5	1.3	4.5
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Through	....	....	....	....	....	....	....	....	....
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31	2.0	—0.5	0.5	0	—2.5	1.5	1.0	0.2	4.5
Total 31	43.5	40.0	33.0	51.0	45.5	43.0	256.0		93.1
Avg(2)	1.4	1.3	1.1	1.6	1.5	1.4			
R (2)	4.0	6.0	3.5	5.0	6.0	7.0		$\bar{R}$ (1)	= 3.00

$$\bar{R}(2) = \frac{31.5}{6} = 5.25$$

$$\bar{R}(1) = \frac{93.1}{31} = 3.00$$

$$\sigma' = \frac{\bar{R}}{d_2} = \frac{3.00}{2.534} = 1.18$$

Results are recorded as units and half units with a negative sign indicating the lightweight packages. An algebraic sum, average and range is calculated for each set and for each individual bucket. The range is defined as the total difference (in units) between the lightest and heaviest item in the set or series.

At this point there is some difference of opinion as to the proper evaluation of the results. Strictly speaking the correct way to evaluate the results would be to consider each bucket separately since each bucket can be individually adjusted. How-

ever, this would necessitate a separate control chart for each bucket. Sampling and plotting results would be complicated and too time consuming for efficient control. The danger of mathematical errors or lack of full understanding on the part of employees could more than offset the slight difference between this approach and the simplified method outlined as follows.

In actual practice the four or six buckets empty in sequence for each cycle. If an inspection by Federal or State agencies is made at a retail outlet for sugar, all of the sugar in that lot can usually be assumed to have come off the production line during some continuous period from a few minutes to a few hours. If twenty to twenty-five packages are examined they may well represent only three to four cycles very close together. It is preferable in our opinion to consider the calculation of the scale unit accuracy as sets consisting of four or six packages, one from each scale bucket in sequence.

An examination of Table 1 will usually reveal a wider range within buckets than between the separate buckets. This fact has the effect of tightening the control slightly which will be more evident when the Control Chart Development is completed.

Since we are considering the accuracy of the equipment as an entire unit and not as individual components, it is next necessary to calculate the sum of the set ranges and an average range for the number of sets involved. The standard deviation (V) of the set ranges is either calculated or taken from Statistical Tables for Range. Table 2 reproduces in part the necessary factors involved. By using the values in Table 2

$$(1) \quad \sigma' = \frac{\bar{R}}{d_2}$$

σ′ is, therefore, an expression for the scale accuracy or the measure of the dispersion about the Range Mean.

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Selection of the Amount of Average Overweight

Unfortunately, it is not permissible for a producer of a packaged commodity to market a product which averages the stated net weight stamped on the bag and be in agreement with the various State regulations. If this were so, 50% of the packages would conceivably be slightly overweight packages and 50% slightly underweight. In a broad sense the average accepted weight must not be less than the stated net weight and only a reasonable amount or percent of packages can be underweight. The actual amount of underweight expressed as a percent of the net on those underweight packages must not be excessive. The definition of "excessive" is rather vague at this point.

In addition, the average weight from any production line rises and falls over a period of time. This is caused by machine inaccuracies or product fluctuations or both.

The decision as to the percent underweight a company will decide to produce determines the average over-fill sacrificed in order to meet the specifications. The precision of the packaging equipment enters into the discussion at this point. Equipment which produces reliable weights within narrow limits or dispersion permits the company to bring the average weight closer to the net weight than less reliable equipment yielding a wide variation in weights of product.

Generally speaking, the underweight percentage will be between 10 and 35%. As a rule of thumb, it is generally permissible to increase the percent underweight as the package net weight is increased. This reasoning can be used to standardize the percent over-fill on total product for all size packages a company is willing to accept providing the equipment permits the producer to comply with regulations.

Table 2.—Condensed table of factors for establishing control chart limits.

No. of Items in Set	Factors			Percent underweight desired	Factor Z
	d_2	A_2	D_4		
2	1.128	1.880	3.267	0	3.00
3	1.693	1.023	2.575	2.5	1.96
4	2.059	0.729	2.282	5.0	1.64
5	2.326	0.577	2.115	7.5	1.41
6	2.534	0.483	2.004	10.0	1.28
				12.5	1.15
				15.0	1.04
				17.5	.93
				20.0	0.84
				25.0	0.67
				30.0	0.52
				35.0	0.39

$\sigma' = \frac{\bar{R}}{d_2}$

$\bar{X} = \sigma' / z$

3 σ Limits for Average

UCL $= \bar{\bar{X}} + \bar{R} A_2$

LCL $= \bar{\bar{X}} - \bar{R} A_2$

3 σ Limit for Range

UCL $= \bar{R} D_4$

Note: Values for σ' $\bar{X}$ and $\bar{R}$ apply to system where 0 = stated Net Weight on Package and values of statistics are in appropriate units as used with check scales.

Source: Complete Tables for above values are found in ASTM Manual on Quality Control of Materials. 1951, and in Probability Tables.

For example let us assume that it is decided to produce $12\frac{1}{2}$% underweight packages as a reasonable amount. In Table 2 under the column Z the value of 1.15 relates $12\frac{1}{2}$% of the one tail area under the normal curve to the standard deviation of the

machine accuracy calculated from Table 1, according to the formula:

$$(2) \quad Z = -5- \text{ or } X = Zo\text{-}'$$

where X establishes the average overweight necessary to produce 12i4% lightweight packages 99% of the time.

Using the values from Table 1 and Table 2 and 12i/% desired underweight, we find from formula (1) </ = ^ 3.00 **F Q T** ^ ^ ^

from formula (2) X = 1.15 X 1.18 = 1.36

since the data are in units of 5 grams we now know that we must have an average overweight of 6.8 grams or nearly one-fourth ounce for each five-pound package produced.

For a 100,200 pound car of five-pound packages this means that the company must give away 300 pounds of sugar in order to conform to the regulation. If the equipment or lack of control of package weights is such that the giveaway is more than 300 pounds, the economics of the situation are readily apparent. If, on the other hand, too many underweight packages are produced in this lot, you are in trouble with the FDA and again an expensive situation develops.

Consequently, the answer lies in being able to control this figure within reasonable limits. To do this a Control Chart is set up for the purpose of recording test results and to give the operator a basis upon which machine adjustments can be made.

Control Chart Limits

A Control Chart consists of two parts: One upon which the average of a sample set is plotted and the other upon which the range of the set is plotted (Figure 1).

Upper and lower control limits are established for the averages based on the scale information in Table 1 and upon the average overweight determined. These limits designated as three sigma (3<T) values encompass all chance results in over 99% of the trials providing there has been no shift in the average or some outside influence has not affected the mechanism. A 3a- upper limit is also calculated for the range section of the Control Chart.

By using information already obtained, i.e., the average range and X, the control limits are calculated as follows using values from Table 2:

Control Limits for Average

Upper Control Limit **(UCL)** = $X + A_2 R$

where A_2 corresponds to the factor for 6 packages comprising the sample

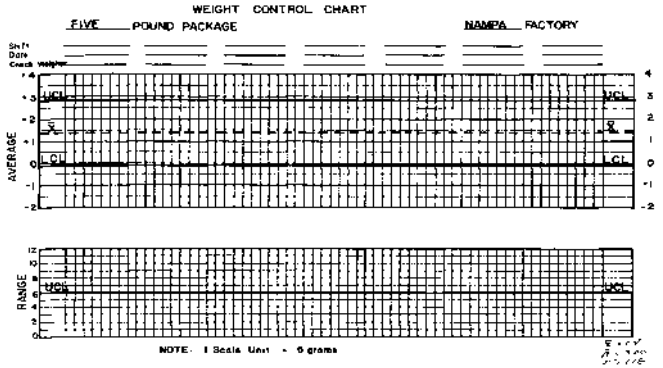

Figure 1.

Lower Control Limit for Average (LCL) $= \overline{X} - A_2 \overline{R}$.

The Upper Control Limit for the range is equal to $D_4 \overline{R}$ where D_4 corresponds to six measurements per set.

These calculations, therefore, give us by substituting the predetermined figures:

For Average

UCL $= 1.36 + 0.483\ (3.00) = 2.8$ (units)

LCL $= 1.36 - 0.483\ (3.00) = -0.1$ (units)

For Range

UCL $= 3.00 \times 2.004 = 6.00$ (units)

The $\overline{X}$, UCL and LCL lines are drawn on the Control Chart on the Average section. The UCL for Range is also drawn in.

The Control Chart is now ready for use in actual testing and control of package weights.

Use of Control Chart and Interpretation of Results

A suitable work sheet should be made up on which can be shown the plus or minus values for each package comprising a sample set. Provision should be made for the total, average and range with an extra line to be used to indicate adjustments, if required.

A sample is withdrawn from the production line consisting of one package from each scale bucket in sequence. The packages are individually weighed on the check scales and the over or under weight in units is recorded. The sum, average and range are calculated. Identity of the individual packages and the corresponding bucket must be maintained.

The average and range are then plotted in the appropriate place on the Control Chart.

If the plot for the average is between the upper and lower control lines the system is judged to be in control or operating in a normal fashion. If the range plot is between zero and the upper control limit (UCL) the spread between individual buckets is assumed to be normal.

It should be pointed out that it is possible for the average to be *out* of control and the range to be *in* control. It is also possible for the average to be *in* control and for the range to be *out* of control. If either situation develops, an inspection of the work sheet will tell the operator where the fault lies and what adjustment is required to bring the system back into control.

For example:

Range In Control, Average Out of Control
This situation points to the fact that two or more buckets are weighing either too heavy or too light depending upon the location of the average plot. The buckets at fault are inspected and adjusted. Note of the change is recorded on the work sheet and another sample set is taken after the operator is satisfied the weighing response to the adjustment has stabilized (usually within 2 - 3 cycles). Results of the next sample are plotted to determine whether or not the correction step was sufficient to bring the system back into control.

Average In Control, Range Out of Control
In this case there is usually one one or possibly two buckets which have gone out of control in opposite directions. The work sheet will show what corrective action is required. A notation is made and another sample is taken.

If both range and average plots are within the control limit lines, no adjustment is made on any of the buckets. The package line is known to be performing normally. There is only one chance in a hundred that a sample set will indicate lack of control when actually the system is in control. The following discussion points to an exception to this rule and is part of the interpretation of results which should give all concerned additional confidence in Control Chart Technique.

After the Control Chart method has been in operation for only a short time, the graphical trends shown by lines connecting the consecutive sample plotting points yield a clear picture of the behavior of the packaging equipment.

If all of the average points fall below or above the X line but still between the LCL and UCL lines, there is an indication that

the average of the production has shifted either down or upward. The result is that you are either producing more underweight units than desired or are giving away more sugar than is required to meet the weight regulations.

The appropriate small adjustment is then made to correct this small but significant trend. A well-controlled system will reveal points falling both above and below the X line in a sine wave pattern. No more than three to five points in succession should be all below or all above the X line.

Sampling Frequency and Personnel

The frequency of sampling depends upon and governs the degree of accuracy you wish to maintain. A sample should always be taken as soon as practical after the production line has started up after a shutdown or at the beginning of a shift. Samples should then be taken at thirty-minute or not more than sixty-minute intervals thereafter during the shift. Time must also be allowed for additional sampling after an adjustment has been made.

We have found that additional labor is not required to carry out a weight control program such as this. It is preferable to appoint one man on each shift who is thoroughly familiar with the machinery and operation to be responsible for the check weighing. This should be the primary job of the employee.

All foremen, and other supervisors should be familiar with the program and understand the objectives and interpretation of the results. As an additional aid to supervisors it has been recommended that the weight Control Chart on a particular package be continuous even though the production is intermittent. Notation as to date and shift can be made above the plotting for a particular time interval of operation. If more than one crew is used on production, the graphic plotting for the first shift can be in red pencil, second shift, blue and third shift, yellow.

It has been our experience that wherever Control Chart systems are used that all personnel involved take more interest in maintaining good weights of packages and better maintenance of equipment is achieved.

In addition, should an inspection of the plant be made by FDA representatives, or should an adverse situation arise, a good weight control system presented as evidence will go a long way toward assuring the public that you are endeavoring to maintain satisfactory weights in their behalf.

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Control of Seedling Diseases of Sugar Beets With Dexon and Dexon PCNB Mixture[1]

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Seedling diseases or root rots are of considerable importance in the growing of sugar beets, especially in heavy irrigated soils in Montana. Investigations showed (5)[3] that the following fungi are involved in the complex of seedling diseases of beets in Montana: *Aphanomyces, Pythiurn, Rhizoctonia, Phoma, Fusarium* and some others. However, evidence indicates that *Aphanomyces cochlioides* probably is the most important pathogen of young sugar beets in heavy soils.

In an attempt to control these diseases, several soil and seed treatments have been investigated with sugar beets since 1939 at the Huntley Branch Station and also in the greenhouse in Bozeman using soil from the Huntley Station (1, 2, 3, 4). In general, seed treatments were found to be only slightly beneficial in controlling seedling diseases of beets. However, soil treatments with fertilizers were found to be of great importance in controlling these diseases. Sugar beets planted in soil well fertilized with manure, nitrogen and phosphorus always had only small amounts of seedling disease as compared to those grown in soil poor in nutrients and organic matter.

New interest in this work arose recently when the Chemagro Corporation introduced a new seed and soil fungicide called Dexon (p-dimethylaminohenzenediazo sodium sulfonate) which has demonstrated an ability to protect plants from a damping-off root rot complex involving species of *Pythiurn, Aphanomyces,* and *Phytophthora* fungi. It was suggested by Chemagro that if *Rhizoctonia* was also involved in the complex, the addition of PCNB (pentachloronitrobenzene) would control that pathogen also. In testing the above-mentioned compounds for control of seedling diseases of sugar beets, several greenhouse tests were conducted in Bozeman, Montana, using heavy Huntley soil. Soil flats were planted with segmented seeds of The Great Western Sugar Company's variety GW359.

Experimental Procedure

In some tests the soil was not sterilized and was either inoculated with *Aphanomyces* or not inoculated. In the other tests, soil was sterilized and inoculated either with *Aphanomyces* or *Rhizoctonia* alone or with a combination of these fungi.

[1] Contribution from Montana State College, Agricultural Experiment Station, Bozeman, Montana, Paper No. 569, Journal Series.

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[3] Numbers in parentheses refer to literature cited.

The following seed and soil treatments were used:

1. Seed treatment—70% Dexon, 2 oz per 100 lbs of seed.
2. Seed treatment—Dexon-PCNB, (35-35), 4 oz. per 100 lbs of seed.
3. Soil treatment—Dexon 5%, regular[4], 1 lb active per acre.
4. Soil treatment—Dexon 5%, coated[1], 1 lb active per acre.
5. Soil treatment—Dexon 5%, regular, 2 lbs active per acre.
6. Soil treatment—Dexon 5%> coated, 2 lbs active per acre.
7. Soil treatment—Dexon 5%, regular, 4 lbs active per acre.
8. Soil treatment—Dexon 5%, coated, 4 lbs active per acre.
9. Check soil.

In the first experiment only some of the soil treatments were used. In soil treatments, Dexon was applied in bands along the beet rows and was well mixed into the soil. Readings of healthy and diseased beet seedlings were made at regular intervals during the duration of the test and final readings were taken at harvest time. Beets were allowed to grow about a month after they emerged.

In the first test non-sterilized soil was used. In one series the soil was not inoculated, but in the other series, inoculum of *Aphanomyces cochlioides* obtained from four petri dish cultures, was added to each flat of soil. Three rows of beets were planted in each flat with 50 segmented seeds per row[7]. Dexon was used as a soil treatment only at the rates of one and two pounds of active material per acre (Table 1).

Table 1.—Soil and seed treatment experiment for controlling *Aphanotnyces* seedling disease of sugar beets—1960.

Seed and soil treatments	Sugar beet seedlings	
	Non-inoc. soil Healthy percent	Inoc. soil Healthy percent
1. Seed—70% Dexon, 2 oz/100 lbs seed	1.0	34.9
2. Seed—Dexon-PCNB, (35-35), 4 oz/100 lbs seed	92.6	88.0
3. Soil—Dexon 5%, regular, 1 lb/acre	58.5	88.8
4. Soil—Dexon 5%, coated, 1 lb/acre	57.4	79.5
5. Soil—Dexon 5%, regular, 2 lbs/acre	91.8	90.7
6. Soil—Dexon 5%, coated, 2 lbs/acre	73.4	94.0
7. Check Soil	46.9	23.0

[4] Two forms of Dexon manufactured by the Chemagro Corporation.

Results and Discussion

The results presented in Table 1 show that seed treatment with Dexon alone was not beneficial to sugar beets planted in non-inoculated soil and the amount of disease in non-inoculated soil was even greater than in the check soil. In the inoculated soil this treatment also was of little benefit and the amount of disease was high; however, it was slightly below the check soil. It is possible that some other fungi in addition to *Aphanomyces* may have contributed to the higher degree of disease in non-inoculated soil.

Sugar beet seedlings grown from seeds treated with Dexon - PCNB combination had a rather low amount of disease in both types of soil. Since this soil was known to be infested with *Rhizoctonia,* and since PCNB is quite effective against this fungus, it is possible that the low incidence of the disease was due to the combined effect of Dexon and PCNB on *Aphanomyces* and *Rhizoctonia,* respectively.

The amount of disease of beets in the non-inoculated soil treated with 1 lb of both kinds of Dexon was relatively high and similar in both cases. Considerably less disease occurred in the same treatments in the inoculated soil. Treatments of soil with 2 lbs of Dexon produced considerable reduction in beet diseases in both types of soil. Beets grown in the inoculated check soil had a high amount of disease.

In the above-mentioned test, non-sterilized soil was used. This soil undoubtedly was infested with several plant pathogens like *Aphanomyces, Pythium, Rhizoctonia,* and possibly others, all of which could produce seedling diseases of sugar beets. It is practically impossible to identify the causal organism responsible for disease of those seedlings on the basis of symptom expression alone. For this reason it is difficult to make any conclusions regarding the specific action of Dexon or Dexon-PCNB combination for controlling any specific disease of beet seedlings caused by a certain organism.

To obtain more information on this subject, another test was conducted in which the soil was sterilized and inoculated with *Aphanomyces cochlioides.* This test made it possible to investigate the effect of Dexon in controlling this particular disease of sugar beets. In this test nine flats of soil were used. Three petri dish cultures of *Aphanomyces* were added to each flat of soil. Three rows were planted in each flat of soil with 30 segmented seeds per row.

Results presented in Table 2 show that both seed treatments producd only a slight reduction in disease and Dexon alone was

Table 2.—Soil and seed treatment experiments for controlling *Aphanomyces* seedling disease of sugar beets—1960.

Soil and seed treatments	Sugar beet seedlings Healthy percent
1. Seed—70% Dexon, 2 oz/100 lbs seed	27.5
2. Seed—Dexon-PCNB, (35-35). 4 oz/100 lbs seed	16.2
3. Soil—Dexon 5%, regular, 1 lb/acre	91.8
4. Soil—Dexon *5%.* coated, 1 lb/acre	100.0
5. Soil—Dexon 5%, regular, 2 lbs/acre	93.1
6. Soil—Dexon 5%, coated, 2 lbs/acre	9(i.4
7. Soil—Dexon 5%, regular, 4 lbs/acre	83.5
8. Soil—Dexon 5%, coated, 4 lbs/acre	79.1
9. Check Soil—Inoculated	0.0

more effective than Dexon-PCNB combinations. All soil treatments were effective in controlling this disease. A slightly lower percentage of beet seedlings remained healthy in the soil which received the highest application of Dexon as compared to the other dosages. It is possible that some of the seedlings were lost in these flats due to Dexon toxicity.

It was mentioned above that in addition to *Aphanomyces cochlioides,* there are present in Montana soils other fungi which can infect young sugar beets and cause disease. It is believed that under field conditions *Rhizoctonia* is probably the next in importance to *Aphanomyces* in causing seedling diseases of sugar beets in Montana.

In the following experiment an attempt was made to investigate the fungicidal effect of Dexon alone and in combination with PCNB, on the control of seedling diseases of beets caused by *Aphanomyces* and *Rhizoctonia* alone and also in combination.

Three parallel series of flats with sterilized soil were used. Each series consisted of nine flats. One set was inoculated with *Aphanomyces,* the other with *Rhizoctonia* and the third was inoculated with a combination of these two organisms. Three petri dish cultures of *Aphanomyces* or *Rhizoctonia* were added to each flat inoculated with these organisms singly. The same amounts of inoculum were added to flats inoculated with both of these fungi. Three rows of beets were planted in each flat of soil with 30 segmented seeds per row. Results of this test are presented in Table 3.

A very high percentage of sugar beet seedlings remained healthy in a set inoculated only with *Aphanomyces* in both soil and seed treatments. Plants grown from seed treated with a combination of Dexon and PCNB showed slightly more disease than beets with the other seed treatment. Check beets in this set had a very high percentage of disease. It is quite evident that

Table 3.—Soil and seed treatment experiments for controlling *Aphanomyces* and *Rhizoctonia* seedling diseases of sugar beets—1960.

Soil and seed treatments	Sugar beet seedlings grown in soil inoculated with: Aphanomyces Healthy %	Rhizoctonia Healthy %	Aphanomyces and Rhizoctonia Healthy %
1 Seed—70% Dexon, 2 oz/100 lbs seed	99.3	62.3	56.4
2 seed—Dexon-PCNB, (35-35), 4 oz/100 lbs seed	82.9	90.8	92.6
3 Soil—Dexon 5%, regular, 1 lb/acre	96.1	60.0	64.7
4 Soil—Dexon 5%, coated, 1 lb/acre	97.2	43.2	71.1
5 Soil—Dexon 5%, regular, 2 lbs/acre	96.9	37.8	58.9
6 soil—Dexon *5%,* coated, 2 lbs/acre	92.8	51.2	50.0
7 Soil—Dexon 5%, regular. 4 lbs/acre	93.0	43.8	38.1
8 Soil—Dexon 5%, coated, 4 lbs/acre	100.0	56.1	61.5
9. Check Soil—Inoculated	19.7	52.3	8.8

Dexon treatments produced a beneficial effect on the control of disease of beets caused by *Aphanomyces.*

Sugar beet seedlings grown in soil inoculated only with *Rhizoctonia* had a considerable amount of disease in all treatments except one where seed was treated with a combination of Dexon and PCNB, in which 90.8 percent of plants remained healthy. The percentage of healthy plants in all treatments, including the check, varied, and was either slightly above or below 50 percent. These results indicate that Dexon was not very effective against *Rhizoctonia.* However, where PCNB was used in combination with Dexon as a seed treatment it definitely produced a beneficial effect in the control of this disease.

Percentages of healthy beet plants, grown in flats inoculated with a combination of *Aphanomyces* and *Rhizoctonia,* were quite comparable to those in the set inoculated with *Rhizoctonia* alone, except that only a few healthy plants remained in the check soil. This undoubtedly was caused by an addition of *Aphanomyces* to the inoculum.

General Conclusions

It appears that Dexon used as a soil treatment is quite effective in controlling the disease of sugar beets caused by *Aphanomyces,* but it is not reliable against *Rhizoctonia.* On the other hand PCNB is quite effective against the disease caused by *Rhizoctonia.* In controlling seedling disease of beets caused by *Aphanomyces,* seed treatments either with Dexon alone or Dexon-PCNB combination are not as reliable as soil treatmnt with Dexon.

Since there may be present in the soil various pathogenic fungi which can cause seedling diseases of sugar beets, it would

be advisable to use combinations of Dexon and PCNB for their control.

As far as soil treatment is concerned it appears that Dexon applied at the rate of 1 lb per acre (active material) to flats of soil in the greenhouse is not toxic to beets. However, a 2-lb rate of this substance may produce a slight degree of toxicity and 4 lbs of Dexon is definitely toxic. Even at a 4-lb rate of Dexon most of the beets survived until harvest. Under field conditions this toxicity would probably not be as evident as it was in the very limited amount of soil in flats.

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Notes Section

Wind Damage Control in the Red River Valley Using a Small Grain Cover Crop. The Red River Valley is a level area susceptible to wind erosion. With summer fallow being a prerequisite of sugar beet culture, it is readily recognized that soil erosion and wind damage to the beet seedlings are problems of the beet grower.

Drainage in the Red River Valley is primarily dependent on surface ditching. These ditches must be kept free of blow-dirt in order to adequately handle excess water.

A light seeding of small grain ($1/_2$ bushel per acre) planted the first week of September provides adequate growth before freezing to anchor the top soil during the winter months and will prevent the drainage ditches from becoming filled with blow-dirt. In the spring, preparing; the cover cropped fields for planting involves incorporating the residue of the cover crop with the top soil to prevent blow-out injury to the beet seedlings. Shallow seed-bed preparation with a single disc or field cultivator provides the soil-plant mixture required. Residue from the cover crop does not interfere with planting or cultivating operations.

Planting beets directly in the cover crop with no seedbed preparation has, also, proven satisfactory. This practice allows earlier planting and prevents any loss of moisture through seedbed preparation.

Use of additional fertilizer to compensate for the fertilizer utilization by the cover crop is not required inasmuch as the cover crop does not reach maturity.

The above described practices appear to be the best and most economical program for (1) saving surface soil, (2) preventing the beet seedlings from blowing out, and (3) keeping drainage ditches clean.

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Lagooning and Treatment of Waste Water

W. W. BARR[1]

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The disposal of waste waters from factories of the sugar beet industry has been given increasing attention by various federal, state and municipal agencies. With the growth of municipalities and the industries in and about them, the need for water supplies of domestic quality becomes more acute for all competing interests. The results of the enactment of legislation over the years to combat stream pollution are evident in most areas served by our industry. The beet sugar industry can rest assured that within a few years, adequate facilities for the treatment of our waste waters will be in effect in all areas.

The American Crystal Sugar Company, in the operation of nine plants in six states, has become fully aware of the problems entailed in waste water disposal and stream pollution. Laboratory facilities for the sanitary analysis of waste water are presently provided at six of our plants. The states of Iowa and Minnesota have recognized for quite some time that the direct return of waste water to a receiving stream (even though it may be screened and subjected to a sedimentation treatment) will not, by dilution, effect a desirable oxygen balance in that stream. Although Minnesota possesses adequate ground-water reserves, river flow in the areas of American Crystal Sugar Company's plants is quite variable and on the average is not of large volume.

A practical solution to the problem of handling high volume beet sugar wastes, high in suspended and soluble organic matter, has been the use of stabilization ponds. The construction of trickling filters, activated sludge systems, clarifiers and the like, even for treatment of relatively small volumes of highly contaminated water, does not appear to be entirely suitable from either a cost or an operational standpoint, particularly under adverse weather conditions. With campaigns extending to 130 days, one can expect a number of days when the maximum temperature is zero or below, necessitating the processing of storage beets with a certain percentage frozen. A continuing increase in the strength of the waste water, measured as biochemical oxygen demand (BOD), is noted as the campaign progresses.

The first steps by the American Crystal Sugar Company in the lagooning of waste water over winter, were taken at the Mason City, Iowa, plant in the early 1920's. A study was instituted there in 1946 to obtain information on sedimentation and the oxidation

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rate of impounded wastes during the early part of the season when biological activity would be in effect. Segregating the water from early campaign in one pond, the study was continued into the spring months during the period of discharge. On the basis of this work, authorities of Minnesota and with the sanction of North Dakota, granted a permit for construction and approved the operation of a lagoon system at the new Moorhead plant for the 1948 campaign. Similarly, a like plan was put into effect in 1954 for the Crookston plant.

Since the major rivers in the area provide the means of final disposal of waste waters, some facts on these waterways are in order. The Red River of the North, formed by the confluence of two smaller streams in southeastern North Dakota, forms most of the boundary between North Dakota and Minnesota in its 394-mile flow to the international boundary and thence into Lake Winnipeg. The river distance is about twice that of the road distance. The one-half foot drop per river mile causes a relatively sluggish condition which effects considerable buildup of sludge deposits during periods of normal flow. Anaerobic decomposition of such sludge causes some material to go into solution while partially oxidized matter goes into suspension. This is of some concern in determining the allowable or calculated volume of waste water that can be safely released since the effect is an increased BOD of the river and decreased dissolved oxygen content. Such a condition, particularly during periods of ice coverage, produces a serious negative oxygen balance.

The Moorhead plant draws water from the Red River just above the Moorhead sewage treatment plant and discharges its waste about a mile below. The Fargo sewage treatment plant discharge is approximately six miles below our outfall. Overloading of both municipal plants curtails our discharge numerous times during the course of a year.

The East Grand Forks plant, situated at the junction of the Red River and Red Lake River, draws its water from the latter as does the Crookston plant 48 river miles upstream. Our plant and the City of Crookston, both employ lagoons and discharge their effluents into the Red Lake River. The twin cities of Grand Forks and East Grand Forks and our plant at East Grand Forks all discharge into the Red River. A new lagoon system constructed at the East Grand Forks plant in 1961 replaced the sedimentation pond constructed in 1934 which allowed only about one day's retention. The city has had a lagoon system in operation for about a year. River flow there has averaged about 2,300 cfs as compared to about 500 cfs at Fargo-Moorhead. There are extended periods during the operating season when the river flow at Moorhead-Crookston ranges from 50 to 100 cfs.

Since none of our Minnesota plants employ the Steffen process, we are not confronted with that waste problem. The three plants are each provided with separate pond areas for lime flume wastes. Facilities are such that after stabilization the effluent may be discharged into the lagoon system.

Some change is to be noted during the last few years in the provisions of the Minnesota Water Pollution Control Commission's approval for construction and permit for the operation of sewerage facilities. The provisions for both the Moorhead and Crookston plants are basically as follows:

1. That no liquid wastes will be discharged from the lagoons during periods of ice coverage in the stream.
2. That wastes will be held in the lagoons and the rates and conditions under which the wastes are discharged will be controlled so that:
   (a) Dissolved oxygen (DO) of the water in the river below the outlet of the plant wastes will not be reduced below 3.0 parts per million;
   (b) A positive oxygen balance (dissolved oxygen greater than the 5-day 20° C biochemical oxygen demand) will be maintained at all times;
   (c) The wastes will not have a deleterious effect on domestic water supplies taken from the river below the plant.

The conditions for the new system at East Grand Forks stipulate that:

1. The total available discharge capacity will be sufficient to allow discharge of the contents of the pond within a period of one month.
2. That the release of the wastes be controlled as may be necessary to avoid depleting the dissolved oxygen of the receiving waters below 3 mgm/liter at any time as shown by adequate sampling records, and that there be no deleterious effect on domestic water supplies, or any material interference with other established uses of the river below the point of discharge.

These latter provisions imply that no discharge should be made to the river during periods of ice coverage. The second item in the East Grand Forks permit is based in part on studies made during the spring months of 1960 and 1961. Investigations were made to determine the effect of releasing large volumes of waste water to the river with high initial DO, during the period of high run-off and low water temperature, in terms of dissolved oxygen, BOD, and total dissolved solids. Control was aimed at

maintaining a river DO of about 5ppm residual, determination of threshold odor concentrations or odor quality and establishing the location of the sag point in respect to DO. The Moorhead factory established a series of 15 sampling stations extending 90 river miles downstream, where the plant at East Grand Forks took over the survey and continued for about 90 river miles beyond the city. With initial pond BOD's in the order of 1,000 and 1,700 at Moorhead and Crookston, respectively, maximum discharge ranged from 15 to 40 M.G.D. in March 1960. The fact that both the Red River and Red Lake River flows were adequate to absorb a high volume rate of discharge of the factory wastes, as indicated by a favorable DO content, was viewed with enthusiasm by our company as well as state authorities. On the basis of previous control measures only a fraction of that volume could be released. Prior to this the rate was governed to concur with the stipulations of the permits and estimated from graphic charts furnished by the Minnesota Department of Health. These charts, showing allowable discharge at various BOD levels of waste, correlated to stream flow, are based on this fomula:

$$\frac{(\text{M.G.D. stream flow} - 15)\ 6}{\text{ppm BOD of Waste}} = \text{M.G.D. allowable waste discharge at river temp. 4° C, open water}$$

In the above case applicable for Moorhead. 15 is the minimum stream flow at or below which the river should receive no waste. This value for Crookston is 35. Six ppm dissolved oxygen are indicated to be available for BOD stabilization in water, at 4° C, saturated in respect to oxygen. With a receding river and increase in river water temperature, control measures revert from the basis of maintaining a residual *3* - 5 ppm DO to that of positive oxygen balance. It is hoped that with adequate treatment and stabilization, along with sufficient river flow, that the bulk of the wastes can be returned during early spring. With the inception of this development, it has also been to the company's advantage to follow this latest method in mid-campaign prior to river freeze-up.

Each of the factories pump 5 to 6 M.G.D. on the average from the rivers. In the case of Moorhead and Crookston approximately 0.2-0.3 M.G.D. go to the pulp drier ash Hume system, where the screened water overflows directly back to the river. With the use of fresh water or seal tank water the BOD is normally in the range of 15 to 25 ppm. The re-use of pulp press water in diffuser supply has materially reduced the load to the ponds. Pulp press water, with BOD values of 2,000 - 4,000 ppm, and a volume of 130 - 160 gallons per ton of beets, can be quite offensive in a lagoon.

Recirculation of faenger water at East Grand Forks has also aided in reducing the over-all load. In addition, recirculation of flume water from 15 to 55%, has decreased the water requirements. However, this practice does tend to increase the BOD of the final water entering the ponds. A traveling screen arrangement at Moorhead and Crookston and a traveling drag at East Grand Forks remove a great portion of the bulky solids. Admittedly, a vibrating screen system would be more effective. Increased re-usage of partially stabilized pond water in beet fluming and segregation of condenser water and re-use alter cooling are quite probable as the problem of waste disposal becomes more acute.

The Moorhead factory lagoon system consists of two areas 45-acres by 13-feet deep, one 50-acres by 10 feet, one 43-acres by 6 feet, and a limepond of 34-acres by 6-feet. The arrangement is such that all water leaves through one pond into a 24-inch Parshall flume and hence into a 30" pipeline to the river. Crookston is provided with three 48-acre waste water ponds by 13-feet deep and an 11-acre lime pond. The water discharges through an open ditch to the river.

The new East Grand Forks lagoon of 840 M.G. total volume, has a surface area of 155 acres and effective depth of 17 feet. It is divided by dikes and breakers into eleven bays in series. The breakers are arranged to allow for maximum water travel from inlet to outlet. The lagoon is laid out with a three-foot barrow pit which allows for the retention of about 110 M.G. to provide seeding action for the next campaign. The outlet on through a Parshall flume is designed for a maximum discharge of about 17 M.G.D. Average DO and BOD results for the greater part of the 1961 campaign at East Grand Forks are presented in Table 1.

The lagoon system of waste water disposal is not problem free. Cities are closing in on the once rural areas and the public is generally not in favor of these installations, largely because of the odor problem. It has been found that except for the lime ponds, this nuisance is usually noticeable the first week of the spring discharge (as the result of anaerobic decomposition). The reduction of various sulfur-containing compounds to hydrogen sulfide does create an offensive odor. The gas appears to build up a substantial pressure under ice cover as evidenced by the turbulence at the outlet. Light to moderate winds frequently carry the odors over the urban areas while strong winds, particularly after the ice is out, raise havoc with the dikes. Other objectionable features are dike maintenance, solids removal and seepage to adjoining properties. Probably the main concern is the build-

up and retention of the flora and fauna responsible for the necessary biological and bacteriological activity.

An intensive study on the lagoon method of disposal of beet sugar wastes was performed at Moorhead in 1949 - 1951 by the Minnesota Department of Health in collaboration with the U. S. Public Health Service. The purpose was to determine the amount of pollutional constituents in the waste and the degree of reduction of these constituents which occurred by lagooning the waste water during campaign and after winter storage.

The following conclusions were reached:

1. The initial rapid settling of suspended matter appeared to account for all the reduction found in total amount of 5-day BOD, COD, and total solids over the entire period of lagoon storage. The studies indicated that 53% of the 5-day BOD, 87% of the total solids, and 97% of the suspended solids were removed by lagooning.
2. A high ratio between total solids and suspended solids indicates that a large part of the waste present in the lagoons are in true solution even at low temperatures.
3. The total amount of constituents as BOD or solids remain essentially the same over winter lagooning. A BOD of 32 for the ice phase was used in the calculations.
4. An average of 20,300 pounds of 5-day BOD per day were discharged to the lagoons. This was equivalent to 7.1 pounds of 5-day BOD per ton of beets sliced and an average 5-day BOD concentration of 455 ppm.

   Total solids of the waste water to the lagoons averaged 6,470 ppm, equivalent to 96 pounds per ton of beets sliced, while suspended solids averaged 4,920 ppm, equivalent to 75 pounds per ton of beet sliced.
5. There was little or no biological activity effective in reducing the strength of the waste at the near or below freezing temperature.
6. The rate constant k appears to be of the same order as that for domestic sewage.

It has generally been established that larger forms of aquatic flora and fauna originally present in the lagoons tend to disappear as the season progresses. Likewise it has been noted that the coliform group of bacteria decrease, for example, from a maximum MPN per 100 ml of 350 x 10° in mid-September to .13 x 10^6 in mid-March of the following year. Similarly, the well-established volumes of plankton show a trend toward extinction as campaign progresses. We usually find that as the depth of the water is lowered, allowing for sunlight penetration, that by the middle of May a red algal growth becomes quite pronounced.

This condition continues for about a period of a month when the green algae take over. As this growth increases a definite rise in pH is noted, and provided the BOD has already decreased to about 100 ppm, a super-saturation in respect to dissolved oxygen to the extent of 20 ppm is likely to occur. This, of course, brings about a rapid BOD reduction. For example, a pond of about 250 ppm BOD on June 1 dropped to 00 ppm BOD on June 10. This may be attributed at least in part to the condition of aerobic decomposition in the shallower water, with the ensuing production of carbon dioxide, carbonates, nitrites, and nitrates, which have been reported to stimulate the growth of algae.

To obtain as much bio-activity as possible during the warmer months of campaign and a continuation in the spring months, an application of a commercial enzyme material was first tried during the 1958 campaign. At that time 300 pounds were added to the south pond. However, since the enzyme systems were reported to be inactive at temperatures below 40° F, little benefit was expected that fall. The following spring in a series of analyses on the treated and untreated ponds and extending from April 15 through June 5, the untreated pond BOD average was 370 ppm as compared to 225 ppm in the treated south pond. With this note of encouragement, the treatment was extended in 1959 campaign, when 700 pounds of the material were applied to all ponds, including 300 pounds to the lime pond. The application to the final pond, through which a small discharge was maintained, was withheld until November 1. Again during the 1960 campaign a further quantity of the enzyme material was added concentrating more on the lime pond and the two ponds farther out. Of the 700 pounds used this past campaign, over half was applied to the lime pond.

The significant reductions in BOD at Moorhead prompted us to expand the program this past season to include Crookston and East Grand Forks. A total of 700 pounds of the material was added at the East Grand Forks lagoons at a rate of 50 pounds per day. Unfortunately, the comparative survey may not be as gratifying as we had anticipated, since each factory discharged from 120 - 190 M.G. of waste water during this campaign. We trust that the material Avas distributed so that a good portion remains.

For the program at Crookston, where high lagoon BOD values have been most prevalent, one ton of Milorganite, a product of the Sewage Commission of Milwaukee, Wisconsin, was used. It is difficult to get specific technical data on the commercial enzyme product, other than advice on the use and application. The supplier advised that the enzyme-containing material was com-

pounded specifically for the type of waste material and pond conditions existing at our plants. It has been formulated to give the best action in darkness and under anaerobic conditions, with the water depth at least 3.3 feet. An individual in the fermentation industry suggested that it is basically a dried activated sewage sludge, with very little enzyme activity and a relatively low count of viable organisms. This has been somewhat confirmed by our bacteriologist who reported finding 7,400 bacteria, 0 yeasts and 10 molds, per gram. Somewhat disappointing were the lower results on a sample of Milorganite, with 220 bacteria, 0 yeasts, and 20 molds per gram. It may not be possible to obtain as satisfactory results with Milorganite. However, the fact remains that although we know little of the actual mechanism or the constituents responsible for the decomposition and stabilization of the waste products, the enzyme material has aided in providing a good biological environment.

Although these lagoons are usually considered to function under anaerobic conditions, the reaction in them cannot be so considered at all times because of the relatively large surface area. The over-all process of decomposition involves a number of factors. The various microorganisms consume the colloidal and dissolved solids for cell division and metabolism and, in the process, secrete enzymes which are capable of peptizing or liquifying collodial particles. It is reported that certain oxidative enzymes of microorganisms may be effective whether the organisms are alive or dead, provided that the enzymes have not been destroyed by the killing action. Such systems could be a contributing factor in our lagoon stabilization. Protozoa and various macroorganisms are capable of ingesting particles of organic matter. Upon occasion, during the summer months when the lagoon is quite active, large populations of organisms, such as the Crustacea have been seen. It has been suggested by wild life authorities that the many migrating birds who rest at the lagoon, contribute to the pond seeding.

We can detect to some degree the relative activity within a lagoon by the conventional BOD analysis. By this we do not. mean to imply just the comparison of results as the days or weeks progress, but another factor. This is the comparison of incubated dilution samples seeded and unseeded, using an acclimated seed source. For a true indication of the BOD of the lagoons, we find it necessary to seed the samples in the early months when a more sterile condition prevails.

It is normal to expect a gradual build-up of the BOD concentration in the waste water both to and from the lagoons, as

campaign progresses. Input values range from 150 to 250 ppm, increasing to 1,200 to 1,500 ppm. After a certain stage is reached, the biological activity declines and the effective BOD removal is mainly that of a decrease in the total and suspended solids. It is felt that the new Fast Grand Forks system functioned exceptionally well in the first year of operation. A tabulation of the data presented (Table 1) summarizes the analysis for most of the campaign. Based on the average of 445 ppm BOD of water entering the lagoons from September 80 to November 29 and a BOD of 195 ppm at the outlet during the October 19 to November 29 period of discharge, a 56% reduction was effected.

Some positive indications as to the merit of enzyme treatment is provided by some examples. A series of eleven comparative tests was made at Moorhead from October 16 to December 16, 1959, during which time the BOD loading to three ponds was approximately the same. The south pond had been treated with 300 pounds in 1958 and 100 pounds in 1959, the east pond with 200 pounds in 1959, and the north pond with 100 pounds late in the season of 1959. The south pond, with well-established bioactivity, averaged 252 ppm BOD compared to 563 ppm in the north and 380 ppm in the east. With a large volume discharge from the lagoons in the spring of 1960, the water level was down to the barrow pit or permanent retention volume before any further significant reduction in BOD could occur. However, with the well-seeded water remaining and the additional enzyme treatment that fall, the south pond BOD values were reduced to, and remained at, the lowest levels in our experience. These BOD values ranged from 18 to 135 ppm up to the middle of December, while only a 150 ppm BOD was noted on March 22, 1961, compared to 875 ppm in the north pond. As this north pond provides the direct connection from all other ponds to the outlet, it could be expected to be well provided with seed from the other ponds, but the effectiveness is lost on the discharge to the river before any appreciable action can occur.

The continued application of the enzyme material to the Lime pond area at Moorhead has also proven to eliminate a high BOD and odor problem. It is not uncommon for settled lime flume water with a highly variable initial BOD to increase to some extent as the pond starts working. Lime water BOD values have been as high as 8,000 ppm, decreasing by natural action over summer to 50 ppm or less. It was noted at Moorhead in the spring of 1960 that this pond BOD dropped from 5,900 ppm to 2,000 ppm in the three-week period of May 26 to June 16. On May 25, 1961, the BOD was 750 ppm and the pond area com-

Table 1.—DO and BOD averages, East Grant Forks — 1961 campaign.

	Red River												Waste water				
Dates	Junction with Red Lake Riv. DO[1]	BOD[2]	Above dam DO[1]	BOD[2]	300 Yards below dam DO[1]	BOD[2]	3 Miles below dam 1)0'	BOD[2]	16 Miles below dam DO'	BOD[2]	28 Miles below dam DO[1]	BOD[2]	To ponds BOD[2]	Bays 1-2 BOD[2]	Bays 5-6 BOD-	Bays 9-10 BOD[2]	Outlet at ponds BOD[2]	Outfall at river BOD[2]
9/30-10/18	9.4	4.8	8.6	5.9	9.7	5.1	8.7	7.6					400					
10/19-11/29	13.6	5.7	11.2	7.6	11.9	11.9	11.0	20.0					464				195	210
11/30- 1/12	9.1	3.8	8.8	8.8	11.3	7.2	9.4	16.3					619					10
9/30- 1/12	11.4	5.0	10.0	7.5	11.3	9.1	10.1	17.0	8.1	9.2	8.1	7.9	499	535	468	349		166
Minimum	0.5	1.4	1.8	2.8	7.8	3.8	5.0	5.8	1.4	3.8	0.9	3.2	230	384	300	100	20	
Maximum	21.7	9.8	17.3	13.8	14.6	22.4	14.6	34.8	12.6	18.0	12.4	24.4	1320	1020	848	777	543	

[1] DO = ppm Dissolved oxygen

[2] BOD = ppm 5-day Biochemical oxygen demand

pletely devoid of odor. As of January 24, 1962, a BOD of 910 ppm Avas reported, a significantly lower value for this period than noted in the earlier years.

In conclusion, it is possible to effect rather dramatic results with a treated lagoon system. This should be as true if not more so in areas not subjected to such severe winters and where the waste water can be so diverted to maintain higher lagooned water temperatures over an extended period of time. Although our company has been able to discharge factory waste water before a practical maximum BOD reduction has been obtained, the loading has been in the range of 3.5 to 4 pounds of 5-day BOD per ton of beets sliced. This is certainly the maximum that could be realized under proper conditions and with sufficient lagoon capacity.

Effects of Defoliation and Reduction of Stand on Yield of Sugar Beets in Southern Alberta[1]

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In southern Alberta insect pests often cause serious damage to roots or foliage of sugar beets. Prior to thinning, flea beetles, *Phyllotreta* spp., may cause serious defoliation. After beets are thinned the sugar beet webworm, *Loxostege sticticalis* (L.), the beet leaf miner, *Pegotnya betae* Curtis, and the spinach carrion beetle, *Silpha bituberosa* Lec., may cause extensive damage to the leaves. The sugar beet root maggot, *Tetanops myopaeformis* (Roder), the red-backed cutworm, *Euxoa ochrogaster* (Guen.), and the three wireworms *Limonius californicus* (Mann.), *Ctenicera destructor* (Brown), and *Hypolithus bicolor* Esch. attack the root and may kill young beets. There is little information concerning the amount of damage the plants can withstand or the level of protection required, and therefore the value of treating with insecticides cannot be adequately estimated beforehand.

In England, Jones *et al*.. (5)[3] found that 50, 75, and 100% defoliation of sugar beets in the 4- and 8-leaf stages reduced yields by 5, 10, and 27%, respectively.

In Montana, Morris (6) found that complete defoliation of sugar beets in late June or early July reduced yield by $^1/_4$ and 50% defoliation reduced yield by 1 6. Afanasiev *et al.* (1), working in the same area, reported that up to 75% defoliation reduced yield of roots by amounts not exceeding 6% and yield of tops by amounts not exceeding 20%,. Complete defoliation resulted in reductions in foliage weight of up to 80% and a 23 to 27% reduction in beet yield. The greatest loss of top weight occurred when plants were injured late in the season.

The following experiments were conducted to determine the effects of defoliation and of reduction of stand on the yields of sugar beets grown in southern Alberta so that the economic significance of damage caused by sugar beet insects could be assessed.

Materials and Methods

In 1960 and 1961 experiments were carried out on irrigated land near Lethbridge. The soil was a silty clay loam with a pH of 7.7. Plots had been summer fallowed the previous year and each had received an application of ammonium phosphate (11-48-0) at 100 pounds per acre prior to seeding. The sugar beets

[1] Contribution from the Entomology Section. Canada Agriculture Research Station. Lethbridge, Alberta.

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[3] Numbers in parentheses refer to literature cited.

were seeded in rows spaced 22 inches apart at a rate of 6 to 7 pounds of seed per acre, using a commercial shoe drill. Tn 1960 seeding was done on May 9 but in 1961 the necessity of irrigating an abnormally dry seedbed followed by inclement weather delayed seeding until May 24. The stands were thinned to 120 beets per 100 feet of row in 1960 and, because of reduced germination, to 100 beets per 100 feet of row in 1961.

After thinning the stand was divided into randomized blocks containing plots 35 feet long and 4-rows wide. In 1960 the treatments were replicated four times and the plots irrigated four times. In 1961 two tests were set out as follows: one, consisting of five replications, was irrigated four times during the growing season; and the other, consisting of four replications, was irrigated twice. In 1960, 8 inches of irrigation water were applied to the plots. In 1961 the experimental area was irrigated with 1 inch of water prior to seeding. During the growing season the plots, irrigated four times, received 9 inches of water while the ones irrigated twice received 4 inches.

To determine the effects of defoliation, treatments were carried out 45, 60, and 75 days after seeding. On each date 25, 50, and 75% of the foliage of every beet in separate plots was removed. Transverse cuts were made through, each leaf to remove the appropriate amount of leaf area. The effect of stand density on leaf- and root-yield was determined by removing every second or every fourth beet in other plots 60 days after seeding.

In 1960 flea beetles were controlled with insecticides. In 1961 the sugar beet root maggot was found for the first time in beet plots at the Research Station. Beets that were attacked bv this pest were removed together with the adjacent soil and replaced with healthy transplants at the same stage of development.

Each year, harvesting was carried out earlv in September before the tops were fro/en. Immediately before harvest the rows were trimmed to 25 feet and the whole plot harvested. The foliage was weighed immediately in the field. The roots were washed and weighed and then sampled with a multi-saw rasp for sugar determination.

All data were compared at the 5% level of significance by a multiple range test (3).

Results

When plots were irrigated four times during the growing season defoliation did not cause significant differences in foliage vields (Tables 1 and 2). A significant reduction in yield of foliage occurred only where the stand was reduced by 50%.

In 1960, 75% defoliation 60 days after seeding resulted in a yield of roots significantly lower than those of the check plots.

Table 1.—Effect of defoliation or stand reduction on yield of foliage, roots, and sugar of sugar beets, Lethbridge, Alberta, 1960.

Treatment	No. of beets (100 row ft)	Treatment	Foliage (lb/ plot)	[1] Treatment	Roots (lb/ plot)	Treatment	Sugar (lb/ plot)
25% defoliation (45 days)	119	25% defoliation (45 days)	262.1	25%, stand reduction	195.8	25%, stand reduction	25.8
50% defoliation (45 days)	123	75% defoliation (75 days)	257.8	25% defoliation (45 days)	190.6	25%, defoliation (45 days)	25.3
75% defoliation (45 days)	119	50% defoliation (45 days)	257.3	50%, stand reduction	185.5	25%, defoliation (75 days)	25.1
25% defoliation (60 days)	121	50%, defoliation (60 days)	249.1	50% defoliation (45 days)	181.8	Check	24.4
50% defoliation (60days)	119	25% defoliation (75 days)	248.3	25% defoliation (75 (lavs)	182.0	50%, defoliation (45 days)	**24.0**
75% defoliation (60 days)	122	75% defoliation (60days)	244.8	Check	**180.8**	50%, stand reduction	23.5
25% stand reduction	94	75% defoliation (45 days)	240.9	50%, defoliation (60 days)	178.5	50%, defoliation (75days)	**23.2**
50% stand reduction	61	Check	239.2	75%, defoliation (75 da\s)	173.4	75%, defoliation (45 days)	23.0
25% defoliation (75 days)	121	25% defoliation (60 days)	222.8	50%, defoliation (75 days)	170.9	50% defoliation (60 days)	23.0
50% defoliation (75 days)	121	50% defoliation (75 days)	221.3	25%, defoliation (60 days)	170.6	25%, defoliation (60 days)	22.5
75% defoliation (75 days)	125	25% stand reduction	217.1	75%, defoliation (45 days)	168.5	75%, defoliation (75 days)	22.5
Check	120	50% stand reduction	176.0	75%, defoliation (60 days)	159.3	75%, defoliation (60 days)	21.6

[1] Means connected by the same vertical line are not significantly different at P = .05.

However, the yields of roots from plots where the number of beets had been reduced from 120 to 94 were significantly higher than where the plants had been defoliated 25% at 60 days, 50% at 60 and 75 days, and 75% at all dates after seeding. It appears that under the growing conditions encountered, 94 beets per plot more closely approached an optimum stand than 120.

In 1961 at the higher level of irrigation a 50%, reduction in stand resulted in a significant decrease in root yield. Root yields from plots in which beets had been defoliated 25%, at 60 days, 50% at 45 days, and 75%, at 45, 60, and 75 days after seeding were also lower than those from the check plots (Table 2). It should be noted that the check plots contained an average of 93 beets in 1961, which was almost the same as that of the stand that had been reduced by 25% in 1960.

A comparison of the total numbers of heat units[3] between the various dates of defoliation and harvest in 1960 and 1961 is shown below:

Year	Total no. of heat units ' growing season	No. of heat units between each defoliation and harvest		
		First (45 days)	Second (60 days)	Third (75 days)
1960	1637	1335	1128	793
1961	1830	1037	767	512

The shorter growing periods available to plants for recovery from defoliation in 1961 may account in part for the enhanced effect of defoliation on yield of roots.

At the lower level of irrigation there were no significant differences in yields of roots regardless of treatments. Beets irrigated four times, however, produced greater yields of roots than did those irrigated twice. With the two additional applications of water, yields in the check plots were increased by 60.7 pounds per plot (54.9%).

There were no significant differences in percentage sugar among treatments at any level of irrigation. In 1961, however, percentages of sugar were higher at the lower level of irrigation.

In 1960 yields of sugar from check plots (120 beets per plot) were significantly higher than those from plots defoliated 75%, 60 days after seeding. Yields of sugar from plots in which beet stands had been thinned by 25%, (94 beets per plot), however, were significantly higher than yields from stands subjected to

[4] One heat unit is one degree above 50° I- for 24 hours and is based on the mean of 24 hourly temperature readings.

Table 2.—Effects of defoliation or stand reduction at two levels of irrigation on yield of foliage, roots, and sugar of sugar beets, Lethbridge, Alberta, 1961.

Treatment	No. of beets (100 ft)	row-Treatment	Foliage (lb/ plot)	Treatment	Roots (lb/ plot)	Treatment	Sugar (lb/ plot)
			Four Applications ofWater				
25% defoliation (45 days)	90	Check	281.8	Check	171.2	Check	19.6
50% defoliation (45 days)	90	25% defoliation (15 days)	280.9	25% defoliation (75 days)	168.1	25%, defoliation {lb days)	19.2
75% defoliation (45 days)	95	25% (lelioliation (75 days)	278.1	25% defoliation (45 days)	165.3	25% defoliation (45 days)	18.8
25% defoliation (60 days)	90	50% defoliation (60 days)	271.8	25% stand reduction	159.4	25%, defoliation (60 days)	17.7
50% defoliation (60 days)	92	25% defoliation (60 days)	268.8	50% defoliation (60 days)	159.0	25%, stand reduction	17.2
75% defoliation (60 days)	98	50% defoliation (45 days)	268.7	50% defoliation (75 days)	158.8	50%.defoliation (tit) days)	17.1
25% stand reduction	73	75°;, (lelioliation (45 days)	264.0	25% defoliation (60 days)	157.2	50% stand reduction	16.9
50% stand reduction	52	75% defoliation (75 days)	262.6	50% defoliation (45 days)	154.5	50%, defoliation (75 days)	16.9
25% defoliation (75 days)	96	50% defoliation (75 days)	254.9	75% defoliation (45 days)	149.6	50%, defoliation (45 days)	16.5
50% defoliation (75 days)	96	75% defoliation (60 days)	248.0	50% stand reduction	149.5	75%, defoliation (75 days)	16.0
75% defoliation (75 days)	95	25% stand reduction	247.2	75% defoliation (60 days)	145.0	75% defoliation (45 days)	15.7
Check	93	50% stand reduction	202.0	75% defoliation (75 days)	142.8	75%.defoliation (60 days)	15.4
			Two Applications of Water				
25% defoliation (45 days)	97	Check	204.4	25% defoliation (60 days)	120.5	25°;, defoliation (45 days)	16.4
50% defoliation (45 days)	99	25% defoliation (60 davs)	203.0	25% defoliation (45 days)	115.5	25",', defoliation (60 days)	15.7
75% defoliation (45 days)	98	25%, defoliation (75 days)	196.1	Check	110.5	75%, defoliation (45 days)	14.9
25% defoliation (60 days)	98	50% defoliation (45 days)	183.9	75% defoliation (45 days)	108.5	Check	14.1
50% defoliation (60 days)	100	25% defoliation (45 days)	182.7	25% defoliation (75 days)	106.0	25%, defoliation (75 davs)	13.8
75% defoliation (60 days)	101	75% defoliation (60 days)	178.5	75% defoliation (60 days)	100.0	50% defoliation (75 davs)	13.1
25% stand reduction	78	25% stand reduction	175.3	50% defoliation (60 days)	99.3	50% stand reduction	13.1
50% stand reduction	54	50% defoliation (60 days)	172.8	50% stand reduction	97.8	50%defoliation (45 days)	13.0
25% defoliation (75 days)	97	50% defoliation (75 days)	168.4	75% defoliation (75 days)	97.3	50% defoliation (60 days)	12.9
50% defoliation (75 days)	95	75%, defoliation (45 days)	167.8	50% defoliation (45 days)	97.1	75% (lelioliation (60 day)	12.9
75% defoliation (75 days)	96	75% defoliation (75 davs)	161.1	25% stand reduction	96.8	75% defoliation (75 day)	12.2
check	96	50% stand reduction	127.0	50% defoliation (75 davs)	96.8	25%stand reduction	12.0

[1]Means connected by the same vertical line are not significatly different at P= .05

25% defoliation at 60 days, 50% defoliation at 60 and 75 days, 75% defoliation at 45, 60, and 75 days, and 50% reduction at 60 days after seeding.

In 1961, at the higher level of irrigation, yields of sugar from check plots (93 beets per plot) were significantly higher than yields from stands reduced by 25 and 50% or from beets subjected to 50% defoliation at 45, 60, and 75 days and 75% defoliation at 45, 60, and 75 days after planting. Yields from beets defoliated 25% at 45, 60, and 75 days after planting were not significantly lower than those of the check plots.

At the lower level of irrigation there were no significant differences in yields of sugar.

Discussion

It is evident that at higher levels of irrigation, defoliation may have an effect on yield of beets. Its importance will vary with extent of injury, stage of plant development at time of injury, growing conditions immediately following injury, and length of growing season.

Results in 1961 indicated that at the lower level of irrigation defoliation or stand reduction seemed to have no adverse effect on plant growth. During the hot weather that prevailed at times the defoliated plants probably benefited from reduced transpiration while each beet in the thinned stands would have access to more moisture and nutrients and increased light intensity.

Swanson (7) found that less leaf area was required to produce a bushel of sorghum in a dry year than in a wet year but that the highest yields were obtained in seasons of abundant rainfall because there was greater leaf area even though it was less efficient. Eldredge (4) reported that loss of leaves was less detrimental tinder drought conditions and that a moderate degree of defoliation could even increase yields of corn.

Watson (8) reported that the rate of dry-matter production by sugar beets apparently increases as the leaf-area index (leaf area per unit area of land) increases until an optimum value is reached. As the index increases further the rate of dry matter production will decline, probably because the lowermost leaves become so heavily shaded at high leaf-index that their photosynthetic contribution is less than their respiration.

Chester (2) stated that the full complement of leaves functions at a relatively low efficiency and he used the results of other workers to prove that the first leaves lost are dispensable, their removal causing less damage to the plant than further equal in-

crements of defoliation. As more leaves are lost those remaining function more efficiently and their loss is more detrimental to the plant. He also reported that losses in yield are greatest when plants are defoliated in midseason. At this critical stage the foliage has not yet served its photosynthetic function, yet it is too late for a new set of leaves to be produced to compensate for those lost.

The results of the present experiments indicate that sugar beets are able to recover from light to moderate defoliation or stand reduction with no decrease in weight of tops and with little or no decrease in yields of roots and sugar. It appears that an insect infestation causing 25% or less defoliation of beets generally will prove to be of no economic importance. During late June, July, and early August an infestation should be controlled if the beets are defoliated 50% or more. Even when the leaves have been subjected to 75% defoliation it is still possible to obtain a reasonably good crop.

The results of stand reduction indicated that in the Lethbridge area 90 to 100 beets per 100 feet of row were probably closer to an optimum stand than 120. A relatively uniform reduction of stand to as low as (31 beets per 100 feet of row gave a yield as high as that from 1 10 to 120 beets. Thus, where stands are lowered due to insect feeding or other factors such as poor seed germination or phytotoxicity from the use of insecticides or fertilizers, it would seem advisable to leave any reasonably uniform stand containing at least 60 to 65 beets per 100 feet of row rather than reseedthe field.

The results also indicate that there would probably be no increase in yield from controlling insect infestations if moisture were a limiting factor in the development of the sugar beet crop.

Summary

To simulate insect injury sugar beets were defoliated 25, 50, and 75% at 45, 60, and 75 days after planting. Yields of roots and tops of defoliated plants were compared with those of undefoliated plants grown at the same stand density and also with those of uninjured plants from stands thinned by 25 and 50%.

Yields of foliage were the same for all treatments in plots irrigated twice during the growing season and were lower only where stands had been reduced by 50% in plots irrigated four times.

In 1960 in plots irrigated four times 75% defoliation 60 days after seeding resulted in reduced yields of roots. In 1961 yields of roots from plots irrigated four times were significantly reduced

when beets were defoliated 25% at 60 days, 50% at 45 days, or 75% at 45, 60, and 75 days alter seeding. Decreasing stand by 50% in 1961 also reduced yield of roots. At a lower level of irrigation the defoliation and thinning treatment had no effect on root yields. Root yields from check plots irrigated four times during the growing season were higher by 60.7 pounds per 100 feet of row than those from check plots irrigated twice.

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Determination of Amino Nitrogen, Pyrrolidone Carboxylic Acid Nitrogen, and Total Nitrogen With Ninhydrin

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Introduction

For a general picture of type impurities in beet juices, it is important to know the total amino acid constitution and so to have a rapid and accurate method for this determination.

Ninhydrin (1,2,3 triketohydrindene) is probably the most useful general reagent for amino acids. But for colorimetric procedures, it long had the drawback of giving different color depths for different amino acids and was not considered suitable for total amino acid determinations (10)- except by the unwieldly measurement of CO, released in the reaction (11).

However, in the early 1950's, several workers had developed the technique to a point that gave 100% color development from the majority of common amino acids. Troll and Caiman (13) used a phenol-alcohol-pyridine system with a combination of ninhydrin and its reduction product, hydrindantin. Moore and Stein (8) improved their previous method, wherein stannous chloride was used to form reduced ninhydrin, by adding hydrindantin itself with ninhydrin in a methyl cellosolve system buffered with 4N sodium acetate at pH 5.5. Yemm and Cocking (15) used KCN to form hydrindantin directly in a methyl cellosolve system containing ninhydrin and buffered with 0.2 M sodium citrate at pH 5.0.

Although these methods are suitable for total amino nitrogen evaluations in most circumstances, beet juices in processing present an additional problem. Here large amounts of pyrrolidone carboxylic acid (PCA), a compound insensitive to ninhydrin, are formed at the expense of glutamine. The ammonia released in this conversion does react to a significant extent in the ninhydrin systems mentioned.

PCA probably derives entirely from glutamine (4,12,6,2); so that the amount of PCA present in the juices directly reflects the amount of glutamine present in the original beet. So, for all practical purposes, PCA must be considered in the amino acid spectrum of factory juices:

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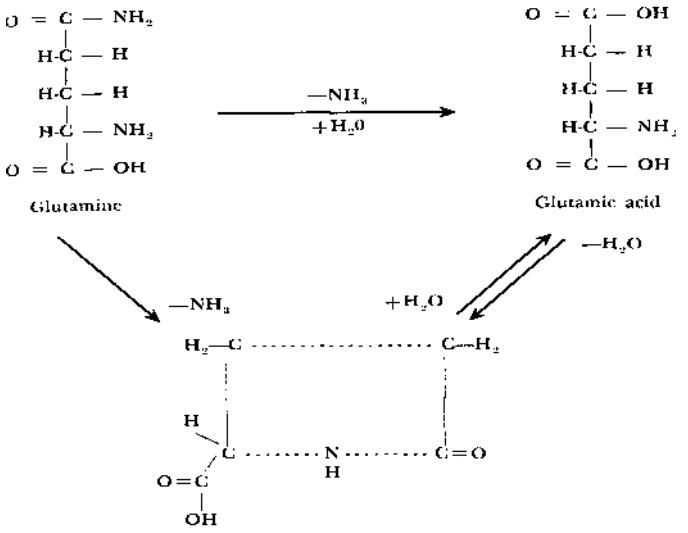
- Numbers in parentheses refer to literature cited.



The method described here is based on the procedure of Yemm and Cocking (15). The important difference is in the buffer system and the way it is used.

To determine total amino nitrogen, a two component acetate buffer is used. For the simultaneous determination of PCA, the caustic fraction of the buffer is used to accomplish rapid and complete hydolysis of PCA to glutamic acid; the acid fraction is then added; the sample is made to proper volume; and the ninhydrin reaction is carried out and colorimetric evaluation made.

Ammonium nitrogen was noted to give a constant response to ninhydrin. This led to development of a technique for Total Nitrogen that compares in accuracy to the Kjeldahl method, but requires no distillation step.

Methods

Determination of Amino Nitrogen

Equipment

Pyrex test tubes, 20 X 150 mm, calibrated at the 2-1/2 ml level.

#2 rubber stoppers

Metal test tube racks
Constant-level boiling-water bath. Some means should be provided to support the racks above tne bottom of the bath —such as a coiled aluminum strip.
Suitable pipettes, burettes, and volumetric flasks.
Colorimeter

Reagents

0.01 M KCN stock solution - 0.1628g KCN made to 250 ml with distilled water.
0.0002M KCN working solution - Dilute 5 ml of stock solution to 250 ml with methyl cellosolve (2 - methoxyethanol)
5% ninhydrin solution - Weight volume solution in methyl cellosolve.
Ninhydrin - KCN working solution (optional) - Mix 50 ml of the 5% nnihydrin solution with 250 ml of the KCN working solution. Allow to stand a few hours before using. Stable about a week.
20% NaOH - Weight volume solution in distilled water.
20% acetic acid - volume-volume solution in distilled water.
Buffer solution - Mix 1 part of the 20%, NaOH with 2 parts of the 20% acetic: acid. pH should be 5.0 - 5.05.
50% isopropanol - Equal volumes of isopropanol and distilled water.

Procedure

To establish a standard curve, prepare a series of dilutions of pure glutamic acid to contain 0 to 0.01 mg of amino N per ml. These are run through the procedure described below for juice samples. A perfectly straight line results when mg of N are plotted against % transmittance on 2-cycle semi-log graph paper.

Juice samples are diluted to contain between 2 and 10 gamma of amino N per ml. Readings are usually within this optimum range with dilutions of 1 to 50-100 on juices through 2nd carbonation and thin, 1 to 100-200 on thick juices and 1 to 500-1000 on molasses.

Pipette 1 ml of diluted sample (or standard) to test tube, add l-i/£ nil of buffer and mix by shaking.

Now add 1.2 ml of ninhydrin-KCN working solution (or 1.0 ml of KCN and 0.2 ml of ninhydrin may be added separately). Mix thoroughly and stopper the tube loosely to prevent undue evaporation.

React the mixture by placing the racked test tube in the boiling water bath for 15 minutes. Then cool in running tap water for about 5 minutes, dilute with 10 ml of 50% isopropanol. and mix.

Read % transmittance on the colorimeter at 570 millimicrons, using distilled water lor 100% transmittance. Translate the amount of amino nitrogen from the standard curve.

Under the conditions of this procedure, ammonium nitrogen gives about 45% as much color as does amino nitrogen. Consequently, if the sample contains much ammonium nitrogen, its amount should be determined and the proper correction made. We determine ammonia by a 10 minute distillation, under vacuum at 55-60° C, from a pH 10.0 borate buffer (14).

To illustrate: a diluted molasses sample contained 1.002 mg of dry matter per ml. One ml gave 32.8% transmittance on the B and L "Spectronic 20". This reads as .0037 mg of amino N from the standard curve - or 0.369%, amino N on dry matter.

The ammonium X had been determined as 0.006%, on dry matter. Therefore, it contributed to the amino X determination in the amount of 0.006 X -45 or 0.003%,.

The actual amino X content was, therefore, 0.369 — .003 0.366% on dry matter.

Obviously ammonium nitrogen is not a significant factor in molasses, or even in thick juice. In all other juices, though— from diffusion juice through thin—it is quite significant and, unless accounted for, can lead to 20 to 30%, error in the amino nitrogen determination.

Determination of Total Amino plus PC A Nitrogen

Equipment

As for Amino X

Reagents

40% XaOH — Weight/volume
Others as for Amino N

Procedure

From a suitably graduated pipette or burette, add 0.25 ml of 40%, XaOH to the test tube containing 1 ml of sample. Place the open tube in a test tube rack in the boiling-water bath for at least 20 minutes. Within this time any ammonia is expelled and PGA is quantitatively converted to glutamic acid.

Cool the sample, add 1 ml of 20% acetic acid, and adjust the volume to the 2-1/4 ml mark with distilled water. At this point the tube contains 1 ml of sample and 1-1/4 ml of buffer exactly as used in the amino nitrogen determination.

Proceed exactly as for amino nitrogen: add 1.2 ml of ninhydrin-KCX; mix; stopper loosely; react for 15 minutes in the boiling-water bath; cool; dilute with 10 ml of 50%, isopropanol; read at 570 millimicrons.

The color intensity now reflects the amount of original amino nitrogen plus the amount of PCA nitrogen.

For example, the molasses sample cited previously gave a reading of 11.7% transmittance after hydrolysis. From the standard curve, this translates to .00744 mg of total amino nitrogen—or 0.743% on dry matter.

Since the original amino nitrogen content was 0.366%, it is apparent, by subtraction, that 0.377% was present as PCA nitrogen.

Determination of Total Nitrogen

Under the conditions of the test just described, ammonium N was observed to give about 45% as much color as an equivalent amount of amino N. The reaction is not completed within the 15 minute time interval, but progresses as time of heating is extended. However, the constancy of the reaction was the clue for possible adjustment of reagents and conditions to allow determination of Total Nitrogen.

Okada and Hanafusi (9) developed an ultramicro-determination of total organic N with ninhydrin. Their method requires the usual digestion and distillation, with ammonia being determined on the distillate.

The procedure devised here requires no distillation. It has given consistently excellent agreement with duplicatd Kjeldahl determinations on pure nitrogenous compounds, molasses and beet juices, and on various feedstufts. In 1958, seventy molasses samples of the previous campaign were divided for the ninhydrin analysis to be used at this laboratory and the Kjeldahl analysis to be made at another laboratory. Results agreed so well, that we now use the ninhydrin procedure for all total N determinations.

After the digestion step the procedure is basically the same as for the determination of amino N, and can be done on the same "production line" basis.

Equipment

Digestion facilities; digestion flasks of 100 ml capacity or large pyrex test tubes and 100 ml Kohlrausch flasks.

Other materials as for Amino N determination.

Reagents

Cone. H_2SO; nitrogen-free Na.. $SO_,$; catalytic mixture of lOOg K_2SO_4+10g HgO+5g selenium

0.1% Methyl red indicator in ethanol

Dilute H_2S0_4 — 0.2 — 0.5 N

20% NaOH

10% ninhydrin in methyl cellosolve

0.0002M KCN in methyl cellosolve (as for amino N)

Ninhydrin - KCN working solution, Mix one volume of ninhydrin with 2 volumes of .0002M KCN.
Buffer (as for amino N) 1 part 20% NaOH - 2 parts 20% 20% acetic acid.
50% isopropanol (as for Amino N)

Procedure

Digestion and preparation of sample

Transfer sample, containing 1 mg N or less, to digestion flask.

Add about 500 mg Na_2SO_4 20 mg of the digestion mixture, and 1-2 ml of **H_2SO_4**

Place flask over reduced heat until any H_2O is boiled off and foaming subsides, then boil rapidly to a water white solution, (about 20 minutes.)

Cool flask sufficiently to add 60-70 ml of H_2O. Then add 2 to 3 drops of methyl red and neutralize as follows: Add 20%, XaOH until the red color is just discharged. Then add dilute H_2SO_4 until the solution is just acid to methyl red. A little excess is of no consequence, but a large excess should be avoided.

Cool to room temperature and make to the 100 ml mark.

Determination of N

Transfer 1 ml of the neutralized digest to test tube. Add 1-1/4 ml of buffer and mix, then add 1-1/6 ml of the ninhydrin KCN solution and mix.

Stopper loosely and place in the test tube rack in the boiling water bath for 15 minutes.

Cool, add 10 ml of 50% isopropanol, and mix.

Read transmittance at 570 millimicrons and translate the reading to mg of N from a standard curve.

To establish the standard curve, any pure N-contaming compound—such as recrystallized hippuric acid— may be run through the digestion step. However, $(NH.,)_2SO._t$ is more easily used and the digestion step can be omitted if desired. Transmittance from dilutions containing 0 to .01 mg N per ml plot as a straight line on 2-cycle semi-log graph paper.

Discussion

These procedures are carried out on several samples simultaneously. For example, our racks each hold 16 test tubes which allow eight simultaneous, duplicated, determinations. While one rack of samples is reacting, another is being prepared. The reacted and cooled samples may be held several hours without color deterioration. Even after dilution with isopropanol, the color is stable two or three hours.

The extreme sensitivity of the reagent demands the use of well cleaned glassware. It is found that occasional boiling in alconox solution is very satisfactory.

The ninhydrin and KCN are mixed as a time-saving measure when many determinations are being made. The mixture is less stable than the individual solutions, and will begin showing a weaker reaction after about a week. It may still be used, provided standards are re-run and readings taken from the new curve. However, once started, the deterioration progresses rapidly and a new solution must be made to restore readings to the original curve.

Occasional standards should be run, but deviations are minor so long as the same cellosolve is being used in the re agents. Since a little color develops from impurities in the cellosolve, the curve must be rechecked when a different batch of this reagent is used. It is customary to re-distill each batch of cellosolve to minimize this impurity interference.

Standard amino acid solutions must be watched carefully. Stock solutions containing 0.1 mg X per ml seem to retain their strength for several days, but standard solutions containing 0.01 mg N per mg, or less, invariably begin showing a weaker reaction after two or three days, even when kept refrigerated. The cause of this deterioration is not known, but it has been encountered by other workers (5).

Most amino acids, based on equivalent amino nitrogen content, fall exactly on the standard curve for glutamic. A few amino acids that give less than 100% reaction by the procedure of Yemm and Cocking (15) were found to give 100%, reaction in the system described here (Tyrosine, for example), or to more nearly approach 100% color formation, such as asparagine.

The exceptions to 100%, color formation are minor, and do not result in gross inaccuracies in analysis of beet juices. Gamma-amino-butyric acid gives about 90%, as much color as glutamic acid. In the total amino acid spectrum of the sugar beet, the error introduced is insignificant.

Asparagine gives about 63% as much color as glutamic acid, on an equivalent amino nitrogen basis. Carruthers *et al. (2)* found asparagine generally less than 1/10 as abundant as glutamine in raw juice. On many California beets we have found slightly higher proportions. Nonetheless, even in raw juice, the error introduced could scarcely exceed 4%. In processing, asparagine is nearly all converted to aspartic acid, which gives 100% color formation in this analysis. Goodban *et al.* (4) report only traces

of asparagine in diffusion juice and molasses. Freed and Hibbert *(3)* show about .07% asparagine or dry matter in thin and thick juices. It is believed, therefore, that small amounts of asparagine existing in beet juice will contribute but minute errors in the determinations for total amino nitrogen. In determination of total amino plus PCA nitrogen, asparagine would convert to asparatic acid to give 100% color formation.

The procedures set down here are not inflexible. Adaptations can be made to suit laboratory conditions or preferences. For example, the curve can reach far beyond the 10 gamma limit by greater dilution of the reactants so long as standards have been set up exactly the same way, Volumes used here may be varied. It seems only important that enough ninhydrin be present; that there be not too much KCN; that pH is proper and that samples are run exactly as are standards. Carruthers *el: al.* (1) have preferred to carry out the alkaline hydrolysis for the PCA determination on a more concentrated sample in a large volume, which is then diluted further and the Moore and Stein procedure applied to an aliquot of the diluted hydrolysate.

In this laboratory, extensive use of the method led to a time saving adaptation that eliminates the necessity of diluting samples containing less than 1.0 mg amino N per ml. Discs of about 8 mm diameter are bored from a pad of filter paper. Pressure of the drill press makes the discs convex. These are laid, convex side up, on a clean surface and a 10 lambda aliquot of the sample is applied with a micro-pipette. The disc will adhere to the tip of the pipette as the sample begins to absorb, which allows it to be lifted over the mouth of the test tube before any of the sample reaches the periphery of the disc. When all the sample has been drawn out, the impregnated disc falls into the test tube. This may be left indefinitely until analysis is ready to be run. If only amino N is being run, 1 ml of water is added and the analysis completed normally. If amino plus PCA nitrogens are being run, hydrolysis is done with 1/2 ml of 20% NaOH or with a little water and the usual 1/4 ml of 40% NaOH.

Thus far there has been no occasion in this laboratory to apply the impregnated disc technique in the determination of Total Nitrogen. One would expect the technique to be usable. However, higher blank readings might occur, and certainly a longer digestion time would be required.

The digestion for Total Nitrogen may be carried out in large Pvrex test tubes (1"x8") and the digest washed into 100 ml Kohlrausch flasks for neutralization. In this manner a clamp arrangement for the tube allows up to 18 simultaneous digestions on a 6 unit electric digestion apparatus.

Volume measurement of the digestion salts is recommended for speed. Suitable measuring devices are easily made.

Several metallic cations interfere with the ninhydrin reaction in an n-butanol system, including the mercuric ion (7). However the digestion mixture used here does not effect the color formation in the cellosolve system. Copper very markedly inhibits the reaction and so cannot be used as a digestion catalyst.

As in the amino N determination, the KCN and ninhydrin may be added separately (1ml of .0002M KCN and 0.5 ml of 10% ninhydrin) to give the same color as the mixture.

Here again some deviation from the procedure as described are possible. For example, it is not essential to use 10% ninhydrin. However, with the stronger solution, color formation has nearly ceased after 15 minutes; so there is less chance of error in the time element.

Summary

A colorimetric method for determining total amino nitrogen is described wherein a simple 2-component acetate buffer is used. The reaction is carried out in a buffered methyl cellosolve system with ninhydrin and KCN. Most amino acids, on an equivalent nitrogen basis, show the same amount of color formation after 15 minutes in a boiling water bath when read at 570 millimicrons.

A method is described for including pyrrolidone carboxylic acid in the total amino determination by using the NaOH component of the buffer in a brief hydrolysis step, followed by the addition of the acetic acid component and carrying out the remainder of the amino-N procedure.

A method is described for the determination of total nitrogen by using a micro-digestion procedure, neutralizing and diluting the digestion mixture, and then performing the ninhydrin reaction on an aliquot of the diluted digest.

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Response of Sugar Beet to Date of Planting and Infection by Yellows Viruses in Northern California

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A common observation in Northern California in recent years has been that sugar beets planted May 1 and later appeared free of the yellows viruses, whereas, early planted beets were usually severely diseased. In 1958 beet yields in California were generally low and symptoms of yellows diseases abundant. In that year, an extensive survey by the Spreckels Sugar Company of beet fields in California central valleys indicated 12% greater root production and 2.1 percentage points higher sucrose concentration of crops planted *in* May compared to those planted in April (Lauren Burtch, unpublished data). Lange, in a five-year study of aphid flight patterns, has found that the number of alate green peach aphids increases abruptly in March and April at Davis, and then declines sharply, dropping to low levels in early May (W. H. Lange, Jr., unpublished data). These observations indicate that late planted fields yield higher in certain years because they escape infection by yellows viruses. An experiment was conducted at Davis, California in 1961 to determine the effect of date of planting on sugar beet production under disease and disease-free conditions. Plants iniected and not infected by the beet yellows virus were compared at three dates of planting.

Procedure

Six treatments were planned, three dates of planting with disease-free and inoculated plants at each date. The variety used was Spreckels Sugar 202H. The planting dates were March 2, March 29 and May 2. The experimental design was a randomized complete block with five replications. Plots were four beds wide (2 rows/40-inch bed) and at least 60 feet long. Two beds were left unplanted between each plot to facilitate irrigating adjacent plots at different times and to reduce the danger of aphid movement between plots. All plots were sprayed with demeton (6 to 8 oz in 40 gal H_20 per acre) at weekly intervals from emergence through the first week of June, resulting in 11,7 and 4 applications respectively, for sugar beets planted March 2, 29 and May 2.

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This technique has been used successfully in other areas of California to keep plants relatively free of naturally occurring aphid-borne yellows viruses (l)[2].

When the plants of each planting date attained 10 to 14 leaves (see Table 1 for dates of inoculation plants of the middle 30 feet of the center four rows of appropriate plots were inoculated with strain 5 of the beet yellows virus. The technique used was similar to that described by Bennett, *et al.* (1). Green peach aphids were reared on radish in aphid-tight cages. Colonies were transferred to New Zealand spinach carrying strain 5 of the beet yellows virus 12 to 24 hours prior to use in the field. Portions of spinach leaves carrying *ca* 5 aphids were clipped and placed in the crown of each sugar beet inoculated. Subsequent indexing of aphids used for inoculating sugar beets of the May 2 planting indicated that they were also carrying the beet western yellows virus.

Table 1.—Responses of sugar beets to date of planting and inoculation with beet yellows virus at Davis, California, 1961. Plants were inoculated at the 10. to 14 leaf stage and harvested October 26. Values given are means of five replications. Variety—Spreckels Sugar 202H.

Date planted	Date inoculated	Yellows 8 August	Tons per acre, fresh wt.		% Sucrose
			roots	tops	
March 2	Not inoculated	100	19.7	21.7	11.4
	Mav 8	99	19.8	23.9	9.7
March 29	Not inoculated	79	24.6	21.2	11.3
	Mav 31	89*	21.8	22.9	9.5
May 2	Not inoculated	6	35.4	23.8	12.1
	June 24'	43*	28.9	20.7	11.9
LSD 5%			2.4	ns	1.6

*Significantlv diffc rent at the 5"'\, level from non-inoculated plants of the same plant date.
[1] Subsequent indexing indicated the aphids used for inoculation were also carrying beet western yellows virus.

On April 24 all plots were sidedressed with 190 pounds N/acre by using ammonium nitrate. It was estimated that this amount of nitrogen would be sufficient to prevent a nitrogen deficiency in plants of any planting date. Leaf samples were collected periodically to determine nitrogen status (7). Plants of the early to late planting were thinned April 14, May 11, and June 6, respectively. Percent plants infected with yellows viruses was determined by counting 25 plants in each of the four center rows of each plot. These data were transformed to arc sines before statistical analysis.

On August 31 and again on September 28 two sub-plots (each - rows X 15 feet) were selected from each plot, one from each ' tid outside the middle 30 feet of the center four rows, and harvested. Fifteen roots were taken from each for sucrose and tare

lumbers in parentheses refer to literature cited.

determinations. On October 26 the center 25 feet of the center four rows were harvested. Two 15-root samples were taken from each plot. Data were evaluated by analysis of variance procedures.

Results

Unusually heavy flights of the green peach aphid during March and April made it impossible to maintain disease-free plants of the first two planting dates. By mid-May, however, aphid flights ceased and beets of the May 2 planting remained relatively free of yellows diseases. Visual differences in color of plants of different dates of planting were evident throughout the season. Naturally infected plants of the March 2 planting were severely yellowed by May 31. Beets planted March 29 appeared less yellow but decidedly more so than the non-inoculated plants of the May 2 planting which remained green throughout the season. Table 1 presents the effect of date of planting and inoculation on yellows symptoms and sugar beet production. Table 2 presents the growth and sucrose concentration during the fall harvest season of naturally infected plants of the non-inoculated plots of each planting date. Table 3 shows the nitrogen status of plants at four dates.

Discussion

The original objective, to compare diseased with disease-free plants at each planting date, was not fulfilled except for the May 2 planting date. The experiment did, however, afford an opportunity to estimate the effect of date of planting on sugar beet production under conditions of different levels of natural yellows infection. Decreasing root yield and higher levels of natural virus infection with early planting indicated the severe effect of naturally occurring viruses in this season (Table 1.).

Based on knowledge of how the sugar beet grows with respect to length of the growing period *(5)* and the results of other dates of planting experiments in California (2) and elsewhere (4), one would expect beets planted in March and harvested in October to yield 20 to 40% more than tliose planted in May instead of 44% less as in this experiment (Table 1). Further evidence of the severe effect of naturally occurring viruses was seen in the failure of plots with a high incidence of infected plants to increase in root yield from August 31 to October 26 while plots with plants relatively free of virus increased at the rate of 1.3 tons/acre per week over this period (Table 2).

A measure of the effect of the beet yellows virus in combination with the beet western yellows virus was obtained from the May planting dates where plants remained relatively disease free and inoculation resulted in 43% infection. This level of infection

Table 2.—Root and top production and sucrose concentration at three planting and harvesting dates. Values given are means of five replications of plots naturally infected with yellow viruses.

Date planted	Date of harvest Aug. 31	Sept. 28	Oct. 26
		Roots, tons/acre, fresh wt.	
March 2	19.4	20.6	19.7
March 29	23.1	25.8	24.6
May 2	24.8	30.7	35.4
LSD 5%:	Between plant dates for any harvesting date - 2.8		
	Between harvest dates for a given planting date - 2.7		
		Sucrose %	
March 2	9.5	10.4	11.4
March 29	10.2	10.7	11.3
Mav 2	11.3	11.4	12.1
LSD 5%:	Between plant dates tor any harvesting date - 1.4		
	Between harvest dates for a given plaining date - 1.0		
		Tops, tons/ acre, fresh wt.	
March 2	51.5	29.6	21.7
March 29	28.8	28.8	21.2
Mav 2	24.9	26.0	23.8
LSD 5%:	Between plant dates for any harvesting date - 5.1		
	Between harvest dates for a given planting date 3.4		

caused an 18% loss of root yield compared to non-inoculated plants of the same planting date. The rate of loss per week of infection was 1%. One might expect that 100% infection would have about doubled the rate of loss to 2% per week, a figure that agrees with losses estimated by Bennett due to inoculation with a severe strain of the beet yellows virus (1). Based on this rate of loss and considering plants of the March 2 planting date to have been infected by thinning time a root yield of 49.2 tons acre is estimated if plants of that planting date had remained disease free. A similar estimate for root yield of disease-free, March 2 planted beets of the current experiment is obtained by multiplying the yields of May 2 planted beets by a factor obtained from data of Ulrich and Ririe, in an experiment conducted at Davis in 1954 wherein beets planted March 1 and May 1 remained free of yellows symptoms and were harvested October 15. The ratio of root growth of the March 1 to May 1 planting was 1.39 (6). Under the conditions of the current experiment the loss in root yield of beets planted March 1 and 100% infected with naturally occurring viruses by thinning time is estimated to be 60% (49.2 - 19.7/49.2).

The loss of 2.8 tons of roots/acre, resulting from an increase in yellows infection in the April planting from 79 to 89%, is a further indication of damage that can be caused by severe strains of the beet yellows virus (Table 1).

Table 3.—Nitrogen status of sugar beet plants at four sampling dates. Values are means of five replications and are ppm (dry weight basis) NO_3-N in petioles of recently matured leaves.

Date planted	Date inoculated	Date sampled			
		24 April[1]	18 June	10 Aug.	25 Oct]
March 2	Not inoc.	6600	12400	10400	1600-
	May 8	8800	13800	12400	6700
Marcn 29	Not inoc.		13900	10800	3400
	May 31		15900	14700	5700
May 2	Not inoc.		17200	10300	6600
	June 24		16800	6400	59U0[3]
LSD 5% Level		ns	ns	ns	ns

[1] Just before fertilizing with 190 pounds of N/acre
[2] Two plots less than lOOOppm
[3] One plot less than lOOOppm

Reduction in sucrose concentrations associated with artificial inoculation with yellows viruses (Table 1) were not readily explained by differences in nitrogen (Table 3) or relative growth rates (Table 2). Roots of disease-free plants of the May 2 planting date which were growing most rapidly and taking up larger amounts of nitrate had the highest sucrose concentration. It appears that the effects of the viruses on sucrose accumulation are due to other factors, among which may be destruction of chloroplasts and phloem tissue as described by Esau (3), or increased respiration due to virus multiplication.

Summary

A date of planting study was conducted at Davis, California in 1961. An attempt was made to maintain plants free of yellows viruses at each of three planting dates to compare with plants inoculated with the beet yellows virus. Heavy aphid nights made it impossible to maintain yellows-free plants of early and late March plantings. Aphid flights were greatly reduced by mid-May and non-inoculated plants of that planting date were relatively yellows free. The yield of roots of beets planted May 2 exceeded the yield from beets planted March 2 and March 29 by 15.7 and 10.6 tons/acre respectively. The reduced yields were associated with high levels of infection by yellows viruses. Sugar beets of the March 2 and 29 plantings made little or no root growth from August 31 to October 26, while those planted May 2 increased in root yield at the rate of 1.3 tons/acre per week. May 2 plantings inoculated with beet yellows and beet western yellows viruses were reduced in root yield 18%, with 43% of the plants showing virus symptoms compared to plants of the same planting date relatively free of yellows viruses.

Acknowledgement

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Affiliation of Low Raw Beet Sugar

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Introduction

Affiliation is a process by which sugar can be upgraded in purity without melting and recrystallizing.

The process of affination is used in the sugar refining industry as the first important step in the processing of raw sugar into its finished products. It is accomplished by mixing the raw sugar, which contains a thin layer of mother liquor on each of its crystals, with a saturated refinery syrup. The saturated syrup has little effect upon the sucrose crystals, but takes into solution all of the mother liquor surrounding the crystals. The resulting magma is then spun in centrifugals and washed up to a very high purity. This combination of affination and centrifuging gives almost complete separation of sugar from nonsugar.

Affination, as it is used in the cane sugar industry, is obviously a simple procedure. Its simplicity tends to obscure its value as a step in the process, but its effect upon the economy of refining raw sugar cannot be overlooked.

The advantages and simplicity of affination have not as yet found application in the beet sugar industry of this country. This is due perhaps to the fact that the domestic industry does not have a counterpart for raw sugar. Most raw sugar comes from sugar cane, but in many parts of the world is made from beets. It is a partially refined product that usually is produced in a raw sugar mill where a complete refining job is not attempted. It then has to be shipped to a refinery for further processing into its finished products.

Beet sugar, on the other hand, is manufactured into its finished products in the same plant where the beets are sliced.

Affination need not be reserved for raw sugar as described above. It can be used to process any grade of sugar that has suitable characteristics. For affination a sugar should contain a fairly large and even size grain that will permit washing in a centrifugal without some of the crystals passing through the screen. Also, its nonsugars should be such that they will be taken into solution by the affinating syrup. Low raw beet sugar is such a product except that in many plants, in this country, the crystal sizes are too small and too irregular in size to permit efficient washing in a centrifugal. It follows then, that if the crystals of

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low raw beet sugar could be enlarged sufficiently, affination and centrifuging could be used as a short cut step in the processing of beet sugar.

In 1960 a sugar boiling program was begun at Moses Lake, Washington, to improve the yield on all the pans and to thereby increase the capacity of the sugar end. This program was crowned with considerable success and from it was learned that with proper Draining and boiling procedures a larger grain in low raw massecuite will result.

Encouraged by this knowledge, equipment to affiliate low raw sugar on an experimental basis was installed in the plant for operation during the 1961-62 campaign.

It is the purpose of this paper to report on that work.

Objective

1. To upgrade low raw sugar to high raw sugar purity without recrystallization or excessive washing.
2. To reduce the amount of high raw massecuite boiled.
3. To improve high raw sugar quality by providing more boiling time.
4. Reduce circulating load on the sugar end.
5. To effect steam economy.

Machinery and Methods

The machinery used for affination of low raw sugar at Moses Lake is shown in Figure 1. Low raw sugar is boiled in the low raw pan (1) in the upper right hand corner of the figure.

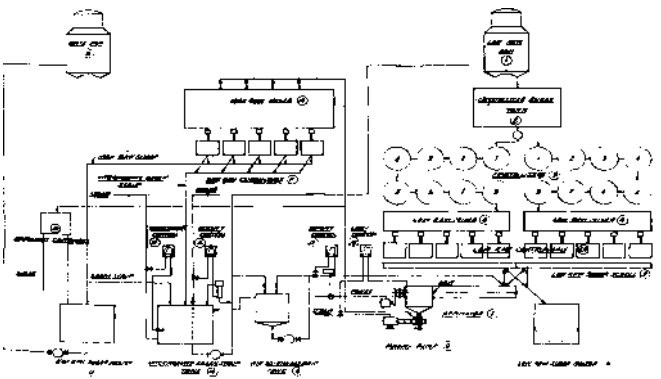

Figure 1

Massecuite from this pan is dropped into a surge tank (2) from which it is pumped into two banks of eight conventional crystal lizers each (3) which are connected in series for continuous opera tion. From here the cured massecuite passes into two low raw mixers (4) each of which accommodates five 42" X 24" X 1600 RPM low raw centrifugal machines (4-a).

The sugar from these machines is discharged into the low raw sugar scroll (5) which brings the sugar to a central point between the two banks of machines. At this point the sugar can be directed from either or both sets of machines into the low raw sugar melter (6) or into the aflinator (7) as desired.

In the affinator, which consists of a 24" scroll case 12 feet long attached to a surge tank and equipped with shaft and set of spiral paddles, the sugar is mixed with intermediate green syrup. In the end of the afhnator, between the scroll case and the surge tank, is a dam designed to keep the paddles of the affinator sub-merged. From the afhnator surge tank the resulting mixture is pumped by a magma pump (9) into the high raw mixer (10) which is equipped with a mixing apparatus. Here the magma is either spun on a continuous centrifugal (19) or mingled with the high raw massecuite and spun on the high raw centrifugals (11). The sugar from the high raw and continuous centrifugals is melted in the high raw sugar melter (12). The intermediate green syrup from these centrifugals enters the intermediate green tank (14) where it is heated and adjusted for R.D.S. before it is pumped through the air disengagement tank (8) on its way back to the affinator (7) and on to the low raw pan (1).

Controls for the affinator consist of: an intermediate green temperature control (15); an intermediate green density control (16); an affinator density controller (17); and a surge tank level control (18).

Methods and Operation

Successful operation of an aflinator is contingent upon there being available a suitable saturated or near saturated syrup for mixing with the sugar. Not knowing which centrifugal machine syrup would work best, provision was made for using either inter-mediate green syrup, high green syrup, or standard liquor. It soon developed that the intermediate green syrup was best adapted for the purpose. This material, however, was laden with air as it came from the centrifugals and the air had to be separated before it could be used. This required heating of the syrup to 90°C in the intermediate green tank and passing it through an air disengagement tank. Before leaving the intermediate green tank the syrup was adjusted to 78 to 80 RDS which assisted in the removal of air and appeared to be the right range of density for

best operation. Through this range of density and temperature the syrup was slightly undersaturated, but when mixed with the low raw sugar and cooled to magma temperature the material was again saturated and there was no significant melting of sugar observed in the affinator.

The "mean aperture" (MA) of the low raw sugar affinated ranged from .0100 to as high as .0142 with the average around .0115. It seemed to mix well in the affinator except that it had a light golden color indicating that air was mixed with it. The RDS of the magma was 91.5 but when this material was mingled in the high raw mixer with the high raw massecuite which has a like density, and was fed into the centrifugals it appeared to have a density very much lighter than this. Further investigation showed the magma to have a weight of only 55 pounds per cubic loot compared to 93 pounds per cubic foot for a massecuite of this apparent density. The difference in the two weights was due to the entrapped air.

There was another effect from this air. It seemed to stay in the Avail of the sugar in the high raw centrifugals and to prevent the wash water from passing through. The result was that the sugar would slump to the bottom of the basket as soon as the machine was stopped.

Laboratory work on methods of mixing low raw sugar and intermediate green syrup pointed the way for remodeling the affinator. After remodeling, the weight of the magma was increased to 76 to 80 pounds per cubic foot. This still was not good, but it made it possible to better load the centrifugals and to better spin the product. The low raw sugar could then be upgraded to a 99 plus purity either when spun by itself or when spun as a mixture with the high raw massecuite. There were times, however, when it was impossible to spin a full load of sugar in the baskets and to maintain the sugar quality at such a high point. There were indications that in addition to the trouble caused by air there was also trouble caused by the smearing action of two sizes of grain. This was confirmed in the laboratory and by the fact that troubles were less whenever the MA of the high raw and low raw sugar were near the same value.

At this point it was felt that both the trouble from air and the trouble from mixed grain sizes could be overcome if the magma could be spun by itself in a continuous centrifugal. The air would be easily disengaged as it spread over the screen of the machine to only a few crystals depth, and the problem of mixed grain size would be overcome if there were no mixing of high raw massecuite with the affiliation magma.

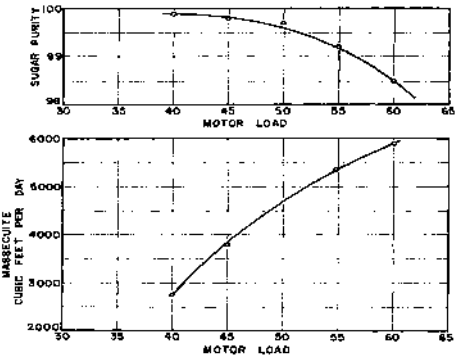

Figure 2

Another thing in favor of spinning this material on a continuous centrifugal is its density. Affination magma need not be as heavy as the massecuite dropped from a white or high raw-pan—it can be anything from 88 to 92 RDS if saturated syrup is used, the difference being only in the amount of intermediate green syrup circulating between the centrifugals and the affinator. The amount of air entrapped in the magma is considerably less at the lower RDS.

For the final two weeks of campaign a Silver continuous centrifugal was used to process the magma from the affinator. After a few adjustments, this centrifugal handled the magma very well. The air in the magma did not seem to hinder the purging ability in any way and varying the magma RDS did not seem to have any effect either. The performance is best given in the form of Table 3 and Figure 2. The machine handled approximately 5000 cubic feet of magma per day, producing a green of approximately the same purity as the green going to the affinator and sugar of approximately 99.0 purity. If no wash water were used in the centrifugal, the purity of the green syrup from the machine would be lower than the purity of the feed syrup to the affinator. This is because the film of low purity syrup around the low raw sugar crystals is dissolved in the feed syrup, lowering its purity When the sugar is washed in the continuous centrifugal, a small portion of the crystals is melted, tending to raise the green purity This explains why the feed syrup to the affinator and the green from the continuous centrifugal end up approximately the same

purity—the impurities on the 95 purity sugar lower the purity, hut the wash raises it back up again.

The amount of magma going to the machine was rather difficult to obtain; however, the figures shown in Table 3 were obtained by weighing the green from the machine and then from the amount of wash water, green **RDS,** and magma RDS, calculating the amount of magma.

Results

Affiliation, was carried out continuously at Moses Lake for a period of thirty-five days. During parts of the first two weeks of this period, all of the low raw sugar from the plant was affinated. For the remainder of the time, only the sugar from one bank of centrifugals or slightly more than half of the low raw sugar from the plant was processed. Table 1 is a tabulation of the results.

Table 1.—Affinator

	Sugar purity		Color			Cubit feet per day	
Week	Slicing Rate	High raw	Low raw	Evap. thick	Standard liquor	Affinator magma	High raw math. spun	High raw mass. boiled
20 Oct. 61	4688	99.0	94.4	71	40		23,800	23,800
27 Oct. 61	4736	99.2	94.4	62	40		23.400	23,400
3 Nov. 61	4675	99.0	94.7	67	4 3		22,100	22,100
10 Nov. 61	4766	99.2	95.1	59	11		22,500	22.500
17 Nov. 61	4773	99.1	94.8	70	52	7,730	16,720	24.450
24 Nov. 61	4661	99.0	95.0	74	53	5.620	16,590	22.210
1 Dec. 61	4664	98.8	95.3	80	55	5.660	17,450	23.110
8 Dec. 61	4711	99.3	95.3	76	54	5.120	18,470	23,590
15 Dec. 61	4642	99.2	95.2	80	58	5,270	19,760	25.030

It can be noted in this tabulation that the amount of high raw massecuita boiled was reduced by as much as 31.5%.

Table 2 shows a chronological comparison between slicing rate, high raw massecuite boiled, and purities of high and low[1]

Table 2.—Affinator

year	Slicing rate	High raw sugar	Purity low raw sugar	Cubic feet High raw filmas % on beets
1961-1962	4620	99.1	94.6	21.7%[1]
1960-1961	4069	99.0	93.0	33.4%
1959 1960	3579	98.6	93.1	36.0%
1958-1959	3539	98.3	92.0	34.9%
1957-1958	3473	98.7	92.8	30.7%
1956-1957	3415	97.6	92.4	30.8%

[1]To date for Campaign

Table 3.—Continuous centrifugal on affinator

Affinator				Feed Syrup			Machine Syrup				
R1)S	Purity	Temp	Ft[1] per day	RDS	Purity	Temp	RDS	Purity	MA	CV	Purity
89.9	88.8	58		75.9	80.7	88	76.4	80.1	.0115	37	99.3
89.9	90.0	56	4908	78.1	70.8	88	76.4	80.5	.....	.....	99.8
90.0	89.9	60	4707	76.8	78.0	95	76.6	80.2	...	....	99.3
90.8	88.8	50	6519	75.1	77.6	96	76.8	78.7	.....	.....	99.4
89.4	89.4	56	5403	72.4	80.8	96	76.4	80.4	.....	.....	98.5
92.1	88.7	60		76.4	78.7	95	78.1	78.1	.0103	25	99.4
80.7	89.9	60	3806	65.8	82.8	96	75.1	81.2	.....	.....	99.8
89.1	90.5	60	4861	75.5	82.2	95	75.9	82.0	.0114	33	99.7
89.4	89.3	60	5837	75.5	80.8	95	75.9	81.7	.....	.....	96.8
89.9	90.2	60	6008	72.8	80.0	92	76.8	81.0	.....	.....	98.5
89.4	89.2	62	5820	75.9	82.4	93	76.8	81.4	.....	.....	98.4
90.3	91.0	60		74.6	81.7	95	76.4	81.7	.....	.....	98.8
90.3	89.2	60	2756	76.8	81.2	98	77.3	80.9	.0128	41	99.9
92.1	88.0	58		72.8	81.8	98	74.1	81.5	.0126	43	99.8

raw sugar. Here again a decrease in the amount of high raw massecuite boiled can be noted for the year 1960-61 and 1961-62. The reduction for the 1961-62 campaign can be attributed mostly ro the process of affiliation but for the reduction in both years some credit must be given to the oxer-all sugar boiling program that was carried on at the plant.

Table 3 shows the results obtained from using the continuous centrifugal on the magma.

Table 4.—Continuous centrifugal, effect of varying amounts of wash water.

Motor Load	Steam	Asp. Steam	Water	Purity Sugar
55	34	60	1.50	09.1
55	34	60	1.25	99.8
55	34	60	1.00	99.2
55	34	60	1.00	99.4
55	34	60	.75	99.5
55	34	60	.50	99.8
55	20	40	1.00	99.6
55	20	40	.30	99.3

Conclusions

1. That low raw beet sugar is a suitable product for affination and that it can be upgraded to 99 plus purity, provided the "mean aperture" (MA) of the sugar is kept above .0100 and the coefficient of variation is kept below about thirty.

2. That the amount of high raw massecuite boiled can be reduced by as much as 32%.

3. That the MA of high raw sugar can be materially increased by utilizing the additional boiling time made available by affination of low raw sugar.

4. That the circulating load on the sugar end can be reduced materially by affination. The low raw sugar, instead of being melted and reboiled, is short-circuited into the white pan.

5. That the amount of coloring matter returning to the white pan is no greater when affinating and continuous centrifuging is in use than when die low raw sugar is remelted and recrystalhzed in the high raw pan. The degradation of sugar is less due to the omission of one boiling step in the process.

6. That there are important steam economies associated with affination. These economies result from the reduction in the amount of high raw sugar boiled and from elimination of dilution and heating in the low raw sugar melter. With affination there is no need for operating the low raw melter.

7. Affination, as it was originally set up and tried at Moses lake, left several things to be desired. First of all, the spinning of magma with the high raw massecuite was a mistake. Under

best operating conditions, variations of MA for both the high raw and low raw sugar were observed. When the values for the two were near the same figure, the results were very encouraging. When the values for the two were quite different the results were poor and discouraging.

Mixed grain and air in the centrifugal feed required light loading on the high raw machines and resulted in a reduction in capacity of the high raw centrifugal station. Since this station is called upon to handle the same amount of material, whether affinating or not, this reduction in capacity became a serious handicap.

8. Affiliation of low raw sugar was much more encouraging after the continuous centrifugal was put into operation. Consistently the machine handled around 5000 cubic feet of magma per day, producing a green of approximately the same purity as the green going to the afrinator. The purity of the sugar coming from the centrifugal averaged over 99 purity, which was the desired result.

Methods of Preparation and Results of Field Planting of Various Types of Processed Monogerm Sugar Beet Seed

P. B. SMITH AND G. E. WALTERS[1]

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The plant breeders have given the beet sugar industry single germ beet seed which is the greatest single boost toward attaining complete mechanization of the sugar beet crop.

In itself, the total benefits of monogerm seed are not completely realized until the seed is properly prepared and properly drilled. Only a small part of the potentiality of this new seed can be realized by simply grading the unpolished seed for size.

Typical single germ beet seed is rather flat with five projections in a star-shaped periphery. The rough shape significantly interferes with the uniform planting of the seed. Also, this rough-shaped cork, which varies in amount with varieties and climate, contains inhibitors that cause irregularity in germination and emergence unless the cork is evenly removed by processing.

When The Great Western Sugar Company first obtained a sufficient amount of single germ seed for study, work commenced on developing devices that might remove this corky material on the periphery of the monogerm seed units. First efforts were with segmenting machines, then decorticating equipment, and from this, progressively to a cylinder with large carborundum stones placed close together on a variable incline arranged so as to rub the seed as it traveled through the drum. This latter piece of equipment had some possibility, but it took as much cork off of the flat seed surfaces as it did off of the harder edge of the periphery. This modification did, however, improve planting ability of the seed by taking off some projections. After being sized in 2/64-inch portions, the finished seed still did not produce the satisfactory metering wanted when tested in drills.

It was found that by taking the monogerm seeds and rubbing them between the palms of our hands, a seed shape something like was sought could be produced. A machine with two continuous rubber belts about 20 inches wide, with one running adjustably at a higher speed was then constructed. The contact surfaces were pressed together with varying amounts of pressure. This turned the seed over and over and, in some respects, accomplished what could be done by rubbing it between the palms of your hands. Thousands of acres were planted with this type of

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preparation but, still, the results were not satisfyng although a seed that planted fairly well was obtained.

The use of a machine that gently removed the skin from rice kernels was the next idea suggested. After observing a commercial rice installation, a McGill miller was purchased to study how much cork need be taken off of the monogerm seed and how much could be removed without damaging germination and emergence.

The next step was the testing of the commercial Engelberg rice polisher which did very gentle polishing without appreciable germ injury. A total of seven or eight different kinds of equipment had been tested before the technique using the Engelberg machine was settled upon. Speeds for operating the huller were worked out with different settings to get the best results with various lots and strains of seed, along with other changes in processing equipment.

A study of typical monogerm seed shown in Figure 1 well explains the progress in removing the corky material.

Polishing the seed makes proper sizing and removal of non-germinating seed pieces mandatory in order to produce the best seed possible for the final purpose of accurate planting. Various divisions of the finished product were all submitted to final planter tests with three makes of drills, which gave us a gauge as to how well they might perform in the field.

Two of our seed processing plants have been entirely rebuilt. Changes include individually-driven Engelberg hullers, new six-screen clipper cleaners, two new Oliver gravity tables, new elevators, and drum separators for edge separation of any double germ seeds. In addition to fungicide and insecticide treatments, a graphite treatment is being added in 1962 universally on monogerm seed for smoother drill operation and more uniform flow of seed as proved in 1961 in commercial testing.

The next question asked was, "How close or narrow should the seed sizes be?" Segmented seed of The Great Western Sugar Company for many years has been 7-10 64 of an inch. This question of segmented seed sizing was subjected to test many years ago, in which sizes were separated into 1/64-inch, 2/64-inch and 3/64-inch size limits. It was found that, with the segmented seed, to get a uniform pattern of singles and a strong, uniform pattern of emergence, a combination of 3/64-inch sizes was needed.

With the monogerm seed, however, the situation is quite different. The germ of monogerm presently is 50 percent heavier by weight than the average bare germs in multigerm seed. This fact has given a much greater proportion of seedlings emerged,

as shown in Table 1 for 100 percent, 90 percent, and 80 percent germinating segmented and monogerm seed.

Great Western monogerm unprocessed strains initially vary between about 85 to 97 percent singles, and Table 1 shows an average of 90. Later field-emergence tables will show slightly more than 50 percent emergence of the monogerm, which then would be compared with about 33 percent emergence for multigerm. For example, 90 percent blotter germination would give five seedlings emerged at ten seeds per foot, while multigerm would have 4.4 plants or less, even though 33 percent more actual germs were planted.

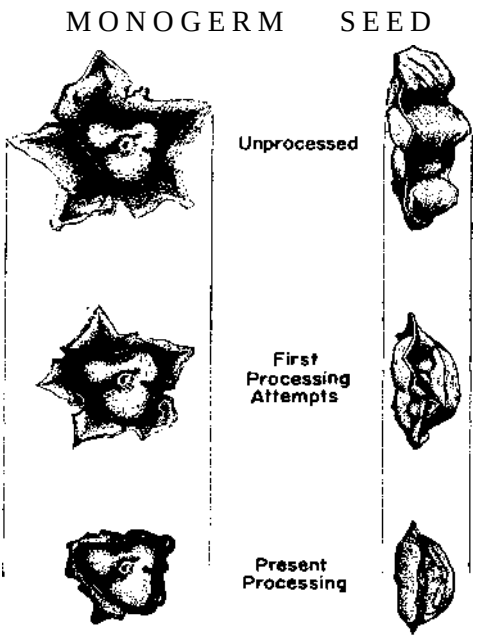

Figure 1

Table 1. — Field emergence comparison of 1960 processed mono—and multigerm seed.

Blotter germination	Actual germs per 100 seeds		Actual germs at 10 seeds per foot		Emerged plants at 10 seeds per loot				Emerged plants at 6 seeds per foot			
					50% Emergence		33% Emergence		50% Emergence		33% Emergence	
	Monogerm	Multigerm	Monogerm	Multigerm	Monogerm	Multigerm	Monogerm	Multigerm	Monogerm	Multigerm	Monogerm	Multigerm
100	110	147	11.0	14.7	5..5	7.4	3.7	4.9	3.3	4.4	2.2	2.9
90	99	132	9.9	13.2	5.0	6.6	3.3	4.4	3.0	4.0	2.0	2.6
80	88	118	8.8	11.8	4.4	5.9	2.9	3.9	2.6	3.5	1.7	2.3

A study on emergence results was started in 1958 with monogerm seed, exploring the possibilities of a range of 1/64, 2/64 and 3/64 sizes. Skips were found in the beet row when sizes were widened beyond a range size of 1/64. When testing seed sizes in drills in the laboratory, more grinding was observed when as much as 2/64-inch seed size range was used and germination suffered as shown by blotter tests. Similar work was done at Colorado State University and by several implement engineering staffs in efforts to match seed plates and rotors to the finely-graded seed.

Accuracy in planting is confounded when monogerm seed is spaced two inches apart in the row and the planter is driven at a speed of three miles an hour, which means actually dropping 27 seeds per second. While the beet drill is not a discussion of this paper, engineers have proved that the beet seed needs to fit, not only the cell diameter of the plates, but also the thickness of the plate. Otherwise, too much cell fill (or too little cell fill) or grinding resulted. Many trials indicate the need for care in the sizing of seed, calibration, speed, etc., as well as careful machining and fitting of drill parts.

Testing of some seven devices for removing the outer cork, shows that a perfect round sphere can not be made out of all the seeds. You will note from Figure 1 that some of the seeds still have a slight amount of projecting cork attached. This means that in one direction those seeds may go through the same cell diameter just as well as a perfectly round polished seed. In another direction the projection will prohibit this and cause the seed to remain in the hopper. The best way to overcome this projection error is to dump the seed cans every eight to ten acres. The quantity of seed will not be great, but it will assist the grower in preventing skips in the field. In the past three years commercial plantings of some 280,000 acres of polished monogerm seed have given large-scale testing among growers.

The Great Western program was for the monogerm seed era to progress gradually with a policy of favoring growers who would agree to provide the type of drilling equipment necessary and adopt some chemical and mechanical practices to reduce labor requirements. This has made it possible to progress more rapidly in the direction toward total elimination of the need for field workers. In fact, in the last five years actual experience shows that Mexican Nationals, for example, now cover 36 percent more acreage during the thinning period than in the year just preceding these years. This type of program has kept failures with the new seed to a minimum.

Table 2.—Various factors as they affect labor performance.

Plot	Seeds per foot	Weed chemical	Inches <containing beets	Percent singles	Machine work	Stand after thinning	No. hrs. labor per acre		
							Thinning	Weeding	Total
I	3	1 Lb. Endothal	12.6	94.45	No	109.8	0	14.5	14.5
2	8	1 Lb. Endothal	27.2	91.91	No	122.0	5.3	8.0	13.3
3	10	1 Lb. Endothal	32.3	83.90	Yes.	131.2	5.5	3.1	8.6
4	10	None	34.3	84.55	Yes	119.4	6.0	4.0	10.0
5	10	None	34.3	84.55	No	123.6	9.1	5.3	14.4
6	10	None	30.8	61.69	Yes	113.6	14.1	3.3	17.4

Plots 1 to 5, 7-8/64" Monogerm Seed; Plot 1, Weeded Once.
Plot 6, 7-10/64" Segmented Seed: Plots 2 to 6. Hoe Thinned 8 Weeded

Table 3.—Comparative results, polished monogerm vs. pellets.

No. of comparisons	Polished Monogerm			Pellets		
	Size	Inches/100 with beets	% Singles	Size	Inches% with beets	% Singles
13	6-7/64	26.7	89.6	1 0/64	23.9	94.1
2	7-8/64	29.7	89.9	12/64	25.3	91.4
22	7-8/64	26.5	87.5	10/64	22.3	91.3
9	8-9/64	24.9	81.9	12/64	23.6	79.1
8	9-10/64	26.5	82.8	10/64	20.2	92.0

Table 4.—Field comparison of processed seed.

Number of Tests	Polished monogerm sized to 1/64" range		10/64" Pellets[1]		7-10/64" Segmented	
	Inches containing beets	Percent singles	Inches containing beets	Percent singles	Inches containing beets	Percent singles
8	28.7[2]	90.6[2]	25.1	94.2		
22	26.5[3]	87.5[3]	22.3	91.3		
9	26.5[4]	82.8[1]	20.2	87.2		
4			25.7	88.9	21.6	61.9

[1]Pellets Made from 7-8/64" Polished Monogerm
[2]6-7/64" Polished Monogerm
[3]7-8/64" Polished Monogerm
[4]8-9/64" Polished Monogerm

Table 2 shows a comparison of five monogerm and one segmented seed plantings with different seeding rates. Three were herbicide-sprayed and three were machine-thinned. A 20-acre field was devoted to this work. The Endothal chemical gave full weed control up to normal thinning size. As can be seen in the column showing total hours for labor, planting to a stand and using Endothal required more time than when more seeds were planted and the mechanical thinner was used.

In 1959 there were 54 field comparisons of four sizes of single germ seed and two sizes of pellets (Table 3). In total, over one thousand 100-inch counts were made. At the same planting rates, the polished monogerm came up an average of 2.45 days quicker and gave 16.3 percent more emerged seedlings, although with slightly less singles in consequence.

In 1960, on 43 field tests, polished monogerm (in four 1/64-inch size ranges) was compared with 10/64-inch size pellets (coated 6-7 64-inch polished monogerm) and 7-10/64-inch segmented multigerm seed. These were planted in Montana, Wyoming, Nebraska and Colorado. As shown in Table 4, the bare monogerm again was highest in emergence and comparable in singles with the coated seed.

Averages of 1960 Results

Seed	% Stand	% Singles	Complete emergence
Polished Monogerm	26.95	87.05	53.9
10/64" Pellets	22.70	90.70	45.4
7-10/64" Segmented	21.60	61.90	27.7

Conclusions

1. After several years of laboratory field tests and commercial use on large acreages, The Great Western Sugar Company is convinced that it is possible to plant the new rice huller polished monogerm seed with considerable success if the seed is sized carefully and the drill seed plate or rotor used has proper tolerances, both in depth and width.
2. It has been advantageous to size the open-pollinated, back-cross-bred monogerm seed to 1/64-inch size ranges in order to have close tolerances for proper drilling which results in precision seed distribution.

3. Seeds less than 4/64-inch in thickness are removed by careful operation of the Oliver Steele gravity table. Each size seed is put over the gravity table before final treatment to bring about maximum germination.

4. Multiple or double germ units are separated from the singles easily in a Carter drum separator.

5. Treatment of the seed with graphite, in additon to ordinary fungicide and insecticide, improves flowability and drill operation.

6. Removal of most of the corky material reduces the effect of inhibitors retarding germination, and uniformly speeds emergence of seedlings two to three days faster than original seed.

7. In over one hundred field comparisons, the rice huller polished seed proved superior to both segmented and coated monogerm in percentage of seedlings emerged.

Effect of Soil Moisture, Nitrogen Fertilization, Variety, and Harvest Date on Root Yields and Sucrose Content of Sugar Beets[1]

D. G. WOOLLEY AND W. H. BENNETT[2]

Received for publication May 24, 1962

The effect of soil moisture on sugar beet yields has been a subject of considerable controversy. Doneen (2)[3] reported that the yields of roots and sucrose were independent of soil moisture when the soil in contact with the roots was maintained above the permanent wilting percentage. Marcum *et al.* (7) maintained soil moisture at several levels above the wilting percentage and were unable to demonstrate differences in root yields. These conclusions are supported by Dahlberg and Maxson (1) and Edlefsen *et al.* (3).

Nuckols (8) increased sugar production substantially by maintaining soil moisture above the 50% available level. With an application of three inches of water in each of six irrigations, he obtained the greatest efficiency in water and soil use. Haddock and Kelly (5) and Haddock (4) obtained marked differences in yield and quality of sugar beets under several soil moisture regimes. Sucrose percentage increased with heavy, frequent irrigations and a deficiency of available nitrogen. Light irrigation and heavy nitrogen fertilization depressed the sucrose percentage.

Hills *et al.* (6) delayed harvest 34 days beyond normal and increased root and sugar yields 4.7 and 0.84 tons per acre, respectively. Sucrose percentage was increased 0.8 percent.

Materials and Methods

A field experiment was conducted at North Logan, Utah, to determine the effects of soil moisture, nitrogen fertilization, harvest date, and variety on the root yields, sucrose, and glutamic acid content of sugar beets. The glutamic acid data are reported elsewhere (11) and the reader is referred there for details of the experiment and the methods used in procuring the data.

Contribution from Agronomy Department, Utah Agricultural Experiment Station, Logan Utah- Journal Paper No. 256. Part of a thesis submitted by Dr. Woolley in partial fulfillment of the requirements of an M.S. degree at Utah State University. The work carried out in cooperation with Western Utilization and Research Division, ARS.

[2]Former graduate student and Dean of Agriculture, respectively, Utah Agricultural Experiment Station, Logan, Utah. The authors are indebted to J. L. Haddock, Research Soil Scientist, Agricultural Research Service; Bliss Crandall, former Statistician, Utah Agricultural Experiment Station; and Rex L. Hurst, Head, Department of Applied Statistics. Utah State University for their assistance in planning and conducting the experiments.

[4]Numbers in parentheses refer to literature cited.

Table 1.—Effects of moisture levels, nitrogen levels, varieties, and harvest dates on sugar beet root and sucrose yields, Logan, Utah, 1955.

Treatment	Root yields tons per acre	Sucrose %	Sugar tons per acre
Moisture			
M_0	22.33	15.34	3.43
M_1	23.21	15.90	3.69
M_2	24.14	16.10	3.89
L.S.D. (.05)	1.06	0.53	0.19
Nitrogen			
N_0	22.61	16.21	3.67
N_1	23.52	15.74	3.70
N_2	23.55	15.39	3.62
L.S.D. (.05)	0.81	0.29	0.08
Varieties			
SP 53101-0	22.77	15.46	3.52
US 22/3	23.69	16.10	3.81
L.S.D. (.05)	0.87	0.31	0.13
Harvest Dates			
Oct. 8	21.20	15.26	3.24
Nov. 11	25.25	16.30	4.12
L.S.D. (.05)	0.55	0.23	0.05

Sucrose content was determined in accordance with the Official Methods of Analysis (9) and with the digestion procedure as suggested by Osborne (10). Sucrose percentages were determined polariscopically.

Results and Discussion

The effects of the various treatments on the root and sucrose yields are shown in Table 1. The M_2 level (80% available moisture) was the only moisture treatment that significantly increased root yields. The M_1 (50% available moisture) and M_2 treatments significantly increased the sucrose percentage over the M_0 (25% available moisture) treatment. Each increase in soil moisture produced a significant increase in total sugar production. The root yields, sucrose percentage, and sugar yields all responded in a linear manner with increasing soil moisture.

The application of 80 pounds of nitrogen (N_1) increased root yields and reduced the sucrose percentage significantly. The N_2 (250 pounds per acre) treatment significantly increased root yields over the N_0 (no nitrogen applied) treatment, reduced percent sucrose compared to the N_0 and N_1 treatments, and reduced the total sugar production compared to the N_1 treatment.

The use of a moderate amount of nitrogen fertilizer with an irrigation schedule that allowed the soil moisture to be maintained near field capacity produced the highest yield of roots and

sugar. Increasing soil moisture above the 50% available level increased root and sugar yields more than did the application of additional nitrogen fertilizer.

Variety US 22/3 was significantly superior to SP 53104-0 in root and sugar production. This result was expected because US 22/3 had been developed for commercial use in the intermountain region, whereas variety SP 53104-0 had been selected primarily for resistance to foliar diseases.

The marked increase in root and sugar production due to the delayed harvest is worthy of consideration. The average increase of 4.05 tons of roots and 0.88 tons of sugar per acre agrees favorably with the results of Hills *et al.* (6) and should warrant a practical appraisal of the risks involved in a delayed harvest. Over the 34-day period, these increases represent average increases of 0.12 tons of roots and 0.026 tons of sugar per acre per day.

The combined effects of the moisture and nitrogen treatments on root and sugar yields are shown in Figures 1 and 2. Both

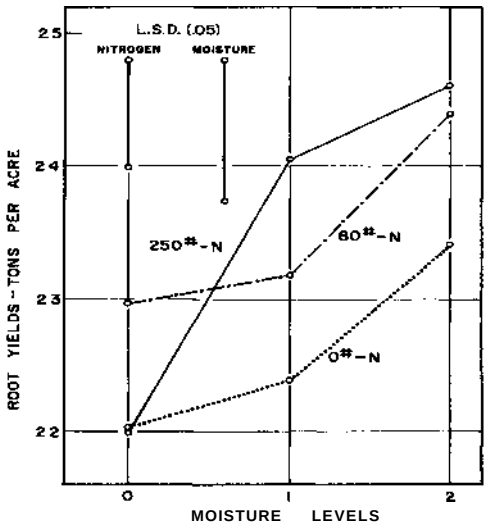

Figure 1. --Effects of soil moisture and nitrogen fertilization on the root yields of sugar beets, North Logan, Utah, 1955.

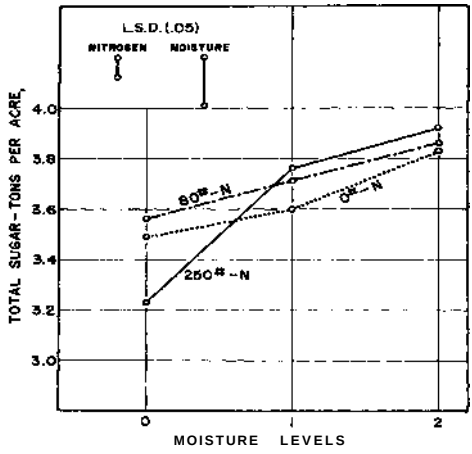

Figure 2.—Effects of soil moisture and nitrogen fertilization on the total sugar production of sugar beets, North Logan, Utah, 1955.

figures point up the importance of the moisture treatments in determining the reaction to the nitrogen treatments. The use of 250 pounds of nitrogen per acre depressed yields below the check when the moisture level was allowed to drop to 25% available before each irrigation. Applying 80 pounds of nitrogen per acre increased root yields at all moisture levels but significantly increased sugar yields at the M_1 level only.

These results agree with Haddock (4) that for any given irrigation regime there is a nitrogen level best calculated to give maximum sugar production. The 27 inches of water applied in the M_0 treatment was sufficient to produce an above average beet crop, yet increasing the amount to 34 inches and tripling the number of irrigations significantly increased root and sugar yields. The amount of water applied above 27 inches does not appear to be as important in increasing yields as the timing of the water applications.

Summary

Two varieties of sugar beets were subjected to three irrigation schedules and three nitrogen fertility levels, and were harvested

on two dates, one month apart. Varieties and harvest dates accounted for significant differences in root and sucrose yields. Specific moisture treatments significantly increased root yield, percent sucrose, and total sugar "production. Nitrogen fertilization increased root yields and total sugar but depressed percent sucrose.

The interaction of soil moisture and nitrogen fertilization suggest that some specific nitrogen level will give best results for any given soil moisture treatment.

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Salt Elimination During Diffusion of Sugar Beets

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For the production of white sugar from sugar beets, sugar must be separated from other soluble impurities in the factory juices. The ease with which this can be done, the ultimate yield of sugar, and the cost of production are largely determined by the amount and character of these impurities. Most of the non-sugars come from the beets, but some are introduced with the water used for diffusion. All of the soluble impurities in the battery supply water do not leave the diffuser in the juice, because a portion diffuses into the pulp and is discarded at the tail of the diffuser. The extent of juice contamination is dependent upon the quality of the diffusion supply water, the draft (9)[2], and the equilibrium distribution of impurities between juice and pulp (9). The contamination of juice by ash constituents of the supply water has been estimated by various authors to amount to 33 to 50% of that present in the water (1,5,6), and water containing 250 ppm of chloride is considered to be unsuitable for diffusion (1).

The present study was undertaken to assess the magnitude of the problem, and to devise a system to reduce the contamination of the juice by ash constituents of the diffusion supply water. Consideration of the theoretical distribution of a soluble additive in a diffuser leads us to the belief that, since the water used for diffusion usually is made up of two kinds, one of which is free of ash constituents, it should be possible to alter the fraction of supply-water solids that are eliminated with the pulp.

Experimental

The diffuser used in these experiments is a laboratory Bruniche-Olsen continuous countercurrent diffuser (4). Beets were obtained from the Woodland factory of the Soreckels Sugar Company. They were washed and then stored in moist pine shavings at 1° C prior to use (4). Sodium was determined by use of a flame photometer attachment on a Beckman DU spectrophotometer, and chloride by means of an Aminco automatic chloride titrator.

For each test in the diffuser, about 200 pounds of beets were removed from storage and sliced into standard cossettes. then mixed in a plastic-lined cement mixer, in order to have a homogeneous supply during the day. Two runs were made in the

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[2] Numbers in parentheses refer to literature cited.

diffuser, the first to establish the distribution of salt from the supply water between pulp and juice, and the second to determine the effect of altered supply water management on this distribution. Feed rates to the diffuser were 9.0 kg. of cossettes and 12.0 liters of water per hour. Product rates were 6.6 kg. of pulp and 12.6 liters of juice per hour. The draft was 146 and diffusion temperature 70° C, with a retention time of 57 minutes for beets and 27 minutes for juice. Measurements were made on grab samples of pulp and juice for sodium chloride, retractometric solids, and polarization sugar.

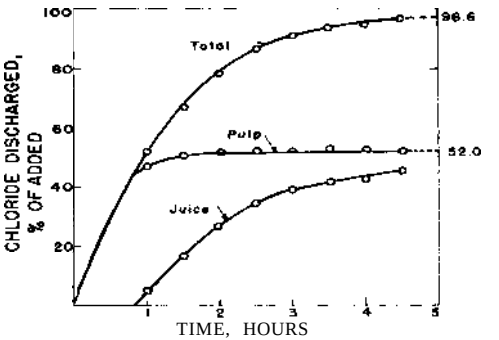

Figure 1.—Distribution of added chloride during normal operation.

For the first run, L21, the diffuser was operated with distilled water as the battery supply until the pulp and juice solids became constant, in order to establish a base concentration of sodium and chloride in the pulp and juice. The battery supply was then changed to 5% NaCl and diffusion continued without other changes. Figure 1 shows the resulting distribution of chloride in the pulp and juice, expressed as a percentage of the chloride added, after subtraction of the base concentration of chloride introduced by the beets. Equilibrium concentration of chloride was reached in the pulp much more quickly than in the juice. Of more interest is the observation that although the chloride in the pulp water reached essentially the same concentration as that in the battery supply, about 48% of the added chloride was carried over into the juice, because the battery supply volume was almost double the pulp water volume. Countercurrent diffusion works very well in reverse, and pulp was shown to be an efficient extractor of chloride ions, but the large excess of

water in the juice over water in the pulp permitted a great deal of salt to leave the process with the juice.

One way to increase the removal of salt by the pulp is to decrease the draft, but this is not desirable because it would severely decrease the extent of sugar extraction. An alternative is to split the battery supply into two streams, introducing one containing salt at the tail end, and the other containing no salt nearer the head end. This would give the desired low draft at the tail end, to favor diffusion of salt into the pulp, and the desired high draft at the head end to favor diffusion of sugar out of the cossettes. Accordingly, a second run (L22) was made in the same way as Run L21, except that the battery supply water was split into two streams. One half of the battery supply was 5% sodium chloride, introduced at the tail end, and the other half of the battery supply was distilled water, introduced through a hole in the trough cover, 12 inches forward of the tail. The effective diffusion length of the apparatus is about 38 inches, therefore, the distilled water was introduced about one third of the length of the diffuser from the tail. The results of Run L22 are shown in Figure 2. The flow rate of the salt supply dropped at 2.5 hours, but equilibrium was reached by 5 hours, as shown by the total recovery figure of 99.8%. Splitting the battery supply into two streams resulted in a much more favorable distribution of salt between the pulp and the juice.

A comparison of Runs L21 and L22 is given in Table 1, in order to evaluate the effect of the split stream. The amount of chloride in the juice at equilibrium is given as the difference

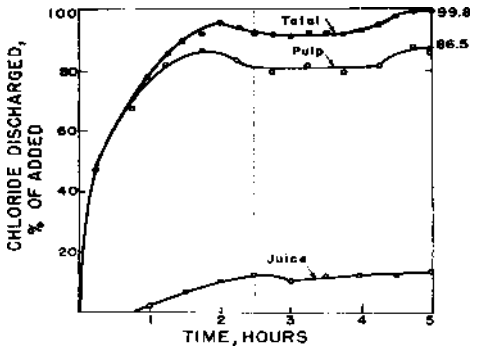

Figure 2.—Distribution of added chloride using dual battery supply.

between 100 and the pulp chloride. Figure 1 shows that the pulp chloride has reached a constant value, but that the juice concentration is still increasing slowly, and should be 48% at equilibrium. The material balance is as good on L22 as on L21, so that the change in flow rate of salt during L22 apparently did not prevent equilibration. There was an increase in pulp sugar with the split stream under the conditions of this experiment. We would expect that loss in a commercial battery would be less, because it would be possible to introduce the pure water at a point proportionally closer to the tail end of the battery, so that the desired high draft would hold for a greater fraction of the total diffuser length.

Table 3.—Equilibrium distribution of NaCl introduced in battery supply water.

	Single battery supply Expt. L2I	Dual[battery supply Expt. L22
Pulp chloride. % of added	52	86.5
Juice chloride, % of added	48'	13.51
Pulp sucrose. pol	**0.21**	0.32
C1:Na ratio pulp	**1.49**	1.53
juice	**1.63**	2.34
Excess Na in pulp, g/kg	0.95	0.85
Excess Na in pulp, Meq/kg	41	37

[1] Estimated equilibrium value. Measured values were 44.6% for **1.21** and 13.3 for 1.22.

The results for sodium are not quite the same as those for chloride. It is apparent that a disproportionate amount of sodium is carried out with the pulp in each case. The sodium deficit in the juice is more apparent in Run L22 where the Cl:Na ratio is 2.34 instead of 1.54 as in NaCl. The total amount of sodium in excess of the chloride in the pulp is about the same in each run. The explanation for this exchange capacity of the pulp is the presence of uronic acid polymers in the pulp (2,3,8). We have found previously that the pulp solids are about 20% anhydrouronic acid (a measure of uronic acid polymers), and approximately two thirds of the acid groups are free carboxyls (7). Since wet pulp is about 5 to 6% solids, this means there are about 37 to 45 milliequivalents (meq) of free acid per kilo of wet pulp. This would mean a total capacity for cation exchange amounting to 0.85 to 1.03 g Na/kg wet pulp. This calculated value agrees with the observed exchange value, even though the excess sodium figure is calculated from a small difference of lather large numbers.

To verify this exchange capacity, a composite sample of pulp from the distilled water portion of Runs L21 and L22 was heated at 70° C for 30 minutes with an equal weight of water containing $CaCl_2$ or NaCl. The calcium ion concentration was then

measured in the supernatant. The results in Table 2 show that there was a cation exchange in the pulp and, furthermore, that sodium could displace the calcium that was already bound by the pulp. The total exchange capacity of this pulp is estimated to be 47 meq, kg of wet pulp (28.8 plus 17.9). This is not a precise estimate for two reasons. First, it is quite possible that a single batch equilibration with sodium ion is not sufficient to displace all of the calcium bound by the polyuronide carboxyl groups. Second, the free carboxyl groups of 28.8 meq kg shown by the more concentrated calcium solution could be high, because some of the calcium may be bound as $(CaCl)^{+}$ instead of Ca^{++} These two effects are in opposite directions and tend to cancel each other. The indicated capacity is sufficient to explain the observed effect of sodium exchange by the pulp in the diffuser.

Table 2.—Exchange capacity of pulp.

Salt added[1] meq/kg pulp		Ca^{++} exchange observed meq/kg pulp	
$CaCl_2$	70.6	28.8	adsorbed
$CaCl_2$	**21.1**	14.2	adsorbed
NaCl	850.0	17.9	desorbed

' Exhausted pulp heated 30 minutes at 70° C with equal weight of salt solution.

The theoretical distribution of a soluble additive in a diffuser has been studied by Stilt. (9). Equations were developed predicting the concentration of additive at any point in the diffuser in terms of the number of cells, the relative volume of liquid in the juice and in the cossettes, the rate of movement of juice and beets, and the point in the diffuser where the additive is introduced. The present data have been compared with the results predicted from these equations. One of the assumptions made in developing the equations was that *d*, the ratio of juice volume to beet liquor volume, remained constant throughout the diffuser. This was not true for these runs, for *d* was 1.92 at the tail of the diffuser, and 1.55 at the head end. The explanation for this may be that when the beets are heated, their capacity for retention of juice is reduced. This reduction of volume occurs simultaneously with diffusion. The reduction in volume is calculated to be about 23% beginning-to-end of the diffusion. The water associated with the pulp was reduced from 18.2 to 15.5 g of water per gram of marc, a reduction of 15%. Thus, it may be seen that part of the reduction in volume is due to trie loss of soluble solids from the water inside the beets. If it is assumed that the volume ratio of 1.92 holds for the portion of the diffuse where active diffusion is occurring, the number of theoretical

cells calculated from the pulp sugar loss of 1.2% is 5.6 cells. This leads to the prediction for Run L21 that the pulp will remove 31.5% of the added chloride while the observed value was 52%.

In the split stream experiment, the battery supply at the tail end was 6 liters per hour of salt water, and the pulp water volume was 6.24 liters per hour, so that d equals 0.96. Using this ratio, and 5.6 theoretical cells, the calculated fraction of chloride in the pulp is 81.1%. The observed value is 86.5%. The advantage of the dual water supply scheme is that in order to achieve this elimination of chloride in the pulp with a single supply of water it would, be necessary to reduce d to 0.96 for the entire length of the diffuser, in which case the pulp sugar loss would be 16.2% instead of 1.9%, as found for Run L22.

In order to apply this system of water management to a factory diffuser, it is necessary to have two sources of water, one containing salts and the other free of salts. Fortunately, this is the case in factories where there is no return of pulp press water, for some of the battery supply is make-up water from outside the factory, and the rest is condensate from the evaporators. In this ease, the make-up water would be added at the tail of the battery, and the condensate some distance forward. The idea of using more than one stream of water into the diffuser can also be applied to the case of pulp press water return, but not in the same way. In this case, the press water contains some sugar which can be saved, and the object then is to *reduce* the amount of soluble solids from the water which will be lost in the pulp. To accomplish this, the pulp water is introduced ahead of the condensate water, at a point where the sugar is slightly higher in the cossettes than in the pulp water. In either case, the application would consist of supplying two different sources of water at two points in the diffuser instead of mixing them outside the diffuser and supplying the mixture at the tail end.

Summary

It has been shown that diffusion water salts can be eliminated in the sugar beet pulp by altering the method of introducing water to the diffuser. In a small continuous diffuser of 5.6 theoretical cells, the salt elimination in the pulp is increased from 52% to 86% by supplying the water containing salt at the rail end, distilled water about one third of the way forward in the diffuser. The increased sugar loss in the pulp is very small by this procedure. The results were found to be in good agreement with the theory developed by Stitt (9). Application of a similar system to the return of pulp press water in order to increase the amount of sugar in the juice is also discussed.

Acknowledgment

The authors wish to thank Mr. Harold Lukens for the sodium analyses, and E. J. Barta, K. Smith, and R. L. Patterson for assistance with the diffuser.

Reference to a company or product name does not imply approval or recommendation of the product by the U. S. Department of Agriculture to the exclusion of others that may be suitable.

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Symposium on New Methods, Procedures and Instruments for Research and Control Laboratories*

Simplicity in Analytical Methods—W. A. HARRIS AND L. W. NORMAN[1] The importance of analytical methods in the sugar beet industry needs no emphasizing. Obviously it is only through analytical procedures that the complex nature of the sugar beet is revealed, and some understanding is obtained of the effects of the various chemical constituents on the growth of the beet and the problems they present in the extraction of pure and marketable sugar.

Through continued development of new techniques, our understanding of the agronomic and processing problems can be broadened, and improved guidance and controls can be instituted in the agricultural and processing phases of sugar production.

Any method of analysis must give reproducible results and an accuracy that is suitable to the problem at hand. But simplicity and speed must be the keynote. This is necessary for routine factory control, or for the handling of the many samples necessary in procuring data in the study of a particular problem.

The fact that a method of analysis has been accepted as standard should not preclude an appraisal of other possible approaches or other techniques. For example, is the calculation of raffinose—from direct and invert polarizations—more satisfactory than its evaluation from a paper chromatogram? Certainly for a large number of determinations the chromatographic approach offers speed and simplicity—along with reasonable accuracy. Again, are the long-used gravimetric and titrimetric methods for invert sugars more preferable than simpler chromatographic or colorimetric techniques? Certainly those methods are subject to inaccuracies if other reducing substances are present. The chromatographic evaluation has even more possibilities now that the Eli Lilly Company has introduced a new reagent that seems to be absolutely specific for glucose.

We know there are materials in the beet that we should be more cognizant of in our efforts to pinpoint individual factors in making beet selections in our breeding programs. We are aware that some compounds or groups of compounds need further study

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[2] Numbers in parentheses refer to references.

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for their effects on juice behavior in processing. For lack of simple techniques, we are inclined to omit analyses of this kind in our studies.

Betaine, for instance, is an abundant nitrogenous component that we really don't know too much about. Techniques for its determination have been somewhat unwieldy for routine analyses. True, the development of the colorimetric determination of betaine reineckate has helped considerably (1)[2], but the determination does require considerable time and manipulation. A simpler technique would certainly be an invitation to include more betaine determinations in our studies.

We think there is a fair possibility that a chromatographic procedure can be developed for betaine. Preliminary work that we have done along this line may offer a clue to something that could lead to such a development.

We have tried, using a variety of solvents, to move betaine reineckate on paper, but instead of moving as a unit, the complex dissociates and only the reinecke is revealed with a ferric chloride spray—and even this seems to fragment into two or three spots. Since some alkaloid reineckates have been separated on aluminum oxide columns *(2),* one wonders if some carrier other than paper might allow the betaine reineckate to move intact. Here the new technique of thin layer chromatography would have application.

At the moment it appears that if paper chromatography could be used, a reagent must be found that will reveal the betaine spot with adequate sensitivity. So far as we have pursued the matter, a solution of about 1% iodine in a water-free solvent—such as absolute ethanol or ethyl ether—has been the most effective reagent. We have been able to detect known betaine spots in the range of 15 micrograms per 15 microliter spot. However, spot intensities have not been uniform. Further, short runs in an isopropanol-benzene-butanol-water solvent failed to separate betaine from interferences. It is hoped that further efforts, by ourselves or one of you, will be fruitful.

An ever-present problem in the industry is that of the tendency of some sugars to form floc in carbonated beverages. Testing for floc is imperative for the proper marketing of our sugars.

Probably the most used and reliable measurements is by the well known "Spreckels Test"—or some variation of it. Yet this test has obvious disadvantages. Precipitation of floc with quaternary amines (3) has not been entirely acceptable.

It has long been known that traces of saponin carrying through to the final product may be held responsible for floc formation (4)—at least to some extent. Consequently, methods have been

devised tor the colorimetric measurement of saponin. These involve the precipitation of saponin from acidic sugar solution, removal of the precipitate on a fine-fritted glass funnel by suction nitration, extraction from the funnel with a suitable solvent, and color development. Antimony pentachloride has been used after extraction with glacial acetic acid (5), and concentrated H_2S0_4 heated with a methanol extract gives a color reaction with saponin ***(6).***

Hibbert and associates (7) recently pointed out the desirability of a more general method of surface active impurities, and have adopted the "polarographic purity" method on Vavruch (8). This method employs the fact that minute quantities of surface active materials strongly suppress the so-called oxygen maxima that are encountered in polargraphic current-voltage curves. On evaluating sugar solutions, the amount of suppression of the peak height indicates the amount of surface active materials present.

The method is rapid—certainly a great advantage for determining immediately whether a strike is suitable for bottlers' trade. It would appear likely that this may be the most suitable and accurate of the objective methods now available.

However, it may be difficult to justify the expense of polarographic equipment, for control purposes, at each factory producing bottlers' sugar, if other means of evaluating floc can keep us out of trouble.

Recently we have started to investigate possibilities of simplifying chemical methods. Two or three things have come to light which appear to offer potential.

First, the filtered floc from an acidified sugar solution may be extracted with H_2S0_4 of 80 to 85% concentration. Heating the extract gave color gradations according to the amount of saponin present-—very much like that obtained with heating a methanol extract with an equal volume of concentrated H_2S0_4 as described by Bauserman and Hanzas (6). This would eliminate the ticklish procedure of adding H_2S0_4 to methanol, cooling and making to volume. Possibly, the drying step would be unnecessary.

Secondly, a much more intense color was obtained when this acid extract was heated for 10 minutes, a few drops of potassium chromate or dichromate added and heating continued for 10 minutes, then chromotropic acid added. Here, only 10 grams of sugar in solution was required to show good differentiation between samples of different saponin content.

Another approach is based on the observation that saponin has reducing properties that might be utilized. The ferric ion, for instance, is reduced to the ferrous ion—which responds to the very sensitive reagent ortho-phenanthroline. The reaction was found to occur in aqueous solutions that were neutral or slightly alkaline, in alcoholic solution, or in methyl cellosolve solutions. The orange-red color developed with about 5 minutes of heating in a boiling water bath and was proportional to the amount of saponin present.

Thus the precipitated and filtered floc could be extracted with methyl cellosolve, a few drops of 1 to 2% solution of o-phenanthroline containing a small amount of ferric chloride or ferric ammonium sulfate added, and the color developed with a few minutes heating. We don't know yet if the reaction is sensitive enough that smaller amounts of sugar solution can be used to cut down on filtration time but obviously the procedure would eliminate some steps in saponin determinations, and requires no unpleasant chemicals.

The most desirable situation, of course, would be to carry out this reaction directly in the sugar solution. Indeed, we did find that 5 ml. aliquots of 40% sugar solutions having different saponin content gave color intensities according to the amount of saponin present. However, all colors were darker than those produced with the isolated saponins. It is probable that reducing sugars would interfere in such a simple scheme.

These are all very preliminary visual observations. As yet we have made no colorimetric measurements to check reproducibility. So we do not offer a new method for floc determinations, but rather, the hope that a simple system can be devised that will lend itself more readily to rapid routine evaluations.

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Dual Laboratory Continuous Dorr System **First Carbonation Apparatus**—F. G. Eis[1] A laboratory continuous Dorr system first carbonation apparatus developed by Dr. R. A. McGinnis has been described in previous publications (2) (3) (4)-. Results of unquestionable significance obtained with the use of such apparatuses have been reported (1) (2) (4).

Various European investigators have preferred using dual carbonators. The determination of the effects of carbonation variables on processing using a single unit requires special procedures for assurance of uniformity of the raw materials being treated. Raw juice is known to be susceptible to changes during retention which have an influence on processing. Increased numbers of tests are often required to compensate for the variability of raw juice when using a single carbonation unit.

Even though the fundamental effects of carbonation variables are well known, at times it is desirable to check the effects of these variables as the processing characteristics of beets are subject to changes. Beets with abnormal processing characteristics sometimes cause operating difficulties and speed in obtaining data is essential for checking the effect of variables to assure factory operation at optimum conditions.

In order to obtain data in as short an elapsed time as possible and to bypass the effects of changes in composition of raw materials, a dual laboratory continuous first carbonation apparatus was constructed. Each unit was built to the design of the original tested apparatus and the units constructed to operate in parallel with separate control of any desired operating variable.

Parallel operations allow a direct comparison of carbonation effluents and a direct measure of the effect of the variable under investigation. Possible inherent differences between the two units, even though constructed as nearly alike as possible, can be compensated for by alternating the test variable by units.

The time required to reach steady state conditions is normally an appreciable part of the total time required for a test. In determining the effect of a variable, a dual unit allows an apprec-

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[2] Numbers in parentheses refer to literature cited.

iable savings in elapsed time as well as man-hours since one man is required tor operation of a single unit but can operate the dual unit without difficulty.

The results of a test on the effect of the point of lime addition to first carbonation can be used to illustrate the performance of the dual carbonation unit.

The apparatus was set up in the factory with raw juice, saccharate milk, and carbon dioxide supplies common to both units. The units were adjusted to operate at a recirculation ratio of 8 to 1 with an equivalent of 2% CaO in first carbonation effluent. Carbonation at 80°C was controlled at an alkalinity of 0.085% CaO. Saccharate milk was fed into the secondary carbonation tank, the gassing tank, of one unit in the normal manner and into the primary tank of the other unit. Samples were taken for analysis after reaching steady state conditions. Settling tests were made on the first carbonation effluent by the Dorr-Kynch method as described by Talmage and Fitch (5). The lime salts and color of thin juice were determined after a batch second carbonation with gassing at the boiling point for three minutes followed by five minutes of boiling. The point of lime addition was reversed between units after the first two samples were taken.

The data indicate that the point of lime addition has a major effect on the results of first carbonation. Addition of lime to the primary carbonation tank rather than the secondary, decreases the settling rate of the first carbonation sludge, and causes an increase in the lime salts and a decrease in the color of thin juice at equal Dorr retention periods.

It is interesting to note that the data on lime salts would not have been considered statistically different at the 95% level of

Effect of point of lime addition in first carbonation

Sample No.	Point of lime addition	Settling capacity lbs. solids/sq. ft/hr	Thin juice Lime salts CaO/100 rds	Thin juice Color 100 (log Tb.) 10 rds, 5 cm cell
1	Primary	13	.221	40
	Secondary	40	.215	47
2	Primary	14	.242	37
	Secondary	56	.178	40
3	Primary	15	.192	37
	Secondary	40	.178	44
4	Primary	15	.254	32
	Secondary	38	.228	50
5	Primary	16	.290	38
	Secondary	40	.237	46
Average	Primary	15	.240	37
	Secondary	43	.207	45

confidence if the samples could not have been paired for statistical analysis. The difference between averages is 0.033 while the LSD_{93} is 0.038 without inherent pairing.

The results of the test reported illustrate the reason for the Dorr Company's choice of the point of lime addition to carbonation. Data were not recorded for rates of filtration but it was observed that filtration of both first and second carbonation juices was decreased by lime addition to the primary rather than the secondary carbonation tank.

The dual laboratory carbonation unit has been found to be highly satisfactory. Its use has allowed a significant savings in elapsed time and man-hours required for testing. Statistically significant results are more readily obtained on factory feed materials since the eflects of changes in composition of the feed are minimized.

Ratio of values: lime addition to primary tank/secondary tank

Sample	Settling capacity	Lime salts	Color
1	.315	1.03	.85
2	.25	1.30	.93
3	.38	1.08	.84
	.39	1.11	.64
5	.40	1.22	.83
Ratio Average	.35	1.10	.82
Difference of ratio from 1.0	.65	0.16	.18
LDS_{95}, from ratio of 1.0	.06	0.13	0.10

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Wet Screening of Sugar Crystals from Low Purity Massecuites and Sugars

—ROBERT R. WEST AND ROBERT S. CADDIE[1]

In any program involving raw sugar boiling improvement (l)[2], it is of great advantage to be able to make routine determination of size and degree of uniformity of sugar crystals in samples of

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[2] Numbers in parentheses refer to literature cited.

sugars and massecuites. Saint and Trott (3), working with raw cane sugar, developed a wet screening method which, with suitable modifications, can be applied to low purity beet house products containing very small crystals.

The test consists of successive washings of the sample with an ethyl alcohol-water solution saturated with sugar at the temperature of the test, before transferring the washed crystals to the top sieve of the selected series.

Pre-treatment Before Screening

High Raw Sugar

Sufficient sample to yield 8 to 10 g of final dried crystals is placed in an evaporating dish and 20 to 30 ml of 90% sugar-saturated alcohol is added. A rubber policeman is used to break up all lumps and mingle the sugar thoroughly with the alcohol so that each crystal is separated and washed by the alcohol. The alcohol is carefully decanted and the washing repeated with a second 20 to 30 ml portion of the 90% alcohol. Usually two washings are sufficient, but if the syrup film on the crystals is not completely removed, a third may be used. A final washing is made with sugar-saturated undiluted alcohol.

High Raw Massecuite, how Raw Massecuite as Spun and Low Raw Sugar

Sufficient sample to yield 8 to 10 g of the final dried crystals is washed as above with successive portions of 20 to 30 ml of 80% sugar-saturated alcohol until no further extraction of color into the alcohol is observed. At least one washing with 90% alcohol is performed with a final washing with undiluted sugar-saturated alcohol.

Low Raw Massecuite as Dropped from the Pan

One washing with 80% alcohol which has been heated to 60°-65°C and saturated with sugar at that temperature is required. To the hot sample (sufficient to yield 6 to 8 g of final dried sugar crystals) direct from the pan is added 20 to 30 ml of the hot 80% alcohol. The mixture is mingled thoroughly with the rubber policeman and decanted as soon is generally sufficient to render the massecuite amenable to further washings with room temperature sugar-saturated 80%, then 90%, and finally undiluted alcohol.

The washed sugar in each case is transferred to the top sieve of the selected series of tared 3-inch Tyler stainless steel sieves immersed in undiluted sugar-saturated alcohol in a $3^{1/2}$ inch cylinder fitted with a gasketed, bolted cover. The cylinder is placed in a Tyler Ro-tap shaker (115 to 120 TPM) and

shaken for 30 minutes. If the washing has been performed properly, this time will be sufficient for all materials. It is important that the alcohol in the retainer is sugar-saturated at the temperature at which the sample will be *shaken,* as the long shaking period results in the retainer and its contents assuming the ambient temperature of the shaker and its surroundings. After shaking, the sieves are removed, separated, and after being allowed to drain, dried with their contents at 105°-110°C for 20 minutes. The screens are cooled, weighed, individual fractions added to obtain a total weight, and the percentage retained on each screen calculated. Generally we express size and uniformity of grain by the Powers method (2)—that is, in terms of "Mean Aperture" (M.A.) and "Coefficient of Variation" (CV).

Notes

1. In making dilutions of alcohol with water, the water normally present in the alcohol is ignored; the 90 + 10 and 80 + 20 dilutions are made volumetrically.

2. The alcohol-water solutions, except in the case of the hot solution used for the first washing of low raw massecuite from the pan, must be saturated with sucrose at the temperature of the area where the washings and otlier manipulations will be performed.

3. If at any time changing from one alcohol concentration to a higher concentration causes the sugar to ball together and refuse to disperse, it indicates that the preliminary washing has not been sufficient, and it is necessary to rewash with the lower concentration of alcohol.

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Process Liquor Color Determination in the Sugar Factory Control Laboratory—ROBERT R. WEST AND ROBERT S. GADDIE[1]

There are many methods used in control laboratories *in* the sugar industry for routine determination of color in process juices. We have tried several over the years, but none has been

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wholly satisfactory. Such a test, to be useful, particularly on dark-colored juices, should accomplish the following:

1. The test should give reasonably accurate results and good reproducibility. This involves a photo-colorimeter, preferably a spectrophotometer.

2. The instrument must be simple to operate, rugged, and not too costly.

3. The test must be quick and simple to perform.

4. If results are to be useful, the color indexes must be equated to constant RDS.

The instrument chosen was the Bausch and Lomb Spectronic "20" spectrophotometer, a grating-dispersion instrument of constant band-width which had been in use for some time in several of the factory laboratories and had proven to be accurate and reliable. To determine the feasibility of such a procedure, we determined first, the absorbance curve of representative standard liquors from 350 to 850 mu; second, the conformity with Beers kaw at numerous points on the curve; and third, the reliability of a table which was to be computed to convert observed colors at any RDS to equivalent color values at a constant RDS.

The absorbance curve showed a plateau of high absorbance in the ultraviolet, falling rapidly to a valley between 550 and 675 mu, then rising again to a sharp peak at 800 mu. Wave lengths in the 775 to 825 mu (red) region did not closely follow Beers Law, but at 440 in the blue region a point was found where divergence from Beers Law over a concentration range of 5:1 was in the order of only 2%. The color density was varied by diluting standard liquors with liquid sugars of equivalent RDS. RDS was varied by simple dilution with distilled water on a weight basis (verified by refractometer). As a result of checking 5 different standard liquor colors at three different densities, it was decided that a table correcting for RDS variations could be calculated with adequate accuracy. The table, correcting all observed colors to 70 RDS was then prepared.

If readings were to be made on the absorbance or optical density scale, it would only be necessary to multiply the reading by a factor $\frac{70}{\text{obs RDS}}$ to correct the absorbance to what it would be if that same sample had been at 70 RDS. In view of the fact that the absorbance scale is logarithmic and necessarily has non-uniform divisions and subdivisions, we decided to use the % Transmittance scale which is linear in calibration and therefore much less subject to misreading by an inexperienced operator.

The table actually converts the %T to a color number which represents (absorbance X 100) of the sample corrected to a standard RDS.

The change to refractometer control for purities made available a 1-normal solution of process liquors, and these solutions are used in the analysis. Excess turbidity in the sample, as might be expected, gives color values which are too high, but the error for factory control purposes usually is not serious.

The procedure established for the factory laboratories is as follows:

The determination is made on the 1-normal solution (26 grams of syrup made up to 100 ml.) prepared for purity and RDS determination. The color is determined using the Spectronic "20" equipped with the blue-sensitive photo cell and matched %" test tubes. The %T at 440 mu is observed relative to distilled water. Using the %T of the 1-normal syrup and the RDS of the undiluted syrup, the color index is obtained from a table. This color index is directly comparable with any other regardless of the original RDS of the sample. The observation must be made at the time RDS and polarization are being determined, as standing for long periods in the diluted state will cause an appreciable alteration in the readings.

Insecticide Residue in Sugar **Beet By-Products—J. R.** JOHNSON AND S. E. RICHSET[1] Interest has mounted rapidly in the past few years in what can be termed side effects or long time effects from the increasing use of standard and new or experimental pesticides, fungicides and herbicides that are used on agricultural field crops resulting in a residual carryover into foods for human consumption. The chlorinated hydrocarbon, DDT, has been given a tolerance of essentially zero in milk by the Food and Drug Administration.

Alarmingly high amounts of DDT have been found in milk in some isolated areas. The source of DDT was traced to alfalfa feed which had been exposed to aerial spraying either directly or by wind drift. This finding focused attention on all livestock feed in that particular area. Other incidents of a similar nature have made it expedient to know something about, the possible level of DDT in dried beet pulp.

The USDA and The Amalgamated Sugar Company, agronomy section, at Twin Falls undertook a series of tests to determine the level of DDT in beet roots grown in soil treated prior to planting with an exceptionally heavy application of **DDT** amounting upwards to 400 pounds of 5% dust, or 20 pounds

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active DDT per acre. The roots and foliage were analyzed after harvest for DDT and were found to contain less than 0.2 ppm DDT which was the lower limit of accuracy of the method employed.

DDT is now registered for use as a soil treatment on sugar beets. However, it is often used for other crops and is known to remain active for several years in soil and will build up due to repeated treatment.

Since DDT was not positively found in the roots, it is logical to assume that there would be a small chance at best for any DDT to survive processing and be carried over into pulp or sugar itself. However, unless the product as sold, pulp in this case, has been tested for DDT it is impossible to state that its presence is negative. Consequently, as a precaution, we have adopted the policy of analyzing weekly composites of all dried molasses beet pulp produced at each of our three pulp driers.

There is no simple analytical method available for the positive quantitative estimation of DDT in ranges of less than 1.0 ppm. We will not attempt to prescribe any particular method at this time. Instead, an attempt will be made to point out a few of the aspects of several methods which may be used to estimate the level of DDT contamination in dried pulp if any is present.

Extraction, cleanup and concentration procedures are common to any method chosen. In the case of dried molasses beet pulp we have adopted the procedure of extracting 50 grams of pulp with 400 mls of U.S.P. chloroform using a Waring blender. The blender is operated through a Power-stat in order to control the apparatus to slow speeds. Mixing is started and stopped on 30 second intervals for a total mixing time of two minutes.

Chloroform is a satisfactory extracting solvent for DDT, hexane, benzene and benzene-acetone or alcohol mixtures are also recommended in the literature.

After extracting, filtering and washing the pulp, the solvent volume is quite large and must be evaporated to a volume of approximately 25 mls. The solvent is then transferred to a 50 ml glass stoppered flask and made to volume with n-hexane.

The cleanup procedure is designed to remove fats, waxes, moisture and any other material soluble in the solvent which may interfere with the subsequent determination. A florisil column topped with anhydrous sodium sulfate is recommended for DDT. The column is first pre-wetted with n-hexane. An aliquot of the DDT suspected solution is added to the column. The column is then eluted with n-hexane at a rate of 8 to 10 ml per minute. 250 mls of n-hexane is sufficient to elute the column thoroughly.

If the colorimetric method of Stiff and Castillo (1) is to be used, treatment of the eluate consists only of evaporation to 1 to 2 mls, transferring to a graduated test tube and further evaporation to dryness with an air stream at room temperature prior to further handling.

If the DDT is to be determined by paper chromatographic procedures, additional cleanup with an acetonitrile extraction is required. This step is necessary in order to yield an essentially pure compound for the chromatogram.

In general the cleanup procedures required for any type of clilorinated liydrocarbon assay for trace quantities is the most important part of the analysis. Plant materials contain a large number of organic compounds all more or less soluble in the solvents used for extraction. In many cases these compounds are present in larger amounts than the pesticide residue sought. For this reason, it is extremely difficult to develop specific methods that are accurate to less than 1 ppm.

After extraction, cleanup and concentration there are several courses open for the analysis of DDT or other chlorinated hydrocarbons. Three will be briefly mentioned in this report.

1. Colorimetric Method.

The only colorimetric method we have used is a modification of the Stiff and Castillo (4)[2] method which is specific for DDT and one of the analogs of DDT. The p,p'-DDT yields slightly more color than o,p'-DDT. Color development depends upon the reaction between DDT and the xanthydrolpyridine - KOH reagent. This reaction is negative for DDT. The lower limit of accuracy is about 0.2 ppm.

This method is satisfactory for control purposes where only four or five determinations a week are required. It will take one technician approximately two days a week for the analysis.

2. Paper Chromatographic Method (2[s]) (3).

In order to prepare a concentrated extract for paper chromatography, the cleanup procedures are somewhat more elaborate. The chlorinated hydrocarbon must be essentially free of other organic materials in order to evaluate the developed chromatogram.

Chromatographic procedures enable us to determine quantitatively some thirteen pesticides if required. Modifications and the proper technique can be extended to the identification of some 114 chlorinated organic pesticides (3). Here again the lower limit of accuracy is in the range of 0.2 ppm.

[2] Numbers in parentheses refer to references.

3. Gas Chromatography.

Gas Chromatography is being developed rapidly for the determination of pesticides. Recent equipment modifications and the development of improved sensing components has made it possible to screen a large number of pesticide residues. The extraction procedure is designed for a catch-all type reaction and cleanup procedures if necessary at all can be rather crude.

Gas chromatographic equipment however is expensive and may not be used extensively by sugar factory laboratories for some time to come.

The following is a list of references which point to the interest and mass of work being done in the pesticide residue field. Many others can be found in 1960 and 1961 issues of Ag. and Food Chem.

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The Interaction of Rates of Phosphate Application With Fertilizer Placement and Fertilizer Applied at Planting Time on the Chemical Composition of Sugar Beet Tissue, Yield, Percent Sucrose, and Apparent Purity of Sugar Beet Roots[1]

J. F. DAVIS, GRANT NICHOL, AND DON THURLOW[2]

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An experiment was initiated in 1959[3] with the following objectives: 1) determining the minimum amount of phosphate fertilizer to be applied at planting time in relation to the total application without significantly decreasing the early growth or final yield of roots, and 2) the amount of preplant phosphate required to best complement the planting-time fertilizer. An additional objective was included in 1961 in which the method of placement of the planting-time fertilizer and its interaction with the amount of preplant phosphate was investigated.

Materials and Methods

This experiment was established at the Monitor Sugar Company farm located near Bay City, Michigan, on a calcareous Kawkawlin-Wisner loam soil complex with pH of 7.5. For the sugar beet crop, four rates of $P_2 0_5$, were broadcast ahead of planting: 0, 200, 400, and 800 pounds per acre. A basic application of 200 pounds of KC1 was plowed under. Treatments were replicated three times in the east-west direction of the field. Superimposed on these areas were three rates of fertilizer: 0, 150, and 300 pounds of 6-24-12 per acre. The 6-24-12 fertilizer contained 2% manganese and 1/2% boron. The fertilizers were applied in two ways: 1) in a band $1^{1/2}$ inches to the side and 3 inches below the seed, and 2) 3 inches directly below the seed. The rows were planted across the plots where $P_2 0_5$, was broadcast. The planting-time fertilizer application was replicated three times. Sixty pounds of N per acre as anhydrous ammonia was applied as a sidedressing to all plots.

[1]Contribution from the Soil Science Department. Michigan Agricultural Experiment Station, East Lansing, Michigan, and Agricultural Department. Monitor Sugar Companv, Bay City, Michigan, and approved by the Director as Journal Article Xo. 2932.

[2] Professor of Soil Science. Agronomist, Monitor Sugar Company, and Assistant Instructor of Soil Science, respectively.

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Plot size was four 28-inch rows by 66 feet. Monogerm beet seed variety SL122 X SP5460 was planted April 13. Stand counts were taken May 11. Plant samples of 100 plants per plot were taken June 2, 1961, oven-dried at 65 degrees Centigrade, weighed and analyzed for phosphorus, potassium, and calcium. Petiole samples were taken at three different dates, July 11, August 3 and September 11. A portion of each sample was extracted with a 10% sodium acetate in 3% acetic acid solution (1 to 20 ratio of green tissue to solution) and the percent of phosphorus determined. The remainder of the tissue was dried and analyzed for total calcium, phosphorus and potassium using a perchloric acid digestion procedure. The beets were harvested October 10, and the number, yield, percent sucrose, and percent apparent purity were determined.

Results and Discussion

The number of plants for the various treatments per 50 feet of row was as follows: 193 plants where 300 pounds of 6-24-12 fertilizer was applied 3 inches below the seed as compared to 184 plants where 300 pounds was applied $1^{1/2}$ inches to the side and 3 inches below the seed. For the 150 pound rate, the number of plants was 190 and 182, respectively. Disregarding placement the average number of plants where 0, 150, and 300 pounds of 6-24-12 were applied was 185, 186 and 189 plants per 50 feet of row.

The effect of fertilizer treatment and the weight and chemical composition of the plant samples taken June 6 are recorded in Table 1.

Fertilizer applied at planting time increased the early growth of the plant, the percent phosphorus in the tissue, and the uptake of phosphorus. Phosphate plowed down, in general, caused similar effects. As preplant phosphate was increased, the effect of fertilizer applied at planting time on the percentage of phosphorus in the tissue was decreased.

There was a marked effect of the placement of the 6-24-12 planting-time fertilizer on the early growth and nutrient uptake by the beets. Fertilizer applied 3 inches below the seed increased the weight of beets and the uptake of each of the nutrients. However, there was a trend for the percentage of P and Ca in the tissue to decrease, although not always significantly SO^4 where the fertilizer was applied directly below the seed as compared to fertilizer applied $1^{1/2}$ inches to the side of and 3 inches below the seed.

* Where the term "significant," applying to differences, is mentioned, the 5% level is indicated.

Table 1.—The effect of time and method of application of fertilizer on the weight and chemical composition of sugar beet plants at blocking time (6-2-61). (Monitor Sugar Co., 1961)

P_2O_5 plowed under	6-24-12 planting time	Placement[1] of fertilizer	Grams per-100 plants	Nutrient on dry weight basis			Total uptake in grams		
Lbs./acre				% P	% Ca	% K	P	Ca	K
0	0	. . .	10.2 i	.377 g	2.42 a	5.14 de	.038	.247	0.52
	150	Side	16.8 kl	.535 ef	2.13 b	5.55 cde	.090	.358	0.93
		Under	39.6 def	.527 f	1.99 bcd	5.07 de	.209	.788	2.18
	300	Side	20.4 ijk	.537 ef	2.14 ab	6.24 ab	.110	.436	1.3
		Under	52.5 bc	.575 cdef	1.83 d	6.75 ab	.302	.961	3.5
200	0	. . ..	17.2 kl	.552 def	2.12 be	4.97 de	.095	.365	0.86
	150	Side	28.8 ghij	.618 abc	2.06 bcd	5.69 cde	.178	.593	1.63
		Under	44.2 cde	.579 cdef	1.90 bcd	5.60 cde	.256	.880	2.48
	300	Side	29.5 ghí	.632 abc	2.13 b	5.95 bcd	.186	.628	1.76
		Under	61.7 ab	.588 bcde	1.88 bcd	7.13 a	.363	1.160	4.40
400	0	.. ..	20.2 jk	.624 abc	2.05 bcd	4.64 e	.126	.422	0.94
	150	Side	31.3 fgh	.615 abc	2.04 bcd	5.27 cde	.192	.648	1.66
		Under	45.8 cd	.603 abcd	2.00 bcd	5.05 de	.276	.916	2.31
	300	Side	34.4 fgh	.657 a	1.93 bcd	5.93 bcd	.226	.664	2.04
		Under	57.5 ab	.617 abc	1.99 bcd	6.92 ab	.355	1.144	3.98
800	0	.....	25.4 high	.633 abc	1.97 bcd	4.59 e	.161	.500	1.17
	150	Side	35.9 efg	.642 ab	2.04 bcd	4.98 de	.230	.732	1.79
		Under	56.0 b	.603 abcd	2.00 bcd	5.47 cde	.338	1.120	3.06
	300	Side	40.7 def	.622 abc	1.95 bcd	5.44 cde	.253	.794	2.21
		Under	66.7 a	.595 bcd	1.84 cd	6.97 ab	.396	1.224	4.64

[1] Side — 11/2" to side of and 3" below seed.
Under — 3" below seed.

[2] All data with the same literal postscripts are not significantly different from each other at the 5% level.

Table 2.—The effect of time and method of application of fertilizers on the phosphorus composition of sugar beet petioles at three sampling dates. (Monitor Sugar Co., 1961)

P_2O_5 plowed under	6-24-12 planting time	Placement[1] of fertilizer	7-16-61		8-3-61		9-11-61	
Lbs./acre			P in green tissue	Total P	P in green tissue	Total P	P in green tissue	Total P
0	0	.....	.111 d	.182 c	.051 i	.143 ef	.102 d	.198 cd
	150	Side	.097 d	.145 d	.053 hi	.162 def	.121 d	.187 d
		Under	.111 d	.170 cd	.059 hi	.152 ef	.114 d	.188 d
	300	Side	.112 d	.165 cd	.040 i	.123 f	.125 d	.212 cd
		Under	.114 d	.168 cd	.056 i	.162 def	.107 d	.213 cd
200	0	.....	.222 abc	.292 ab	.096 fg	.204 cde	.250 abc	.307 a
	150	Side	.225 abc	.285 ab	.116 bcdefg	.205 cde	.198 c	.247 bc
		Under	.212 abc	.276 b	.090 gh	.224 bcd	.229 abc	.298 ab
	300	Side	.238 abc	.272 b	.105 efg	.232 bc	.213 bc	.295 ab
		Under	.200 c	.269 b	.113 cdefg	.233 bc	.213 bc	.285 ab
400	0	.....	.245 ab	.315 a	.122 abcdefg	.260 abc	.284 a	.315 a
	150	Side	.236 abc	.303 ab	.105 efg	.238 bc	.236 abc	.270 ab
		Under	.228 abc	.291 ab	.127 abcdef	.256 abc	.264 ab	.323 a
	300	Side	.220 abc	.283 ab	.109 defg	.249 abc	.263 ab	.298 ab
		Under	.209 abc	.272 b	.125 abcdef	.270 abc	.234 abc	.288 ab
800	0	..	.250 a	.313 a	.147 abc	.292 ab	.254 abc	.307 a
	150	Side	.244 abc	.303 ab	.155 a	.312 a	.255 abc	.290 ab
		Under	.226 abc	.300 ab	.136 abcde	.265 abc	.253 abc	.306 a
	300	Side	.205 bc	.287 ab	.141 abcd	.287 ab	.251 abc	.307 a
		Under	.234 abc	.298 ab	.150 ab	.256 abc	.243 abc	.317 a

1 Side — $1^{1/2}$ to side of and 3" below seed.
Under — 3" below seed.

[2]All values reported on an oven-dry weight basis. Green tissue extracted with a 10% sodium acetate in 3% acetic acid solution (1:20 ratio of green tissue to solution). Moisture content of green tissue approximated 90%.

Potassium applied at planting time increased the percent potassium in the tissue and also the total uptake. More potassium was taken up where the 300 pounds of 6-24-12 was applied than where the 150 pounds was used. There was a trend for a higher amount of potassium both percentagewise and total uptake to occur where the fertilizer was applied directly below the seed than where it was applied to the side and below the seed, particularly where the higher rate of potash was applied.

There was a trend for the calcium in the tissue to decrease as the weight of the tissue increased. The percent of calcium in plant tissue was decreased when fertilizer was applied. The lowest percentage of calcium was found where the fertilizer was placed in a band directly below the seed. However, the total uptake of calcium was more dependent on the yield of the beet tissue at blocking time than on the amount of fertilizers applied either at planting time or when preplanting applications were made.

The phosphorus composition of the petioles (Table 2) increased as the phosphate applied prior to planting increased. The amount of phosphate applied at planting time did not appreciably affect the percent of phosphorus in the petioles. The amount of phosphorus in the tissue decreased from that contained in the tissue at blocking time for all subsequent sampling dates. The phosphorus content of the tissue taken September 11 was about equal to that obtained from the July 1 1 sampling whereas the P content of tissue sampled August 3 was lower than that sampled at any other date. Placement of planting-time fertilizer did not materially affect the percent of phosphorus in the tissue at any of the sampling dates.

A larger percentage of the total phosphorus in the plant was accounted for in the green tissue where a preplanting application of phosphate was made.

The percent of potassium (Table 3) in the sugar beet petioles decreased with each successive sampling date. In general, the percent potassium was highest in the petioles of the beet plants where no planting-time fertilizer was applied. The method of applying the planting-time fertilizer did not have any definite effect on the percent of potassium in the petioles. The percent of calcium (Table 3) in the tissue was lower at the July II sampling date than at blocking time but was higher at the August 3 sampling date. It then decreased on September 11 sampling date below that obtained at any other of the sampling periods. The amount of fertilizer whether plowed down or applied at planting time did not materially affect the percent of calcium in the tissue.

Table 3.—The effect of time and method of application of fertilizer on the calcium and potassium contents of sugar beet petioles at three sampling dates (Monitor Sugar Co., 1961)

P_2O_5 plowed under	6-24-12 planting time	Placement[1] of fertilizer	7-11-61[2]		8-3-61		9-11-61	
Lbs./acre			Total K	Total Ca	Total K	Total Ca	Total K	Total Ca
0	0	.....	5.15 a	1.29 bc	4.34 a	1.53 abc	3.99 abc	1.03 abc
	150	Side	4.75 abc	1.66 a	4.14 abc	1.36 abcd	3.87 abc	1.15 ab
		Under	4.77 abc	1.29 bc	3.82 abc	1.44 abcd	3.59 abc	.91 abc
	300	Side	4.86 ab	1.28 bc	3.95 abc	1.39 a	4.12 abc	1.18 a
		Under	4.49 abcd	1.32 b	4.18 abc	1.57 ab	4.39 a	.99 abc
200	0	.....	4.35 bcd	1.11 bc	3.58 c	1.21 cd	4.10 abc	.86 bc
	150	Side	4.22 bcd	1.15 bc	4.15 abc	1.42 abcd	3.51 bc	.99 abc
		Under	4.28 bcd	1.14 bc	3.87 abc	1.43 abcd	4.12 abc	.87 bc
	300	Side	4.32 bcd	1.20 bc	3.95 abc	1.37 abcd	4.09 abc	1.01 abc
		Under	4.17 bcd	1.18 bc	3.92 abc	1.30 abcd	4.33 ab	1.05 abc
400	0	.....	4.35 bcd	1.27 bc	3.66 bc	1.24 bcd	3.78 abc	.93 abc
	150	Side	4.14 cd	1.21 bc	3.69 abc	1.32 abcd	3.79 abc	1.00 abc
		Under	4.01 d	1.20 bc	3.93 abc	1.35 abcd	4.22 ab	.91 abc
	300	Side	4.19 bcd	1.18 bc	3.74 abc	1.41 abcd	4.24 ab	1.04 abc
		Under	3.99 d	1.18 bc	3.88 abc	1.28 abcd	4.00 abc	.91 abc
800	0	.....	3.91 d	1.14 bc	4.03 abc	1.16 d	3.79 abc	.86 bc
	150	Side	4.20 bcd	1.14 bc	4.29 ab	1.25 abcd	3.37 c	.90 bc
		Under	4.04 d	1.10 bc	3.89 abc	1.15 d	4.00 abc	.84 c
	300	Side	4.15 cd	1.05 bc	4.04 abc	1.24 bcd	3.97 abc	.87 bc
		Under	4.20 bcd	1.02 c	4.10 abc	1.24 bcd	4.15 abc	.93 abc

[1]Side — 1 1/2" to side of and 3" below seed.
Under — 3" below seed.

[2]All percentage values based on oven-dry weights.

The number of beets harvested per plot (128 feet of row) was significantly influenced by fertilizer treatments. The number of beets per plot where 0, 200, 400, and 800 pounds of P_2O_5 were plowed under was 119, 124, 127, and 128, respectively. The number was higher where phosphate was plowed under. Similarly, 6-24-12 fertilizer applied at planting time significantly increased the number of harvested beets per plot from 118 where no fertilizer was used to 127 where fertilizer was used. There were 127 beets per plot where the fertilizer was placed 3 inches below the seed as compared to 121 beets for the side band placement ($1^{1/2}$" X 3"). This difference was significant.

Data in Table 4 show that the highest yield of beets was obtained where the maximum amount of fertilizer was applied. The effect of planting-time fertilizer was greatest where no pieplant application of phosphate was made. However, there was a trend for planting-time fertilizer to increase beet yields regardless of whether phosphate had been plowed under or not. This effect decreased as the amount of phosphate plowed under increased.

Table 4.—The effect of time and method of application of fertilizer on the yield, percent sucrose and percent purity of sugar beets (Monitor Sugar Co., 1961).

P_2O_5 plowed under	6-24-12 planting time	Placement* of fertilizer	Tons per acre		Percent sucrose	Percent purity	Lbs. gross sugar per acre
Lbs./acre							
0	0		13.2	i	14.5	87.7	3357
	150	Side	14.3	hi			
		Under	17.9	abedef	15.0	89.4	4800
	300	Side	15.3	fghi	15.4	89.0	4194
		Under	17.3	bedefg	14.7	88.9	4522
200	0		15.0	ghi	15.2	90.7	4136
	150	Side	15.4	fghi			
		Under	17.0	cdefg	15.3	91.1	4739
	300	Side	16.0	efgh	14.3	87.7	4013
		Under	16.5	defgh	15.7	91.2	4725
400	0		14.7	ghi	15.4	90.6	4102
	150	Side	16.6	cdefgh			
		Under	18.0	abedef	14.8	85.3	4545
	300	Side	16.8	cdelg	15.1	87.4	4434
		Under	18.4	abede	15.2	89.2	-1989
800	0		17.9	abedef	15.6	86.4	4825
	150	Side	19.2	abed			
		Under	20.0	ab	15.5	89.2	5530
	300	Side	19.3	a be	15.2	88.1	5169
		Under	20.6	a	15.1	86.9	5406

Side — $1^{1/2}$" to side of and 3" below seed.
Under — 3" below seed.

There was a marked effect of fertilizer placement on the yield of beets. Fertilizer placed directly below the seed caused a greater increase in yield than where it was placed to the side and below the seed. This trend was noted on all areas where different amounts of phosphate were plowed under prior to planting.

Fertilizers or methods of application did not have a significant effect on the percent sucrose or percent apparent purity of the sugar beet roots.

There was a slight indication, however, that as the amount of phosphate plowed under increased, the percent sucrose in the beets increased. This increase was from 14.5% to 15.6%.

Summary

The effect of time and method of application of fertilizers on sugar beets was investigated on a Kawkawlin-Wisner loam soil complex. Four rates of phosphate fertilizer, 0, 200, 400, and 800 pounds of P_2O_5 per acre, were plowed under the fall before planting the beets. Three rates of 6-24-12 fertilizer, 0, 150, and 300 pounds per acre, were applied in two methods; in a band 3 inches below the seed, and in a band $1^{1/2}$ inches to the side and 3 inches below the seed. A basic application of 200 pounds of KC1 was applied on all plots.

The data can be briefly summarized as follows: 1) there was a marked response of early growth, phosphorus content of tissue and yield of beets to phosphate application; 2) planting-time applications of 6-24-12 fertilizer increased early growth of beets at each of the four levels of plowed down phosphate fertilization, increased the phosphorus and potassium contents of plants at blocking time, but did not increase the phosphorus content in the petioles of the leaves at any of the sampling dates; 3) Planting-time fertilizer placed in a band directly below the seed markedly increased the early growth of the plant over that where the fertilizer was placed $1^{1/2}$ inches to the side and 3 inches below the seed. In general, the percent of phosphorus in the tissue at blocking time was higher where the fertilizer was placed to the side of the seed than directly below the seed. The phosphorus uptake at time of blocking was greatest where the planting-time fertilizer was applied in a band directly below the seed. Similarly, the uptake of calcium and potassium was highest where the fertilizer was placed in a band directly below the seed than when placed in a band to the side and below the seed. Placing the planting-time fertilizer in a band directly below the seed in-

creased yields at both levels of application, 150 and 300 pounds per acre. The increase was least on the plots where 800 pounds of phosphate had been plowed down previously; 4) The percent of phosphorus in the tissue decreased with the first two sampling dates, July 11 and August 3, and increased when the petioles were sampled on September 11. The data suggest that the percent extractable P in the petioles should not be allowed to fall much below 0.15 at any time during the season to insure an adequate supply of P for highest yields; 5) The potassium content of the petioles tended to decrease with each successive sampling date. However, the calcium content of the petioles was lower than that of the beet tissue sampled at blocking time for all three sampling dates. It was higher on August 3 than on July 1 1 or September 11; 6) There was no significant effect on the percent of sucrose or percent apparent purity due to the fertilizer applications; 7) Yield of gross sugar increased as the amount of phosphate applied increased.

Selection for Seed Size in Monogerm Varieties

C. W. DOXTATOR AND R. H. HELMERICK[1]

Received fur publication March 23, 1962

Selection tor seed size in sugar beets has not been of importance in multigerm varieties because ot* the association of large seed ball size with a large number of germs. However, with true-breeding single germ seed now available (1)[2] seed size and other characters can be studied as in other crop plants.

Since the size of monogerm seed is positively correlated with germ size (2), seed size can be important in obtaining better field stands of beets. With this thought in mind a breeding project was set up in 1958 to determine what changes could be made in seed size by selection and what effect these changes might have on seed yield.

Materials and Methods

Two varieties, 58-401 and 58-413 were selected for study. The variety 58-401 was a mass selection of SLC 15, and 58-413 was a recovered monogerm variety from crosses of sclerotium (Sclerotium rolfsii Sacc.) resistant multigerm types with SLC 15 and was a steckling group.

Polycross seed was harvested from 200 plants of each variety individually. The 400 seed lots were lightly polished by hand and cleaned over a small Clipper cleaner equipped with a bottom or retaining screen having 6/64" round hole perforations. The seed thus prepared was graded by hand using 12" X 12" dockage screens having round hole perforations of 8/64", 10 64", 12/64" and 14/64". The resulting five size fractions were weighed and a weighted average seed size Avas obtained for each plant.

From the 200 plant progenies of each variety a 15-plant selection was made for large and for small seed size. All plants selected were good seed producers, having produced 90 or more grams per plant. Seed of these progenies was planted in August 1958 at Rocky Ford, Colorado, in four space isolation groups for overwinter seed production. In each group 20 hills spaced 30" X 30" were planted with each seed lot in a 20 replication design. The following spring the hills were thinned to single plants. Plants were harvested individually and average seed size obtained as described previously. From these data 15 plant progenies from each of the four groups were selected and planted at Phoenix, Arizona, in August 1959. Thirty-six stecklings of each line were transplanted at Rocky Ford in four groups in

[1] Plant Breeders, respectively, American Crystal Sugar Company, Rocky Ford, Colorado.
[2] Numbers in parentheses refer to literature cited.

1960. However, a June hailstorm severely damaged the flowering plants and made it impossible to obtain reliable size data. It was then decided to make selection of the large seed sizes in the two varieties for a breeders stock seed increase.

From the 1958-59 Rocky Ford groups which were thinned to single plants, there was a large surplus of stecklings, which was saved by variety and by seed size, for a replicated test to determine differences of size and yield. These stecklings were graded in 3 sizes—large, medium and small—and planted in a split-split plot test of 10 replications. Plots were single rows 20 feet in length and 44 inches apart. Steckling sizes were the main plots (four-rows wide) and were made up of two rows of each variety, with seed sizes in adjacent rows.

Experimental Results

Excellent overwinter stands were obtained in the four group isolations in 1958; a nearly perfect thinning stand was available for seed production the following year. However, it was necessary to rogue out some double flowering types. Curly top was present and further discards had to be made at harvest. In all, there were 722 plants harvested from the four groups out of a possible 1200. Table 1 gives the seed size data for the 1958 selection and the 1959 progenies as well as data on the 1959 selection.

Table I.—Average seed size of the 1958 parents, the 1959 progenies, and the 1959 selection.

		1958 Parents		1959 Progeny		1959 Selection
Variety	Seed size selection	No. plants selected	Avg. size in 64th"	Xo. plants harvested	Avg. size in 64th"	Avg. seed size of selected plants
58-401	Large	15	13.12	275	10.76	13.02
	Small	15	9.40	238	9.94	9.33
58-413	Large	15	12.61	91	11.28	12.72
	Small	15	10.75	118	10.45	9.77

As shown in Table 1, the difference between the large and small seed selection in 1958 was large for both varieties. The progenies also differed in size, with the selection for large seed producing large seed and the selection for small seed producing small seed. The trend for large seed plants to produce larger seed than those selected for small was great enough to indicate a substantial parent-progeny correlation. The relationship is shown in Table 2.

As seen in Table 2, the parent-progeny correlation for large seed size in 58-401 is highly significant and is suggestively large for 58-413. Although both correlations for small seed were posi-

tive, neither was significant. Further evidence of a parent-progeny relationship was found from a survey of the individvial plant progenies, as follows:

Table 2.—Correlations between seed si/.e of parents and the average seed size of their progenies.

Variety	Seed size selection	Number of parent plants	Avg. no. of progeny plants from each parent	Correlation
58-401	Large	15	18.3	+0.64*
	Small	15	15.9	+ 0.05
58-413	Large	15	6.1	+0.40
	Small	15	7.9	+0.12

* Significant beyond the 1% point

Table 3.—Difference in seed size obtained from selection for steckling size and seed size, in two monogerm varieties.

Steckling size	No. of comparisons	Avg. seed size F Value	64th inches
Large			11.92
Medium	40	18.15* *	11.79
Small			11.16
		(Sign. Diff.)	.23
Variety			
58-413		6.24*	11.85
58-401	00		11.61
		(Sign. Diff.)	.20
Seed Size			
Large		63.74* *	11.99
Small	60		11.46
		(Sign. Diff.)	.22

* Significant beyond the 5% point
** Significant beyond the 1% point

1. 58-401 Large. The largest seed progeny of the 275 plants harvested came from the second largest parent.

2. 58-401 Small. The smallest seed progeny of the 238 plants harvested came from the second smallest parent.

3. 58-413 Large. The largest seed progeny of the 91 plants harvested came from the third largest parent.

4. 58-413 Small. The smallest seed progeny of the 118 plants harvested came from the smallest seed parent.

5. Large seed progenies had fewer seeds per pound than small seed progenies.

As mentioned previously, all stecklings thinned from the 1959 seed groups were graded into large, medium and small sizes for a seed size and yield test, in a split-split plot design. The average

steckling weight for the three classes was: large—.16 pounds, medium—.09 pounds, and small—.03 pounds. The analysis of variance in this test for seed size is given in Table 3.

It will be observed that size of stecklings affected seed size. The differences obtained were highly significant. The two varieties also differed significantly in seed size, 58-413 being the larger. It is of interest to note that during the flowering period the observation was made in the isolated seed fields that this variety had larger flower buds, but was attributed at that time to possible differences of soil *in* the different isolated fields.

By far the most significant was, however, the difference in seed size due to selection. As indicated in Tables 1 and *2,* one open-pollinated plant selection for large and for small seed, significantly divided the varieties for the seed size character.

The analysis of variance is given in Table 4 for seed yield obtained in the same test.

There were no significant differences in this test for seed yield as shown in Table 4. There was, however, an indication of a slight trend for large stecklings to produce more seed. Although the seed sizes yielded alike, the number of seeds per pound ranged from 19,500 for the largest to 72,000 for the smallest size.

Table 4.—Differences in seed yield obtained from selections for steckling size and seed size, in two monogerm varieties.

Steckling size	No. of comparisons	F Valle	Av. grams seed per plant
Large			67.7
Medium	40	1.15 (NS)	64.8
Small			59.8
Variety			
58-401	60	2.00 (NS)	66.4
58-413			61.0
Large	60	2.22 (NS)	63.8
Small			63.5

Discussion

The results obtained in this experiment indicate that seed size in monogerm varieties can be easily improved by ordinary mother line selection. Although it was not possible to test progenies of the second selection, the results of the first progeny test were so satisfactory that it can be expected that further differences were obtained in the later selections.

One of the most important discoveries made was the effect of steckling size on seed size. Because of this discovery, stecklings of the second selection were grown at Phoenix, Arizona, and after thermal induction were graded as nearly as possible to the same

size and extra care was used in transplanting at Rocky Ford, so that uniform conditions for regrowth would he obtained. In the 1958-59 overwinter planting, where the stands were thinned to one beet per hill there was no possibility of obtaining a uniform size of stecklings and consequently some of the recorded seed sizes from these plants may have been in error due to this environmental factor.

The lack of a parent-progeny relationship in small seed size (Table 2) is distinctly different than that obtained with the large seed size. This lack of relationship can be due to at least two environmental factors: first, plants were all harvested at the same time, and some of the plants may not have been as mature as others. This would tend to reduce seed size on some plants and cause errors in classification; secondly, a mild epidemic of curly top occurred in all groups except 58-401 Large, and it was necessary to discard many plants in these three groups. Others may have been affected. If curly top affects seed size this would also cause errors in classification.

The effect of seed size on seed yield is an important consideration, and was studied in this experiment. Since both sizes yielded alike, it is apparent that the difference between the two sizes must have been in number of seeds per pound.

It is evident in the two varieties studied in this experiment that there is a wide range in seed size due to heritable factors. Since uniformity of seed size is important for maximum recovery and drillability of commercial monogerm beet seed, it would seem important that selection work be conducted for the size of seed desired.

Summary

1. A selection for large and small seed was made in two varieties of monogerm sugar beets using the "mother" line method of breeding.

2. Progeny tests showed that in general, large seed parents produced large seed progenies and small seed parents produced small seed progenies.

3. It was found that large stecklings produced larger seed than small stecklings.

4. Yield of seed per plant was not affected by selection for seed size. However, selection for large size reduced the number of seeds per pound.

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Correlations of Pre-Harvest Samples and Cultural Practices With Final Yield and Quality of Sugar Beets[1]

R. E. FRIEHAUF, H. L. BUSH AND E. E. REMMENGA[2]

Received for publication February S, 1962

A method for taking pre-harvest samples as an aid for estimating the tonnage and sugar content of the sugar beet crop in The Great Western Sugar Company territory has been devised and described by Brewbaker and Bush (1) (2) (3)[3] Samples are taken the first week in September and again the third week of the same month by this method. Each sample consists of 10 feet of row taken at random for each 90 acres of beets being grown in a factory district. This method has been used each year since its innovation and the results obtained have led to highly accurate estimates of the amount of sugar to be produced.

Seven years ago The Great Western Sugar Company decided to obtain information relative to the effect different cultural practices might have on the beet crop. The study pertained to those farms chosen for the regular pre-harvest sampling. The information was obtained through a questionnaire which was especially designed so that the information could be readily recorded on IBM cards. This provided for a rapid analysis of relationships existing between the various practices. Many questions were answered in a categorical form while actual results were recorded in some cases.

In 1957, the pre-harvest sampling idea was extended to include an early pre-harvest sample taken about July 25. This sample was taken according to the same procedures employed in taking the regular September pre-harvest samples, except that no sugar analyses were made and both root and top weights were taken in July.

Data have been recorded from approximately 2500 farms represented by all fieldman in the Great Western organization and calculations were performed at the Colorado State University Statistical Laboratory.

For the purposes of this study, the territories served by The Great Western Sugar Company have been divided as follows:

[1] The writers are indebted to Western Data Processing Center at UCLA for the use of their computing facilities in analyzing the data.

[2] Graduate Student, Colorado State University, Statistical Laboratory, Fort Collins, Colorado; Statistician-Agronomist, The Great Western Experiment Station, Iongmont, Colorado; and Associate Professor of Statistics, Colorado State University, Fort Collins, Colorado, respectively.

[3] Numbers in parentheses refer to literature cited.

Area 1, Northern Colorado; Area 2, Northeastern Colorado; Area 3, Nebraska; Area 4, Montana and Wyoming; Area 5, Ohio.

No attempt will be made here to discuss all of the studies which are possible from such a questionnaire; only some of the results which may be of general interest will be presented. Further, only 1960 results will be discussed except for few occasions where the years 1957-1960 were combined for a total effect.

It must be emphasized that these results are based on agricultural practices under wide-scale field conditions and a wide range of management levels. They do not represent basic agronomic relationships, but indicate results and conclusions that can be obtained from farm practices, as indicated by the characters which can be, at least, partially measured.

In making a study of this type the experimenter is faced with several unavoidable complications. There is no way of measuring the effects of weather, sugar beet diseases and insects. Without these complications the results presumably would be more accurate or would show greater significance.

Yield-Stand Relationship

One of the more interesting graphs plotted from the results of the survey deals with the effect thinning methcxis may have on the final stand, the 1960 results being presented in Figure 1. It

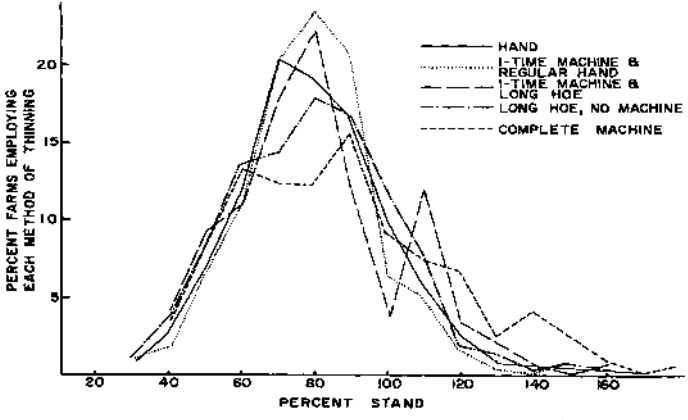

Figure 1.—Total Great Western district, 1960—method of thinning and resulting stand.

is apparent that all thinning methods gave essentially the same final stand except for small deviations in one time machine followed by long-handle hoes and complete machine. The fact exists that less than 5.5% of the farms used complete machine, while over 60% used hand thinning.

The relation of stand to final yield is presented in Figures *2*, 3, 4, 5, for Areas 1, *2*, 3, and 4. Here the predictions are calculated from regression equations and steady increases in percent stand are accompanied by increases in yield. Maximum yields are obtained at approximate stands of 150 beets per 100 decrease. Insufficient data are available beyond this point for adequate analysis.

Yield Predictions

Correlations between final yield of beets and weight of roots at the various pre-harvest dates are presented in Table 1. Computing on a linear basis, the final yield will be 14.84 tons per acre plus an additional 0.2633 ton for each ton of the early September sample.

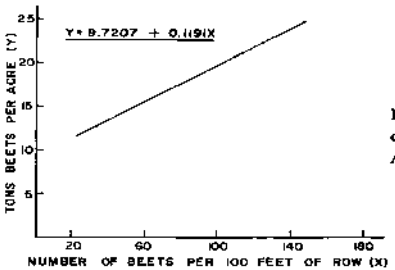

Figure 2.—The Great Western Sugar Company, 1960—Area 1.

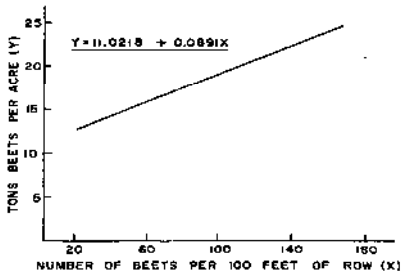

Figure 3.—The Great Western Sugar Company, 1960—Area 2.

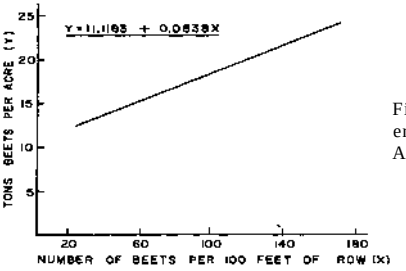

Figure 4.—The Great Western Sugar Company, 1960—Area 3.

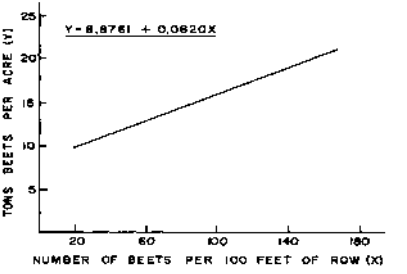

Figure 5.—The Great Western Sugar Company, 1960—Area 4.

As would be expected the correlation between September root yield and the final yield is significant. The relationship (significant at the 5% level) between weight of tops in July and final yield of beets is also given in Table 1. It is quite possible, since this high correlation exists, that top weights taken in July might be substituted for the second September sampling as a predicter of final yield. A prediction from this sample, if satisfactory, would be much more useful because it is taken earlier.

Sugar Content Predictions

The correlations between the sugar content indicated by the preharvest samples and final results appear in Table 1. The correlation coefficients are high enough to indicate that a fairly accurate prediction as to the final sugar content can be obtained through the use of this type of data.

Effects of Fertilizer Practices by the Farmer

The effects of fertilizers, as applied by the farmer, on sugar content and purity are shown by the simple correlation coefficients and coefficients of multiple determination in Table 2.

TABLE 1

RELATION — Average of Areas 1,2,3,4'	r	byx	Predictor equation
1. Wt. of beets in early Sept. (X) to final yield of beets (V)	0.3079	0.2633	Y = 14.838 + 0.2633X
2. Wt. of beets in late Sept. (X) to final yield of beets (Y)	0.3764	0.3210	Y = 12.743 + 0.321 OX
3. Wt. of tops in inid-July (X) to final vield of beets (V)	0.3878	0.2627	Y = -18.241 + 0.2627X
4. Sugar content in early Sept. (X) to final sugar content (Y)	0.4256	0.2831	Y z^ 13.761 + 0.283 IX
5. Sugar content in late Sept. (X) to final sugar content (Y)	0.5032	0.3670	Y 1 2.1 74 + 0.3670X

Except for relations 4 and 5 which is the average of Areas **1,** 2, and 3 only

TABLE 2.—**Current crop,** areas **1,** 2, 3.

Year	Fertilizer	Simple Correlation Coefficients % Sugar	% Purity
1960	N	-0.1147	-0.1023
57, 58, 59, 60	N	-0.0621	0.0917
1960	P_2O_5	0.0094	0.0224
57, 58, 59. 60	P_2O_5	0.0123	-0.0094
1960	K_2O	0.0857	0.1143
57, 58, 59, 60	K_2O	0.0738	-0.0754

Coef. of Multiple Determination (R^2)

	Tons/Acre		
1960	N, P, K	0.0439	0.1336

Simple correlations (r) between the various fertilizer elements with both sugar content and purity have been calculated, first as fertilizers applied in 1960 and secondly by combining all years over which the study was made and by combining Areas 1, 2, and 3. The amounts of fertilizers applied to the crop are calculated from both commercial fertilizers and organic manures.

The correlation coefficients all appear non-significant—basing significance on coefficients of 0.15 or greater—and in the cases of N and K_2O are negative, indicating a slight decrease in sugar content and in purity. Considering a coefficient of multiple determination value (R^2) of .04 or greater as significant for a sample of this size it was found that these values are significant. These coefficients give us the estimated percent decrease in variance in predicting percent sugar and purity given the factors N, P, K and final yield on which to base our predictions. In the case of predicting sugar, the variance is decreased by 4% and by 13%

in predicting percent purity. In evaluating these data it should be kept in mind that only a narrow range of fertilizer applications is represented. For example, nearly 50% of the farmers applied between 50 and 100 lbs of N per acre and over 45% of the farmers applied between 50 and 100 lbs of P_2O_5 whereas less than 3% failed to apply either of these fertilizers. Nearly 50% of the farmers did not apply K_2O.

The coefficient of multiple determination indicates a combined improvement in both sugar content and purity, thus meaning that the present farm fertilizer practices are nearly correct. However, the application of an additional amount of fertilizer might be safe without affecting the quality of the beets.

Some Indications from the Statistical Analysis of Data Taken from Pre-Harvest Sample Studies 1960

Results

1. Contrary to results observed in controlled experiments, fertilizers appeared to have little or no effect on yield, percent sugar, or percent purity. The fertilizer effects are at least partially masked by the random uncontrolled variables such as management, weather and native fertility. The fact that a few of the farmers did not apply some fertilizers may have had a profound effect on the statistical analysis.

2. Percent stand shows a positive, constant, independent, significant effect on yield. The effect of percent stand on percent sugar shows a positive, significant effect, this effect being much smaller than that on yield.

3. The weight per sample, taken September 4 as the first, pre-harvest sample is highly associated with final yield and is valuable in predicting final yield. The sampling error for these ten-foot samples is large and the result is lower correlation values. The logical assumption is that the yield prediction should be better for samples taken closer to the time of harvest. Thus the second pre-harvest sample taken the third week of September shows a greater correlation between sample weight and final yield.

Discussion

The data recorded on the "Pre-harvest Sample Field Data Sheet" for 1960 have been stored on IBM punch cards and a preliminary report of some of the results has been made.

Both simple and multiple correlations have been calculated for certain factors, the results of which are herein discussed.

As these results are considered, it must be realized that they are for only one year's data and in some cases the number of observations is too low for accurate conclusions to be drawn for

a study of this type. Also, a high standard error must be assumed for each figure, as reported. Some conclusions are indicated which apply to the different areas in which the Great Western territory has been divided for this study. These areas being: northern Colorado (Brighton, Eaton, Fort Collins, Greeley, Longmont, Loveland, and Windsor); northeastern Colorado (Brush, Fort Morgan, Ovid, and Sterling); Nebraska (all Nebraska factories and Wheatland, Wyoming); Billings-Lovell; and Ohio (Fremont and Findlay).

In all of the studies herein discussed, fertilizers N, $P_2 0_5$, $K_2 0$ and certain other variables are combined to show their multiple effect on final tonnage, sugar content, and purity. These are compared with simple correlation values for each of the variates.

The following relationships are discussed concerning Areas 1, 2, 3, and 4 (with the omission of relationship three from Area 4). Discussion of these relationships for Ohio Area will be omitted because of an insufficient amount of data for this study.

1. N, P, K, %Stand with Final Tons Beets per Acre
2. N, P, K, %Stand with Final %Sugar
3. N, P, K, %Stand with Final %Purity
4. N, P, K, Tons Beets/Acre 1st Sample with Final Tons Beets/Acre
5. N, P, K, %Sugar Sept. 4 with Final %Sugar

N. P. K. %Stand—Tons per Acre

NORTHERN COLORADO

There is a slight significant positive relationship between application of N and tons beets per acre while no significant relationship was caused by the application of $P_2 0_5$, and $K_2 0$ in regards to yield of beets. Percent stand appears to have a large effect on final yield of beets. The combined effects of N, $P_2 0_5$, and $K_2 O$ on tons beets per acre gives a small, but positive effect which is significant. Combining the fertilizers with percent stand gives a large correlation (R = 0.5299) that definitely shows percent stand is a deciding factor in determining tons per acre and that it is also independent of fertilizers.

NORTHEASTERN COLORADO

In the single comparisons N and $K_2 0$ have a significant positive effect on tons per acre, whereas $P_2 0_5$ shows a positive but non-significant effect.

There is a non-significant negative relation of fertilizers with percent stand in the single comparisons but a highly significant effect of percent stand on tons per acre.

The combined effects of N, $P_2 0_5$, $K_2 0$ and percent stand on tons per acre is indicated by a significant correlation which leads to the same conclusions as that for Northern Colorado.

NEBRASKA

The single comparisons show a positive significant effect on yield. These comparisons also show that the fertilizers do not have a significant relation with percent stand.

The combined effects of the fertilizers and percent stand show[7] a significant effect on tons per acre with a multiple correlation of 6.5846.

N, P, K, %Stand—%Sugar

NORTHERN COLORADO

From single comparisons of the fertilizers to percent sugar, there appears to be a non-significant negative effect. Percent stand shows a significant but small effect on percent sugar.

The fertilizers combined with percent stand have a positive significant effect on percent sugar.

NORTHEASTERN COLORADO

The single comparisons show that the fertilizers have a negative non-significant effect on both percent stand and percent sugar, and percent stand has a significant effect upon percent sugar.

When percent stand is included in the analysis with the fertilizers, the effects of the fertilizers are changed very little and the multiple correlation coefficient shows little significant effect on percent sugar.

NEBRASKA

The single comparisons of the fertilizers with percent sugar show that N has a positive but non-significant effect on percent sugar, whereas, $P_2 0_5$ and $K_2 0$ show negative non-significant effects. The effect of percent stand on percent sugar is positive but shows little significance. The combined analysis of the fertilizers and percent stand with percent sugar shows a positive significant relationship and that the effect of percent stand is independent of the effect of the fertilizers.

BILLINGS-LOVELL

The fertilizers in single comparisons have positive but almost no effect on percent sugar. Percent stand also shows a positive but non-significant effect on percent sugar.

The effect of fertilizers combined with percent stand on percent sugar is positive, but insignificant.

N, P_2O_5, K_2O %Stand—%Purity

NORTHERN COLORADO

The fertilizers in single comparisons have a negative effect on percent purity, whereas, percent stand shows a positive but non-significant effect.

In combining the fertilizers and percent stand it is found that they have a positive significant effect on percent purity.

NORTHEASTERN COLORADO

Here again the fertilizers have a negative non-significant effect on percent purity. The fertilizers also have a negative effect on percent stand but there is no significant effect for any of the fertilizers in this comparison.

The effect of the fertilizers on percent purity increases when combined with percent stand. The multiple correlation coefficient appears to be significant when combining all of the above factors.

NEBRASKA

The fertilizers show a negative effect on percent purity but only in the case of K_20 is there any significance. The single comparisons also show that percent stand has a small positive effect on percent purity.

The combined effects of the fertilizers with percent stand on percent purity show a positive effect with a multiple correlation of 0.4407.

N, P_20_5, K_20, Weight per Beet (1st PHS)[4] Final Yield

NORTHERN COLORADO

In the single comparisons, fertilizers show a positive but non-significant relationship to weight per beet in early sample and to final sample weight.

The effect of weight per beet (1st PHS) on tons per acre is significant and increases in relationship from pre-harvest sample to final yield.

The multiple comparison shows that a significant relationship exists.

NORTHEASTERN COLORADO

In single comparisons N and K_2,0 appear to have a significant effect on weight per beet (1st PHS), whereas, P_2O_5, shows no significance on weight per beet. In this comparison, weight per beet has a significant effect on tons per acre. When the weight per beet is combined with N, P_2O_5, K_2O in the analysis the effect on tons per acre is highly significant, R = 0.5046.

[4] 1st PHS refers to pre-harvest sample taken in early September.

NEBRASKA

The single comparisons show that there is no significant relationship between fertilizers and weight per beet but that weight per beet has a significant positive effect on final tons per acre. This effect holds in the combined analysis of N, P_2O_5, K_20 and weight per beet (1st PHS) to tons per acre.

BILLINGS LOVELL

The fertilizers appear to have no significant effect on weight per beet, however, weight per beet (1st PHS) appears to have a significant effect on final yield.

The combination of these factors also shows a positive sig- per beet, however, weight per beet (1st PHS) has a significant effect on final yield.

N, P_2O_5 K_2O, %Sugar (1st PHS)—Final %Sugar

NORTHERN COLORADO

The effect of fertilizers on percent sugar (1st PHS) appears to be negative and non-significant and their effect on final percent sugar is negative but appears to have a significant relationship. The percent sugar (1st PHS) appears to give a good prediction of final percent, sugar and is independent of fertilizers.

NORTHEASTERN COLORADO

The single comparisons show that the fertilizers have a negative significant effect on percent sugar (1st PHS) and final percent sugar. However, percent sugar (1st PHS) is a good predictor of final percent sugar.

The fertilizers combined with percent sugar 1st PHS) have a positive significant relationship to final percent sugar. Percent sugar (1st PHS) also appears to be independent of the fertilizers.

NEBRASKA

The single comparisons show that fertilizers have no significant effect on either percent sugar (1st PHS) or final percent sugar and that the relationship between percent sugar (1st PHS) and final percent sugar is highly significant. The combined effects of fertilizer and percent sugar (1st PHS) on final percent sugar appear to be highly significant with percent sugar from the preharvest sample being the most dominant factor.

BILLINGS-LOVELL

The effect of fertilizer on percent sugar (1st PHS) appears to be greater than the effect of fertilizers on final percent sugar. However, percent sugar (1st PHS) appears to have a significant relationship to final percent sugar.

The correlation between percent sugar (1st PHS) and final percent sugar is almost as large as that of the combined effects of fertilizers and percent sugar (1st PHS) on final percent sugar. This indicates that fertilizers have relatively no effect on final percent sugar.

Summary

Prepared for The Great Western Sugar Company Managers

1. Perhaps because of the narrow range of fertilizer applications as observed in these studies, fertilizers appear to show little or no effect on yield, percent sugar, or percent purity. For example, over 50% of the farmers applied between 50 and 100 lbs of N per acre with very few applying amounts which might be considered excessive and only a small number of farmers not applying any fertilizer.

Their results lead to the conclusion that the present farm fertilizer practices are nearly correct, although it might be safe to use a small additional amount of N fertilizer, if applied early in the season, without materially affecting beet quality.

2. Percent stand shows a positive effect on yield with maximum yields obtained from approximately 150 beets per 100 feet of row.

3. Results from early September sampling give a good prediction of final yield and sugar content, but the later September sampling taken closer to final harvest, gives a slightly better prediction than the early sample.

However, a slightly higher correlation was found between top weight in July and final yield than was obtained between September beet weights and final yield. This indicates that a satisfactory yield estimate can be made from July top weights.

Regression formulae for calculating predicted yields from samples taken at the various dates are presented.

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The Effect of Phosphate Applications on Soil Tests and on Subsequent Yield of Field Beans and Wheat[1]

DONALD THURLOW, GRANT NICHOL, AND J. F. DAVIS[2]

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The differential response of various crops to residual fertilizers and the effect of applied fertilizers on soil tests is of considerable interest. These effects have been studied for the past three years in conjunction with an experiment established near Bay City, Michigan, on a calcareous Kawkawlin-Wisner loam soil complex, **pH** 7.5.

Materials and Methods

A rotation of beets, beans, and wheat with a companion crop of sweet clover in the wheat was established at the above location in 1959. Each crop in the rotation appeared each year. For the sugar beet crop, four rates of $P_2 0_5$ were broadcast and plowed under ahead of planting, 0, 200, 400, and 800 pounds per acre. A basic application of 200 pounds of 60 percent muriate of potash was plowed under. At planting time a starter fertilizer of either 5-20-10 or 6-24-12 was used on the beets.

The wheat and bean crops were fertilized with 150 pounds of either 5-20-10 or 6-24-12 per acre.

Soil samples were taken (20 cores per plot) and were analyzed for phosphorus using .025 normal HC1 plus .003 normal NH_4F extractant with a one to eight soil to extractant ratio. The area was divided into three sections—Section A, where the first application of phosphate was made in the spring of 1959. Soil samples were taken on July 30, 1959, and again on August 16, 1961. The phosphate fertilizer was plowed down on Section B in the fall of 1959, and the soils were sampled on July 8, 1960, and August 16, 1961. The phosphate was applied on Section C in the fall of 1960 and the soils were sampled on August 16, 1961.

Sanilac variety of field beans was planted following the sugar beet crop. The sequence of crops was beets, beans, and wheat. Phosphate treatments were replicated three times and four subplots (28' X 66') out of each main plot were sampled.

Results and Discussion

The data in Table 1 show that the amount of phosphate applied was reflected by the soil tests. The greater the amount of phosphate applied the higher the soil test. The relationships

[1] Contribution from the Soil Science Department, Michigan Agricultural Experiment Station, East Lansing, Michigan, and Agricultural Department, Monitor Sugar Company, Bay City, Michigan. Approved by the director as Journal Article No. 2937.

[2] Assistant Instructor of Soil Science, Michigan State University, Agronomist, Monitor Sugar Company, and Professor of Soil Science, Michigan State University, respectively.

Table 1.—The effect of phosphate applications on phosphorus soil tests (Monitor Sugar Co., 1961).

Lbs P_2O_5./acre	Pounds P per acre[1]				
	Section A		Section B		Section C
	P_2O_5, applied spring, 1959		P_2O_5, applied fall, 1959		P_2O_5 applied fa'l, 1960
	7-30-59[2]	8-16-61[2]	7-8-60[2]	8-16-61[2]	8-16-61[2]
0	24	27	23	29	26
200	40	40	50	41	48
400	70	63	89	63	79
800	119	122	130	98	117
L.S.I). (5% level) (1% level)	16	9	18	8	l3

[1] P determintd by extracting the soil sample with Brays P. extracting solution (.025 NHCl + .003 X NH_4F) 1:8 soil to solution ratio.
[2] Dates of sampling.

in general appear to be linear. The variability in the data is indicated by the pounds of P required for significance between treatments. This difference amounted to 16 pounds per acre for the sampling date of July 30, 1959, where one plowing had intervened between the time of application of phosphate and the time the samples were taken. The difference was significant (1% level) for all treatments. However, when the soils were sampled on August 16, 1961, the least significant difference required between means had decreased to 9. Similar results were obtained for the samples taken from Section R. There is no consistent decrease between the two sampling dates in soil tests over the three-year period in Section A or the two-year period in Section B. There was a great similarity between the soil test values on the various sections indicating that the soil was fairly uniform as far as phosphorus content was concerned.

One of the objectives of soil testing is to set up a threshold value above which small increases in yield would be expected other than those obtained from the use of planting time or starter fertilizer. Now as these data might suggest, as far as the sugar beet crop is concerned, this value has not been attained, because yields of sugar beets in 1961 were substantially higher where 800 pounds of phosphate was plowed under than where 400 was applied (Table 2). Bean yields were significantly reduced (1% level) in 1961 where additional phosphate was applied, that is, 400 and 800 pounds per acre. However, there was no significant increase in yields of wheat due to the phosphate that was plowed under for the sugar beet crop. This indicates that crops differ with respect to their nutritional needs for phosphorus.

Table 2.—The effect of phosphate applications for the sugar beet crop on the yields of subsequent crops of beans and wheat (Monitor Sugar Co., 1961).

	Section A			Section B		Section C
Lbs P_2O_5/acre Plowed under for sugar beets	Sugar beets 1959 Tons/acre	Beans 1960 Bu/acre	Wheat 1961 Bu/acre	Sugar beets 1960 Tons/acre	Beans 1961 Bu/acre	1961 Sugar beets Tons/acre
0	14.7	25.0	55.5	9.5	26.3	15.2
200	16.8	27.2	58.9	10.7	26.8	15.8
400	18.9	29.8	59.6	11.3	21.4	16.5
800	18.8	24.0	58.7	12.5	15.0	19.2
L.S.D. (5% level)	1.9	N.S.	N.S.	1.5	3.9[1]	2.0

[1] Significant at the 1% level.

While the difference in yield of beans due to phosphate applications was not significant in 1960, nevertheless, the lowest yield was obtained where 800 pounds of phosphate had been plowed under. In 1961, a very striking situation developed. Just prior to blossoming time, about six weeks after planting the beans, browning symptoms on the leaves developed. This was progressively worse as the amount of phosphate plowed under increased and significant reductions in yields resulted. The beans from the plots that had received the two higher rates of phosphate application were small and apparently did not develop normally. When this condition was noted, several minor elements, including zinc, were applied on the plot, but no noticeable result was indicated in the appearance of the plant or in the final yield. Judging from past experiences with corn, possibly zinc should be applied in the starter fertilizer to correct zinc deficiency. There are several instances reported by farmers and others who state that beans in some cases do not do well after sugar beets. It is suggested that this condition may have been due to a zinc deficiency caused by the tie-up of zinc by the phosphate, in that zinc phosphate is one of the most insoluble phosphate compounds. The observation concerning beans following sugar beets has also been made in USDA Leaflet No. 495 entitled "Zinc Deficiency of Field and Vegetable Crops in the West."

Summary

The amount of phosphate applied was reflected in soil tests. The greater the amount applied the higher the soil test.

Less variability between the data was found after the soil had been plowed three times after an application of phosphate fertilizer than one time after plowing. Apparently the subsequent mixing in the soil of the phosphate permitted more precise sampling.

Crops vary in their response to phosphate. Wheat apparently will produce well at relatively low phosphate levels between 40 and 50 pounds per acre, whereas sugar beets produce the highest yield where the soil tests were above 100 pounds per acre, approximately 125 pounds per acre.

The appearance and behavior of a bean crop, particularly in 1961, suggests that a possible zinc deficiency is being induced where high rates of phosphate fertilizer are used. Bean yields were 11.3 bushels of beans lower where 800 pounds of phosphate had been applied as compared to where no phosphate had been plowed under.

Low Raw Sugar Crystallization in Connection With Affination

HAL L. MEMMOTT AND E. CLARK JONES[1]

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Introduction

Since the advent of the modern sugar industry, crystallization has always been given major consideration. In recent years many investigators have studied the subject with reference to sugar boiling. The objectives have been many and varied such as: pan design, steam economy, sugar quality, pan yield, workability of low grade products and overall sugar recovery. In 1960, a sugar boiling program was started at the Utah-Idaho Sugar Company. The objective was to improve the quality and yield of low raw sugar with the final objective of affinating this sugar. The end result desired was to increase sugar end capacity without major expenditures for additional equipment. The plan was to standardize on improved graining and boiling procedures that would give higher purity sugar with larger and more even crystals. This better sugar should purge more readily and thus give the needed increase in plant capacity. The Moses Fake, Washington, factory was chosen for the initial experimental work on this problem.

Description of Equipment

The Moses Lake factory is equipped with two 11-foot diameter calandria pans of 1,200 cubic feet capacity (Figure 1*)* used to boil the low raw sugar. These pans are cross-connected with an 8-inch line and valve for splitting pans. Both are equipped with mechanical circulators driven from the bottom. The controls (See Figure 2) include an absolute pressure controller, a density controller, a BPR (boiling point rise) recorder, and a level recorder. Attached to the pan is a microscope so that the inside contents of the pan can be magnified and viewed during; the whole boiling period. The microscope has a light source inside the pan that shines through the juice and crystals, giving an excellent illuminated field.

The massecuite is dropped into a surge tank where the RDS is adjusted to the crystallizer RDS before it is pumped into the continuous crystallizers. This requires that the pan RDS be determined in the laboratory and the amount of water to be added to the surge tank calculated from the RDS of the pan as dropped. After the proper amount of water has been added and mixed with the massecuite, the massecuite is pumped to the continuous

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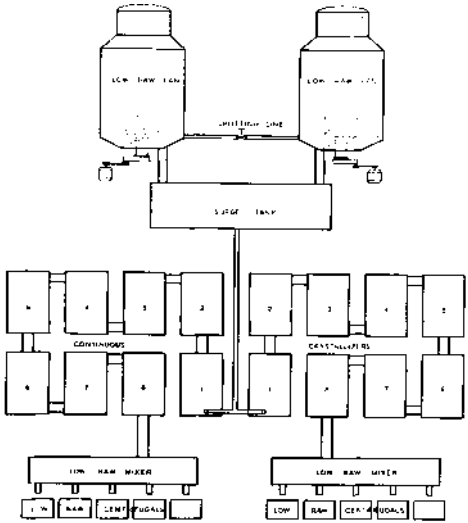

Figure 1.—Diagram showing equipment used in low raw sugar crystallization.

crystallizers. These are the conventional jacketed-type crystallizers equipped with internal cooling connected together so that the massecuite flows in parallel through two sets of eight single crystallizers connected in series. Both the jacket and cooling arms are used to cool the massecuite. The arms turn at 1 RPM and the cooling water is about 20° C. From the bottom of the last or eighth crystallizer on each side, the massecuite drops into two separate mixers—one for each set of crystallizers. Ten 42 inch X 24 inch Roberts centrifugals, five under each mixer, operating at 1,600 RPM, handle all the low raw massecuite.

Procedure

From the start of the sugar boiling program it was evident that in order to make each and every pan produce good low raw sugar, the proper amount of grain had to be established in each pan. The method previously used for graining - powdered sugar and air - left much to be desired. Several graining methods were attempted including: fondant and isopropyl alcohol, fondant and

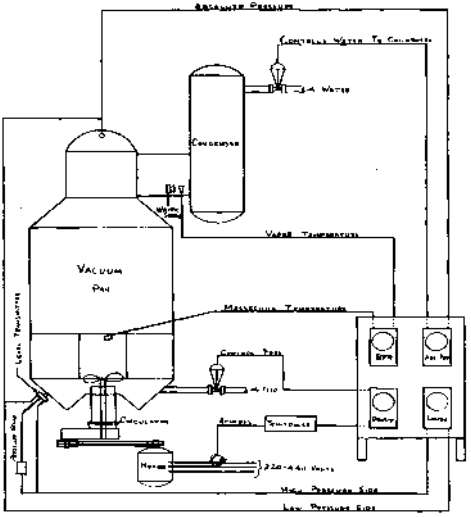

Figure 2.—Instrument diagram for calandria pans.

saturated liquid sugar, and finally milled seed. Gillett (2)[2] has explained in detail the first two methods, but the latter method mentioned perhaps needs some explanation. The idea originated in the Hawaiian Islands. It entails merely grinding two pounds of sugar with two liters of anhydrous isopropyl alcohol in a one gallon ball mill tor twenty-four hours—hence the name milled seed. This mixture contains approximately 1.6 X 10^9 neuclei per milliliter which means that about 300 milliliters are required to seed a low raw pan or about 125 milliliters to seed an average white pan. Milled seed has the following advantages: 1. Is is a stable mixture. 2. It requires only a small quantity, 300 milliliters, to seed a pan. 3. It gives approximately the same number of grain each time. 4. It permits a very simple seeding operation.

The boiling procedure starts by taking a 400 cubic foot graining charge of high green into a clean tight pan. The amount of graining charge is constant from pan to pan by the use of the level recorder. By using high green, the seed crystals grow very rapidly in the graining charge resulting in a finished pan in which

[2] Numbers in parentheses refer to references.

the crystals are of maximum size. As soon as the charge is pulled into the pan, the agitator is started and the steam is turned on. The charge is boiled down until the boiling point rise reads 9° C. This supersaturation point can be observed through the pan microscope by putting a few coarse crystals in the graining charge just prior to the time when saturation is reached and noting when the crystals just develop sharp edges.

At this point the pan is seeded with 275 to 300 milliliters of milled seed (See Figure 3) and then as soon as the crystals take distinct shape as observed through the pan microscope (BPR 11° C), the steam valve to the calandria is turned back to $^{1/2}$

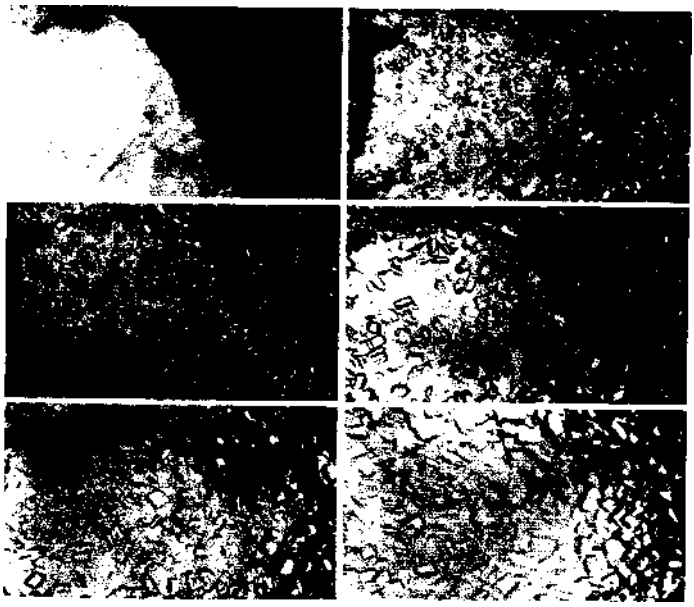

Figure 3.—Photographs of crystals as they appear through the pan microscope. Upper left—crystals at seeding time, upper right—crystals when steam rate is reduced, center left—crystals when feed is started, center right—crystals when pans are split, lower left—crystals when feed is shut off, lower right—crystals when pan is brixed.

round open. The crystals now are allowed to grow unmolested for approximately fifteen minutes until the BPR starts down. This point is verified through the microscope as being the time when the crystal width equals the distance between crystals. At this time the feed of intermediate green is started and the steam is turned back on the calandria. The pan feed is controlled by the load on the agitator motor operating the feed valve. When the massecuite volume reaches 1000 cubic feet, half of it is drawn into a second similar pan. The pan which is used for graining is fed at a fast evaporating rate while the other pan is taken up more slowly. The obvious reason for this is to balance the boiling schedule.

When the final pan volume is reached, the feed is shut off and the pan Brixed to the required RDS as indicated by the motor load. Both pans are boiled at a constant absolute pressure of four inches of mercury and the boiling time is approximately four hours per pan. This method of boiling requires a mechanical agitator to provide circulation while the pan is held at a slow evaporating rate. The purpose of this is to establish a good grain footing on which to build the sugar crystals. By slowing the pan down, the small crystals are allowed to get the growth and crystal area necessary to take more sucrose in the form of feed liquor.

Table 1.—Table showing results of low raw sugar boiling program.

Year	Cut	True purity low raw pan	Massecuites % on beets	Cubic: feet/day	True purity molasses	Purity low raw-sugar
1961-1962	4620	79.40	12.4	12,200	60.67	94.6
1960-1961	4069	78.36	12.2	10.700	59.95	93.0
1959-1960	3579	76.58	11.4	8.800	60.70	93.1
1958-1959	3539	76.74	11.7	8,900	62.25	92.0
1957-1958	3473	77.07	10.9	8.100	63.33	92.8
1956-1957	3415	77.41	11.5	8.400	62.76	92.4

To control grain size on low raw sugar is was necessary to know the MA and CV of this sugar. This could most easily be done by developing a practical method for wet screening the low raw sugar. It would also be helpful if such a method could be used to get an estimate of the MA and CV on the crystals in the massecuite as dropped from the pan. Such a procedure was developed (1). It is an adaptation of the method of Saint and Trott (3) and consists of washing the raw sugar or massecuite free of syrup with a sugar saturated alcohol solution and then wet screening in more of the same solution. The crystals become separate and distinct and may be photographed under magnifica-

tion with a polaroid camera. The pictures and screen analyses give good guides to the sugar boilers and the result is a better quality of low raw sugar.

A table showing the MA and CV of the low raw sugar can perhaps best demonstrate the use of this control. A special chemist was employed during the sugar boiling work mainly to run the analysis on the low raw sugar for MA and CV. The results of one month's operation are shown in Table 2 for the Moses Lake Factory and the same table has the results of one week's operation at the Toppenish Factory. It is interesting that on about the 20th of October the MA went down and CV up, making the massecuite considerably harder to spin. These low MA's were caused by returning to the former sugar boiling practices. A return to the standard boiling procedure corrected the trouble as evidenced by the MA of .0128 on the 23rd of October.

From all indications the success of this method of boiling is dependent upon the use of a high purity graining charge and the growth of the crystals in this material as long as possible. The BPR will actually start down before any feed is added to the pan. This indicates that the sugar crystals have taken enough sugar from the mother liquor to actually lower the super-saturation. When the BPR starts down, more sugar must be made available for crystallization in the form of feed liquor. In actual practice, 60% or more of the crystal width may be attained in the graining charge before any feed is added to the pan. With the crystals firmly established, the remainder of the growth time can be spent getting as much sugar out of the molasses as possible.

Results

The standard boiling procedure increased the MA on the low raw sugar to as high as .0143 and an average of about .0115. At the start of the program the MA was about .0050. The plant capacity at Moses Lake increased from 3,579 tons per day in 1959 to 4,620 tons per day in 1961 using the same existing sugar end equipment. The purity of the low raw sugar increased from 93.1 to 94.6. These larger crystals and higher purity sugar permitted the affination of the low raw sugar to 99 purity and its return via the affinator to the white pan.

This program could not have been carried out or made successful without the cooperation and helpful suggestions of the sugar boilers, sugar end foremen, and particularly the factory supervisory personnel.

Table 2.—Table MA and CV of low raw sugars.

Date	Moses Lake North Crystallizer		Moses Lake South Crystallizer		Date	Toppenish Low raw sugar	
	MA	CV	MA	CV		MA	CV
1 Oct. 61	.0083	38	.0079	38	18 Jan. 62	.0093	24
2 Oct. 61			.0092	44	19 Jan. 62	.0090	34
3 Oct. 61			.0078	55		.0066	78
4 Oct. 61			.0085	46	20 Jan. 62	.0105	38
6 Oct. 61	.0107	45	.0097	53		.0102	38
7 Oct. 61	.0113	48	.0113	53	21 Jan. 62	.0125	33
9 Oct. 61	.0104	45	.0110	45		.0115	39
10 Oct. 61			.0117	36	22 Jan. 62	.0140	33
11 Oct. 61			.0117	39		.0144	22
12 Oct. 61	.0116	38	.0117	30	23 Jan. 62	.0115	37
13 Oct. 61	.0111	38	.0097	33		.0122	36
14 Oct. 61			.0113	32			
15 Oct. 61	.0143	31	.0124	29	24 Jan. 62	.0106	27
36 Oct. 61	.0143	31	.0143	31		.0130	41
18 Oct. 61	.0135	28	.0131	29	25 Jan. 62	.0143	37
19 Oct. 61	.0104	34	.0107	33	26 Jan. 62	.0130	34
20 Oct. 61	.0086	51					
21 Oct. 61	.0094	36	.0092	37			
23 Oct. 61	.0128	28	.0128	30			
24 Oct. 61	.0140	27	.0136	28			
25 Oct. 61	.0121	35	.0124	31			
26 Oct. 61	.0112	35	.0123	28			
28 Oct. 61	.0112	35	.0119	32			
29 Oct. 61	.0117	33	.0124	29			
30 Oct. 61	.0115	32	.0117	30			
31 Oct. 61	.0104	34	.0111	37			

Conclusion

A sugar boiling program of improved crystallization techniques and standard graining and boiling procedures, along with better pan instrumentation and controls, can give benefits by 1. increasing the capacity of the low raw sugar handling equipment, 2. increasing the purity of the low raw sugar, and 3. producing a quality sugar that is adaptable to affiliation.

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Effects of Nabam Solutions on Emergence of Larvae from Cysts of Heterodera schachtii in Aqueous Solutions and in Soil

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In areas infested with the sugar-beet nematode, economic production of sugar beets is made possible through discriminant use of rotation systems. Production of sugar beets is limited to once in three to six years, depending on the severity of infestation and local conditions that determine the persistency of this nematode pest.

Methods that will accelerate hatching of larvae from soil-borne cysts in the absence of host plants may enable shortening of rotations or increase the efficiency of control by rotations.

In previously reported tests, Steele (2)[2] found that solutions containing 1,000 parts per million of disodium ethylene bis dithiocarbamate (nabam) increased hatching of *Heterodera schachtii* Schmidt larvae as compared with tap water controls but only 60% as much as sugar beet root diffusate. Addition of 1,197 ppm of manganese sulphate or 1,316 ppm of zinc sulphate reduced the action to about the same as tap water. Nabam at concentrations of 2,000 ppm inhibited hatching of sugar-beet nematode larvae.

Nabam has recently been marketed as a wettable powder (Dithane A-40)[3]. A test was undertaken to compare the effects of this material and liquid nabam. In addition, attempts were made to determine whether hatching is permanently inhibited by concentrations of nabam exceeding 2,000 ppm and whether stimulatory effects of 1,000 ppm nabam would be retained after removal of the treatment solution.

Materials and Methods

In the first of two tests, seven treatments were checked for their effects on emergence of larvae from cysts of *Heterodera schachtii.* Four replications, each consisting of 40 cysts, were treated for 7 weeks with either tap water, beet root diffusate, 1,000 or 4,000 ppm nabam (Dithane D-14, a liquid formulation), or 1,000 ppm nabam (Dithane A-40). Equal numbers of cysts

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[2] Numbers in parentheses refer to literature cited.

[3] Dithane A-40 (93% nabam) and Dithane D-14 (22% nabam) were supplied by Rohm and Haas Company. Use of trade names and company names is for identification only and does not imply indorsement by the Department of Agriculture over similar ones not mentioned.

were treated for one week with either 1,000 or 4,000 ppm nabam after which the cysts were transferred to tap water where they remained an additional 6 weeks. Collection of diffusate and conduct of the test were essentially the same as described by Golden (1). Counts of larvae emerged from cysts are listed in Table 1.

A second test consisted of drenching various treatment solutions on soil contained in cylindrical paper cartons of 2 quart capacity. Each carton measured 3.5 by 13.5 inches. Before application of the treatments, three tea bags, each containing 50 cysts, were placed in each of 36 cartons 1, 6, or 12 inches below the soil surface.

Treatments consisted of tap water, beet root diffusate, 1,000 or 2,000 ppm nabam, 1,000 ppm nabam plus 658 ppm zinc sulphate, or 2,000 ppm nabam plus 1,316 ppm zinc: sulphate. The treated cartons were kept in a utility room. The temperature of the treated soil remained at about 65° F. Ten days after treatment the cartons were removed to the laboratory, where the cysts were recovered and placed in Syracuse watch glasses containing about 15 ml of beet diffusate. The cysts were treated with diffusate for three weeks to induce hatching and emergence of the remaining- larvae from the cysts. Counts of the remaining larvae are listed in Table 2. Data of both tests were analysed for statistical significance by the "analysis of variance" method.

Results and Discussion

Treatment of sugar beet nematode cysts with 4,000 ppm of nabam inhibited emergence of larvae (Table 1, treatment 1). However, considerable numbers of larvae emerged from cysts in tap water after they were removed from the 4,000 ppm solution

Table I.—Numbers of larvae emerging from cysts of *Heterodera schachtii* in 7 weeks with treatments as detailed in text.

Treatment		Replic ations 1	2	3	4	Total	Average	Hatch of 1st week[c]
1 Nabam	4.000	0	12	1	17	36	9.0	. 25.0
2 Tap water		433	1,320	654	552	2.959	739.8	60.3
3 Nabam[a]	1,000	2,384	1.364	1.398	2.431	7,577	1.894.3	78.3
4 Nabam"	4.000	2,232	1,438	2.784	2,676	9.130	2,282.5	.2
5 Nabam*	1,000	3.541	3,676	4 639	3,604	15.460	3.865.0	46.6
6 Nabam	1.000	4.790	4,661	4,380	5,258	19,089	4,772.3	27.7
7 Beet cliff.		6.952	6,997	7.201	8,857	30,007	7,501.8	34.0
Significance							**	
LSD .05							793.1	

[a] Cysts of treatments 3 and 4 were treated one week with the indicated solutions followed by treatment with tap water for 6 weeks.
[b] Treatment 5 was with Dithane A-40. All other nabam treatments were Dithane D-14.
[c] Expressed as a percent of the total number of larvae emerged from cysts in 7-week period.

(Table 1, treatment 4), indicating that nabam or a breakdown product of nabam is probably responsible for the inhibiting effect. Dithane A-40 (Table 1, treatment 5) gave greater hatches during the first week than did Dithane D-14 (Table 1, treatment 6).

Table 2.—Emergence of larvae remaining in cysts of *Heterodera schachtii* after recovery from 2 weeks in soil treated with nabam and beet diffusate. Average number of larvae per cyst from 6 replications.

Depth of cysts (inches)	Tap water	Nabam 1,000 ppm	Nabam 2,000 PP"»	Nabam 1,0(M) ppm + 658 ppm Zn SO4	Nabam 2,000 ppm + 1,316 ppm Zn SOi	Beet diffusate
1	326.3	311.1	322.0	299.6	319.0	126.4
6	313.2	316.3	298.4	346.9	313.4	83.7
12	319.5	344.7	339.8	347.6	341.8	309.4
Total	959.0	972.1	960.2	994.1	974.2	519.5
Average	319.7	324.0	320.1	331.4	324.7	173.2
Least significant mean difference (.05)						72.6

Results of the second test (Table 2) indicate that soil drenches of beet diffusate stimulated emergence of larvae from cysts. The effects of all nabam treatments to the soil were similar to the effects of tap water drenches. Absorption of nabam by soil or decomposition in soil may be contributing factors.

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Evaluation of Diffusates and Juice of Asparagus Roots for Their Nematocidal Effects on Heterodera schachtii

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Rohde and Jenkins (2)- isolated a compound from root diffusate and expressed root juice of *Asparagus officinalis* var. *altilis* L. toxic to several nematode species, fuice expressed from fibrous roots of asparagus killed 100% of *Trichodorus christiei* Allen 1957 after 18 hours, and solutions of the toxic material, drenched into the soil or sprayed directly on leaves of growing tomatoes, decreased *T. christiei* populations. Chemical tests led the authors to conclude that the toxic compound was a glycoside with the aglycone component of low molecular weight.

This paper reports tests to determine whether root diffusate or root juice of asparagus is toxic also to the sugar beet nematode *(Heterodera schachtii* Schmidt 1871).

Materials and Methods

Root diffusates of *Asparagus officinalis,* Golden State lettuce *(Lactuca sativa* L.), and sugar beet *(Beta vulgaris L.)* var. US 75 were tested to determine their effect on hatching of beet nematode larvae. Diffusates of the latter two plants were controls. Methods used in this test to obtain diffusates and cyst material and the procedures of the hatching test are described by Golden (1). The test was continued for six weeks. Treatments were replicated 4 times in individual watch classes containing 40 nematode cysts each. The nematodes emerging from cysts were counted and the data were analysed for statistical significance.

A second test was initiated, to determine the effects of various treatments on populations of *H. schachtii* in soil. The roots of asparagus plants were thoroughly fragmented in a blendor, the resulting material filtered, and the filtrate saved for use in the tests. Six-inch clay pots filled with soil containing an average of 25 cysts of *Heterodera schachtii* per gram received either 200 ml of asparagus juice, or 200 ml of tap water, or single seedling transplants of asparagus or lettuce. Pots to which asparagus juice or tap water was added were left fallow in the initial phase of the experiment. All treatments were replicated seven times, making 28 pots in all.

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[2] Numbers in parentheses refer to literature cited.

The asparagus and lettuce plants were allowed to grow four months, after which, they were removed from pots and discarded. At this time one sugar beet seedling was transplanted to each of the 28 pots. Fifty days later the plants were removed and examined for mature female nematodes.

Results and Discussion

In the first experiment, the average numbers of larvae emerged in the various treatments were, lettuce root diffusate 3,100, tap water 3,260, asparagus root diffusate 2,750, and sugar-beet root diffusate 10,870. Since the least significant difference at the 5% level was 1,070, it was concluded that neither lettuce nor asparagus root diffusate had any effect on hatching, while sugar-beet root diffusate had the usual stimulatory effect.

In the second experiment, the number of adult female nematodes observed on the roots of sugar beets grown 50 days in infested soil were not significantly different. It was concluded that treatments with asparagus root juice had no measurable effects on *H. schachtii.*

Summary

Soil drench treatments of asparagus juice or asparagus grown 4 months in infested soil did not decrease populations of *Heierodera schachtii.* Asparagus-root diffusate did not stimulate emergence of larvae from cysts of the beet nematode.

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Processing and Drill Performance of Monogerm Beet Seed

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Many attempts have been made in the United States since 1935 to reduce the hand labor required to thin beets to satisfactory field stand. Only partial success has been obtained, however, because until 1948 all sugar beet seed was multigerm in character. During the period 1940-1945 Bainer (1, 2)[2] developed segmenting and decorticating machines for multigerm seed, and this seed when graded to size and planted in good drills greatly reduced thinning labor. Possibility of a further reduction in labor came in 1948 with the discovery of monogerm beet seed by Savitsky (4).

For precision planting, monogerm seed must be polished to remove adhering flower parts before grading to size. Various types of polishers have been used—beet seed decorticators, barley debearders, and specially constructed polishers (3). The purpose of this paper is to report on the processing of monogerm seed using the Engleburg rice huller, the grading of seed for size, and the drillability of this seed in three makes of drills.

Monogerm Seed Processing and Grading

In 1960 an Engleburg huller was installed in the Western Seed Production Corporation cleaning plant at Cashion, Arizona, and 25 test runs were made, using monogerm varieties produced in this area. Data from these 25 runs are found in Table 1.

Table 1.—Characteristics of natural and polished monogerm seed.

Character	Type of Seed	
	Natural	Polished
Seeds per Pound	49.405	62,716
Percent Germination	82.7	82.4
Weight Per Bushel (pounds)	21.06	33.85
Percent Polishing Loss (by weight)		19.27

American #3 N polished seed had similar characteristics to those listed in Table 1 and was selected for size grading and planter tests. Seed of this variety was graded over round-hole screens with size perforations of 6 64", 7/64", 8/64", 9/64" and 10/64"; 200 pounds were sent to The Simon-Carter Company of Minneapolis, Minnesota for two dimensional gradings—diameter

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[2] Numbers in parentheses refer to literature cited.

and thickness. The diameter sizes were the same as those listed above, and the thickness grades were obtained using slot screens with the following widths of slot: 4/64", 5/64", 6/64" and 7/64". The proportions falling into the various classes for the two dimensions separately, are given in Table 2.

Table 2.—Percent of seeds by weight divided into 1/64 inch fractions for both diameter and thickness.

Diameter	-6	6-7	7-8	8-9	9-10	+10	Total
	5.8	16.5	32.83	24.08	12.02	8.07	100%
			85.43%				
Thickness							
	-4	3.81					
	4-5	43.44	> 72.25%				
	5-6	28.81					
	6-7	8.11					
	+7	1.26					
		85.43					

As shown in Table 2, 85.43 percent of the polished seed was from 6 to 10/64 inches in diameter, and of this size range, 72.25 percent was from 4 to 6/64's inches in thickness.

The characteristics of twelve of the two dimensional fractions are given in Table 3.

Table 3.—Percent recovery, percent germination and percent multigerm in twelve sizes of polished monogerm seed.

Thickness 64th inch	Diameter 64th inch	Percent multigerm	Percent germination	Percent recovery
-4	6-10	0	25.0	3.81
4-5	6-7	0	90.3	12.50
4-5	7-8	0	90.2	21.00
4-5	8-10	0	90.0	9.94
5-6	6-7	29.0	86.5	1.28
5-6	7-8	12.0	91.0	9.07
5-6	8-9	2.0	89.0	11.86
5-6	9-10	1.5	90.0	6.60
6-7	6-10	20-95	90.2	8.11
+ 7		100	90.6	1.26
	-6			5.80
	+ 10			8.77
				100.00

From these data, the following observations can be made:

1. -4/64" thickness seed was too low in germination to be used.

2. The 4 to 5/64" thickness sizes were 100 percent monogerm, regardless of diameter size.

3. The 5 to 6/64" thickness size contained some double germ seed, with the percentage decreasing with increasing seed diameter.

4. The 6+/64" thickness sizes were mostly double germ.

5. Seed of 6-7/64" diameter and 5-6 thickness is not usable because this seed being nearly round, has a high percentage of doubles and triples.

6. In regard to thickness, the usable portion of the seed was in the 4 to 6/64" range.

7. Diameter sizes 6-8 and 8-10/64" appeared to be the most usable fractions.

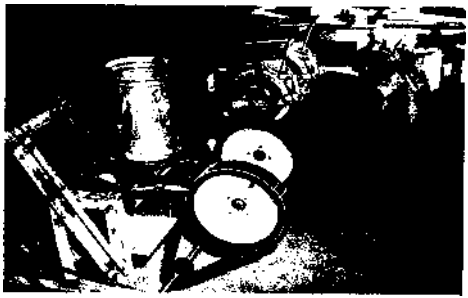

Figure 1.—Picture of drill test rack mounted with units of three makes of drills used in experiments at Rocky Ford, Colorado.

Drill Experiments

With two polished monogerm seed lots on hand, one of which was diameter graded, and the other graded for both diameter and thickness, drill tests were conducted with one unit of each of the following three makes of drills: International 185, John Deere 70, and the Milton. All three drill units were set up on a "rack" and driven at as nearly the same speed as possible (See Figure 1). The drill testing consisted of three 5-minute test runs averaged to obtain the percent cell fill. The method used for determining cell fill is outlined as follows:

1. Obtain the number of seeds per gram.

2. Plant the seed through the drill for 5 minutes and weigh in grams.

3. Convert the weight of seeds planted to number of seeds planted.

4. Determine the number of cells in the seed plate which pass the "cutoff" in the drill can in 5 minutes.

5. Calculate the cell fill by dividing the number of seeds planted, by the number of cells available, and express in percent.

It is well known that errors can be obtained by the use of this method of calculating cell fill, since if one cell receives two seeds, and one receives no seed, the result is 100% cell fill. However, the error is minimized to the point of little importance when accurately sized seed is planted through plate cells of appropriate diameter and thickness. If seed size is not correct for the cell size of the drill plate, excessive grinding of seed will be obtained.

The first drill tests were made on seed graded for diameter only, using a variety of plate cell sizes and thicknesses, using the International and the John Deere. These tests were run on seed sized at 1/64 inch size differences, and on combinations of two sizes. In all of these preliminary trials excessive grinding was obtained, with the exception of the 6 to 7/64" diameter seed. Close inspection of the seed sizes and shapes indicated that seed thickness was of more importance than had been expected.

Most of the usable dimensional graded seed (Table 3) fell into 3 sizes: 6 to 7/64" diameter and 4 to 5/64" thickness; 7 to 8/64" diameter and 4 to 5/64" thickness; and 8 to 9/64" diameter and 5 to 6/64" thickness. After repeated drill tests it became clear that two thickness grades could not be combined and still produce the best precision planting. However, within the two thickness grades a tolerance of 2/64" diameter size was possible. The sizes finally determined were 4 to 5/64" thickness and 6 to 8/64" in diameter and 5-6/64" thickness and 8-10 64" in diameter. The smaller seed was designated as Number 1 and the larger as Number 2. Percent recovery, seeds per pound and percent germination are given in Table 4 for these sizes and also on the polished and unpolished seed.

The change in seed characteristics made from polished seed with the Engleburg huller is clearly indicated in Table 4. Weight per bushel was greatly increased by polishing as well as by grading to size. Percent germination was increased from 87.0 percent for the unpolished seed to 92.5 and 94.8 for the No. 1 and No. 2

Table 4.—Seed characteristics of unpolished seed, polished seed and the two dimensional sizes designated as No. 1 and No. 2 (Am #3N).

Seed type	Seeds per pound	Weight per bushel	Percent germination	Percent recovery
Unpolished	49,405	21.1	87.0	100.0
Polished	62,716	33.9	88.0	80.27
No. 1	72,000	43.5	92.5	26.87)41.69*
No. 2	44,500	36.7	94.8	14.82)

* on unpolished basis

usable fractions, respectively. A comparison of number of seeds per pound indicates that the large seed in the original sample may not have been polished as much as the small seed.

Results of Drill Tests on No. 1 and No. 2 Polished Seed

In the testing of the drills, only certain seed plates were available in cell diameter size and thickness for the International and John Deere drills. Since International plates were easily machined to various thicknesses this drill was used extensively. In no case were cell diameters changed on plates of either drill.

Since both International and John Deere drills performed very similarly, some of the data on cell fill is a combination of the results obtained with both drills. Table 5 gives the percent cell fill data using seed plates of .083 thickness on No. 1 seed with different cell diameters, travel rates, and planting rates.

Table 5.—Percent cell fill on No. 1 seed with .083 seed plate thickness with different cell diameters, travel rates and planting rates.
(John Deere and International Combined)

Diameter	Miles per hour	Seeds planted per row foot	Percent cell fill
81/2/64"	2.5	6.0	90.0
	2.5	8.5	85.5
(.133")	3.0	6.0	87.7
	3.0	8.5	84.5
9/64"	2.5	6.0	100.6
	2.5	8.5	98.1
(.141")	3.0	6.0	100.1
	3.0	8.5	98.3

In Table 5 many comparisons can be made; but the first conclusion to be reached is that cells of 81/4/64" diameter are not large enough for seed with maximum diameter of 8/64". In speed of travel, 2.5 miles per hour was slightly better than 3.0, especially when cell size of $8^{1/2}$/64" was used. As an average, a planting rate of 6 seeds per foot of row gave a percent cell fill of 94.6 as compared with 91.6 for the planting rates of 8.5 seeds per foot of row. This result is to be expected, since increased planting rate is obtained by increased speed of seed plate travel, which is the equivalent of increasing the miles per hour travel rate. Thus if 8.5 seeds are planted per foot of row at 2.5 miles per hour instead of 6 seeds, seed plate travel converted to miles per hour travel rate will be: $= 3\frac{8.5}{6.0} \times 2.5$ miles per hour. The data given in Table 5 indicate that very satisfactory cell fill was obtained with plates of .083 thickness and cell diameter of 9/64" when planting rate was 6 seeds per foot of row at both 2.5 and 3.0 miles per hour travel rate.

Table 6.—The effect of cell size and shape on percent cell fill with the Milton Drill.

Type of seed wheel		Seeds planted per row foot	Miles per hour travel	Percent cell fill
Size of cell in 64th inch	Number of cells			
$8^{1/2}$-$5^{1/2}$-$7^{1/2}$	140	6.0	2.79	92.6
8-6-7	180	6.0	2.94	98.9
8-6-8	140	6.0	2.79	100.4

The effect of cell size on percent cell fill was also determined for the Milton drill using No. 1 sized sized seed (Table 6).

The effect of plate thickness on percent cell fill was also studied. Due to the difficulty in obtaining seed plates in the various thicknesses, these tests were limited to No. 2 seed (8 to 10/64" diameter, 5 to 6/64" thickness), using the International drill. Cell diameter for these tests was 11 64". The results are given in Table 7.

Table 7.—Comparisons of plate thickness on percent cell fill, with No. 2 seed.

Plate thickness in inches	Cell diameter 64th inch	Miles per hour	Percent cell fill
.090	11/64	2.96	82.0
.103	11/64	2.96	100.4
.110	11/64	2.96	111.5

These results indicate the great importance of seed plate thickness on percent cell fill. In this test, a difference in plate thickness of .020" made a difference of 29.5 percent cell fill.

In Table 5 data were given on the effect of speed of travel on percent cell fill. This was investigated further with all three test drills using No. 1 seed, and the John Deere and International on No. 2 seed. The plates used for the two seed sizes were those which had been found to be satisfactory for both cell size and thickness and are listed as follows:

Seed Type	Drill	Cell Diameter	Thickness
No. 1	John Deere	9/64"	.083"
	International	9/64"	.083"
	Milton	8-6-8 /64 "	
No. 2	John Deere	11/64"	.103"
	International	11/64"	.103"

Tables 8 and 9 give the effect of rate of travel for both seed sizes at 6 seeds per row foot planting rate, on percent cell fill.

As shown in Table 8, all three drills planting No. 1 seed showed a significant reduction in percent cell fill for each increased planting speed. In Table 9 the data show the same trend

with No. 2 seed, but with little difference between the two lower speeds. With speeds nearing 4 miles per hour there was a definite drop in percent cell fill.

Table 8.—The effect of travel speed with three different drills planting six seeds per row foot of No. 1 size on percent cell fill.

Miles per hour	Drill make		Percent cell fill
2.56	International		103.7
3.01			101.0
3.91			99.5
2.56	John Deere		101.1
2.96			99.8
3.89			96.4
2.48	Milton		99.5
2.92			96.4
3.83			88.7
F. Value		89.5	
Sign. Diff. (19:1)			1.21

Table 9.—The effect of travel speed with two different drills planting six seeds per i- foot of No. 2 size on percent cell fill.

Miles per hour	Drill make		Percent cell fill
2.56	International		100.8
3.01			100.4
3.91			96.4
2.56	John Deere		101.0
2.96			100.3
3.89			96.5
F. Value		13.23	
Sign. Biff. (19:1)			.46

Summary of Results

1. Seed used in these experiments was polished with the Engleburg rice huller and graded to size (a) over round hole perforated screens for diameter and (b) over round hole and slot screens for diameter and thickness.

2. Preliminary drill tests indicated that both diameter and thickness grading was necessary for accurate planting of seed.

3. Two sizes of polished seed, representing 41.7 percent of the total per acre yield of seed were considered satisfactory for precision planting:

No. 1—6 to 8/64 inch in diameter; 4 to 5/64 inch in thickness

No. 2—8 to 10/64 inch in diameter; 5 to 6/64 inch in thickness.

4. Drill tests of these two sizes indicated that thickness of seed plate was most important. Cell diameter was also important.

5. Travel speed of approximately 3 miles per hour gave approximately 100 percent cell fill with three beet seed drills when equipped with the correct seed plates, with a planting rate of 6 seeds per row foot.

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Interrupted Nitrogen Nutrition Effects On Growth, Sucrose Accumulation and Foliar Development of the Sugar Beet Plant[1]

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Under natural conditions, nitrogen supply is frequently limiting to the growth of plants. This may result from seasonal variations in the nitrogen content of soils and in rates of nitrogen absorption and assimilation by plants as well as from a low supply of native nitrogen. In agricultural environments, nitrogen nutrition may be maintained at an optimum level with suitable fertilizer practices. This usually involves more than simply supplying sufficient nitrogen for luxury consumption since the yield and quality of the economically useful yield of many crop species are maximal when nitrogen has been in marginal or deficient supply during critical stages of plant development (8)[3]. Thus, the response of plants to fluctuations in nitrogen supply is of importance agriculturally as well as biologically.

Sugar beet is well suited to studies on the effects of fluctuating nitrogen nutrition on plant growth. It has an indeterminate vegetative growth habit and tissue analysis procedures (15) have been developed for assessing plant nutrient status. Considerable information has been obtained concerning the responses of sugar beet to nitrogen starvation, (4,5,6,9,11,12) but, except for an experiment by Ulrich (9), much less is known about its recovery from the deficient condition. Such information should have ecological significance as well as having practical application to commercial production.

The experiment reported here was designed to provide information on the time course of the nitrogen starvation and recovery responses of sugar beet. Particular attention was given to changes in leaf growth.

Methods and Materials

Sugar beet plants were grown outdoors in 10-gallon pots at Davis, California, during the 1960 season; environmental data are summarized in Figure 1. Air temperature was recorded at 4.5 feet with a thermocouple and a recording potentiometer. Solar radiation data, obtained with a horizontally exposed Eppley pyrheliometer, were supplied by the Davis weather station.

[1] This study was supported in part by a grant from the beet sugar companies operating in California and the California Beet Growers Association, Ltd.

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[3] Numbers in parentheses refer to literature cited.

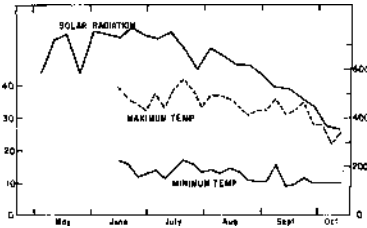

Figure 1.—Maximum and minimum air temperature (5-day means) and solar radiation (7-day means) in the experimental environment.

Cultural procedures were similar to those previously employed (4, 5). The pots (used carbide cans, 32 cm diameter and 52 cm high) were provided with bottom drainage and were filled with no. 2 grade vermiculite. On May 1, 10 seed units (variety MS NB1 X NB4)[4] were planted per pot. The seedling plants were thinned at regular intervals, so that by June 10 only two plants remained per pot. The pots were spaced a minimum of 50 cm apart to prevent foliar competition between pots.

An excess of half-strength modified Hoagland's solution made up with tap water (4) was supplied daily to each pot. This level of nutrition was maintained until July 23 when differential treatments were begun. The treatments (Figure 2) consisted of supplying the plants for various lengths of time with a solution free of added nitrogen. The tap water contained 0.04 mMole NO_3-N per liter. Experience has shown that the severity of a nitrogen deficiency is not measurably influenced by using solutions ranging from 0.00 to 0.08 mM $N0_3$-N per liter and the low-nitrogen solution used here will be referred to as the minus-nitrogen solution. Tn this minus-nitrogen solution, $CaCl_2$. was substituted for $Ca(NO_3)_2$ and K_2SO_4 for KNO_3. The vermiculite was leached with tap water at the start of a nitrogen-deficiency period.

Data on foliar development were collected for key treatments throughout the growing season. Leaf appearance rate was obtained by tagging weekly the smallest leaf over 5 cm in length, and counting the number of leaves between it and the previously tagged leaf. Leaf area per plant was estimated weekly by tracing every fifth living leaf and determining the area with a planimeter. Dead leaves were collected and counted at weekly intervals.

The plants in ten pots from each of various treatments, as indicated in Figure 2, were harvested at three-week intervals

[4] Dr. J. S. McFarlane, ARS, U. S. Department of Agriculture. Salinas, California, provided the seed.

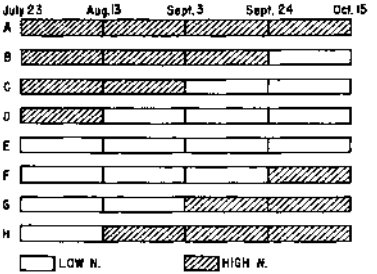

Figure 2.—Treatment combinations. All plants were supplied with high nitrogen until July 23. Minus-nitrogen solution was then used for varying periods of time as indicated. Ten pots of each treatment were harvested at 3-week intervals as follows: July 23—A; August 13 and September 3—A and B; September 24—A, B, C, and G; October 15—all treatments.

beginning July 23. The final harvest on October 15 included 10 pots from every treatment. Eight recently mature leaves were selected from each pot for tissue analysis (3). Roots of harvested plants were separated from tops at the base of the oldest living leaf, washed to remove vermiculite and secondary roots, and the crowns were then cut from the roots at the lowest leaf scar. Fresh weights were recorded for individual roots, tops, and crowns. Dry weight of tops was obtained after drying at 70° C.

The two roots in each pot were pulped together and three 26-gram samples of pulp were frozen on dry ice. These were analyzed later for sucrose (with hot water digestion) by the Sachs-le Docte procedure (2)[5] One 26-gram sample was taken for dry weight. Sucrose yields were calculated on total beet weight (root -j- crown) with the assumption that crowns had the same sucrose concentration as roots (4).

Results

Plant nutrient status

Incipient nitrogen-deficiency symptoms usually were apparent within 2 weeks after a group of plants had been changed to the minus-nitrogen solution. After 3 weeks, the older leaves were lighter green and the expanding leaves were smaller than on high-nitrogen plants. Otherwise the general appearance of high- and minus-nitrogen plants was similar. Extended nitrogen deficiency resulted in fewer leaves with short petioles and small, green leaf blades occurring in flattened rosettes. Deficient plants responded

[5] Sucrose analyses on the frozen samples were made with the assistance of the Spreckels Sugar Company, Woodland, California.

rapidly to nitrogen return with the renewed production of new leaves. Plant tissue analyses (Figure 3) confirmed the rapid development of nitrogen deficiencies and the equally rapid recovery.

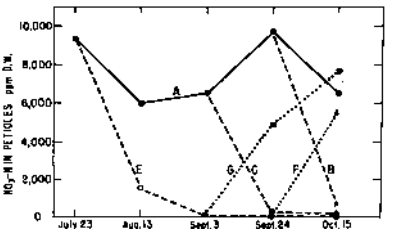

Figure 3.—NO_3-N content (ppm dry basis) in petioles of recently mature sugar beet leaves. Critical nutrient concentration is 1000 ppm. Letters refer to treatments.

Growth with high and minus nitrogen

In Figure 4, the nitrogen-deficiency responses shown by treatment E are compared with those of the high-nitrogen control (treatment A). These results are similar to the patterns depicted by Bouillene et al. (1) and Ulrich (10, 11) for high-nitrogen plants and by Ulrich (11) for nitrogen-deficient plants. With high nitrogen, weight of tops and of storage roots increased throughout the 12-week period from July 23 to October 15. On August 13, 3 weeks after nitrogen cut-off, the minus-nitrogen plants could be distinguished from the high-nitrogen plants by appearance, but the weights of tops and of storage roots were equal for the two treatments. Growth rates of tops and of storage roots were sharply reduced by nitrogen deficiency after August 13 and no further increase in root size was observed after September 13. By October 15, high-nitrogen roots were nearly twice as large as those obtained with continuous nitrogen deficiency. The growth of new leaves was reduced and top weight declined as the older leaves died. Size of crowns was greatest with high nitrogen reflecting the greater amount of top growth which had occurred (Table 1).

The sucrose concentration in roots of plants maintained at high nitrogen was relatively constant during the season at about 12% (fresh weight basis). Since this equilibrium concentration has been found to be inversely related to night temperature (10, 12, 14), slightly higher concentrations were anticipated in the fall (Figure 1, Table 2). However, the midsummer sucrose levels

were slightly, but significantly, higher than later values, suggesting that solar radiation (Figure 1) or root size (5) may have been the controlling factor in sucrose concentration.

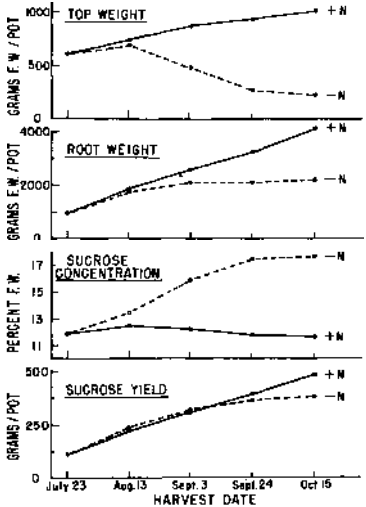

Figure 4.—Time course of sugar beet growth at high (treatment A) and minus nitrogen (treatment E).

Table 1.—Harvest results on October 15 from sugar beet plants grown with varying nitrogen nutrition. (Means of 10 replications).

Treatment	Nitrogen cutoff date	Nitrogen return date	Beet root plus crown Fresh wt g/pot	Crown %	Sucrose %	Sucrose g/pot	Tops Fresh wt g/pot	Height cm	Diameter cm
A			4100	6.1	1 1.7	~~480	1020	30	~"62
B	9/24		3730	6.3	12.5	468	854	30	60
C	9/3		3460	4.8	13.9	480	582	24	52
D	8/13		2880	4.5	15.7	452	431	19	51
E	7/23		2160	3.2	17.6	379	207	13	48
F	7/23	9/24	2140	3.6	15.7	336	280	18	43
G	7/23	9/3	2550	5.4	13.6	347	407	20	46
H	7/23	8/13	3340	6.9	11.9	396	869	30	59
	LSDos		205	0.9	0.4	28	120	[2]	3
	Ii		100.4**	14.9**	188.3**	37.0*	49.6**	67.2**	44.9**
	Error M.S. (72 df)		53,023	1.173	0.2353	964.7	18,176	6.291	10.92
	C.V. %		7.6	21.2	3.4	7.4	23.2	10.9	6.1

[1] Required $F_05 = 2.14$; $F_{01} = 2.90$

Table 2.—Summations of minimum temperatures for 4 weeks prior to various harvest dates and the sucrose concentrations in beet roots observed with high-nitrogen nutrition.

Harvest date	Heat sum[1] °C-days	Sucrose
July 23	411	**11.9**
August 13	412	**12.4**
September 3	367	**12.1**
September 24	313	**11.8**
October 15	296	**11.7**
LSD.-,		0.3
F[2]		6.33**
Error M.S. (45 df)		0.12
C.V. %		2.9

[1] Minimum temperatures above 0° summed daily for 28 days prior to harvest.
[3] Required F_{05} = 2.58; F_{01} = 3.77

Nitrogen deficiency caused sucrose concentration to increase gradually at first and then more rapidly; a maximum of 17.4% was reached after 9 weeks of deficiency (Figure 4). Sucrose yields were the same in high- and minus-nitrogen treatments with up to 6 weeks of deficiency indicating that the increase in sucrose concentration compensated for the reduction in root size.

Nitrogen-deficiency treatments (B, C, D) beginning August 13, September 3, and September 24 gave response patterns similar to those shown in Figure 4 for treatment E, but the changes were not as great. As an example, while 17.4% sucrose was attained 9 weeks after the July 23 cutoff, only 15.7% was reached 9 weeks after the August 13 cutoff. On October 15, the high-nitrogen treatment (A), and the 3- and 6-week terminal deficiency treatments (B, C), all yielded similar amounts of sucrose, and while higher sucrose concentrations were obtained after 9 and 12 weeks of deficiency, root weights from these treatments (D, E) were reduced to the extent that less total sugar was produced.

Nitrogen return responses

The influence of a midseason interruption in nitrogen nutrition may be assessed most easily from data obtained on October 15 (Table 1). Root weights were reduced 25%, when plants were deficient for 3 weeks (July 23 to August 13; H), even though nitrogen was available throughout the remainder of the growth period. Since this reduction was not apparent on August 13 (Figure 4), it occurred after nitrogen was returned. A 6-week midseason nitrogen deficiency (G) resulted in an even greater reduction in beet root weight. Beet root weight was the same with a 9-week deficiency followed by a 3-week nitrogen return (F) as with 12 weeks continuous deficiency (E). Apparently a brief period of nitrogen return did not effectively stimulate root growth once growth stoppage had occurred.

Sucrose concentrations in the storage roots on October 15 (Table 1) were also influenced by nitrogen return. In general, each 3 weeks of nitrogen-return reduced sucrose concentration approximately 2 percentage units below what would have been attained with continuous minus nitrogen. Thus, 15.9% sucrose was attained by September 3 after 6 weeks of nitrogen deficiency and this was reduced to 13.6% during the subsequent 6 weeks at high nitrogen (G). The increase in sucrose concentration noted after a midseason deficiency of 3 weeks (H) was not retained when nitrogen was returned and this treatment yielded less sucrose per pot than the high-nitrogen control because of the 25% reduction in root weight. At the other extreme, returning nitrogen on September 24 to plants which had been deficient for 9 weeks (F) lowered sucrose concentration but did not increase root weight and this treatment yielded less sucrose than was obtained with continuous nitrogen deficiency (E).

Top development

Number of leaves The rate of new leaf appearance, as shown for treatments A, E, and G in Figure 5, was markedly affected by nitrogen deficiency. With adequate nitrogen, 4 ± 1 new leaves appeared per plant each week. This rate was maintained throughout the season and was not greatly influenced by changes in plant age or climate. A lower rate of leaf appearance was noted during the second week following induction of nitrogen deficiency; the rate continued to decline to a minimum of < 1 per week by the sixth week of deficiency. Nitrogen-deficient plants maintained this rate for the remainder of the season, apparently by utilizing the small amount of nitrogen in the solution and nitrogen supplied to the apical meristem from other parts of the plant. With later nitrogen deficiency dates (B, C, and D; data not shown), the leaf-appearance rate declined even more rapidly. A greater rate of nitrogen utilization by the larger plants may have accounted for this, as evidenced by tissue analysis date (Figure 3). When nitrogen was returned to deficient plants the rate of leaf appearance increased within 2 weeks to 4 ± 1 per week.

Since the leaf-appearance rate was constant when the plants were supplied with a high level of nitrogen, there was a constant increase in the accumulated total of leaves. By October 15, high-nitrogen plants (A) had produced an average of 74 leaves while plants which had been supplied with minus-nitrogen solution after July 23, produced only 47 leaves. Of particular interest was the observation that no compensatory increase in number of leaves occurred after nitrogen return (G; Figure 5), i.e., the leaf-appearance rate did not exceed 4 per week. A compensatory

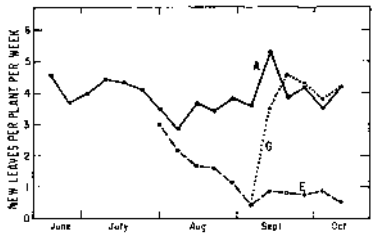

Figure 5.—Number of new leaves per plant which appeared each week with high (A) and minus (E) nitrogen and nitrogen return (G).

increase would be expected if during nitrogen deficiency leaf initiation had continued at the normal rate while leaf expansion and hence leaf-appearance rate were inhibited. Microscopic examination of terminal apices from high- and minus-nitrogen plants failed to reveal any marked differences in the number of leaves less than 5 cm long indicating that leaf appearance and leaf initiation rates were equal.

The number of dead leaves collected was not significantly affected by nitrogen nutrition. There was a tendency for leaves which had matured under adequate nitrogen, to undergo earlier senescence as evidenced by yellowing and to die somewhat sooner when subjected to nitrogen deficiency. However, final counts on October 15 showed equal numbers of dead leaves in both high- and minus-nitrogen treatments. Thus, because of the lower rate of leaf appearance, the nitrogen-deficient plants had only about one half the number of living leaves as the high-nitrogen plants.

Leaf area With high nitrogen, leaf area remained less than 1 dm^2 per plant during the first month after planting (Figure 6). It increased rapidly thereafter to a maximum of 38 dm^2 per plant in September and then declined. A decrease in leaf area, which continued throughout the remainder of the season, was apparent after 2 weeks of growth for plants on minus-nitrogen solution. A return to high nitrogen following a 6-week deficiency tended to slow the rate of decline.

The decline in leaf area in late September observed with high nitrogen was due to a smaller size of the new leaves as shown by the leaf growth curves in Figure 7 which are representative of the observations for treatments A, E and G. With high nitrogen (A), the maximum areas of leaves 15-20 were typically about twice those attained by later leaves. In addition, leaves initiated during July and August had slower growth rates than those

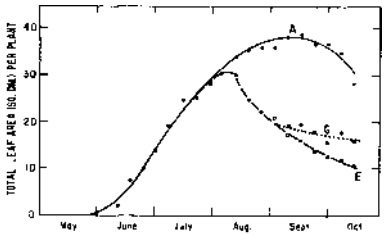

Figure 6.—Total area of living leaves per plant at high (A) and minus (E) nitrogen and with nitrogen return (G).

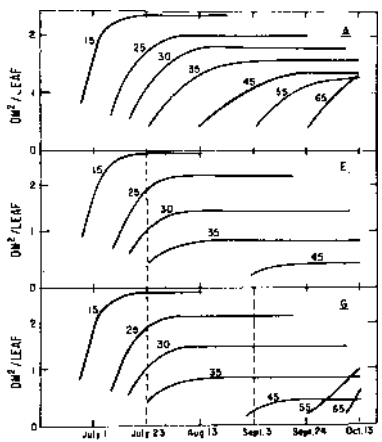

Figure 7.—Stylized growth curves for various leaves at high. (A) and minus (E) nitrogen and with nitrogen return (G). These curves are representative of the changes observed for the 20 plants in each treatment.

produced earlier or later. Within 3 weeks after the change to minus-nitrogen solution (E), the enlargement of expanding leaves ceased, and the new leaves which appeared subsequently had small blades and short petioles. When nitrogen was returned to such plants, they continued to produce only small leaves during the subsequent 2 weeks, although the leaf-appearance rate did increase. These new leaves formed a flattened rosette of new

growth within the whorl of older leaves. Only those leaves which appeared 3 or more weeks after nitrogen return enlarged normally.

Biological yields

Dry weights of tops and roots were determined to provide estimates of biological yields. Percent dry matter of both tops and roots increased with nitrogen deficiency. Total dry weight of the whole plant (dead leaves and fibrous roots excluded) at final harvest varied with nitrogen treatment (Table 3). The highest production was obtained with continuous high nitrogen. Production was reduced 5 to 10% by terminal deficiencies of 3 and 6 weeks, although sucrose yields remained unchanged. Thus, the proportion of the total dry weight which occurred as sucrose (coefficient of economic yield; 7) was increased from 48% with high nitrogen to 58% with 6 weeks of nitrogen deficiency prior to October 15.

Table 3.—Distribution of dry matter in sugar beet plants on October 15 as influenced by previous nitrogen nutrition. (Means of 10 replications.)

Treatment	Root plus crown g/pot	Green tops g/pot	Whole plant g/pot	Accumulated old leaves g/pot	Season total g/ Pot	K[1] %
A	766	143	908	96	1004	48
B	733	125	858			
C	727	86	813	93	906	53
D	665	69	734			
E	543	37	580	79	659	58
F	496	49	545			
G	533	64	597	76	673	52
H	631	122	753			
LSDor,	41	11	49	11	51	
F	49.3[2]	112.3[2]	68.9[2]	7.43	90.3*	
Error M.S.	2140	134.5	2684	137.7	3266	
C.V. %	7.1	13.3	7.2	13.6	7.0	

[1] K_v (Coefficient of economic yield) = $\frac{\text{sucrose yield}}{\text{total season dry wt}} \times 100$; calculated from treatment means.

[2] Required F05, = 2.14; F01 = 2.90. 72 df for error.

[3] Required F05 = 2.86; F01 = 4.38. 36 df for error.

The net assimilation data (Table 4) indicate that photosynthetic activity was reduced greatly by extended nitrogen deficiency. Net assimilation rates were equal for both high- and minus-nitrogen plants for the first 6 weeks; net assimilation rates for the minus-nitrogen plants then declined to a very low level. Gross assimilation rates cannot be obtained from these data since respiration losses were not estimated. Whole plant respiration was probably less with nitrogen deficiency due to the reductions in growth rate and the smaller size of tops and roots. However, because of the sharp reduction in leaf area, the proportion of the gross photosynthate used in respiration may have increased.

Table 4.—Dry matter accumulation by sugar beet plants at high- and minus-nitrogen status. (Calculations based on means of 10 replications.)

	Continuous high nitrogen (Treatment A)			Continuous minus-nitrogen (Treatment E)		
Interval	Dry wt. increase of tops and roots g/pot	Mean[1] leaf area dmVpot	Mean-' N.A.R. g/dm^2 day	Dry wt. increase of tops and roots g/pot	Mean[1] leaf area dm^2/pot	Mean-N.A.R. g/dm^2 day
7/23 - 8/13	154	.58	.126	151	58	.124
8/13 - 9/3	188	70	.128	131	49	.127
9/3 - 9/24	137	74	.088	12	35	.016
9/24 - 10/15	158	68	.111	14	22	.030

[1] Mean leaf area per pot represents the mean of the 4 weekly measurements obtained during the 3-week growth period. See Figure 6 for time course of leaf area changes in treatments A and E.

[2] N.A.R. (net assimilation rate) defined as grains dry matter accumulated per dm- leaf area per day.

Discussion

The sequence of leaf shapes and sizes observed at high nitrogen is of particular interest. Bouillenne et al. (1) found that "juvenile" sugar beet plants produce a series of leaves with broad, rounded blades and short petioles, while "adult" plants (storage root enlarging) produce leaves with small, narrow blades on longer petioles. Ulrich (13) found that leaf shape is influenced by climate, and that these two shapes are produced in cool and warm climates, respectively. In the present experiment, temperature differences (Figure 1) between early and late summer do not appear to account for differences in the size and shape of leaves; leaves produced during cool weather in September were adult shape, while leaves produced during slightlv warmer weather in June were juvenile shape. All leaves were light green with light-colored petioles and thus corresponded to warm climate leaves by Ulrich's criteria (13).

As leaf area per pot increased, transpiration increased and wider fluctuations in water content probablv led to internal diurnal water deficits of increasing intensity. While brief wilting was noted only on extremely hot or windy days, moisture variations in the upper half of the available moisture range will influence leaf enlargement (C B. Shah and R. S. Loomis, unpublished). We have observed that one third of the available moisture in a 10-gallon pot may be used within 24 hours after watering. Thus the tendency towards smaller leaf size and slower leaf growth during August can be attributed in part to moisture deficits. However, this does not explain the continued production of small leaves in September and October when such deficits would have been much less pronounced. It seems likely that physiological age or plant size in some way controlled leaf size.

Nitrogen deficiency reduced leaf initiation and leaf enlargement but only leaf initiation recovered quickly when nitrogen was returned. This suggests that cell division within the developing leaf was reduced in the nitrogen-starved plants in agreement with the observations of Morton and Watson (6). It also suggests that cell division in the apex, which accounts for the initiation of new leaf primordia, was renewed while cell division within the expanding leaf was not renewed.

The physiological bases for the continued suppression of leaf enlargement and storage root growth after nitrogen return are not apparent. Particularly puzzling is the large effect that a 3-week interruption in nitrogen nutrition had on subsequent growth. It may be that nitrogen deficiency has such a general debilitating effect at all levels of metabolism and organization that considerable time is required for recovery. Our speculations have also included two, more specific, possibilities.

1. Nitrogen was resupplied as nitrate and reduction to the ammonium level must precede its utilization. Limitations in nitrate reductase activity (the enzyme is adaptive; 8) or in distribution of the assimilated nitrogen might effectively extend the period of nitrogen starvation in some plant tissues even in the presence of abundant nitrate. If this were the case, different recovery responses would be obtained if nitrogen were resupplied in a reduced form.
2. There is also a possibility that nitrogen deficiency caused injury to meristematic tissues in the root, and that some or all of the active supernumerary cambia failed to recover or were slow to resume activity when nitrogen was resupplied. The plate meristems in the expanding leaves appear to have behaved similarly while the terminal meristem recovered rapidly.

The results of the present experiment may also serve as basis for predicting optimum nitrogen-management practices for commercial production. With abrupt removal of nitrogen from the rooting medium, the transition from luxury level to deficient level of nitrogen nutrition required about 3 weeks. Maximum sucrose concentration, i.e., maximum quality, was attained after an additional 3 to 6 weeks with sucrose yields equal or higher than obtained with high nitrogen. As in earlier experiments *(4,* 5), these results indicate that field-grown sugar beet plants should be permitted to become nitrogen deficient at least 6 weeks prior to harvest. In fact, a longer period may be desirable for plants grown in soil where roots are able to continue growing into undepleted media and where nitrogen continually becomes available through nitrification processes.

Since plants which were returned to high nitrogen after 6 or 9 weeks of deficiency yielded less sucrose than either the continuous-high or continuous-minus nitrogen treatments, a further conclusion of practical significance appears warranted; viz., if nitrogen deficiency occurs within 3 months of harvest it would be better to continue the deficiency than to apply additional nitrogen. A similar response would be obtained if the resupply occurred naturally, e.g., if nitrogen, which had accumulated in the surface soil as a result of furrow irrigations during a dry season, was moved downward into the root zone by late season rains.

Summary

The responses of sugar beet to high- and minus-nitrogen nutrition, and during recovery from the deficient condition, were studied using nutrient cultures. Particular attention was given to growth of storage roots and of individual leaves, and to changes in sucrose content of storage roots.

At high nitrogen, new leaves were initiated at a relatively constant rate which was influenced little by other environmental factors. Leaf area per plant reached a maximum by mid-September and then declined due to a progressively smaller size of the new leaves. Nitrogen deficiency reduced root growth, rate of leaf initiation, leaf area, and dry matter accumulation, and increased sucrose concentration in the root. The degree of these responses was dependent upon the length of the nitrogen-deficiency period with sucrose concentration reaching a maximum after 9 weeks. The increase in sucrose was sufficient to compensate for the smaller size of roots so that equal amounts of sucrose were obtained from high- and minus-nitrogen plants during the first 9 weeks of the deficiency.

When nitrogen was returned after a brief deficiency, leaf initiation was renewed and the amount of sucrose accumulated in the roots declined. However, root growth and leaf expansion continued to be limited during the period of recovery at high nitrogen. Lower sucrose yields were obtained by returning nitrogen to the deficient plants than by allowing the deficiency to continue.

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Greenhouse Chambers for Small Seed Increases

J. S. MCFARLANE AND I. O. SKOYEN[1] [2]

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The sugar beet breeder must frequently make small seed increases of breeding lines and hybrid combinations. These increases can be made in field isolations, but costs are high and great care is required to avoid contamination from outside pollen. Greenhouse chambers ventilated with filtered air are widely used in Europe for producing small quantities of seed. Wood et al. (l)[3] developed a compartmented greenhouse at Longmont, Colorado, which uses the principle of negative pressure for ventilating and cooling. After a study of these facilities a group of 12 compartments were constructed at the U. S. Agricultural Research Station, Salinas, California, in 1961.

A prefabricated, aluminum-framed greenhouse without door or roof vents was used as the basic unit (Figure 1). The greenhouse measured 32 X 9 feet and was divided into 6 sections by using standard commercial partitions. Each section was subdivided into 2 equal-sized chambers by cross partitions constructed of vinyl plastic film. The planting area within the chambers measured 57 X 50 inches. All seams and joints between chambers were sealed with a caulking compound or plastic-cement. Each compartment was entered through modified, commercial ventilating sash hinged at the eave line (Figure 1).

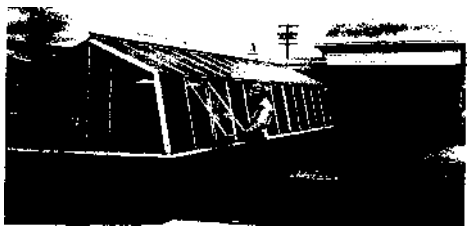

Figure 1.—Compartmented greenhouse used for production of sugar-beet seed at Salinas, California. Filtered air from fan house (top, right) is supplied to each chamber through underground ducts.

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[2] The authors express their appreciation to F. A. Araujo of the U. S. Agricultural Research Station, Salinas, California, for help in designing and constructing the isolation chambers.

[3] Number in parentheses refers to reference.

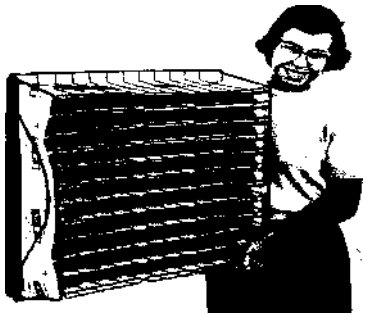

Figure 2.—Filter unit consisting of a replaceable, pleated-cotton, filter cartridge and a metal retainer.

Filtered air was provided from a fan house constructed at the head of the greenhouse. The fan was located in a pollen-tight room and air was drawn through filters fitted with a cotton medium capable of removing air contaminants below the size of sugar beet pollen (Figure 2). The filtered air was directed through an underground duct constructed of concrete pipe and parallel to the greenhouse. Junction boxes were placed at 15-foot intervals in the concrete pipe and 4-inch transite pipe was used to carry the filtered air from the junction boxes to the individual chambers. The air How was adjusted to provide a change of air at least every 2 minutes. Air escaped through flutter valves located in the outside wall of each chamber.

Thermally induced beet roots were planted in beds formed by placing soil to a depth of 1 foot inside the chamber foundations. The plants were furrow-irrigated, and the flow of water was controlled by valves located just outside the chambers. Supplementary light was furnished from a 150-watt incandescent bulb in each chamber and controlled by a time clock. Fumigants for insect control were introduced through the air outlets.

The plants grew vigorously and flowered normally in the chambers. Because some difficulty was experienced with pollen distribution, provisions for shaking the plants during pollination would be desirable. Seed yields as high as $2^{3}/_{4}$ pounds per chamber were obtained. Very little contamination occurred from pollen introduced from the outside. This was determined by planting one chamber entirely to male-sterile plants and count-

ing the seeds formed at the end of the pollinating season. Only 17 seeds were identified as having arisen from fertilization with outside pollen.

Two crops of seed were grown in each chamber in 1961. Three seed crops per year should be possible by carefully co-ordinating the supply of thermally induced roots with the dates the chambers are available for planting*

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Status of Sugar Color and Turbidity Measurements

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Introduction

Color has long been used in the sugar industry as a measure of a class of impurities. Although the word "color" in common usage connotes visual appearance, and some sugar technologists insist (9)[2] that this is what is meaningful, the ultimate interpretation of sugar color from a chemical viewpoint must be based on a measure of the amount of impurity that causes the visual appearance. Color is used both to monitor the sugar manufacturing or refining process, and to rate the final product at the consumer level. In both, the final interpretation is undoubtedly in terms of a measure of impurity. Even the housewife who looks at a yellowish trace of color interprets this (perhaps unconsciously) as a sure sign of less-than-pure sugar.

The distinction between visual appearance and the amount of impurity that causes the appearance is important because it influences the optical measurement and the method of expressing the results. The visual color is a three-dimensional entity that involves the entire visible spectrum and the response of the human eye. This measurement has had very limited acceptance by the sugar industry. The amount of impurity, on the other hand, can be related to the transmittance at one wave length and is a much less complex entity. The point of view taken in this paper is that the amount of impurities has more significance to sugar evaluation than does visual appearance. Accordingly, one object of this paper is to examine the factors whereby a measure of the amount of impurities can be gained from optical measurements on solutions.

However, since the word "color" will undoubtedly continue to be used to mean either visual appearance or amount of impurity in a very ambiguous manner, if a scale could be chosen that would be a good measure of amount of impurity and at least a fair indication of the visual appearance, then everyone would be happy, and a major source of confusion would be gone.

The sugar impurities which influence the optical measurement are of two classes, dissolved and suspended. Little is known of the composition of the suspensoids. They contain both high molecular weight organic and inorganic components, the latter being probably highly siliceous. Also, relatively little is known

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[2] Numbers in parentheses refer to littrature cited.

about the molecular composition and structure of the dissolved impurities. A large number of different colored compounds have been isolated in cane juice and raw sugar, but these account for only a small fraction of the total color.

Two fundamental optical measurements can be made in sugar solutions, absorption and scattering. Problems in interpretation arise when attempts are made to correlate these optical measurements with the non-sucrose constituents that are dissolved and suspended. The presence of strong chromophore groups in certain dissolved materials can strongly influence the absorption and large suspended or colloidally dispersed particles contribute predominately to light scattering. Between these extremes are many materials for which this interpretation is not so distinct. Nevertheless, it is useful to divide the sugar impurities into two groups: a colorant fraction that contains the summation of all constituents that contribute to absorption, and a scattering material that has the corresponding light scattering behavior. This is an obvious simplification of the actual state of affairs, but it is pursued as a working hypothesis until a better method is required. Optical measurements obtained under a specified set of conditions will be used as a measure of the colorant and scattering material. The conditions will be chosen to provide the best measure of impurities and also for convenience, speed, ease, precision, or for any other good reason that arises, such as minimization of undesirable side effects.

Optical Properties of Sugar Solutions

When a light beam is passed through a solution, the transmittance is defined as the ratio of the transmitted flux to the incident flux, corrections, if any, for reflections and cell walls having already been made. Denoting the value of transmittance for solutions and solvent by $T_{soln.}$ and $T_{solv.}$ respectively, the transmittancy, T, is $T_{soln.}/T_{solv.}$. This solvent is properly sucrose and water at the same concentration as the solution. However, pure sucrose and water are both quite transparent in the ultraviolet, blue and yellow regions (230 to 700 mμ). Water absorbs more than sucrose in the deep red (5). Therefore, for practical purposes, pure water makes a highly satisfactory reference solvent. It should be recognized, however, that the difference in refractive index between water and sugar solution will also have an important effect that will be discussed later. Transmittancy measurements constitute one class of primary data whereby the influence of the colorant and scattering material in sugar products may be studied.

In solutions containing light absorbing material only, the transmittancy is related to the cell depth, b, and to the concentration of colorant, c_i, by the familiar Lambert-Beer law,

$$-\log T_i = a_i bc_i$$

The constant of proportionality, a_i, is known as the absorbancy index (also, extinction coefficient) and this is a physical constant for a pure material. The subscript, $_i$, refers to any one of the sugar impurities that absorb. Since the concentration or even the identity of these impurities is unknown, it is common practice in the sugar industry to define an "absorbancy index" as follows:

$$\frac{-\log T}{bc_s} = \frac{\Sigma a_i c_i}{c_s} = a_s$$

where c_s is the concentration of sugar. This terminology is incorrect inasmuch as a measure of the light absorbed by one constituent (impurity) is divided by the concentration of a different constituent (sugar). However, the value of a_s is proportional to the relative concentration of the colorant impurities to the concentration of the sugar ($\Sigma a_i c_i / c_s$) and this is precisely what the sugar technologist wants to know.

About ten years ago (5) it was pointed out that the colloidal materials in commercial sugar liquors contributed significantly by scattering to the transmittancy measurement. The term "attenuation index" (a*) was proposed in order to distinguish a transmittancy measurement in which scattering was not negligible and this was expressed as:

$$\frac{-\log T}{bc} = a^*$$

The attenuation index is the sum of absorbancy index, a_s, and scattering index, s,

$$a^* = a_s + s$$

In the absence of scattering,

$$\frac{-\log T}{bc} = a^* = a_s$$

In the absence of absorption,

$$\frac{-\log T}{bc} = a^* = s$$

Implicit in this concept is the independence of absorption and scattering. The scattering index, s, is related to the more familiar turbidity, T, by the relation:

$$\frac{\tau}{2.3\ c} = s$$

Turbidity may also be evaluated as the sum of the light scattered in all directions:

$$\tau = 2\pi \int_0^{\pi} R_\Theta \sin \Theta \, d\Theta,$$

where Θ is the angle of scattering and R_Θ is the Rayleigh ratio expressed as

$$R_\Theta = \frac{i_\Theta \, r^2}{I_0 \, V}$$

where r is the distance between the small scattering volume, (V), and the observer, i_Θ is the intensity of scattered light, and I_0 is the intensity of the incident light. The angular variation in the intensity of scattered light is expressed by a scattering envelope. Rieger and Carpenter (16) showed that these envelopes for sugar liquors (see Figure 1) were dominantly forward and of a similar shape. This similarity in shape, which has been thoroughly established, permits the estimate of the entire scattering envelope from a measurement at any one angle. Thus, the total turbidity

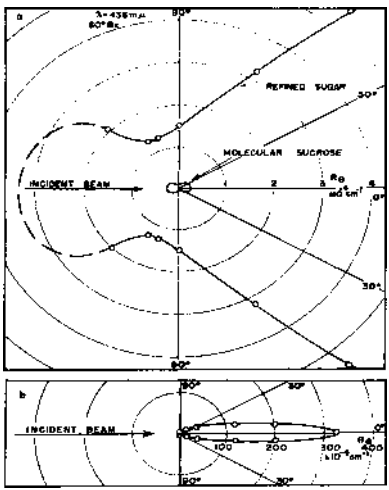

Figure 1.—Scattering envelope of a refined sugar compared with that of molecular sucrose in polar coordinates. In Figure lb the scale has been decreased 100-fold to show the complete envelope of the refined sugar.

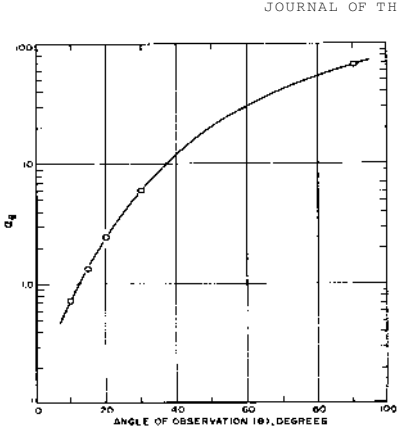

Figure 2.—Dependence of a_θ on angle of observation.

of a sugar solution can be closely estimated from a single scattering of measurement

$$\tau = a_\theta \quad R_\theta$$

Values of a_θ are shown graphically in Figure 2. This appears to be a property of sugar solutions in general and is not a function of any particular instrument or geometry used to measure the scattering. The angle of about 20° was selected as most suitable, tin which case $a_{20} = 2.45$ and

$$\tau = 2.45 \quad R_{20}$$

The turbidity can also be determined with equal facility by measuring all the light scattered within an integrating sphere (2). In either case, the scattering index is evaluated by a method that is independent of the transmission measurement, and the attenuation can be "corrected" for scattering to obtain by difference the true absorption as follows:

$$a_s = a^* - s$$

Table 1 gives some examples of this separation of "color" and "turbidity". It is seen that in commercial sugar liquors, the fraction of light scattered is seldom negligible.

Factors Influencing Optical Properties

It is of considerable interest to review some of the various factors that influence the absorbancy and scattering indices of commercial sugar solutions.

Table 1.—Separation of absorption and scattering of sucrose solutions. A = 436 mn; concentration ~ —' 35° Brix

Type	a* $\frac{-\log T}{bc}$	s $\frac{r}{2.303c}$	a (a* —s)	Percent light lost by scattering
Granulated:				
Medium	0.0315	0.0238	0.0077	75.5
Medium	.0134	.0066	.0068	49.3
Fine	.0722	.0391	.0331	54.2
Tablets	.0452	.0182	.0270	40.3
Washed	.7072	.428	.2792	60.5
Washed	.6154	.400	.2154	65.0
soft	22.66	2.16	20.5	9.53
Hawaiian raw	4.04	0.507	3.53	12.5
Cuban raw	5.56	.782	4.78	**14.1**

Dependence Upon Wave Length

The dependence of optical measurements with wave length for sugar solutions is well known. Attenuation index curves for some typical sugar products over the visible and ultraviolet spectrum are shown in Figure 3. It is noted that all sugars behave in a quite similar manner, showing no maximum. In some cases inflections are found at 280 mμ.

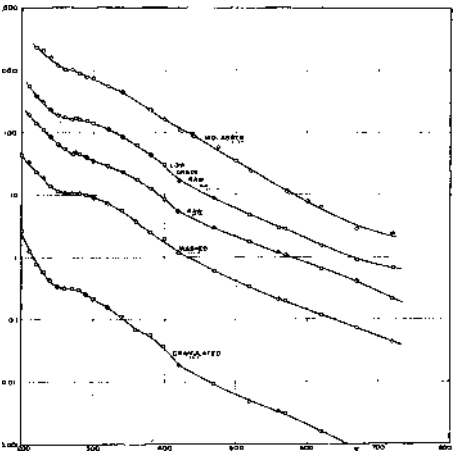

Figure 3.—Attenuation indices for typical sugars.

The dependence of scattering in sugar liquors upon wave length has not been as completely investigated as the other aspects of scattering, but it is safe to say that the effect is not nearly as steep as in the case of absorption. This fact is the basis of the so-called "subtractive" turbidity corrections which will be considered later.

Refractive Index

The effect of refractive index on turbidity is shown in Figure 4. These data were obtained by adding a constant small amount of a raw sugar solution to purified sucrose solutions of different

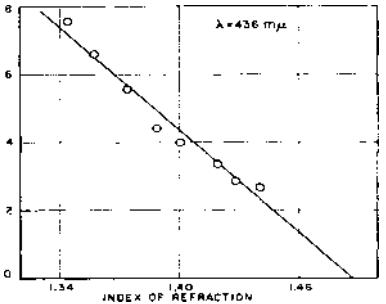

Figure 4.—Effect of refractive index on scattering at constant concentration of scattering particles.

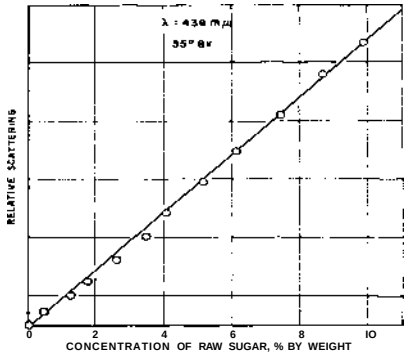

Figure 5.—Effect of concentration of scattering particles at constant refractive index.

densities. As the refractive index of the medium approaches that of the colloidal material causing the scattering, the turbidity approaches zero. For this reason, not all colloidally dispersed materials scatter light.

At constant refractive index, the turbidity increases directly with the number of scattering particles. This is shown in Figure 5 where the data were obtained by adding known small amounts of a raw sugar solution to a purified sucrose solution. When a commercial sugar is dissolved in water at various concentrations, the effect is the product of these two as shown in Figure 6. At low concentrations the turbidity increases with the concentration of sugar solids and reaches a maximum at about 35° Brix. At higher concentrations there is a decrease in the turbidity due to

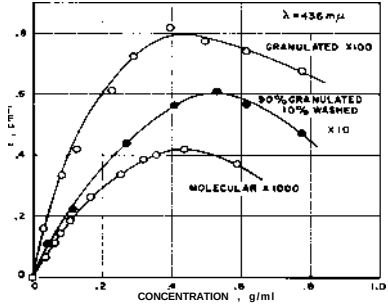

Figure 6.—Effect of sucrose concentration on turbidity.

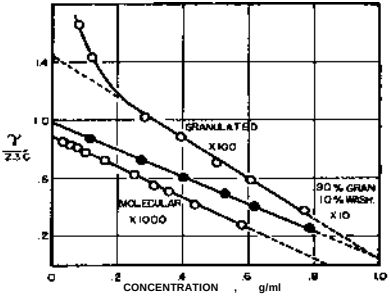

Figure 7.—Effect of sugar concentration on the scattering index.

the increase in refractive index. The same effect is observed for highly purified sucrose, granulated sugar, and for liquors in process. The turbidity may be expressed as a scattering index, which is the more pertinent method of expressing light scattering data because it is additive with absorbancy and attenuancy. The turbidity data of Figure 6 are thus expressed as scattering index in Figure 7. This decreasing behavior with increase of solids concentration is characteristic of light scattering in sugars. It can be traced to the refractive index effect. The attenuation index behaves in similar manner. The upper curve in Figure 8 is the

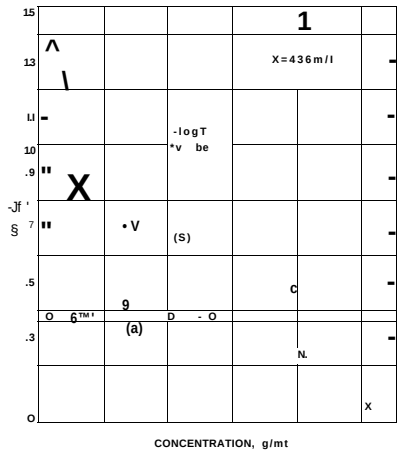

Figure 8.—Effect of concentration on attenuation, absorption, and scattering indices for a typical washed sugar.

attenuation index as determined from a transmission measurement. The middle curve is the scattering index as determined from forward scattering measurements. Both of these curves show the same sharp dependence upon concentration (refractive index). Their difference, which is the true absorption and directly correlates with colorant, is independent of concentration as it should be according to the Lambert-Beer law.

Highly turbid liquors often exhibit multiple scattering. This can lead to serious errors and there is no adequate theory to account for it. Multiple scattering can be recognized by a very high apparent turbidity. It can be avoided by dilution with

extra pure sucrose solution of the standard density. Dilution until the turbidity is less than 0.1/cm will always eliminate it.

Dependence on pH

A very important behavior of the sugar impurity is the dependence of the attenuation index upon pH. The visual appearance of many sugar solutions is strongly dependent upon the pH. The strong variation of a* at various wave lengths is shown in Figure 9 and this is characterized by a maximum change with pH in the neighborhood of pH = 7. It has been found that the scattering index is almost independent of the pH of the liquors and, hence, the entire effect noted in Figure 9 is due to changes in the absorbancy index. This may be due to changes in molecular form of the impurities. When a determination of the amount of impurity is desired, the pH must be brought always to the same level, since obviously, a change in pH does not change the total amount of impurity.

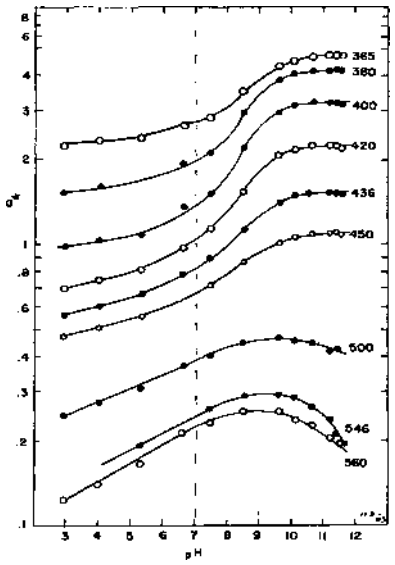

Figure 9.—Dependence of a* on pH for solution of a washed Cuban raw observed at 38° Brix over a range of wave length.

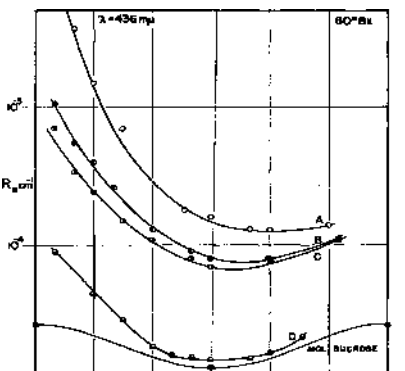

Figure 10.—Angular scattering by solutions of a granulated sugar filtered through various media: A — Coarse sintered glass; B — 0.8 μ Millipore; C — 0.45 μ Millipore; D — Powdered carbon on a 0.45 μ Millipore.

Filtration and Centrifligation

It is important to note at this point, the effect of nitration upon light scattering. Figure 10 shows the angular scattering from solutions of a granulated sugar filtered through various media. As the porosity decreased, thus removing greater fractions of the scattering particles in the filtration, the amount of scattering decreased. The scattering decreased about equally at all angles (further indication of the constancy of shape of the scattering envelope). Notice especially that even a filtration through a 0.45 μ Millipore filter left a very appreciable amount of scattering in the effluent. Such solutions are definitely not to be called "turbidity free." Finally, with the addition of an absorbent, activated carbon, the scattering was reduced almost to that predicted by theory for molecular sucrose. This indicates that part of the light scattering was caused by dissolved material that could never be removed by filtration.

Centrifuge action at high gravity fields has been found to modify the turbidity of sugar liquors. Plots of the attenuation index of the resulting liquors under different conditions are shown in Figure 11 starting in each case with the same Cuban raw. The attenuation index decreased with increase in field and after 150,000 times gravity it was considerably less dependent on

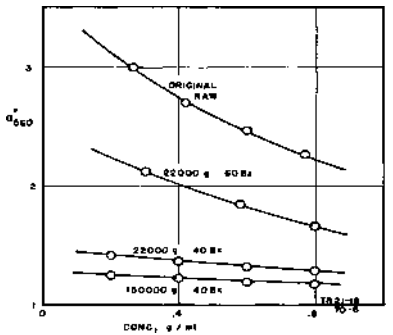

Figure 11.—Factors that influence the centrifuge action on a Cuban raw (°Brix, gravity field).

sugar solids concentration which indicates less scattering. A deposit was observed at the top, sides and bottom of the centrifuge tube. Obviously, the colloidal material in the sugar liquor is a mixture of varying degrees of buoyancy. A high speed ultra centrifuge is not a simple means of separating the soluble colorant from the material responsible for the high degree of forward scattering. The most helpful technique is to measure the turbidity.

Fluorescence

One other feature of commercial sugar solutions that must be mentioned in order to complete this discussion is the strong fluorescence in impure sugars. Since the fluorescent light is always at a longer wave length than the exciting wave length, an error *in* transmittancy or scattering measurements is introduced only when the detector responds to wave lengths longer than the exciting beam. It can be eliminated very simply by inserting a filter in front of the detector. An inexpensive color glass is generally satisfactory because the fluorescent wave length is somewhat removed from the exciting wave length. The use of fluorescence as a measure of impurities in commercial sugar liquors has not been adequately studied.

Instrument Error

Scattering also influences the measurement of sugar color in other ways. As was already mentioned, the attenuation index is the measure of the amount of light removed from the incident beam of both absorption and scattering. However, the scattered

light is mostly scattered forward through only a very small angle and thus emerges from the cell very close to the transmitted beam. If the aperture in front of the photocell is made a little large to avoid the need for careful optical alignment, as is usually the case, then part of this light that was scattered from the beam may be included in the measurement as if it were part of the beam, as illustrated in Figure 12. Thus, the instrument sees less removal of light than was actually the case and an error results in that the indicated attenuation is reported too low.

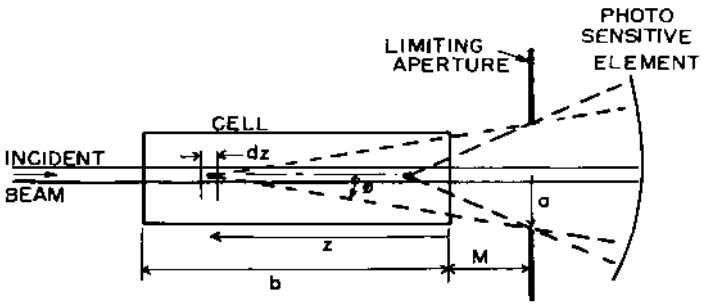

Figure 12.—Geometry of transmission measuring instruments showing how forward scattered light is measured along with transmitted beam: a = radius of limiting aperture; M = distance from exit end of cell to limiting aperture; b = length of cell; z = distance from exit end of cell to scattering particle.

It is essential, therefore, to evaluate the amount of forward-scattered light that is mistakenly measured with the transmitted beam. This error is inherent in all transmission measurements on commercial sugar liquors since these always contain some residual turbidity. It is obvious that the geometry of the instrument used for the transmission measurements is important. The detector element of different instruments can vary as to the limiting aperture with which the scattered light is received. This subject has been carefully studied and is discussed in another publication (14). Errors by as much as a factor of 2 are not uncommon, but all can be reduced by a slight instrument modification.

Choice of Conditions for Color Evaluation

From the foregoing discussion of the factors that influence the optical measurements, it should be evident that the complete optical evaluation of a sugar solution requires the independent evaluation of both attenuation and scattering. The absorbancy

index can then be determined by difference. However, the scattering should be looked upon as more than a mere correction to obtain absorption; it is by itself an additional measure of another class of impurities. Because a suitable light-scattering instrument is not yet available at a reasonable cost, most laboratories will have to rely on the attenuation measurement. It must be appreciated that this measurement always includes light scattering. The conditions of measurement which must be specified are the wave length, pH, and sugar concentration.

Choice of Wave Length

In regard to wave length, a monochromatic source in the blue, violet, or very near ultraviolet undoubtedly has real advantages. One guide in the selection of a particular wave length is the desirability that it correlate somewhat with visual appearance; thus, the measurement is made to serve a dual nature. In the charts for evaluating the E_{NBS} unit (6) of visual appearance, it is readily seen that the value obtained depends more upon the reading at 420 mμ than the one at 560 mμ. In spite of the fact that the dominant wave length for sugars is about 580 mμ, and that the greatest response of the human eye occurs at 550 mμ, the skewed attenuancy curve for sugars brings the single wave length for best correlation with visual appearance well into the blue. Any wave length in the blue appears to serve quite well, but the higher attenuancy in the extreme blue assists in the precision. This can become a factor for highly refined sugars that exhibit little color. The wave length of 420 mμ or the mercury lines at 365, 405 or 436 mμ appear as feasible choices.

The attenuancy increases with decrease in wave length according to an inverse 3rd to 8th power law. Such a steep dependence requires a close specification as to the wave length when reproducibility and precision are desired. It is not adequate to isolate the desired spectral region by means of optical filters. In addition to prism or grating spectrometers, interference filters (transmission type) may be used as a source of monochromatic light. For greater intensities the emission lines of the mercury arc are very useful as a monochromatic source.

Choice of pH

In sugar processing it is essential to keep the pH in the range of acid-base neutrality for the majority of the time. Lower values than pH 7 are avoided because of sucrose inversion and higher values are undesirable because of chemical reactions leading to alkaline degradation at the processing temperatures. However, there are serious disadvantages to the use of pH 7 in the optical evaluation of sugar liquors. It is difficult in a short time to obtain

a precise measure of the pH because of the slow approach to steady state by a pH meter. This is caused mainly by slow diffusion at the salt bridge of the reference electrode in highly viscous media. Moreover, the slope of the a* versus pH curve at 420 mμ in the neighborhood of pH —- 7.0 is so steep that an error of 0.1 in pH results in an uncertainty in a* of 5% or more. It is, therefore, highly desirable to use a value where a slight error in pH will produce a minimum change in the absorbancy or attenuancy. This occurs obviously in the neighborhood where the curves of Figure 9 have zero slope. At 560 mμ this is at pH 9 while at 420 mμ or thereabouts this occurs at pH 10 to 12 and apparently again below pH 3. A high pH gives a greater attenuancy to measure, but otherwise any fiat portion on the curves of Figure 9 is a good region to use. There may be a chemical instability of the colloidal scattering particles at extreme values of pH that would influence the scattering reading. However, this can be circumvented by making the readings promptly after the pH adjustment is made.

Choice of Concentration

The absorption index can be properly measured at any concentration, but the attenuation index, because of its dependence upon scattering (which is in turn dependent upon the refractive index) is dependent upon the concentration. Any convenient solids concentration is satisfactory, but all measurements should be made at the same concentration. Liquors of 60° Brix and above are most difficult to handle in optical cells due to the need to eliminate striations. Liquors of 50° Brix and below do not present this difficulty. The turbidity is greatest and easiest to measure at 35° Brix, but a greater dilution is to be avoided since this decreases the magnitude of the measurement. All these factors influence the choice of concentration, but once it is selected it should be rigidly adhered to for all measurements. Further dilution of very dark products such as molasses, should be made with extra pure sucrose of the selected concentration instead of water to keep the same refractive index.

A Recommended Procedure

This paper could hardly be considered complete without recommending a procedure that meets the requirements set forth. The conditions of measurement which have been found most suitable are:

Wave length	365 mμ
pH	11 ± 1
Brix	35 db 1

These conditions were chosen to define a unique attenuation index in a procedure that is quick, easy, precise, and a good

measure of the concentration of colorant. The procedure for attenuation index only is as follows:

1. Prepare a solution of the sugar to be tested at 35 ± 1° Brix by diluting with 0.1N NaOH. If the original solution is 60° Brix, dilution with an equal volume falls within the range. Other original densities require slightly different dilutions. This procedure automatically adjusts the pH to 11 ± 1.
2. Place the solution in an absorption cell whose path length was chosen to keep the measurement between 10 and 90% transmission. Further dilution, if required, must be made with a high-quality granulated sugar solution of 35° Brix. A cell depth of 5 cm is adequate for even the lightest colored sugar.
3. Measure the attenuancy at 365 mμ wave length using an instrument which was designed or modified to exclude as much as possible of the forward-scattered light from being measured along with the transmitted beam.
4. Report the results as the attenuation index: $a^*_{365\text{-}11\text{-}35} = (-\log T)/bc$

When a light-scattering instrument is available, the same conditions are used, and both attenuation and scattering are measured in the same instrument. The procedure is extremely simple once the calibration has been made. The measurement is made at a cell depth required by the instrument and always at the same concentration of 35° Brix, and the same pH of 11 ± 1. Highly turbid liquors require further dilution with a purified sucrose solution of 35° Brix in order to eliminate multiple scattering.

After the instrument is properly adjusted, the cell containing the sugar liquor is placed in the measuring compartment and two readings made, one of the transmitted light G^T and one of the scattered light G_s at a well defined angle. The scattering index is then calculated as:

The constant k is a very complex function of: (A) the concentration of the solution, (B) the refractive index of the solution, (C) the angle of the scattering observation, (D) the width of various beam-defining slits in the instrument, and (E) the optical density of a filter in the transmitted beam. Fortunately, several calibration methods are available and the value of the constant is easily determined (13, 15).

The attenuation index is determined at the same time from the transmission reading and an additional reading for water, $G_{T(water)}$. The transmittancy is then evaluated:

$$T = \frac{G_T}{G_{T(water)}}$$

The attenuation index is calculated in the usual manner and the true absorbancy obtained by difference.

Discussion: Critique of Various Methods for Measuring Sugar Color

In view of the optical properties of sugar solutions, it is of interest to examine those procedures that have already been proposed to measure sugar color in order to see how they comply with the necessary conditions.

Visual Appearance

Visual appearance methods, of which the tristimulus values, Lovibond, E_{NBS}, and the method of Brice (4) are examples, are based on a mistaken emphasis and it is the thesis of this paper that visual appearance is not the most important aspect of the problem. Visual appearance is inadequate to deal with commercial sugar solutions. Each method enumerated above was designed for non-turbid solutions and either ignores turbidity, requires a low level of turbidity, or makes a crude "correction" for turbidity. Yet turbidity is definitely a part of visual appearance. Visual appearance methods are sometimes used at a prescribed concentration and cell depth and if the pH were also specified, as is sometimes done, then these methods may produce an approximate measure of amount of impurity. The scattering has not been sufficiently considered and the effective wave length is a peculiar "average" over some of the visible region.

Visual Comparators

The visual comparison methods (8, 19) such as Stammer, Home, Scott-Klett and C & H, in which the sugar solution is compared with a standard glass or colored solution of inorganic salts, depend on an empirical standard. These should not be confused with visual appearance methods. The eye is used only to detect differences. Very precise results could be obtained, but there is considerable difficulty in reproducing any of these standards. It is almost impossible to reproduce various melts of a colored glass and it is not practical to obtain a good match in hue with actual sugars. Even the chemical solutions of inorganic salts (8, 19) have limited reproducibility and have the added inconvenience of frequent liquid manipulations in filling the

reference cell. Furthermore, a light source having a broad band of wave lengths is used and small differences in the light source, or among individual observers, can produce errors. The methods proposed in the past have not always specified a reference concentration or pH. Also, light scattering was not adequately considered. Each observer tends to "correct" a little more or a little less for the turbidity so the value actually obtained appears to be somewhere between the attenuancy and the absorbancy.

White Light Transmission

Attempts to get a better "average" concentration of impurities, or to obtain a measure of visual appearance, have prompted some workers to use white light. In any broad band colorimetry, the wave length distribution of the source, and response of the detector are all important factors. The problem of duplicating the source and the detector is a greatly added burden on the already complex problem of sugar colors. The interpretation is more difficult because of the unknown "averaging characteristics" over the spectrum range. The appropriate procedure to account for scattering in white light has not been worked out. The use of white light with filters at the detector to approximate the standard observer can be traced to the mistaken emphasis on visual appearance. Fortunately, there is now general agreement by many investigators to use a monochromatic light source.

Monochromatic Light Transmission

Transmission measurements at the wave lengths of 720, 680, 560, 545, 485, 436, 435, 420, 405, and 365 mμ have been used in various investigations. Only those in the violet (i.e. 485 to 365) meet the desirable conditions proposed above. Failure to specify pH and Brix has sometimes given rather ambiguous measurements but this could be easily corrected in the future. When pH was specified, it was often 7.0 which is not good. The procedure (9) which employs 420 mμ, pH 7, and 50° Brix comes the closest to meeting all the requirements.

Prefiltration

Several attempts (3, 21, 1) have been made to eliminate the turbidity effect by a prefiltration operation. These have not been altogether successful because of the variability in the tightness of the nitration media. However, all such efforts are of only very limited value, because they are based on the false premise that the scattering is caused entirely by suspended particulate matter. Actually, a part of the scattering is caused by dissolved material that filtration can never remove. This is another example of errors that have arisen from false concepts of the optical properties of commercial sugar solutions.

Subtractive Methods

Attempts have been made to use transmission measurements at long wave lengths as a measure of "turbidity". Examples are those of Keane and Brice (10) who used a band defined by a red filter (Corning traffic red, No. 245), and Gillett, Meads and Holven (7) who proposed the wave length of 720 mμ.

Thus, the attenuation in the red (or a constant times this value) subtracted from that in blue was considered to correct the latter reading for turbidity. This method is applicable only if the sugar solution has essentially no absorption in the red, and at the same time be so turbid as to have a large scattering in the red. This description fits only some granulated sugars. The major failing of the method occurs when attempts are made to extend it to sugar liquors containing higher levels of impurities. In general, the attenuation, absorption and scattering indices of different sugars have different wave length dependences. For instance, the wave length exponent of attenuation has been observed to range between 3 and 8. This is indeed a very large change. It is not too surprising, therefore, that these subtractive corrections for turbidity sometimes give paradoxical results, such as negative color.

Concluding Remarks

Three optical properties of sugar liquors are currently used extensively in evaluating sugar liquors: optical rotation as a measure of sucrose concentration, refractive index as a measure of total solids, and spectrophotometry absorption as a measure of a class of impurities. Scattering has too long been considered solely as an interference in the absorption measurement and only recently has it been recognized as an additional indpendent measure of impurities.

A more complete approach to the color problem has been suggested by Liggett and Deitz (12) who used the Kubelka (11) theoretical solutions relating the absorption and scattering properties of pigments. Evaluation of both the absorption coefficients and scattering coefficients of both the dissolved and suspended material, would provide a more complete understanding of the nature of the optical phenomena. This would result in four parameters, instead of the two which now make up the attenuation index. Such a complication could hardly be justified in the sugar color application at present.

An example of what might be done is shown in Figure 13. The bottom part of the figure represents a plausible arbitrary distribution of particle sizes ranging from molecular magnitudes for degradation fragments having high absorbancy to the colloidal magnitudes showing large forward scattering. The middle curves

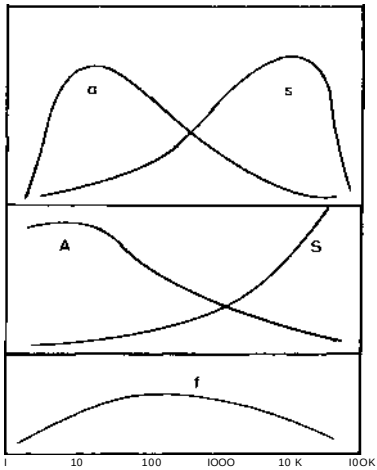

Figure 13.—Simplified dependence on particle size: f = distribution function; A = absorption coefficient; S = scattering coefficient; a = absorbancy index; s = scattering index.

of Figure 13 illustrate possible magnitudes of the absorption and scattering coefficients. The products give the indices in the top curve. The two principal maxima are the justification for the present division of the overall problem into only two parameters, namely the colorant and the scatterer. A more complete evaluation would have to consider the entire curves.

Absorbancy values at particular wave lengths have been proposed by many as a measure of single constituents or small groups of constituents, but, with one exception, the results have been very discouraging. The exception is the detection of HMF (hydroxymethylfurfural) in acid-hydrolysis products (17, 18, 20). Apparently, the attenuation index versus wave length of all the sugar impurities are so nearly the same that there is little hope of distinguishing among them optically. On the other hand, much of the work was done without a sufficient appreciation for scattering, and, if true absorption were properly evaluated, more significant progress might have been made.

An examination of the general shape of the whole spectrophotometric attenuancy curve shows that definite differences or trends can be found among commercial sugars. This observation has been expressed in various ways, one of which is by the wave length exponent (12). In general, more turbid sugars have a lower exponent than the less turbid sugars. But again, many of these observations were made without a satisfactory differentiation between absorption and scattering. Further studies in the light of present day knowledge might well prove fruitful.

Fluorescence is very weak in very highly refined sucrose and strong in raw sugars. This readily-measured optical property most certainly has promise as an additional measure of a class of impurities. Virtually no work has been done in this field with commercial sugar products and a wide open opportunity may await a careful investigator.

The effect of pH on absorbancy has possibilities as another measure of a class of impurities. Some sugars show a greater pH effect than others. A distinction could be made between pH sensitive colorant and pH insensitive colorant by measuring a* at two different pH's. However, the absolute amount cannot be determined from transmission measurements alone due to the contribution of the scattering index as illustrated in Figure 14.

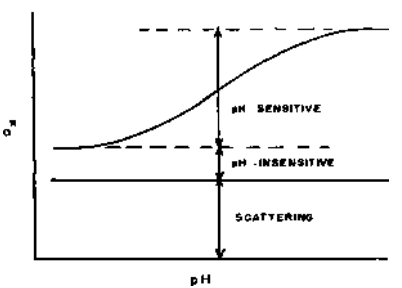

Figure 14.—Three aspects of the dependence of a* on pH at constant wave length.

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Control of Sugar Beet Nematode With 1,3-Dichloropropenes in Irrigation Water

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Introduction

Sugar beet nematode *(Heterodera schachtii Schmidt)* has been observed in all the important sugar beet producing areas of the United States and Europe (10)[2]. Because of the protective cysts that enable this pest to survive long periods of adversity, economic controls are difficult. Rotation and early planting are recommended in California as reported by Hart (4). Tliorne and Jensen (9) , Oftedal (6), Altman and Fitzgerald (1), Turner (11) and others have reported effective controls of sugar beet nematode using preplant injection fumigations with 1,3-dichloropropenes at the rate of 200-250 lb per acre made in the fall or spring before planting. This work, along with other research, resulted in recommendations as reported by Bischoff (2) in the Mountain States to combine soil fumigation with crop rotations.

Soil fumigation for sugar beet nematode control is practiced generally where there is emphasis on sugar beets as a cash crop or where the climate does not permit fall and winter plantings. In California, even with rotations and early plantings, widespread damage often occurs; these may range from nearly a complete loss to a slightly reduced yield.

With chisel injections, distribution through the soil mass depends upon gaseous diffusion. If the soil is too wet, such as after winter rains, dispersion will be limited to a few inches around the line of injection. Organic matter above 3% also may limit gaseous diffusion (3).

Experiments with 1,2-dibromo-3-chloropropane in irrigation water reported by Morton (5) and Warren (12) have given good control of root knot *(Meloidogyne* spp.), root lesion *(Pratylenchus* spp.) and other nematodes. It was decided that water applications might be more efficacious than the chisel injections in distributing toxicants to control sugar beet nematode through the soil mass, particularly in the heavier soils. This paper presents the results of experiments designed to determine the response of sugar beets to the control of sugar beet nematode with the application of 1,3-dichloropropenes as Telone® in water using ditferent

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[2] Numbers in parentheses refer to references.

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methods or irrigation. Since the high ratio of 1,3-dichloropropenes to other constituents is unique to Telone, the trade name will be used hereafter in this report.

Methods

In two of the experiments reported chisel and water applications were compared. Injections were made with conventional chisels in lines 12 in apart and 8 to 9 in deep followed by cultipacking to "seal" the surface. In one experiment, the chemical was deposited in a "sheet" 4 to 5 in deep using 12-in "duckfoot" sweeps 12 in apart modified as shown in Figures 1 and la by attaching an outlet at the lower back of the shank to direct a flat fan nozzle horizontally.

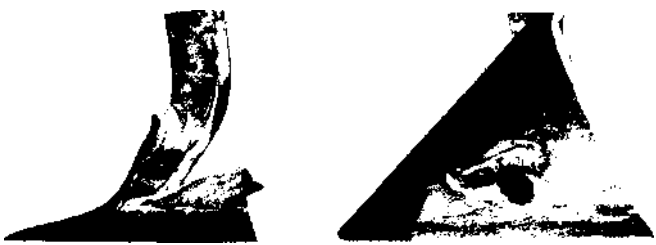

Figure 1.—Side view (left) and bottom view (right) of 12-in duckfoot sweep with flat fan nozzle attached to back of shank and directed backward under a shield over wings.

For irrigation applications, the chemical was metered into the water stream at the suction side of a centrifugal pump with a known output and dispensed into a ditch or pipe (Figure 2) to be carried to the individual plots. Initially the amount of water in acre-inches was selected to penetrate somewhat beyond the depth to which control was desired, about 24 in. A Spraying Systems Flow Regulator with an appropriate orifice was used to regulate the flow of chemical into the water stream. An emulsifier, at 5 percent of the Telone, was added to insure adequate dispersion in the first experiment. Physical data sheets (8) indicate that Telone is soluble in water at over 1000 ppm. Simple agitation tests determined that Telone at 200 ppm would dissolve readily in water if vigorously agitated. As a true solution it would not settle out. Therefore, unformulated Telone was used in the other experiments reported.

Figure 2.—Telone being added to water at suction of centrifugal pump.

Treatments were applied in October, December and May; previous results with chisel injections have shown that these differences in time of treatment should not affect the control. The sugar beets were planted by the cooperating growers from February to June and were grown under normal culture. They were harvested by weighing several 20 ft to 96 ft lengths of the center 3 to 8 rows in each plot. Samples for tare and sucrose percent were analyzed by the sugar companies concerned. Pounds of sucrose per acre were calculated from individual plot weights and sugar sample percentages.

Treatments were replicated in randomized blocks or strips. The data were analyzed using analysis of variance techniques according to Snedecor (7) and lowest significant difference values are indicated with the tables of results.

The soils in these experiments were clay-loams with 2 to 15 percent organic matter. Details are presented with each experiment.

Treatment Data arid Results

In December, 1959, Telone at 15 to 25 gallons per acre was applied as chisel and irrigation treatments to an Egbert muck soil on the Gardiner Ranch near Isleton, California with 10 to 15 percent organic matter at 6- to 17-in depths. The mineral fraction was 20 percent clay, 60 percent silt and 20 percent sand. Moisture was 15 to 19 percent, which was somewhat above the wilting point; air space was 17 percent at 7 in and 49 percent

at 17 in. Temperature at 6 in was 48°F. The plots were 20 ft wide by 100 ft long, quadruplicated. The chemical to which an emulsifier was added at 5 percent by volume was dispersed in 7 acre-inches of water by flooding for the irrigation treatments. Chisel injections were made by the Harvey Lyman Chemical Company. Spreckels variety 601 beets were planted on April 12.

Eight center rows were harvested from each plot on September 12. Differences in growth were obvious from the time of emergence through the season. The percent sucrose and tare and yields of roots and sucrose are presented in Table 1.

Table].—Comparison of chisel and basin irrigation methods of applying fumigants in organic soil to control sugar beet nematode.

Telone per acre	Application method	Percent tare	Tons beets per acre	Percent sucrose	Pounds sucrose per acre
0 gal	Basin irrigation	10.4	7.4	12.7	186?
15 gal	7 acre-in water	8.7	15.8	16.3	5160
20 gal	7 acre-in water	5.1	19.1	16.7	6392
25 gal	7 acre-in water	8.1	15.4	14.6	4512
0 gal	injection	21.9	5.9	10.4	1280
15 gal	12 in spac.	14.5	8.6	13.1	2220
20 gal	12 in spac.	11.2	10.4	13.0	2700
25 gal	12 in spac.	12.0	10.6	12.9	2736
LSD *5%*		4.2	2.9	1.6	1028
1%		5.8	4.0	2.2	1402

Note that the tare has been reduced commensurate to the nematode control. It is noteworthy that, in addition to the increase in tonnage of roots, the sucrose percentage is higher in the irrigation treatments than in the untreated or injection plots where control was poor. The return to the grower amounted to over a threefold increase in actual sucrose with 15 to 20 gallons of Telone in water per acre. The short growing period probably prevented attainment of the maximum yield that could be de rived from this treatment.

After harvest, samples of soil were collected in the irrigation treatments from 6 to 12 in and 18 to 24 in deep at 2 points per plot and potted into 1 gallon cans. Rape was planted and maintained in a greenhouse above 58°F. After about three months, the roots were examined for "pearls" with the results shown in Table 2.

The reduction in the number of cysts is correlated directly with the amount of Telone applied, but there is doubt that a second good crop of beets could have been grown without retreatment. The fact that the 25 gallons per acre treatment had the lowest cyst count, but not the best yield may indicate some

Table 2.—Populations of sugar beet nematode in soil after one sugar beet crop following treatment with telone in basin irrigation.

Telone per acre	Cysts per can[1] 6-12 in	18-24 in
0 gal	7.3	7.5
15 gal	5.5	**4.1**
20 gal	3.0	4.7
25 gal	2.5	2.8

[1] Rating scale : 0 - 10
where 10 = profuse "pearls"; 0 = none.

chemical phytotoxicity, despite the long period from December to planting in April.

In October, 1960, a strip plot experiment was established on the Hunn, Merwin & Merwin Ranch, 10 miles southwest of Clarksburg, having a Sacramento clay-loam soil with about 2 percent organic matter; moisture was in the low part of the available range; air space at 7 in was 28 percent, at 17 in 13 percent. The texture range was 49 percent clay, 45 percent silt and 6 percent sand. Duplicate strip areas 20 ft by 1250 ft were set up. The plot basins were 20 ft by 100 ft and 7 acre-inches of water was used for the flooded treatments. The chisel injections were made by the Harvey Lyman Chemical Company with straight chisels and the duckfoot sweeps. Rainbird sprinklers were set up following the sweep treatments in less than 2 hours to apply 4 acre-inches of rain at $^1/_3$ acre-inch per hour. Temperature at 6 in was 62°F. American No. 5 sugar beets were planted on February 26, 1961, and harvested September 13, 1961. Sections 26 ft long were taken from 3 rows per plot, weighed, and analyzed for sucrose by the American Crystal Sugar Company. The percent sucrose and yields of roots and sucrose are compared in Table 3.

Table 3.—Control of sugar beet nematode using chisel and basin irrigation application of Telone on clay-loam soil.

Telone per acre	Application method	Tons beets per acre	Percent sucrose	Pounds sucrose per acre
0 gal	flood	9.5	13.9	2641
10 gal	7 acre-in	**18.0**	**14.5**	5183
15 gal	7 acre-in	**15.2**	**14.3**	4223
20 gal	7 acre-in	**16.4**	**14.1**	4588
0 gal	injection	**10.4**	**14.4**	2995
15 gal	chisels	15.9	**14.3**	4539
25 gal	12 in spacing	**17.0**	**13.7**	4658
0 gal	sweeps	**12.4**	13.2	3289
15 gal	**4 acre-in rain**	**16.3**	**14.4**	4680
25 gal	**4 acre-in rain**	15.8	**14.0**	4429
LSD 5%		**2.3**	**0.64**	866
		3.1	0.85	**1158**

As in Table 1, the treated plots all produced much better yields than the untreated, but there was no difference between treatments except that Telone at 10 gallons per acre in water gave the best yields. This may indicate that this low rate in water is adequate for a good response. Prior to harvest, a pronounced reduction in watergrass *(Echinochloa crusgalli)* and pigweed *(Amaranthus* spp.) was noted where the treatments had been applied; the cleanest plots were those with Telone at 15 and 20 gallons per acre in water.

The failure to show better yields with the flooding treatments compared to the injections probably was the result of very favorable soil conditions for gaseous diffusion of the fumigants. A serious infestation of virus yellows also undoubtedly caused some reduction in yield and sucrose percentage. The treated beets at harvest time had very few visible cysts compared to large numbers in the untreated plots.

Another experiment was established on a McClusky clay-loam in the Spreckels sugar beet nematode nursery at Salinas, California to determine the efficacy of Telone in a furrow irrigation. In May, 1961, the beds were formed on 40 in centers as for normal planting. After these beds had dried out so the treated water could "sub" into them, the treatments were applied on May 23 in 4 acre-inches of water. The plots were 200 ft long and contained two full 2-row beds and a 1/2 bed border row on

Figure 3.—Growth of beets on treated versus untreated sides of a bed.

each side. Treatments were randomized in triplicated strips. The water was backed up to nearly the top of the ridge to soak the beds as rapidly as possible. Soil temperature at 6 in was 64°F. Soil moisture was 15 percent at 6 in and 19 percent at 16 in. Air space was 38 percent at 6 in and 26 percent at 16 in. Spreckels S-l beets were planted June 1 and maintained in good growing condition through the season. Growth in the treated plots was much better than in the untreated plots from emergence on through the season. Figure 3 shows a 2-row bed with the treated and untreated rows. Although the treated beets obviously were not mature, on November 15, four 20-ft sections of row from the record beds were harvested from head, center and lower ends of each plot. They were weighed and analyzed for sucrose by the Spreckels Sugar Company. The sucrose percentages and yields of roots and sucrose are compared in Table 4.

Table 4.—Control of sugar beet nematode with Telone-treated water in furrow irrigation in clay-loam soil.

Chemical	Gallons per acre	Yield tons per acre	Percent sucrose	Pounds sucrose per acre
None	0	5.6	13.4	1500
Telone	15	12.0	12.9	3222
Telone	20	13.6	12.4	3300
Telone	25	13.7	11.8	3262
LSD 5%		1.4	1.4	453
1%		1.8	1.9	606

Although the Telone-treated beets responded dramatically to the treatment throughout the season and produced over twice as much sucrose as did the untreated beets, the yields obtained were too low to be acceptable commercially. The treated beets had a lush green color at the time of harvest and obviously had not exhausted the nitrogen. The reduced sugar percentages of the treated beets is evidence that they had not reached "maturity"; it is believed that, with additional growing time, these yields and sucrose percentages would have improved markedly. There were again very few cysts in the root zone at harvest time and there was no root proliferation in the treated plots; the stand was reduced and roots were extremely distorted by nematode action in the untreated plots.

In this experiment, the weed populations in the treated plots again were considerably less than in the untreated.

Discussion

These experiments demonstrate that 1,3-dichloropropenes applied in irrigation water will give control of sugar beet nematode in certain situations where control has been difficult with

chisel injections. These are principally in the finer textured and organic soils.

Compounds dissolved in the water will move with the water over the surface and be carried into the soil a distance that depends on certain relationships between the soil fractions and the solute. Distribution over the surface can be accomplished by basin or furrow irrigation or sprinklers. The last method exposes the chemical to high losses by volatilization as the droplets fly through the air. Flooding offers the opportunity to achieve nearly 100 percent kill of the nematodes because the cysts in the top 2 to 3 in (as well as deeper) would be contacted readily by the chemical. Although the extent of control with the furrow method would be somewhat less than with the basin technique, growers may prefer the easier preparation. Local soil conditions will influence results considerably.

Although 1,3-dichloropropenes are not considered water soluble, as mentioned above, their solubility is over 2000 ppm. The amount required to control encysted sugar beet nematodes in the laboratory is *25 to 100 ppm* (3). Dilution by the soil moisture will require a somewhat higher concentration in irrigation water. There still is, however, an ample safety factor to effect solution of Telone in the applied water.

Since the Telone is heavier than water (sp gr = 1.21) and not readily water soluble, vigorous agitation is required to insure complete solution before settling out. This can be accomplished by introducing the chemical into the suction line of a centrifugal pump that provides all the irrigation water as shown in Figure 2. As an alternate method, a smaller pump can withdraw part of the water which is treated with enough material for all the flow and ejected back into the main stream. Other methods of adding Telone with sufficiently vigorous agitation to effect uniform dispersion in *all* the water also would be satisfactory.

Uniform horizontal distribution of treated water in the soil can result only from uniform dispersion on the surface. Sufficient treated water should be used to penetrate about 50 percent beyond the depth to which control is desired. This factor may vary with the amount of water in the soil and soil texture. Dilution by existing soil moisture and some adsorption of the toxicant by organic matter accounts for this requirement.

An optimum time to make the irrigation applications is in early fall in order to have the soil dried out somewhat and leave time to reshape the soil for over-wintering. There is no reason that a flooded field has to remain flat after the soil has dried out. The waiting period to plant beets after application of Telone at

20 gallons per acre would be satisfied by the time the soil dries out sufficiently to be worked.

The weed control displayed by Telone in water is probably the result of both some kill of weed seeds and the competition of better crop growth.

Experience indicates that soil moisture and air space in the heavier mineral soils are often sub-optimal. The narrow range for good gaseous dispersion of Telone between too dry and too moist is difficult to realize at practicable treatment times. Chisel injections, therefore, are undependable in these soils. They are of even less value in the organic soils (over 3 to 4 percent organic matter).

If good controls can be realized consistently with the sweep method of applying a "sheet" of chemical followed by sprinklers, this practice may prove to be more acceptable to some growers than the basin irrigation application. The optimum time to wait between injection and sprinkling will depend on the temperature, output of the sprinklers, the amount of moisture in the soil and the seal following injection. Probably 2 to 4 hours before sprinkling would be suitable.

Certain aspects of nematode control on soils with high organic matter remain to be determined. The moisture in the soil at treatment may influence results. It seems that here an appreciable amount of moisture may be desirable as contrasted to the mineral soils. The high moisture holding capacity and adsorption capacity for the 1,3-dichloropropene may prevent adequate dispersal of the treated water through sufficient soil volume if the soil is too dry before application. A grower could either pre-irrigate or await the first fall rains before treating.

Further research should be pursued to establish the value of water treatments of Telone in furrow irrigations and in higher organic soils. Also since sprinklers are used in some areas the "sweep" chisel technique should be investigated further.

Where careful control of irrigation water is possible, Telone at 15 to 20 gal. per acre in 4 to 7 acre-inches of water can be recommended using basin irrigation applications.

Acknowledgments

The assistance of the following in pursuing various facets of these experiments is gratefully acknowledged: John Bryan and George Wheatley of Spreckels Sugar Company; Norman Lawlor and Curzon Kay of American Crystal Sugar Company; Louis Kloor and A. Laiblan of Holly Sugar Corporation; other field, laboratory and managerial personnel of these companies; Harvey

Lyman Chemical Company and the growers who cooperated in these experiments.

Summary

Treatments of 1,3-dichloropropenes as Telone at 10 to 25 gallons per acre in fall basin applications oi: irrigation water were compared with chisel injections in clay-loam soils with *2* to 15 percent organic matter. One experiment included injections with a modified sweep chisel ('sheet") followed by 4 acre-inches of water through sprinklers. In another experiment, furrow applications of Telone in water were compared at dosages of 15 to 25 gallons per acre in spring. Results were as follows:

1. In the higher organic soils, the treatments with Telone in 7 acre-inches of water produced increases over chisel injections or untreated plots:
   A. Of several sucrose percentage points;
   B. In percent clean beets (reduced tare);
   C. In sucrose yields of 200 to 300 percent.
2. Soil samples from the above irrigation plots after harvest contained about 1/3 as many "pearls" in the Telone treatments as in the untreated soil.
3. In a clay-loam soil with optimum conditions for gaseous diffusion, comparisons of chisel injections, "sheet" injections followed by rain, and basin irrigation application indicated Telone at 10 to 25 gallons per acre produced similar increases with all treatments over the checks.
4. A furrow irrigation with Telone at 15 to 25 gallons per acre in 4 acre-inches of water produced excellent increases in yield.
5. Weeds always were less in the treated plots than in the checks or poor treatments—probably a combination of some seed kill by Telone and of competition from the more vigorous beets.
6. Further research on certain phases of the water application of Telone should be pursued to enable growers in all irrigated areas to take advantage of this activity.

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Notes Section

Control of yeasts in sucrose syrup by control of syrup pH.

A major problem associated with liquid sugar use has been that of contamination by yeasts. The large volume of publications in various journals on the subject of microbiology in liquid sugar attests the concern of industry concerning spoilage due to yeasts in liquid sugar and products in which liquid sugar is used.

The main difficulties have been associated with sucrose syrup and with sucrose syrup-corn syrup blends. Difficulties with yeast growth in blends have been far greater than in sucrose syrup. The problems are relatively minor in corn syrup itself and in the more dense invert syrups.

The effect of the pH value of sucrose syrup on yeast metabolism was investigated.

Changes in yeast counts, syrup pH, and taste and odor of the syrup during storage can be used as indexes of metabolism. Sucrose syrup at 7.25 pH was inoculated with 7000 spoilage yeasts per 10 grams dry substance equivalent (dse), adjusted to various pH values with HC1 or NaOH and stored for 25 days. The syrup adjusted to 4.00 pH had a fermented taste and odor at the end of the period, the pH decreased to 3.40 and yeasts were too numerous to count (TNTC) using 1 g dse on a Millipore membrane. The original syrup at 7.25 pH also became fermented, the pH decreased to (5.25 and yeasts were TUTC. Syrup adjusted to 8.15 showed no decrease in pH throughout the period, no fermentation could be detected and the yeast count had decreased to 40 per 10 g dse. Further testing using lower levels of inoculation showed that the rate of metabolism decreased with increasing pH values up to about pH 8. At pH 8 and above it was found that yeast metabolism stopped and yeasts present in the syrup died during storage. These findings suggested a simple expedient for control of yeasts in sucrose syrup through control of the pH of syrup production.

Since high quality granulated sugar dissolved in properly treated water has a very low buffering capacity, an increase in pH value of liquid sugar can readily be obtained by increasing the hydroxyl alkalinity of the water used for solution of the sugar. An increase of about 5 ppm in the hydroxyl alkalinity of the sucrose syrup has been found to increase the pH of the syrup from about 7 pH to about 8 pH. Thus the equivalent of about 10 ppm NaOH added to sucrose syrup changes the product from

one in which yeasts can grow to one in which they die. The slight increase in hydroxyl alkalinity was found to have little or no discernible effect on such quality factors as syrup color and resistance to color formation, taste, odor, etc.

The activity of yeasts in sucrose syrup-corn syrup blends can also be reduced by increasing the pH value of the blend but what may be an objectionable increase in color occurs.

Laboratory results have been confirmed on a commercial scale and a major improvement in the microbiological quality of sucrose syrup, without changes in physical or usage qualities, has been obtained by control of sucrose syrup produced at a pH value of 8. Additional processing costs are insignificant.

Patent coverage has been applied for.

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Harvesting and delivering beets 24 hours a day. Harvesting and delivering of sugar beets in the Red River Valley of Minnesota and North Dakota must normally be completed between September 20 and October 25. Earlier harvest is not practical because of beet growth and later harvest is affected materially by cold weather, snow, and freezing conditions which adversely influence the recovery and storage quality of beets.

During the harvest season of 1960 and 1961, delivery of beets at Moorhead, Crookston and East Grand Forks, Minnesota, was extended from 14 to 24 hours a day in order to complete harvest by October 25. An increase in the acreage contracted, increased yields, use of multiple-row and multiple-unit harvesters by the growers, and weather limitations established the need for such a change. Modification and improvement of pilers did not increase receiving speed sufficiently to overcome an increasing speed of delivery by growers. Similarly, an extension of receiving hours beyond a 14-hour day did not serve to reduce long truck lines ahead of pilers and idle hours of field crews waiting for trucks to return.

As a result, a second piler crew was hired, permitting two shifts of 12 hours each, starting at noon and midnight. To equalize any advantage, the shifts were changed upon 50% completion

of harvest. Grower deliveries to local stations were divided into two groups representing approximately equal acreage for each shift. Harvest was not controlled, but delivery within a shift was identified with truck windshield stickers.

Growers have enthusiastically accepted the program with many reporting a 40% reduction in harvest costs. The number of days needed for harvest has been reduced by more than one third. An average truck hauls 10 loads per shift compared with 6 loads under the 14-hour day.

Most of the rotobeating and topping are done by the growers during daylight hours; lifting and loading are generally done after dark. Lighting for night operation has been no problem. A few use special generator units but most growers use regular truck and tractor generators.

Beets delivered under this system have been well topped, clean, fresh, crisp and cool. The face of storage piles is always fresh which creates no storage problems from dehydrated, frozen or warm beets. The only trouble spots in piles were in 1960 when a 24-hour stop in delivery was allowed when the shifts alternated. This was eliminated in 1961 with an 8-hour stop.

Piler maintenance improved with the advent of scheduled 15-minute stops for greasing. Repairs that were formerly put off until night are now made immediately. Increased lighting has improved working conditions and no increase in accident rate has occurred.

The rate of night delivery is approximately the same as in daylight. The average daily delivery at four end-dump local receiving stations has increased from 1,450 truck loads under the 14-hour system to 2,340 under the 24-hour system. This is more than a 60% increase in the receiving rate. With the increased delivery rate and reduction in overtime, an approximate 20% decrease in receiving costs has been noted.

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TABLE OF CONTENTS

Ion Exclusion Purification of Sugar Juices

LLOYD NORMAN, GUY RORABAUGH, AND HAROLD KELLER[1]

Received for publication February 19, 1962

Preface

In preface to this article the authors would like to point out that ion exclusion as a commercial reality is not an accomplished fact. Both in the laboratory and the pilot plant it has been demonstrated that 50% or more of the impurities which escape carbonation can be eliminated by ion exclusion. The process as herein described is new—beset with the usual "bugs" (also mentioned) which threaten and could preclude development into commercial feasibility. Because the economic potential is so great, (as calculation of the value of recovery of 50% of the sugar lost in molasses will reveal), and because the need is so great (as the trend of sugar extraction in recent years will show), it is felt that the principles of ion exclusion and the machinery by which these principles may be applied will be of interest to all who are engaged in the production of sugar.

Introduction

Although the literature is replete with methods for purifying sugar juices, the basic, century-old system of clarification with lime (plus carbon dioxide in the case of beet juices) remains the accepted industrial procedure. This is not to say that others are inoperative, but the simplicity, the relative effectiveness, and particularly the economy of lime purification have so far withstood all efforts to replace it.

Despite its advantages, the process of carbonation has severe limitations. Such juice impurities as monovalent mineral salts and the anions of amino and certain other organic acids are present in large amounts, yet are relatively untouched by carbonation purification. Many are highly mellassigenic.

Decreased sugar recovery and increased molasses production over the past several years point to a need for better elimination of the impurity load we now process. Undoubtedly progress can and will be made on improving the quality of our beets in such areas as better varieties, better topping and storage, and more judicious use of nitrogenous fertilizers. But that is another story. As processing men, we must obtain the maximum sugar recovery from the beets as delivered. With other losses being normal, only improved impurity removal can accomplish this, and since car-

[1] Research Laboratory Manager and General Chemist-Director of Research, respectively, Holly Sugar Corporation and Assistant Research Director, Illinois Water Treatment Company.

bonation cannot presently be eliminated due to its inherent economy, supplementary purification seems to be indicated.

Shortly after World War II the new process of ion exchange seemed to be the solution to our problem. However, the process failed at that time, not because of technical inability to do the job, but because of such cost factors as rising regenerant costs, rising freight rates, cost of cooling the juice, and rising molasses value. Improvements in resins and development of new techniques make ion exchange worthy of continued consideration, although the newer process of ion exclusion reported here appears to have more promise.

Procedure

The process of ion exclusion has been known for several years, having been introduced by the Dow Chemical Company as early as 1953 (1)[2] and subject to a patent by this group (2). Early evaluation of the process, as applied to a fixed bed column, was discouraging because excessive dilution was indicated. Nevertheless, commercial utilization is being made of fixed bed ion exclusion for purification of such products as glycerine (3).

It remained for the advent of the Higgins continuous contactor (4) to bring about serious consideration of ion exclusion for purification of sugar juices. Holly Sugar Corporation in cooperation with the Illinois Water Treatment Company, and the Dow Chemical Company has conducted pilot-scale experiments with an 8-inch diameter Higgins loop.

Ion exclusion, while utilizing an ion exchange resin to effect a separation of both ionic and non-ionic materials, does not involve a true exchange reaction. A strongly acidic ion exchange resin, such as DOWEX 50W, is made by the nuclear sulfonation of styrene-divinyl benzene beads and variations can be made in these resins by changing and controlling the amount of cross-linkage in the resins. The degree of cross-linkage in a styrene-divinyl benzene bead refers to the amount of divinyl benzene it contains. A resin containing 4% divinyl benzene and 96% styrene would be said to have 4% cross-linkage. The amount of cross-linkage influences the physical-chemical properties of the resin. As the cross-linkage is increased the diffusion path becomes small enough to bar the entrance of large ions or molecules. By control of the size of these diffusion paths it then becomes possible to separate by size. If the cross-linkage of the resin is controlled to the right degree, the sugar molecule will enter the bead, but larger molecules, such as color bodies, will be excluded or screened out.

[2] Numbers in parentheses refer to references.

At the same time, ionizable compounds, such as sodium and potassium salts, amino acids, etc., are excluded because of the Donnan membrane effect. Ionic substances in equilibrium with resin will tend to have a higher concentration in solution, external to the resin bead, than that of the liquid phase inside the bead. The non-ionic substances will have the same concentration, both external and internal, or perhaps greater internal concentration due to adsorption. If an impure sugar solution is contacted with resin, such as DOWEX 50-YV, in the salt form, the sugar concentration inside will be the same as or greater than outside, but the impurity concentration (ionic impurities) will be less inside than outside. If the beads are then eluted with water, purification will have been accomplished relative to ionic materials, even though the molecular size of the ionic compounds is smaller than the non-ionic sugar.

Ion exclusion offers a way of removing ionic constituents and separating large organic molecules from sugar solutions without the use of power or chemical regenerants. The separation can be shown graphically. If an impure sugar solution is passed through a column of resin in the salt form followed by a water rinse as shown in Figure 1, it will be seen that the salt is displaced from the column first with the purified sugar solution lagging behind.

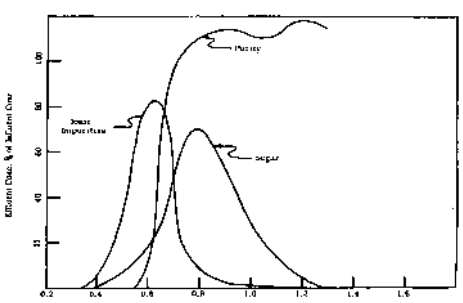

Figure 1.—Fixed bed exclusion of sugar juice.

It has been found that the sugar juices should be softened or converted to monovalent form prior to exclusion. This phase of the process has been patented by the Illinois Water Treatment Company (5). If this is not done the resin will eventually become loaded with multivalent ions such as calcium and the effectiveness

of the process will be impaired. Fixed bed columns of DOWEX 50-W in the salt form are used for juice softening.

Figure 2 is a schematic diagram of the Higgins contactor as adapted for ion exclusion purification of sugar juices. The operation of the loop is semi-continuous or cyclic in nature. At the start of each cycle the loop is filled with resin from point A on the diagram clockwise all the way around to the valve at point V_1. Water fills the remainder of the loop between V_1 and A. In order to move or pulse the resin, valves V_1 and V_3 are opened with V_2 remaining closed. Water under a pressure of about 60 psi is introduced at point B. The resin is moved clockwise for a definite, predetermined distance in slug type, positive displacement motion. The water displaced by the resin as it moves through valve V_1 is withdrawn at point C and can be re-used for subsequent

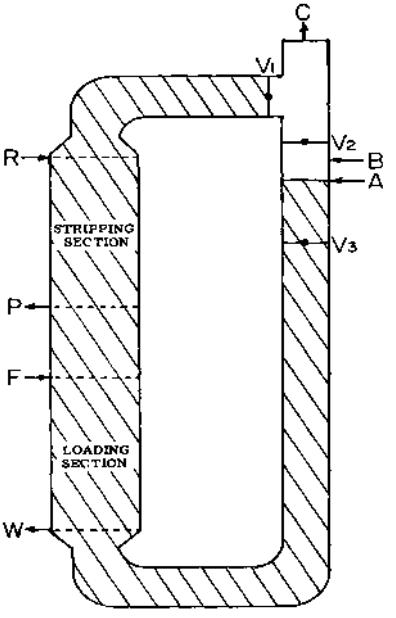

Figure 2.—Higgins continuous contactor.

pulses. When the pulse is completed, valves V_1 and V_3 are closed and V_2 opened to allow the resin to settle back to point A preparatory for the next pulse.

As soon as valves V_1 and V_3 are closed, the service cycle is begun. Feed juice is introduced at point F and rinse at point R, while product is withdrawn at point P and waste at point W.

Since a definite volume of resin is moved with each pulse, the maximum volume of feed is thereby determined because only a fixed amount of internal resin pore space will be available to accommodate the volume of feed. If more than this is fed, sugar will be lost in the waste stream. The degree of purification is also affected by the feed to resin ratio, so that an optimum must be sought with both volume throughout and purification being considered.

If the loading section between F and W is too short in length, sugar may be lost at W even though the ratio of feed to resin is satisfactory. This section length must also be a function of the flow rates involved, since the equilibrium considerations are a function of time.

Because the sugar preferentially penetrates the resin beads, it will move with the resin while the impurities move against the resin flow and separation is obtained. To recover the purified sugar solution it must be displaced from the resin interior. This, of course, is accomplished in the stripping section of the contactor by means of water introduced at point R. Only the minimum amount of water required to strip all sugar from the resin is added; any additional water will serve no purpose but will cause dilution. Adequate length in the stripping section is necessary to allow complete removal of sugar with the minimum water volume.

No transfer to or from the beads takes place in the center section between points P and F. However, as the resin is moved up, the void volume of juice between the resin beads is also moved up. Since the juice originally at point F is of feed composition it must be displaced back to point F during the service cycle or eventually the product at point P will be contaminated by juice of feed composition and purification will be impaired.

From the foregoing discussion it will be seen that the cycle of operation is divided into two parts: 1. the pulse in which the resin is moved and 2. the service cycle during which liquid flows are accomplished. Careful control of resin movement as well as liquid flows is necessary to maintain separation and throughout at optimum conditions.

The positive values of ion exclusion to the sugar processor are several. We would like to enumerate these and discuss each briefly:

1. Ion exclusion can eliminate ionic impurities not removable by carbonation. Sugar extraction can be increased. The degree of purification (about 50%) will be such that some buffer capacity in the juice will be maintained.
2. Considerable color is removed from the juice. Improved sugar color should thus be realized.
3. Because calcium is removed in the softening step, scaling of evaporator tubes should be eliminated.
4. Possible improvement in crystallization is anticipated, since many impurities of high molecular weight are eliminated.
5. The process can be operated continuously with the Higgins contactor. It can be made completely automatic with a minimum of supervision necessary.
6. Operation is at high temperature—no costly cooling and reheating required.
7. No regenerant chemicals are required. Only water is necessary to strip the sugar from the resin.
8. High throughput rates may be possible with resultant savings in equipment cost. Operation is at 40 Brix. More solids per gallon also help lower equipment size and cost.
9. Dilution is minimized. Countercurrent flows and high Brix feed keep added evaporation costs low.
10. Resin employed is most stable type known. This allows operation at high temperature and minimizes attrition losses. Mesh size of resin is 50-100. This also helps keep resin losses and make-up at reasonable levels.

Just as with demineralization by ion exchange, the process of ion exclusion will purify commercial juices. We need only develop the equipment and technique to do the job economically enough to be commercially feasible. Juice throughput must be high to keep down equipment size and capital costs. Degree of purification must be kept at a high figure to realize maximum benefits. Losses of sugar must be minimized. Dilution must be kept at low levels to prevent excessive re-evaporation costs. Water and waste quantities must be within reasonable limits to allow efficient and economic handling thereof. Obviously, simultaneous maximization of all these objectives is incompatible, so that compromise must be made to optimize operating conditions.

Our pilot plant studies have encountered the usual problems. Most of these have been mechanical in nature. In order to maintain control of physical conditions inside the contactor loop all flows must be precisely controlled from cycle to cycle. Resin flow in particular has been difficult to control. Pressure drop across valves and past internal obstructions such as distributors has been a major factor in erratic resin movement. Resin expansion and contraction due to temperature variations and juice flow changes is thought to be another factor. Wall effects of the loop itself may be still another.

Control of all fluid streams into and out of the loop is extremely important, not only from the effect on steady state conditions of the exclusion phenomenon, but also because of the effect on resin flow just mentioned. The solution of flow control in our pilot contactor has not yet been found, though we feel that progress is being made and that the answer will be found.

At the present state of development many problems still remain to be solved before ion exclusion purification can become a commercial reality. We feel that the following operating conditions must be met:

Flow Rate—3 gpm/ft^2 of contactor cross section (with 40 Brix juice)

Separation—50% removal of impurities

Dilution—10% or less

Sugar losses—undetermined, but as low as possible

Water requirements—no more than 300% by volume on juice flow

Waste—roughly equivalent in volume to water requirements

Some of these conditions have been achieved in our pilot plant operations; some have not, but our experience leads us to believe that these objectives can and will be realized.

We have focused our attention on application of this new process to factory thick juice. This is the logical point of attack if full benefits are to be realized throughout the entire sugar end of the factory. However, the dictates of optimum economic return require the examination of other possibilities. Processing of high greens or machine syrups—or even molasses—may in some circumstances prove more economical, depending on such factors as pan capacities, purity considerations, and equipment costs. For instance the quantity of machine syrup for any given factory would be much less than thick juice. If an equivalent amount of impurities can be eliminated at this point, the low raw load and molasses production could be equally reduced with extraction

correspondingly increased. Equipment cost would be less, though facilities would be required to handle the recycle juice at the purity and Brix attainable.

Also, we have mentioned little about ion retardation, a process similar to exclusion in which the ionic impurities would travel with the resin movement while sugar would travel counter to resin flow. An advantage of increased throughput may be gained by using retardation. Exclusion is favored at this time because a special, more costly resin is required for retardation, a resin which may not have the stability of the strong acid cation used in exclusion. Furthermore, little or no color removal is expected with retardation, a benefit of exclusion which is difficult to evaluate economically, but which will undoubtedly be of much value in some areas.

We have briefly described the process of continuous ion exclusion as it might be applied to the sugar industry. It will be supplementary to, but will not eliminate carbonation. Hopefully, it can increase sugar extraction and quality by eliminating melassigenic impurities and color from juice, while requiring no regeneration reagents other than water. Necessary equipment is complex, but automatic in operation and relatively high in potential throughput, with reasonable labor and capital costs being indicated. The need for a process which will economically allow increased sugar extraction from juices continuing to deteriorate in quality from year to vear is clearly indicated. Ion exclusion promises to fill this need.

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Some Physico-Chemical Factors of the Fruit Influencing Speed of Germination of Sugar Beet Seed[1]

F. W. SNYDER[2]

Received for publication April g, 1962

Speed of germination of the sugar beet seed is controlled largely by the physico-chemical characteristics of fruit tissue which surrounds the true seed (2)[3]. In the commercial varieties examined, chemical composition of the fruit seemed to play the major role in regulating the rate of germination (2). Sedlmayr (1) has demonstrated that seeds harvested from different plants of a sugar beet variety may not germinate at the same rate and that this germination characteristic is heritable.

Tests have been conducted to determine some factors which control speed of germination of open-pollinated seed from individual plants. This paper describes techniques which indicate the potential rate of germination of seed samples as well as the correlations between the techniques and actual germination.

Methods and Materials

Ripe seeds were harvested from 65 plants of five progeny groups of US 401. Samples were harvested as they matured over 54 days, but the majority were collected between August 9 and September 6, 1957. The seedballs on a given plant were harvested when at least 80 percent were dry and straw-colored. Normally three weeks were required for maturing, and since only traces of precipitation were recorded between July 23 and August 23, seedballs from some plants were not exposed to rain before harvest. Although the seedballs on plants within a progeny group tended to mature about the same time, some were exposed to more rain than others.

The speed of germination for each seed sample was determined by two methods: 1—The *liquid-contact method* (2) involved germinating the seeds while the seedballs were in contact with a mineral nutrient solution of 10.1 atmospheres osmotic pressure. Eighty seedballs (each considered as a single unit and appearance of first seedling foot indicating germination) were used for each sample. Percentage of germination were recorded for 2, 3, and

[1] Cooperative investigations of the Crops Research Division, Agricultural Research Service, U. S. Department of Agriculture, and the Michigan Agricultural Experiment Station. Approved for publication as Journal Article #2620, Michigan Agricultural Experiment Station.

[2] Plant Physiologist, Crops Research Division, Agricultural Researcn Service, U. S. Department of Agriculture, East Lansing, Michigan.

[3] Numbers in parentheses refer to literature cited.

5 days; 2—In the *blotter method,* 40 seedballs were placed on a blotter (4 1/2 X 4 3/4 inches) moistened with tap water.

Speed of germination data (Table 1) are coded for simplicity of comparison and designated as a speed-of-germination index; the first digit representing the 2-day germination, the second the 3-day, and the third the 5-day germination. The coding is as follows: 1 represents 0 to 19 percent germination; 2, 20 to 29; 3, 30 to 39; 4, 40 to 49; 5, 50 to 59; 6, 60 to 69; 7, 70 to 79; 8, 80 to 89; and 9, 90 to 100. For example, for clone 318 the speed-of-germination index for the liquid-contact method is 136. This means that the 2-day germination is between 0 and 19 percent, since the first digit is 1. The second digit, "3", represents a percentage between 30 and 39 for the 3-day germination and the digit "6" a germination between 60 and 70 percent for the 5-day value. Thus, a speed-of-germination index of 999 represents a very rapid and complete germination and an index of 111 a very slow and incomplete germination as of the fifth day.

In both the liquid-contact and the blotter methods of germination, the speed of germination represents the integrated physical and chemical effect of the fruit on the germinating seed, as well as the physical and chemical attributes of the seed itself. An additional test was used to separate the effect of the chemical factors from that of the physical factors in the fruit. The germination and growth of wheat on water extracts of seedballs were selected to indicate differences in their chemical composition. The procedure for the *wheat test* was as follows: Air-dried seedballs (1 gram for 10 milliliters of distilled water) were soaked for 18 hours in the refrigerator. The extract was decanted. Four milliliters of extract were added to 25 kernels of wheat (Genesee variety, certified) placed on a filter paper in a Petri dish. After 96 hours the fresh weight of the wheat grown on the extract was compared with that grown on distilled water. The average of four replications was then expressed as a percentage of the fresh weight on distilled water. The average weight of the 25 dry wheat kernels was subtracted from the total fresh weight of wheat seedlings to obtain a more precise value for water absorption during the growing period.

The specific conductance of the seedball extract, which is a measure of the quantity of electrolytes in solution, was determined for each sample by diluting 10 milliliters of the extract to 90. The pH also was recorded. Since speed of germination is known to be affected by osmotic stress, the relations between specific conductance of the seedball extract and the other tests were established.

Table 1.—Comparison of germination and certain seedball characteristics of 65 seed samples of sugar beet variety US 401.

Progeny and plant numbers	Speed-of-germination index[1]		Specific conductance (mhos x 10—3)	Wheat test on seedball extract (% of growth on water)	Seedballs shedding seedcaps (percent)	Harvest date (days after Aug. 8, 1957)
	Liquid-contact	Blotter				
986:						
318	136	799	22	94	0	22
256	489	699	33	95	3	21
266	179	699	26	106	2	28
243	138	499	34	76	2	21
288	134	478	41	57	1	27
305	899	189	48	39	1	8
256	157	169	44	69	0	13
257	124	155	54	45	0	32
295	488	149	14	95	8	27
312	117	114	52	41	2	11
311	785	113	52	53	36	54
998:						
469	788	999	25	102	23	27
514	999	899	28	94	25	27
528	999	899	29	94	11	28
524	899	799	36	100	42	29
472	899	689	31	83	20	22
413	999	489	17	106	30	28
426	777	479	52	88	8	29
493	999	357	24	98	9	22
525	677	268	49	89	4	29
477	999	159	40	103	44	29
433	167	146	19	103	0	29
451	134	134	69	31	2	54
519	499	128	29	103	21	29
517	999	118	79	40	7	13
457	459	117	60	50	4	13
141:						
581	178	999	21	96	0	22
547	999	799	37	95	74	27
551	799	799	21	100	8	27
521	899	799	30	92	39	22
553	899	699	28	92	62	27
550	677	599	37	89	32	27
565	179	579	28	97	7	29
596	688	489	40	67	2	27
560	168	479	44	62	1	27
584	999	479	25	92	42	22
568	899	478	38	99	48	29
617	289	269	22	95	16	29
577	167	258	29	97	0	22
549	399	129	56	36	22	13
907:						
226	999	579	23	92	23	28
204	999	358	42	89	58	27
144	599	199	40	43	0	1
188	189	159	61	38	0	1

[1] Index for 2-, 3-, and 5-day germination values. See details for methods of coding in the text.

(Continued on next page)

Table 1. (continued)

Table 1.—Comparison of germination and certain seedball characteristics of 65 seed samples of sugar beet variety US 401.

Progeny and plant numbers	Speed-of-germination Index[1]		Specific conductance (Mhos x 10^{-3})	Wheat test on seedball extract (% of growth on water)	Seedballs shedding seedcaps (percent)	Harvest date (days after Aug. 8, 1957)
	Liquid-contact	Blotter				
197	188	159	65	41	0	11
136	156	139	43	50	2	21
140	179	139	51	56	1	8
172	244	197	46	63	0	11
128	114	114	51	35	0	11
211	134	114	23	99	5	21
194	135	112	61	30	0	1
615:						
368	134	899	14	106	3	27
401	899	899	14	103	2	28
342	577	689	22	104	9	27
372	999	378	29	103	52	27
358	255	357	25	98	0	28
379	999	159	40	103	4	27
403	157	148	26	75	0	22
361	146	138	48	62	0	11
360	111	128	55	51	0	11
353	157	127	37	80	1	22
362	358	116	59	37	4	11
383	134	115	50	58	0	13
015	138	113	52	60	0	1
393	389	113	55	46	0	11

[1] Index for 2-, 3-, and 5-day germination values. See details for methods of coding in the text.

Table 2.—Correlation coefficients between speed of germination and certain other seedball attributes of 65 selected samples of US 401.

	Specific conductance	% shed seedcaps	Liquid-contact - 5 day	Liquid-contact - 3 day	Liquid-contact - 2 day	Blotter - 5 day	Blotter - 3 day	Blotter - 2 day
Wheat test	—0.85**	0.40**	0.28*	0.42**	0.41**	0.43**	0.59**	0.73**
Blotter - 2 day	—0.71**	0.39**	0.27*	0.45**	0.43**	0.67**	0.90**	
Blotter - 3 day	—0.60**	0.29*	0.27*	0.46**	0.38**	0.85**		
Blotter - 5 day	—0.45**	0.27*	0.46**	0.57**	0.39**			
Liquid-contact - 2 day	—0.21	0.66**	0.67**	0.83**				
Liquid-contact - 3 day	—0.28*	0.56**	0.88**					
Liquid-contact - 5 day	—0.18	0.45**						
% shed seedcaps	—0.21							
Date harvest	—0.37**							

* Indicates r-value greater than that required for significance (0.25) at the 5% level.

** Indicates rvalue greater than that required for significance (0.32) at the 1% level.

Some measure of the physical attributes of the seedball may be derived from the tightness of the seedcaps or lids covering the ovarian cavities. Thus, the percentage of seedballs having lost or shed seedcaps was determined by examination of 200 seedballs per sample.

The six values which characterized each of the 65 seed samples (Table 1), were employed in calculating the coefficients of correlation in Table 2. Actual percentages of germination were employed in the calculations.

Discussion of Results

All correlations were significant, except three involving specific conductance. Specific conductance was correlated inversely with the other attributes.

A coefficient of determination[4], which indicates the per cent of the variation in two variables that is concomitant or simultaneous, may be calculated by squaring the coefficient of correlation and multiplying the result by 100. Expressed in this manner, many of the relations appear less significant. Only seven of the 37 coefficients of determination equal or exceed 50 percent. As might be expected for either the liquid-contact or the blotter methods of germination, the coefficients of determination for the 2- versus 3-day and the 3- versus 5-day comparisons were between 69 and 80. However, coefficients for the 2- versus 5-day comparisons were less than 50. The coefficient of determination for 2-day blotter germination versus specific conductance was 50 and versus the wheat test 54, while the coefficient for specific conductance versus the wheat test was 72. Thus, the blotter method of germination, the specific conductance, and the wheat test appeared to measure the same variables in approximately the same way.

Since the three tests appear to give the same general information, a choice of tests would be permitted. In contrast, the liquid-contact method of germination apparently measured a different set of factors which contributed to the speed of germination. Of the attributes examined, speed of germination by the liquid-contact method correlated best with the percentage of seedballs shedding seedcaps. The relative tightness with which the seedcaps are attached may affect speed of germination by physically restricting the flow of water and oxygen to the seed.

The electrolytes in the seedball, as measured by specific conductance, correlated significantly (—0.71 for 2-day) with speed of germination by the blotter method. The differential response

[4] Koch, E. J. Presentation of Experimental Results. Symposium sponsored by Amer. Soc. Hort. Sci. anl Biometric Soc. at Pennsylvania State Univ. August, 1959.

may be the result of the osmotic stress imposed by the solution employed in the liquid-contact method which may mask the influence ot electrolytes in the seedball.

The pH values for the seedball extracts ranged from 6.2 to 7.3. These deviations from neutrality appeared to have no significant effect on the germination response.

Although no data are presently available to relate the quantity of organic inhibitors to the specific conductance values, they may be closely related, since the speed of germination does not appear to be controlled solely by the quantity of electrolytes in the seedball. Exposing ripe or nearly ripe seedballs to rain leaches soluble organic substances as well as inorganic electrolytes from them. The significant correlation between date of harvest and specific conductance probably reflects the leaching effect of rain on the seedballs harvested later in the season (Table 3).

Table 3.—Relation of precipitation in 3-week period before harvesting of seed and the specific conductance of the seedball extract.

Day of Harvest period	Number of samples	Rainfall in 3-week period before harvest Inches	Average specific conductance Mhos x 10^{-6}
1st (August 9)	4	1.35*	54
8th to 13th	15	Trace	55
21st	4	0.90	38
22nd	9	1.07	27
27th to 32nd	31	1.47	34
54th	2	0.75	61

* Precipitation occurred on the 19th day before harvest.

Summary

Seed harvested from 65 US 401 sugar beet plants, and which had been selected for a range of germination characteristics, was germinated by the liquid-contact and blotter methods. The specific conductance and pH of the seedball extract for each sample were measured. Wheat was grown on a portion of the extract for 96 hours and its fresh weight was expressed in terms of growth of wheat on distilled water. The percentage of seedballs shedding seedcaps was determined for each sample. Coefficients of correlation between the various tests were calculated.

Most of the tests were significantly correlated. All tests, except specific conductance, were positively correlated. Although the liquid-contact and blotter methods of germination were significantly correlated, the coefficient of determination indicated that they do not measure precisely the same attributes. On the basis of simplicity and amount of information derived from a test, the

blotter method of germination and the specific conductance of the seedball extract are suggested as the most useful tests to evaluate the speed of germination.

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(1) SEDLMAYR, T. E. 1900. Genetic studies on speed of germination in sugar beets *(Beta vulgaris* L.). Doctoral Dissertation, Department of Farm Crops, Michigan State University, East Lansing, Michigan.

(2) SNYDER, F. W. 1959. Influence of the seedball on speed of germination of sugar beet seeds. Amer. Soc. Sugar Beet Technol. J. 10(6): 513-520.

Effect of Solids Recirculation On Purification of Raw Juices

F. GALE[1]

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Introduction

Purification of raw juice extracted from beet cossettes is universally performed by means of lime and carbonic acid treatment in various steps. A large variety of processes are employed, all using the same chemicals for eliminating as many impurities as possible.

These notes deal with a particular aspect of the juice purification process, namely, with the recycle of calcium carbonate particles and the various effects obtained. It is beyond the limits of this paper to examine solids recycle from a strictly chemical point of view, although a thorough study in this direction is highly recommended. Existing trends in Europe, as far as solids recycle is concerned, are briefly reviewed with special regard to those with which the writer has had direct experience or has been able to obtain firsthand information. Recycle of clean solids, as successfully practiced in some Italian factories, is briefly described and some qualitative results are given.

Historical Background

The Dorr System of continuous first and second cardonation includes, in the broad use of the term, the steps of liming, gassing, mud thickening and filtering prior to evaporation. This system is the heart of all modern juice purification processes, all of which include some recirculation of carbonated juice within the saturation step. This recirculation is necessary in order to facilitate filtering of the carbonated juice and sweetening off of the cake on continuous rotary filters. Batch carbonation, it is well known, produces saturated juices that are very difficult to thicken and/or filter with any type of equipment. A typical flowsheet of the Dorr Continuous Carbonation System is shown in Figure 1.

The basic Dorr Carbonation System, first practiced commercially about 1928, has now undergone numerous modifications. One such modification is shown in Figure 2. In this system, in order to adapt it to a particular purification need, continuous preliming and separate main liming were included. This system is often practiced in some European countries. Other modifications of the basic system are being used as will be covered later.

[1] Senior staff engineer, Dorr-Oliver S.p.A. Milan, Italy; Technical Adviser, Dorr-Oliver Companies of Belgium, France, Germany, Italy.

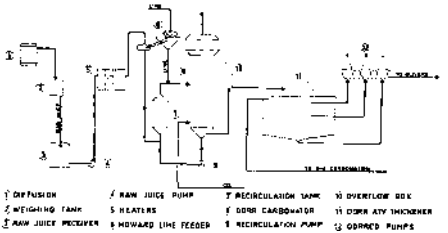

Figure 1.—A typical flowsheet of the Dorr Continuous Carbonation System.

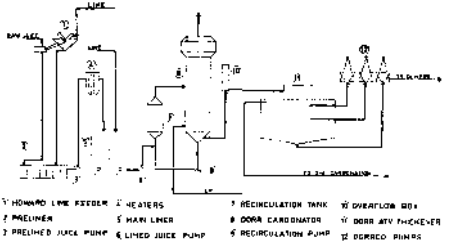

Figure 2.—A modified Dorr Continuous Carbonation System incorporating continuous preliming and separate main liming.

If one studies the various purification systems as described in patents and technical publications, it is quickly discovered that they are usually a compromise between two distinct and contrasting requirements: 1. Highest elimination of impurities and; 2. best possible filtration and sweetening off of calcium carbonate muds.

It is interesting to note that in order to cope with both requirements, some solids recirculation is considered necessary in all of the flowsheets. This very simple consideration led the writer to the investigation of a modification of the basic system utilizing recycle of clean calcium carbonate particles.

Early DorrClone Tests in Europe On First and Second Carbonation Juices

Porcelain DorrClones[2] of 50- and 100- mm diameter are widely used in European factories for degritting milk of lime. Because they make separations on grit in the range 20 to 30 microns, they are much more effective than conventional machines such as rotary and vibrating screens.

[2] Trademark for hydrocyclone manufactured by Dorr-Oliver Companies.

This use of DorrClones led to consideration of the use of smaller diameter DorrClones (10 and 15 mm) for clarification of carbonation juices. Several flowsheets have been considered (Figures 3 and 4) and tests have been conducted in Germany and Italy.

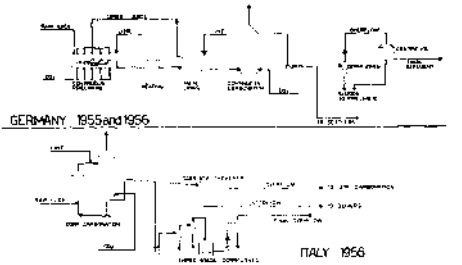

Figure 3.—Experimental flowsheets incorporating Dorr-Clone clarification in the first carbonation step.

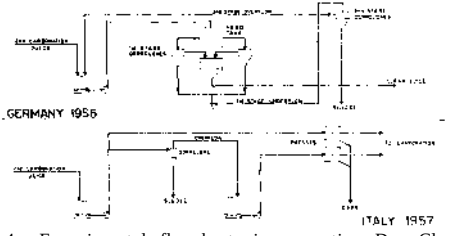

Figure 4.—Experimental flowsheets incorporating Dorr-Clone clarification in second carbonation step.

On first carbonation juice it was found relatively easy to obtain high solids removals at high concentrations but overflow clarities comparable to thickener overflows were never achieved. The cloudiness of the DorrClone overflows was mainly due to colloidal particles. Attempts to polish DorrClone overflow with disc: centrifuges were not successful because of the inability of the centrifuge to make adequate separations at reasonable capacities With polishing filters, low filtration rates and cloth blinding were encountered.

Better results were obtained when DorrClones were tested on second carbonation juices. Although the clarification was not

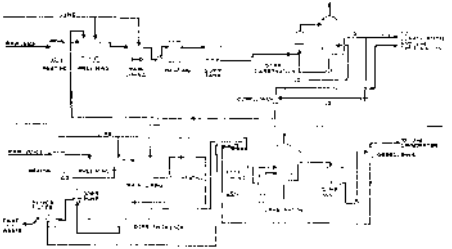

Figure 5.—First carbonation flowsheets currently in use in Europe incorporating DorrClones to recycle selective fractions of the carbonate mud.

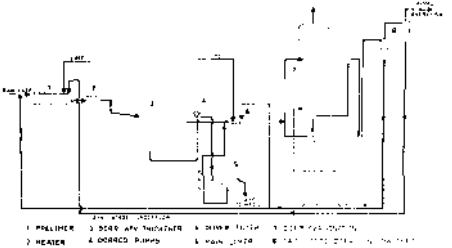

Figure 6.—Experimental flowsheet incorporating two-stage DorrClone separation of first carbonation juice and recirculation of solids to raw juice and prelimer.

complete, the length of the cycles on the second carbonation pressure filters was increased. Results from the Italian experiences indicate that the capacity of the plate-and-frame filters increased from two to five times when they were fed with the Dorr-Clone overflow. DorrCIone underflow is sent back to the raw juice tank.

Although DorrCloncs were not satisfactory for clarification, they do perform a useful function when used in various ways within the carbonation process. They enable selective fractions of the carbonate mud to be recycled so as to improve mud settling and filtering characteristics. For example, Figure 5 shows some flowsheets which are being used in Europe and other flowsheets to be shown later also embody this use of DorrClones.

Research Work In Belgium On Recirculation of Solids

An interesting flowsheet (Figure 6) has been tested in Belgium, at the suggestion of A. Schaus[3]. The objectives are to

[3] Chief Chemist Dorr-Oliver S.A., Brussels.

reduce lime consumption and simplify the separation of solids from the first carbonation step. In this process a horizontal prelimer of the multicompartment type and a prelime thickener were installed ahead of the carbonation step. All of the first carbonation juice was passed through a two-stage DorrClone battery. The final overflow was sent to second carbonation while the two underflows from the DorrClones were recycled into the preliming step as shown in the flowsheet. The coarser particles were added to the raw juice as it entered the prelimer while the fines, contained in the second underflow, were recycled to the last compartment of the prelimer.

After heating, the prelimed juice was clarified in a Dorr Thickener. This overflow was then heated, limed, and carbonated. The underflow of the prelime thickener was sweetened off on an Oliver Filter and finally the cake was discarded from the system. According to the theory behind this, carbonation conducted in the presence of as much carbonate as possible increased the particle size and facilitated the separation of solids. Particles over 40 microns in size have been observed and measured microscopically. This thinking was supported by several tests on first carbonation juice which showed a 3- to 5-fold increase of the filtration rate when DorrClone underflows were recycled.

This flowsheet, which gave encouraging results when processing high purity juices, encountered some difficulty when the amount of impurities to be removed was relatively high. Tests in Italy have indicated that while processing juices of, say, 82 to 84 purity the overflow from the DorrClone was still cloudy, even when a 3-stage arrangement was used. It was found that about 80% of this turbidity was due to organic impurities in colloidal form. These organics could not be removed by any simple polishing filtration and would prove detrimental if conveyed to the second carbonation vessel.

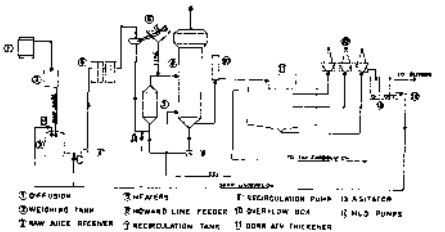

Figure 7.—First carbonation flowsheet, widely used in England and Italy, incorporating recycle of Dorr Thickener underflow.

Recirculation of Thickener Underflow Ahead of Purification Station as Practiced in Several European Factories

Recycle of Dorr Thickener underflow has been used for years in Europe, principally in England and Italy where it has become a standard practice in most plants. Although results obtained in various factories are not strictly comparable, it has been established that recycling of Dorr Thickener underflow is effective in reducing the color formation within the thickener and in improving settling and filtration.

A typical flowsheet utilizing this recycle is shown in Figure 7. Tests have been made in which the Dorr Thickener underflow was returned to three different points ahead of the carbonator. These were:

Point A—into raw juice pipe just before it entered the recirculation tube.

Point B—into the raw juice tank where the diffusion juice flowed from the weighing tanks.

Point C—into the suction line of the supply pump.

Recycling to Point A did not show very good results except for color reduction, whereas other positive advantages were obtained when mud was recycled in Points B and C, although no significant differences were noted between these two.

The advantages obtained may be summarized as follows: 1. Color reduction of thickener overflow ranging from 25% to 30%; 2. improved settling with higher settling rates and smaller mud volumes thus reducing the actual unit area requirement to 3.9 sq ft/short ton solids/day which is equivalent to 0.24 sq ft/short ton beets /day under the conditions of the plant where the tests were made in Italy and; 3. improved filtration due to porous filter cake. The solids handling capacity of the Oliver Filters was increased by 1/3 as shown in the following figures developed at the Italian installation.

Capacity	Without Mud Recycling	With Mud Recycling
Sq ft/short ton beets/day	0.171	0.113
Sq ft/short ton solids/day	4.40	1.88

Although the overflow turbidity increased with mud recycling from about 50 to 150 ppm the polishing filtration ahead of second carbonation was greatly enhanced. Filtration cycles of 75 to 80 hours were experienced with porous ceramic candle filters while, without mud recycling, the cycles were not more than 10 to 20 hours.

Another minor but consistent advantage was the reduction of foam in the raw juice tank where the Dorr underflow was continuously added.

Certainly, when filtration on rotary drum filters is a serious problem the recycle of Dorr underflow is a simple and inexpensive solution without increasing the total lime consumption. In some cases an increase in lime salts has been experienced compared to the conventional Dorr-Oliver scheme, especially when the first carbonation (phenolpthalein) alkalinity is relatively high (over 0.10 grams CaO/100 cubic centimeters).

A curious phenomenon which has been experienced is the poisoning of the calcium carbonate being recycled after a time ranging from 1 to 3 weeks. The color reduction decreases progressively and the slurry becomes darker and darker. The recirculation of Dorr underflow must then be interrupted for two or three shifts in order to purge the system completely.

At this point a question might be raised as to how much Dorr underflow should be recycled in order to obtain the best results. Any figure concerning the volumetric percentage of the recycled underflow would be misleading if not supported by other data, such as sludge and juice density. As it is rather difficult to measure and control continuously the volume or weight of Dorr underflow in commercial installations, it seems advisable to express the recycle in terms of equivalent grams of CaO recycle into raw juice before any further treatment. Our experience in Europe indicates that the optimum is in the range of 0.7 to 1.0 grams of CaO per 100 cubic centimeters of juice, depending on local conditions. Higher alkalinities in first carbonation, thicker raw juices, and lower purity juices demand a higher percentage. A maximum value of 1.2% CaO has been found necessary in a plant where extremely rich beets are processed (percent sugar in the cossettes over 22%).

All the above considerations apply also when disc type pressure filters are used as intermittent thickeners.

Pilot Plant Work in the United States With Preliming and Solids Recirculation

A few years ago, extensive test work was carried out in a pilot plant erected by Dorr-Oliver at Betteravia, California, with the cooperation of the Union Sugar Division.

The beet juice purification process tested in California is shown in Figure 8. The following processing steps were used: 1. Stabilization of the slightly acid raw juices by massive recirculation of carbonation thickener underflow; 2. progressive preliming with milk of lime and partial recirculation of prelime juice; 3. separation of coagulated impurities (nonsugars) by sedimentation; 4. addition of carbnate solids to the clear juice; 5. mainliming, carbonation, and thickening and; 6. sweetening off of carbonate cake.

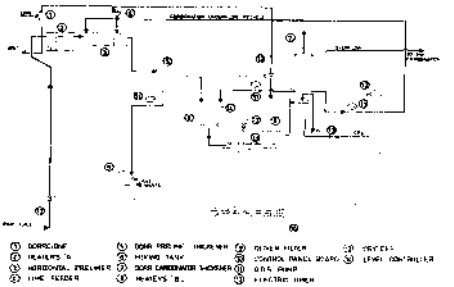

Figure 8.—Beet juice purification pilot plant used in cooperative test program at the Union Sugar Company's factory at Betteravia, California.

According to several authors (Dedek, Vasatko, Teatini, Brieghel-Miiller, Salani and others) an improved purification can be accomplished with progressive preliming because the impurities (nonsugars) present in the raw juice will coagulate, flocculate, and segregate at different pH values. It would seem desirable to remove these segregated impurities as soon as they precipitate and before any further treatment. In this experimental process, therefore, a prelime thickener was used for separating the coagulated impurities. A massive recirculation of carbonation sludge was needed for this purpose. Otherwise these colloidal solids would not settle satisfactorily. The prelime thickener overflow was then treated according to standard practice,, i.e., it was limed and saturated with CO_2. In order to obtain good settling rates in the carbonation thickener, carbonation sludge was added before main-liming and carbonation to maintain a controlled concentration. This recirculation promoted growth of large crystals.

In normal operation, Heater A (Figure 8) was not used. Juice coming from the diffuser was fed into the first compartment of the Brieghel-Miiller prelimer at about 55-60°C. Milk of lime was added in the next-to-last compartment at about 0.25-0.35% CaO on juice. A large paddle agitator and the surface baffles provided a certain backward recirculation of prelimed juice from each compartment to the preceding one, thus achieving progressive preliming. Practically all of the carbonation thickener underflow was added into the first compartment of the prelimer.

The prelimed juice was then fed to the prelime thickener. The underflow from the thickener was sweetened off on an Oliver filter, while the overflow was sent to a mixing tank where a

controlled quantity of carbonation thickener underflow was added before heating, main-liming and saturation. Overflow from the carbonation thickener was sent to second carbonation. The underflow was recirculated to the first compartment of the prelimer and also to the mixing tank ahead of carbonation.

Average results obtained during the period October 1-24, 1958, are tabulated below:

Juice	Purity[1]	Color[2]
Diffusion	82.15	—
Pilot Plant Effluent	88.07	0.2030
Factory Dorr Overflow	87.78	0.2642
Factory Kelly Filtrate	87.79	0.2327

[1] Apparent Purity
[2] Optical Density at 420 Angstrom. — These values times 1000 equal Spekker degrees—the color classification used in British factories.

Operating conditions were as follows: Flow Rate—75 U.S. gallon/minute: Preliming—0.25% CaO on beets, 55-60°C; Carbonation—1.75% CaO on beets, 70°C.

During the period October 29 to November 11, 1958, the temperature after the preliming was raised to 70°C, obtaining the following results:

Juice	Purity[1]	Color[2]
Diffusion	82-15	—
Pilot Plant Effluent	87.01	0.2350
Factory Dorr OverFlow	86.36	0.2540
Factory Kelly Filtrate	86.28	0.2210

[1] Apparent Purity
[2] Optical Density at 420 Angstrom. — These values times 1000 equal Spekker degrees—the color classification used in British factories

Under both operating conditions mentioned, unit areas in the thickeners were as follows: Prelime Thickener—0.309 sq Ft/short ton beets/day; Carbonation Thickener—0.215 sq Ft/short ton beets/day.

Statistical analysts of the date tabulated above indicate a high level of confidence.

In summary, it can be said that this pilot plant achieved a 0.3 point purity increase over the Factory Dorr overflow, with a color reduction of 30% and a lime saving of 20%. By heating prelimed juice, the lime saving was maintained and a purity rise of 0.7 point was achieved but the color did not improve. The tests therefore demonstrate the improvements which can be obtained with preliming and removal of prelime solids. Although a thickener was used for removal of prelime solids, other removal devices, such as a Webtrol belt filter might also be used.

Present Recirculation Practice With Dorrclones in Italy

From all of the experiences briefly summarized up to this point, as well as from a survey of the patents and of the various processes which have been described from time to time in the technical press, the basic requirements for improving the filterability of carbonation juices can be said to consist of:

1. The presence of calcium carbonate particles in the raw juice before it is submitted to treatment by heating or liming. Organics that are coagulated only by the action of heat and lime do not form aggregates with the calcium carbonate which is precipitated later in the gassing step. The failure to form such aggregates renders the subsequent clarification operations more difficult regardless of whether this clarification step consists of settling followed by vacuum filtration or by direct filtration with either pressure or vacuum filters.

2. The calcium carbonate particles added to the raw juice should be as clean, as hard, and as large as possible. The addition to the raw juice of calcium carbonate, from which the coagulated organics and fine particles have not been separated, does not produce optimum results.

Other considerations that contributed to the process described in this section are:

1. Only a small part of the total lime usually added to the juice is necessary for reacting with those nonsugars which can be precipitated by liming and carbonating. The balance is needed only for creating solid nuclei within the liquor to be clarified.

For instance, it can be said that 1% CaO on beets, from a strictly chemical point of view, would be just as effective as 2%, all other conditions remaining the same.

2. First carbonation cake as discharged from Oliver drum filters is an excellent source of calcium carbonate which has been formed in the liquid being treated—the sugar solution.

Being inexpensive, and a waste product unless employed for soil conditioning purposes, it is logical to make use of it.

The addition of finely ground limestone to raw juice has been tried but without satisfactory results, to the best of our knowledge, for reasons that are beyond the scope of these notes.

It has also been suggested that calcium carbonate for use in raw juice might be obtained by washing and classifying carbonation cake in a hydroseparator, but this has not been tested[3].

3. Porcelain DorrClones were extremely effective in thickening first carbonation juice but their overflow was always slightly turbid on account of the suspended fine solids.

[3] Suggested by R. C Campbell, retired sugar technologist of Dorr-Oliver Inc., Stamford, Conn.

Since 80% of these fines were colloids, it was reasonable to take advantage of this apparently negative result for removing coagulated impurities from a slurry of carbonation cake. DorrClones were expected in this use to overflow the finest solid particles so that the larger particle sizes could be recovered in the underflow. The intense shearing action which occurs within the cylindriconical DorrClone body was expected to separate some organics not fully ama/gamated with the particles but adhering to their surfaces.

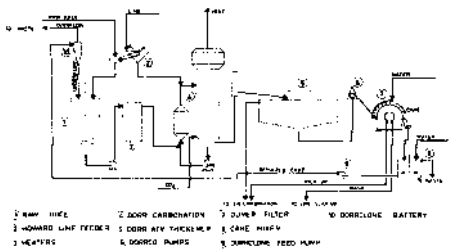

Figure 9.—First carbonation flowsheet incorporating recycle of filter cake after washing and classifying in DorrClones.

As a result of all the above considerations, the simple flowsheet shown on Figure 9 was developed[4]. It includes repulping first carbonation filter cake with clean water, pumping the resulting slurry through a battery of DorrClones which classifies out and discards the organic impurities and the finest calcium carbonate particles, and yields a clean calcium carbonate in the underflow. This product is returned to the raw juice prior to any heating or liming treatment. Among the several purification processes using DorrClones for facilitating the handling of first carbonation juice, this one has two unique features: 1. Washing of the first carbonation cake by repulping in water and; 2. classifying the suspended solids and discarding the fines which are the most detrimental, chemically and physically.

The addition of a slurry containing clean and classified calcium carbonate particles to the raw juice produces results which, according to the available information, include:

1. Stabilization of the raw juice. Its pH is increased from a value below 7 to 8 - 8.5 by the alkaline slurry which still contains a little active lime. This stabilization enables the

[4] Patents applied for.

juice to be heated ahead of liming without fear of inversion or of coagulation of nonsugars, such as occurs when raw juice, free of lime, is heated.

This is particularly important with raw juice from certain types of continuous diffusers which tend to increase the extraction of organic impurities that are usually very difficult to remove without a large excess of lime.

2. Foam in the raw juice tank is easily controlled by adding the recycled carbonate slurry as a spray. The addition of conventional defoaming chemicals is unnecessary except in emergencies.

3. The lime consumption is consistently reduced because it is possible to build up a stock of calcium carbonate within the system of any desired concentration. The solids handling capacity of the equipment is the limiting factor. A maximum reduction of 40% in the lime consumption was obtained in a factory operating in accordance with the flowsheet shown on Figure 10A.

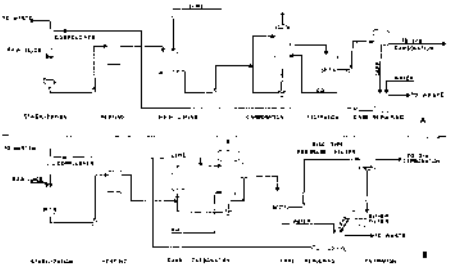

Figure 10.—Flowsheet A resulted in recovery of 50 tons of sugar in a 60-day campaign with a 40% reduction in total lime consumption. Same factory later adopted flowsheet B, with comparable results.

It can be said that the governing factor for the clarification of carbonation juice is the weight ratio of the calcium carbonate solids to the nonsugars, all other conditions, such as temperature concentration, viscosity, alkalinity, and detention times, being held constant. The recirculation of clean and classified calcium carbonate permits a reduction in the fresh lime addition while maintaining, or even increasing, this weight ratio.

4. Sweetening off of the carbonate cake is greatly improved as it contains mostly large particles. The fines are restricted to those originating from the carbonation of the reduced addition of fresh lime.

The carbonate particles are continuously washed, classified, and recirculated within the system, allowing the nascent calcium carbonate to grow on large size nuclei of 15 to 35 microns. The filter cake is thus unusually porous and requires less water for sweetening off.

Wash water dilution of the juice amounting to 6% on beets was sufficient to reduce sugar losses on press cake to about 0.6 to 0.7% whereas before the installation of the process, the dilution was 12% on beets and still left about 1% sugar on cake at 50% moisture. Press operators agree that the cake is extremely porous and not sticky, so that it drops easily, on opening the presses, without any manual help.

On Oliver drum filters the dilution by wash water required to reduce the sugar loss to 0.4% on wet cake is about 4% or less on beets.

Like everything born of the human mind, this system also has some minor disadvantages:

1. By recycling a water-suspended slurry there is a small increase in juice dilution which in turn increases evaporation costs. It is possible that a reduction in sweet water production at the first carbonation filter station, as described above, will be sufficient to offset this disadvantage but, as yet, there are not enough data to fully support this belief.

2. Because the dilute DorrClone overflow must be added to the factory effluent waters there is more effluent to be disposed of. This disadvantage may be serious where local conditions limit the amount of effluent which can be discharged into public waters or existing ponds. It should be remembered, however, that the total amount of waste solids has been reduced, as compared to conventional processes, because of the reduction in the fresh lime consumption.

No significant change for better or worse has been experienced to date in juice purity or color.

Although it does have a limited amount of chemical activity, the carbonate recycled is not essentially a chemical reagent. Consequently, this recirculation scheme, although conceived as a logical improvement to the standard Dorr Carbonation Process, can be adapted to any juice purification process without losing the unique characteristics of that process.

The recirculation of cleaned and classified calcium carbonate from the DorrClones improves settling rates as well as filtration

rates on thickened sludge. Bulk settling rates ranging from 18-23 feet/hour, or double the rate found without recirculation, were obtained in most of the many tests made. After 30 minutes detention, the sludge volumes never exceeded 20%, of the unsettled juice volumes.

The settling and filtration rates obtained in commercial installations were in agreement with the laboratory tests and checked the values obtained with other systems of solids recirculation described earlier. Consequently in new plants equipment sizes can be reduced and, in existing plants, a higher factor of safety is obtained or plant capacity may be increased. The reduction in fresh lime consumption extends these advantages to the lime kiln, slaking, and degritting stations and the gas pumps.

No detailed figures are given with regard to economics since the unit costs of lime, fuel, beets, and sugar vary from one country to another and, even in the same country, from one factory to another. A factory slicing 1200 tons beets per day operating according to the flowsheet shown on Figure 10A recovered about 50 tons of sugar in a 60-day campaign, because of reduced filter cake losses and in addition, other savings resulting from a 40% reduction in lime consumption.

After two campaigns, this same factory adopted the flowsheet shown on Figure 10B, using leaf-type pressure filters from which the cake is sweetened off on Olivers, and maintained the same advantages.

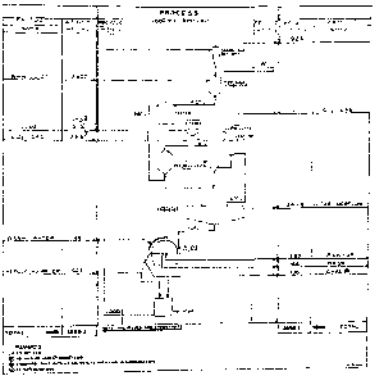

Figure 11.—Simplified material balance for 2,000-ton per day beet sugar factory incorporating DorrClones for recycle of filter cake.

A simplified material balance for a factory with a capacity of 2,000 tons of beets per day is detailed on Figure 11 from which economic calculations can be made using existing unit costs for any particular case along with the following factors: 1. Reduced lime consumption, with a maximum of 40%; 2. reduced cake losses to about 0.4% on cake at 50% moisture; 3. reduced wash water dilution to about 4% on beets; 4. increased load on the evaporators of about 2% and; 5. increased amount of waste effluent depending on present practice.

Installation and operating costs must, of course, be calculated for each individual case.

Summary

To summarize, a number of modifications of the basic Dorr Carbonation System have been reviewed. All include a scheme of solids recirculation. Several systems using preliming in Brieghel-Miiller type prelimers together with solids recirculation showed possible advantages in improved color, settling, and filter rates and a reduction in lime consumption.

Through the use of DorrClones in a novel flowsheet developed in Italy, cleaned and sized calcium carbonate particles are produced from filter cake and recycled to raw juice prior to heating or liming. Results from installations show substantial advantages.

A material balance is presented which permits economic calculations for specific installations.

Chemical Genetic and Soils Studies Involving Thirteen Characters in Sugar Beets[1, 6]

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The increased use of nitrogen fertilizers in the production of sugar beets emphasizes the importance of chemical-genetic and soils studies pertaining to processing quality in sugar beets. Of particular interest are the interrelations of weight per root, percentage sucrose, and percentage apparent purity and chemical characters in the thin juice and in the petioles as influenced by certain fertilizer practices. The purpose of this article is to study the interrelations of the different characters. Some of the information has been reported in previous articles, Payne et al. (15, 16) and Powers et al. (19, 20, 21).

Achard was the first to show that percentage sucrose in the sugar beet can be increased by breeding and hence is subject to genetic control (see Coons 4). That chemical characters other than sucrose are subject to genetic control and hence can be manipulated by breeding procedures has been shown by Dahlberg (6), Doxtator and Bauserman (7) , and others (10, 19, 22, 24, 29). Rorabaugh (23) discusses the effects of impurities on crystallization of sucrose and Carruthers and Oldfield (3) give methods for the assessment of beet quality. Haddock, Linton, and Hurst (13) discuss the association between certain nitrogenous constituents and sucrose percentage and purity of sugar beets.

Materials, Experimental Design, Methods, and Analyses

These studies were conducted during 1956 and 1958, and 1960 and 1961.

[1] Cooperative investigations of the Colorado Agricultural Experiment Station, the Crops Research Division, Agricultural Research Service, U. S. Department of Agriculture, and the Beet Sugar Development Foundation. The Colorado State University gratefully acknowledges financial support from the James G. Boswell Foundation administered by the Agricultural Research Center of Stanford Research Institute, the National Institutes of Health, the Beet Sugar Development Foundation, the National Plant Food Institute, and contract-research-funds [12-14-100-4549(34)] from the Agricultural Research Service of the U. S. Department of Agriculture. These studies would not have been possible if these funds had not been grant el. Approved by the Colorado Agricultural Experiment Station for publication as Scientific Series Article No. 789.

[2] Geneticist, Crops Research Division, Agricultural Research Service, U. S. Department of Agriculture.

[3] Agronomist, Colorado Experiment Station, Coloralo State University.

[4] Professor of Biological Statistics, Cornell University, on leave from Cornell University.

[5] Professor of Chemistry and Chemist for Colorado State Agricultural Experiment Station, Colorado State University.

[6] The writers are indebted to R. Ralph Wood of The Great Western Sugar Company for obtaining thin juice samples bv an oxalate method standard with his company and also acknowledge the cooperation of D. W. Robertson and Robert S. Whitney in conducting the field experiment. The writers are indebtel to the Western Data Processing Center at the University of California at Los Angeles for use of the computing facilities for analyzing data, Job' No. 398.

For 1956 the materials are populations A54-1, A54-1BB, 50-406BB, 50-406, F_1 hybrid, and 52-307. A54-1 is a commercial variety and A54-1BB resulted from seed harvested from 25 mother beets of A54-1. The 25 mother beets giving rise to population A54-1BB were grown in an isolated seed plot along with 25 mother beets from each of 22 other populations. Seed was harvested from all mother beets on an individual plant basis and seed from each mother beet of A54-1 was bulked to produce the seed for population A54-1BB. Seed saved from mother beets of 50-406 and handled in a similar manner produced the population designated as 50-406BB. Hence, 50-406BB is a top-cross hybrid. Populations 50-406 and 52-307 are inbreds and the F_1 hybrid population resulted from crossing them.

For 1958 the populations were A56-5BB, A56-5BB,, A54-1, 52-430, F_1 hybrid, and 55-5307. A56-5BB is the population produced from the seed of 120 mother beets of A56-5 surrounded by mother beets from A54-1, AC No. 2, MW 391, US 201, SL 028, and Janasz. A54-1 is a commercial variety, 52-430 and 55-5307 are inbred lines, and the F_1 hybrid resulted from crossing these two inbreds. Since 50-406 and 52-430 have green hypocotyls (rr) and the inbreds 52-307 and 55-5307 have red hypocotyls (RR) the hybrid populations for both 1956 and 1958 were obtained by leaving only red hypocotyl beets during thinning. The same was true for 1960 and 1961. For 1960 the populations are an F_1 hybrid and A56-3 and for 1961 the populations are three F_1 hybrids and A56-3.

For 1956 there were two treatments, non-fertilized and fertilized. The fertilized plots received one surface application of 100 pounds of N (in the form of ammonium nitrate) and one surface application of 250 pounds of P_2O_5 per acre on April 4, 1956. The fertilizer was cultivated under with a rototiller. The experiment was planted on April 10 and 11. On June 26, another 100 pounds of N per acre were drilled in the center of each space between rows of the fertilized plots.

For 1958, 250 pounds of P_20_5, were applied to all plots on April 3 and disked deeply into the soil. On March 4, 50 pounds of N were applied to all plots and harrowed under. On March 8 an additional 100 pounds of N were applied to one third of the plots and 250 pounds to another one third of the plots. This made three applications of nitrogen, 50 pounds, 150 pounds, and 300 pounds of N (in the form of ammonium nitrate) per acre. For 1960 and 1961, 100 pounds of N (in the form of ammonium nitrate) and 250 pounds of P_20_5 per acre were applied on the entire experimental area.

In 1956 and 1961 every plot was bordered by a row of A54-1 to provide uniform competition. After thinning, each plot had

12 plants in 1956 and 24 plants in 1961. In 1956 and 1958 data were taken on only eight plants, the extra plants at each end of the row being discarded at time of harvest. In 1960 all the plants per plot were harvested and in 1961, 15 competitive plants per plot were harvested. The stands were excellent in all years. The petiole samples were taken just prior to harvesting of the roots.

Percentage of apparent purity and concentrations of total nitrogen, glutamic acid, betaine, and chlorides were determined from thin juice samples only. However, the associations between chemical determinations in the thin juice and press juice have been found to be extremely high (see 22). The chemical characters are expressed as milligrams per 100 milliliters of thin juice equated to a refractometer reading of 10. Potassium and sodium were determined from both thin juice and petiole samples. In the thin juice they are expressed as parts per 100,000 and in the petioles as parts per million. Finally, nitrate nitrogen and phosphorus were determined only from petiole samples and are expressed as parts per million (for methods see Johnson and Ulrich, 14). For more details of the method used to determine total nitrogen see Payne et al. (15). Standard methods were used in making the chemical determinations for the other characters (see Payne et al. 16).

In 1958 the thin juice samples from the individual plants were composited to make five replication groups. For both 1956 and 1958, the petioles taken from each plant were composited to form 10 replication groups.

The methods used in analyzing the data are those given by Powers et al. (19, 20, 21), Federer (8), and Federer et al. (9). For the details of the methods of calculation the reader is referred to these articles. Primarily, the analyses were made by employing the analysis of variance, components of variance methods of genetic analysis, and regression. In this article the biology and not the statistical procedures will be emphasized.

A summary of the experimental designs, materials, etc. are given in Table **1.**

Results

This article is concerned, primarily, with the interrelations of the characters as determined by a study of the means and simple correlation coefficients, with phenotypic-dominance phenomena, and dates of harvest and combining ability tests.

Means

The means will be studied first as the determination of phenotypic-dominance involves a comparison of the means. The means for populations within treatments for 1956 are listed in Table 2.

Table 1.—Summary of experimental designs and of treatments, populations, and characters studied.

Year	Designs of experiment	Number of replications	Number of plants per plot after thinning	Number of plants harvested	Space (inches)		Treatments	Populations	Characters recorded for root	Characters recorded for petioles
					Plants	Rows				
1956	Split-plot with treatments as whole plots and populations as split plots	**40**	**12**	**8**	**20**	**22**	Non-fertilizwd Fertilized 200 lb N 250 lbs $p_2 0_5$	A 54-1 A51-IBB 50-406 50-406BB F_1 52-307	Wt. /root, lb Sucrose, % Puritv, % Total N, mg Glutamic acid, mg Betaine, mg Potassium, mg Sodium, mg	Nitrate N, ppm Phosphorus, ppm Potassium, ppm Sodium, ppm
1958	Split-plot with treatments as whole plots and populations as split plots	30	**24**	**8**	10	**44**	501b N 150 **lb** N 300 1l) N	A56-5BB A56-5BBi A54-1 52-430 F_1 55-5307	Wt. /root, lb Sucrose, % Puritv, % Total N, mg Glutamic acid, mg Belainc, mg K,pp 100,000 Na.pp 100,000 Chlorides, mg	Nitrate N, ppm Phosphorus, ppm Potassium, ppm Sodium, ppm
1960	Randomized complete block	30	**24**	**all**	**10**	**22**		F_1 A56-3	Wt./root, kg Sucrose, % Purity, %	
1961	Randomized complete block	**10**	**24**	**15**	10	**22**		F. (1) F_1 (2) F1 (3) A56-3	Wt. /plot, kg Sucrose, % Sugar/plot, kg Puritv, % Total N, mg	Nitrate N, ppm

Table 2.—Means for populations within treatments, 1956.

Treatment and population	Weight[1,2]	Sucrose[1,2]	Purity	Thin juice[1,2] Nitrogen	Gluamic acid	Betaine	Potashim	Sodium	Petioles[3] Nitrate N	Phosphorus	Potassium	Sodium
	lb	%	%	mg	mg	mg	pp 100,000	pp 100,000	ppm	ppm	ppm	ppm
Non-fertilized												
A54-1	1.93	17.9	96.1	18.8	12.8	89.8	122.6	27.0	1736	1798	29275	10940
A54-1BB	1.71	17.8	96.5	16.9	7.6	85.5	114.4	25.0	1592	1902	27160	11280
50-406BB	1.40	17.6	97.2	12.6	6.4	69.9	99.3	19.5	1491	1556	25505	8490
50-406	0.75	17.4	95.9	14.6	8.0	73.4	103.8	17.6	962	1388	23315	6875
Fi	1.54	17.6	97.6	9.8	3.5	51.3	96.0	19.9	1284	1899	31665	9010
52-307	0.58	16.5	96.4	11.1	3.0	79.8	106.8	25.8	1253	2616	33520	10455
Fertilized												
A54-1	2.60	16.8	93.2	46.8	80.1	116.6	133.1	47.9	4233	1598	17695	15265
A54-1BB	2.64	16.7	93.0	44.8	41.4	115.1	129.7	49.6	4884	1628	18795	15050
50-406BB	2.04	17.3	94.4	33.6	45.8	106.1	112.5	31.5	4267	1335	17500	11460
50-406	1.02	16.1	92.6	31.2	17.3	125.3	122.6	33.4	2297	1362	17650	9570
Fi	2.23	17.6	95.3	21.3	18.0	101.3	101.9	33.5	3060	1545	18595	10570
52-307	1.16	16.6	94.9	18.6	10.0	108.7	99.4	43.4	3644	2234	24690	14995
LSI) at 5% level'	0.12	0.3	0.4	3.2	8.0	6.1	5.1	3.4	817	159	2020	1121
LSD at 5% level*	0.14	0.4	0.7	3.7	8.5	6.9	5.3	4.2				

1,2 These least significant differences are calculalcd using appropriate variance formula (see Fedcrer, 8).

3 These least significant differences are calculated from replications condensed to 10 groups.

4 For comparing populations within treatments.

5 For comparing treatments within populations.

Populations A54-1 and A54-1BB have the largest weights per root on both the non-fertilized and the fertilized plots. Also, these two populations have the highest percentage sucrose on the non-fertilized plots. This is not true of the fertilized plots as the F_1 and 50-406BB have the highest percentage sucrose. They are intermediate in weight per root. On the non-fertilized plots, the lowest weight per root and the lowest percentage sucrose occur in population 52-307 and on the fertilized plots in population 50-406. Definitely, there is a genotype-environment interaction as regards the interrelation of weight per root and percentage sucrose. The fact that higher weight per root and higher percentage sucrose can be combined is of economic importance. The F_1 and 50-406BB are highest in percentage apparent purity on the non-fertilized plots and the F_1 52-307, and 50-406BB are highest in percentage apparent purity on the fertilized plots. It is interesting to note that both the F_1 and 50-406BB are hybrid populations, the F_1 being a single-cross hybrid and 50-406BB being a top-cross hybrid.

Total nitrogen in the thin juice is lowest for the F_1 and 52-307 and highest for A54-1 and A54-1BB. This is true of both the non-fertilized and fertilized plots. On the fertilized plots glutamic acid averages highest for A54-1 and lowest for 52-307, 50-406, and the F_1 A54-1 has more than twice as much total nitrogen as 52-307 and eight times as much glutamic acid on the fertilized plots. A54-1BB and 50-406BB are intermediate in glutamic acid and do not differ significantly from each other. The F_1 is lowest in betaine on both the non-fertilized and fertilized plots. A54-1 and A54-1BB are highest in betaine on the non-fertilized plots and 52-307, 50-406, and 50-406BB are intermediate. On the fertilized plots 50-406 is highest in betaine and A54-1 and A54-1BB are next highest with 52-307 and 50-406BB having still lower betaine but having more betaine than the F_1 hybrid.

On the non-fertilized plots and for the thin juice the F_1 and 52-307 are lower in potassium than A54-1 and A54-1BB but they are higher in potassium in the petioles. On the fertilized plots 52-307 is again lower than A54-1 in potassium in the thin juice but higher in potassium in the petioles. It is clear that comparative concentrations of potassium in the petioles are not necessarily indicative of the comparative concentrations of potassium in the thin juice. On the non-fertilized plots all but populations A54-1BB and 50-406BB, and 52-307 and the F_1 differ significantly from each other as regards potassium in the petioles; whereas, on the fertilized plots only 52-307 differs significantly from the others. Such facts as these must be kept in mind when

interpreting the genetic correlation coefficients which are presented later in this article.

Comparisons involving sodium in the thin juice and sodium in the petioles agree fairly well, both on the non-fertilized and fertilized plots. In these comparisons 50-406BB, 50-406, and the F_1 have low sodium and A54-1, A54-1BB, and 52-307 have high sodium. Population 50-406 has significantly lower sodium in the petioles than any other population with the possible exception of the F_1 hybrid on the fertilized plots.

On the non-fertilized plots there are no statistically significant differences at the 5% level in the concentrations of nitrate nitrogen in the petioles. On the fertilized plots 50-406 and the F_1 are lowest in nitrate nitrogen in the petioles, 52-307 is intermediate, and A54-1, A54-1BB, and 50-406BB are highest.

The comparisons between total nitrogen in the thin juice and nitrate nitrogen in the petioles for the two inbreds and their corresponding F, are informative. Populations 52-307 and the F_1 have lower total nitrogen than 50-406 in the thin juice and higher nitrate nitrogen in the petioles. The latter comparison between 52-307 and 50-406 on the fertilized plots is statistically significant at the 5% level. It is apparent that comparative concentrations of nitrate nitrogen in the petioles cannot be taken as indicative of comparative concentrations of total nitrogen in the thin juice.

Population 52-307 is highest in phosphorus on both the non-fertilized and fertilized plots, whereas 50-406 and 50-406BB are the lowest. Populations A54-1, A54-1BB, and the F, are intermediate and do not differ materially. These data were taken on the petiole samples. The comparisons are essentially the same for the non-fertilized and fertilized plots.

A comparison of the treatment means of Table 2 reveals that for 1956, on the fertilized plots as compared with the non-fertilized plots, there is an increase in weight per root, total nitrogen, glutamic acid, betaine, potassium in the thin juice, sodium in the thin juice and petioles, and nitrate nitrogen in the petioles. The same comparisons reveal that there is a decrease on the fertilized as compared with non-fertilized plots for percentage sucrose, percentage apparent purity, and phosphorus and potassium in the petioles. Potassium on an average is higher in the thin juice on the fertilized plots; whereas, the reverse is true for potassium in the petioles. It is clear that comparative concentrations of potassium in the thin juice cannot be taken as indicative of comparative concentrations of potassium in the petioles. Also, it should be noted that there is a genotype-environmental interaction as regards potassium. Population 52-307 is significantly lower in potassium in both the thin juice

and petioles on the fertilized plots than on the non-fertilized plots, whereas all the other varieties have higher potassium in the thin juice on the fertilized plots as compared with the non-fertilized plots and lower potassium in the petioles of the fertilized plots as compared with the non-fertilized plots. This is an interaction involving genotype, fertilizer treatments, and location in the plant (tops or roots). This behavior would be expected if some populations retain greater amounts of potassium in the tops whereas for other populations it is translocated to the roots, resulting in greater concentrations of potassium in the thin juice.

The 1958 means for populations within treatments are listed in Table 3, the means for populations over all treatments in Table 4, and the means for treatments over populations in Table 5.

Population A54-1 has the largest weight per root fallowed by A56-5BB, and A56-5BB. The F_1, hybrid is fourth in weight per root and 55-5307 and 52-430 fifth and sixth, respectively. Populations A54-1 and 52-430 average highest in percentage sucrose and the F_1 is third followed by A56-5BB and A56-5BB,. Population 55-5307 is lowest in percentage sucrose. The F_1 is highest in apparent purity and 55-5307 is lowest. These data show that high weight per root and high percentage sucrose are not mutually exclusive as the highest percentages sucrose are for A54-1 and 52-430, the former having the largest weight per root and the latter having the smallest weight per root.

Population 55-5307 averages highest in total nitrogen and A54-1 and the F_1 are next highest. A56-BB, A56-5BB_1 and 52-430 are lowest. Glutamic acid follows nearly the same pattern. Population 55-5307 is highest in betaine followed by 52-430 and A54-1. The F_1 is next in order of magnitude and A56-5BB and A56-5BB, are lowest.

The level of chlorides is low for all populations and the F_1 has the lowest chlorides of all populations. Population 55-5307 is next lowest followed in order bv 52-430, A54-1, A56-5BB, and A56-5BB_1.

Populations 55-5307 and 52-430 are lowest in nitrate nitrogen in the petioles, whereas A56-5BB and A56-5BB, are highest. As regards phosphorus, population 55-5307 is lowest and 52-430 is highest. The F_1 and the other three populations do not differ materially as regards parts per million of phosphorus in the petioles. Population 55-5307 is lowest in potassium in the petioles, the F_1 and 52-430 are next lowest, A54-1 is intermediate, and A56-5BB and A56-5BB, are highest. Populations 52-430 and the F_1 are lowest in sodium, whereas the other four populations are not materially different.

Table 3.—Means for populations and treatments, 1958.

Treatment and populations	Weight	Sucrose	Thin Juice					Petioles			
			Purity	Nitrogen	Glutamic acid	Betaine	Chlorides[2]	Nitrate N	Phosphorus	Potassium	Sodium
	lb	%	%	mg	mg	mg	mg	ppm	ppm	ppm	ppm
50 lb N.											
A56-5BB	2.52	13.4	89.5	57.2	44.2	121.3	3.14	3433	1400	33315	19095
A56-5BB$_1$	2.59	13.4	89.6	55.2	44.0	122.2	3.24	3724	1390	32055	19265
A54-1	2.99	14.6	89.2	62.9	72.0	154.9	3.12	2683	1318	27745	19430
52-430	1.39	14.4	89.4	63.1	49.4	169.1	2.73	1493	2018	19990	16135
F_1	2.32	13.9	89.7	59.3	54.8	151.5	2.19	2220	1380	17065	16890
55-5307	1.62	12.8	85.8	86.9	132.6	181.8	2.48	1392	1241	9870	21335
150 lb N											
A56-5BB	2.58	13.0	87.4	70.6	75.0	133.2	2.35	5626	1348	30380	19095
A56-5BB.	2.58	13.0	86.5	75.3	92.6	148.8	2.69	4726	1300	29540	19295
A54-1	2.84	14.0	86.4	79.7	115.2	166.5	2.52	4626	1261	22465	18310
52-430	1.37	14.0	87.0	68.9	67.8	199.1	2.41	2648	1651	15665	15845
Fi	2.21	13.7	87.9	76.0	93.8	158.4	1.83	3358	1269	14235	16970
55-5307	1.70	12.1	82.6	117.8	158.4	222.8	2.24	1929	1052	6560	17775
300 lb N											
A56-5BB	2.58	12.1	84.0	85.7	108.0	142.5	2.65	9660	1239	27630	19605
A56-5BBJ	2.53	11.9	83.6	88.0	131.2	151.3	2.87	9262	1129	27750	19745
A54-1	3.03	13.0	82.7	103.5	158.4	190.4	2.36	7968	1238	21960	18115
52-430	1.29	13.4	85.0	89.5	95.6	214.0	2.47	5099	1486	14860	15030
Fi	2.32	13.0	84.8	102.0	132.2	177.2	1.82	6386	1045	14720	15225
55-5307	1.67	11.3	78.8	144.5	228.8	229.5	2.28	5274	886	9970	17205
LSD at 5% level[1]	0.21	0.4	0.7	8.3	25.5	17.7	0.32	885	200	1784	2249

[1] These LSD's are calculated from the replications condensed to 5 groups.

2 The writers are indebted to Lynn Gardner, Research Assistant, Department of Chemistry, Colorado State University, for chloride determinations.

Table 4.—Means for populations, average of three fertility treatments, 1958.

			Thin Juice					Peticles			
	Weight			Nitro-gen	Glutamic acid	Betaine	Chlor-ides	Nitrate N	Phos-phorus	Potas-sium	Sodium
Populations	**lb**	**%**	**%**	mg	mg	mg	**mg**	ppm	ppm	ppm	ppm
A56-5BB	2.56	12.8	87.0	71.2	75.7	132.3	2.71	6240	1329	30442	19265
A56-5BBi	2.57	12.8	86.6	72.8	89.3	140.8	2.93	5904	1273	29782	19435
A54-1	2.95	13.9	86.1	82.0	115.2	170.6	2.67	5092	1272	24057	18618
52-430	1.35	13.9	87.1	73.8	70.9	194.1	2.54	3080	1718	16838	15670
Fi	2.28	13.5	87.5	79.1	93.6	162.4	1.95	3988	1231	15340	16362
55-5307	1.66	12.1	82.4	116.4	173.3	211.4	2.33	2865	1060	8800	18772
LSD at 5% level	0.12	0.2	0.4	4.8	14.7	10.2	0.18	511	115	1030	1298

Table 5.—Means for treatments, avreages of all populations, 1958.

				Thin Juice				Petioles			
Treatment	**Weight**	**Sucrose**	**Purity**	Nitro-gen	Glutamic acid	Betaine	Chlor-ides	Nitrate N	Phos-phorus	Potas-sium	Sodium
	lb	**%**	**%**	mg	mg	mg	**mg**	ppm	ppm	ppm	ppm
50 **lb N**	2.24	13.7	88.9	64.1	66.2	150.1	2.82	2491	1458	23340	18692
150 **lb N**	2.21	13.3	86.3	81.4	100.5	171.4	2.34	3819	1313	19808	**17882**
300 **lb N**	2.24	12.4	83.2	102.2	142.4	184.2	2.40	7275	1170	19482	17488
LSD at 5% level	0.08	0.2	0.3	3.4	10.4	7.2	0.13	361	81	728	918

The relations between total nitrogen and the nitrogenous compounds in the thin juice and nitrate nitrogen in the petioles are interesting. Population 55-5307 is materially higher than A56-5BB in total nitrogen, glutamic acid, and betaine in the thin juice and is decidedly lower than A56-5BB in nitrate nitrogen in the petioles. Further, population 55-5307 is materially higher than 52-430 in total nitrogen, glutamic acid, and betaine in the thin juice but does not differ materially from 52-430 as regards nitrate nitrogen in the petioles. These findings make it clear that the comparative concentrations of nitrate nitrogen in the petioles cannot be taken as an indication of the comparative concentrations of total nitrogen and nitrogenous compounds in the thin juice.

The treatment means for each population in 1958 are listed in Table 3 and the treatment means averaged over all populations in Table 5. It is clear from a study of these two tables that the weights per root do not differ materially with the amounts of nitrogen added as fertilizer. However, there are decreases in percentage sucrose, percentage apparent purity, phosphorus, potassium and sodium with increases in the amounts of nitrogen applied as fertilizer. Also as might be expected, there are increases in total nitrogen, glutamic acid, betaine, and nitrate nitrogen with increases in amounts of nitrogen applied as fertilizer.

One interaction of Table 3 involving total nitrogen in the thin juice is particularly interesting. At the 50 lb application of nitrogen, total nitrogen in the thin juice, percentage sucrose, and percentage apparent purity are not significantly different for populations A54-1 and 52-430. At 150 lb application of nitrogen, A54-1 compared with 52-430 is significantly higher in total nitrogen in the thin juice, has the same percentage sucrose, and approaches statistical significance in having lower percentage apparent purity. At the 300 lb application of nitrogen, A54-1 compared with 52-430 is significantly higher in total nitrogen, and significantly lower in both percentage sucrose and percentage purity. This confirms the findings for 1956 that genotypes differ in the concentrations of nitrogen in the thin juice grown under identical applications of fertilizer and that total nitrogen in the thin juice is negatively associated with percentage sucrose and percentage apparent purity.

Three interactions in Table 4 are of particular interest. First, the F_1 is significantly higher than A56-5BB, in total nitrogen in the thin juice, percentage sucrose, and percentage apparent purity. This shows that some genotypes may have higher nitrogen in the thin juice than other genotypes and at the same time have higher percentages of sucrose and purity. The second interaction

involves genotypes and total nitrogen in the thin juice as compared to nitrate nitrogen in the petioles. Population 55-5307 has the highest concentration for any population of total nitrogen in the thin juice and the lowest concentration for any population of nitrate nitrogen in the petioles; whereas A56-5BB has the lowest concentration for any population of total nitrogen in the thin juice and the highest concentration for any population of nitrate nitrogen in the petioles. It is evident that concentrations of nitrate nitrogen in the petioles cannot always be taken as a reliable indicator of comparative concentrations of total nitrogen in the thin juice. These first two interactions confirm findings for the 1956 data. Finally, 52-430 compared with the F_1 has a significantly higher concentration of betaine in the thin juice, has significantly higher sucrose, and is significantly lower in percentage purity.

Phenotypic Dominance

Phenotypic-dominance phenomena derived from a comparison of the means of the inbreds and their corresponding F_1 hybrids (see Tables 2, 3, and 4) are listed in Tables 6 and 7. The terms used in classifying the dominance phenomena are heterosis-!-, dominance +, partial dominance +, intermediate, partial dominance—, dominance — , and heterosis—. A plus following the designation indicates that the greater expression of the character exhibits the phenomenon tabulated. A minus following the designation indicates the smaller expression of the character exhibits the phenomenon listed. Heterosis is used when the expression

Table 6.—Phenotypic dominance for weight per root and chemical characters in the thin juice and in the petioles of the sugar beet, comparisons involve the inbreds and F_1 of 50-406 and 52-307, 1956.

Material and character	Non-fertilized[1] [2]	Fertilized[1,2] -
Weight per root	Heterosis +	Heterosis +
Sucrose	Heterosis +	Heterosis +
Thin juice		
Purity	Heterosis +	Heterosis +
Nitrogen	Heterosis—	Partial dominance—
Glutamic acid	Dominance—	Dominance-H
Betaine	Heterosis—	Heterosis—
Potassium	Heterosis—	Dominance—
Sodium	Partial dominance—	Dominance—
Petioles		
Nitrate nitrogen	Dorninance +	Intermediate
Phosphorus	Partial dominance—	Partial dominance—
Potassium	Partial dominance+	Partial dominance—
Sodium	Partial dominance +	Partial dominance—

[1] A plus following the designation shows that the greater expression of the character shows either partial dominance, dominance, or heterosis.

[2] A minus following the designation shows that the lesser expression of the character shows either partial dominance, dominance, or heterosis.

of the character goes beyond that of either parent, dominance when the character is not significantly different from one or the other parent, partial dominance when the expression of the character lies between the two parents but closer to the mean of one of the parents, and intermediate when the expression of the character is not significantly different from the mean of the two parents.

The data for 1956 are for the inbreds and the F_1 hybrid 50-406 X 52-307.

As would be expected weight per root exhibits heterosis in a plus direction on both the fertilized and non-fertilized plots. Likewise percentage sucrose and percentage apparent purity exhibit plus heterosis on both the non-fertilized and fertilized plots.

Considering the thin juice samples total nitrogen exhibits minus heterosis on the non-fertilized plots and minus partial dominance on the fertilized plots. Glutamic acid exhibits minus dominance on the non-fertilized plots and plus dominance on the fertilized plots. However, the differences on the non-fertilized plots are not significant at the 5% level. This behavior indicates a genotype-environment interaction involving comparisons between the two inbreds and the F_1. Betaine exhibits minus heterosis on both the non-fertilized and the fertilized plots. Potassium exhibits minus heterosis on the non-fertilized plots and minus dominance on the fertilized plots. Sodium exhibits minus partial dominance on the non-fertilized plots and minus dominance on the fertilized plots. Again the difference between the low sodium parent and the F_1, is not significant at the 5% level.

Table 7. Phenotypic dominance for weight per root and chemical characters in the thin juice and in the petioles of the sugar beet, comparisons involve the inbreds and Fi of 52-430 and 55-5307, 1958, averages for the three levels of nitrogen application.

Material and character	Average[1,2]
Weight per root	Heterosis+
Sucrose	Partial dominance+
Thin juice	
Purity	Dominance+
Nitrogen	Partial dominance—
Glutamic acid	Partial dominance—
Betaine	Heterosis—
Chlorides	Heterosis—
Petioles	
Nitrate nitrogen	Heterosis +
Phosphorus	Partial dominance—
Potassium	Partial dominance+
Sodium	Partial dominance—

[1] A plus following the designation shows that the greater expression of the character shows either partial dominance, dominance, or heterosis.
[2] A minus following the designation shows that the lesser expression of the character shows either partial dominance, dominance, or heterosis.

Turning to a consideration of the chemical characters taken on the petioles, nitrate nitrogen exhibits plus dominance on the non-fertilized plots and intermediate dominance on the fertilized plots. Phosphorus exhibits minus partial dominance on both the non-fertilized plots and the fertilized plots. Both potassium and sodium exhibit plus partial dominance on the non-fertilized plots and minus partial dominance on the fertilized plots. These are genotype-environment interactions and they are well established statistically.

The phenotypic-dominance phenomena for 1958 are tabulated in Table 7. Comparisons involve the inbreds and F_1 derived from crossing 52-430 and 55-5307.

Again weight per root exhibits plus heterosis as was expected. Percentage sucrose exhibits plus partial dominance and percentage apparent purity exhibits plus dominance. Nitrogen and glutamic acid exhibit minus partial dominance and betaine and chlorides exhibit minus heterosis. The data for the latter four characters are from thin juice samples as were the data for purity.

Again nitrate nitrogen, phosphorus, potassium and sodium were determined from petiole samples. Nitrate nitrogen showed plus heterosis, phosphorus minus partial dominance, potassium plus partial dominance, and sodium minus partial dominance.

The phenotypic-dominance phenomena tabulated in Tables 6 and 7 are of interest to the beet sugar industry even though in these two tables only two F_1 hybrids and their corresponding inbred parents are involved. The main characters are weight per root, percentage sucrose, and percentage apparent purity. In the F_1 of 50-406 X 52-307, all of these three characters exhibited heterosis. As previously pointed out this was to be expected for weight per root. The fact that percentage sucrose and percentage apparent purity also exhibit heterosis indicates that hybrids or at least some form of breeding utilizing heterosis will play a dominant role in the production of sugar from beets. Also it is significant that in the thin juice nitrogen, betaine, potassium, and sodium exhibited minus partial dominance, minus heterosis, minus dominance, and minus dominance respectively on the fertilized plots. On the non-fertilized plots these four characters and glutamic acid showed minus reactions. Such behavior would lead to comparatively smaller amounts of these chemicals in the thin juice. Such reactions in turn would be expected to result in higher percentages sucrose and purities. Therefore from this behavior of some of the impurities in the thin juice, plus heterosis for percentage apparent purity might have been anticipated.

Correlation Coefficients

The relations noted between characters in Tables 2 to 5, inclusive, and Table 8 can be expressed as correlation coefficients.

The simple correlation coefficients express average relations between two characters; that is measure the average association between two characters. It is important that this be kept in mind while interpreting the data, especially when associations differ materially depending upon the environment, genotype, location in the plant, or all three. It is equally important to keep in mind the limitations of the data. For example, the data of Table 8 include only two years and five fertilizer treatments. This is a small sample of years and fertilizer treatments. Likewise, the number of populations studied is 11 for the two years, 1956 and 1958. Such being the case when interactions are involved, it is necessary to study in detail the data in Tables 2 to 5, inclusive, and Table 8 in order to make correct deductions.

As is the case with the variances, the covariances can be divided into that portion attributable primarily to differences in the environment and that attributable primarily to genotypic differences. In these studies the environmental variances include a negligible amount of genetic variance and the genetic variances include a negligible amount of environmental variability. The environmental correlation coefficients will be considered first.

Environmental Variability

The means for treatments within years at five levels of total nitrogen in the thin juice for 1956 and 1958 and for population A54-1 are given in Table 8. These data permit a study of the interrelations of characters under partially controlled environmental conditions. Fertilizer practices were controlled within each year. As can be seen from an examination of Table 8 there were five levels of total nitrogen in the thin juice starting with the non-fertilized plots in 1956 and progressing to the 300 lb application of nitrogen in 1958. These five levels of total nitrogen in the thin juice resulted from applications of known amounts of nitrogen (in the form of ammonium nitrate) and from possible year effects. Known amounts of P_2O_5 were added to the fertilized plots in 1956, and to all of the plots in 1958. It should be noted that the data in Table 8 are only for population A54-1 and hence, genetic differences due to populations are non-existent.

A study of the means listed in Table 8 shows that the decreases in percentages sucrose and apparent purity, starting with the non-fertilized plots in 1956 and progressing to the 300 lb application of N in 1958, are accompanied by increases in total nitrogen, glutamic acid and betaine in the thin juice. In 1956 potassium and sodium in the thin juice increased also. As published in a previous article nitrogen, glutamic acid and betaine are highly associated as regards the environmental variability (16).

Table 8.—Means for treatments, within years, five levels of total nitrogen in the thin juice, 1956 and 1958, population A54-I.

Year and treatment	Thin juice								Petioles			
	Weight	Sucrose	Purity	Nitrogen	Glutamic acid	Betaine	Chlorides	Potassium	Sodium	Nitrate N	Phosphorus	Potassium	Sodium
	lb	%	%	mg	mg	mg	mg	pp 100,000	pp 100,000	ppm	ppm	ppm	ppm
1956													
Non-fertilized	1.93	17.9	96.1	18.8	12.8	89.8		122.6	27.0	1795	1798	29275	10940
Fertilized	2.60	16.8	93.2	46.8	80.1	116.6		133.1	47.9	4233	1598	17695	15265
1958													
50 lb N	2.99	14.6	89.2	62.9	72.0	154.9	3.12			2683	1318	27745	19430
150 lb N	2.84	14.0	86.4	70.7	115.2	166.5	2.52			4626	1201	22465	18510
300 lb N	3.03	13.0	82.7	103.5	158.4	190.4	2.36			7968	1238	21960	18115
LSD[1]	0.14	0.4	0.7	3.7	8.5	6.9		5.3	4.2	817	159	2020	1121
LSD[2]	0.21	0.4	0.7	8.3	25.5	17.7	0.32			885	200	1784	2249

[1] LSD at 5% level, 1956.
[2] LSD at 5% level, 1958.

First, the interrelations of weight per root, percentage sucrose, and percentage apparent purity will be considered. A study of Table 8 reveals that the differences in weight per root at the three levels of nitrogen application are not significantly different for 1958. This is true even though there are increases of total nitrogen in the thin juice and nitrate nitrogen in the petioles. Apparently each of the three applications of fertilizer provided sufficient nitrogen for maximum weight of root under the conditions of the experiment in 1958. There was a difference in weight per root as regards fertilizer applications in 1956 and a difference between the years, the weight per root being higher in 1958 than in 1956. These findings should be kept in mind when interpreting the simple correlation coefficients.

The correlation coefficients and percentages of the variances accounted for by regression (r^2 X 100) are listed in Table 9. A study of the data in the columns headed "Sucrose" and headed "'Purity" reveals that as weight per root increased percentage sucrose and percentage apparent purity decreased. The correlation coefficient for weight per root and percentage sucrose is — 0.90. Hence 81% of the environmental variability of percentage sucrose is accounted for by regression or covariance. The correlation coefficient for weight per root and percentage apparent purity is —0.86 and 74% of the environmental variability is accounted for by regression. One of the larger percentages of the environmental variability accounted for by regression is that between percentage sucrose and percentage apparent purity, 98% of the environmental variability being accounted for by regression. Hence, under the conditions of this experiment increased applications of nitrogen fertilizer resulted in no further increase in weight per root but resulted in a material reduction in both percentage sucrose and percentage apparent purity.

The correlation coefficients and the percentages of the variances accounted for by regression of weight per root, percentage sucrose, and percentage apparent purity on other chemical characters are listed in Table 9, also. The regressions of weight per root, percentage sucrose, and percentage apparent purity on total nitrogen in the thin juice account for 77, 94, and 98%, respectively of the environmental variability of these three characters. The relations with glutamic acid and betaine are very similar as are those with total nitrogen. The correlation coefficient involving weight per root is positive, whereas those involving percentage sucrose and percentage apparent purity and total nitrogen are negative. Nitrate nitrogen in the petioles is positively associated with weight per root and negatively associated with percentage sucrose and percentage apparent purity. Regression accounted for 41, 56, and 71% of the environmental variability.

Table 9.—Correlation coefficients for ten characters and percentages of the variances accounted for by regression, differences between fertilizer treatments and between years, 1956 and 1958[1].

Character	Thin juice										Petioles					
	Sucrose		Purity		Nitrogen		Glutamic acid		Betaine		Nitrate N		Phosphorus		Potassium		Sodium	
	r	r^2(100)	r	r^2(100)	r	r^2(100)	r	r^2(100)	r	r^2(100)	r	r^2(100)	r	r^2(100)	r	r^2(100)	r	r^2(100)
Weight	—0.90	81	—0.86	74	0.88	77	0.83	69	0.91	83	0.64	41	—0.95	90	—0.39	15	0.98	96
Sucrose			0.99	98	—0.97	94	—0.90	81	—0.997	99	—0.75	56	0.98	96	0.22	5	—0.88	77
Purity (thin juice)					—0.99	98	—0.94	88	—0.99	98	—0.84	71	0.94	88	0.29	8	—0.81	66
Nitrogen (thin juice)							0.97	94	0.99	98	0.88	77	—0.94	88	—0.40	16	0.82	67
Glutamic acid (thin juice)									0.92	85	0.94	88	—0.85	72	—0.58	34	0.74	55
Betaine (thin juice)											0.78	61	—0.97	94	—0.27	7	0.88	77
Nitrate nitrogen (petioles)													—0.65	42	—0.61	37	0.49	24
Phosphorus (petioles)															0.24	6	—0.95	90
Potassium (petioles)																	—0.26	7

[1] The value of r at the 5% level approximates 0.17.

Weight per root is negatively associated with phosphorus, whereas percentage sucrose and percentage apparent purity are positively associated with phosphorus. Regression accounted for 90, 96, and 88% of the environmental variability. Relatively minor portions of the environmental variability of weight per root, percentage sucrose, and percentage apparent purity are accounted for by the regressions of these characters on potassium; the values being 15, 5, and 8%, respectively. The percentages of the environmental variability accounted for by regressions of weight per root, percent sucrose and percent apparent purity on sodium are 96, 77, and 66, respectively. The relations are negative for percentage sucrose and percentage apparent purity. Total nitrogen and the nitrogenous compounds in the thin juice, and phosphorus and sodium in the petioles are most closely associated with weight per root, percentage sucrose, and percentage apparent purity.

The correlation coefficients for nitrogen, glutamic acid, betaine, nitrate nitrogen, phosphorus, potassium, and sodium, and the percentages of the variances accounted for by regression are listed in Table 9. It should be kept in mind they are calculated from the data for A54-1 for 1956 and 1958. Total nitrogen in the thin juice is very closely associated with glutamic acid and betaine and is rather closely associated with nitrate nitrogen, phosphorus, and sodium. The percents of the environmental variability accounted for by regression are 94, 98, 77, 88, and 67, respectively. The associations are positive for glutamic acid, betaine, nitrate nitrogen, and sodium, and negative for phosphorus and potassium.

The only other close associations are between betaine in the thin juice and phosphorus in the petioles and phosphorus and sodium in the petioles, 94 and 90% of the variability being accounted for by regression. The associations are negative. The relation between potassium and sodium is negative and only 7% of the variances are accounted for by regression. The negative relation is due to the differential behavior of these two characters in 1956 on the non-fertilized as compared with the fertilized plots. There was a decrease in potassium and an increase in sodium in going from the non-fertilized to the fertilized plots in 1956 (see Table 8). For the 1958 data both decrease with increased applications of nitrogen in the fertilizer. When such interactions occur, it must be kept in mind that the correlation coefficients present average relations and to obtain all of the information, data such as fisted in Table 8, must be studied in detail.

Genetic Variability

The average interrelations attributable to genetic variability are shown by the correlation coefficients and the percentages of

the variances accounted for by regression (r^2 X 100) in Tables 10 and 11. These constants measure the degree of association between characters whose variability is attributable, primarily, to differences between populations and hence are primarily genetic. The variability of each character contains only a negligible amount due to environmental differences.

The data for both the fertilized and non-fertilized plots are given in Table 10. The relation between weight per root and percentage sucrose is positive on both the non-fertilized and fertilized plots, 76% of the variability being accounted for in the former case and 29% in the latter. The association between weight per root and purity is not close on both fertilizer treatments, only 7% of the variability being accounted for by regression on the non-fertilized plots and practically none on the fertilized plots. On the fertilized plots regression accounted for 53% of the variability involving percentage sucrose and percentage apparent purity. This represents a decided increase in the proportion of the variance accounted for by regression calculated from the fertilized plots compared with the proportion accounted for by regression calculated from the non-fertilized plots. As was the case for the environmental variability, the relation is positive.

The correlation coefficients and the percentages of the variances accounted for by regression of weight per root, percentage sucrose, and percentage apparent purity on other chemical characters are listed in Table 10 also. They were calculated from differences between populations and are from the 1956 data.

A study of Table 10 reveals that the strongest association of characters other than sucrose for weight per root is with nitrate nitrogen in the petioles. Here for the non-fertilized and fertilized plots 69% and 52% of the variability are accounted for by regression. The next strongest associations as regards weight per root are with glutamic acid and total nitrogen in the thin juice. The percents of the variability accounted for by regression for the non-fertilized and fertilized plots are 31 and 52, and 27 and 44, respectively.

For percentage sucrose the closest association other than with weight per root on the non-fertilized plots is with phosphorus. It is negative and only 49% of the variability is accounted for by regression. The next strongest associations on the non-fertilized plots are with glutamic acid and total nitrogen in the thin juice. For the fertilized plots the strongest association is with betaine in the thin juice and is negative. Here 81% of the variability is accounted for by regression.

Table 10.—Correlation coefficients tor 12 characters and percentages of the variances accounted for by regression, differences between populations, 1956[1]

Character and treatment	Thin juice												Petioles					
	Sucrose		Purity		Nitrogen		Glutamic acid		Betaine		Potassium		Sodium		Nitrate N		Phosphorus		Potassium		Sodium	
	r	r^2(100)	r	r^2(100)	r	r^2(100)	r	r^2(100)	r	r^2(100)	r	r^2(100)	r	r^2(100)	r	r^2(100)	r	r^2(100)	r	r^2(100)	r	r^2(100)
Weight																						
Non-fertilized	0.87	76	0.27	7	0.52	27	0.56	31	0.13	2	0.43	18	0.31	10	0.83	69	—0.29	8	—0.03	0	0.48	23
Fertilized	0.54	29	—0.06	0	0.66	44	0.72	52	—0.27	7	0.47	22	0.45	20	0.72	52	—0.26	7	—0.46	21	0.41	17
Sucrose																						
Non-fertilized			0.12	1	0.58	34	0.66	44	0.04	0	0.29	8	—0.08	1	0.54	29	—0.70	49	—0.48	23	0.06	0
Fertilized			0.73	53	—0.19	4	0.12	1	—0.90	81	—0.42	18	—0.30	9	0.26	7	—0.17	3	—0.18	3	—0.13	2
Purity (thin juice)																						
Non-fertilized					—0.67	45	—0.57	32	—0.78	61	—0.68	46	—0.32	10	0.10	1	0.04	0	0.30	9	—0.05	0
Fertilized					—0.75	56	—0.40	16	—0.93	86	—0.90	81	—0.35	12	—0.05	0	0.39	15	0.47	22	—0.08	1
Nitrogen (thin juice)																						
Non-fertilized							0.93	86	0.81	66	0.89	79	0.46	21	0.56	31	—0.33	11	—0.41	17	0.38	14
Fertilized							0.86	72	0.49	24	0.95	90	0.55	30	0.60	36	—0.42	18	—0.60	36	0.40	16
Glutamic acid (thin juice)																						
Non-fertilized									0.64	41	0.77	59	0.29	8	0.52	27	—0.50	25	—0.45	20	0.18	3
Fertilized									0.17	3	0.73	53	0.45	20	0.61	37	—0.32	10	—0.52	27	0.45	20
Betaine (thin juice)																						
Non-fertilized											0.90	81	0.72	52	0.49	24	0.16	3	—0.11	1	0.57	32
Fertilized											0.71	50	0.25	6	—0.22	5	—0.22	5	—0.26	7	—0.01	0
Potassium (thin juice)																						
Non-fertilized													0.80	64	0.68	46	0.13	2	0.03	0	0.68	46
Fertilized													0.50	25	0.30	13	—0.44	19	—0.60	36	0.28	8
Sodium (thin juice)																						
Non-fertilized															0.68	46	0.66	44	0.57	32	0.94	88
Fertilized															0.64	41	0.50	25	0.28	8	0.92	85
Nitrate nitrogen (petioles)																						
Non-fertilized																	0.09	1	0.17	3	0.77	59
Fertilized																	0.14	2	0.00	0	0.75	56
Phosphorus (petioles)																						
Non-fertilized																			0.89	79	0.62	38
Fertilized																			0.96	92	0.64	41
Potassium (petioles)																						
Non-fertilized																					0.57	32
Fertilized																					0.45	20

[1] The value of r at the 5% level approximates 0.16.

Table 11.—Correlation coefficients for eleven characters and percentages of the variances accounted for by regression, differences between populations, average of three treatments, 1958.[1]

	Thin juice										Petioles							
	Sucrose		Purity		Nitrogen		Glutamic acid		Betaine		Chlorides		Nitrate N		Phosphorus		Potassium		Sodium	
Character	r	r^2(100)	r	r^2(100)	r	r^2(100)	r	r^2(100)	r	r^2(100)	r	r^2(100)	r	r^2(100)	r	r^2(100)	r	r^2(100)	r	r^2(100)
Weight	0.12	1	0.31	10	—0.37	14	—0.15	2	—0.73	53	0.36	13	0.83	69	—0.38	14	0.71	50	0.59	35
Sucrose			0.68	46	—0.59	35	—0.57	32	0.00	0	—0.07	0	—0.04	0	0.66	44	0.12	1	—0.63	40
Purity					—0.96	92	—0.95	90	—0.66	44	0.09	1	0.46	21	0.59	35	0.56	31	—0.37	14
Nitrogen (thin juice)							0.97	94	0.74	55	—0.37	14	—0.61	37	—0.62	38	—0.75	56	0.17	3
Glutamic acid (thin juice)									0.64	41	—0.27	7	—0.46	21	—0.74	55	—0.62	38	0.32	10
Betaine (thin juice)											—0.42	18	—0.94	88	0.03	0	—0.89	79	—0.42	18
Chlorides (thin juice)													0.64	41	0.22	5	0.76	58	0.61	37
Nitrate N (petioles)															—0.11	1	0.96	92	0.65	42
Phosphorus (petioles)																	0.16	3	—0.61	37
Potassium (petioles)																			0.52	27

[1] The value of r at the 5% level approximates 0.12.

For percentage apparent purity the strongest associations are with betaine, potassium, and total nitrogen in the thin juice, regression accounting for 86, 81, and 56% of the variability on the fertilized plots and 61, 46, and 45% on the non-fertilized plots, respectively. With the possible exception of glutamic acid, the percentages of the variances accounted for by regression of purity and the other characters both on the non-fertilized and fertilized plots are comparatively small. The closest association between percentage apparent purity and any character taken from petiole samples is with potassium. The association is positive. The relation between potassium and percentage apparent purity in the thin juice was negative. These data definitely show that as regards the genetic variability the association between purity and potassium in the thin juice is the reverse of that in the petioles. The same conclusion can be drawn for weight of root and potassium in the thin juice and in the petioles with the exception that the relations are reversed; that is, the relations are positive in the thin juice and negative in the petioles or non-significant, the correlation coefficient being only —0.03 for the non-fertilized plots. It is clear that the relations found existing between potassium in the petioles and the three characters weight per root, percentage sucrose, and percentage apparent purity in the petioles cannot be taken as a measure of the relations between potassium in the thin juice and these same three characters.

The genetic correlation coefficients and percentages of the variances accounted for by regression for total nitrogen, glutamic acid, betaine, potassium (thin juice), sodium (thin juice), nitrate nitrogen, phosphorus, potassium (petioles) and sodium (petioles) are listed in Table 10 also. The data are for 1956.

Total nitrogen, glutamic acid, and betaine in the thin juice are positively associated with potassium in the thin juice on both the non-fertilized and fertilized plots and are negatively associated with potassium in the petioles. In the thin juice 79 and 90, 59 and 53, and 81 and 50 percents of the variances are accounted for by regression on the non-fertilized and fertilized plots, respectively, whereas in the petioles the values are 17 and 36, 20 and 27, and 1 and 7 percents, respectively. These relations can be accounted for if both total nitrogen and potassium are being accumulated in the roots at the expense of the tops. If such is the case one would expect a negative correlation between potassium in the thin juice and potassium in the petioles. There is no significant relation between potassium in the thin juice and potassium in the petioles on the non-fertilized plots but there is a relation between them on the fertilized plots and it is negative as expected.

Total nitrogen, glutamic acid, and betaine in the thin juice and sodium in both the thin juice and petioles are positively correlated with the exception of betaine and sodium in the petioles on the fertilized plots. The percents of the variances accounted for by regression are 21 and 30, 8 and 20, and 52 and 6 in the thin juice and 14 and 16, 3 and 20, and 32 and 0 in the petioles.

Total nitrogen in the thin juice and nitrate nitrogen in the petioles are positively correlated and the percents of the variances accounted for by regression are 31 and 36, respectively, for the non-fertilized and the fertilized plots. Phosphorus in the petioles is negatively associated with total nitrogen. This is true for both fertilizer treatments and the percentages of the variances accounted for by regression are 11 and 18%, respectively, for the non-fertilized and fertilized treatments.

All of the percentages of the variances accounted for by regression of total nitrogen on each of the other characters listed in Table 10 are significantly different from zero. This follows from the fact that the correlation coefficients are significantly different from zero.

On both the non-fertilized and the fertilized plots potassium in the thin juice is significantly associated with all the other characters with the exceptions of phosphorus and potassium in the petioles on the non-fertilized plots. Potassium in the thin juice is negatively associated with purity on both the fertilized and non-fertilized plots, with sucrose on the fertilized plots, and with phosphorus and potassium in the petioles on the fertilized plots. Sodium in the thin juice is positively associated with all other characters listed in Table 10 excepting sucrose and purity, the association being strongest for sodium in the thin juice and sodium in the petioles. This is in striking contrast with the behavior of potassium in the thin juice which shows a negative association with potassium in the petioles.

Nitrate nitrogen in Table 10 for the statistically significant values shows positive relations with all characters except purity and betaine on the fertilized plots, the association with purity being negligible. Also the associations with phosphorus and potassium in the petioles are negligible. Nitrate nitrogen in the petioles is rather closely associated with sodium in the petioles, 59 and 56% of the variances being accounted for by regression on the non-fertilized and fertilized plots, respectively.

Phosphorus and potassium in the petioles will be considered together as their relations with all the other characters in Table 10 are very similar. This is also true of the environmental relations shown in Table 9. Both of these chemical characters are

negatively associated with nitrogen in the thin juice, are negatively associated with potassium in the thin juice on the fertilized plots and show no significant relation with this character on the non-fertilized plots (see Table 10). Both are positively associated with sodium in the thin juice and in the petioles. Neither phosphorus nor potassium in the petioles show significant associations with nitrate nitrogen in the petioles. The closest relations of these two characters are with each other, 79 and 92% of the variances being accounted for by regression.

Both sodium in the thin juice and sodium in the petioles show significant positive associations with all other characters listed in Table 10, excepting sucrose and purity on the non-fertilized plots and excepting sodium in the petioles with betaine on the fertilized plots. Their behavior patterns are very similar and as would be expected, the closest associations are between sodium in the thin juice and sodium in the petioles, 88 and 85% of the variances being accounted for by regression. The next strongest association for sodium in the petioles is with nitrate nitrogen in the petioles.

The genetic correlation coefficients and the percentages of the variances accounted for bv regression for the eleven characters studied in 1958 are listed in Table 11.

Weight of root and percentage sucrose show very little association; only one percent of the variances being accounted for by regression. Also weisrht of root and percentage apparent purity are associated positively; but only 10% of the variances are accounted for by regression. The association between percentage sucrose and percentage apparent purity is positive and 46% of the genetic variances are accounted for by regression. This latter association is sufficiently close to aid materially in breeding sugar beets both high in sucrose and purity when it is found to occur.

The correlation coefficients and percentages of the variances accounted for by regression for weight per root, percentage sucrose, and percentage apparent purity with total nitrogen, glutamic acid, betaine, and chlorides in the thin juice; and nitrate nitrogen, phosphorus, potassium, and sodium in the petioles are listed in Table 11, also. All of the correlation coefficients and hence, percentages of the variances accounted for by regression involving weight per root. are significant at the 5% level. The associations with total nitrogen and the other nitrogenous compounds in the thin juice and phosphorus in the petioles are negative, whereas, those with chlorides in the thin iuice and nitrate nitrogen, potassium, and sodium in the petioles are positive. For betaine, nitrate nitrogen, and potassium with weight per root the associations are fairly close, regression account-

ing for 53, 69, and 50% of the genetic variances, respectively. Sucrose and purity are negatively associated with total nitrogen and the nitrogenous compounds in the thin juice and with sodium in the petioles. The associations with chlorides are not statistically significant. The same is true for the association of sucrose and betaine. The other associations with the exceptions of sucrose and nitrate nitrogen in the petioles are positive. However the correlation involving percentage sucrose and nitrate nitrogen is not significantly different from zero. All of the correlation coefficients with percentage apparent purity excepting the one with chlorides are significantly different from zero at the 5% level. The associations between purity and total nitrogen and glutamic acid are very close, 92 and 90% of the genetic variances being accounted for by regression. An examination of Table 4 shows that the high correlation coefficient is almost entirely due to the low purity and high total nitrogen of inbred 55-5307 as compared with the other populations. In none of the other relations of sucrose and purity with the chemical characters is as much as 50% of the genetic variance accounted for by regression.

The correlation coefficients and percentages of the variances accounted for by regression for the chemical characters other than sucrose for 1958 are listed in Table 11 also. Again these values are calculated from differences between populations and hence are genetic. Nitrogen in the thin juice is negatively associated with chlorides in the thin juice, nitrate nitrogen, phosphorus, and potassium in the petioles. The percentages of the variances accounted for by regression are 14, 37, 38, and 56, respectively. With the possible exceptions of nitrate nitrogen, potassium, and sodium, all in the petioles, the associations involving chlorides are not close. However, all are statistically significant excepting those with sucrose and purity. The percentage of the variance accounted for by the regression of nitrate nitrogen on phosphorus is negligible, on potassium is 92% and on sodium is 42%. The associations between the latter two and nitrate nitrogen are positive. The association between phosphorus and potassium is negligible and between phosphorus and sodium is negative, 37% of the genetic variability being accounted for by regression. The association between potassium and sodium is positive, 27% of the variability being accounted for by regression.

Combining Ability and Dates of Harvest Tests

Some results from combining ability and dates of harvest studies are of special interest in connection with the results previously presented in this article. The population genetic

studies conducted in 1956 provided data pertaining to whether populations differ in their ability to produce maximum yields together with maximum percentages of sucrose under the conditions of this experiment. Maximum weights, percentages sucrose, and corresponding percentages apparent purity, concentrations of total nitrogen in the thin juice and nitrate nitrogen in the petioles for populations A54-1, 50-406BB, and the F_1 (50-406 X 52-307) are listed in Table 12. These may be considered as combining ability tests of the top-cross and F_1 hybrid compared with the commercial variety A54-1. These data are from Powers et al. (21, Table 8).

To facilitate the interpretation of the data listed in Table 12 an explanation as to how these data were compiled is needed. In Table 8 of literature citation (21) there are 5 replication groups obtained by combining the data from 8 consecutive replications. Replications 1 to 8, inclusive, were grouped to produce the first replication group, 9 to 16 to produce the second replication group, and so forth on up to 33 to 40 to produce the 5th and last replication group. From the data in Table 8 of the article cited (21) it can be seen that some replication groups thus constructed differed materially in the parts per million of nitrate nitrogen in the petioles. The data of Table 8 were examined and the replication group having the maximum weight was determined and the data tor that replication group was tabulated in Table 12. Then, the replication group having the maximum percent sucrose was determined and the data for that replication group was also tabulated in Table 12. In the event that the maximum weight per root and the maximum percent sucrose occurred in the same replication group and in the same fertilizer treatment, then the data were taken from the replication group having the next highest percent sucrose, regardless of the treatment.

A study of Table 12 reveals the following:

Weight per root for A54-1 in the replication group having the greatest weight is 2.90 lb and is accompanied by 17.1% sucrose and occurs on the fertilized plots. The maximum percentage sucrose for any replication group is 18.4% and is accompanied by a root weight of 1.66 lb and occurs on the nonfertilized plots. For 50-406BB the maximum weight per root for any replication group is 2.24 lb and is accompanied by 18.1% sucrose which is also the maximum percent sucrose for this population. The next highest percent sucrose for this population is 18.0 and it is accompanied by a root weight of 1.43 lb. For the F_1 hybrid the maximum weight per root for any replication group is 2.49 lb and it is accompanied by 18.5% sucrose. This is also the maximum for percentage sucrose. The next highest percent

Table 12.—Maximums for weight per root, percentage sucrose and corresponding percentage apparent purity, concentrations of nitrogen in the thin juice, and concentrations of nitrate nitrogen in the petioles at time of harvest for populations A54-1, 50-406BB, and F_1 (50-406 X 52-307), 1956.

Population and treatment	Weight per root	Sucrose	Apparent purity	Nitrogen	Nitrate nitrogen
	lb	%	%	mg	ppm
A54-1					
Non-fertilized	1.66	18.4	97.0	12.6	452
Fertilized	2.90	17.1	94.2	38.6	1724
50-406BB					
Non-fertilized	1.43	18.0	97.5	10.8	406
Fertilized	2.24	18.1	95.3	25.7	1050
F_1(50-406 X 52-307)					
Non-fertilized	1.39	18.3	97.8	8.8	374
Fertilized	2.49	18.5	95.5	17.8	390
LSD at 5% level	0.12	0.3	0.4	3.2	817

sucrose of any replication group is 18.3 on the non-fertilized plots and is accompanied by a root weight of 1.39 lb. From these data it is evident that as regards weight per root and percentage sucrose A54-1 is responding differently to the high fertility level than are 50-406BB and the F_1 hybrid. That is, both of these hybrids have the maximum weight per root and maximum percentage sucrose occurring in the same replication group and occurring on the fertilized plots. This is not true of A54-1.

The F_1 produces as high percentage sucrose on the fertilized plots as does A54-1 on the non-fertilized plots and the weight per root of the F_1 on the fertilized plot is 50% greater than that of the F_1 on the non-fertilized plot. However, on the fertilized plots A54-1 outyields the F_1 hybrid in weight per root by 16%.

On the other hand maximum yields and maximum sucroses are not accompanied by the highest purities listed in Table 12. In every case the higher purities occur on the non-fertilized plots and the highest purity is for the F_1 hybrid. A study of the data in Table 12 reveals that the purities are closely associated with total nitrogen in the thin juice, and concentrations of nitrate nitrogen in the petioles, but with the latter to a smaller degree.

It is clear that the breeder can do much to improve both percentage sucrose and percentage apparent purity. The improvement in percentage sucrose will result from the fact that for certain hybrids, higher concentrations of nitrogen in the thin juice, up to certain limits, are not accompanied by reductions in sucrose content, as compared with other populations, and to the fact that certain hybrids have lower concentrations of total nitrogen in the thin juice, as compared with other populations, under the same fertilizer practices (see Tables 2 and 12). The

increase in percentage apparent purity is associated with the latter phenomenon; that is, some populations have less total nitrogen in the thin juice than other populations.

These findings raise the question as to whether genotypes (populations) might not differ as to the length of the growing season required to obtain acceptable percentages of sucrose and acceptable weights per root. In 1961 an experiment was conducted to determine whether such might be the case. Part of the data are tabulated in Table 13.

Table 13.—Means of weight per plot, percentage sucrose, and sugar per plot for three dates of harvest, 1961.

Population	Weight per plot			Percentage sucrose			Sugar per plot		
	Sept. 14	Oct. 3	Oct. 16	Sept. 14	Oct. 3	Oct. 16	Sept. 14	Oct. 3	Oct. 16
	kg	kg	kg	%	%	%	kg	kg	kg
52-430 × 54-565 F_1	4.45	5.02	4.94	15.5	16.0	17.3	0.6881	0.8018	0.8567
52-430 × 54-846 F_1	5.30	5.54	5.36	14.6	16.3	17.7	0.7982	0.9011	0.9528
52-430 × 52-408 F_1	7.11	7.48	7.49	14.4	15.5	16.7	1.0218	1.1618	1.2446
A56-3	7.54	8.02	7.97	12.5	14.2	15.2	0.9313	1.1290	1.2092
LSD at 5% level	0.59	0.77	0.81	0.6	0.6	0.5	0.0955	0.1195	0.1355

The data show that undoubtedly there are four levels of yield represented by the four populations. The F_1 hybrid 52-430 X 52-408 averages 6.2% lower weight per plot than does the commercial variety, A56-3. The yields of all populations increased from September 14 to October 3, but none of the population yields increased after October 3.

The data for percentage sucrose reveal that, when harvested September 14, all of the F_1 hybrids have from 1.9 to 3.0% more sucrose than does A56-3, the commercial variety with which they are compared. Moreover, all of the hybrids had higher percentage sucrose harvested on September 14 than did the commercial variety harvested on October 3 and the sucrose content of 52-430 X 54-565 was significantly higher than that of A56-3. In fact, this hybrid had as high a percentage sucrose content harvested on September 14 as did the commercial variety harvested on October 16. Likewise, all of the hybrids had higher percentages sucrose harvested on October 3 than did the commercial variety harvested on October 16. In the case of the first two hybrids listed, they averaged about 1% higher sucrose harvested on October 3 than did the commercial variety harvested on October 16. Finally, the F_1 hybrids harvested on October 16 had from 1.5 to 2.5% higher sucrose than did the commercial variety. It is clear that hybrid populations can be obtained that will have

as high a percentage sucrose as this commercial variety when harvested from two weeks to one month earlier.

A study of the data for yield of sugar per plot reveals that hybrid 52-430 X 52-408 produced more sugar per plot for all three dates of harvest than did the commercial variety, and the difference for September 14 approaches statistical significance. Moreover, this hybrid harvested on September 14 produced within 9% as much sugar per plot as did the commercial variety harvested on October 3, and within 15% as much sugar per plot as did the commercial variety harvested on October 16, approximately one month later. Hybrid 52-430 X 52-408 harvested on October 3 produced within 4% as much sugar as did the commercial variety harvested on October 16.

The fact that the hybrids studied in these researches do not have as great a weight per root as the commercial variety raises the problem whether the physiological relations between weight per root, percentage sucrose, and percentage apparent purity are such that hybrid populations cannot be obtained that will exceed the commercial variety in all three characters. Some information pertaining to a solution of this problem is provided by the data listed in Table 14. The F_1 hybrid exceeds the commercial variety, A54-1, in weight per root, percentage sucrose and percentage apparent purity. The differences are statistically significant at the 5% level. The question still remains as to what extent the plant breeder can increase these three characters simultaneously. It is apparent from these data that they can be increased simultaneously under the conditions which this experiment was conducted in 1960. An amount of nitrogen was applied in the spring of the year which it was hoped would result in optimum amounts of N being available for the production of near maximum yield of roots. That is, 100 lb of N and 250 lb of P_2O_5 were applied per acre. The 250 lb of P_2O_5 were applied in the previous autumn and plowed under.

Table 14.—Means and their standard errors for weight per root, percentage sucrose, and percentage apparent purity, 1960[1].

Population	Weight	Sucrose	Purity
	kg	%	%
A54-1	1.12±0.031	16.9±0.178	91.9±0.258
F_1 (52-430 × 52-307)	1.21±0.025	17.8±0.140	92.9±0.196
Difference	0.09±0.040	0.9±0.226	1.0±0.324

[1] The estimates of the standard errors include differences between replication means and therefore are over estimates.

Discussion and Conclusions

The discussion and conclusions will be divided into environmental variability, genetic variability, phenotypic-dominance phenomena, and combining ability and dates of harvest tests.

The characters studied of greatest agronomic importance are weight per root, percentage sucrose, and percentage apparent purity. The chemical determinations made on thin juice samples are total nitrogen, glutamic acid, betaine, potassium, sodium, and chloride. Total nitrogen, glutamic acid, betaine, and chloride are expressed as milligrams per 100 milliliters of thin juice equated to a refractometer reading of 10. In the thin juice, potassium and sodium are expressed as parts per 100,000. The determinations made from petiole samples are nitrate nitrogen, phosphorus, potassium, and sodium. They are expressed as parts per million.

Environmental Variability

The associations attributable to environmental variability will be considered first. It will be remembered that two years and five fertilizer treatments contributed much of the environmental variability. Weight per root is negatively associated with percentage sucrose and percentage apparent purity, 81 and 74% of the variability, respectively, of sucrose and purity being accounted for by that of weight per root. That is, on an average, the roots having the smaller weights tended to have the higher percentages sucrose and the higher percentages apparent purity.

As would be expected total nitrogen in the thin juice is positively associated with weight per root, 77% of the variability of one being accounted for by that of the other. However, after a certain concentration of nitrogen in the thin juice had been reached, no further increase in weight per root resulted. An increase from 18.8 mg of nitrogen per 100 ml of thin juice to 46.8 mg is associated with an increase in weight per root from 1.93 lb to 2.60 lb. An increase from 46.8 mg of nitrogen in the thin juice to 62.9 is accompanied by an increase in weight per root of only 0.39 lb and by decreases in percent sucrose and percent apparent purity of 2.2 and 4.0, respectively. Above 62.9 mg of nitrogen per 100 ml of thin juice, there is no further increase in weight per root. This is true even though a 300 lb application of nitrogen (in the form of ammonium nitrate) in the fertilizer increased the concentration of total nitrogen to 103.5 mg per 100 ml of thin juice. Then, in this experiment, the maximum weight per root was reached with a concentration of total nitrogen in the thin juice lying somewhere between 46.8 and 62.9 mg per 100 ml of thin juice equated to a refractometer

reading of 10. It should be kept in mind that two years are involved.

Weight per root is positively associated with nitrate nitrogen and sodium in the petioles and negatively associated with phosphorus and potassium in the petioles. The association is high for phosphorus and sodium, 90 and 96% respectively, of the variability of weight per root being accounted for by the variability of these two characters.

Percentage sucrose and percentage apparent purity will be considered together. These two characters are very closely associated as regards the environmental variability, 98% of the variability of one being accounted for by that of the other. The association is positive. That is, an increase in percentage sucrose is accompanied, on an average, by an increase in percentage apparent purity. The characters most closely associated with sucrose and purity are total nitrogen and betaine in the thin juice and phosphorus in the petioles, 94 and 98, 99 and 98, and 96 and 88 percents, respectively, of the environmental variabilities of percentage sucrose and percentage apparent purity being accounted for by regression. The associations are negative with total nitrogen and betaine and positive with phosphorus. In going from 18.8 mg of total nitrogen in the thin juice to 103.5, there is a decrease from 17.9% sucrose to 13.0% and a corresponding decrease in percentage purity from 96.1% to 82.7%. The corresponding changes in betaine are from 89.8 mg to 190.4 mg and for phosphorus are from 1798 ppm to 1238 ppm.

In relation to sucrose and apparent purity potassium in the petioles follows a behavior pattern very similar to that of phosphorus. However, the associations are not so strong.

Sodium in the petioles is highly associated with sucrose and purity, and the relations are negative. The relations are almost entirely due to differences between years and the differential behavior of sodium for the two years. In going from the nonfertilized plots in 1956 to the fertilized plots, sodium increased from 10940 ppm of sodium in the petioles to 15265 ppm; whereas, in 1958 in going from a 50 lb application of nitrogen in the fertilizer to a 300 lb application, sodium decreased in the petioles from 19430 ppm to 18115 ppm. Since, with increased applications of nitrogen in the fertilizer, sucrose and apparent purity percentages decrease both within and between years, the associations with sodium and these two characters are negative. These results emphasize the importance of studying the data in detail when interactions are occurring, such as noted for sodium, years, and treatments.

The interrelations of the chemical characters total nitrogen, glutamic acid, and betaine in the thin juice and nitrate nitrogen,

phosphorus, potassium, and sodium, all in the petioles, follow two behavior patterns. Total nitrogen, glutamic acid, and betaine in the thin juice, and nitrate nitrogen and sodium in the petioles are positively associated with each other and negatively associated with phosphorus and potassium in the petioles. Hence, as might be expected, phosphorus and potassium are positively associated with each other and negatively associated with total nitrogen, glutamic acid, betaine, nitrate nitrogen, and sodium. As has been shown, these same two behavior patterns exist as regards the relations of these elements with weight per root, percentage sucrose, and percentage apparent purity. Weight per root was positively associated with total nitrogen, glutamic acid, betaine, nitrate nitrogen, and sodium and negatively associated with phosphorus and potassium. The reverse was true of the relation between these same chemical constituents and sucrose and purity.

The data provide information as to the concentrations of nitrogen and betaine in the thin juice and concentrations of phosphorus in the petioles at which marked reductions in percentage sucrose and percentage apparent purity occur. Ulrich (28) gives critical nitrate levels estimated from analysis of petioles and blades for yields and for sucrose percentages. In our studies sharp reductions in percentage sucrose and percentage apparent purity occur in going from total nitrogen concentrations of 46.8 mg to 62.9 mg, from betaine concentrations of 116.6 mg to 154.9 mg, and from phosphorus concentrations of 1598 ppm to 1318 ppm.

When interpreting these findings it must be kept in mind that both percentage sucrose and percentage apparent purity are highly associated with each other and with each of the three chemical characters listed. Also it is equally important to keep in mind that the three chemicals are closely associated with each other. The associations are positive for sucrose, purity and phosphorus and negative for each of these three characters with total nitrogen and betaine. The association between nitrogen and betaine is positive. Hence, for the environmental variability, on an average, a change in concentration of any one of the chemicals (nitrogen, betaine, and phosphorus) would be expected to change the limits at which decided reductions in percentage sucrose and percentage apparent purity occur. Also it must be kept in mind that these values are not the same for all populations, as interactions involving genotype, environment, and location of the chemicals in the plant (thin juice or petioles) were found to be playing a part.

Genetic Variability

Finkner et al. (11) in studying genetic variability found that both aspartic acid and glutamine content can be increased or

decreased in the beet root according to direction of selection. Selection for low amino acid content was accompanied by increases in percentage sucrose and percentage apparent purity and increases in concentrations of raffinose, galactinol, and sodium. They concluded that selection for either aspartic acid or glutamine content in the beet root could be used to improve populations of sugar beets. However glutamine selection was slightly more effective in spreading the populations into separate groups.

The genetic variability for 1956 represents differences between populations on the fertilized plots. Weight of root and percentage sucrose are positively associated but only 29% of the variability of one is accounted for by that of the other. This positive relation is due to the fact that the two hybrid populations have both a greater weight per root and a higher percentage sucrose than the two inbreds. This shows that on the fertilized plots both higher weight per root and higher percentage sucrose can be obtained if certain populations are grown. However, A54-1 and A54-1BB have greater weights per root and lower percentages of sucrose than the two hybrid populations. Hence, the question still remains as to what extent both greater weight of root and higher percentage sucrose can be combined. The association between weight per root and percentage apparent purity was negligible. This indicates that by appropriate breeding procedures it should be possible to increase both weight of root and percentage apparent purity.

Percentage sucrose and percentage apparent purity, as is true for the environmental variability, are again rather highly associated positively. Fifty-three percent of the genetic variability of one was associated with that of the other. Since as percentage sucrose increased there was, on the average, an increase in apparent purity, the task of increasing both of these characters by breeding would be easier than if no relation existed.

In studying the genetic variability it was found that total nitrogen in the thin juice is negatively associated with both percentage sucrose and apparent purity. There is a decided interaction between genotype and fertilizer treatment for percentage sucrose. This is shown by comparing A54-1 and the F_1 on both fertilizer treatments. For A54-1 there was a reduction of 1.1% in sucrose on the fertilized plots compared with the non-fertilized plots, whereas the percentage sucrose was the same for the F_1 hybrid on both fertilizer treatments. A comparison of these same populations as regards total nitrogen in the thin juice and percentage sucrose is informative. For A54-1 the percents sucrose on the non-fertilized plots and the fertilized plots were 17.9 and 16.8 accompanied by concentrations of nitrogen in the thin juice of 18.8 and 46.8. The same relations for the F_1 are sucrose 17.6

and 17.6 and nitrogen 9.8 and 21.3. It should be noted that the F_1 on the fertilized plots does not have a materially greater concentration of total nitrogen in the thin juice, than does A54-1 on the non-fertilized plots, the comparison being 21.3 to 18.8. Neither are the percents sucrose materially different, 17.6 compared with 17.9. This would indicate that these two genotypes under the environmental conditions of this experiment react similarly to total nitrogen in the thin juice. However, A54-1 has considerably more total nitrogen in the thin juice than does the F_1 and 52-307 on the fertilized plots. Hence, populations differ as to total nitrogen in the thin juice under identical fertilizer treatments. These same conclusions hold for glutamic acid and betaine but they are not so marked for betaine.

A comparison of 50-406BB and its female parent 50-406 shows that populations do not always react the same to total nitrogen in the thin juice as regards percentage sucrose. On the non-fertilized and fertilized plots the percents sucrose for 50-406BB are 17.6 and 17.3, respectively, and the corresponding values for 50-406 are 17.4 and 16.1. On the non-fertilized and fertilized plots the concentrations of total nitrogen for 50-406BB are 12.6 and 33.6, respectively, and the corresponding values for 50-406 are 14.6 and 31.2. This definitely represents a genotype-environment interaction, as an increase in milligrams of total nitrogen per 100 ml of thin juice to 33.6 for 50-406BB was accompanied by only a decrease in sucrose of 0.3%, whereas, for 50-406 a corresponding increase to 31.2 was accompanied by a decrease in sucrose of 1.3%. Obviously a higher concentration of total nitrogen in the thin juice is accompanied by a considerably smaller decrease in percentage sucrose for 50-406BB than is the case for 50-406. Stating it another way, at about the same concentration of total nitrogen in the thin juice, 33.6 and 31.2 mg per 100 ml of thin juice, 50-406BB has 17.3% sucrose as compared with 16.1% for 50-406. The corresponding values involving; percentage apparent purity rather than percentage sucrose are 94.4 and 92.6. Hence, not only do some populations have smaller concentrations of total nitrogen in the thin juice under identical fertilizer treatments but some populations have a higher percentage sucrose and higher percentage apparent purity than others at the same level of total nitrogen concentration in the thin juice. These conclusions hold for glutamic acid but not for betaine on the fertilized plots. However, they do hold for betaine and apparent purity on the non-fertilized plots. These findings warrant detailed discussion of the data as regards the interrelation of chemical characters and their association with percentage sucrose and percentage apparent purity.

The data in Tables 2 and 4 show that some populations have lower concentrations of total nitrogen, glutamic acid and betaine in the thin juice and that these lower concentrations are accompanied by increases in percentage sucrose and percentage apparent purity. It was found that, on the fertilized plots, the F_1 as compared with the commercial variety has lower concentrations of total nitrogen, glutamic acid, and betaine in the thin juice. These lower concentrations are accompanied by an increase of 0.8% in sucrose and 2.1% in apparent purity. In 1958 for an average of all three fertility levels inbred 55-5307 averaged considerably higher than the F_1 in concentrations of total nitrogen, glutamic acid and betaine. The lower concentrations of these three chemicals in the F_1 were accompanied by increases of 1.4% in sucrose and 5.1% in apparent purity. The same was true of comparisons involving the F_1 and A54-1 on the fertilized plots in 1956. However the accompanying increases in percent sucrose and apparent purity are not so large.

For 1956 on the fertilized plots 50-406BB as compared with 50-406 had about the same concentration of total nitrogen, was materially higher in glutamic acid and materially lower in betaine. This reduction in the concentration of betaine was accompanied by an increase of 1.2% in sucrose and 1.8% in apparent purity.

It was found that 52-430 compared with A54-1 at the 300 lb application of nitrogen has lower concentrations of total nitrogen and glutamic acid in the thin juice and a higher concentration of betaine. These lower concentrations of total nitrogen and glutamic acid in the thin juice are accompanied by increases of 0.4% sucrose and 2.3% apparent purity. A comparison of the F_1 and 50-406 for 1956 shows that the former has lower concentrations of total nitrogen and betaine in the thin juice and does not differ materially as regards concentrations of glutamic acid. The lower concentrations of total nitrogen and betaine are accompanied by increases of 1.5% in sucrose and of 2.7% in purity. Hence total nitrogen, as might be expected, is associated with both glutamic acid and betaine in giving favorable or unfavorable reactions as to percentages sucrose and apparent purity.

So far lower concentrations in the cases cited have resulted in benefits in both higher sucrose and higher apparent purities. However, such is not necessarily always true, as the following examples show. A comparison of the means for populations A54-1 and 52-430 showed that for the average of the three fertility levels in 1958 decreases in the concentrations of total nitrogen and glutamic acid and an increase in concentration of betaine were accompanied by an increase of one percent in apparent purity but that the percent sucrose was the same for both populations. Population 52-430 is not significantly different from A56-

5BB in percentage apparent purity and concentrations of total nitrogen and glutamic acid but has 1.1% more sucrose and a considerably higher concentration of betaine. Finally, population A54-1BB is not significantly different from A54-1 in weight per root, percentage sucrose, percentage apparent purity, and concentrations of total nitrogen and betaine in the thin juice, but it has only about one half as much glutamic acid.

From the above study of genetic differences (comparisons between populations) it is apparent that the greatest increases in percentage sucrose and in percentage apparent purity are found in those genotypes which have lower concentrations of total nitrogen, glutamic acid, and betaine as compared with genotypes which have higher concentrations of these three chemicals. It is equally clear that sometimes increases in percentage sucrose and percentage apparent purity are associated with decreases in concentrations of glutamic acid even though there has been an increase in betaine. In other comparisons increases in percentage sucrose and percentage apparent purity are accompanied by decreases in betaine even though there has been an increase in glutamic acid. Finally, whenever there have been increases in percentage sucrose and percentage apparent purity accompanied by a decrease in total nitrogen there has also been a decrease in either glutamic acid or betaine. or both glutamic acid and betaine. This might be expected since both glutamic acid and betaine are nitrogenous compounds. However, increases in percentage sucrose and percentage purity may be accompanied by decreases of either glutamic acid or betaine without there being a decrease in total nitrogen. Finally, increases in percentage sucrose are not necessarily accompanied by increases in percentage apparent purity, nor are increases in apparent purity necessarily accompanied by increases in percentage sucrose.

These findings show that for some genotypes increases in both sucrose and purity as compared with other genotypes are associated with the lower concentrations of total nitrogen and the nitrogenous compounds in the thin juice when all are grown at the same fertility level. Also some genotypes as compared with other genotypes have higher percentage sucrose and percentage apparent purity even though the concentrations of total nitrogen and one or more of the nitrogenous compounds in the thin mice may be higher. Hence genetic improvement of sucrose and puritv may be brought about by either breeding populations having higher percentages of sucrose and apparent purity associated with lower concentrations of total nitrogen and the nitrogenous compounds in the thin juice or by breeding genotypes which have higher percentages of sucrose and apparent purity even though the concentrations of total nitrogen and nitrogenous compounds are

comparatively high. In these studies the greatest gains in percentage sucrose and percentage apparent purity were obtained in those populations having lower concentrations of total nitrogen and the nitrogenous compounds in the thin juice and for which there was a reduction in all. However gains in percentage sucrose were obtained in those genotypes having a reduction in one or the other of the nitrogenous compounds. The greatest gains in percentage sucrose and percentage apparent purity will be for those genotypes which possess both lower concentrations of nitrogen in the thin juice and possess the ability to produce higher percentages of sucrose and apparent purity even at higher levels of concentration of nitrogen and nitrogenous compounds in the thin juice. Also, these latter genotypes are expected to be adapted over a greater environmental range.

Further these studies show that genotypes can be bred which have higher percentages sucrose but do not have higher percentages purity and vice versa. However, the greatest improvement in quality will result from breeding genotypes that are both high in percentage sucrose and percentage apparent purity.

Another interesting association is that involving total nitrogen in the thin juice and nitrate nitrogen in the petioles on the fertilized plots. The populations that will be considered are A54-1, 50-406, F_1, and 52-307. A54-1 is highest in total nitrogen in the thin iuice (46.8) and highest in nitrate nitrogen in the petioles (4233), 50-406 is next highest in total nitrogen in the thin juice (31.2) and lowest in nitrate nitrogen in the petioles (2297), the F_1 is second lowest in total nitrogen in the thin juice (21.3) and intermediate in nitrate nitrogen in the petioles (3060) and finally, 52-307 is lowest in total nitrogen in the thin juice (18.6) and second highest in nitrate nitrogen in the petioles (3644). That these data do not represent an exceptional case is shown by comparing populations A56-5BB, 52-430, and 55-5307 grown in 1958. A56-5BB has 71.2 mg of total nitrogen per 100 ml of thin juice, whereas, the corresponding concentration of nitrate nitrogen in the petioles is 6240, 52-430 has 73.8 mg of total nitrogen per 100 ml of thin juice, whereas, the corresponding concentration of nitrate nitrogen in the petioles is 3080, and finally 55-5307 has 116.4 mg of total nitrogen per 100 ml of thin juice; whereas, the corresponding concentration of nitrate nitrogen in the petioles is 2865. Of considerable importance is the fact that the percentages of sucrose and percentages of apparent purity are very closely associated with total nitrogen in the thin iuice and not necessarily with nitrate nitrogen in the petioles. This conclusion holds for both the environmental variability and the genetic variability.

These data show that, as regards the genetic variability, high nitrate nitrogen in the petioles is not necessarily associated with high total nitrogen in the thin juice. Nor is high nitrate nitrogen in the petioles necessarily associated with low total nitrogen in the thin juice. Also the same was found to hold for the environmental variability, but the associations were closer between degrees of concentration of total nitrogen in the thin juice and concentrations of nitrate nitrogen in the petioles. That is, the exceptions to an increase in one being accompanied by an increase in the other were fewer for the environmental variability than for the genetic variability. It is clear that relative concentrations of nitrate nitrogen in the petioles cannot always be taken as indicative of relative concentrations of total nitrogen in the thin juice. Further, it was found that relative concentrations of potassium in the petioles cannot always be taken as an indication of relative concentrations of potassium in the thin juice. For example, 52-307 had 24690 ppm of potassium in the petioles and only 99.4 parts per 100,000 of potassium in the thin juice; whereas, A54-1 had only 17695 ppm of potassium in the petioles and 133.1 parts per 100,000 in the thin juice. This genetically controlled decrease of 133.1 to 99.4 parts per 100,000 of potassium in the thin juice was accompanied by an increase of 1.7% in purity.

Before leaving the associations noted between characters due to environmental variability and those noted due to genetic variability, patterns of behavior common to both should be considered. It was found that nitrate nitrogen and sodium in the petioles followed one behavior pattern as regards their associations with other characters and phosphorus and potassium in the petioles another behavior pattern. In brief, nitrate nitrogen and sodium in the petioles are positively associated with each other and negatively associated with phosphorus and potassium in the petioles. Likewise, phosphorus and potassium in the petioles are positively associated with each other and negatively associated with nitrate nitrogen and sodium.

These associations are apparently related, at least in part, to interionic effects in the soil on nutrient absorption by the plant. Steward (25) has pointed out that absorption of one ion is affected by other ions; the general principal is that A) the entry of an ion will be aided by an ion of opposite sign having similar or greater mobility in water or in the cell membranes, and B) the entry of an ion may compete with the absorption of another ion of similar sign. In the field experiments reported here, increasing fertilizer rates would increase nitrate uptake by the

plant and tend to increase the rate of absorption of a mobile cation. In the 1956 experiment the absorption of sodium was enhanced rather than potassium. This might be expected since Sutcliffe (26) has reported red beet tissue shows preferential absorption of sodium over potassium when the ions are present in equivalent amounts. Conversely, the absorption of nitrate and phosphate ions would tend to be competitive and would explain the negative association between these ions. Arnon (1) has reported that the absorption of the phosphate ion may be depressed by the presence in the nutrient medium of high concentrations of rapidly absorbable nitrate ions. It must be recognized, however, that interionic effects in the nutrient medium is but one of several factors which affect mineral absorption by plants; in a different environment other factors may exert a greater influence on nutrient uptake and mask the apparent interionic effects noted in these experiments.

Finally, the associations noted for total nitrogen in the thin juice of the sugar beet root and potassium in the thin juice and phosphorus and potassium in the petioles are those expected if phosphorus and potassium are involved in the metabolic and translocation processes that result in increased amounts of total nitrogen in the thin juice. This postulation is supported by the negative relation between potassium in the thin juice and potassium in the petioles on the fertilized plots. A comprehensive review of the literature and conclusions pertaining to translocation in plants is given by Crafts (5). Also, Arnon (1) presents an excellent discussion of the translocation of phosphorus in plants.

From a study of the means in Tables 2 to 5, inclusive, it is apparent that all of the 13 characters listed possess both genetic and environmental variability. This is important as the environment can be controlled to some extent by fertilizer and cultural practices and the genotype can be controlled to a considerable extent by breeding. It is also clear that there are interactions between the genotypes and the environments; that is, all genotypes are not reacting the same to all environments. Hence, to obtain high yields of sugar per acre and to obtain beets high in processing quality the genotype and environment, and the genotype-environment interactions must be taken into account. A study of the means makes it clear that genotype-environment interactions are of very considerable importance in determining the advances that can be made by the plant breeder.

The fact that, as regards both the environmental and genetic variabilities, percentage apparent purity is negatively associated with all other characters in the thin juice with the possible exception of chlorides and that predominantly the same is true of

percentage sucrose with the exception of purity, warrants further discussion of the genetic implications of these findings. First, it is evident that since the characters differ and since there are a number of different genotype environment interactions, these characters must differ also in some of the genes controlling their differentiation and production. Hence the least number of genes differentiating and controlling differences in percentages of sucrose and differences in percentages of purity would be the number of such characters studied in the thin juice samples.

For the genetic variability, considering both years, the number of different characters studied in thin juice samples is 7. Probably the number should be increased to 8 as percentage sucrose and percentage apparent purity are somewhat closely associated in both years as regards the genetic variability. This does not mean that in any given segregating population the least number of genes differentiating any given population is 8. For example, hybrid populations derived by hybridizing closely related inbreds could be segregating for only one or a few of the genes differentiating one of the chemical characters. Going to the other extreme, segregating populations derived from very diverse genetic material would be expected to have many more than eight gene pairs differentiating percentage sucrose and differentiating percentage apparent purity. Undoubtedly genetic linkages both favorable and unfavorable to the recombination of genes tending to produce higher percentages of sucrose and to the recombination of genes tending to produce higher percentages of purity are occurring in the transmission of genes in these segregating populations derived from hybridization of extremely diverse genetic material.

The progress that can be made in percentage sucrose and percentage apparent purity is important. These data do not provide information on the exact advances that can be made, but they do provide information from which generalizations can be drawn. From the discussion given in the preceding paragraph and from a review of the literature it is apoarent that numerous genes are involved in the control and differentiation of both percentage sucrose and percentage apparent purity in the genus *Beta.* A number of sources of inter-fertile genetic and breeding material are available. For example, stock beets, garden beets, swiss chard, and some species such as *Beta maritima* hybridize readily with sugar beets *(Beta vulgaris)* and the offspring are fertile. These populations undoubtedly differ in the chemical characters studied in this article and in other chemical characters not studied which are associated with both percentage sucrose and percentage apparent purity.

These studies show that the genes conditioning and differentiating those chemical characters studied can be recombined, and hence desirable combinations of characters favorable to the production of higher percentage sucrose and higher percentage apparent purity can be obtained. Since so many genes and so many characters are involved progress may be expected to be gradual. However, on the other hand, because there are many genes and characters associated with quality it seems certain that considerable progress can be made in breeding populations of sugar beets adapted to production at higher levels of soil fertility and adapted to other climatic and cultural conditions. In fact such populations would be expected to have wide adaptability as regards producing satisfactory percentages of sucrose and apparent purity.

Such being the case it seems highly improbable that the populations being grown commercially today have obtained the maximum possible as regards either percentage sucrose or percentage apparent purity. Tt is still more improbable that the populations being grown commercially today have obtained the maximum possible in recombining high weight per root, high percentage sucrose, and high percentage apparent purity. Some of the researches promising the greatest remuneration to the beet sugar industry involve fundamental studies on the genetics of and methods of breeding for these three characters.

In any breeding program criteria for selection are important and the expense of the breeding program can be reduced materially if the number of characters used as criteria for evaluating individual plants, populations, inbreds, etc., can be reduced. The closeness of the association between percentage sucrose and percentage apparent purity with each other and with total nitrogen and betaine in the thin juice indicates that perhaps either of the latter would be a rather effective criterion for use in breeding populations having higher percentages of sucrose and higher percentages of apparent purity. Of course it would be still better to use all four characters as criteria for evaluating material to be used in breeding programs for improving percentage sucrose and percentage apparent purity of populations grown for the production of beet sugar. If only one criterion is employed the determination should be for percentage sucrose or percentage purity depending on the character for which improvement is sought. If both characters are being bred for simultaneously the determinations for both characters should be made.

Phenotypic-Dominance Phenomena

Phenotypic-dominance phenomena can be determined for weight per root, percentage sucrose, percentage apparent purity and for the concentrations of the chemicals determined from an

analysis of the thin juice and those determined from an analysis of the petioles.

The F_1 hybrid grown in 1956 exhibits heterosis for weight per root, percentage sucrose, and percentage apparent purity. Heterosis for weight per root was expected. That is, the F_1 was expected to possess hybrid vigor. That the F_1 would also exceed either parent in both percentage sucrose and percentage apparent purity was not expected, and undoubtedly is true of only certain hybrids. In the thin juice, total nitrogen, betaine, potassium and sodium ranged in expression of the character from minus partial dominance to minus heterosis. Only glutamic acid exhibited plus dominance and this was for the fertilized plots.

Since in general a decrease in all of those chemical characters in the thin juice, both as regards environmental and genetic-variability, tends to be associated with an increase in both percentage sucrose and percentage apparent purity, it is apparent that the dominance phenomena exhibited by the F_1 for total nitrogen, betaine, potassium, and sodium in the thin juice are favorable. They would be conducive to expression of heterosis for higher sucrose and higher purity as actually was found to be the case for this F_1. The plus dominance noted for glutamic acid would tend to offset, somewhat, these favorable dominance reactions noted for the other chemical characters measured in the thin juice.

It is interesting to note that nitrate nitrogen in the petioles on the fertilized plots is intermediate, but that all the other chemical characters measured in the petioles exhibit minus partial dominance. It is apparent that, in general, the dominance phenomena shown by the chemical characters studied for this hybrid on the fertilized plots are favorable to both high percentage purity and to high percentage sucrose. This would indicate that by employing those methods of breeding designed to utilize heterosis, hybrid populations can be bred that are superior in weight per root, percentage sucrose, and percentage apparent purity to those varieties now grown for the manufacture of beet sugar.

Heterosis, dominance, partial dominance, and intermediate dominance are different degrees of expression of a given character due to physiological-genetic reactions and interactions (Powers 18). That the expression of dominance is dependent upon both the genotype and environment has been demonstrated by a number of workers (Goldschmidt 12); and that dominance may be shifted to heterosis or vice versa, by varying either the genotype or the environment has been shown by Powers (17). The data in Tables 6 and 7 substantiate these deductions. Then in summary it may be said that heterosis and dominance are different

degrees of expression of the same physiological-genetic phenomena and are dependent upon both the genotype and environment and upon the interactions within and between them. Also these studies emphasize the importance of the chemical characters and their interrelations in determining the phenotypic-dominance reactions of weight per root, percentage sucrose, and percentage apparent purity. In turn these findings have a very practical application in breeding superior populations for use in the production of beet sugar.

Combining Ability and Dates of Harvest Tests

It was found from a study of the 1956 data that the two hybrid populations compared with a commercial variety, A54-1, showed no decrease in percentage sucrose or very little decrease grown on the fertilized plots, as compared with the non-fertilized. A54-1, however, showed a decrease in percentage sucrose when grown on the fertilized plots. The replication groups in this experiment varied considerably in nitrogen fertility level. This was shown by concentrations of nitrate nitrogen in the petioles. For A54-1 (see 21, Table 8), the replication groups varied from 829 parts per million of nitrate nitrogen in the petioles to 13,778 on the fertilized plots. The range was similar for the other populations. The range for A-54-1 on the non-fertilized plots was from 452 parts per million of nitrate nitrogen in the petioles to 6055. Such being the situation, an opportunity was provided to determine whether some populations, as regards these replication groups, are able to reach, simultaneously, the maximum mean sucrose content and maximum mean weight per root, 'whereas other populations are not able to do so. The data for populations A54-1, 50-406BB, and the F_1 (50-406 X 52-307) are taken from Table 12.

The maximum mean weight per root of A54-1 for any of the replication groups was 2.90 lb and the corresponding mean sucrose content was 17.1%. The maximum sucrose content was 18.4% and the corresponding weight per root was 1.66 lb. Hence, maximum weight per root is accompanied by a comparative decrease in percentage sucrose.

For 50-406BB, the maximum mean weight per root for any of the replication groups was 2.24 lb and the mean sucrose content was 18.1%. The maximum mean percent sucrose content was 18.0 on the non-fertilized plots and the mean weight per root was 1.43 lb. It is apparent that for this top-cross hybrid and under the conditions of this experiment, maximum mean weight per root and maximum mean percentage sucrose occur together. It will be recalled that this top-cross, hybrid 50-406BB, had a higher concentration of total nitrogen in the thin juice as compared with the inbred parent, but that this higher concentration

was not accompanied by a material reduction in percentage sucrose.

The maximum weight per root for the F_1 hybrid was 2.49 lb and the corresponding percent sucrose for this replication group was 18.5, which was also the maximum percent sucrose of any of the replication groups either on the fertilized or non-fertilized plots. The maximum percent sucrose on the non-fertilized plots was 18.3 and the corresponding weight per root was 1.89 lb. Hence, the maximum weight per root and the maximum percentage sucrose occur in the same replication group on the fertilized plots. It will be recalled that the F_1 hybrid had a total nitrogen concentration of only 21.3 mg per 100 ml of thin juice as compared with a concentration of 46.8 for A54-1 grown under the same fertilizer treatment (fertilized plots).

It is clear that the breeder can do much to improve both percentage sucrose and percentage apparent purity. The improvement in percentage sucrose will result from the fact that for certain hybrids, higher concentrations of nitrogen in the thin juice up to certain limits are not accompanied by a reduction in sucrose content and to the fact, that certain hybrids have lower concentrations of total nitrogen in the thin juice as compared with other populations under the same fertilizer practices. The same is true for percentage apparent purity. However the increase in percentage apparent purity is largely associated with the latter phenomenon; that is, some populations have less total nitrogen in the thin juice than other populations.

These findings raise the question as to whether populations (genotypes) might not differ as to the length of the growing season required to obtain acceptable percentages of sucrose and weight per root. In 1961, an experiment was conducted to determine whether such might be the case. The data are taken from Table 13.

The data on weight per root show that, undoubtedly, there are four levels of yield represented by the four populations. The F_1 hybrid 52-430 X 52-408 averages 6.2% lower weight per plot than does the commercial variety. The yields of all populations increased from September 14 to October 3 but none of the population yields increased after October 3.

The data for percentage sucrose reveal that, when harvested September 14, all of the F_1 hybrids have from 1.9 to 3.0% more sucrose than does A56-3, the commercial variety with which they are compared. Moreover, all of the hybrids had higher percentage sucrose harvested on September 14, than did the commercial variety harvested on October 3 and the sucrose content of 52-430 X 54-565 was significantly higher than that of A56-3. In fact, this hybrid had as high a percentage sucrose content harvested

on September 14 as did the commercial variety harvested on October 16. Likewise, all of the hybrids had higher percentages of sucrose content harvested on October 3 than did the commercial variety harvested on October 16. In the case of the first two hybrids listed, they averaged about one percent higher sucrose harvested on October 3 than did the commercial variety harvested on October 16. Finally, the F_1 hybrids harvested on October 16 had from 1.5% to 2.5% higher sucrose than did the commercial variety. It is clear that hybrid populations can be obtained that will have as high a percentage sucrose as this commercial variety when harvested from two weeks to one month earlier.

A study of the data for yield of sugar per plot reveals that hybrid 52-430 X 52-408 produces more sugar per plot for all 3 dates of harvest than does the commercial variety and the difference for September 14 approaches statistical significance. Moreover, this hybrid harvested on September 14 produced within 9% as much sugar per plot as did the commercial variety harvested on October 3 and within 15% as much sugar per plot as did the commercial variety harvested on October 16, approximately one month later. Hybrid 52-430 X 52-408 harvested on October 3 produced within 4% as much sugar as did the commercial variety harvested on October 16.

The importance of these responses of F_1 hybrids lies in the fact that by growing them, the beet sugar factories can be operated over a long period of time. By longer operation of the factories fewer beets would be piled and storage losses would be reduced materially. The production of hybrids that can be harvested from a month to two weeks earlier would also reduce, considerably, the expense of harvesting beets, as by growing such hybrids the farmer would have more choice of the conditions under which the harvest would be conducted. Consequently, unfavorable weather conditions could be avoided, particularly those occurring late in the fall.

The fact that the hybrids studied in these researches do not have as great a weight per root as the commercial variety raised the problem whether the physiological relations between weight per root, percentage sucrose, and percentage apparent purity are such that hybrid populations cannot be obtained that will exceed the commercial variety in all three characters.

In 1960, an experiment was conducted that provided information as to such a physiological possibility. A54-1 and the F_1 hybrid 52-430 X 52-307 were grown in this study. It was found that the F_1 hybrid exceeds the commercial variety in weight per root, percentage sucrose, and percentage apparent purity. This does not prove that this F_1 hybrid will be superior

in all three of these important agronomic characters under all environmental conditions but it does prove that increases in all three of these characters can be obtained simultaneously. This experiment was conducted on plots all of which had received an application of 100 lb of N and 250 lb of P_2O_5.

These results could have been predicted from the chemical-genetic studies summarized in this article. That is, the genotype-environmental interactions, the associations of characters as regards both the environmental and genetic variability, and the phenotypic-dominance phenomena are more favorable on the average for certain hybrid populations.

One of the more important findings from these researches is that certain hybrid populations are more likely to have higher percentage sucrose and higher percentage apparent purity over a wider range of environmental conditions than are commercial varieties. That is, they would be expected to perform more favorably over a period of years, over diverse climatic conditions represented by locations, and under different fertilizer and other cultural practices. This is indicated by the following findings: First, there is a close negative association between total nitrogen in the thin juice and the nitrogenous compounds and both higher percentage sucrose and higher percentage apparent purity. Second, some genotypes (populations) have higher percentage sucrose associated with higher levels of total nitrogen in the thin juice than do some of our commercial varieties. Finally, some F_1 hybrids under conditions conducive to high nitrogen in the thin juice do not have as high a concentration of nitrogen in the thin juice as some of the commercial varieties. Other chemical characters showed similar behavior patterns.

Also of considerable importance is the finding that some genotypes, comparatively, may have high concentrations of nitrate nitrogen in the petioles and low concentrations of total nitrogen in the thin juice. The same was found to be true for concentrations of potassium. That is, some genotypes, comparatively, had high concentrations of potassium in the petioles and low concentrations of potassium in the thin juice. This has added interest when considered in the light of the findings of Ulrich (27). He states that "In order to obtain maximum sugar formation, a large supply of nitrogen must be available continuously early in the season. The available nitrogen must be utilized completely at the time of harvest, otherwise beets of a relatively low sugar-percentage will be obtained. By carefully controlling the nitrogen supply, beets both high in yield and in sugar percentage may be grown." Our results indicate that we might add to this statement of Ulrich's that by genetically controlling the location (roots or tops) of the higher concentrations of the chemical con-

stituents and chemical compounds found to be undesirably associated in the thin juice (in our studies nitrogen and potassium), beets high in yield, percentage sucrose, percentage apparent purity, and processing quality may be grown.

These findings led to the postulation that some F_1 hybrids would have higher percentage sucrose at different dates of harvest than the commercial variety and such was found to be the case. One hybrid harvested on September 14 had 0.3% higher sucrose than did the commercial variety on October 16. These results show that by growing certain hybrids the number of poor quality years would become less frequent. Moreover, when they did occur quality would not be as poor as it would have been if the old commercial varieties had been grown. Equally, if not more important is the fact that the hybrids would be expected to yield higher quality beets over a wider range of soil and climatic conditions.

Summary

1. The most important agronomic characters studied are weight per root, percentage sucrose, and percentage apparent purity. Characters studied in the thin juice are total nitrogen, glutamic acid, betaine, potassium, sodium, and chlorides. Characters studied in the petioles are nitrate nitrogen, phosphorus, potassium, and sodium. These make a total of 13 characters.

2. Primarily, this article is concerned with the interrelations of the characters as determined by a study of the means and simple correlation coefficients, with phenotypic-dominance phenomena and with dates of harvest and combining ability tests.

3. The variabilities studied were those attributable to environmental differences and those attributable to genetic differences.

4. As regards the environmental variability weight per root, percentage sucrose and percentage apparent purity are very closely associated with each other; and with total nitrogen, glutamic acid, and betaine in the thin juice and with phosphorus and sodium in the petioles. Weight per root is negatively associated with sucrose, purity, phosphorus, and potassium in the petioles. It is positively associated with total nitrogen, glutamic acid, and betaine in the thin juice and nitrate nitrogen and sodium in the petioles. Sucrose and purity are negatively associated with those characters with which weight per root is positively associated and are positively associated with those characters with which weight per root is negatively associated.

5. As regards the environmental variability total nitrogen, glutamic acid, and betaine in the thin juice and nitrate nitrogen and sodium in the petioles are closely associated with each other. The associations are positive. They are negatively associated with

phosphorus and potassium in the petioles, the association with phosphorus being close.

6. Phosphorus and potassium in the petioles are positively associated with each other. However, the association is not close, only 6% of the environmental variability of one being accounted for by that of the other.

7. For the environmental variability nitrate nitrogen and sodium in the petioles were found to form one behavior pattern as regards their associations with all other characters and with each other, and phosphorus and potassium in the petioles another behavior pattern. All of the characters with which nitrate nitrogen and sodium are associated positively, phosphorus and potassium are associated with negatively. Further, all the characters with which nitrate nitrogen and sodium are associated with negatively, phosphorus and potassium are associated with positively.

8. For 1956 data, the genetic variability was studied on the basis of the two fertilizer treatments, non-fertilized and fertilized. With a few exceptions, the characters are not nearly so closely associated as they are for the environmental variability. The only somewhat close associations for weight per root occur on the non-fertilized plots and are with percentage sucrose and with nitrate nitrogen in the petioles. The relation is positive. The only very close association for sucrose is with betaine on the fertilized plots and the relation is negative. The only very close associations for percentage apparent purity are with betaine and potassium in the thin juice. The relations are negative and are for the fertilized plots. The associations between purity and sucrose, and purity and total nitrogen in the thin juice are also somewhat close, being positive in the first case and negative in the second. Also, it should be noted that purity is negatively associated with all the other five characters of the thin juice.

9. The genetic associations between total nitrogen in the thin juice, glutamic acid, and potassium in the thin juice are very close on both the fertilized and non-fertilized plots. The relations are positive.

10. As regards genetic associations with other characters, potassium and sodium in the thin juice for 1956 follow very similar behavior patterns, excepting the associations with phosphorus and potassium in the petioles, and therefore will be summarized together. The only significant negative associations except as noted below are with purity on both the non-fertilized and fertilized plots and with percentage sucrose on the fertilized plots. The associations of potassium in the thin juice with phosphorus and potassium in the petioles on the fertilized plots are negative, whereas the associations of sodium in the thin juice with these same characters are positive. It is interesting to note that po-

tassium in the thin juice is negatively associated with potassium in the petioles, whereas, sodium in the thin juice is very closely associated with sodium in the petioles and the relation is positive.

11. As was noted for the environmental variability, nitrate nitrogen and sodium in the petioles, as regards the genetic variability for 1956, follow one behavior pattern and phosphorus and potassium in the petioles another. The behavior patterns are not as marked for the genetic variability as they are for the environmental variability. Sodium in the petioles is positively associated with phosphorus and potassium in the petioles, and the correlation coefficients are statistically significant at the 5% level. Also, the associations of nitrate nitrogen with these same two characters are positive, but they are negligible. Not only are phosphorus and potassium following very similar behavior patterns, as regards their association with other characters, but they are very closely associated with each other, 79 and 92 percents of the variances of one being accounted for by that of the other on the non-fertilized and fertilized plots, respectively.

12. In 1958 weight per root is negatively associated with total nitrogen in the thin juice, whereas, in 1956 the association is positive. It is clear that ail the populations grown in 1958 are not behaving the same as the populations grown in 1956 as regards the relation between weight per root and total nitrogen in the thin juice.

13. The data for both 1958 and for 1956 on the fertilized plots show that percentages sucrose and percentages apparent purity are negatively associated with total nitrogen in the thin juice. In 1958 both of these characters are positively associated with phosphorus and potassium in the petioles and negatively associated with sodium in the petioles. As regards the genetic variability in 1956 sucrose is negatively associated with phosphorus and potassium in the petioles and the relation with sodium is negligible.

14. For 1958 total nitrogen in the thin juice is negatively associated with nitrate nitrogen, phosphorus, and potassium in the petioles and positively associated with sodium. However, the association with sodium is negligible. The genetic association of total nitrogen in the thin juice and nitrate nitrogen in the petioles is the opposite of that for these same two characters in 1956. It must be kept in mind that the populations which provide the differences giving rise to the genetic variability are not the same for the two years.

15. Nitrate nitrogen in 1958 is very closely associated genetically with potassium in the petioles and to a smaller degree with sodium. The associations of nitrate nitrogen with phosphorus are

negligible in both 1958 and 1956. Also, the association with potassium is negligible in 1956.

16. For the genetic variability, phosphorus and potassium in the petioles are positively associated in both 1958 and 1956, the association being negligible in 1958 and very close in 1956. It must be kept in mind that the populations differed for the two years. Phosphorus and sodium are negatively associated in 1958 and positively associated in 1956. Potassium and sodium are positively associated in both years and to about the same degree.

17. Considering all of the associations studied, both environmental and genetic, the most consistent relations involving percentage sucrose and percentage apparent purity are those with each other and those with total nitrogen, glutamic acid, and betaine in the thin juice. With each other the associations are positive and with total nitrogen, glutamic acid, and betaine the associations are predominantly negative. For the environmental variability the degree of association is very high, the least amount of the variability accounted for by regression being 94%. For the genetic variability in 1956 the association between sucrose and total nitrogen is negligible on the fertilized plots. The same is true of sucrose and betaine in 1958. This indicates that some populations produce higher percentage sucrose at the same or higher concentrations of total nitrogen and betaine in the thin juice than do other populations. From a study of the means such was found to be the case. The same was found to be true as regards the associations of purity with total nitrogen and betaine.

18. Also, a study of the means reveals that some populations have as low concentrations of total nitrogen in the thin juice on the fertilized plots as other populations have on the non-fertilized plots. Further for such populations there was no material reduction in the percentages of sucrose in going from the non-fertilized to the fertilized plots.

19. The data provide information as to the concentrations of nitrogen and betaine in the thin juice and concentrations of phosphorus in the petioles at which marked reductions in percentage sucrose and percentage apparent purity occur. Sharp reductions in percentage sucrose and percentage apparent purity for A54-1 (Table 8) occur in going from total nitrogen concentrations of 46.8 mg to 62.9 mg, from betaine concentrations of 116.6 mg to 154.9 mg, and from phosphorus concentrations of 1598 ppm to 1318 ppm. These limits pertain to the genetic and environmental conditions prevailing during this study.

20. For these studies, as regards environmental differences (Table 8), increases in total nitrogen were accompanied by marked increases in weight per root up to concentrations of 46.8 milligrams per 100 milliliters of thin juice. However, increases

in concentrations of total nitrogen were not accompanied by increases in weight per root above a value lying somewhere between 46.8 and 62.9 mg per 100 ml of thin juice.

21. Both percentage sucrose and percentage apparent purity, as regards the environmental variability, continued to decrease with an increase in concentration of total nitrogen in the thin juice. In going from 18.8 mg of total nitrogen in the thin juice to 103.5 mg the accompanying decrease in sucrose was 4.9% (from 17.9 to 13.0) and the accompanying decrease in percentage purity was 13.4% (from 96.1 to 82.7).

22. It is apparent that under the environmental conditions of this experiment maximum yields are reached with high applications of nitrogen fertilizer and that applications greater than necessary for maximum yield usually result in material reductions in both percentages of sucrose and percentages of purity.

23. For some genotypes as compared with other genotypes decreases in concentrations of total nitrogen, glutamic acid, and betaine are accompanied by increases in both percentage sucrose and percentage purity when grown at the same fertility levels. This might be expected.

24. Some genotypes have lower concentrations of total nitrogen in the thin juice than other genotypes when grown at the same fertility level and these lower concentrations are accompanied by increases in both percentage sucrose and percentage apparent purity.

25. Some genotypes have higher percentage sucrose and higher percentage purity than other genotypes even though the concentrations of total nitrogen in the thin juice are not materially different.

26. For some genotypes as compared with other genotypes increases in concentrations of total nitrogen and glutamic acid and a decrease in concentrations of betaine are accompanied by increases in both percentage sucrose and percentage purity.

27. For some genotypes as compared with other genotypes decreases in concentrations of both total nitrogen and glutamic acid and an increase in concentration of betaine are accompanied by increases in both percentage sucrose and percentage purity.

28. For some genotypes as compared with other genotypes increases in concentrations of total nitrogen and betaine and a decrease in concentration of glutamic acid are accompanied by increases in both percentage sucrose and percentage apparent purity.

29. For some genotypes as compared with other genotypes increases in concentrations of total nitrogen, glutamic acid, and betaine are accompanied by increases in both percentage sucrose and percentage purity.

30. For some genotypes as compared with other genotypes increases in percentage sucrose are not necessarily accompanied by increases in percentage apparent purity. Likewise for some genotypes increases in percentage apparent purity are not necessarily accompanied by increases in percentage sucrose.

31. It is clear that from the immediately above cited facts concerning the behavior of these characters and their associations the genes conditioning them can be recombined. As a consequence recombination of and different degrees of expression of the characters result. It is apparent that such findings have a very important bearing on breeding populations of sugar beets that possess high weight per root, high percentage sucrose, and high percentage apparent purity.

32. It was found that relative concentrations of nitrate nitrogen in the petioles cannot always be taken as an indication of relative concentrations of total nitrogen in the thin juice. For example. 55-5307 grown in 1958 has the lowest concentration (2865 ppm) of nitrate nitrogen in the petioles and the highest concentration (116.4 mg) of total nitrogen in the thin juice; whereas, A56-5BB has the highest concentration (6240 ppm) of nitrate nitrogen in the petioles and the lowest concentration (71.2 mg) of total nitrogen in the thin juice. This was not a lone case as similar behavior patterns were found for populations grown in 1956. This finding has an important bearing; on the breeding of high quality sugar beets. It means that populations can be bred which have comparatively higher concentrations of nitrate nitrogen in the petioles and lower concentrations of total nitrogen in the thin juice without a material reduction of percentage sucrose and percentage purity as compared with other populations. It will be recalled that lower concentrations of total nitrogen in the thin juice is very closely associated with both higher percentages of sucrose and of apparent purity.

33. Also, it was found that relative concentrations of potassium in the petioles cannot always be taken as an indication of relative concentrations of potassium in the thin juice. For example, the F, had 18595 ppm of potassium in the petioles and only 101.9 parts per 100,000 of potassium in the thin juice; whereas, A54-1 had 17695 ppm of potassium in the petioles and 133.1 parts per 100,000 of potassium in the thin juice. This genetically controlled decrease of 133.1 to 101.9 parts per 100,000 of potassium in the thin juice was accompanied by increases of 0.8% in sucrose and 2.1% in purity.

34. In these studies relative concentrations of sodium in the petioles are a fairly reliable indicator of relative concentrations of sodium in the thin juice.

35. With the exception of glutamic acid on the fertilized plots in 1956, the phenotypic-dominance phenomena of the chemical characters in the thin juice are favorable to both higher percentage sucrose and higher percentage apparent purity. That is, the F_1 as compared to the two inbreds showed partial dominance, complete dominance, or heterosis for lower concentrations of the respective chemical in the thin juice. This was true for both of the F_1 hybrids grown in the two different years.

36. These results involving studies of the genetic variability and phenotypic-dominance phenomena indicate that it is possible to breed hybrids that have higher percentages of sucrose than our better commercial varieties at earlier dates of harvest. Such was found to be the case. One hybrid had as high percentage sucrose when harvested on September 14, 1961 as did A56-3, a commercial variety, when harvested on October 16, approximately one month later. None of the F, hybrids were equal to A56-3 in weight per root, but one was fully the equal of A56-3 in yield of sugar per plot.

37. The studies reported in this article indicate that it should be possible to breed hybrid populations that are superior to A56-3 in weight per root, percentage sucrose, and percentage apparent purity. Such was found to be the case for one hybrid grown in 1960. It surpassed A56-3 in all three characters and the differences were statistically significant.

38. From these studies it is apparent that to obtain maximum returns for both the farmer and the processor of beet sugar, populations of sugar beets adapted to production at higher fertility levels and adapted to other environmental conditions, such as different climates and locations must be bred. If the maximum returns are to be realized, proper fertilizer and other cultural practices must be followed in growing of these superior populations of sugar beets. It does not seem likely, if at all possible, that populations, hybrids or otherwise, can be bred that will produce satisfactory percentages of sucrose and satisfactory percentages of apparent purity when indiscriminate use of nitrogenous fertilizers is practiced. It seems almost certain that the populations grown in the future to produce beet sugar will be varieties or hybrids bred to take advantage of the favorable phenotypic-dominance phenomena found from these studies to be occurring.

39. It seems highly improbable that the populations being grown commercially today have obtained the maximum possible in weight per root, percentage sucrose, or percentage apparent purity. It is still more improbable that the populations being grown today have obtained the maximum possible in recombining high weight per root, high percentage sucrose, and high per-

centage apparent purity. Some of the researches promising the greatest remuneration to the beet sugar industry involve fundamental studies on the genetics of and methods of breeding for these three characters.

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An Improved Paper Chromatography Method for the Determination of Raffinose and Kestose in Beet Root Samples

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Introduction

Historically, the determination of raffinose by paper chromatography was introduced by deWhalley (4)[2] using the method developed by N. Albon and D. Gross in the Tate and Lyle Research Laboratories. Initial raffinose determinations were made on raw beet sugars. Later, Brown (3) introduced an adaptation of the original procedure for the quantitative determination of raffinose in beet root press juice. Modifications included deionization of press juice before the sample was applied to the paper. Quantitative evaluation of raffinose concentration was accomplished by visual comparison with spots of known raffinose concentration. Kestose spots were noted but no attempt was made to evaluate them quantitatively.

The method presented in this paper describes preparation of press juice samples, evaluation and selection of an improved solvent system and color indicator. Several different papers were evaluated on the basis of component resolution within a specified time. The quantitative determination of raffinose using optical density measurements is described. Kestose is determined indirectly by reference to standard raffinose concentrations.

Experimental Procedure

From 200 to 300 grams of representative rasped pulp is obtained from the beet root in question. A 200-gram sample of pulp is placed in a high speed Waring blender, 175 ml of deionized water is added and the pulp is blended at high speed for a period of 7 minutes. The pulp slurry is filtered through a tight weave linen in a 10 cm Biichner funnel under gentle vacuum. Fifty ml of the filtrate is pipetted into a 100 ml Kholrausch flask, 2 to 5 ml of 55 brix basic lead acetate is added to the Kholrausch flask. The flask is diluted to the mark with deionized water, mixed by inverting several times and filtered through a Reeve Angel No. 201, 8" X 8" square filter paper with a little celite filter aid added. The clarified leaded filtrate, after dilution to a standard sugar concentration, is applied to the chromatography paper.

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[2] Numbers in parentheses refer to literature cited.

The developing solvent is prepared by mixing n-butanol, glacial acetic acid and deionized water, (4 : 1 : 5) v/v in a separatory funnel of suitable volume. After standing for 15 minutes, the bottom aqueous phase is discarded. The top organic phase is used as the partition solvent. From a 22.5 X 18.5 in sheet of Schleicher and Schuell No. 2043-b, three 7.5 X 18.5 in sheets of paper are cut. Machine direction of the paper should correspond with solvent flow. The cut sheets are marked with seven pencil dots 2.5 in from the edge at 1 in intervals. The clarified leaded filtrate is adjusted to a standard 3% sucrose concentration by the following simple calculation:

1. (200 m.m. Pol. tube reading on leaded filtrate X 2.6 = % sugar in solution)

2. $\frac{(\%\ \text{Sugar in solution}) \times (10)}{3.0}$ = The final volume an initial 10 ml portion of leaded filtrate must be diluted to.

A raffinose standard is prepared by weighing 48.0 mg of anhydrous raffinose into a 100 ml volumetric flask and diluting to the mark with deionized water at 20°C.

Standards are spotted on the paper in the 1, 2, 4, 6 and 7 spot marks. Positions 3 and 5 are reserved for the unknown raffinose and kestose samples. A 2.5 microliter application of the standard solution equals 0.2% raffinose on 20 microliters of a 3%, sucrose unknown solution. The following spot pattern is utilized: 0.2%, 0.2%, unknown, 0.4%, unknown, 0.6%, 0.6%. These seven spots correspond to amounts of 2.5, 2.5, 20, 5, 20, 7.5, 7.5 microliters respectively. Applications are made in 2.5 microliter increments with a Micro Chemical Specialties Company 2.5 microliter No. 282-A self-filling micropipet. Spots are allowed to dry completely under a heat lamp between applications.

The finished sheets are placed in any standard chromatography chamber in the descending or ascending position. Equilibration of the sheets with the vapor phase in the cabinet is not necessary. The development solvent is added to the troughs and the cabinet is sealed. After 16 to 18 hours, the sheets are removed and dried in a forced air hood for a two-hour period. Seven 1-inch strips are cut from the length of the sheet, which include the five standards and two unknowns. The strips are individually dipped in a small shallow tray containing the color developer. The color developer consists of 0.5 grams resorcinol and 15 grams trichloracetic acid dissolved in 100 ml of anhydrous-ethyl acetate. The strips are dried in a vertical hanging position for 30 minutes in a forced air hood. The dried strips are heated in a forced air drying oven at 110°C for 7 minutes.

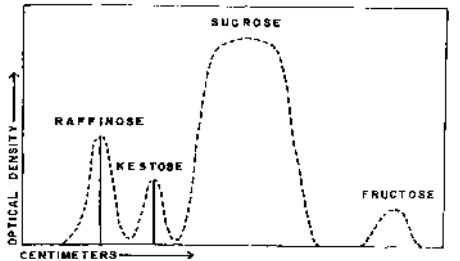

Figure 1.—A typical plotted scan pattern on an unknown run.

The developed strips are scanned immediately with a densitometer, Photovolt Model No. 425, with attached high sensitivity galvonometer. A 490 millimicron narrow band glass filter is used in the scanning head in conjunction with a 0.1 cm X 0.6 cm light slit. Figure 1 shows a typical plotted scan pattern on an unknown run. Maximum optical densities of the standards vs. the log of the standard concentrations are plotted. Unknown raffinose and kestose concentrations are determined from the standard curves on each individual chromatography sheet (1). This is necessary to compensate for small Rf. variations in different sheets. Kestose spots are evaluated on the raffinose standard curve (6). The raffinose equivalents obtained are divided by a factor of two which gives a true approximation of the percent kestose on sucrose. Results are reported directly as percent raffinose or kestose on sugar.

Table 1.—Comparisons of eight replications with 0.2 and 0.4% raffinose on sucrose added.

	1	2	3
Run No.	Original % Raff. on Sugar	Original Sample + 0.2% Raff. on Sugar	Original Sample + 0.4% Raff. on Sugar
1	0.43	0.60	0.87
2	0.40	0.58	0.83
3	0.39	0.63	0.83
4	0.41	0.60	0.80
5	0.40	0.61	0.79
6	0.40	0.57	0.79
7	0.38	0.56	0.81
8	0.39	0.60	0.79
Average	0.400	0.594	0.814

Standard Deviation — 0.0228

Statistical comparison of the original juice samples in Columnl with results in Columns 2 and 3 and 3 minus added Raffinose shows no significant difference between results in Columns 1, 2 and 3 at the 95% confidence level.

Results

Table 1 shows 8 replications of a sample of beet root press juice. Averages and standard deviation figures are given at the 0.2 and 0.4% raffinose on sugar addition levels. Unfortunately, there is no standard test known in the industry with which to compare this determination. It is of interest to note that statistical comparison of the original sample percent raffinose on sugar with the values found in columns 2 and 3 show no significant difference at the 95% confidence level when they are corrected for the amounts of raffinose added to the original sample.

Discussion

Several factors contribute to the successful resolution and quantitative evaluation of carbohydrate compounds in mixture.

Solvent: Three solvent systems were evaluated in conjunction with this study. They were n-butyl alcohol-pyridine-water-benzene (5 : 3 : 3 : .45, v/v), ethyl acetate-acetic acid-water (3 : 1 : 3, v/v) and n-butyl alcohol-acetic acid-water (4 : 1 : 5, v/v). Evaluation was based on an 18- to 20-hour run corresponding to an oxer-night duration. Solvent system selection was based upon maxi-mum resolution in the shortest possible spot run. Figure 1 in-dicates that complete component resolution has been obtained with a minimum of run from the starting line in the run time specified. This is of particular importance because of the en-largement of spots due to diffusion as run length increases (2). Therefore, to obtain maximum optical density readings with a standard light slit, it is necessary to resolve the carbohydrate mix-ture in the shortest spot run length possible. Upon accomplishing this, maximum sensitivity in the concentration determination is assured. The latter solvent system listed was found to fulfill the cited requirements.

Chromatography Paper: The following chromatography papers were evaluated in this study. Whatman 1, 2; Schleicher and Schuell 2040b, 2040a, 2045b, 2045a and 2043b. Papers were evaluated on a basis of component resolution, spot deformation and uniformity of paper density. Schleicher and Schuell 2043b was selected as suitable for this determination. Whatman papers 1 and 2 were found unsuitable due to characteristic W. V and N shaped raffinose and kestose spots. Quantitative evaluation of spots displaying irregular shapes is not possible with a densito-meter. Developed spots should be either circular or ovoid in shape. Schleicher and Schuell 2043b displayed uniform ovoid spots, good resolution and maximum variation in optical density of ±0.15 O.D. units. If blank densitometer readings can be main-tained at zero optical density throughout the scan length of the

strip, then it can be assumed that the maximum optical density readings on the spots are valid. Unfortunately, all papers suffer from some variation in density. Variations from ±0.10 to ±0.25 optical density were noted in the papers evaluated.

Color Developer: A partial list of the more common carbohydrate color reagents evaluated are included in this paper. Aniline hydrogen phthalate, alpha-naphthol, m-phenylene diamine, p-anisidine HC1, triphenyltetrazolium chloride, (used after trichloracetic acid hydrolysis of raffinose and kestose), benzidine, p-dimethylamino aniline, orcinol and resorcinol. Resorcinol was the color reagent of choice. Selection was based on background color produced and sensitivity. The resorcinol reagent gave little or no background coloration and was sensitive to as little as 1.5 micrograms of raffinose. Developed colors produced were stable up to 1 hour. Faint spots may be easily detected under ultraviolet light.

Raffinose and Kestose Quantitative Evaluation: The resorcinol reagent reacts only with the fructose moiety in the raffinose molecule. Tests performed with fructose, sucrose and raffinose have proven that only the fructose moiety of the polysaccharide is involved in quantitative color production. Kestose is described by Freed et al (5) to be a trisaccharide composed of two fructose molecules and one glucose molecule. This was verified by isolating a very small quantity of kestose from actual press juice, hydrolyzing and submitting the hydrolysis products to paper chromatography. Complete hydrolysis yielded fructose and glucose. Comparison of the hydrolysis products with known glucose and fructose standards indicates a kestose molecular composition of two fructose molecules to one glucose molecule. It is therefore assumed that an indirect quantitative determination of kestose can be obtained by dividing the raffinose equivalent obtained from the standard raffinose curve by a factor of two. Kestose is not available from any commercial source. In order to estimate kestose it is necessary to use indirect means rather than a direct comparison with known standards.

Summary

An improved chromatographic procedure has been presented for the determination of raffinose and kestose in beet root samples. It has been found that deionization of sample juices in preparation for chromatography is unnecessary. Colloidal material present in press juice must be removed by clarification. Organic and inorganic cations and anions left after wet lead clarification do not affect the resolution of carbohydrate mixtures. Use of optical density to measure the concentration of carbohydrates in

question decreases the human error predominate in quantitative estimations based on the visual comparison method. Statistical analyses indicate that the method presented shows excellent precision. Intercomparison of the results presented shows suitable accuracy within the method itself.

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Winter Protection of Piled Sugar Beet Roots[1]

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In Michigan and Ohio, sugar beet processing in recent years has been concentrated in about one half of the factories in operation a few years ago. This has resulted in a significant lengthening of the processing period and increasing difficulty in the processing of the last 10 to 15% of the beets. Not only has there been an increase in raffinose, which reduces extraction, but complete loss of a considerable volume of beet roots in the more unfavorable seasons.

In abnormally warm storage seasons, wilting may be prominent in beets on the outside or near the edges of the piles. In average seasons, freezing and thawing of beets to a depth of several feet is expected on the west slopes of the piles. In colder seasons, very deep freezing during- cold periods is often followed bv thawing in warm periods, particularly when beets remain in the piles much past the first of the year. In the process of fluming and washing, some of these thawed beets disintegrate entirely, and others are removed by discarding the soft beets in the process of loading them into trucks when transporting from the pile to the flumes. In any case, the problem of satisfactorily disposing of the spoiled beets is a formidable one, whether in the settling pond or in the piling area.

Experimental method in Michigan

In an attempt to reduce this apparent loss, financial and otherwise, parts of two piles of beets were covered with plastic sheets from November 28, 1960, to January 27, 1961, at Sebewaing, Michigan. Experience had shown that the prevailing westerly winds caused the greatest losses on the west slopes of piles running north and south. Such a pile was selected for most of the experiment. The pile was the standard truncated pvramid, about 600 X 120 feet on the base. 560 X 80 on the top and 19 to 20 feet deep. Plastic sheets 40 feet wide were used, both clear and black, in a thickness of 6 mils. Plastic was used on the west face only, except in one trial when a strip of plastic was used on the south face of a pile running east and west.

In some cases, straw, in various amounts, was used under the plastic. In one trial, straw only was used. In another trial, the

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plastic extended from the top to about 10 feet from the ground. The plastic was held in position by old tires and/or discarded twine fish nets. In each case, an uncovered strip was left adjacent to the covered one. In each segment of the pile, both covered and not covered thermometers were buried in three positions:

1. Six feet deep in the center of the top of the pile (never covered);
2. Six feet deep in the beets, about one-third of the way down the side, in both covered and not covered beets;
3. Six feet deep in the beets, about two-thirds of the way down the side, in both covered and not covered beets.

Other thermometers, outside the piles, gave air temperatures. All thermometers were read at 8 A.M. each day.

Results in Michigan

Figure 1 shows an over-all view of the experiment at Sebewaing. Figure 2 shows one segment of the pile (40 feet wide) covered with plastic. In order to condense the volume of data,

Figure 1.—An overall view of the experiment, Sebewaing, Michigan.

Figure 2.—A close-up of plastic over a covering of straw.

temperature readings for about every fifth day are given in Table 1. The days were selected to show the minimum temperatures attained in the "body" of the beets. "Body" temperatures are the average of the two side temperatures, 6 feet deep. "Top" temperatures are those taken 6 feet deep in the center of the uncovered top of the pile. For brevity, average temperatures for 5 closely agreeing checks are given, for 4 plastic-covered strips—two with and two without straw—and for one strip with straw only.

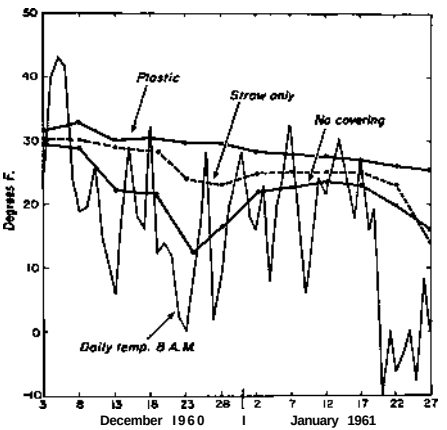

Figure 3.—Daily air temperatures at 8 A.M., Sebewaing, Michigan, and temperatures on the west side of the beet pile, six feet deep in the uncovered beets, six feet deep in the beets covered with straw only, and six feet deep in the beets covered with plastic.

In Figure 3, daily air temperatures are shown, together with (1) average "body" temperatures for the 4 strips fully covered with plastic, whether or not straw and twine were used, since these made almost no difference, (2) "body" temperature under straw only and (3) average "body" temperature of the 5 uncovered check strips.

The body temperatures of the plastic covered area and in the top of the pile running east and west were regularly from 2 to 3 degrees higher than that in the pile running north and south (Data not shown).

Table 1.—Temperatures (F°) 6 feet deep in the center of the uncovered "top", 6 feet deep in the uncovered beets ("body" temperatures) and 6 feet under the coverings on the west sides of the piles ("body" temperatures).

	Temperature F°												
Date*	**11/28**	**12/3**	**12/6**	**12/13**	**12/18**	**12/24**	**12/28**	**1/3**	**1/7**	**1/2**	**1/17**	**1/22**	**1/27**
Average 5 checks													
Top	**47**	32	**32**	30.5	**31**	**29**	**31**	**29**	**30**	**29**	**29**	**28**	27.5
Body	**45**	29.5	**29**	**22**	**22**	**12.5**	**16.5**	22	22.5	23.5	23	**20**	**16**
Average 4 plastic-covered													
Top	**47**	33	32	**31**	30.5	**30.5**	**30**	**31**	30.5	**30**	32	**29**	**29**
Body	**44**	**32**	33	**30**	30.5	**29.5**	29.5	28.5	**28**	**27.5**	**27**	**26**	**26**
Straw only													
Top	**50**	**37**	35	**32**	**31**	**31**	**31**	32	32	32	33	**30**	**30**
Body	**44**	**30**	**30**	**29**	**28**	**24**	23	25	**25**	**25**	25	23	**14**

* Temperatures at 5 day intervals, or on days of minimum temperature in "body" of beets.

Discussion

From Table 1 it can be seen that all top temperatures were within a degree or two of 30°F, no matter what the daily temperature, until the end of the experiment. This was due to convection of warmer air from the interior of the pile.

From figure 3, it is plain that covering with plastic held the beets at a rather uniform "body" temperature that gradually fell to 26°F by January 27. The uncovered body area is shown to vary widely in temperature, depending upon the weather, and to be from about 5 degrees to 18° F colder than when covered with plastic. The straw covered strip was intermediate in body temperature, and was readily affected by air temperatures. When plastic reached only to within about 10 feet of the base, effectiveness was greatly reduced (Data not shown).

The fact that uncovered strips interrupted the covering probably somewhat reduced the effectiveness of the coverings. Even so, the covered beets were frozen to not nearly the extent as those not covered, approximately 2 to 3 feet in the plastic covered vs. 12 to 14 feet in the uncovered check. Alternate freezing and thawing was equally reduced. We are informed that frozen beets can be sliced and extracted without appreciable trouble. If this is the case, protection from weather damage was almost complete with plastic covering.

Method in Ohio

In a similar trial at Fremont, Ohio, from November 9 to December 23, 1961, a strip about 100 feet long and 30 feet wide on one side of a pile of beets was covered with 4 mil clear plastic, and held in place by blowing asphalt-impregnated straw on top of the plastic. A machine used for stabilizing grass seedings along highways was rented for use in this experiment. One ton of straw was used in a laver 4 to 10 inches thick, together with 30 <?-allons of asphalt. This treatment proved adequate to hold the plastic in place even in heavy winds. Costs for labor ($15), straw ($12), asphalt and machine rent ($22) and plastic ($26) totaled $75, or about $1 \frac{1}{9}$ cents per ton of beets on approximately 5000 tons covered. It is felt that this could be reduced to something like 1 cent per ton, if operating on a lareer scale, since the blower could cover a much larger oile area (75,000 tons of beets) at the same rental in an 8-hour day. Removal of the plastic and straw layer was rapid and easy.

Previous to covering the pile, 27 weighed beet samples, in numbered nylon mesh bags, were placed at depths of 4, 8 and 12 feet in the pile. On November 9, 27 duplicate samples were analyzed with the crown on the beet, for comparison with the

buried samples when they were removed from the pile 47 days later.

Results in Ohio

All except 2 of the 27 buried samples were recovered, weighed and analyzed for sugar. Since all samples were from one farmer's field, sugar percentage and shrinkage were remarkably constant. In averaging the results from the 25 recovered samples and their duplicates, the following results were obtained:

Avg Wt when placed in pile	Removed	Weight Shrinkage	Percentage sugar into pile	Percentage sugar removed	Percentage total sugar apparently lost
233.8 oz	230.8 oz	1-28%	13.82	13.78	1.57%

A portion of the pile was sliced as follows on December 23. The covered beets were handled *in* the first shift, and the two following shifts continued on the same pile with uncovered beets.

	First shift covered beets	Second shift not covered	Third shift not covered
Tons sliced	588	580	528
Avg purity	85.0	84.3	84.6
Avg sugar content	14.10	13.94	13.50
Total recovery per ton beets	239.6	235.0	228.4
Lb sugar per shift	140906	136300	120595

It is recognized that these plant operation figures are not adequate for accurate cost accounting. Since the calculated recovery of sugar per ton was 239.6 pounds for the covered beets and 231.7 pounds for those not covered, there appears to have been about 8 pounds extra recoverable sugar per ton in the covered beets. But, assuming a cost of about 1 cent per ton for the plastic protection, these figures indicate a recovery of perhaps 800 pounds of sugar per dollar expended in this experiment.

In this season in Ohio, the weather was never severely cold before the beets were removed from the piles and no freezing occurred. In the 47 day period, only three minimum night temperatures were below 20°F and only 8 were below 25°F. In contrast with the experiment in Michigan in 1960, where protection from deep freezing and thawing was a problem, here the main observable difference was in the degree of wilting of beets near the edges of the pile. The daily temperature of the covered beets averaged about 39°F during the last month, while the uncovered beets were about 4 degrees cooler, although with much greater fluctuation than was found in the covered beets.

Covering with plastic, while preventing penetration of rain and melted snow water into the pile, may also concentrate this water in limited areas, and in depressions in the plastic. This should be avoided. In these experiments molding of the beets was a very minor problem, but had the temperatures been higher, ventilation of the beets might have been advisable.

Summary of Ohio Experiment

Sugar beets in large piles were covered on one side with plastic sheets to protect the beets from wilting and freezing. It was found convenient to hold the plastic in position with asphalt-impregnated straw, blown into place. Costs, for labor and materials in covering about 5000 tons approximated 1 1/2 cents per ton, but could be lowered if on a larger scale. Such covering reduced wilting of beets, and gave great protection from freezing and thawing. Factory operation for one day, indicated a slightly (8 pounds per ton) greater recovery of sugar from the covered beets.

Conclusions

These two "pilot plant" experiments in the use of plastic to prevent undue weather damage to piled sugar beets show considerable promise. Larger scale experiments in which factory operations could compare covered and uncovered beets for longer periods would yield valuable data on costs and sugar recovery and could lead to refinements in the technique. While, in these two experiments, the emphasis Avas mainly on protection from damage from prolonged freezing weather, it might be discovered that protection from dehydration, from mid-day warm winds, or from excessive rainfall might be of equal or greater importance in the conservation of sugar beet quality.

Experiments in Vacuum Pan Control

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The primary purpose of the present work was to increase pan floor capacity by improved vacuum pan operation and to develop a control system that would enable any person to boil consistently good strikes.

A material balance showed that it was first necessary to improve white pan yields. An increase from the normal 40% to 60% could reduce total fillmass almost 50%. It would cut white fillmass by one third and high raw by an astonishing two thirds.

We were fortunate to have available pan microscopes, first a foreign model and later very excellent domestic units. The picture they gave of crystal growth within the pans themselves exploded some conventional theories and helped prove others. By watching the course of strikes boiled by even skilled sugar boilers it was apparent that there was a great deal of room for improvement.

In order to increase yield of finished sugar per strike it was necessary to boil grain of more uniform size and with a minimum of conglomerates. Such clean strikes around 60% yield were found to purge better and require less wash than poor strikes with yields below 40%. Better control of mean aperture was required which indicated the need for full seeding rather than by shock.

Rate of crystal growth depends upon supersaturation and syrup purity. In a typical standard liquor at maximum safe supersaturation, crystals can grow at a rate of about 0.016" per hour measured on the mean dimension. This is equivalent to about 3.5 microns per minute on each face. The pan microscopes disproved the existence of a supersaturation zone in which crystals form spontaneously only in the presence of other grain; above a very definite supersaturation, about 1.50, grain would form in syrup or at any stage of the strike. This simplifies the picture in that only one zone between 1.00 and 1.50 supersaturation is of interest in sugar boiling. If clean strikes were to be produced from an original seed crop, it was imperative that the upper limit never be exceeded; maximum rate of growth, however, would be realized just under this limiting supersaturation. In the interval just after graining when crystal area was low, it was easy to exceed the safe value and form more grain. In spite of

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the vastly increased crystal area during final brixing the limit could again be reached since cutting off feed liberated sugar some eight times as fast. Fine grain formed at this time goes"' through the centrifugal screen instead of being deposited on the existing crystals and is lost from the pan yield. On coil pans, premature addition of one coil to many in the middle of the strike could also create a smear.

Interestingly, the pan microscope disproved the notion that fine grain formed during the course of a strike can be washed out by a large drink of feed. Grain will only dissolve in liquor below saturation and it would require a volume of 70 brix feed almost equal to the fillmass volume at any time to reduce a pulled together strike to saturation. What actually happens is that the fine grain conglomerates and grows rapidly so that in a few minutes the strike looks clean again in the sight glasses or on a slide but in the microscope the new ones are all there growing along with the larger original crystals.

Conventional methods of measuring supersaturation were found inadequate for the precision boiling we sought. Boiling point measured in the side or center-well of a pan is affected by material that bypasses and reaches the bulb without dropping to the temperature and pressure at the fillmass surface. A means was developed to measure temperature at the surface which is the most highly supersaturated region; this coupled with a precisely controlled absolute pressure gave a reliable reading of supersaturation throughout the strike.

The actual absolute pressure at which a strike is boiled seems to be of secondary importance since equally good strikes can be produced over quite a range of pressures. The value selected is determined more by considerations of water supply and steam pressure. At any given absolute pressure, the temperature corresponding to the supersaturation limit can be determined approximately from the alignment chart of Figure 1 which is based on the data of Brown and Nees (1). The actual value for the limit on a particular syrup is precisely determined by means of the pan microscope which is a necessary part of a precision boiling system. The most direct way to fix the value is to gradually raise the pan temperature until the appearance of new grain shows that the limit has been exceeded; they are visible within seconds after they form. Or the saturation point can be determined by introducing a bit of powdered sugar into the graining charge as it is being concentrated and noting the temperature at which the crystals first show corners.

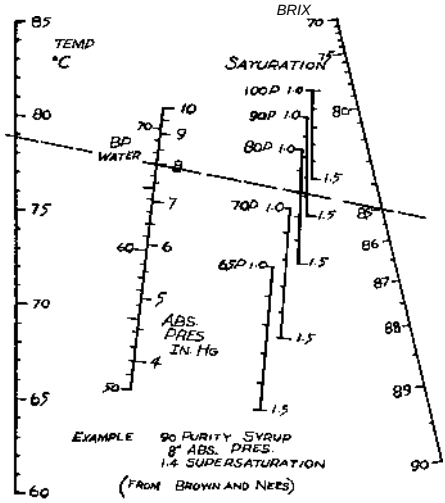

Figure 1.—Supersaturation chart for beet sugar syrups of various purities. At a controlled absolute pressure in a pan, the boiling temperature at the fillmass surface indicates the degree of supersaturation; it must be held below 1.5 to prevent formation of new grain.

Boiling time is generally fixed by the heat transfer rate in a given pan but it was found that the pan cycles could be shortened by bringing the pan together as soon as possible, and carrying an optimum tightness during the feeding period. This seeming paradox that a tight strike is "looser" than a loose strike is probably due to the fact that for the same supersaturation gradient, the average syrup concenration is reduced as the crystal faces are brought closer together with corresponding decrease in viscosity. If tightened excessively, the overall fluidity is reduced by the increasing crystal concentration and heat transfer is reduced. A probe has been developed to record tightness and control feed to maintain it at the optimum which is of the order of 20% yield. A 10 to 20% reduction in boiling time can be realized by so doing.

The problem of conglomeration was not an easy one bu[1] some of the factors contributing to the formation of multipk

grain were established. Their complete elimination was not realized but it was possible to reduce the number to a small fraction of the total grain.

Observation showed that most conglomeration takes place when the crystals are quite small, 0.001 in to 0.003 in size. Before and after this dangerous age there is almost none. Carrying the strike at lower supersaturation or looser during this interval has little effect. Some improvement was noted by boiling at higher absolute pressures. Purity has a great deal of influence; conglomeration is almost no problem at all in the lower purity syrups. One plantation in Hawaii noted for its excellent boiling house work grains in low purity syrup and then switches to feed of higher purity.

Apparently vigorous circulation during the conglomeration period is the only cure. Mechanical circulation is very helpful but needs to be supplemented by some boiling. Open steam is useful in pans without mechanical circulation but is much less effective than the same amount of steam flow to the heating surface probably because of the local circulation created by the formation and liberation of vapor bubbles. Surface within the pan over which the boiling material can shower and spread further deters conglomeration; a coil pan is better in this regard than a calandria pan at twice the boiling rate.

In pans without mechanical circulation, conglomeration can be held down by rapid boiling but the conglomeration period occurs when the crystal area is too small to absorb the sugar liberated by the boiling. This dilemma is solved by reducing the steam flow only to a value that discourages conglomeration and feeding water to prevent the supersaturation from exceeding the upper limit. Within a few minutes, as the crystal area increases, the water flow is reduced to zero and boiling rate can be increased.

As these techniques were developed it became possible to boil consistently good strikes with low CV values, obtain high yields of well-formed grain and do them in minimum time. The final problem of introducing the correct seed crop to produce the desired final crystal size was solved by borrowing a technique that has been used in Hawaiian mills. Laboratory ball mills are charged with sugar and iso-propyl alcohol in the proportion of Mb to 1 liter. After grinding for 24 hours, the particle size has stabilized at an average of about 4.5 microns and the resulting density is around 2.5 X 10^9 particles per milliliter. Approximately 200 ml of this "milk" is sufficient to seed 1,000 cubic feet

of white fillmass for 0.015 in M.A. The actual amount for a particular pan can be adjusted until the required size is obtained and will repeat very well thereafter. Graining procedure is standardized by maintaining the same graining volume for each strike and introducing seed at the same supersaturation each time.

The immediate object of this work was not to produce a "push-button" pan control system but rather one which would make possible precision boiling of the most high-quality sugar from a given pan in the least possible time. The operations of dropping and steaming out as well as introducing seed have been left to the sugar boiler since the additional complexity and cost seem hardly justified. Nevertheless, the system, though only semiautomatic does reduce the time and attention normally required to a great extent. As the controls are arranged, variations in feed concentration or steam pressure are taken care of automatically. No adjustment of the supersaturation limit is required except in the event of a drastic change in syrup purity. The same system is applicable to white, high raw or low raw pans since the problems are the same. A pan with mechanical circulator does not require a water make-up valve to hold supersaturation.

On the pan control panel there are four controllers, absolute pressure, level, supersaturation and tightness. Steam flow is indicated so that the optimum tightness value may be easily determined and checked. The strike is initiated by turning a switch which opens feed and condenser water valves. When level reaches the graining volume, steam comes on to concentrate the charge and the level is maintained. An alarm sounds when supersaturation rises to 1.3; the sugar boiler acknowledges the alarm and seeds the pan with a measured quantity of the wet milled fondant. As supersaturation rises to the 1.5 limit, steam is throttled so the limit will not be exceeded. As the pan comes together, the increasing tightness opens the feed valve to hold it constant. Whenever the combination of feed and increased crystal area cause the supersaturation to fall away from the limit, the steam valve opens to maximum.

Boiling proceeds until the pan reaches maximum set level, the feed valve throttles to prevent further rise and the pan begins to brix up. If at this time, the supersaturation increases to the set limit, evaporation will be reduced to prevent grain formation This is a most important period since sugar is being deposited at the rate of many bags per minute; if hurried, it can only result in sugar being lost with the syrup and recirculate through the house.

As the tightness reaches the dropping point, the steam valve closes and an alarm notifies the sugar boiler that the strike is finished. He turns the switch to "off" and drops the strike.

The techniques and controls developed by this work achieved the original objective of increasing pan floor production. Reduced reboiling of syrups added economy dividends. To the less cost-conscious person, the sparkling sugar that emerged from the granulator and from the high and low raw machines was a delight to the eye.

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Symposium on Instrumentation in the Sugar Industry, Problems and Application*

Instrument Maintenance in the Sugar Factory—Hal L. Memmott and Park Gaugh[1]

Introduction

The increase in the use of instruments and controls in the modern sugar factory has necessitated a corresponding increase in instrument maintenance. An over-all yearly program of sound instrument maintenance becomes increasingly important. During operating periods, a program of preventive maintenance should be supplemented with an over-all inspection, cleaning and repairing program during intercampaign. More efficient plant operation can result from a planned instrument program, noting trouble spots during campaign and using the intercampaign time for improved instrument application.

Methods

Prior to the time the factory starts slicing beets, each instrument should be thoroughly cleaned and inspected. The cleaning involves freeing all air passages and moving parts. Oil and sludge residue are particularly troublesome in air relays and baffle units. Because most instruments use air and direct the air through small parts, passages, and nozzles, not enough emphasis can be placed upon clean air. Every effort possible should be made to ensure a clean dry air supply. Oil, water, or dirt in the air can easily cause the malfunction of an instrument. The instruments should have a separate air supply system with a special compressor. The supply air to the compressor should come from a clean source, preferably from outside the factory. The compressor should be located in a cool place and must be kept in the best of condition to insure that no oil gets into the air. Special high-grade compressor oil should be used at all times.

Thorough cleaning of the instruments is also beneficial in that all parts of the instrument may be inspected at that time for wear and damage. In particular, the condition of diaphragms, bellows assemblies, connecting linkages and nozzles can be observed and worn or damaged parts replaced when necessary. Tests for proper function of instrument components should be made

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by simulating operating conditions. Correcting any instrument faults at this time will result in a smoother factory start-up when campaign arrives.

The instrument normally measures thru its primary measuring element or unit and these should be checked for calibration. Thermal systems should be checked against a reliable mercury thermometer. Thermocouples should be calibrated using a reliable potentiometer. The thermal element capillaries are easily damaged by arc welding, rough handling, kinking or excessive vibration. Providing some kind of protection for these capillaries can help, but the system should always be calibrated. The bulb sometimes becomes insulated with dirt or scale causing an error in the reading, so these should be properly cleaned. Even if a thermal system is properly calibrated, errors can result from improper immersion depth, liquid level, temperature stratifying, bulb location or the misapplication of the thermal system. These problems will probably be observed during campaign, but they should be corrected—possibly during intercampaign. The solutions to these problems are obvious once the problems are recognized. For example, stratifying can be overcome by agitation, either mechanical, or in the case of smaller tanks just a tangential entry of the supply stream.

A dead-weight tester may be used to check and calibrate pressure systems to ensure their accuracy. Air purges will reduce corrosion from vapors where such is a problem with units measuring pressure. Bubble tubes have as the receiver a pressure measuring unit so these systems should be calibrated in a like manner to pressure systems. The greatest problems with bubble tubes are leaks and blocked air passages, so generally all tubing and capillaries should be inspected for kinks, and flattened or damaged areas. Using one-inch pipe or copper pipe for bubble tubes will reduce errors and troubles caused by solid material build-up and also reduce channeling in heavy syrups. Raising a bubble tube a few inches from the bottom of a tank such as raw juice tank or pulp water tank may prevent bubble tube errors. Bubble tube operation in heavy slurries can be helped by using two concentric-tubes with a small water purge running into the outer tube. In this case the outer tube or pipe should be l 1/2-inch or 2-inch pipe. Bubble tubes in some locations such as the concentrating pans or tail-end evaporators can be given a hot water purge simply by drilling and installing a small water line in the high and low pressure bubble tubes at an elevation above the normal juice level. This will melt out troublesome sugar or salt build-up that is the source of errors in these level readings.

The importance of the control section of the instrument cannot be overlooked and so the control unit must be properly aligned and adjusted. A few tests of the responses—proportional response, reset rate, and proportional derivative—can verify their proper operation.

Perhaps most of the time required for instrument maintenance is actually spent on actuating motor and valve overhaul. Pneumatic motor operated valves all need cleaning and usually the packing gland needs repacking. Here too, the cleaning is almost second in importance to a thorough inspection for wear, loose valve seats and rough stems. At least part of the repacking of glands during campaign can be overcome by the careful selection of the proper packing. One type of packing that has been particularly successful for valves operating in heavy syrups is Garlock #5733 Teflon braid packing. The proper lubricant can reduce stem wear and also increase packing life. For example, for valves in hot juices and syrups, the use of Rockwell-Nordstrom lubricant #555 gives excellent results.

Chattering causes excessive wear on valves and operators and results in a high maintenance cost. Of course, the best way to get away from chattering is by the proper planning and designing ahead of time so that the correct valve is used in a line of the correct size. If a chattering valve is already in service and cannot be replaced with the correct valve, a hydraulic snubber just ahead of the pneumatic operator will stop the chattering in most cases. However, this method cannot be used to cure troubles caused by misapplication and poor installation of control valves.

Closed butterfly valves have the force of the upstream pressure against the whole face of the disc. This condition can cause problems such as bent shafts, stuck valves, and leaky packing. While cleaning this type of valve, careful inspection of the valve shaft and operator stem should be made to make sure these parts are straight. If a bent stem is located it should be replaced with a larger stem to prevent further trouble. Because of the high torque required to operate these valves, every effort should be made to make sure that the valve and operator are correctly aligned.

Instrument accessories are an integral part of an instrument and these accessories also require maintenance. The supply air regulators and filters require complete cleaning and replacement of filter cartridges where necessary. Some pot metal types of regulators and filters react to the moisture in the air and this adds to the contamination of these parts. Climax Type 245 combination filter and regulator gives excellent results under

adverse air conditions. The moving parts and air passages in this unit are large enough to function efficiently even in very dirty air.

This covers a part of the maintenance problems in **a** factory, but a few other items should be mentioned, such as spare parts, etc. An adequate supply of the inexpensive parts such as gaskets, o-rings, diaphragms, diaphragm material, small relay parts, springs, etc., for both the instruments and the accessory equipment is essential. An excellent thing to keep in mind in purchasing instruments is to buy from a company that produces all the various kinds of instruments required in the factory. The ever-changing sugar factory requires an instrument that can be changed from one job to another with the simple substitution of one or two components. This requires that the instruments have unity, be as simple as possible and be adaptable to different application. The importance of these considerations *is* realized when determining the number and cost of spare parts. Regardless of cost, some of the expensive components, such as sensitivity units, absolute pressure bellows, etc., must be kept on hand. The more applicable these parts are to several different instrument types the more useful a stock of parts will be at a nominal cost. Standardizing on instruments from a reputable company helps to reduce the parts stock also.

Summary

1. Clean air is necessary for successful instrument operation.
2. Planned installation and sensible application is a big factor in instrument maintenance.
3. A planned program of cleaning, inspecting and adjusting all instruments is necessary.
4. Use preventive maintenance where possible.
5. Buy instruments from a dependable manufacturer.

Cost Reduction through Instrumentation Improvement in Steam Balance, Fuel Savings and Other Material Savings—Harold C. Dyer[1]

Cost reduction through instrumentation

About fifteen years ago, I was introduced to the problems of instrumentation in the beet sugar industry as the result of an instrument failing to perform on the process it was intended to control. No one was responsible for the satisfactory operation of any instrument and no one seemed to care too much whether

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or not the instrument worked properly. At the time I became interested in the problem, I found that many of the installation failures were the result of salesmen's errors in selling instruments without knowing the process to be controlled and the apathy of the operators toward automatic controls.

During the intervening years this condition has changed considerably. Instrument salesmen today are generally engineers and have become more familiar with the problems of the beet sugar process. They are now better qualified to recommend the proper type of instrument, and some of the plant personnel have become more interested in the satisfactory operation of control systems.

I have been invited to discuss with you in this paper "Cost reduction through instrumentation, improvement in steam balance, fuel savings, and other material savings." Unfortunately, instrumentation in our industry has lagged behind other industries. We have now learned that not only can labor be saved, but often as much or more benefit can be obtained in improved operations. In the past, in many instances operators lacked confidence in instruments and were reluctant to allow instruments to function completely on their own. In other cases, instruments could not be effectively applied to existing equipment . Until the equipment could be economically replaced with a type adaptable to instrumentation, automation had to be delayed.

Improvement in steam balance

Probably the most familiar control system for this purpose being used in the sugar industry is the evaporator control with which many of you are familiar. We have a total of seven factories equipped with evaporator control systems. In most of our installations these controls are used on a quintuple-effect evaporator, in which the exhaust steam from the main generator turbine is admitted to the steam chest of the first effect where it gives up most of its heat to the thin juice, causing the juice to boil. Vapor from the boiling juice cools in the evaporator dome and is piped to the steam chest of the second effect.

This process continues to the fifth effect where the vapor is condensed in a barometric condenser. The instruments used generally for the control of this system are designed to maintain the correct juice level in each effect, maintain constant density of the thick juice, hold first and third vapors constant, thereby maintaining second vapors at a relatively constant pressure, and to keep the evaporating rate equal to the demand for evaporation. This latter function is accomplished by raising and lowering the absolute pressure of the fifth effect.

The first and third vapor pressure controller, controls the pressure of the first and third vapors by operating a butterfly valve in the exhaust steam line to the first effect and a butterfly valve located between the second and third effect in the vapor line. This controller is a double unit type; one unit controls the pressure of the first effect, and the other unit controls the pressure of the third effect.

The absolute pressure controller measures and controls the absolute pressure within the last effect by operating a valve in the water line to the barometric condenser. The control point of this instrument is pneumatically adjusted by the thin juice supply tank controller.

The density controller measures and controls the density of thick juice leaving the last effect by operating a valve in the outlet line from the thick juice pump, thus throttling the flow from the pump.

The level instruments measure and control the level of the respective bodies by operating a valve in the inlet juice line to the respective body. Measurement of level is accomplished by bleeding air into two level taps located on the bodies.

The exhaust steam pressure controller measures and controls the pressure of the exhaust steam by operating in sequence a make-up valve and a relief valve. By the use of controllers the vapor pressures can be maintained for use elsewhere in the process.

Fuel saving through boiler controls

In the operation of a boiler steam generating svstem there are several essential elements which must be controlled for efficient and economical operation, resulting in fuel savings. These mainly are steam pressure, rate of fuel feed, fuel-air ratio and draft. We will consider these in this order.

Steam pressure

Steam pressure is a direct indication of the load demand on the boiler. If the load increases, the pressure will drop; conversely, if the load decreases, the pressure will rise. It is necessary to keep the pressure relatively constant in the main header to have an adequate supply of steam at a given temperature available to the process equipment. Varying pressures and temperatures are often injurious to the process. For this control a master controller is used. This controller measures the steam header pressure and sends a loading impulse to the fuel feed controller to maintain a constant steam pressure.

Rate of fuel feed

In order to maintain a constant pressure in the main header, it is necessary to change the rate of fuel feed to the boiler. As the lead increases, the fuel feed rate must be increased proportionately and conversely, decreased with decreasing load. For this control a fuel feed controller is used. This controller receives the loading impulse from the master and changes the fuel feed to correspond to the master loading.

Fuel-air ratio

As the fuel feed rate changes it is desirable to change the rate of air flow to the boiler proportionately. As fuel is burned, the carbon combines with the oxygen in the air, thus forming CO2 in the flue gases. If too much air is supplied to the furnace, it dilutes the gases and the CO, content will be low. Also, the boiler cutlet gas temperature will be excessively high. If not enough air is admitted to the furnace, then all of the fuel is not burned and we get CO, and CO in the outlet gases. CO is unburned fuel which is wasted out of the stack. In normal operation it is desirable to operate with a small amount of excess air. In the case of natural gas fuel, the excess air should be between 10 and 20%. This will give approximately 9 to 10% CO, and 3 to 5% O_2 in the flue gases. With fuel oils and coal, these readings are higher. For this process a fuel-air ratio controller is used. This controller measures the fuel flow and changes the air flow (forced draft fan) to correspond to the rate of fuel feed, thus maintaining the desired ratio for efficient operation.

Draft

When considering draft, we think of the negative pressures in the boiler. The usual boiler is built for balanced draft and there are leaks in the setting which would allow gases to escape into the boiler room if the pressure in the boiler exceeded the pressure in the room. It is desirable to keep the furnace pressure at a point lower than the room so any leaks will be from the room into the boiler instead of in the opposite direction. For this process a draft controller is used. This controller measures the pressure in the furnace and operates the outlet damper to maintain the furnace pressure at a point slightly lower than that in the boiler room.

There are several types of systems which may be used to accomplish this control which operate pneumatically, hydraulically, or electrically.

In addition to the above controllers the following instruments are used on different functions: steam flow meters that record the load on the boiler; temperature recorder for recording the air and flue gas temperatures to assist in determining the efficiency of the boiler; CO_2 and oxygen recorders that analyze the flue gases and tell how efficiently the fuel is being burned; draft gauges that measure the pressures and drafts in the air and gas systems, and tell the condition of the boiler from the standpoint of fan operation and whether or not the boiler is becoming dirty due to soot build-up.

Other material savings

There are many types of instruments which may be used in the beet sugar industry which not only will result in reduced operating costs and material savings but also in better product control. Some of these are pH control for carbonation, sulphitation, and predefication, milk of lime control, density control of melters, humidity control for sugar storage, closed circuit T.V., and many others.

In addition to the control of the process through the use of instruments, it is possible to use automatic electrical controls at many locations. We have used many of these and have built some rather extensive push-button control panels used for automatic control of the following: limerock and coke handling, operating of the lime kiln, wash house and beet handling, beet slicers, continuous diffuser, centrifugals, bulk sugar storage and loading, and many others which time does not permit discussing at present.

I have not attempted *in* the time allotted for this paper to give any statistics of specific dollar savings through instrumentation but all of the above applications have made their contribution to the reduction of costs through instrumentation in the beet sugar industry.

In closing I wish to say that I feel that the application of instrumentation and automatic controls in the beet sugar industry is still in its infancy. The opportunity of applying automatic controls to this industry is unlimited, and if we in this industry expect to compete with rising production costs in the years ahead we must continue to use more and more automatic controls. I feel that the engineers are ready to design a fully automatic beet sugar factory as soon as they are given the opportunity to do so.

Marginal Nitrogen Deficiency of Sugar Beets and the Problem of Diagnosis

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In recent years much has been learned concerning the response of sugar beets to changes in environment. Ulrich (9)[2], in a controlled climate facility, has shown that low night temperatures coupled with nitrogen deficiency can result in beet roots with 18% sucrose. This response is also apparent with sugar beets grown in pots outdoors; plants which had been nitrogen deficient for 6 to 8 weeks during an early fall growing period, produced as much total sucrose as comparable plants well supplied with nitrogen (5). Both the above studies involved growing sugar beet plants in vermiculite and watering with culture solution. In such a system it is possible to bring about a high degree of nitrogen deficiency as evidenced by a rapid decrease in growth rate of tops and roots within three weeks after nitrogen was removed from the culture solution.

Under field conditions lesser degrees of nitrogen deficiency are likely, depending on the balance between nitrogen demand (plant growth) and nitrogen supply (rate of nitrification and the nitrogen status of soil into which roots are extending). Early experiences with nitrogen fertilization of sugar beets in California, however, also indicated that fairly sharp nitrogen deficiency responses were obtained. This reflected the low residual nitrogen fertility of the soils at that time (6, 7). Under such conditions the length of the deficiency period prior to harvest appeared to be the most important factor in determining the quality of the harvested crop. This picture appears to have changed as the result of the great increase in the use of the nitrogenous fertilizers on field crops in California. More recent experiences with fields of high residual nitrosren fertility (3, and Loomis and Worker, unpublished) have indicated that sharp nitrogen deficiencies are not obtained under such conditions and that degree of deficiency may be as important as the length of deficiency.

This paper concerns the results from the first of a series of field experiments designed to assay the degree of nitrogen deficiency in several California soils and to relate the degree of

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[2] Numbers in parentheses refer to literature cited.

deficiency to diagnostic techniques. The experiment was conducted in a field of commercial sugar beets in Kern County. In this area sugar beets are usually planted in late winter (January and February) and harvested in midsummer (July and August). Crops usually produce excellent root yields (20 to 35 tons per acre) but with low sucrose concentrations (12 to 14%).

Procedure

The field selected was on Hesperia fine sandy loam. The sugar beet variety Spreckels 202H was planted in 30-inch single-row beds in early January, 1961. Four rates of nitrogen (0, 80, 160 and 320 pounds N per acre) and five dates of harvest (May 24, June 16, July 6, 27, and August 17) were arranged in a split-block design (1). Nitrogen rates were main plots randomized in a 4 X 4 Latin square and dates of harvest were subplots 60 feet long X 4 rows wide. The subplots were randomized the full length of each column of main plots. On March 7, shortly after thinning, 80 pounds of N per acre were applied to all except control plots. On April 8, 80 and 160 pounds N per acre were applied, establishing the 160- and 240-pound rates, respectively; on June 7 an additional 80 pounds were applied to the plots which had already received 240 pounds to establish the 320-pound rate. The object was to provide N levels that would allow plants to become deficient at different times and to maintain one level where plants would remain adequately supplied all season. Fertilizer nitrogen was applied as ammonium sulfate except on June 7 when ammonium nitrate was used. Starting March 25, 15 to 20 petioles of recently matured leaves were collected at two-week intervals from the center two rows of each subplot and oven-dried for subsequent analysis for NO_3-N (2). At each harvest, beets of the center 50 feet of the two center rows of appropriate subplots were harvested. Fresh weights of roots and tops were determined and two samples of 15 roots each were taken for tare and sucrose determinations.

To determine "days of nitrogen deficiency prior to harvest," NO_3-N values of each subplot were plotted against dates. The number of days below 1000 ppm NO_3-N (dry basis) were averaged for replicates of the same nitrogen level and date of harvest.

Results

Table 1 gives the mean NO_3-N concentration in petioles for several sampling dates of subplots of each harvest and means for top yield, root yield and percent sucrose in roots for subplots of the respective harvest dates. In general the plants showed deficiency symptoms, and the concentration of NO_3-N reached the

Table 1.—Effect of nitrogen fertilization on growth of sugar beets, sucrose concentration of roots and on nitrate-nitrogen concentration of petioles of recently matured leaves. Values are means of four replications.

Fertilizer nitrogen[1] (pounds/ acre)	ppm (dry wt. basis) NO_3-N in petioles									Tons/acre fresh wt.		% Sucrose in roots
	4/8	4/22	5/5	5/21	6/3	6/16	7/6	7/26	8/16	Tops	Roots	
						Plots harvested May 24						
0	1880	62	100	130						4.6	5.5	13.9
80	12900	3810	160	160						12.7	9.8	14.0
160	12500	6540	1500	660						19.6	10.2	12.2
320	13100	7900	3430	1820						22.4	9.5	12.1
						Plots harvested June 16						
0	1880	140	110	90	150	240				6.8	11.8	15.2
80	12900	2770	160	130	150	210				13.5	17.0	15.5
160	13500	6170	1530	520	520	270				21.9	18.6	14.2
320	13200	7220	1630	1640	1100	2240				29.6	19.4	12.7
						Plots harvested July 6						
0	1010	120	120	170	190	280	250			8.5	15.3	15.1
80	14500	3000	140	240	240	220	160			12.8	21.4	15.2
160	14800	5800	1320	990	550	830	420			23.4	25.0	14.1
320	12700	6470	2560	2450	1040	3210	3650			29.8	24.2	13.0
						Plots harvested July 27						
0	890	190	130	230	200	240	250	540		8.7	17.4	14.5
80	14200	2770	180	270	260	260	150	200		13.0	26.2	14.2
160	15100	5240	910	920	320	440	290	560		19.4	30.0	13.4
320	13100	7000	1530	1450	1270	3500	3010	1700		25.8	30.5	12.4
						Plots harvested August 17						
0	1030	190	110	180	160	200	300	1500	1910	5.6	19.0	14.7
80	14100	2680	180	230	210	650	200	540	1020	9.2	27.5	14.7
160	15400	6050	1520	640	440	340	400	700	500	12.5	32.3	13.6
320	15600	6670	2700	2060	1280	3140	2800	2200	1580	16.9	31.3	12.7
	L.S.D. 5%: Among N levels for same harvest date									4.4	4.1	0.8
	Among harvest dates for same N level									3.1	3.4	0.8
	Error (e) mean squares									5.09	1.99	0.269
	F values: N x Harvest dates									4.18**	5.59**	0.78
	N levels									75.62**	18.41**	128.62**
	Harvest dates									10.36**	111.89**	14.52**

[1] March 7, 80 pounds of N/acre applied to all but O-N plots. April 8, 80 pounds and 160 pounds of N applied respectively to 160 N and 320 N plots. June 7, 80 pounds of N applied to 320 N plots.

*, ** Value exceeds that required for the 5% and 1% level of signifiance respectively.

critical level, about 1,000 ppm (10), in early April for ON plots, about May 3 for plants of 80 N plots and about May 15 for plants receiving 160 N. Plants fertilized with 320 N remained green and the NO_3-N content of their petioles remained above the critical level throughout the season.

An anomalous situation arose in connection with three subplots harvested on August 17. The NO_3-N concentration in petioles of one of the ON subplots increased rapidly from 325 ppm on June 23 to 670 on July 6, to 5570 on July 26 and to 6830 on August 16. Similarly, the concentration of NO_3-N in two subplots of the 80 N rate rose from an average of 230 ppm on July 6 to 905 on July 26 and 1865 on August 16. The reasons for these increases cannot be precisely explained but were probably due to a sudden increase in soil nitrification in these plots. The result was a reduction in the sucrose concentration in the roots of plants harvested from these plots on August 17 and, therefore, a somewhat lower average sucrose concentration for the O and 80 N rates of this harvest date than would have been the case otherwise.

As top and root production indicate (Table 1) there was a marked response to nitrogen fertilization. Of particular interest is the rapid rate of root growth despite nitrogen deficiencies. As Figure 1 indicates, plants that were unfertilized grew at the rate of 1.1 tons of roots/acre week from June 16 to July 27; those receiving 80 pounds of N/acre grew at the rate of 1.5 tons/acre week. Plants receiving 160 and 320 N had the same root growth during this period of ca. 1.8 tons/acre week despite the fact that plants of the 160 N rate were nitrogen deficient throughout the harvest period while those of the 320 N rate were not..

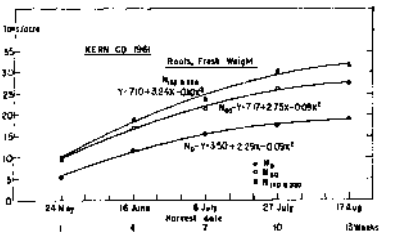
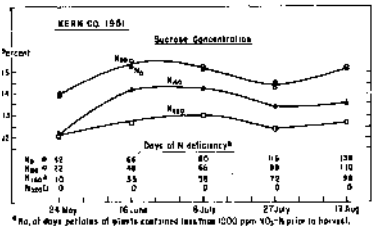


Figure 1.—Root growth as influenced by nitrogen fertilization and time of harvest. $N_{160 \text{ and } 320}$ are means of the combined nitrogen treatments. X in regression equations is the week of harvest.



[a] No. of days petioles of plants contained less than 1000 ppm NO_3-N prior to harvest.

Figure 2.—Sucrose content of beet roots related to time of harvest, nitrogen fertilization and duration of nitrogen deficiency prior to harvest.

Figure 2 illustrates the time course of changes in sucrose concentration as influenced by nitrogen fertilization, and gives the associated days of nitrogen deficiency indicated by petiole analysis. Values for the three high nitrogen plots of the zero and 80 N rates were not included in plotting sucrose concentrations for the August 17 harvest and thus the values shown in the figure more nearly represent a typical situation. Plants not fertilized and those receiving 80 N had essentially the same sucrose concentrations. Both attained maximum concentrations of ca. 15% in mid-June after periods of nitrogen deficiencies of 66 and 46 days respectively. With 160 N the peak sucrose concentration also occurred in mid-June but at a lower level (ca. 14%) and after only 35 days of N deficiency. Thus, extending nitrogen deficiency beyond June 16 did not result in further increases in sucrose concentration suggesting that climate had an overriding influence. The sucrose concentration of roots of plants fertilized with 320 N did not change greatly throughout the season but was highest in early July. There was a general decline in sucrose concentration for all N rates at the July 27 harvest and a partial recovery by August 17.

Figure 3 gives the average weekly maximum and minimum air temperatures (11) and indicates a sharp rise in day and night temperatures in mid-June.

Discussion

From a practical point of view the most prominent feature of responses of sugar beets to nitrogen deficiency is the increase in the sucrose percentage in storage roots. The change may be visualized as resulting from inhibition of vegetative growth which permits a higher proportion of the sucrose produced in the leaves to accumulate in the roots rather than be utilized in growth

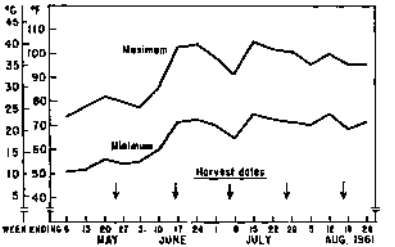


Figure 3.—Weekly mean temperatures, Kern County Airport. Data of U. S. Weather Station.

processes. The degree of the shift in this growth-storage balance is dependent upon many factors. Thus, the increase in sucrose concentration is dependent upon the length and degree of the deficiency, root size, and the amount of photosynthesis, as well as the temperature regime under which the response is studied.

In pot experiments, where the degree of the nitrogen deficiency can be controlled, extreme values may be obtained. Maximum sucrose concentration is usually obtained within 6 to 8 weeks after the beginning of the nitrogen deficiency and increases in sucrose have been found to be inversely proportional to the initial root size (4). The present field trial typifies the kind of nitrogen deficiency responses which are commonly observed on fields with high residual nitrogen. Under these conditions the sugar beet plants continue to make rapid root growth indicating that although they are deficient in nitrogen the degree of deficiency is slight, and the plants appear to be receiving a high percentage of the nitrogen that they require for maximum growth. The results of the present trial clearly indicate that the degree of deficiency is as important or more important than the length of the deficiency period under such conditions. This is particularly evident in the fact that roots of the nitrogen deficient plants of the 160 N rate grew as rapidly as did those fertilized with 320 N.

At present such a situation can be assessed only by observing nitrogen deficiency responses, i.e., by measuring crop growth. Soil analysis procedures which would predict the nitrogen supplying power of a soil, or the rate at which nitrogen might be supplied do not exist. While plant analysis, utilizing the average content of nitrate nitrogen in a group of petioles, serves admir-

ably to predict and measure the date of nitrogen deficiency, it does not indicate the degree of the deficiency. What appears to be needed is a modification of procedure or an additional diagnostic tool that will easily measure the degree of deficiency a particular crop is experiencing. One modification, although an expensive one, would be to analyze petioles separately from individual beets (8), thereby assessing the degree of deficiency among plants composing the grouped petiole sample. With the present technique of determining the NO_3-N content of a composited group of petioles, a value below the critical level of 1,000 ppm can be obtained when 50% or more of the petioles in the sample contain 1,000 or more ppm NO_3-N. This is due to the considerable variability that occurs from plant to plant in a field (8). In such a case the degree of deficiency is less than when a larger percentage of the petioles are below the critical value. Another possibility would be to analyze for other forms of nitrogen; e.g., soluble nitrogen in blade or petiole tissue might more clearly indicate the degree of deficiency.

This experiment affords an interesting comparison of the effect of fertilizer nitrogen on top growth compared to root growth (Table 1). As early as June 16, plants that were fertilized with 320 N produced more tops than those that received 160 N. This growth differential continued through the last harvest on August 17, yet the higher N rate never resulted in more root growth. Thus when soil nitrogen is low it appears that roots take precedence over tops for the use of nitrogen in growth.

Several other important practical conclusions may be drawn from the present experiment. It is of interest to compare the control plants which received no supplemental nitrogen with those that were fertilized. The nonfertilized plants were nitrogen deficient for only about three weeks longer than plants receiving 80 N and about 4 weeks longer than plants receiving 160 N (Table 1) yet their growth rate (Figure 1) during the period of nitrogen deficiency was much less than the fertilized plants. There are many possible explanations for this occurrence, two of which are worth mentioning at this time. The O-N plants became nitrogen deficient about April 10, just as the crop was beginning to make its most rapid growth, as a result these plants never achieved good foliage development, and thus appeared to have insufficient photosynthetic area to support the crop during the subsequent growth at low nitrogen. In addition, plants in these plots may have had poor fibrous root development, and thus did not have access to nitrogen released by the soil during the summer period.

Another important observation relates to the magnitude of the increases in sucrose concentrations which were observed. The plants which received zero and 80 N became nitrogen deficient in April and the small roots rapidly increased in sucrose concentration to over 15%; whereas plants of the 160 N plots which became nitrogen deficient in mid-May with a much larger root size, and with tops and roots growing at a more rapid rate attained a lower maximum of 14.2%. It appears that after June 16 temperature had an overriding effect and the combination of rapid root growth and reduced photosynthetic surfaces prevented further gains in the sucrose concentration of nitrogen deficient plants.

Summary

A field experiment involving four rates of nitrogen fertilization and five dates of harvest was conducted to determine how long sugar beets should be deficient in nitrogen prior to harvest to attain high sucrose concentrations. The sucrose content of roots did not exceed 15.5% even though some plants were deficient for 139 days prior to the last harvest on August 17. With an onset of nitrogen deficiency, maximum sucrose contents were reached in from 4 to 6 weeks. The failure to attain high sucrose concentrations in roots was related to high temperatures, rapid rates of root growth and reduced photosynthetic surfaces of nitrogen deficient plants.

Midseason nitrogen deficiences were readily detected by petiole analyses. However, there was little or no effect of such a deficiency on the rate of root growth indicating that the plants were taking up most of the nitrogen they needed for maximum root growth. Such results indicate the desirability of modifying current procedures or finding a new diagnostic tool to more accurately reflect the degree of nitrogen deficiency.

Acknowledgement

We thank Mr. N. L. Ritchey for furnishing the land for this experiment and for carrying out the cultural practices and the Spreckels Sugar Company for assistance in harvesting and for laboratory analyses of root samples.

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Cultural and Pathogenic Studies of an Isolate of Cercospora beticola Sacc.[1]

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Introduction

Cercospora leaf spot, which is caused by *Cercospora beticola* Sacc. is one of the major problems of sugar beet cultivation. In 1959 and again in 1961 the incidence of leaf spotting of beets was considerably above the usual average of infection in southwestern Ontario and caused renewed interest in the chemical control of this disease. A reliable method of producing conidia was required to supply spores for bioassay and greenhouse tests. Beet leaf agar was reported by Nagel (6)[3] and Vestal (9) to be a satisfactory medium for the growth and sporulation of *C. beticola.* Consequently an isolate obtained from locally-grown infected sugar beets was studied in culture on beet leaf and other agar media.

The incidence of Cercospora leaf spot is of economic concern mainly to the sugar beet industry. However, since cultures isolated from infected sugar beets in other seasons (1,9) were pathogenic to related plants the present isolate was tested against several varieties of sugar beet, mangel and table beet.

The results of field trials for the control of Cercospora leaf spot of sugar beets with protective fungicides have been published separately (2).

Methods and Materials

An isolate of *C. beticola* was obtained from an infected leaf of sugar beet[4] at London, Ontario and maintained on beet leaf agar prepared as follows. A hot water extract (15 minutes boiling) was prepared from 200 g fresh weight of field-grown sugar beet leaves. This was diluted to 1000 ml with distilled water, dispensed and sterilized in 100 nil lots. The beet leaf medium contained 50 ml of the extract, 20 g dextrose and 15 g agar per liter.

Cultures for heavy spore production and for study of the effect of temperature and medium on spore production were prepared by cutting out a 9 mm disk from the center of the agar layer in a 100 mm Petri plate with a sterile cork borer and replacing this with a disk from an established culture of *C. beticola* growing on beet leaf agar.

[1] Contribution #224 from Research Institute. Canada Department of Agriculture, University Sub Post Office, London, Ontario, Canada.

[3] Numbers in parentheses refer to literature cited.

[4] Grown from scarified multigerm seed as supplied by Canada and Dominion Sugar Company, Chatham. Ontario.

The method of Ludwig et al. (4) involving washing the plate cultures for 24 hours in running water and inverting them in a slanted position, was found ideal for spore production after the mycelium had grown over about 5/6 of the surface of the agar medium.

Results

Figure 1 shows an individual leaf and Figure 2 an entire plant infected with Cercospora leaf spot. The infections are isolated on the leaf (Figure 1) with little or no coalescing; several leaves are dead, brown and shrivelled in the entire plant (Figure 2).

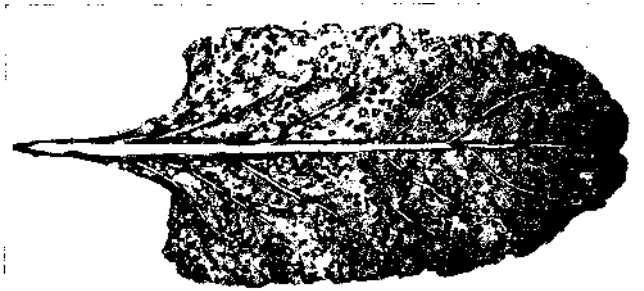

Figure 1.—Cercospora leaf spot infection on leaf of multigerm sugar beet.

Figure 2.—Cercospora leaf spot on entire plant showing advanced stage of the disease with several leaves dead and shrivelled.

The infection sites on the shrivelled leaves are excellent sources of conidia for the airborne dispersion of this organism whenever the relative humidity is high. Our isolate was obtained from a site of this type.

Typical conidia produced on beet leaf agar are illustrated in Figure 3. These were colorless and contained up to 16 septations.

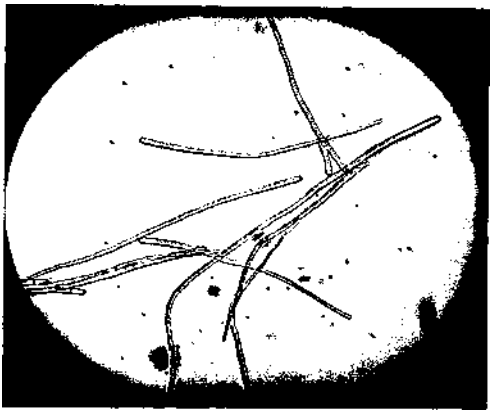

Figure 3.—Representative conidia of *C. beticola* producde in culture on beet leaf agar.

An attempt was made to produce large numbers of C. *beticola* conidia by agar culture. Consequently a comparison was made of the type of culture and yield of conidia produced on four media; peptone agar (PA), potato dextrose agar (PDA), V-8 juice agar (V-8A) and beet leaf agar (BLA). Figure 4 illustrates the difference in colony type and diameter after 22 days incubation at 22 C. The average diameters of 10 colonies were 79.0 mm for PDA, 72.4 mm for BLA, 31.0 mm for PA and 68.4 mm for V-8A. Differences in the development of the cultures are evident. For example in the PA cultures the rate of growth is obviously low compared with the rate in the other media and there is a ring of dense white mycelium at the periphery. The central part of the culture had gray-green mycelium. The PDA culture had an outer gray-green ring with white mycelium toward the center. The V-8A culture had an outer gray-green ring with a circle of dense white mycelium toward the center. There was much sectoring of the cultures with this medium. A sector is obvious

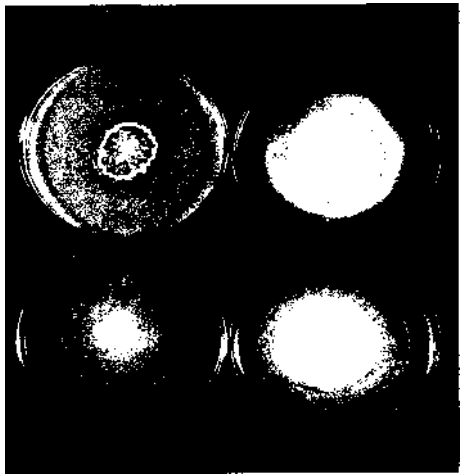

Figure 4.—Cultures of *C. beticola* on peptone agar (upper left), potato dextrose agar (upper right), beet leaf agar (lower left) and V-8 juice agar (lower right).

at the lower left hand edge of the V-8A culture in Figure 4. The best medium for our purpose would be the one producing the greatest number of conidia in the shortest time. Table 1 shows that the BLA was the best medium of those tested for the production of conidia.

The optimum temperature for use with the BLA was determined by comparing rates of culture growth at 18, 22, 25 and 30 C over a period of 27 days. The fastest growth was obtained (Figure 5) at 25 C but the rate of 22 C was only slightly lower. Conidia were harvested from the cultures at 22 and 25 C (12 plates of each, 27 days old) and it was found that when the conidia were suspended in 200 ml that there were between 20- and 30,000 conidia per ml of liquid from cultures at each of the two temperatures.

Table 1.—Sporulation of an isolate of *C. beticola* in culture.

Culture medium	Conidia per ml[1]
Beet leaf agar	40-50,000
Porato dextrose agar	10-20,000
V-8 juice agar	5,000
Peptone agar	5,000

[1] Total from twelve 27-day-old colonies in 200 ml water.

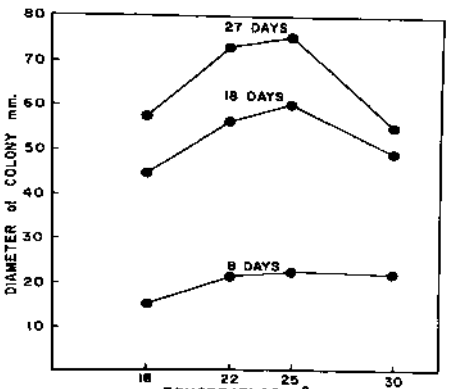

Figure 5.—Growth of *C. beticola* for 27 days on beet leaf agar at 18, 22, 25 and 30 C.

Table 2.—Reaction of various mangel, table beet and sugar beet varieties to *C. beticola.*

	Variety	Normal color of stem	Normal color of leaf	Color of infection site	Wilting due to infection
Mangel	Mammoth long red	red	red veined	red circle	no
Mangel	Giant white sugar	light pink	green	gray	no
Mangel	Yellow globe	yellow or green	green	gray	no
Mangel	Yellow leviathan	yellow	green	gray	no
Mangel	Red leviathan	red	light red or green vein	red or gray circle	no
Mangel	Sludstrap	yellow	green	gray	yes
Table beet	Early flat Egyptian	red	red veined	red circle	no
Table beet	Ruby queen	red	red veined	red circle	no
Table beet	Detroit	red	red veined	red circle	no
Table beet	Asgrow conner	red	red veined	red circle	no
Table beet	Long dark red	red	red veined	red circle	no
Table beet	Early wonder	red	red veined	red circle	no
Sugar beet	C & D[1] scarified 1960	light pink	green	gray	no
Sugar beet	Giant sugar	light red	green	gray	no
Sugar beet	C & D monogerm	light pink, green	green	gray	no
Sugar beet	Czechoslovakian	green	green	gray	yes

[1] C & D = Canada and Dominion Sugar Company

Since this work was conducted with a *C. beticola* isolate from sugar beets in an area not normally seriously affected by this disease, the pathogenicity of this culture was also tested on six varieties of mangel, 6 of table beet and 4 of sugar beet to note any indication of resistance to the organism in any of the varieties.

Table 2 records the color of leaf, stem and infection site. Whenever there is red or pink color normally in the leaf or stem, there is a red circle formed about the infection site. Conversely when the stem and leaf have no red coloring there is a gray circle about the infection. None of the plants tested had resistance to the Cercospora. The mangel Sludstrap and the Czechoslovakia[5] strain of sugar beet wilted as a response to the infection whereas the other varieties did not. This is considered to mean that the aforementioned two varieties are especially susceptible to this isolate of *C. beticola.*

Discussion

Noll (7) found that isolates of *C. beticola* tended to produce variants as 'islands' of whitish, yellowish, pink or abundant white aerial growth. The isolate used in this study produced variants also, most frequently in the cultures on V-8 agar. They were all of the whitish or normal type in color; the white ones producing fewer conidia per given area of culture than was normal. A white variant was stable in that on transfer it produced an entire colony of white mycelium of low conidia production.

Canova (1) in a study of the biology and epidemiology of *C. beticola* found that infection was less active at 30 C than at 25 C and that infection was more active in mature than in young or old leaves. However Vestal (9) and the present authors were able to get heavy infections on young leaves of sugar beet by using a heavy suspension of conidia produced in laboratory culture. Incubation for three days after inoculation at 25 C, 90 to 92% relative humidity, and low intensity illumination fluorescent light was satisfactory.

All of the sugar beet, mangel and table beet varieties tested were susceptible to our *C. beticola* isolate. Vestal (9) has recorded that many weeds found in or around sugar beet fields were rather susceptible to this organism. *Chenopodium album, Amaranthus retroflexus, Malva rotundifolia. Plantago major, Arctium lappa* and *Lactuca sativa* were all easily infected in his tests. Plants of *Plantago major* in our field plot area in 1961 (2) were infected with *C. beticola.* Clearly these host plants could be a serious reservoir of inoculum able to carry the organism through a long period in the absence of sugar beets.

Cerospora leaf spotting is thought to be favored by high temperatures but Hull (3) and Mischke (5) have stated that a minimum temperature of 10 C at night and a minimum of 20 C during the day were favorable to the development of the disease.

[5] The seed of Czechoslovakian sugar beet was obtained from the Canada and Dominion Sugar Company, Chatham, Ontario.

Mischke (5) further concluded from his experimental results that a critical period was reached in a developing infection of sugar beet in the field when there were at least 10 lesions on about 5% of the plants; 3 days or more with relative humidity above 95% for at least 10 hours within the crop; and a minimum temperature in the crop of 10 C, even at night. There is no reason to believe that Mischke's rules for forecasting would not be accurate in southwestern Ontario.

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Yield and Quality of Sugar Beets as Affected by Cropping Systems

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Cropping systems influence yield and quality of sugar beets primarily by affecting the soil's nutrient supplying power and physical properties, and the plant diseases and pests transmitted by the soil. Before the advent of inexpensive commercial fertilizers, the effect of a cropping system on the nutrient supplying capacity was very important. Now, however, all necessary nutrients can be supplied through the use of commercial fertilizers and the other effects of cropping systems need to be evaluated.

Many previous rotation experiments measured the combined effects of the rotation on yield and did not attempt to determine the various factors which affected the yield. In the Morrow rotation plots of Illinois the yield of the continuous corn rotation without nitrogen fertilizer was 22 bushels per acre as compared with 109 bushels per acre for the three-year rotation with a legume before corn. However, recently the experiment was modified and this yield difference was overcome in one year by a heavy application of nitrogen fertilizer (l)[2]. The beneficial effects of alfalfa on succeeding crops have been observed by agronomists (3, 4, 6) on irrigated western soils. Gardner and Robertson (3), however, showed that the major effect of alfalfa on succeeding crops was to increase available nitrogen.

The soils of Imperial Valley are alluvial soils, low in organic matter and nitrogen, and poor in physical structure. To maintain or improve these factors, alfalfa, sesbania (an annual summer legume) and steer manure are commonly used in cropping systems. A field experiment was initiated in 1956 to evaluate the effectiveness of these cropping systems for improving physical properties and increasing soil nitrogen. Only the effect of the cropping systems on the supply of soil nitrogen and its effect on the sugar beet crop are discussed in this paper.

Methods and Materials

The experiment was conducted on a Holtville silty clay, stratified phase, at the Southwestern Irrigation Field Station located in the Imperial Valley near Brawley, California. The plot area had not been manured or planted to alfalfa for over 10 years. In the upper 8 inches of soil the organic matter content

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[2] Numbers in parentheses refer to literature cited.

was about 1% and the nitrogen content about 0.065%. Although phosphate is not limiting to plant growth on this soil, 240 pounds of P_2O_5 per acre were applied in 1956 and 80 pounds of P_2O_5 per acre in 1958.

The experimental design used was a split-plot randomized block with four replications. The cropping systems used as main plots for the first two years of the study are given in Table 1. In the third year of the experiment the entire area was plowed and planted to US 75 sugar beets in September, 1958. Each cropping-system main plot was subdivided into 6 subplots which received the following rates of nitrogen: 0, 60, 120, 180, 300. and 420 pounds per acre. The nitrogen was applied as ammonium nitrate, one third at planting and two thirds at thinning time.

Table 1.—Description of the cropping system treatments used prior to the uniform sugar beet crop planted in September, 1958.

Treatment No.	1956 - 57	1957 - 58
1	Sugar beets (0 lb N)	Barley (0 lb N)
2	Sugar beets (160 lb N)	Barley (80 lb N)
3	Sugar beets (320 lb N)	Barley (160 lb N)
4	Alfalfa	Alfalfa
5	Sugar beets (160 lb N) Sesbania[1]	Barley (80 lb N) Sesbania[1]
6	Sugar beets (160 lb N) + 10 tons manure	Barley (80 lb N) + 10 tons manure

[1] Sesbania *(Sesbania macrocarpa)* is a legume and was grown as a summer green manure crop.

At harvest, June 1959, a 15- to 20-beet sample was taken from each plot to determine sucrose percentage and purity. Sucrose percentage was determined with a saccharimeter using; a method of the Association of Official Agricultural Chemists (2). Total soluble solids for calculating purity were determined with a Brix spindle hydrometer on an aqueous extract of the sugar beet pulp.

Results and Discussion

Cropping systems influenced yield and qualitv of sugar beets by their effect on the nitrogen-supplying ability of the soil during the cropping season. Large amounts of nitrogenous organic matter were added to the soil by cropping systems that included alfalfa (Treatment No. 4), sesbania, (Treatment No. 5) or steer manure (Treatment No. 6). The other systems added only inorganic fertilizer nitrogen (Treatment No. 2 and 3) or no nitrogen (Treatment No. 1). Figure 1 shows how the yield and quality of the sugar beets from all of the cropping systems varied as the rate of nitrogen fertilization increased.

With no additional fertilizer nitrogen, treatments 4, 5 and 6, which added organic matter to the soil, produced marked increases in yield over treatments 1, 2 and 3, which added relatively little organic matter to the soil. The yield of sugar beets from

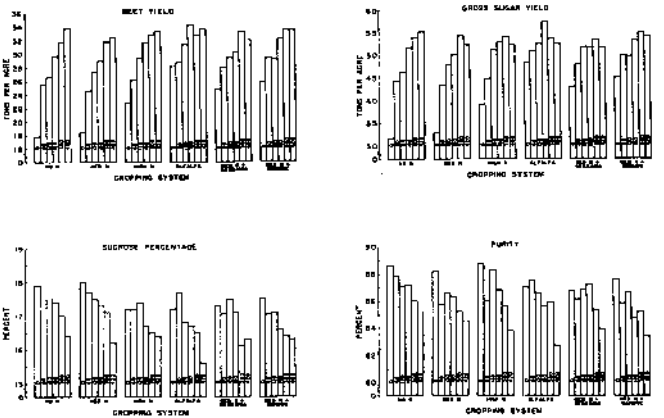

Figure 1.—The effect of different cropping systems on yield and quality of sugar beets in Imperial Valley during the 1958-59 season. Cropping systems designations from left to right refer to treatments 1-6 in Table 1. The LSD at the 5% level of significance is 2.5 tons/acre for yield of beets, 0.42 tons/acre for gross sugar, 1.52% for sucrose percentage, and 2.5% for purity.

the alfalfa treatment was 58% higher than that from the treatment that had received no nitrogen during the preceding two years. The sesbania and manure treatments resulted in a 33 and 45% higher yield, respectively, than the no-nitrogen treatment. The cropping system that had received a high rate of inorganic nitrogen fertilizer (Treatment No. 3) during the preceding two years resulted in a 23% increase in yield.

When additional nitrogen was supplied in the form of fertilizer to the 1958-59 sugar beet crop, the differences in yield among the different cropping systems were decreased. At the 180 pound per acre rate of nitrogen fertilization, the alfalfa treatment yielded only 15% more sugar beets than treatment 1. The yields of the other treatments at this rate of fertilization were not significantly different from each other or from treatment 1 at the 5% level. When 420 pounds of nitrogen per acre were used, the yields of all treatments were practically the same. Only 180 pounds of nitrogen per acre were required to maximize the yields with the alfalfa treatment but 420 pounds of nitrogen per acre were needed to produce the maximum yield in treatment 1.

The cropping system that resulted in the highest yields of sugar beets tended to produce the lowest sucrose percentage and purity. Also, as the amount of nitrogen fertilizer applied was increased the sucrose percentage and purity decreased. Loomis and Ulrich (5) showed that nitrogen nutrition affects the quality of sugar beets. Ample nitrogen throughout the season depresses sucrose percentage whereas beets deficient in nitrogen have a high sucrose content. The results of this experiment indicate that cropping systems 4, 5, and 6 supplied additional nitrogen to the beets. At the higher rates of nitrogen fertilization this additional nitrogen tended to inhibit sugar yield, and depress sucrose percentage and purity.

The nitrogen status of sugar beets throughout the season can be followed very closely by measuring the nitrate concentration in the beet petioles (7). Results of the analysis of sugar beet petiole samples from the alfalfa system and the no-nitrogen system are shown in Figure 2. The availability of additional nitrogen to the sugar beets from the alfalfa system resulted in more nitrate at all dates for comparable nitrogen fertilizer levels. Depletion of petiole nitrate concentrations to the 1000 ppm critical level was delayed approximately one month at each nitrogen rate by the alfalfa cropping system.

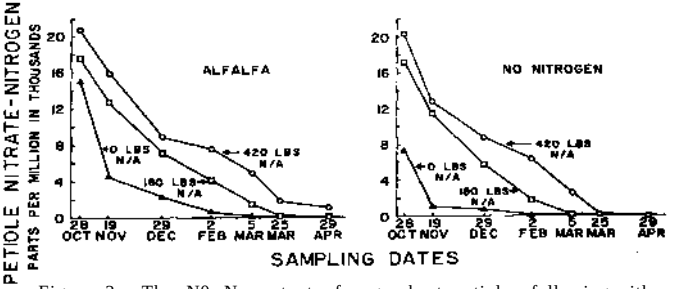

Figure 2.—The NO_3-N content of sugar beet petioles following either alfalfa or two years of beets and barley with no nitrogen.

Summary

From these results it may be concluded that cropping systems influence yield and quality of sugar beets on Holtville silty clay by their influence on the availability and supply of soil nitrogen. The cropping systems which added nitrogenous organic matter or had residual nitrogen from high fertilizer applications in-

creased the supply of available soil nitrogen and increased sugar beet yields, especially at low rates of applied nitrogen. However, these differences in yield due to cropping systms were overcome by the application of additional nitrogen fertilizer. At 420 pounds of nitrogen per acre there was no significant difference in yield for any cropping system. This indicates that the benefits from alfalfa, sesbania or steer manure were mainly due to the addition of nitrogen to this soil. The nitrogen from these organic sources had no apparent advantage over inorganic fertilizer nitrogen. Furthermore, any benefits from these treatments other than nitrogen, were not reflected in yield or quality of sugar beets.

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Growth Rate of Young Sugar Beet Roots as a Measure of Resistance to Virus Yellows

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Introduction

Considerable time may be required to obtain substantial results in breeding for resistance to virus yellows. The lack of criteria for an accurate determination of resistance in individual beets and the apparent absence of wide ranges of variation in resistance in the genetic material available for selection are responsible for this situation (1)[2]. Field tests are unsuitable for precise evaluation of slight differences in resistance to virus yellows among selections. Field evaluation is difficult because of yearly variation in climate, soil fertility, soil moisture and the spread of other virus diseases. A breeding program for increased resistance to virus yellows would be greatly facilitated, therefore, if a more accurate method of determining the relative resistance of beets to yellows were available.

This paper reports experiments, conducted in the greenhouse under controlled conditions, which indicate that the reduction in growth rate of the roots of inoculated plants during an early period of development may be useful in evaluating resistance to virus yellows.

Methods, Results and Discussion

Four boxes were lined with polyethylene and filled with sterilized sand. Seed of variety US 75 was planted on 6-inch centers April 24, and watered with Hoagland's solution containing 100 ppm of nitrogen. Forty days after emergence the plants in two of the boxes were inoculated with a virulent strain of the yellows virus. Sixteen healthy plants were removed from the boxes 3, 6, 8, and 10 weeks after inoculation and the root weights determined. Roots of the infected plants were harvested 8 and 10 weeks after inoculation and their weights determined.

In a similar test, started two weeks later, 100 plants were grown in one large box in another area of the greenhouse and watered with Hoagland's solution. The plants in this test were inoculated 40 days after emergence with the virus strain used in the first test. Half of the plants were removed and the roots were weighed 8 weeks after inoculation; the remaining roots were weighed 11 weeks after inoculation. The results of these two tests are shown in Figure 1 and Table 1.

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[2] Numbers in parentheses refer to literature cited.

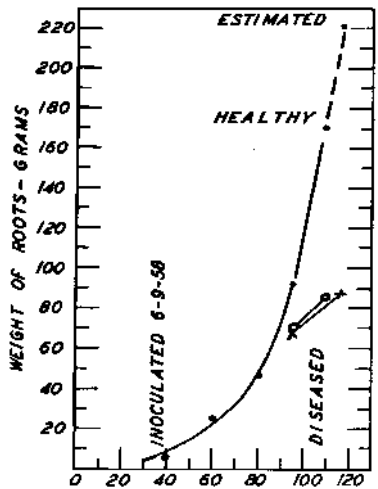

Figure 1.—Growth curve of roots of young healthy and yellows-infected sugar beet plants.

Table 1.—Growth rate of roots of healthy and yellows-inoculated sugar beet plants when inoculated 40 days after emergence.

Growth period from emergence		Growth rate per day	Index of resistance growth rate: Inoc./H × 100
From	To		
Day	Day	Gm	
Healthy Plants (H)			
40	61	0.90	
61	82	1.03	
82	96	3.21	
96	110	5.25	
Inoculated plants: Tests 1 and 2			
96	110	1.07	20
96	117	0.90	17

Leaf samples, taken from the mature leaves of each plant at the time the roots were harvested, showed the amino acid pattern for both the healthy and infected plants of that found in plants of other tests (2).

Under the conditions of these experiments, rapid growth of the roots of the healthy plants started approximately 60 to 80 days after emergence (Figure 1). During the 2-week period (beginning with the 82nd day after emergence) the mean growth rate of the roots of the healthy plants was 3.21 grams per day. The growth rate for the following 2-week period was 5.25 grams per day for the roots of the healthy plants as compared to 1.07 grams per day for the roots of the infected plants in test 1. This amounts to a reduction in the growth rate of 80% due to the disease.

In the second test, the growth rate of the infected roots for the 3-week period, (beginning with the 96th day after emergence) was 0.90 gram per day. This amounts to a reduction in the growth rate of the roots of the infected plants of 83%.

In both tests the reduction in root weight, due to the disease, was approximately 24% at the end of 8 weeks after inoculation. When the infected plants were allowed to grow 2 weeks longer the reduction in root weight of the plants in the first test was approximately 49%. By extending the growth curve of the healthy plants 7 more days, at the established rate of 5.25 grams per day, an estimated weight of 225 grams was obtained for the healthy roots. This weight as compared to 88 grams for the diseased roots shows a reduction of approximately 60% due to the disease.

The reduction in the growth rate of roots of infected plants in the early stages of growth may be an accurate criterion for the determination of resistance of selections to virus yellows. It would be necessary to make the measurements under standardized conditions nearly optimum not only for the growth of the plants but for the expression of symptoms of the disease and during the period when the virus is exerting its maximum influence on the growth of the plant. This period would be when the plant is in the acute stage of the disease. During this period both top and root growth are greatly retarded.

There is evidence that root growth may be retarded for a longer period than the top growth. The reduction in growth rate may depend upon several factors such as age of plants at the time of infection, strain of the virus used and upon the growing conditions. It is possible also that both the resistant and susceptible plants may show the same initial violet reaction to

infection but that resistant plants are able to recover from the acute stage of the disease and resume more nearly normal top and root growth in a shorter period of time than the susceptible plants.

In the tests reported, the growth rate of the roots of the infected plants was determined during the period when the plants were in the acute stage of the disease. Plants inoculated in the 4- to 6-leaf stage (40 days after emergence) and allowed a 77-day growing period before the root weights were taken resulted in a 60% reduction in root weight compared to the roots of healthy control plants. Bennett (1) reported that, in field tests, inoculation of plants in the 12- to 16-leaf stage resulted in a reduction of 34.1% in root weight, whereas inoculation 49 days later resulted in only a 12.5% reduction.

The growth rate of roots of young plants of selections made from US 75 for resistance to virus yellows and the parent was determined by essentially the same method as described. In this test the plants were inoculated with a virulent strain of the virus 60 days after emergence and root weights taken 90 and 111 days after emergence. Two selections having a growth rate superior to that of the parent and another selection which appeared to recover sooner from the acute stage of the disease were tested along with the parent in a replicated field test. The plants in the field test were inoculated in the 4- to 6-leaf stage with the same virulent strain of the yellows virus used in growth-rate test conducted in the greenhouse. The percentage increases in the growth rate of the roots and in yield per acre of roots in the field test are shown in Table 2.

Table 2.—Growth rate of roots of young inoculated greenhouse-grown beet plants of selections in relation to their yield in a replicated field test under severe yellows conditions.

Selection	Growth rate per day	Increase over parent	
		Greenhouse growth rate	Field test root yield
	Gm	Percent	Percent
US 75 (Parent)	1.56		
91DS-9	1.88	20.5	33.3
91DS-23	2.14	37.2	19.3
91DS-22	1.59	1.9	18.0

Two selections showing a superior growth rate of roots among the young inoculated plants yielded 33 and 19% more beets per acre than the parent in a replicated field test. The selection which appeared to recover from the acute stage of the disease sooner than the parent, yielded 18% more than the parent.

If the growth rate, of roots of healthy plants of a suitable variety, was determined under standardized conditions, the value could be used for comparison of the growth rates of roots of infected plants of all selections tested under the same conditions. The ratio (multiplied by 100) of the growth rate of the roots of infected plants, of the selection tested, to the growth rate of the roots of the healthy standard may be called the "relative resistance index" of the selection. For example, if US 75, having a growth rate of 5.25 grams per day for roots of healthy plants (Table 1), is taken as the standard and the growth rate of the roots of infected plants is taken as 1.07 grams per day, then the resistance index would be 20. A relative resistance index of 100 would indicate that the disease had no effect on the growth rate of the roots under the conditions set up. The "absolute resistance index" of a selection would be the ratio of the growth rate of roots of infected plants to the growth rate of roots of healthy plants of the same selection. Using this criterion as a measure of resistance to virus yellows, US 75 would have an "absolute resistance index" of 20 also.

Further tests are necessary to establish the optimum length of the growing period before and after inoculation of the young plants and the length of the interval during which the growth rate of the roots is determined, in order to more clearly identify those selections which may prove to be only slightly superior to the parent under severe yellows conditions in the field.

Summary

Sugar beet plants were grown in sand and watered with Hoagland's solution containing 100 ppm of nitrogen in tests designed to measure the growth rates of roots of healthy and of yellows-infected plants during the early stages of growth. Inoculated plants grown with this concentration of nitrogen showed typical symptoms of virus yellows including necrosis. The amino acid pattern in the leaves was typical for the healthy and infected plants. In two tests, during the growing period from the 96th to the 117th day after emergence, the growth rate of the roots of the infected plants was reduced 80 and 83%, respectively, as compared with that of healthy plants. The over-all reduction in the growth rate of the roots of the infected plants for the 117 days, after emergence resulted *in* a 60% reduction in the weight of the roots. The tests indicate that an accurate evaluation of selections as to their relative resistance to virus yellows may be obtained in approximately 120 days after emergence. The ratio of the growth rate of roots of infected plants of a selection to the growth rate of roots of healthy plants of a selection used as

a standard is suggested as a numerical value which would serve as the "relative resistance index" of the selection in question to virus yellows. Sugar beet selections having a growth rate superior to that of the parent in young inoculated plants grown in the greenhouse, outyielded the parent in a replicated field test under severe yellows conditions.

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Occurrence of Yellows Resistance in the Sugar Beet With an Appraisal of the Opportunities for Developing Resistant Varieties

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Introduction

Beet yellows is a virus disease which occurs in nearly all countries where the sugar beet is grown. In the United States the disease causes serious losses in California and in the Salt River Valley of Arizona. Bennett, Price, and McFarlane (3)[3] found that beet yellows reduced root yields from 13.8 to 53.0% and sucrose content from 0.4 to 2.2 percentage points. Seed yields of commercial sugar beet varieties were reduced as much as 34.9% in Arizona (7) and 44.6% at Salinas, California (2).

Beet western yellows (radish yellows) described by Duffus (4) also causes a yellowing of beets which is difficult to distinguish from yellowing induced by the less virulent strains of beet-yellows virus. Duffus (5) found that western yellows caused losses which were additive to losses produced by beet yellows when the 2 diseases occurred simultaneously. Western yellows is present in most of the beet-producing areas of western United States and in most of these areas more beets are affected by this disease than by beet yellows.

Progress in breeding for resistance to beet yellows has been reported from Europe. In the Netherlands, breeding work has been in progress since 1948 and selections have been developed in which yield reductions do not exceed 14 to 16% (8). Information is not available on resistance of these selections to western yellows.

Nine wild species of *Beta* have been tested for susceptibility to beet yellows (1). Symptoms were produced on all these species and no evidence of a high decree of resistance was found. Some species including *B. macrocarpa* Guss., *B. maritima* L., and *B. patellaris* Moq. were more severely injured than commercial varieties of sugar beet. It seems unlikely that any of the species tested will be of value in a program of breeding for resistance

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[3] Numbers in parentheses refer to literature cited.

to beet yellows. The resistance of wild species of *Beta* to western yellows has not been determined.

Experimental Methods

Replicated field tests were made at Salinas in 1957 and 1958 to determine the relative resistance of our present varieties and breeding stocks to beet yellows. The degree of resistance to yellows was determined by comparing inoculated and noninoculated plots of each variety or breeding stock (Figure 1). Inoculations were made with a virulent strain of the beet-yellows virus by the method described by Bennett, Price, and McFarlane (3) in which leaf pieces containing about 10 green peach aphids, *Myzus persicae* (Sulz.), were removed from source plants and

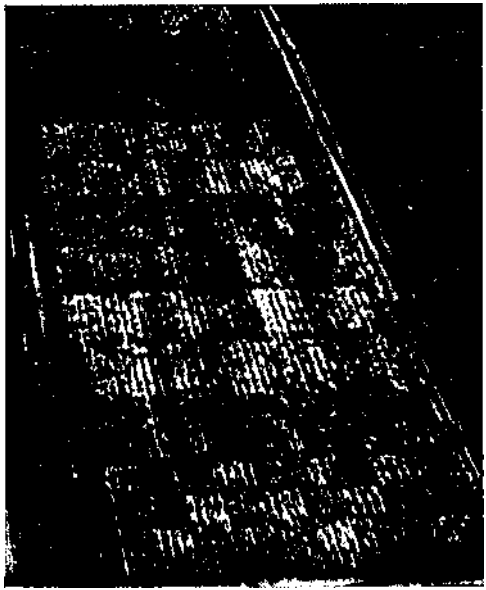

Figure 1.—Aerial view of 1958 beet-yellows resistance evaluation test at Salinas, California. Replications were divided into 2 equal parts 1 of which was inoculated with beet-yellows virus and the other maintained as a noninoculated check. Some natural infection occurred in the noninoculated plots.

placed on the plants being inoculated. Plots were sprayed with an aphicide 24-48 hours after inoculation.

1957 Tests. Plantings were made in December 1956 and in May 1957 to survey the level of resistance in a wide range of varieties and breeding lines. The December planting consisted of bolting-resistant varieties, selections, and inbreds from the United States Department of Agriculture breeding program at Salinas, California. One test included 12 varieties replicated 5 times and a second test consisted of 80 inbreds replicated twice. Each replication of each entry in both tests was divided into 2 adjacent plots, 1 of which was inoculated with yellows virus and the second maintained as a check. The plots were 2 rows wide by 50 feet long in the variety test and 1 row wide by 22 feet long in the inbred test. Spraying to control the aphid vectors was started March 16 and continued at 10- to 14-day intervals through July. The plots were inoculated April 15. The inoculated plots were graded for yellowing and estimates made of percent stunting and necrosis on June 6 and again on June 25. Percent spread of yellow[r]s to the noninoculated plots was also determined on these dates. The tests were harvested August 20-23 and data obtained on root yields and sucrose percentage.

The May planting included 256 varieties, selections, and inbreds furnished by sugar beet breeders in the United States and Europe. Each entry was replicated 2 times and divided into inoculated and noninoculated plots as in the December planting. The plots were 1 row wide by 25 feet long. Spraying for aphid control was started as soon as the plants emerged and continued until August 15. Inoculations were made July 1. The plots were graded for yellowing and estimates made of percent stunting and necrosis on August 9 and again on August 21. The test was harvested September 20-25 and root yields obtained.

1958 Tests. Field tests were planted December 13, 1957, and May 1, 1958, to determine the resistance of additional varieties and breeding lines and to recheck the resistance of lines which showed the least damage in the 1957 tests. The damage from yellows was determined as in 1957 except that the inoculated and noninoculated plots were placed end to end rather than side by side. This end-to-end arrangement permitted one half of each replication to be inoculated as a block.

The December planting included separate tests of 8 bolting-resistant varieties and 8 bolting-resistant inbreds. Both the varieties and the inbreds were replicated 4 times. The plots were 2 rows wide by 40 feet long in the variety test and 2 rows wide by 25 feet long in the inbred test. The entire planting was

sprayed with an aphicide at 7- to 10-day intervals beginning March 3 and ending July 15. Inoculations were made March 4 and the plots were harvested August 13-15.

The May planting included separate tests of 14 varieties or selections and 14 inbred lines. Four replications of each entry were used. The plots were 2 rows wide by 40 feet long in the variety test and 2 rows wide by 25 feet long in the inbred test. Spraying to control aphids was started May 20 and continued at 7- to 10-day intervals through August 15. Inoculations were made June 25 and the plots were harvested September 10 and 11.

Selecting for Resistance. Field and greenhouse selections were made for yellows resistance between 1957 and 1961. The greenhouse selections were from plants grown in 6-inch plots and inoculated with beet yellows virus when the plants were 6 weeks old. Selections were based on relative freedom from yellowing and on root size. Major attention was placed on root size and the selections were made when the plants were approximately 4 months old.

The field selections were from plantings arranged in form of a checkerboard so that each plant occupied an area 28 X 28 inches. This arrangement tended to equalize competition between plants and reduced the danger of selecting large beets which had received an unfair competitive advantage. Inoculations were made when the plants were about 7 weeks old. Selections were based on freedom from top symptoms and on root size with major attention on root size.

Field inoculations were made with a virulent strain of beet-yellows virus through 1960. In 1961 a combination of beet and western-yellows viruses was used to inoculate beets grown for selection purposes.

Results

Resistance to Damage from Yellows. Infection ranging between 90 and 100% was obtained in nearly all inoculated plots in both 1957 and 1958. Aphid populations remained high throughout both growing seasons and yellows gradually spread to the noninoculated plots even though the plantings were sprayed with an aphicide at 10- to 14-day intervals. By harvest time nearly all plants in the noninoculated plots were infected with yellows in both years. Spread to the noninoculated plots occurred more rapidly when they were placed alongside the inoculated plots than when the inoculated and noninoculated plots were placed end to end.

Reduction in yield and sucrose percentage for the 12 varieties in the December 1957 planting are shown in Table 1. The re-

Table 1.—Effect of beet yellows on the performance of sugar beet varieties in a December 17, 1956, planting at Salinas, California.

Variety	Noninoculatd plots (Checks)			Reduction by disease in inoculated plots			Infection in checks
	Gross sugar per acre	Beets per acre	Sucrose	Gross sugar	Beets	Sucrose	
	Pounds	Tons	Percent	Percent	Percent	Percentage points	Percent
461HO × US 201B	9,560	30.1	15.88	26.7	24.7	0.62	22.6
US 56/2	9,800	30.4	16.12	30.1	26.6	0.36	24.7
US 15 × US 22/3	10,300	32.3	15.94	32.1	31.1	0.74	23.6
616[a]	11,750	36.8	15.96	33.4	31.7	0.44	17.5
551SHO × 672	10,300	32.0	16.10	33.7	29.3	1.00	25.3
511[b]	10,030	31.1	16.12	34.1	30.5	0.86	19.2
551SHO × NB4	10,190	33.4	15.26	36.5	34.0	0.60	25.3
US H6	11,890	35.9	16.56	37.8	33.7	1.14	14.8
5570-49-11H1 × 6576	11,710	34.1	17.22	38.1	32.8	1.40	16.5
515[c]	10,260	32.9	15.60	40.0	36.1	0.98	19.2
US 75	10,250	31.9	16.06	41.1	37.6	0.90	25.6
MS of NBL × NB4	10,970	34.1	16.08	41.4	36.8	1.18	13.6
L.S.D. (5%)	1,320	NS	0.65	8.0	7.6	NS	7.7

[a] Field selection from US 75 for beet-yellows resistance made by Charles Price.
[b] Greenhouse selection from US 75 for beet-yellows resistance.
[c] Field selection from US 75 for beet-yellows resistance made by Charles Price.

duction in yield of roots ranged from 24.7 to 37.6% and the difference between varieties was significant at the 1% point. The loss in sucrose percentage in the 12 varieties ranged from 0.36 to 1.40 percentage points, but the difference between varieties was not significant. Yield reductions from beet yellows among 80 inbreds included in 2 replications in the December 1957 inbred test ranged from 10.4 to 55.5%.

Yield reductions in the May 7, 1957, planting were greater than those in the December planting. Yields were reduced from 16.6 to 49.4% in 91 varieties and selections included in 2 replications in the May planting. Yields of 165 inbreds were reduced from 9.0 to 65.1% in the same planting. The performances of representative groups of these varieties and inbreds are shown in Table 2.

Table 2.—Effect of beet yellows on root yield of sugar beet varieties and inbreds in a May 7, 1957, planting at Salinas, California.

Varieties	Acre yield[a] Check	Yellows	Reduction in yield
	Tons	Tons	Percent
A7/S1	30.1	25.1	16.6
IRS 55M9	23.3	19.4	16.7
US 400	24.3	18.2	25.1
US 56/2	21.9	15.8	27.9
MS of NB1 × NB2	26.9	18.5	31.2
US H2	29.1	19.8	32.0
MS of NB1 × NB4	29.2	17.7	39.3
Klein E	28.9	14.6	49.4
Inbreds			
F1 287	21.1	19.2	9.0
TASCO 5-148	20.3	17.6	13.3
F1 282	16.5	11.8	28.5
SL 618	15.4	10.0	35.1
NB4	17.7	9.9	44.1
NB1	18.6	9.7	47.8
NB2	19.7	8.5	56.8
5508-113	10.9	3.8	65.1

[a] Acre yield is an average of two replications.

The 1957 tests demonstrated that a wide range of resistance to beet yellows exists within *Beta vulgaris* I,., but varieties or breeding lines immune or highly resistant were not found. Percent yield reductions varied greatly among replications emphasizing the necessity for adequate replication in resistance-evaluation tests.

Reductions in yield and sucrose percentage of the 8 varieties in the December 1958 planting are shown in Table 3. Root yields were reduced 24.1 to 44.0%. This difference between

Table 3.—Effect of beet yellows on the performance of sugar beet varieties in a December 13, 1957, planting at Salinas, California.

Variety	Noninoculated plots (checks)			Reduction by disease in inoculated plots			Infection in checks
	Gross sugar per acre	Beets per acre	Sucrose	Gross sugar	Beets	Sucrose	
	Pounds	Tons	Percent	Percent	Percent	Percentage points	Percent
MS of NB6 × NB5	10,400	36.1	14.35	20.5	26.2	0	10.5
MS of 515 × 569	9,360	27.7	16.93	27.3	24.1	0.65	14.3
711[a]	9,960	33.2	14.98	28.2	26.0	0.35	13.5
MS of NB1 × NB4	12,000	42.3	14.15	31.6	28.3	0.62	3.9
US 75	10,440	34.6	15.08	36.8	33.2	0.83	17.3
US H5A	11,320	39.3	14.35	37.0	35.2	0.27	10.5
MS of NB1 × NB2	11,710	39.3	14.85	41.2	38.0	0.50	5.4
US 15 selection	11,770	39.5	14.93	46.0	44.0	0.45	17.2
L.S.D. (5%)	880	4.0	1.33	12.2	12.1	NS	7.5

[a] Second successive field selection from US 75 for beet-yellows resistance.

varieties was significant at the 1% point. Sucrose percentages were reduced in the yellows-inoculated plots, but the reductions varied so much from one plot to another that differences between varieties were not significant. The 1957 and 1958 results indicate that yield data give a more accurate measure of beet-yellows resistance than do sucrose data.

Yield reductions for the 14 varieties and 14 inbreds in the May 1958 planting are shown in Table 4. Losses in the varieties ranged from 11.8 to 36.2% and those in the inbreds ranged from 20.4 to 44.2%. Selections made for beet-yellows resistance at the Institute voor Rationele Suikerproductie, Bergen op Zoom, The Netherlands (IRS numbers), showed the least damage.

Table 4.—Effect of beet yellows on the performance of sugar beet varieties and inbreds in a May 1, 1958, planting at Salinas, California.

Varieties	Acre yield		Reduction in yield	Infection in checks
	Check	Yellows		
	Tons	Tons	Percent	Percent
IRS 55M24	22.0	19.4	11.8	13.0
715-1	12.6	11.0	12.7	2.2
IRS 55M9	21.1	18.2	13.7	13.8
IRS 55M14	20.1	16.9	15.9	10.9
MS of NB1 × NB4	27.9	22.1	20.8	8.6
IRS M1-1953	18.9	14.3	24.3	6.7
MS of NB1 × NB2	24.2	18.1	25.2	5.4
Sel. from US 104	25.5	18.7	26.7	13.7
711	26.4	19.8	26.9	15.9
MS of NB6 × NB5	23.9	17.3	27.6	11.0
Sel. from US 104	20.2	14.4	28.7	13.4
US 75	27.7	19.7	28.9	15.8
US 15 selection	26.8	18.4	31.3	18.2
Sel. from US 201	13.8	8.8	36.2	14.0
L.S.D. (5%)	2.4	1.9	7.4	5.3
Inbreds				
55-RF993	23.0	18.3	20.4	11.4
5614	22.1	17.4	21.3	7.2
SL 7807	19.1	14.8	22.5	5.5
SLC 117	14.6	11.1	24.0	15.8
NB4	17.3	13.1	24.3	4.3
NB1	17.3	13.0	24.9	3.4
5577-2	19.5	14.3	26.7	5.3
SL 6509	16.0	11.6	27.5	9.7
5628-24	19.5	14.1	27.7	20.1
NB5	16.5	11.1	28.4	10.8
NB6	19.3	13.6	29.5	5.4
TASCO 6-278	14.3	9.6	32.9	55.3
TASCO 5-148	21.8	13.8	36.7	23.2
NB2	15.6	8.7	44.2	10.5
L.S.D. (5%)	3.8	2.5	8.2	6.7

Varieties and inbreds selected as possessing resistance to beet yellows in the 1957 tests tended to perform well in 1958. There was also reasonably good agreement among the results for the different planting dates. The IRS 55M9 variety showed superior resistance in both 1957 and 1958. The NB2 inbred and the US 15 selection were severely damaged in each of the tests in which they were included. Where disagreement in results occurred, the test with the greater number of replications is considered the more accurate.

Variation in Susceptibility to Infection. The 1957 and 1958 tests provided an opportunity to determine the relative resistance of the varieties and inbreds to natural infection with yellows. Aphid build-ups in the tests were prevented by spraying regularly with an aphicide. Infection in the noninoculated plots was primarily from wind-borne winged aphids and took place at a relatively slow rate. Counts in the December 1956 planting showed that infection in 12 varieties ranged from 13.6 to 25.6% in the noninoculated checks (Table 1). In the December 1957 planting-infection ranged from 3.9 to 17.3 percent (Table 3) and in the May 1958 planting from 2.2 to 35.3 percent (Table 4). Differences between varieties and inbreds were significant at the 5% level.

Counts were also made in an unsprayed variety evaluation test planted in a commercial sugar beet field near Salinas. Only a moderate amount of yellows infection occurred in this field and an accurate determination was made of spread among 12 varieties included in the test. The amount of infection ranged from 15.0 to 34.7% and the difference between varieties was significant at the 1% level.

The results of the 1957 and 1958 tests demonstrate that differences exist among varieties and inbreds in susceptibility to yellows infection. No attempt was made to identify the yellowing virus which caused the natural infection. Western-yellows virus was predominant in the Salinas district in both years and probably much of the natural infection was with this virus.

No relation was found between resistance to infection and resistance to damage from yellows nor was there a clear-cut relation between color of foliage and susceptibility to natural infection with yellows. Inbred lines with dark-green foliage showed a wide range in susceptibility to infection. Inbreds with light-green foliage tended to be susceptible; however, some lines with light-colored foliage showed only moderate infection.

Progress in Selecting for Resistance to Beet Yellows. Some uncertainty exists as to the relative reliability of greenhouse and

field techniques of selecting for beet yellows resistance. Watson and Russell (9) reported that scores for severity of symptoms made in the greenhouse were positively correlated with similar scores made in a field experiment through the use of 2 cultivated and 2 wild beet types. The symptom scores were also positively correlated with losses in root and sugar yields caused by the beet-yellows virus. Observations thus far in California indicate that greater progress can be made by selecting in the field than in the greenhouse. Top symptoms of plants grown and inoculated in the greenhouse tended to be more uniform than those in field plantings. Wide variations in root size occurred in greenhouse-grown plants, but these variations were more closely associated with differences in environment among plants than with differences in resistance.

In the California field program greatest emphasis has been placed on the development of a beet-yellows resistant selection of US 75. Successive selections based primarily on superior root size were compared with the parent variety in 1960 and 1961 replicated tests (Table 5).

Table 5.—Progress in selecting for yellows resistance in US 75 at Salinas, California.

Variety	Acre yield of noninoculated check		Reduction in yield		
			Beet yellows		Beet and west. yel.
	1960	1961	1960	1961	1961
	Tons	Tons	Percent	Percent	Percent
US 75	27.1	19.4	31.7	33.1	42.1
Sel. from US 75	28.8[a]	17.6[b]	25.3	15.8	24.2

[a] Third successive selection for beet-yellows resistance.
[b] Fourth successive selection for beet-yellows resistance.

Both the third and fourth successive selections from US 75 were significantly more resistant to beet yellows than the parent variety. The resistance of the fourth successive selection to the combination of beet and western yellows was also significantly greater than that of US 75. These results indicate that a correlation may exist between resistance to beet and western yellows.

The fourth successive selection and the parent US 75 variety were included in three variety trials in 1961. In each of these trials both the root yield and sucrose percentage were similar in the selection and in US 75.

Correlation between root-yield reduction and top symptoms Correlation coefficients were computed between reduction in root yield from beet yellows and stunting, yellowing, or necrosis of tops. These coefficients were computed separately for varieties and for inbreds in each of the replications of the 1956-57 tests

Table 6.—Correlation coefficients between yield reduction from beet yellows and the top symptoms stunting, yellowing, and necrosis.

Date of Planting	Type of material	Stunting		Yellowing		Necrosis	
		Rep. 1	Rep. 2	Rep. 1	Rep. 2	Rep. 1	Rep. 2
December 1956	Inbreds	.07	.24*	.48	.19	.12	.09
December 1956	Varieties	−.16	.32	.33	.39	.28	.06
May 1957	Inbreds	.46**	.45**	.26**	.02	.36**	.02
May 1957	Varieties	.59**	.52**	.40**	.04	.51**	.02

* Significant at 5% point.
** Significant at 1% point.

(Table 6). In the December planting very little correlation existed between yield reduction and any of the top symptoms. In the May planting a significant positive correlation was found between yield reduction and stunting in both inbred and variety tests. In one replication, yield reduction was also correlated with yellowing and with necrosis.

None of these correlation coefficients was high. Yield reduction was most closely associated with stunting, but even this association varied greatly from one variety or inbred to another.

The results of these tests show that none of these three top symptoms will serve as a reliable selection criterion. Yellowing and stunting are undesirable characters in a sugar beet variety; so preliminary selections can be made for relative freedom from these characters. Unless a reliable biochemical technique is developed (6), true resistance can be determined only through yield comparisons of yellows-infected and noninfected beets. The necessity of using yield measurements to determine resistance limits the size of populations which can be handled in a breeding program and adds greatly to the cost of developing resistant varieties.

Summary

Tests at Salinas, California, in 1957 and 1958 with more than 350 sugar beet varieties and breeding lines showed that a wide range of resistance to beet yellows exists within *Beta vulgaris* L. Yield losses among lines ranged from 9.0 to 65.1%. Immune or highly resistant lines were not found.

Natural infection with yellows (probably largely western yellows) in noninoculated varieties and breeding lines ranged from 2.2 to 35.3% indicating that differences also exist in resistance to yellows infection. Resistance to infection was not related to resistance to damage from yellows nor was there a clear relation between color of foliage and resistance to natural infection.

The yellows resistance of US 75 was improved by selecting in the field from plants inoculated with a virulent strain of beet-

yellows virus. The root yield of the fourth successive selection from US 75 was reduced 15.8% by beet yellows compared with a reduction of 33.1% in the parent variety.

Correlations between reduction in root yield and stunting, yellowing, or necrosis of tops were low in plants affected by beet yellows. None of these three types of top symptoms will serve as a reliable selection criterion. True resistance can be determined only from yield comparisons of diseased and healthy beets.

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Highly Virulent Strains of Curly Top Virus in Sugar Beet in Western United States

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Introduction

It has been known for more than 30 years that beet curly top virus is a complex of strains that vary in virulence, induced symptoms, host range, and perhaps other characteristics. Griddings (2,3,5)[2] described 12 strains of this virus. One strain obtained from Idaho was highly virulent on sugar beet and was designated 'Strain 11" (5). This strain is capable of causing marked injury even on resistant varieties of sugar beet.

During the season of 1960 curly top caused considerable damage to individual plants in some fields in central San Joaquin Valley, but no special study was made of strains of the virus involved. In 1961, curly top symptoms were so severe on plants in certain fields near Shandon, Los Banos, and Tracy that it was thought advisable to compare the virulence of the virus strains involved with that of strains previously isolated. Results of these tests are presented in this report.

Method of Testing

Beet plants affected with curly top were selected from fields near Shandon, Los Banos, and Tracy, and planted in pots at the U. S. Agricultural Experiment Station at Salinas, California. Also, beets received from Wyoming and Colorado were potted and included in the tests. After sufficient top growth was produced on the potted beets, nonviruliferous beet leafhoppers were allowed to feed on the diseased plants 3 days or more and then caged singly on seedling sugar beet plants. To determine the relative virulence of virus from different field beets, tests were made using the susceptible selection SL 742, the resistant variety US 75, and the very resistant selection SL 68. Additional tests and subtransfers were made on US 75 and on hybrid varieties with high degrees of resistance.

Only plants with very severe symptoms were selected from fields in California. Therefore, the virus recovered from these plants probably represents strains with the highest virulence to be found in the respective fields and the results are not necessarily representative of the fields as a whole.

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[2] Numbers in parentheses refer to literature cited.

Results were compared with those obtained with strain 11 on the different varieties and selections used in testing the field beets. Strain 11 was chosen because it is the most virulent of the curly top virus strains described up to the present. This strain was tested at Jerome, Idaho, in 1956 and 1957, on 4 varieties of beets growing in field plots (7). In both years, strain 11 caused substantially greater losses on all varieties than was caused by natural infection. This was true even in 1957 when plots naturally infected yielded only 2.47 to 5.20 tons per acre, and plots inoculated with strain 11 yielded 1.16 to 3.42 tons per acre.

Severity of injury on the test plants by virus from different field plants was estimated on the basis of stunting, leaf curling, and plant survival. A numerical grading system, ranging in ascending order of severity from 1 to 5, inclusive, was used in estimating relative virulence of virus from the different sources.

Results of Transfers of Virus from Field Beets

The results of tests of representative beets from different areas are shown in Table 1. As would be expected, a range of severity of symptoms was produced by virus from different sources. Highly virulent strains were obtained from beets from Shandon, Los Banos, Tracy, and Wyoming. Some of these were obviously more virulent than strain 11 with which they were compared. The

Table 1.—Relative virulence of curly top virus strain 11 and isolates from beets from different areas of western United States, indicated by tests on US 75 sugar beet.

Source of beets from which virus transfers were made	Number of plants of 20 inoculated showing indicated grade of severity					Average severity
	1	2	3	4	5	
Shandon, Calif.	0	0	2	9	7	4.3
Shandon, Calif.	0	3	6	7	2	3.4
Shandon, Calif.	3	2	5	2	4	3.1
Shandon, Calif.	0	0	0	3	11	4.8
Shandon, Calif.	0	0	0	10	7	4.4
Shandon, Calif.	0	0	0	7	7	4.5
Shandon, Calif.	0	1	1	5	11	4.4
Tracy, Calif.	0	0	0	0	11	5.0
Tracy, Calif.	0	0	1	3	13	4.7
Tracy, Calif.	0	2	5	5	0	3.2
Los Banos, Calif.	0	0	0	1	11	4.9
Los Banos, Calif.	0	0	0	1	12	4.9
Los Banos, Calif.	0	0	1	8	9	4.4
Wasco, Calif.	0	2	12	0	0	2.8
Wyoming	0	2	14	0	0	2.9
Wyoming	0	1	1	3	13	4.6
Wyoming	0	6	10	0	0	2.6
Salinas, Calif.-st. 11	0	0	6	11	2	3.8
Salinas, Calif.-st. 11	0	0	2	9	4	4.1
Salinas, Calif.-st. 11	0	0	5	10	2	3.8

relative amounts of dwarfing by an isolate from Shandon and by strain 11 on a hybrid variety (1 X 3) and on US 75 are shown in Figure 1. Several other isolates appeared also to be more virulent than strain 11 when tested on US 75 (Table 1).

Figure 1.—Beet plants inoculated with curly top virus in the cotyledon stage. Top, Selection 1 x 3 inoculated with isolate from Shandon (left) and strain 11 (right). Bottom, US 75 inoculated with isolate from Shandon (left) and strain 11 (right).

Transfers from some of the field beets gave uniformly severe effects. Transfers from others gave a range of severity of symptoms on US 75, indicating that the plants were infected with a mixture of strains. Subtransfers from mildly affected test plants gave predominantly mild symptoms, whereas transfers from severely affected plants gave severe symptoms only or a range of severity of symptoms, indicating that more than one strain of virus had

been transmitted. These results supply further evidence that field beets often are infected with a mixture of curly top virus strains.

Giddings (4) showed that a single beet leafhopper is able to carry a combination of at least 3 strains of virus. When such leafhoppers were allowed short feeding periods on seedling beets, they introduced the strains into the plants singly and in all possible combinations. The beet leafhopper, therefore, may infect beets with more than one strain of virus in a single feeding. Also, plants infected with one strain remain susceptible to infection by other strains. If the second strain is more virulent than the strain already present, symptoms of curly top are increased by the second strain.

Four of the most virulent isolates—one from Shandon, two from Los Banos, and one from Wyoming—were selected for making a series of transfers to different varieties and selections of sugar beets and other plants. These isolates have continued to produce very severe symptoms on resistant varieties and selections, such as US 75 and SL 68. Infected plants of US 75 produced curled and dwarfed leaves, and little growth was produced after the plants showed first symptoms of disease. High percentages of plants inoculated in the cotyledon stage with the 4 virus selections were killed. The virus isolates have maintained their relative degrees of virulence, as compared to strain 11, through 3 or more transfers on US 75. Each of the 4 isolates has appeared to be more virulent than strain 11 on sugar beet.

Damage by Virulent Strains of Virus

It is not possible to assess accurately the damage produced in 1961 by virulent strains of curly top virus in any specific area because injury varied in different fields depending on the time of infection, vigor of plants, and other factors. It was evident, however, that in certain fields yields were greatly reduced.

In the Shandon and Los Banos areas, beet leafhoppers moved into the beet fields a month to six weeks earlier than usual, owing to the earlier drying of desert vegetation which forced the leafhoppers to migrate. In certain areas there also was overwintering of leafhoppers on the floor of the valleys close to beet fields. In some fields leafhoppers were present at thinning time. Where conditions were unfavorable for very rapid growth, leafhoppers multiplied in the beet fields and produced high percentages of infection. In some fields the leafhoppers continued to multiply through the summer. Fields that had 50 or more leafhoppers per plant in June and July were found near Los Banos and Shandon.

Plants in these fields did not attain sufficient size for the foliage to cover the rows. Thus, they were exposed to direct sunlight throughout the summer and remained favorable hosts for multiplication of leafhoppers.

The high summer populations of leafhoppers probably account for the severe damage noted in some fields. The leafhoppers that initially invaded the beet fields from the desert areas undoubtedly carried many different strains of curly top virus ranging in virulence from mild to very severe. Tests over a period of years have indicated that leafhoppers from desert areas predominantly carry mild strains of virus. Giddings (6) suggested that this is due to the fact that virulent strains of virus kill most desert host plants. If this is true, virulent strains of virus that may be developed in the natural breeding grounds of the beet leafhopper tend to be self-eliminating.

After virus is carried from the desert breeding grounds to beet fields by the beet leafhopper, factors involved in strain selection change radically. In beets, the highly virulent strains of virus are best equipped to survive.

The sugar beet plant is an excellent host for increase of the beet leafhopper if plants are small and exposed to full sunlight. If the plants are large and the foliage covers the rows so that shade and high humidity prevail, little leafhopper increase occurs. By stunting the beet plants virulent strains of virus provide more favorable conditions for leafhopper increase. Also, since strains of curly top virus do not afford cross-protection against each other, plants infected with a mild strain of virus remain susceptible to infection with more virulent strains. Where high populations of leafhoppers are present in a field they may continue to spread more virulent strains throughout the season. Curly top, therefore, may become progressively more severe as the season advances. By the end of the season, most of the plants may be infected with the most virulent strains of virus along with any less virulent strains that may be present.

Evidence of progressive spread of more virulent strains of curly top virus in fields already 100% infected was noted in beet fields near Los Banos as late as November 2. Older leaves of many plants showed mild vein swelling, indicating that they had first been infected with a mild strain of virus. On November 2, some of these plants had badly curled young leaves, indicating that the plants had been reinfected with a more virulent virus strain.

As already stated, the strain complex in desert areas apparently has remained more or less stable for many years. No new factors that would change this condition are known to have been introduced. However, if conditions which would permit perpetuation of virus on beets through the year should arise, the percentage of beet plants infected with the more virulent strains would be expected to increase.

Summary and Conclusions

Tests of isolates from field beets in 1961 indicate that strains of the curly top virus capable of causing appreciable damage to resistant varieties of sugar beets were present in widely separated areas of western United States. Some of these isolates have higher degrees of virulence than any of the strains previously described, indicating that strains of increased virulence are being evolved. The findings emphasize the desirability of maintaining and increasing the curly top resistance of new varieties of sugar beets developed for use in areas of western United States where curly top virus and the beet leafhopper are prevalent.

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Use of Tetrazolium Salts in Determining Viability of Sugarbeet Pollen[1, 2]

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The increased use of stored pollen and cytoplasmic male sterility in sugar beet breeding necessitates determining the viability of pollen at the time of its use or at anther dehiscence. Stains commonly used for staining sugar beet pollen, such as acetocarmine or iodine, are not vital stains and hence do not differentiate viable mature pollen from mature pollen which has lost its viability. A stain specific for living mature pollen would be useful to persons working with stored pollen or pollen treated in any possibly lethal manner. Such a stain would also be of value in the classification of plants with different degrees of male sterility.

Tetrazolium salts, which are reduced to insoluble colored products (monoformazans or diformazans by action of dehydrogenase enzymes linked to respiratory processes, seem to offer possibilities for this purpose. The development of 2,3,5-triphenyl tetrazolium chloride (TTC) and its application to biology has been reviewed by Smith (8)[4]. According to Smith this chemical was first prepared by H. von Pechmann and P. Runge in 1894 and was found by R. Kuhn and D. Jerchel in 1941 to cause a red coloration in cells of yeast, bacteria and water cress. Lakon (2,3) in 1942 found it possible to determine germination percentage of cereal grains and corn by treating exposed embryos with TTC. He found that the percentage of those embryos stained red was not different from the percentage which germinated in a standard germination test. However, MacLeod (4) found that under a narrow range of conditions of grain moisture and temperature, TTC results grossly overestimated germination. This overestimation was due to the fact that seed germination was more sensitive to heat damage than was enzymatic activity.

TTC has been used in various tests on many types of tissues. According to Porter, Durrell, and Romm (7), the salt is an

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[3] Geneticist, Crops Research Division, Agricultural Research Service, U. S. Department of Agriculture.

[4] Numbers in parentheses refer to literature cited.

oxidation-reduction indicator, and the development of the non-diffusible red color in a specific tissue is in general indicative of the presence of active respiratory processes.

Vieitez (9) in 1952 reported that a 2% TTC solution at 50°C provided a quick and reliable index of viability of maize pollen. However, Oberle and Watson (5) in 1953 reported that TTC stained to varying degrees certain fruit pollens known to be nonviable and concluded that the chemical was of no value as an indicator of germinability for peach, pear, apple, and grape pollens.

Other tetrazolium salts and derivatives have been formulated. Some of these have been found to be of value in the localization and quantitative measure of certain reducing enzymes. Pearse (6) found 2-(p-iodophenyl)-3-(p-nitrophenvl)-5-phenyl tetrazolium chloride to be rapidly reduced to a red monoformazan under aerobic conditions. He further found 3-(4,5-dimethylthiazolyl-2)-2,5-diphenyl tetrazolium bromide to be rapidly reduced to a blue or purple diformazan.

The present study was conducted to determine whether certain of these newer tetrazolium salts are of value in determining the viability of sugarbeet pollen.

Materials and Methods

A series of eight tetrazolium salts were tested for their vital staining capacity of beet pollen. The eight salts were as follows:

1. 2,3,5-triphenyl tetrazolium chloride
2. tetrazolium blue
3. tetrazolium violet
4. tetrazolium red
5. nitro-blue tetrazolium
6. neotetrazolium chloride
7. 2-(p-iodophenyl)-3-(p-nitrophenyl)-5-phenyl tetrazolium chloride
8. 3-C4,5-dimethylthiazolyl-2)-2,5-diphenyl tetrazolium bromide

Various concentrations of the salts were dissolved in distilled water. Salts 1 and 4 were readily soluble in cold water, 7 and 8 were soluble upon heating nearly to the boiling point and 2. 3, 5, and 6 were soluble upon being brought to the boiling point.

The salts were tested at the following four concentrations and three temperatures: 0.2, 0.5, 1 and 2% each at 20°. 35° and 50°C. The pollen was examined at intervals of 3, 5, 10, 15, 20, 30 and 40 minutes. Preliminary tests were made on pollen from

the stock beet A60-3 as pollen was most abundant on this population at the time of the study. All salts were first tested for vital staining capacity. Those salts which exhibited a vital staining ability were then used in determining the most effective concentration and temperature. The sugar beet populations used in these tests were 52-430 (inbred), 52-307 (inbred), 52-305CMS (cytoplasmic male-sterile inbred), A60-3 (stock beet), and 52-305CMS X A60-3. The population 52-305CMS X A60-3 was segregating for male-sterile, semisterile and fertile types. The semisterile plants were those with yellow shrunken anthers.

The plants used in this study were grown in the greenhouse during the winter of 1961-62. All pollen from the fertile plants was collected about 9 AM from anthers which had just dehisced. Anthers from newly-opened flowers of sterile and semisterile plants were used. Nonviable pollen from four sources was also tested using the vital staining salts at their most effective concentrations and temperatures. One source of nonviable pollen had been collected and stored frozen without humidity control for about 2½ years. This pollen had been previously determined to be ineffective for fertilization of cytoplasmic male-sterile plants. The other nonviable pollen sources were fresh pollen killed in 70% ethanol, fresh pollen heat-killed in an electric oven held at 80°C for 15 minutes and fresh pollen heat-killed by holding it at 110°C for 15 minutes.

The germinability of all pollen was tested on an agar-sucrose culture medium as described by Artschwager and Starrett (1). This medium contained 1.5% agar and 40% sucrose. The incubation period was 7 hours at 32°C.

It was found most convenient to drop the tetrazolium solution on the pollen grains on a glass microscope slide, mix slightly, and cover with a glass cover slip. The slides were then set aside in daylight until examined.

Results and Discussion

Four of the eight tetrazolium salts, 1,4,7 and 8, acted as vital stains on fresh pollen. The staining action of salts 1 and 4 was similar as was 7 and 8 except for their resulting colors. The deepest staining and most rapid reaction in salts 1 and 4 took place in 2% solution at 20°C. Most morphologically mature pollen grains were stained pink to deep red in 25 minutes. Salts 7 and 8 were most effective in a 0.5% solution at 35°C. After 5 minutes most morphologically mature pollen were stained pink to red by salt 7 and purple to deep purple by salt 8. A 2% solution of salts 7 and 8 did not stain. In general the staining by all salts was

more rapid as the temperature increased except at the 2% concentration where the threshold of activity was evidently exceeded. None stained in this concentration at 50°C. The reaction in salts 1 and 4 at all concentrations and temperatures was rather slow and not completely positive; light-pink and nonstained pollen were hard to distinguish. The reaction in salts 7 and 8, particularly 8, was more rapid and much more positive.

Morphologically mature pollen grains ruptured in solutions of salts 1, 4 and 8. Rapidity of the rupture increased with concentration and temperature. This rupture might be primarily due to the low osmotic concentration of the solution. The staining reaction was complete before any cell rupture occurred at any concentration of salt 8. In salts 1 and 4, however, rupture often occurred in unstained or only slightly stained cells. Cell rupture was not noted in salt 7, however, minute insoluble particles in the solution interfered with the observations. The staining reaction in salt 8 was the most positive followed in order by salts *7*, 1 and 4.

The reaction in each salt was the same in all pollen-fertile populations tested. One of the semisterile plants produced about 9% morphologically mature pollen, which stained in the same manner as pollen from the pollen-fertile plants. The abortive pollen did not stain. All pollen from the cytoplasmic male-sterile plants and from all but one of the semisterile plants was abortive in appearance and was not stained in any of the solutions. It will be noted in Table 1 that the nonstaining portion of the fresh pollen from fertile plants ranged from 16..8 to 38.9%. This nonstaining portion consisted primarily of cells which had apparently aborted at an early stage of development.

These same pollen sources were tested for germinability on an agar-sucrose medium. After 7 hours of incubation the pollen tubes of the germinated pollen grains were up to 200 microns in length. The percentage germination varied somewhat with populations but even that of A60-3 was only 13.9. Low germination might be expected because in reviewing this subject Artschwager and Starrett (1) stated that N. Favorsky in 1928 had obtained poor germination of sugar beet pollen, not more than 30%. In their own studies they got pollen to germinate easily and abundantly, but they did not report actual percentages. Work summarized by Artschwager and Starrett (1) and this study indicate that there are additional unexplained factors affecting the germinability of sugarbeet pollen on the artificial medium used.

Optimum conditions for germination have not been accurately determined. Hence, the percentage germination of pollen on the culture medium is not likely to be a good direct measure of pollen viability.

The pollen known to be nonviable was tested in salts 1, 4, 7 and 8 at concentrations and temperatures which produced the most favorable reaction with fresh pollen. Salts 1 and 4 each caused a light-pink color in the 2½ year-old pollen, particularly in pollen grains near the peripherv of the cover slip, while salt 7 stained red about 1 % of the pollen which had been heat-killed at 80°C for 15 minutes. Salt 8 did not stain any of the types of nonviable pollen. None of the pollen germinated when incubated on agar-sucrose medium.

Since salt 8 was the only solution which resulted in no staining of pollen known to be nonviable, it was tested further on pollen exposed at room temperature for 3 and 8 hours.

According to Artschwager and Starrett (1) viability of pollen under Colorado field conditions does not extend beyond a day. They further reported that it often loses its viability in less than 3 hours when stored in a shallow glass dish in daylight at room temperature.

The results for pollen given such treatments as compared with fresh pollen are summarized in table 1 for salt 8.

Exposure of pollen in daylight at room temperature apparently reduced its viability drastically. This is reflected in germination and staining percentages. Pollen neither germinated nor stained after exposure for 8 hours. There would appear to be a relation between germination and stainability.

Although pollen of A60-3 which had been frozen failed to germinate it is doubtful that this pollen was completely nonviable since sugarbeet pollen has remained viable for at least 4 months when stored at low temperatures[5]. Vieitez (9) reported that maize pollen was not stained by TTC after it had been cooled to 0°C and he referred to this as an "enzyme inhibitor treatment". Pollen storage studies indicate, however, that this would not be a permanent enzyme inhibitor treatment in sugarbeets.

Pollen in solutions of salts 1 and 4 was not stained when not covered by a cover slip. Pollen in salts 7 and 8, however, was stained whether covered or uncovered.

[5] Unpublished data of LeRoy Powers and J. W. Dudley in 1958 Sugar Beet Research Report, Sugar Beet Section, Crops Research Division, Agricultural Research Service, U. S. Department of Agriculture.

When the slides were prepared and immediately placed in darkness no staining of pollen was noted in salts 1 and 4. Salts 7 and 8 stained equally well in light or darkness.

After 2 days, solutions of salts 1 and 4 had lost most of their staining capacity. This change cannot be explained by a difference in pollen but could possibly have resulted from a pH change of the solution (not investigated). Salts 7 and 8 maintained their staining ability even after being in solution for 28 days. A slight black precipitate that appeared in salt 8 did not alter its effect.

When germinated pollen was placed in a solution of salt 8 the percentage stained was only slightly less than that recorded in Table 1 for fresh pollen. Hence, many pollen grains were stained but ungerminated. Among the germinated pollen grains the cytoplasm of both the pollen cell and tube was stained. Rarely were there individual pollen grains which had germinated but did not stain.

Table 1.—Germination of sugar beet pollen and development of purple color in a 0.5 percent solution of 3-(4,5-dimethylthia7.olyl-2)-2,5-diphenyl tetrazolium bromide.

Population and treatment[1]	Germination (percent)	Purple color (percent)
52-307:		
None (fresh pollen)	9.1	64.3
Exposure 3 hours	0.0	0.0
Exposure 8 hours	0.0	0.0
52-430:		
None (fresh pollen)	12.3	61.1
Exposure 3 hours	0.1	6.8
Exposure 8 hours	0.0	0.0
A60-3		
None (fresh pollen)	13.9	83.2
Exposure 3 hours	0.2	7.5
Exposure 8 hours	0.0	0.0
Freezing 96 hours	0.0	20.6

[1] Fresh pollen was stained or incubated immediately after collection. Exposed pollen was stored in the collection dish in davlieht at room temperature for the specified period. Fro/en pollen was stored in a tightly corked container at —30°C for 96 hours without humidity control.

Under the conditions of the tests, salt 8 was the only one which did not stain known nonviable pollen. In addition this salt was the most rapid and positive in its staining action. It stained most rapidlv when used in a 0.5% solution at 35°C. But since the reaction is rapid, leading to considerable cell rupture after 15 minutes, it is more convenient to use a 0.5% solution at about 20°C, which leads to a reaction equally as effective but slightly less rapid. This allows greater latitude in the period of examination, which is made most easily after 5 to 20 minutes.

After 30 minutes at 20° C, considerable cell rupture occurs accompanied by draining of the cytoplasm. The empty cells are somewhat difficult to discern from nonstained pollen cells.

This study indicates that 3-(4,5-dimethylthiazolyl-2)-2,5-diphenyl tetrazolium bromide is useful as an indicator of pollen viability in sugar beets. However, there remains the possibility that a narrow range of conditions may exist in which pollen germinability is inhibited but enzymatic activity continues. If this were to occur it could lead to an erroneous conclusion using salt 8 as an indicator. Under the limited set of conditions in this study this possibility was not detected.

Summary

Studies were conducted in an attempt to find a tetrazolium salt which would rapidly and accurately determine the viability of mature sugar beet pollen.

Eight tetrazolium salts were tested for their staining capacity at concentrations of 0.2, 0.5, 1 and 2% and at temperatures of 20°, 35°, and 50°C. Positive results were obtained with four of the salts. Of these four the most positive and effective was 3-(4,5-dimethylthiazolyl-2)-2,5-diphenyl tetrazolium bromide at a concentration of 0.5% at 20°C.

The percentage of pollen grains stained by this compound was related to the percentage germinated on artificial medium but was in all cases greater. It is believed, however, that all viable pollen was not germinated on the artificial medium.

The mature pollen grains assumed to be viable were stained an easily distinguishable purple to deep-purple color. Nonviable mature pollen and abortive pollen from cytoplasmic male sterile plants was not stained.

The results obtained indicate that a 0.5% solution of 3-(4,5-dimethylthiazolyl-2)-2,5-diphenyl tetrazolium bromide, at 20°C provides a specific and rapid means of determining the viability of mature sugarbeet pollen.

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Effects of Root Diffusates of Various Nematode-Resistant and-Susceptible Lines of Sugar Beet (Beta vulgaris L.) on Emergence of Larvae from Cysts of Heterodera schachtii

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Certain selected breeding lines of sugar beets have a considerable degree of resistance to the sugar-beet nematode, *Heterodera schachtii* Schmidt 1871. A test was undertaken to determine if this resistance was due to lack of production of the nematode-hatching factor.

Root diffusates of four lines of nematode-resistant and two varieties of nematode-susceptible sugar beets were tested by the method reported by Golden (l)[2]. Two hundred-ml quantities of root diffusate were leached from 5-inch pots containing 3 plants of a single breeding line or a commercial variety of sugar beets growing in sterilized soil. All diffusates were diluted 1 to 10 to facilitate detection of slight differences in hatching effect. Treatments were replicated 4 times in individual Syracuse watch glasses, each of which contained 40 cysts. At weekly intervals the nematode cysts were transferred to clean watch glasses containing fresh treatment solutions, and the emerged larvae preserved in 5% formalin until counted. Samples which contained large numbers of larvae were aliquoted to expedite counting. Results were analysed for statistical significance by the analysis of variance method.

The numbers of larvae that emerged in the various diffusates were not significantly different. Nematode-resistant lines averaged 10,390; 9,200; 8,410; and 7,990 larvae per replication, whereas the two susceptible varieties averaged 9,060 and 8,130 larvae per replication. The total number of larvae emerging in 4 replications of tap water treatment was 3,090. That is, diffusates of all resistant lines tested contained about the same amount of hatching factor as the susceptible beets. Obviously, resistance in these lines is not due to lack of hatching factor.

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Influence of Age and Supplemental Light On Flowering of Photothermally Induced Sugar Beet Seedlings[1]

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Photothermal induction of young seedlings by prolonged exposure to low temperature and artificial light (1,2,3)[3] is widely used in the United States to expedite sugar beet breeding work. However, the usefulness of this technique has been limited to some extent by a tendency toward reversal of induction where artificial light is not provided, as a supplement to sunlight, during the post-induction period. A striking illustration of this tendency was reported for seedlings removed from the induction chamber on August 2, 1951 (2). Similar results were obtained in a study involving seedlings of the variety GW359 transferred from the induction room into the open on July 21, 1959[4].

In the 1951 and 1959 comparisons, the induction treatment ended when days were relatively long but decreasing in length. In a 1960 study seedlings of two varieties were transferred from the induction chamber into the open on June 2, nearly 3 weeks before the longest day of the year[4]. Duration of the induction treatments were 8 and 14 weeks for GW359 and US 75, respectively. Final counts of flowering plants were made 12 weeks after the end of the induction treatment. In the GW359 population receiving continuous illumination during the post-induction period, 96% of the plants flowered. In the corresponding population receiving no supplemental light, only 53% flowered. For the bolting-resistant variety, US 75, comparable flowering percentages were 83 and 27, respectively. Each of these 4 percentages was based *on* a minimum population of 47 plants.

The 1959 and 1960 results left no doubt as to the need for supplemental light during the post-induction period under the conditions of the experiments. These results reinforced the tentative conclusion, reported for the 1951 investigations (2). that supplemental light tends to counteract the induction-reversing action of high temperature under such conditions.

[1] Report of investigations conducted by the Crops Research Division. Agricultural Research Service, U. S. Department of Agriculture, in cooperation with the Colorado Agricultural Experiment Station; publication Approved by the Experiment Station Director as Scientific Series Article No. 776.

[2] Plant Pathologist, Agricultural Research Service, U. S. Department of Agriculture Assistance of Luther W. Lawson, Agricultural Research Technician, in conducting the experimental work, is acknowledged.

[3] Numbers in parentheses refer to literature cited.

[4] Unpublished results.

In connection with the breeding program at the Fort Collins station, induced seedlings have been used in several instances for production of seed in isolated locations where supplemental light was not available. In this undertaking an attempt was made to avoid the reversing effects of high temperature by earlier transfer of seedlings from the induction chamber to the field plots. The results were conflicting with respect to reversal and suggested the possibility that a relationship exists between the length of the pre-induction growth period in the greenhouse and the ability of a plant to flower when supplemental light is withheld throughout the post-induction period. In the experiments discussed in the preceding paragraphs the seedlings were quite young at the beginning of the induction treatment. Elapsed time from date of planting to the beginning of induction ranged from 9 to 14 days, and it was postulated that the need for supplemental light following the induction period was due in part to small plant size. The remainder of this report pertains to an experiment initiated in December 1960, primarily for the purpose of studying the relationship between the length of the pre-induction growth period and reversal.

Material and Methods

A bolting-resistant variety (US 75) and a variety having so-called "ordinary" bolting tendencies (GW359) were used in this study. Seed was planted in soil in 3-inch pots in the greenhouse, and the seedlings were thinned to 1 plant per pot soon after emergence. In the greenhouse, continuous illumination was provided, incandescent filament lamps being used at night, and temperatures were maintained approximately as follows: 9:00 AM to 4:30 PM, 77°F.; 7:00 PM to 8:00 AM, 60°. For the photothermal induction treatment, the plants were held continuously at a temperature of about 45° (± 3°) with light supplied entirely by means of incandescent-filament lamps.

The basic experimental plan involved induction treatments of 13 weeks for US 75 and 9 weeks for GW359, exposures considered adequate for the respective varieties. These induction treatments were to end on May 4, 1961, the date set for transfer of the plants into the open. Planting of seed was timed to provide pre-induction growth periods in the greenhouse of 2, 4 and 7 weeks for each variety. In the induction room, treatments and varieties were randomized and precautions were taken to avoid detrimental plant competition. Comparable sets of control plants were produced by planting seed in the greenhouse 2, 4 and 7 weeks prior to May 4.

On May 4, representative plants of each treatment within each variety were transferred to 6-inch pots (2 plants per pot) and placed in location 1. This location (outdoors) was covered with 1/4-inch mesh wire screen for hail protection and was divided into 2 comparable sub-locations as follows: (a) supplemental light provided throughout each night by means of two 150-watt, incandescent-filament lamps approximately 3 feet above the pots; and (b) no supplemental light provided at any time. Treatments and varieties were randomized within each sub-location, and precautions again were taken to avoid detrimental plant competition between age groups. The pots in each sub-location occupied an area approximately 6 feet wide and 14 feet long.

Location 2 consisted of field plots on an outlying farm where no supplemental light was provided. A randomized-block experimental design was employed with liberal plant spacing. Seedlings were transferred to this location on May 9. Consequently, induction treatments, with respect to location 2, were 5 days longer

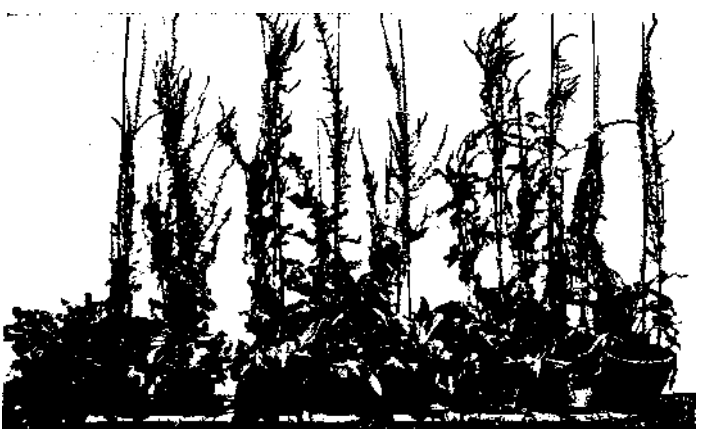

Figure 1.—Representative photothermally induced seedlings of the sugar beet variety US 75, 81 days after the end of induction treatment, showing the influence of age on reproductive development in a natural, long-day, post-induction environment without supplemental light. Each pot (size, 6-inch) contains 2 plants. The lengths of the pre-induction growth periods for plants in the 3 groups of 3 pots each, left to right, were 2, 4 and 7 weeks, respectively.

than indicated above. Likewise each set of control plants going into this location was 5 days older than originally planned.

Results

The appearance of foliage and seedstalks of US 75 in location l-b, near the end of the study, is illustrated in Figure 1. Flowering percentages for all locations, together with information as to treatments and the number of plants in each population, are summarized in Tables 1 and 2.

As expected, none of the control plants of US 75 flowered. Control plants of GW359 flowered to some extent in locations la, 1-b and 2, with a tendency toward higher flowering percentages among the older plants in each location, especially where supplemental light was not supplied. In this connection it is of interest that about one third of all GW359 plants of treatments 3 and 03 produced seedstalks that could be detected readily at the end of 7 weeks' growth under continuous illumination in the greenhouse.

The response of induced, potted seedlings to post-induction supplemental light may be summarized as follows: 1) In each variety, final flowering percentages for the respective age classes were consistently higher where supplemental light was supplied (location 1-a) than in the comparable location receiving no supplemental light (1-b), and the need for such illumination as a condition for flowering obviously was greater in US 75; 2) A tendency toward greater need for supplemental light by younger plants is indicated, especially in US 75 where final flowering percentages, in the absence of supplemental light (location 1-b), were 33, 50 and 67 for treatments 1, 2, and 3, respectively.

The relationship between age and the ability of induced plants to flower in the absence of post-induction supplemental light may be appraised further by inspection of the results obtained from location 2. Final flowering percentages for induced plants of GW359 in that location were confined to the range, 97 to 100, indicating negligible age effects. For comparable plants of US 75, on the other hand, the final flowering percentages for treatments 1, 2 and 3 were *66,* 75 and 94, respectively. This strong trend in US 75, toward higher flowering percentages for older plants, is in agreement with the corresponding results obtained for that variety in location 1-b. Analysis of variance, combining these 2 sets of results, showed that the trend was highly significant. Flowering percentages for this material, 11 weeks after the end of induction, were about the same as at the conclusion of the experiment.

Table 1.—Effects of age and supplemental light on flowering of photothermally induced and non-induced seedlings of the sugar beet variety GW359, Fort Collins, Colo., 1961.

Location and conditions after transplanting			Induc.[a] time (days)	Trans-plant, date	Plant[b] age (days)	No. of plants	Treat-ment no.	Elapsed time after transplant, and cumulative % of plants flowering			
Loc. no.	Soil	Suppl. light						5 wks.	7 wks.	11 wks.	17 wks.
l-a	In pots	Nightlong	63	5/4	14	24	1	4	83	100	100
					28	24	2	21	75	100	100
					49	24	3	33	58	100	100
			0	5/4	14	12	01	0	0	33	33
					28	12	02	0	8	17	25
					49	12	03	33	33	33	42
1-b	In pots	None	63	5/4	14	24	1	4	50	88	88
					28	24	2	21	54	79	83
					49	24	3	21	38	92	96
			0	5/4	14	12	01	0	0	0	0
					28	12	02	0	8	8	8
					49	12	03	42	42	42	42
2	In field	None	68	5/9	14	31	1	3	74	90	97
					28	32	2	9	72	100	100
					49	32	3	22	91	100	100
			0	5/9	19	18	01	0	0	0	0
					33	18	02	22	28	28	28
					54	18	03	28	28	28	28

[a] Photothermal induction treatment ended on date of transplanting.

[b] Age at beginning of induction treatment for induced plants and age at time of transplanting for non-induced plants.

Table 2.—Effects of age and supplemental light on flowering of photothermally induced and non-induced seedlings of the bolting-resistant sugar beet variety US 75, Fort Collins, Colo., 1961.

Location and conditions after transplanting			Induc.[a] time (days)	Trans-plant, date	Plant[b] age (days)	No. of plants	Treat-ment no.	Elapsed time after transplant, and cumulative % of plants flowering			
Loc. no.	Soil	Suppl. light						5 wks.	7 wks.	11 wks.	17 wks.
1-a	In pots	Nightlong	91	5/4	14	24	I	0	63	83	83
					28	24	2	0	58	88	88
					49	24	3	13	46	92	96
			0	5/4	14	12	01	0	0	0	0
					28	12	02	0	0	0	0
					49	12	03	0	0	0	0
1-b	In pots	None	91	5/4	14	24	I	0	21	33	33
					28	24	2	0	33	50	50
					49	24	3	4	29	67	67
			0	5/4	14	10	01	0	0	0	0
					28	12	02	0	0	0	0
					49	12	03	0	0	0	0
2	In field	None	96	5/9	14	32	I	9	47	66	66
					28	32	2	16	53	72	75
					49	32	3	6	63	94	94
			0	5/9	19	17	01	0	0	0	0
					33	18	02	0	0	0	0
					54	17	03	0	0	0	0

[a] Photothermal induction treatment ended on date of transplanting.

[b] Age at beginning of induction treatment for induced plants and age at time of transplanting for non-induced plants.

Discussion

It seems probable that the consistently higher final flowering percentages for the induced plants of each variety in location 2, as contrasted with those for the corresponding material in location 1-b, were due in part to the fact that the plants in location 2 had received slightly longer induction treatments. However, the magnitude of the differences in the case of US 75 suggests that other factors also were involved. In this connection it should be pointed out that the seedlings in location 2 were in field plots whereas those in 1-b were in 6-inch pots. The pots were placed on a hard surface without soil or other packing material between them and were spaced so as to avoid unfair competition between treatments. With the resultant exposure of the pots to sunlight, it is assumed that the daytime temperature of the soil in the vicinity of the crown and upper part of the taproot tended to be higher in the pots than in the corresponding places in the field. Such a temperature difference may have contributed somewhat to the observed contrast between locations 1-b and 2 in degree of flowering.

The results presented in this report indicated rather conclusively that, under conditions such as those prevailing in this experiment, the length of the pre-induction growth period is positively correlated with the ability of induced seedlings of some sugar beet varieties to flower in a natural, long-day, post-induction environment without supplemental light. The nature of this relationship is not clear, and further investigation seems desirable before an explanation is proposed. However, the knowledge that such a relationship exists should be of assistance to those using the seedling photothermal induction technique as a sugar beet breeding tool.

Summary

Seedlings of each of 2 sugar beet varieties were given starting periods in the greenhouse of 2, 4 and 7 weeks followed by photothermal induction treatments (continuous exposure to low temperature and artificial light) considered adequate for the respective varieties. Timing; was such that by May 4, 1961, the plants of GW359 and US 75 had received 9 weeks' and 13 weeks' induction exposure, respectively. On that date, representative seedlings were transplanted in pots in the open. Five days later the remaining plants were transplanted directly in field plots. Half the potted plants of each variety and age class were provided with continuous illumination during the post-induction period. None of the other plants received supplemental light during that

time. Comparable sets of non-induced plants were maintained as controls.

Results were evaluated on the basis of percentage of plants flowering in each population within 17 weeks after the end of the induction treatment. Two conclusions with respect to conditions similar to those prevailing in this study are of special interest: 1) The tendency of young, photothermally induced, sugar beet seedlings to revert to the vegetative phase in a natural, long-day, post-induction environment without supplemental light, apparently varies with variety; and 2) this reversal tendency can be reduced substantially in some bolting resistant material by an increase of several weeks in the length of the pre-induction growth period.

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Effect of Nitrogen Fertilization on Yield and Quality of Sugar Beets[1]

W. R. SCHMEHL, RALPH FINKNER AND JERRE SWINK[2]

Received for publication January 18, 1963

The need for greater efficiency in production of sugar beets has caused an increase in the use of nitrogen fertilizer on this crop. In some areas there has been excessive use of nitrogen which has resulted in lower quality beets. The influence that nitrogen fertilizer has on quality will depend not only upon the rate of fertilization but also upon other management practices as well as variety and season. The objective of this study was to determine the interactions of rate of nitrogen fertilization, date of planting, and plant spacing in the row on yield and quality of the beet.

Materials and Methods

The experiment was conducted on a Rorkv Ford loam located on the Arkansas Valley Branch Station near Rocky Ford, Colorado. Analysis of a representative soil sample from the experimental area showed 1.7% $CaCO_3$ 1-2% organic matter (6)[3], and 40 lb available P_2O_5 per acre by the sodium bicarbonate procedure (4). Previous crops were sorghum in 1959, corn in 1958 and alfalfa in 1957. Thirty-eight pounds of nitrogen was applied in 1959 for sorghum but no fertilizer was used for corn.

The experiment was designed as a factorial with 4 fertility treatments, 2 dates of planting, and 2 stands. There were 4 replications. The fertilizer treatments, expressed on an acre basis, were: 1) no-nitrogen check, 2) 40 lb nitrogen preplant, 3) 120 lb nitrogen preplant, and 4) 120 lb nitrogen side-dressed June 17, at the time of thinning. Preplant nitrogen fertilizer was broadcast in the spring before plowing. The side-dressed fertilizer was applied about five inches from the row in alternate middles not used for irrigation. Ammonium nitrate was the nitrogen source. Concentrated super phosphate, applied at the rate of 100 lb P_2O_5 per acre, was broadcast uniformly over the experimental area before plowing.

[1] Department of Agronomy and Arkansas Vallev Branch Experiment Station. Colorado Agricultural Experiment Station in cooperation with the American Crystal Sugar Company. The research was financed, in part, bv grants from the United States Steel Company and the Southern Colorado Beet Growers Association. Colorado Agricultural Experiment Station Scientific Series No. 841.

[2] Agronomist, Colorado Experiment Station, Manager, Research Station, American Crystal Sugar Company, and Superintendent, Arkansas Valley Branch Station, respectively.

[3] Numbers in parentheses refer to literature cited.

Planting dates were April 7 and May 3, 1960, with a commercial monogerm seed. The sugar beet stands established for the experiment were 1) 12 to 14 inch spacing and 2) 6 to 7 inch spacing of plants in 22-inch rows. The heavy population was not attained in harvested beets, however, and the average stand counts based on beets recovered at harvest were 9 inches for the heavy stand and 13 inches for the normal stand. Some of the smaller beets from the heavy population were lost during harvest which reduced recovery of beets and the apparent stand.

The beets were harvested November 3, and root samples were taken for sugar, purity and analysis of the pressed juice. Crop yields may have been reduced somewhat by a severe hail in June; otherwise, growing conditions were good. At harvest, foliage on the check treatments exhibited marked nitrogen deficiency symptoms and the treatments receiving 40 lb nitrogen showed slight nitrogen deficiency.

Leaf petioles were sampled July 12, August 5 and September 15. The petioles were analyzed for total nitrogen, nitrate-nitrogen, and acetic-acid soluble phosphorus (3). The pressed juice in the beets at harvest was analyzed for nine amino acids by paper chromotography methods as outlined by Hanzas[4]. Galactinol and raffinose were determined by paper chromatography procedures similar to those reported by Brown (2). Potassium and sodium were determined with the flame photometer (1) and total nitrogen by a microkjeldahl procedure (5).

Results and Discussion

Yield and Quality of Roots

Root yields, sucrose content, sucrose production and purity are summarized in Table 1 for the main effects of fertilizer treatment, date of planting and spacing in the row. The analysis of variance (Table 1) shows that the main effects of fertilizer treatment were significant for root yield, sucrose content and purity, but not for sucrose production. The first increment of nitrogen (40 lb) increased the yield of roots, but there was no additional response to the next increment of nitrogen. Both sucrose content and purity decreased with each nitrogen rate. The application of nitrogen did not significantly increase sucrose production at the five percent level of significance. The trend was for a small increase for the 40-lb rate followed by a decrease *in* sucrose for the 120-lb rate of nitrogen.

[4] "A paper chromatographic method for semiquantitative analysis of amino acids found in sugar beet juices" (1957); unpublished report by P. C. Hanzas, Research Station, American Crystal Sugar Company.

Table 1.—Effect of fertilizer treatment, date of planting and plant spacing in the row, on yield and quality of sugar beets.

Treatment	Root yield T/A	Sucrose %	Sucrose production T/A	Purity %
lb N/A				
0 (check)	20.7	16.4	3.39	90.3
40	22.6	15.4	3.48	89.2
120	22.2	14.5	3.22	86.9
120[1]	22.0	15.0	3.30	89.1
Early plant	22.9	15.4	3.53	88.6
Late plant	21.6	14.9	3.22	88.6
9 in spacing	22.3	15.2	3.39	88.4
13 in spacing	22.3	15.2	3.39	88.7
Significance for:				
Fertilizer treatment	**	**	N.S.	**
Planting date	*	+	**	N.S.
Plant spacing	N.S.	N.S.	N.S.	N.S.

\+ Significant at 10% level of significance
* " 5% " "
** " 1% " "
[1] side-dressed at thinning

The early planted beets yielded 1.3 tons more than the late planted beets, and were 0.5% higher in sucrose content (Table 1). Sucrose production was significantly lower for the second date of planting. Purity was not affected by date of planting.

There were no significant effects of plant stand on yield or quality of root (Table 1). The lack of significance may have been caused, at least in part, by too small a difference between the two stands.

The application of 120 lb nitrogen at thinning resulted in about the same yield and quality of beet as the same rate of fertilizer applied preplant. There was no apparent adverse effect on quality.

Of greater interest than the main effects are the interactions between rate of nitrogen and date of planting (Table 2). Planting date had little effect on yield of the check treatment (no nitrogen); on the other hand there was a significant yield response to 40 lb nitrogen for the early planting but not for the late planting. There was no further response to the 120-lb rate of nitrogen for either planting date. When 120 pounds nitrogen was side dressed at thinning for the early or late-planted beets, yield and quality of roots did not differ significantly from results obtained with the same amount of nitrogen applied preplant.

There was a marked decrease in sucrose content when nitrogen fertilizer was applied and the decrease tended to be greater for

Table 2—Influence of nitrogen fertilizer and date of planting on yield and quality of sugar beets.

Fertilizer lb N/A	Planting date	Root yield T/A	Sucrose %	Sucrose T/A	Purity %
0 (Check)	4-7-60	20.5	10.5	3.38	90.6
40	"	23.6**	15.6	3.68*	90.5
120	"	22.9**	14.9*	3.41	86.7*
120[1]	"	22.2**	15.1*	3.35*	88.4*
0 (Check)	5-3-60	20.9	16.3	3.41	90.0
40	"	21.5	15.2	3.27	87.9
120	"	21.6	14.2**	3.08*	87.1
120[1]	"	21.9	14.7*	3.22	89.9

* Differs from check for the same date of planting at 5% level of significance.
** Differs from check for same date of planting al 1% level of significance.
[1] Nitrogen fertilizer side-dressel at thinning.

the late planted beets. The application of nitrogen fertilization caused a decrease in purity of the beet, but the effect was not as pronounced as for sucrose content.

For the early planting, the application of 40 lb nitrogen increased sucrose production whereas there was the trend for the same amount of nitrogen to cause a small decrease in sugar for late-planted beets. The high rate of nitrogen significantly decreased sucrose production for the late planting. If the value of the sugar is calculated as 4c per pound, and the cost of nitrogen is 13c per pound, 0.065 tons sugar per acre is required to equal the cost of 40 lb of nitrogen, and 0.195 tons sucrose is needed to pay for 120 lb nitrogen. On this basis 40 lb nitrogen for the early planting was the only fertilizer treatment (Table 2) that gave an increase in sucrose production large enough to balance or exceed the cost of the fertilizer. The results emphasize the need to consider date of planting when making nitrogen fertilizer recommendations for beets. The recommended rate of fertilization with nitrogen should be flexible, and if planting is delayed it may be necessary to reduce the amount of fertilizer applied.

Petiole Analyses

The results of petiole analyses for nitrate-nitrogen and acid soluble phosphorus are given in Table 3 for three sampling dates. There were no significant differences between stands for petiole nitrogen or phosphorus at any sampling date.

The application of nitrogen fertilizer increased the nitrate-nitrogen in the petioles. The effect persisted throughout the season and was associated, at harvest, with lower sucrose in the root (Table 1). The petiole nitrate was slightly lower for the delayed application of nitrogen than for the same amount of

Table 3.—Effect of fertilizer treatment and date of planting on nitrate-nitrogen and acid-soluble phosphorus in petioles.

Treatment	Nitrate-nitrogen sampling date			Acid-soluble phosphorus sampling date		
	7-11-60	8-5-60	9-9-60	7-11-60	8-5-60	9-9-60
lb N/A		ppm NO3-N			ppm P	
0 (Check)	7810	4600	1140	2400	1600	970
40	9580	4980	1820	2270	1580	1040
120	14610	9490	3 J 50	2180	1560	1050
120[1]	11830	6160	1930	2330	1620	980
Early plant	9370	5200	1330	2070	1510	1050
Late plant	13920	8330	3420	2500	1690	980
Significance for Fertilizer treatment	**	**	**	N.S.	N.S.	N.S.
Date of planting	**	**	**	**	**	N.S.

** Significant F-test at 1% level of significance.
[1] Nitrogen fertilizer side-dressed at thinning.

nitrogen applied at planting; and sucrose content and quality were slightly higher for the delayed treatment (Table 1). The nitrogen for this treatment was side dressed at thinning between alternate unirrigated rows. Placement of the fertilizer in this position may have been less favorable for efficient absorption of nitrogen by the plant than plowing under the fertilizer for the preplant application.

Nitrate nitrogen was higher in the petiole for the late-planted beets at all sampling dates. This was associated with a lower sucrose content of the root for the late planted beets (Table 1). The results show that the late-planted beets did not "ripen" to the same degree as did the early-planted beets.

The acid soluble phosphorus content of the petioles was not affected by the application of nitrogen fertilizer. Phosphorus was higher in petioles from the late-planted beets for the July and August samplings but not for the September sampling. The trends were probably caused, as with nitrate nitrogen, by reduced plant growth and lower crop demands on soil nutrients by the late-planted beets.

Chemical Composition of Pressed Juice

Total nitrogen, total amino acids, sodium, potassium and raffinose were determined in the pressed iuice. The results are summarized in Table 4. The nine individual amino acids were summed for total amino acids in Table 4 since individual statistical analyses showed that they all reacted in a similar way to the imposed treatments.

Table 4.—Effect of fertilizer treatment and date of planting on partial chemical analysis of pressed juice (percent on dry basis).

Treatment	Total N- %	Total amino-acids-f %	Sodium[3] %Na	Potassium[3] %K	Raffinose[2] %	Galactinol[2] %
lb N/A						
0 (Check	0.56	0.71	0.067	0.153	0.40	0.25
40	0.65	0.84	0.086	0.165	0.41	0.25
120	0.74	1.23	0.111	0.164	0.42	0.30
I20[1]	0.68	0.96	0.084	0.157	0.43	0.25
Early plant	0.61	0.81	0.083	0.155	0.43	0.26
Late plant	0.73	1.00	0.097	0.163	0.42	0.27
Significance for:						
Fertilizer treatment	**	**	**	N.S.	N.S.	N.S.
Planting date	**		*	N.S.	N.S.	N.S.

+ Sum of nine amino acids: alanine, asparagine, aspartic acid, glutamine, valine, isoleucine, glycine, glutamic acid, gamma amino butyric acid.
* Significant F-test at five percent level.
** Significant F test at one percent level.
[1] Side-dressed at thinning.
[2] Dissolved substance basis.
[3] On beet root weight.

The total nitrogen and amino acid contents of the pressed juice increased when nitrogen fertilizer was added, and were higher for the late-planted beets. Trends for nitrogen components in the pressed juice were similar to the nitrate-nitrogen in the petiole and were negatively associated with sucrose content of the root.

Sodium in the pressed juice increased with an increase in rate of nitrogen fertilizer and was also higher for the late planting. The sodium content of the pressed juice was positively associated with changes in total nitrogen and amino acids in the pressed juice and with nitrate-nitrogen in the petioles. In experiments conducted in northeastern Colorado, it has also been observed that where application of nitrogen fertilizer increased nitrate-nitrogen in the petiole, the sodium content also increased (7).

The potassium, raffinose, and galactinol contents of the pressed juice were not influenced, at the five percent level of significance, by either nitrogen fertilization or date of planting.

Summary

An experiment was conducted to study the influence of nitrogen fertilization, date of planting, and plant spacing in 22-inch rows on yield and quality of sugar beets.

Early planted beets produced higher yields and more sucrose than late-planted beets.

Forty-pounds nitrogen increased sugar production when the beets were planted April 7, but the same amount of nitrogen applied to beets planted May 3, failed to increase sugar production. Application of 120 lb nitrogen decreased sucrose production for the late planting.

The application of nitrogen fertilizer increased the total nitrogen and amino acids in the pressed juice and nitrate-nitrogen in the petioles and decreased the sucrose content and purity of the root. Sodium content of the pressed juice was associated in a positive relationship with nitrogen content of the pressed juice and nitrate-nitrogen in the petiole.

Plant spacings of 9 and 13 inches in the row had little effect on yield and quality of root.

The experiment demonstrates the need to adjust the rate of nitrogen fertilization with date of planting in climatic areas where the harvest date cannot be extended.

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Effect of Temperature During Anthesis and Seed Maturation on Yield and Germinability of Sugar Beet Seed[1]

F. W. SNYDER AND G. J. HOGABOAM[2]

Received for publication February 14, 1963

Under field conditions, the quantity and quality of seed produced by plants of a given sugar beet variety may vary considerably. Although relatively little is known about the causes of variations, changes in specific environmental factors from year to year seem to be involved. Temperature appears to be responsible for part of the variations.

Temperature markedly affected seed production in garden stock, *Matthiola incana* (L.) R. Br. (2)[3]. Seed was produced on garden stock plants at 55, 65, 75, and 85 F, but the yield of fruits and seeds per plant was greatest at 65 F. At 55 F, less flowers were formed. At 75 F, approximately the same number of flowers were formed, but only 50% developed fruits. The plants at 85 F were sterile because of various abnormalities.

Since temperature is controllable in the greenhouse at certain times of the year, a study of its effect on seed quality of sugar beets was initiated in 1956. This paper reports the effect of temperature during anthesis and seed maturation on yield, germinability, and certain other plant and fruit characters.

Methods and Materials

Sugar beet clones were used to minimize genetic variability, and to evaluate more precisely the effect of the experimental conditions with a relatively small number of plants.

Four experiments were conducted in the greenhouse at East Lansing, Michigan. During the day, the temperatures were regulated by manually opening and closing the vents. Temperatures in excess of approximately 73 F depended on solar radiation. Temperature in the third experiment was less precisely controlled because of the duration of the experiment.

The experimental plants received adequate amounts of a complete mineral nutrient solution and water. In Experiment 1, they were grown in 8-inch pots containing sand. In Experi-

[1] Cooperative investigations of the Crops Research Division. Agricultural Research Service, U. S. Department of Agriculture, and the Michigan Agricultural Experiment Station. Approved for publication as Journal article #2188, Michigan Agricultural Experiment Station.

[2] Plant Physiologist and Research Agronomist, respectively. Crops Research Division. Agricultural Research Service, U. S. Department of Agriculture, East Lansing, Michigan.

[3] Numbers in parentheses refer to literature cited.

ment 2, the sib-fertile plants used as the male clones were grown in 10-inch pots on vermiculite and the self-sterile plants used as the female clone in 4-gallons crocks containing vermiculite. Ten-inch pots containing vermiculite were used in Experiments 3 and 4.

All plants were photothermally induced and maintained at a cool temperature until just before anthesis. They were kept on a long photo-period during anthesis, unless specifically stated otherwise. The experimental temperatures were initiated at first anthesis. For the temperature data for Experiment 1, see Figure 1; for Experiment 2, Figure 2; and for Experiment 3, Table 1.

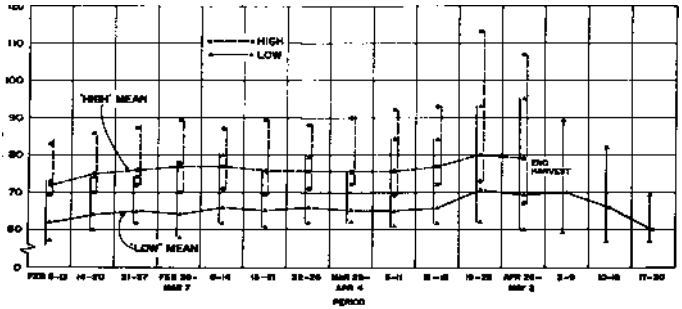

Figure 1.—Weekly mean temperatures and the high and low temperatures for each week during anthesis and seed maturation to which clones of US 401 sugar beet were exposed, Experiment 1.

To minimize the effect of maturity on germination, seed was harvested when at least 80% of the fruits on a plant were straw-colored. Seed was harvested from each of the plants within a temperature treatment. In Experiments 2 and 3, the shoots of the plants were removed approximately 8 inches above the crown and their fresh weights were recorded. The shoots were dried at 90 F and then the seed was removed, cleaned, and weighed.

In Experiments 1 and 2 speed of germination was determined by the liquid-contact method (3), and in Experiment 3 by the blotter method (water).

Experimental details specific to each experiment follow:

Experiment 1. The experimental plants were progeny from open-pollinated seed of one plant of US 401. Thirty pairs of vegetative cuttings from 18 clones were used. In February 1957, just before anthesis, one plant of each pair was kept at a mean temperature of approximately 66 F. These were designated "low-temperature"

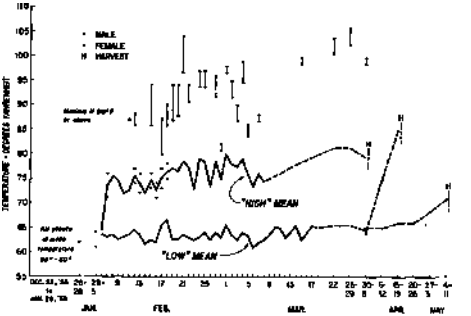

Figure 2.—Air temperatures of the rooms in Experiment 2. Daily mean temperatures (solid lines) were plotted during anthesis. Weekly mean temperatures (broken lines) were plotted for the remainder of the experimental period. During anthesis, temperatures in the low-temperature rooms never exceeded 80 F. "High" mean indicates the average temperature of the high-temperature rooms in which the female and male plants were kept. Deviations between the rooms of more than one degree are indicated by appropriate symbols above and below the mean line. Maximum temperatures in excess of 80 are plotted for the rooms at the higher mean temperature.

plants. The others, designated "high-temperature" plants, were held at a mean temperature of approximately 76. All plants received supplemental light (fluorescent) to approximate 20 hours of light daily between February 5 and 26. After February 26, the high-temperature plants received no supplemental light (natural length of day approximately $12^{1/2}$ hours). The low-temperature plants received supplemental light to maintain a day-length of approximately 14 hours until April 15, when the supplemental light was discontinued. These light and temperature conditions were selected as representative of the environment in the seed-producing areas of California and Oregon. Pollen was dispersed with a hand vacuum cleaner (operated in reverse) during part of the period of anthesis. Dates of initiation of anthesis and harvest were recorded for each plant.

Experiment 2. Seven of the 18 clones employed in Experiment 1 were selected for further study. These clones also served as the pollinators for a self-sterile clone in a study designed to determine how high temperature reduces seed yield.

Before January 28, 1959, all plants were maintained under uniform conditions. The experimental temperatures were initiated on February 6 and 7, just before anthesis. Plants within a

clone were paired for uniformity of development so that each plant would have a counterpart at the other temperature. The 7 sib-fertile clones, each clone consisting of 4 plants, were designated as "male plants". Half of these male plants (2 from each of the 7 clones) were placed in a greenhouse room maintained at a mean temperature approximating 64 F and were designated as "low-temperature male plants." The other group of 14 male plants was placed in an adjacent greenhouse room at a mean temperature approximating 76 and designated as "high-temperature male plants."

The self-sterile clone, consisting of 26 plants, was designated "female." Half of these female plants were isolated in a third greenhouse room maintained at a mean temperature approximating 64 F and were designated as "low-temperature female plants." The other group of female plants was isolated in a fourth greenhouse room at a mean temperature approximating 76 F and designated as "high-temperature female plants." Thus, 13 female plants were placed at each temperature. Within each group of 13 female plants, one plant was isolated in the room to determine how much seed would be produced from stray pollen or an occasional selling. Of the remaining 12, 6 plants were placed on one cart and pollinated by the low-temperature male plants. The other 6 plants were handled similarly and pollinated by the hiqrh-temperature male plants. The factorial procedure outlined diagrammatically in Figure 3 was used in an attempt to separate the effects of temperature on the male and female gametes which cause the decrease in yield at high temperature.

The date of first blossom was recorded for each plant. First anthesis occurred between Februarv 8 and 22. Pollination was initiated on February 9. The female plants were placed in the room with the male plants daily for approximately 15 minutes (the pollen was blown from the male plants onto the female plants by means of a hand vacuum cleaner) and then returned to isolation in their temperature room. The pollinations generally were performed between 11 AM and noon. Temperatures and relative humidities were recorded for the entire experimental period.

Because of the more rapid development at the hiorher temperature, pollen production was greatly reduced by March 8, but at the lower temperature pollen production continued longer. Pollination of the female plants with hicrh-temperature pollen was discontinued on March 9 and with low-temperature pollen on March 19. The grand periods of flowering of plants maintained

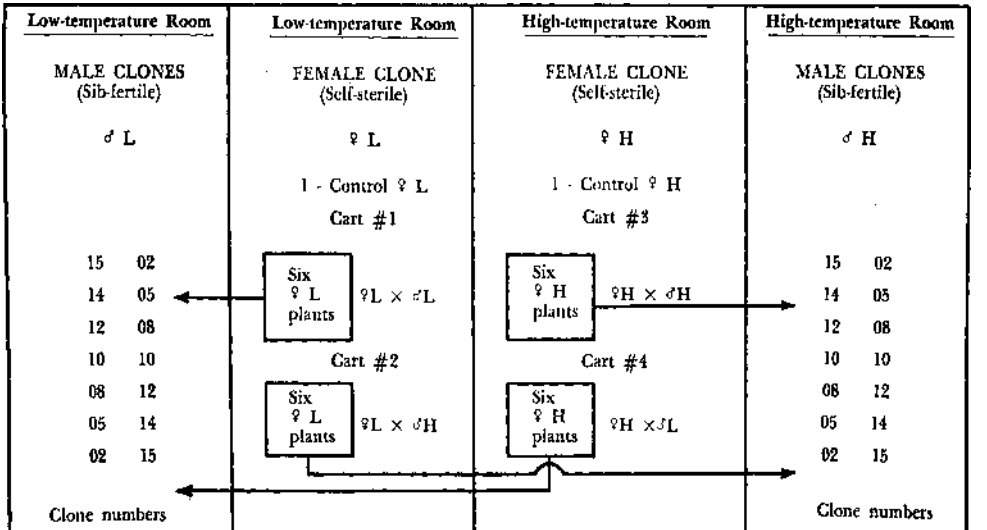

Figure 3.—Procedure for isolation and pollination of female clone used in Experiment 2 to study the effect of temperature on yield of sugar beet seed. Systematic procedures were avoided to minimize the positional effects on plant response.

at different temperatures were not synchronized perfectly in this experiment, but the differences in time of anthesis did not seem to influence the results.

The control female plant of the self-sterile clone, isolated in the low and high temperature rooms to determine the amount of stray pollen and selfing, yielded less than a gram of seed in each location. The magnitude of this yield was considered to be so low that no corrections were applied to the seed yields of the female plants exposed to pollination.

Experiment 3. Roots of the monogerm variety SP 5832-0 were selected from nursery plots at East Lansing, Michigan, in 1959. These roots were grown from open-pollinated seeds harvested from 17 different mother plants. One to 11 roots were selected from each progeny group.

Each root was sawed into nearly equal halves and the halves placed in separate pots. To more nearly synchronize anthesis and seed harvest at the two temperatures, we kept the first member of the paired half-roots to bloom at a mean temperature of approximately 65 F during anthesis, and the second member of the pair at a mean temperature of approximately 75 F. Thus, plants maintained at the lower temperature bloomed earlier, usually a few days but rarely as much as two weeks. During seed development and maturation, the temperature differentials were maintained as long as the out-door temperature did not exceed the lower mean temperature. The higher temperature treatment was initiated on Tanuary 29, 1960. The first plants began anthesis about January 25 and the last on April 17. Seed harvest was completed by June 10.

To determine the range in the size of fruits among the plants of a progeny group in a given temperature environment and between the plants of paired half-roots at the two temperatures, we sized a sample of approximately 20 grams (when available) for fruit diameter and then for fruit thickness, using round-holed and slotted screens, respectively. The percentages by weight were calculated for each size-class. Fruits having more than a single ovarian cavity were removed from the sample and the percentage by weight and number of these fruits were recorded. In addition, to see whether temperature during maturation had any effect on the size of the processed fruits, 25 fruits of given size-classes, which matured at the low and at the high temperature, were hand processed. The size of each of the fruits was determined and average sizes recorded for each temperature.

Experiment 4. Twenty of the monogerm clones of SP 5832-0 used in Experiment .3 were selected for a range in sensitivity to high temperature as measured by yield of seed. All plants were kept at a mean temperature approximating 65 F during anthesis. Between November 11 and December 1, 1960, two flowers were removed daily until 20 had been examined for each plant. The lengths of the pistil (style plus distance to center of ovarian cavity) and the style were recorded for each flower. The average lengths of the styles were correlatd with the yields of seed for 16 of the clones in Experiment 3.

A branch of each plant was isolated in a bag before anthesis to determine the degree of self-fertility at a temperature approximating the lower mean temperature employed in the earlier experiments.

Results

Effect of relative humidity on yield of seed

The relative humidity was recorded in the two temperature regimes of Experiment 2. The humidity usually ranged from 35 to 70% for the daylight hours during anthesis. Although the relative humidity was higher in the low-temperature rooms, differences in humidity between the temperature regimes were not great. The higher relative humidity of the low-temnerature rooms did not appear to affect the setting of seed adversely.

Effects of temperature

The effects reported *in* the following sections are caused mainly by the temperature at which the plants bloomed and the seeds and their surrounding fruits developed and matured.

Time to mature: Temperature affected the length of time required from the start of anthesis until the seed matured. For the 66 F mean temperature in Experiment 1, the average number of days was 71 (range 61 to 90 days), whereas for the 76 F mean temperature the average was 57 (range 44 to 69).

Seed yield: Temperature during anthesis markedly affected the yield of seed. The higher temperature depressed yields to about half of those at the lower (Table 2). The ratio of shoot weight to seed (fruit) weight represents an efficiency index for seed production and indicates the number of grams of shoot (fresh weigrht) required to produce a gram of seed. Plants grown at the lower mean temperature were more efficient than those at the higher. Progeny groups within a variety differ in efficiency of seed production and appear to be differentially affected by temperature during anthesis (Table 3). Individuals within a progeny group of SP 5832-0 differ in efficiency of seed production, particularly at the higher mean temperature (Table 4).

Table 1.—Resume of temperatures (F) in Experiment 3 during anthesis* of clones of monogerm sugar beet variety SP 5832-0.

	Low temperature room							Hiigh temperature room						
	Mean for	Range in mean daily	Maximum temp, in	No. days max. temp, equalled or exceeded			Mean for	Range in mean daily	Maximum temp. in	No. days max. temp. equalled or exceeded				
Period	period	temp.	period	80	85	90	period	temp.	period	80	85	90	95	100
Jan. 29-Feb. 29	61	59-64	78	0	0	0	72	70-75	87	6	2	0	0	0
Mar. 1-31	62	60-67	80	1	0	0	77	71-79	95	26	20	10	1	0
Apr. 1-30	66	61-74	90	5	2	1	77	72-85	104	23	21	15	10	3
May 1-17	65	61-70	84	4	0	0	78	73-85	101	11	9	9	7	1

* Plants blossomed between January 29 and May 17, depending on temperature and the clones themselves. The period of anthesis for a plant was considered to be a 30-day period after opening of the first blossom.

Table 2.—Effect of temperature during anthesis on yield of sugar beet seed.

Exp. no.	Variety	No. of clones	No. of plants	Approx. mean temp. (F)	Avg. fresh weight of shoots per plant (grams)	Avg. weight of seeds per plant (grams)	Ratio of shoot wt. / seed wt.
1	US 401	18	30	66		43.2 ± 14.6#	
			30	76		19.4 ± 4.5#	
2	US 401	7	14	64	782	84.8	9.2
			14	76	485	40.8	11.9
3	SP 5832-0	58	58	65	582	77.9	7.5
			58	76	455	30.5	14.9

\# Standard deviation

Table 3.—Effect of temperature (F) during anthesis on efficiency of seed production by different progeny groups of monogerm sugar beet variety SP 5832-0.

Progeny group	No. of paired half-roots	Shoot wt./seed wt. for plants grown at mean temp, approx. 65 Ratio	65 Rank	76 Ratio	76 Rank
4	5	5.34	1	9.88	1
40	4	6.52	2	12.90	2
137	11	6.89	3	14.68	5
82	3	6.96	4	16.63	7
30	4	6.98	5	13.38	3
52	5	7.18	6	18.07	8
123	7	8.03	7	15.66	6
59	4	8.23	8	23.63	9
70	4	9.85	9	13.48	4

Table 4.—Effect of temperature (F) during anthesis on efficiency of seed production by plants within three progeny groups of sugar beet variety SP 5832-0.

Progeny group and root number	Ratio of shoot wt./seed wt. for each half of paired-root grown at mean temperature approximating 65	76
4:8	4.83	8.91
3	5.10	7.14
1	5.39	12.82
7	5.67	11.90
6	5.72	9.01
52:2	4.93	9.86
5	7.05	9.28
3	7.31	50.56
4	7.44	28.08
1	9.67	32.80
59:5	6.85	11.02
3	8.20	32.78
4	9.35	18.64
2	9.36	53.53

Size, weight, and number of fruits: Temperature at which the fruits developed and matured affected size, weight, and number of fruits (Table 5). Fruits matured on plants at the lower mean temperature were larger, heavier, and greater in number. Temperature influenced monogerm fruits similarly (Table 6). The progeny-group data also indicate considerable genetic variability within a group. Temperature during maturation of the fruits affects the size of the fruits obtained after processing (Table 7). Fruits matured at the higher temperature process to smaller mean dimensions than those matured at the lower temperature.

Tendency for monogermness in the monogerm variety SP 5832-0: When plants of the same clones were grown at two temperatures, they usually produced a greater percentage of fruits having two or more ovarian cavities at the lower temperature

Table 5.—Effect of temperature (F) on size, weight, and number of fruits on US 401 sugar beet plants.

Exp. no.	No. of clones	No. of plants	Approx. mean temp.	Average size of fruit	Average weight of 200 fruits (grams)	Calculated average number of fruits per plant
1	9	13	66	38.6% > 12/64 inch**	3.71**	2,477**
		13	76	11.3% > 12/64 inch	2.70	1,503
2	7	14	64		3.83**	4,390**
		14	76		2.74	2.989

* Statistically significant difference at 1% level.

Table 7.—Influence of temperautrc during maturation of the fruits on the reduction in size of fruits in processing operation. Clones of sugar beet variety SP 5832-0 grown in Experiment 3.

Median size-class* of whole fruits	Number of clones compared	Mean size* of processed fruits matured at		Reductiion in size of fruit due to processing			
		Low temperature	High temperature	Low temperature		High temperature	
				Diam.	Thick.	Diam.	Thick.
				%			
14 x 9.5	3	11.3 X 6.5	10.6 x 6.4	19	32	24	33
14 x 8.3	8	11.1 X 6.1	10.0 x 5.9	21	27	29	29
12 X 9.5	3	10.5 X 6.3	9.6 x 6.1	13	34	20	36
12 X 8.3	12	10.3 x 5.8	9.3 X 5.6	14	30	23	33
12 X 6.8	18	9.7 X 5.3	8.6 x 4.9	19	22	28	28
10 x 6.8	10	8.4 x 5.0	7.6 x 4.8	16	26	24	29

* In 64ths of an inch. First figure is fruit diameter and second is fruit thickness.

Table 6.—Effect of temperature on fruit size in 3 progeny groups of monogerm sugar beet variety SP 5832-0, Experiment 3.

Progeny group and root number	Fruit size*	Percentage by weight of sample on scren			
		Fruit diameter		Thickness	
		Low temp.	High temp.	Low temp.	High temp.
4:3	9 X 7.5	46.8	66.9	5.3	4J5
1		50.4	59.6	0.7	0.0
6		89.5	55.0	34.2	11.5
8		90.4	90.3	36.2	2.3
7		95.1	43.0	49.5	4.1
30:6	11 X 7.5	65.9	39.6	46.5	17.8
7		71.2	50.1	66.9	37.7
1		84.5	46.1	89.0	55.3
4		93.6	54.1	83.4	31.2
40:5	11 X 7.5	2.0	1.0	1.8	0.6
2		82.8	61.6	55.1	23.4
1		86.0	67.9	79.9	44.5
3		89.9	30.3	81.7	27.2

* In 64ths of an inch. First figure is fruit diameter and second is fruit thickness.

Table 8.—Variability for monogermness of plants with a common female parent (No. 123) in sugar beet variety SP 5832-0.

root number	Multigerm fruits in sample from clone grown at			
	Low temperature		High temperature	
	Number	Percent by wt.	Number	Percent by wt.
1	11	1.0	5	0/7
2	2	0.1	2	0.05
3	61	6.4	17	1.6
4	79	10.4	44	4.3
5	238	24.2	154	11.6
6	9	1.0	19	1.7
7	0	0.0	3	0.4

(Table 8). The large variation in tendency toward monogermness within a progeny group at a given temperature is illustrated, too.

Tightness of seedcaps: The seedcaps which cover the ovarian cavities of the fruits usually are cemented tightly enough to remain in position during normal handling of the fruits. However, shedding of seedcaps is observed in many varieties and differences within lines have been noted (1). Fruits of 10 clones from Experiment 1 and 7 from Experiment 2 were examined to determine what percentage of fruits had shed seedcaps. Two hundred fruits were examined for each sample in Table 9. The tendency for a greater percentage of the seedcaps to shed at the higher temperature was marked, but there was an interaction between clones and temperature.

Development of seeds: To determine what may have contributed to the larger multigerm fruits on plants grown at the lower temperature in Experiment 1, we examined 200 fruits per

Table 9.—Effect of temperature (F) on the shedding of seedcaps covering the ovarian cavities of US **401** sugar beet.

Clone number	Number of paired samples examined	Percentage of fruits having shed one or more seedcaps at indicated mean temperature			
		Experiment 1		Experiment 2	
		66	76	64	76
50612	2	17.4	85.2	27.4	87.1
50625	1	2.0	27.5		
50609	2	1.0	7.5		
50611	1	1.5	4.0		
50607	1	0.5	3.5		
50606	1	0.5	2.5		
50602	2	0.0	2.5	27.8	5.0
50601	1	1.5	1.0		
50608	2	10.5	0.5	7.4	9.4
50604	2	0.5	0.0		
50610	2			5.0	16.8
50605	2			0.8	11.3
50614	2			2.1	10.5
50615	2			0.6	2.1

Table 10.—Effect of temperature (F) on percentage of fruits containing developed seeds in each ovarian cavity of clones of US 401 sugar beet.

Clone number	Number of pairs of plants	Percentage of fruits having developed seeds in all ovarian cavities at indicated temp.*	
		66 F	76 F
50602	2	75.0	26.5
50607	1	67.5	28.5
50608	2	61.5	44.0
50625	1	57.0	14.0
50609	2	52.0	25.8
50611	1	49.5	29.5
50606	1	48.0	24.0
50601	1	4:3.0	18.0
50604	2	38.5	9.5
Average		55.3	25.0

* Percentages based on 200 fruits per plant.

plant to determine the number of ovarian cavities per fruit and the number of cavities containing developed seeds. The percentage of fruits which contained a developed seed in each ovarian cavity varied considerably at a given temperature (Table 10). The higher temperature was most detrimental to seed development. Additional data are presented in Table 14.

Speed of germination of seed: For Experiments 1 and 2, we germinated the seed by the liquid-contact method, using a nutrient solution of 10.1 atmospheres osmotic pressure. Eighty seedballs from each plant at each temperature were vised. In Experiment 1, the temperature at which the seed developed and the fruit matured did not affect significantly the speed of germination of the clones as a group. However, 3 clones which grew at the higher temperature produced significantly faster germinat-

Table II.—Effect of temperature during seed development and fruit maturation on speed of germination of seed from 7 clones of US 401 sugar beet, Experiment 2.

Temperature	Percentage of germination in indicated days* 2	3	5
Low	28.1	67.6	77.3
High	56.0	84.6	89.2

* Data based on 1120 seedballs from each temperature (7 clones X 2 plants per clone X 80 seedballs per plant).

ing seeds than their counterparts produced at the lower. The significant interaction (5% level) between temperature and clones suggests that some clones were more sensitive to the temperature employed than others. Regardless of temperature, speed of germination for the clones differed significantly. Of the 3 clones in Experiment 1, which had seeds that germinated more rapidly when produced at the higher temperature, 2 were included in Experiment 2. The germination data for the 7 clones used as pollinators in Experiment 2 are shown in Table 11. The seeds that developed and matured at the higher temperature were significantly faster than those produced at the lower. Seeds of 6 of the 7 clones germinated significantly faster, but those of clone 50602 were essentially unaffected by temperature. Seeds of clones 50605 and 50615 produced at the higher temperature germinated significantly faster in both experiments. Detailed data for *S* of the clones used in Experiment 2 (Table 12) reveal that clones 50605 and 50615 were affected by the high temperature, but clone 50602 was less sensitive and somewhat less consistent.

Table 12.—Effect of temperature during seed development and fruit maturation on speed of germination of seeds of 3 clones of US 401 sugar beet, Experiment 2.

Clone and **plant** pairing **numbers**	Temperature	Percentage germination in indicated days* 2	3	*a*
50602:				
1	High	65	96	99
2	High	33	64	73
1	Low	51	78	79
2	Low	31	76	83
50605:				
1	High	63	90	94
2	High	89	100	100
1	Low	45	91	94
2	Low	39	83	85
50615:				
1	High	35	64	69
2	High	34	61	68
1	Low	4	16	38
2	Low	4	20	31

* Each percentage based on 80 seedballs.

lable 13.—EHect ot low *(L)* and high (H) temperatures on weights (grams) of fresh shoot and air dried seed for the self-steri!e clone (female) of sugar beet pollinated with pollen *(male)* produced at two temperatures, Experiment 2.

	Temperature designation of female plant and pollinator							
	♀ L × ♂ L		♀ L × ♂ H		♀ H × ♂ L		♀ H × ♂ H	
Replication number	Weight ot Shoot	Seedballs	Weight ot Shoot	Seedballs	Weight ot Shoot	Seedballs	Weight ot Shoot	Seedballs
1	500	42.0	565	48.0	520	24.0	500	34.0
2	500	27.5	475	20.0	545	27.0	320	19.5
3	500	30.0	610	29.0	270	20.0	320	18.5
4	545	60.5	610	46.0	635	39.0	590	39.0
5	250	21.5	475	22.0	520	45.0	500	33.0
6	770	52.5	180	23.5	475	34.0	500	38.0
Mean	510.8	39.2	485.8	31.4	494.2	31.5	455.0	30.3
Ratio Shoot wt. / Seed ball wt.	13.0		15.5		15.7		15.0	

The speed of germination data for the clones of monogerm variety SP 5832-0 (Experiment 3) are based upon germinating 40 seeds on a blotter moistened with tap water. The seeds produced at the higher temperature usually germinated more rapidly than those produced at the lower. Alter four days, 53.8% of the seeds produced at the higher temperature had germinated, but only 41.1% of those produced at the lower.

Analysis of the effect of temperature on the junction of male and female gametes in seed yield (Experiment 2): Although the female plants (self-sterile clone) were paired by seedstalk development when the experimental temperatures were initiated, a few plants failed to grow as large as the others. As a result, the fresh weights of the shoots and yields of seedballs in relation to the weight of shoots were not consistent (Table 13). The tendency of the plants in the ? L X ' L treatment to weigh more and yield more seedballs suggested that an analysis of covariance should be used. The analysis revealed no statistically significant differences between treatments. However, the regression between fresh weight of shoot and weight of seedballs was significant at the 1% level.

Since the weights of seedballs per plant were not significantly different, samples of the seedballs were examined to determine the proportion of ovarian cavities containing developed seeds for each of the four treatments. Two random samples of 100 seedballs from each plant were counted and then examined for both the total number of ovarian cavities and the number of cavities containing a developed seed. The percentage of cavities containing a developed seed would indicate the effect of temperature on the functioning of the male and female gametes in seed production. Differences would be valid if the assumption that pollination was accomplished uniformly is valid. Analysis of the percentages of cavities containing a developed seed (Table 14) reveals that temperature significantly affected these percentages. The high temperature during pollen development adversely affected (significant at approximately the 10% level) its subsequent performance on the female at low or high temperature. While the data reveal a highly significant effect of temperature on the female, this cannot be construed solely as an effect on the female, because after pollination the male gamete was at the same temperature as the female. Although the more adverse effect of high temperature could only be observed after pollination, the experimental design did not eliminate the possibility of high temperature injuring the female before pollination.

Table 14.—Effect of low (L) and high (H) temperatures at which the gametes developed on the percentage of ovarian cavities of the self-sterile female clone of sugar beet containing a developed seed, Experiment 2.

Plant repl. number	Percentage of ovarian cavities containing a seed for temperature treatments indicated							
	♀ L × ♂ L		♀ L × ♂ H		♀ H × ♂ L		♀ H × ♂ H	
Sample	1	2	1	2	1	2	1	2
1	82.2	88.0	86.2	84.5	87.0	82.9	74.7	74.8
2	87.3	93.0	80.2	79.7	85.4	71.1	68.2	62.2
3	82.5	87.0	76.5	76.2	80.2	74.1	79.2	72.4
4	87.4	84.2	88.9	91.4	78.0	73.2	81.9	69.4
5	94.2	89.4	82.8	81.1	81.6	75.1	81.5	75.6
6	90.2	85.1	87.4	87.6	81.0	76.2	86.7	79.5
Sums:								
Sample	523.8	526.7	502.0	500.5	493.2	452.6	472.2	433.9
Treatment	1,050.5		1,002.5		945.8		906.1	
Means:								
Treatment*	87.5		83.5		78.8		75.5	

Sums:
♀ L = 2,053.0. ♀ H = 1,851.9, ♂ L = 1,996.3, ♂ H = 1,098.6
♀ L × ♂ L and ♀ H × ♂ H = 1,956.6, ♀ L × ♂ H and ♀ H × ♂ L = 1,948.3

Analysis of variance:

	Sums squares	Degrees freedom	Mean squares		Significance level
Treatment	1,004.20	5	334.73	8.59	1%
♂ L versus ♂ H	160.24	1	160.24	4.11	10%
♀ L versus ♀ H	842.53	1	842.53	21.61	1%
Like versus unlike	1.44	1	1.44		NS
Error	701.81	18	38.99		1% NS

* LSD 10% = 3.6%, 5% = 4.4%, 1% = 6.0%

Relation of length of the style to seed yield at high temperature: While the data of Experiment 2 indicated that the most adverse effect of high temperature on seed set occurred on the female plant, the marked difference in the sensitivity of clones to high temperature, particularly in Experiment 3, suggested that the degree of sensitivity to high temperature may actually reside in the female plant. Among a number of possible causes for fewer seeds developing at high temperature, the length of the style might influence the time-lapse between pollination and fertilization of the different plants. If the style were longer, fertilization might be delayed. The greater respiration at the higher temperature might deplete the limited energy supply available to the sperm so that less fertilization could be accomplished at the higher temperature.

An index of efficiency of seed production for each clone in Experiment 3 was calculated by dividing the number of grams of fresh weight of shoot required to produce a gram of seed

(dried fruits) at the low temperature into the number of grams of fresh weight of shoot required to produce a gram of seed at the high temperature. In Experiment 4, 20 styles per clone were measured for length. The average length of the styles for a clone was correlated with the index of efficiency of seed production in Experiment 3. Because four clones bloomed later than the others and environment might have influenced stylar length, only the 16 clones that bloomed simultaneously were used in the correlation. The correlation coefficient between stylar length and efficiency of seed production was —0.1109 (0.4793 required for the 5% level of significance). A highly significant t-test was obtained for differences in stylar length between a number of the clones. Therefore, stylar length does not seem to be related to the marked reduction in seed yield at the higher temperature.

To observe the degree of self-fertility, we placed a bag on a branch of each clone before anthesis. We estimated the quantity of seeds set to the total number of flowers isolated in the bag, and ranked the clones by percentage of self-fertility. The plants which exhibited the greatest self-fertility also generally exhibited the least reduction in seed yield at the higher temperature.

Discussion

The foregoing data indicate rather clearly the multiplicity of effects of temperature in the production of sugar beet seed. Furthermore, the desirable effects apparently cannot be achieved under any one temperature regime. Because temperature in a given geographic location is essentially uncontrollable, one must choose the location that provides the most favorable temperature.

The need for adequate photothermal induction for seed production is obvious. Then from the standpoint of maximum seed production, a low temperature (mean approximating 65 F) would appear most desirable. In contrast, the seed produced at this temperature does not germinate as rapidly as seed grown at a temperature sufficiently higrher (mean approximating 75 F) to reduce seed yield. Under field conditions, a mean temperature between those used in this study might be most suitable. If seed could be produced where the lower mean temperature could be maintained through most of the grand period of flowering, the yield should not be affected adversely by subsequent high temperatures. Higher temperatures definitely hasten maturity of the seed.

Initially, a study of the effect of temperature was undertaken to test the hypothesis that seed produced at higher temperatures

might germinate faster for the following reasons: 1. The higher temperature would increase respiration and thus might reduce the quantity of reserve materials in the plant and indirectly might reduce the quantity of inhibitory substances in the fruit. 2. The more rapid maturation at the higher temperature would shorten the time in which cementing-substances would be deposited between the tissues of the fruit and the seedcap. Also the quantity of reserve materials available to form cementing-substances may be reduced by the increased respiration. Although seme exceptions occur, the data lend considerable support to this hypothesis, both as indicated by speed of germination and looseness of seedcaps. The precise effects are unknown, however.

Although the data clearly demonstrate the marked reduction in yield of seed at the higher temperature, the exact mode of action is not clear. The following facts are known: 1. The adverse effect occurs during anthesis. 2. Pollen developed and matured at the higher temperature is only slightly less effective in producing seed than pollen matured at the lower temperature. 3. The major portion of the reduction in yield seems to occur immediately after pollination. 4. The length of the style is not correlated with the relative reduction in yield at the higher temperature. 5. The more self-fertile plants appear to be less susceptible to the adverse effects of the higher temperature than those which exhibit less self-fertility. 6. Some of the clones (Experiment 3) yielded only slightly less seed per gram of shoot at the higher temperature while others yielded markedly less than at the lower temperature.

From the foregoing statements, one can deduce that the cause for the reduced yield of seed at the higher temperature resides in the plant on which the seed is produced. Whether the effect of temperature is directly on the female function or indirectly on the male cannot be determined without further investigation.

In at least two instances[4], reduced yields of sugar beet seed in the southwestern United States seemed to be correlated with abnormally high temperatures during anthesis. The high temperatures in the present greenhouse studies were similar to the high temperatures observed in the seed-producing areas.

Seed yields of the multigerm variety US 401 and the monogerm variety SP 5832-0 were similarly affected by high temperature. Temperature during fruit maturation also affected the fruit characteristics similarly.

[4] Communications from Hillas Cole, Farrar-Loomis Seed Company, Heiret, California and H. E. Brewbaker, formerly with The Great Western Sugar Company, Longmont. Colorado.

Breeding programs carried out in the greenhouse may be accelerated by using higher temperatures during the latter part of anthesis and subsequently. The higher temperature during maturation of the seed also may serve as a useful tool to locate progenies with a tendency to produce too many loose seedcaps.

Summary

Clones of sugar beet varieties US 401 and SP 5832-0 were grown in the greenhouse in a relatively uniform environment until just before anthesis. At this time, the plants within clones were paired for uniformity and placed at mean temperatures approximating 65 and 75 F. During anthesis, the plants at the higher temperature were subjected to daily maximums from the middle 80's to the middle 90's on most days. Daily maximums for those at the lower temperature were below 80 and rarely exceeded 75.

The yield of seed at the higher temperature was reduced to about half of that produced at the lower. Some clones were more sensitive to the higher temperature than others. Plants producing seed at the lower temperature had greater fresh weights of shoot; greater weight, size, and number of fruits; and usually a smaller percentage of fruits that had shed seedcaps. The seeds in fruits that matured at the higher temperature germinated more rapidly (usually statistically significant) than those in fruits matured at the lower temperature.

Although pollen matured at the lower temperature was slightly more effective in fertilization than that matured at the higher, the most adverse effect of high temperature occurred after pollination. Length of the style was not correlated with yield of seed at the higher temperature.

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Postemergence Weed Control in Sugar Beets Under California Conditions

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Annual weeds, particularly barnyardgrass *(Echinochloa crus-galli* (L..) Beauv.) and junglerice *(Echinochloa colonum* (L.) Link.) are serious problems in the production of sugar beets in California. The problem continues after the sugar beets have been thinned and the sugar beet foliage has developed to the point that cultivation can no longer be practiced without severe damage to the sugar beet tops. This problem usually occurs from May to October in Central California and from September to March in Southern California.

Preemergence or preplant herbicides have met with considerable success in recent years, but those presently used may last only until the sugar beets are thinned; from this time on weeds must be controlled by cultivation or hand weeding (2)[5]. A number of herbicides has been investigated for use in controlling these late germinating weeds in established sugar beets. Dalapon (2,2-dichloropropionic acid) has been the most successful and most widely used (1, 3). It has been reported to kill annual grasses in sugar beet fields selectively. Although sugar beets do appear to be tolerant to dalapon under some California conditions, yield reductions generally occur when dalapon is applied directly to the foliage of the sugar beet. Some of the injury to sugar beets resulting from applications of dalapon can be avoided by using directed or shielded sprays (5).

Because of the injury generally resulting from applications of dalapon to sugar beets, even from shielded or directed sprays, trials were conducted to find a herbicide that would control these annual weeds when applied postemergence to both the weeds and the sugar beets.

Methods and Materials

Several experiments were conducted to compare the effect of four herbicides on sugar beets and annual weed control over a wide range of environmental conditions in California. A pre-

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liminary screening trial was established to evaluate several herbicides for selective postemergence use in sugar beets. The cliemicals were applied as broadcast topical sprays to sugar beets growing in loamy soil. Application was made with a logarithmic dilution sprayer as indicated: sodium salt of dalapon 20 lb/acre to $1^{1}/_{4}$ lb/acre; disodium salt of endothal (3,6-endoxohexahydrophtalic acid) 24 lb/acre to $1^{1}/_{4}$ lb/acre; reciprocal combinations of dalapon and endothal (dalapon constant at 3 lb/acre with endothal decreasing from 20 lb/acre to $1^{1}/_{4}$ lb/acre: endothal constant at 5 lb/acre with dalapon decreasing from 12 lb/acre to $^{3}/_{4}$ lb/acre); barban (4-chloro-2-butynyl N-(3-chlorophenyl) carbamate) 4 lb/acre to 14 lb/acre; and DPA (3,4-dichloropropionanilide) 12 lb/acre to $^{3}/_{4}$ lb/acre.

Some of these herbicides were further tested in small hand plots set up in a randomized block design; three one acre plots and two one-half acre plots were established using a commercial sprayer with shielded nozzles and a leaf-lifter. The materials used were diuron (3-(3,4-dichlorophenyl)-l,l-dimethylurea) at 2, 3. and 4 lb/acre in the small hand plots and 1.6, 2.4. and 3.2 lb/acre in the large one acre plots; IPC (isopropyl N-phenylcarbamate) at 3 and 6 lb/acre; dalapon (sodium salt) at 4 and 8 lb/acre; endothal (disodium salt) at 3 and 6 lb/acre: and a combination of endothal plus dalapon at 3 and 4 lb/acre, respectively. Applications made with the small hand sprayers included both directed and broadcast topical applications while the large commercially applied plots were all directed spray applications. All rates of herbicides are expressed as pounds active ingredient per acre.

Most of the applications were made in 50 gallons of water per acre when the sugar beets were approximately 12 inches tall. Weed growth varied from emerging plants to 12 inches tall. Soil types were predominantly clay loam with the exception of two trials established on a sandy soil. Care was used in the directed applications to avoid spraying the lower leaves of the sugar beet plants, but some of the older, lower leaves received some spray.

Results

Northern California

The initial hand plots in Northern California were established in an area expected to be severely infested with annual weeds, particularly barn yard grass. The first application was made following a cultivation so the field was free of weeds. This application consisted of diuron and IPC. Following the application, the trial was thoroughly irrigated in order to activate the herbicides.

The beds were subbed completely across until the soil was saturated with moisture. The second application, consisting of endothal, dalapon, and the combination of endothal plus dalapon, was made when the predominant weed, barnyardgrass, had formed its secondary root system. No additional weed control treatments were given the plot area.

Treatments with corresponding yields and weed control for the trial conducted in Northern California are shown in Table 1. The yields and weed control for the plots treated with broadcast topical sprays are not reported here because cf the severe injury that resulted from some treatments and the virtual failure of weed control with others. IPC was the only herbicide that did not cause visual stunting of the sugar beers. The combination of endothal plus dalapon showed only minor stunting at harvest time.

Table I.—Sugar beet yields and percent weed control from directed postemergence herbicide applications in Northern California.

Herbicide	Lb/acre active ingredient	Roots tons/acre	%, Weed control[1] Broadleaved	Grass
Diuron	2	23	90	90
	4	**17**	99	99
IPC	3	**16**	**0**	40
	6	22	**10**	80
Dalapon	4	18	**0**	90
	8	15	20	95
Endothal	3	**18**	60	0
	6	**19**	95	**10**
Endothal + Dalapon	3 + 4	21	95	90
Check	0	23	0	0
LSD P = .05		4		

[1] Percent weed control was based on a visual estimate with 0% indicating approximately 7 to 9 broadleaved weeds per square foot and 20 to 23 grass weeds per square foot.

Southern California (Hand Plots)

The plots in Southern California reported in Table 2 consisted of diuron applied by hand as a directed spray. The area selected was weed free at the time of application, but was infested with annual weed seeds, primarily canarygrass *(Phalaris canariensis* L.), silversheath knotweed *(Polygonum argyrocoleon* Steud.), sour clover *(Melilotus indica (1..)* All.), spiny sow thistle *(Sonchus asper* (I,.) Hill), wild mustard *(Brassica arvensis* **(L.)** B.S.P.), and nettleleaf goosefoot *(Chenopodium murale L.*).

There were no typical diuron symptoms on the sugar beets at 2 lb/acre of diuron, regardless of soil type. However, there was some leaf burn on the sugar beets receiving 4 lb/acre of diuron on sandy soil. This leaf burn was not typical of diuron

Table 2.—Sugar beet yields and percent weed control from hand applied direct postemergence applications of diuron in Southern California.

Trial	Lb/acre active ingredient	Beets/100 ft row	Roots tens/acre	% weed[1] control[1] broadleaved	Grass
Experiment #1	1.6	91	19.0	98	99
	3.2	104	21.2	100	100
	0	91	20.9	0	0
Experiment #2	1.6	188	26.7	75	90
	3.2	176	25.0	90	98
	0	195	28.0	0	0
LSD P = 0.5			NS		

[1] Percent weed control was based on a visual estimate with 0% indicating approximately 10 to 12 weeds per square foot in Experiment #1: 12 to 15 weeds per square foot in Experiment #2.

Table 3.—Sugar beet yields and percent weed contrc?! from field scale plots under Southern California conditions. Directed lay-by applications of diuron were made using commercial equipment.

Trial	Lb/acre active ingredient	Roots tons/acre			
Location #1	1.6	16.9	14.3	18.4	90
Applied 1-17	3.2	13.-1	12.9	34.7	98
Harvested 4-20 & 21	0	16.4	14.1	4(5.4	0
	0	16.6	14.4	48.0	0
Location #2	1.6	13.6	14.0	38.1	80
Applied 1-9	3.2	11.8	11.8	27.9	95
Harvested 4-28 & 30	0	13.7	13.2	36.2	0
	0	12.5	12.5	31.2	0
Location #3	1.6	25.9	14.7	76.2	95
Applied 1-23	2.4	23.0	15.1	69.5	98
Harvested 4-20	0	24.0	14.7	70.5	0
	0	23.8	15.3	72.9	0
LSD P = .05		2.8			

[1] Percent weed control was based on a visual estimate with 0% indicating approximately 3 to 4 weeds per square foot in Location #1; 10 to 12 weeds per square foot in Location #2; over 50 weeds per square foot in Location #3.

injury on other crops. It appeared to be more like a burn or necrosis resulting from drought or a soil condition of excess salt.

Southern California (1/2 and 1 Acre Plots)

Additional trials were established to determine if the herbicides could be applied with commercial equipment with satisfactory results. The results of three of these trials using diuron are reported in Table 3. The other two trials using IPC, endothal, and dalapon were not harvested for yield data. However, IPC did show promise, particularly for the control of canarygrass, with no visual injury to the sugar beets.

The beets were dug with a commercial digger and weighed by truck load. Weed control was satisfactory at all rates. Obvious

injury to the sugar beets occurred only at the 3.2 lb/acre rate of diuron on sandy soil (location #1). However, yield data in Table 3 indicate that some injury at the higher rates may have occurred that was not apparent visually.

Discussion

Postemergence weed control has long been recognized as a desirable practice; but to date, no herbicide has all the necessary requirements. Dalapon may be used as a postemergence treatment for the control of annual grasses in some areas. The rate used will depend on the species, stage of growth, environmental conditions, etc. In California, dalapon is suggested for use only as an emergency measure for heavy grass infestations, as it will usually cause temporary stunting of the sugar beet plants. Unsatisfactory results have been experienced in the desert valleys of Southern California.

When properly applied, approximately 4 lb acre of dalapon are required to control barnyardgrass. The barnyardgrass should not be sprayed until the seedlings produce secondary roots and are growing vigorously. It frequently takes from ten days to two weeks after seedlings first appear before secondary roots develop. Treatment before this time is usually not effective. Treatment after the watergrass has reached the boot stage is likewise not effective. These plants generally are not killed and will produce viable seed. If volunteer barley or wild oats are a problem, higher rates will be required to obtain satisfactory control. These high rates are more likely to injure the sugar beets.

To minimize injury, directed sprays should be used whenever possible, especially if applications are made during periods of high temperature, or when higher rates per acre are applied. If the temperatures are high during application, use directed sprays only, as dalapon is less selective under these conditions and will cause stunting and yellowing of the sugar beet plants.

The combination of endothal plus dalapon provided satisfactory weed control but appeared to cause similar injury symptoms on the sugar beet plants to that caused by dalapon alone, although not as severe. However, the weeds following application showed typical endothal effects except at the low rates of endothal where grass kill was more complete than would be expected from endothal alone.

Injury to the sugar beet plants was questionable with the endothal treatment. While it did not stunt the sugar beets, it did burn some of the foliage. This burning appeared to be the

most severe when application was made to young sugar beet plants during high temperature conditions. Broadleaved weed control was satisfactory, but endothal did not control the grasses.

Further testing of barban and DPA was discontinued because DPA caused mild contact burn to both the sugar beet plants and the weeds at rates of 6 to 12 lb/acre but did not control the weeds. Barban controlled only the oat species but did not visually injure the sugar beets.

IPC showed some promise as a lay-by treatment when applied to weed free sugar beets. It was necessary to activate the herbicide by thorough irrigation. The primary disadvantage of IPC is its relatively short soil residual life (4). By the end of the trial, weeds had started to invade these plots.

Good weed control and no injury to the sugar beets resulted when using 2 lb/acre of diuron. Rates of 3.2 and 4 lb/acre of diuron caused some visual injury to sugar beets on sandy soil. This injury was not evident on heavier soils; however, yield data might indicate a slight reduction at the higher rates. Diuron should be applied as a directed spray to weed free beds and followed by a thorough irrigation. If applied as a broadcast topical spray, serious injury to the sugar beets will result.

A factor to consider in the use of diuron as a lay-by treatment in sugar beets is the long residual life of the herbicide in the soil (6). In one of the trials, sorghum was planted following the sugar beet harvest. Four months had elapsed between treatment and planting. Some stunting of the sorghum seedlings was present in the plots receiving the 1.6 lb/acre rate of diuron. Both stunting and stand reduction were evident in the plots receiving 2.4 lb/acre of diuron, but injury at maturity was not apparent.

Summary and Conclusions

Several trials were conducted to control late season weeds in sugar beets with herbicides applied postemergence to the sugar beets and either pre- or postemergence to the weeds. These results are still experimental and additional work should be governed accordingly.

Dalapon provided some control of grass weeds but caused some stunting and yellowing of the sugar beet plants. This treatment usually causes temporary stunting of the sugar beets and should be used only as a directed spray when possible.

The combination of endothal plus dalapon showed some promise for controlling mixed populations of broadleaved and grass weeds.

Endothal did not control emerged grass weeds satisfactorily but did control the emerged broadleaved weeds.

Diuron showed promise as a lay-by treatment when applied as a directed spray. Rates of 1.6 and 2 lb/acre of diuron gave satisfactory weed control under the conditions of this study without injuring the sugar beets. Consideration should be given to the soil residual life of diuron because of possible injury to succeeding crops.

Acknowledgment

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Sugar-Beet Root Aphid Resistance in Sugar Beet[1]

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The sugar-beet root aphid *(Pemphigus* sp., probably *betae* Doane) has been widely distributed in sugar beet-growing areas in western United States and western Canada for many years. When conditions are favorable for its development, it is capable of causing serious injury to the sugar beet crop (1,2)[3]. Insofar as the writers are aware, the existence of differences among sugar beet strains or varieties, in resistance to this pest, has not been previously reported.

In exploratory studies at Fort Collins, Colorado, in 1961, on control of the sugar-beet root aphid with insecticides, a striking contrast was observed between two sugar beet strains in degree of infestation. Four pairs of phorate-treated and untreated plots occurred in a part of a sugar beet field on the Hospital Farm in which a vigorous, leaf spot-susceptible inbred, SP 471001-0 (Strain A), was growing. A similar set of 4 pairs of plots, occurring in a neighboring area in the same field, contained the leaf spot-resistant commercial variety, GW 674 (Strain B), growing under comparable conditions. On July 27, granular phorate was applied to the center of the foliar rosette of each plant in the plots designated for treatment. The roots of 3 plants in each plot were examined for aphids on August 15, and the results are presented in Table 1. These data show similar differences between strains for both the treated and untreated plots, with strain B averaging only about 2 percent as many aphids per plant as strain A.

In order to study further the question of root aphid resistance, 4 pairs of plots were set up in border areas of the above

Table 1.—Numbers of sugar-beet root aphids per plant, on two sugar beet strains, 19 days after application of phorate granules; results given as 6-plant averages.

Pounds phorate per acre	Strain A (SP 471001-0)	Strain B (GW 674)
0	4.0	0.2
1	5.4	.0
0	10.6	.3
2	1.0	.0

[1] Report of investigations conducted in cooperation with the Colorado Agricultural Experiment Station. Approved by the Experiment Station Director for publication as Scientific Series Article No. 771.

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[3] Numbers in parentheses refer to literature cited.

field where phorate was not applied. The plots were 6 rows wide and 12 feet long, and each pair consisted of contiguous plots of strains A and B. Counts were made of aphids occurring on the roots of 3 plants in each plot on August 21 and September 22, 1961. The plants were dug with approximately 3 inches of soil around the taproot. The soil was carefully removed in the laboratory and the aphids were counted under magnification.

On August 21, aphids were found on all but 1 of the 12 plants of strain A examined, and on only 2 of 12 plants of strain B. On September 22, aphids were found on all 12 plants of strain A and on 6 of strain B. As shown in Table 2, the number of aphids per plant was slightly higher on both strains, at the second count, with a proportionately larger increase on strain B. The average number of aphids per plant, for strain B, was approximately 6 percent of the average for strain A.

Table 2.—Numbers of sugar-beet root aphids per plant on two sugar beet strains; results given as 12-plant averages.

Date	Strain A (SP 471001-0)	Strain B (GW 674)
Aug. 21	8.8	0.3
Sept. 21	9.7	.8

Although the results presented in this report were based on limited observations, the contrasts were sufficiently striking to justify the conclusion that the 2 strains differ substantially in root aphid resistance. It is not known whether the type of resistance carried by strain B actually inhibits root aphid development under commercial field conditions. The observations made in this study showed that the aphids were attracted to strain B in small numbers and were able to multiply on it. If the strain contrasts observed were merely the results of aphid preference, it is conceivable that, in commercial fields where preferred varieties are not available, the resistance of strain B would be of little, if any, practical value.

Because of the preliminary nature of this study, it would not be safe to conclude, on the basis of these results, that breeding for resistance to the sugar-beet root aphid is a potentially valuable tool. However, in view of the importance of that pest in sugar beet production, and since sugar beet strains apparently differ in resistance, investigation of the nature and practical value of such resistance appears to be highly desirable.

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Variety Crosses in Sugar Beets (Beta vulgaris L.) I. Expression of Heterosis and Combining Ability

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Regardless of the crop, the plant breeder must become thoroughly acquainted with his breeding material. Information pertaining to heritability of yield factors, genetic diversity and variability, type of genetic variance and gain expected from selection is a prerequisite before the breeder selects his basic breeding material and designs his program.

Recently there has been an increased interest in heterosis resulting from population crosses in both corn (5, 9, 7)[2] and Drosophila (1, 13, 14). Diallel crosses among six southern corn varieties (9) showed the F_1's to yield on the average 19.9% more than the midparent values and 11.5% more than the high parent. Intercrosses among a group of ten corn belt varieties (5) on the average yielded 8.5% more when compared with the midparent values. This increase in average yield of the F_1's indicates that corn varieties differ in their underlying genetic constitution. If the varieties had been at equilibrium between mutation and selection and the gene frequencies affecting yield similar, no increase in the F_1's would have been evident.

Moll et al. (7) hybridized corn varieties originating from three distinct regions; the southeastern United States, the mid-western United States and Puerto Rico. There were two varieties from each region thus giving the opportunity to compare crosses of varieties from the same region and from different regions. They found that the average of the within-region crosses was 104% of the midparental values, compared with the average of 124% for the between-region crosses. The results of these findings indicate that heterosis, expressed as percent of the midparent, increases with increased genetic diversity.

Variety crosses in sugar beets (2) indicate that specific combining ability exists between varieties. The intercrossing of open-pollinated mother lines selected from a parent variety showed an appreciable amount of hybrid vigor (3). These displays of heterosis would also indicate a genetic difference between varieties and lines selected from a single parental variety.

A program of reciprocal recurrent selection using populations which have a high frequency of many favorable genes and which

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[2] Numbers in parentheses refer to literature cited.

exhibit substantial heterosis when crossed should result in the concentration of yield factors. The probability of selecting superior inbred lines from the derived populations should be much greater than random line selection from varieties. The hybrids derived from these lines should exhibit maximum combining ability as a reciprocal recurrent selection program is designed to capitalize on both additive and nonadditive gene action.

This present study was planned to evaluate genetically five diverse monogerm sugar beet varieties as a guide in selecting stocks for a reciprocal recurrent selection program.

Materials and Methods

Five self-sterile monogerm varieties believed to represent a diverse range of types were included in this study. A brief description of these varieties seems desirable since any implication concerning their cross-performance could reflect on their genetic origin. Two of the varieties, 58-412 and 57-807, are American #2 and American #3N types, respectively, and are believed to possess a reasonable amount of genetic variation. Two USDA varieties, SP 5832-0 produced at Beltsville, Maryland, and SEC 24 produced at Salt Lake City, Utah, were included for their leaf-spot and curly-top tolerance. Variety SP 5832-0 was produced by intercrossing eight monogerm progenies which were equal to US 401 in yield and leaf-spot resistance, while SLC 24 was the F_3 monogerm selection resulting from crosses of curly-top-tolerant monogerm lines to the leaf-spot-tolerant multigerm variety US 201. Both of these varieties probably represent a fairly narrow range of genetic variation. The variety, 58-411, is a broad base extraction from a cross of SEC 15 with seven different pollinators and was included in this study to represent the extreme in genetic variation.

Based on sucrose and weight, 45 of the best mother beets were selected from each of the 5 varieties by the unit block method of mass selection (8). These roots were halved thus making it possible to divide each variety into 2 genetically identical populations making a total of 10 groups. One group of roots from each variety was designated as the male population while the remaining group was termed the female population.

The male populations were individually isolated to serve as a pollen source and as a population advance. Each female population was randomly divided into 4 groups of 10 beets each. The 5 extra beets were used as substitutes in cases of bolting failure. Each group of ten beets was preassigned a specific pollinator.

The inflorescence of each female beet prior to flowering was completely covered with a large manila bag which had a plastic window to observe floral development. Pollen was collected from each male population using a pollen collector powered by a small vacuum sweeper. The pollen was blown into the assigned manila bags through a small hole. The procedure was repeated each day until pollen sheading was complete. This system allowed the production of reciprocal crosses and permitted a twenty-beet sample plus pollen from the 40 male beets to represent the cross population. By this method the diallel series including reciprocals was produced.

The seed from the 10 female plants was bulk harvested. Reciprocals were kept separate and later included as alternate replications for each F, entry in the yield trial.

The field design of the experiment was a 40 replication randomized complete block which included the five varietal populations, their ten diallel crosses and 54-406, a multigerm American #2 type commercial check. Two single cresses were also included but the discussion of their implications is given in another paper (4).

In planting the different populations, the rows were spaced 22 inches apart with 20 inches between hills within the row. Three to 5 seeds were hand planted in the 15 hills of the single row plots and later thinned to 1 beet per hill. At harvest all the beets were selected in order down the row and anv beet showing visual evidence of disease was discarded. The individual weight and sucrose percent were determined and the first 8 beets were included in the data. Thus, 320 beets for each entry-were individually analyzed. The method is similar to that described by Powers (8).

The data included in this article are calculated on a plot mean basis with each plot representing the mean of eight beets.

Experimental Results

A breakdown of the variances associated with varieties and crosses was not particularly enlightening, since these variances were highly significant statistically for both weight and sucrose percent. A reaction of this type would be expected due to the diversity of the basic varieties used in this study. However, most of the significance may be attributed to the higher-than-average root and sugar yield of the variety 58-412. This locally adapted selection did extremely well when compared with the other entries, both as a parental variety and when intercrossed with the other varieties.

Reciprocal Crosses

The mating system by which the intercrosses were produced afforded the chance to test reciprocal crosses. These were included in the experimental design by pairing the cross and its reciprocal, then randomizing the pair. This plan allowed the total of 20 paired comparisons or 20 replications of each of the two crossing types. This system was projected to include the 10 intercrosses reported in this study.

The single degree of freedom analysis for differences between reciprocal crosses indicated that a real difference existed due to the mating system between certain crosses. This unexpected reaction had to be explained before the data could be objectively analyzed.

Table 1 presents the mean yield for the six intercrosses that displayed statistical differences between reciprocals for either weight per root or sucrose percent. These differences between reciprocals are difficult to explain by chance due to the magnitude of the mean squares and the number of crosses showing differences. A cytoplasmic genotype interaction could be a possible explanation, however the acceptance of this theory is not universal nor satisfactory. Self-fertility in one or more of the varieties is probably a more acceptable explanation for differences between reciprocal crosses. An examination of the frequency distributions and means of the populations exhibiting reciprocal differences left no doubt that there were two populations within each cross fluctuating around their common mean.

The yields of the crosses exhibiting reciprocal differences are shown in Table 1. The mean yield for both root weight and sucrose percent for variety SP 5832-0 was lower in all crosses where this variety was used as the female. A reaction of this type would indicate that the seed harvested from these female plants was predominantly self-fertilized instead of the desired varietal hybrid seed. Thus, the differences between reciprocals involving SP 5832-0 can be explained by assuming that the variety is self-fertile.

Table 1.—Intercrosses displaying statistical differences between reciprocals.

CROSSES		Weight			Sucrose		
Female	Male	Column 1 Female		Column 2 Female	Column 1 Female		Column2 Female
SP 5832-0	57-807	2.13	*	2.55	10.80	*	11.58
SP 5832-0	58-412	1.70	• •	3.74	9.59	**	12.80
SP 5832-0	SLC #24	2.29		2.53	10.01	**	11.15
SP 5832-0	58-411	2.51		2.63	10.35	**	11.36
58-411	58-412	2.40	**	3.87	11.49	**	13.02
SLC 24	58-411	2.59	**	3.20	11.28		11.17

The differences observed between the reciprocals of the cross, 58-411 X 58-412, can be explained with similar reasoning;. The quantity of pollen collected from variety 58-412 was sharply reduced due: to a high percent of male steriles in the population. Thus, when 58-411 was bagged as the female of the cross and subjected to limited pollen, this variety tended to be self-fertile.

Additional evidence which further substantiates the limited pollen and self-fertility theory is the reciprocal cross data from the SP 5832-0 X 58-412 hybrid. If 58-412 pollen was limited, we would expect the seed harvested from the 5832-0 plants to be mostly self-pollinated, thus a reduction in yield would be expected in the cross where SP 5832-0 was used as the female in combination with 58-412. This theory was verified from the results obtained from the reciprocal data of the SP 5832-0 X 58-412 cross.

The cross, SLC 24 X 58-411, displayed differences between reciprocal*i* for weight of root. There is no apparent logical explanation for this response but it was assumed, based on the previous experience, that self-fertility was responsible for this reaction.

Due tc• these reciprocal differences and the evidence for self-fertility in certain crosses, the reciprocal cross displaying the lowest mean yield was deleted from the data. Tin's assumes that the higher yielding reciprocal is composed primarily of the desirable hybrid seed. This deletion reduced the populations of certain crosses from 320 to 160 beets.

General and Specific Combining Ability

The statistical model used to evaluate diallel crosses in corn (6, 10, 11) can also be applied to the variety crosses observed in this study. Tables 2 and 3 show the mean yield of the parents and intercrosses together with the estimates of general and specific

Table 2.—Mean yields (weight per beet) of the parental varieties (parentheses) and their Fi intercross combinations together with their general and specific combining ability.

Parents	57-807	SP 5832-0	58-412	SLC 24	58-411	F_1 mean	^	^
57-807	(2^30)	2.55*	3^18	2Ti	2JH	2^60	.2232	.0012
SP 5832-0		(2 33)	3 74*	2.53*	2.63*	2.86	.0139	.0593
58-412			(3.33)	3.16	3.87*	3.49	.5108	.0263
SLC 24				(2.52)	3.20*	2.75	.0694	.0626
58-411					(2.71)	3.06	.0187	.0839
Means						2.95	.1672	.0467

LSD (0.05) = .34
LSD (0.01) = .26
•Mean of 160 Beets

Table 3.—Mean yields (percent sucrose per beet) of the parental varieties (parentheses) and their F_1 intercross combinations together with their general and specific combining ability.

Parents	37-807	SP 5832-0	58-412	SLC 24	58-411	F_1 mean	$\sigma_G 2$	$\sigma_S 2$
57-807	(11.40)	11.58*	12.80	10.93	11.06	11.59	.0939	.0079
SP 5832-0		(10.21)	12.80*	11.15*	11.36*	11.72	.0125	.0005
58-412			(13.23)	12.46	13.02*	12.77	1.5534	-.0041
SLC 24				**(11.21)**	11.17*	11.43	.2826	—.0160
58-411					(11.29)	11.65	.4891	.0091
Means						11.83	.1863	-.0005

LSD (0.05) = .52
LSD (0.01) = .68
•Mean of 160 Beets

combining ability for the five varieties used in this study. These combining ability estimates are relative and dependent upon the particular group of varieties involved in the hybrids under test. The average performance of the varieties in crosses provides a measure of their general combining ability or the additive effects. Deviations of a particular cross from expectations based on the average of its two parents provides a measure of specific combining ability or the nonadditive effects. Low values for $\sigma_G 2$ indicates that the particular variety in question is average in its general combining ability while laree values of $\sigma_G 2$ mav indicate that the particular variety is either much better or much poorer than the remaining varieties with which it is compared. Low values of $\sigma_G 2$ indicate that the hybrids involving: the particular variety are responding; as expected based on their general combining ability. High values of $\sigma_G 2$ indicate that some combinations did relatively better or poorer than expected.

The greneral and specific combining; ability means for the varieties indicate that the additive effects are much more important than the nonadditive effects for both weight of root and sucrose percent. This response is to be expected considering the heterogeneous nature of these varieties. However, there are some nonadditive effects indicated bv the high specific combining ability values for certain crosses. Both effects must be considered in planning a breeding program utilizing these open-pollinated varieties.

Heterotic Responses

The parental and F_1 means together with F_1 yields expressed as percentages relative to the midparent, high parent and constant parent are presented in Tables 4 and 5. The averagre relative root yield of the F_1 intercrosses compared to the respective midparent yield was **111.7%.** When the F_1's are compared with the higher **parent** of each cross and the constant parent, the

average relative yield was 102.4% and 112.7%, respectively (Table 4).

The average relative sucrose yields for the midparent, high parent and constant parent were 103.2%, 97.9% and 103.6%, respectively (Table 5).

These results indicate the presence of considerable genetic diversity between certain varieties for root weight but little diversity for percent sucrose.

Table 4.—Weight per root of parent varieties and their F_1 means as determined in all possible cross combinations together with the F_1 means expressed as a percent of the mid-parent, higher parent and constant parent yields.

Population	Parental means	Fi means	Mean F_1 yields, % relative to Mid-parent	Higher parent	Common parent
57-807	2.30	2.60	103.6	95.6	113.0
SP 5832-0	2.33	2.86	113.5	105.1	122.7
58-412	3.33	3.49	120.3	104.8	104.8
SLC 21	2.52	2.75	106.2	99.3	109.1
58-411	2.71	3.06	114.8	107.0	113.9
Means	2.64	3.00	111.7	102.4	112.7
54-406	3.00				

Table 5.—Percent sucrose of parent varieties and their mean F_1 as determined in all possible cross combinations together with the F_1 means expressed as a percent of mid-parent, higher parent and constant parent yields.

Population	Parental means	Ft means	Mean F_1 yields, % relative to Mid-parent	Higher parent	Common parent
57-807	11.40	11.59	101.3	97.7	101.7
SP 5832-0	10.21	11.72	106.5	99.5	114.8
58-412	13.23	12.77	105.3	965	96.5
SLC 24	11.21	11.43	100.5	97.0	102.0
58-411	11.29	11.65	102.2	98.7	103.2

Variety Evaluation

Based on the information obtained, a brief description of the relative merits of each variety will perhaps aid in the interpretation of the data. Due to the nature of the experiment, comparison and estimates of combining ability and heterotic responses are relative only to the varieties and hybrids under test.

The variety, 57-807 is an American #3N type and is a monogerm extraction without selection from the commercial multigerm American #3N. Based on the F_1 performance when compared to the average midparent value and the common parent, some genetic diversity and heterosis is evident for weight of root.

The general combining ability of this variety is about average for percent sucrose but below average when compared with the other varieties for weight of root. The specific combining ability values for both yield factors are low indicating that the particular variety is responding as expected based on its general combining ability. Some heterosis is evident for root weight and sucrose percent when 57-807 is crossed with 58-412. This yield increase could also be due to exceptional yield of the variety 58-412 which performed above average in all crosses where it was included as a parent.

The reciprocal cross data indicated that the USDA variety SP 5832-0 was self-fertile when subjected to conditions of limited pollen. The sugar percent for this variety is 10.21% which could indicate inbreeding in the population or a variety that is inherently low in sucrose. The root weight of 2.33 pounds per beet is close to the average of the five populations and would not indicate a substantial amount of inbreeding. When the F_1's are compared with the common parent the increase in yields were 22.7% and 14.8% for root weight and sucrose percent, respectively. These results were exceptional and would indicate the possible existence of inbreeding in the parental population.

The sreneral and soecific combining ability estimates indicate that variety SP 5832-0 is an average combiner with some specific combining: ability with 58-412 for weisrht of root. The root yield of the F/s was 5.1% better than the average of the hisrh parent when SP 5832-0 was one of the narents in the varietal hybrid. A 6.5%) increase in sucrose was displayed when the F_1's were compared with the average midparent values.

The data indicate that variety 58-412 is the best variety included in the experiment. The high parental yields of this variety when compared to the other varieties mav contribute to the exceptional heterotic response of this population. The high variances for sreneral combining ability for both sucrose and weight are indications of the excellent combining ability of this variety.

It should be pointed out that 58-412 was the only locally adapted variety in the test. This adaptation may have contributed to the exceptional yields of the variety and its intercrosses.

The performance of SLC 24 is averasre for sreneral and specific combining ability when compared with the other varieties. Slight heterotic responses are evident for weisrht of root when the F_1's were compared with the midparent and common parent. A *2%* increase in sucrose percent was indicated when the hybrids involving this cross were compared with this variety. The below

average yield for both root weight and sucrose percent coupled with the low heterotic responses do not make this variety particularly attractive for breeding purposes.

Variety 58-411 produced the largest average heterotic response for root yield when the mean of the F_1's was compared with the higher parent. The broad genetic base of this variety probably contributed to the average values obtained for general and specific combining ability.

Discussion

The monogerm varieties intercrossed in this study were selected to include diverse types of genetic material originating from American Crystal and USDA sources. The genetic variation within varieties was also considered to include a range of varieties from an eight progeny synthetic to a variety crossed with seven different pollinators. The intercrosses would be expected to display varietal heterosis providing actual genetical differences existed between these varieties. The 11.7% increase in root yield of the F_1 when compared with the midparent would indicate that the varieties are genetically diverse for this character.

The same comparison indicates a 3.2% increase for sucrose percent. The average heterotic response if measured by the mean F_1 yields exceeding the higher parent was 2.4% for root weight, with none exceeding the higher parent for sucrose percent. The data indicate the presence of heterosis in crosses between varieties with specific crosses having considerable yield advantage. The plant breeder developing a hybrid program should expect the higher yields to result from increased root weight rather than from increased sucrose percent. There will no doubt be exceptions to this observation but on the average the data indicate that the increased yield resulting from a hybrid program will result from more tons per acre.

The average additive effects were greater than the non-additive effects which is to be expected due to the heterogeneous nature of the varieties. However, certain varieties possess better-than-average values for these components. A program designed to exploit these effects to the maximum would be expected to give maximum yield. In designing such a program, several breeding methods may be necessary to accomplish the goal.

If the five varieties used in this study represent a random sample of unselected monogerm varieties the following program should result in maximum yield. Since the data indicate that the monogerm varieties lack substantial genetic diversity, measured by the heterotic response when the F_1's are compared

with the higher parent, the first point of consideration should be in the development of several diverse populations of approximately equal yield potential.

These populations could be accumulated by selecting varieties or by combining similar geographical types. Selections from Europe would enhance the possibility of obtaining genetic diversity in the populations. Any number of populations could be selected or developed but the population should be chosen to secure maximum genetic variation and diversity coupled with desirable agronomic traits.

One or two mass selections using the unit block procedure within the populations should serve as an adaptive selection. In order not to seriously reduce the genetic variability a large number of beets should be selected to form the next population.

A diallel series involving these populations should reveal their genetic diversity and combining ability. A recurrent selection program should be initiated in those varieties displaying the greatest amount of additive gene action. The two varieties displaying the greatest amount of general and specific: combining ability for yield factors should be incorporated in a reciprocal recurrent selection program which is designed to capitalize on both additive and nonadditive genetic effects. The selection of inbred lines from these synthetic populations to be incorporated in hybrid combinations should result in maximum yields.

The data indicate that the variety 58-412 could be improved in both root weight and sucrose percent by a recurrent selection program. The selection of two varieties for a reciprocal recurrent selection program is more difficult because 58-412 was the only variety tested which displayed both high-additive and nonadditive effects for root weight and sugar percent. The best prospects would be the use of the varieties 58-412 and 58-411. The F_1 intercross between these two varieties gave the highest yield of all the hybrids.

Doxtator (2) reported that the best F, originating from a variety cross—American #4 X American 1936—gave a 23.1% increase in root yield and a 4.1% increase in sugar percent over the high parent based on the heterotic effect. These multigerm varieties were apparently quite diverse. This display of heterosis is certainly greater than heterosis displayed by the varieties selected for this experiment. Perhaps, selection over the years and the conversion to monogermness has depleted the genetic diversity in our monogerms. These data, if comparable certainly point in that direction. The sugar beet breeder should perhaps

re-evaluate these old varieties, if available, and capitalize on this additional source of genetic diversity.

Summary

Five open-pollinated sugar beet varieties representing a range of diverse types were crossed in a diallel series. The parental varieties together with their ten intercross populations were grown in a yield trial *in* 1959.

The average yields of the parents ranged from 2.3 to 3.3 pounds per beet and from 10.2% to 13.2% sucrose. The F_1 yield ranged from 2.1 to 3.9 pounds for root weight and 10.9% to 13.0% for sucrose. The mean F_1's relative to the midparent was 117.7% for root weight and 103.2%, for sugar percent.

Estimates of general combining ability (additive effect) and specific combining ability (nonadditive effect) were calculated for root weight and percent sucrose. The average additive effects were greater than the nonadditive effects.

Application of the results is discussed and a proposed breeding program outlined.

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Variety Crosses in Sugar Beets (Beta vulgaris L) II. Estimation of Environmental and Genetic Variances for Weight Per Root and Sucrose Percent

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The primary purpose of this variety crossing program was to thoroughly evaluate several monogerm varieties originating from diverse sources. The information obtained would enable the plant breeder to re-evaluate the converted monogerms and serve to guide his decisions when designing a breeding program. The implications of the general and specific combining ability, together with the heterotic responses were discussed in a previous report (3)[2]. The estimations of the environmental and genetic variances for the parental varieties and their intercrosses will be included in this paper.

Materials and Methods

A detailed description of the varieties and the testing technique were included in a previous article (3). In addition to the five parental varieties and their intercrosses previously reported, two single-cross hybrids and a commercial multigerm, American #2 Check were included in the field plots. The single-cross hybrids, (NB_1 X NB_4) were produced by Dr. McFarlane and 58-9061 (52-430 X 52-407) produced by Dr. Powers were used to obtain the estimates of environmental variation. The check variety was included to compare the relative performance of monogerm and multigerm seed. The data included in this article are on an individual plant basis. The methods of statistical analysis are essentially those outlined by Powers (I, 2).

Estimating Environmental Variances

Two single crosses were included in this study to estimate the environmental variances. Table 1 gives the means and within-population variances for root weight and sucrose percent for these nonsegregating populations. The within-population variances are essentially the environmental variances. No problem exists when only one nonsegregating population is included *in* the experiment because its within-population variance is the only estimate of the environmental variance available. When two or more nonsegregating populations are included, the ex-

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[2] Numbers in parentheses refer to literature cited.

perimenter hopes that these estimates will be fairly uniform and that he can use the mean to estimate the environmental variance. An examination of the within-population variances for the non-segregating populations included in this experiment indicated that considerable differences existed between the two populations for root weight, Table 1.

Table 1.—Means and within population variances for root weight and sucrose percent for the nonsegregating populations.

Nonsegregating populations	Root Weight		Sucrose per cent	
	Mean	Within-population variances	Mean	Within-population variances
NB_1 X NB_4	2.38	0.695	10.20	2.177
58-9061	2.80	1.241	12.18	2.019
Regression of means and Variances	67.2%		58.2 %	

Powers (2) reported that a linear relationship exists between population means and the within-plot variances for root weight. He was able to show that the mean weight per root accounted for 99.7% of the sums of squares for variances. Based on these results and the apparent positive association between the means and variances in these data the regression of the within-plot variances on the means was calculated. The 40 replications for each nonsegregating population Avere divided into four groups, i.e., replication 1-10, 11-20, 21-30 and 31-40. The means of each group were used to calculate the regression.

The "t" test indicated that the regression value 0.8188 was significantly different from zero at the one percent level. (D.F. = n—2 = 6.) Thus a true relationship existed between the means and the variances for root weight. The mean weight per root, however, accounted for only 67.2% of the sums of squares for variance, the remaining 32.8% of the variance was due to the interactions. This failure to account for a greater percentage of the variation seriously questioned the advisability of using regression to estimate the environmental variances of the entries in this test.

Several alternative methods for estimating the genetic variances were considered. These included the mean of the two estimates, the lower or higher estimate, either estimate as long as constant and the variance associated with the mean closest to the mean of the entry. Because the "t" test had indicated an association between the means and variances, the within-population variance for the single cross whose mean was closest to

the mean of the variety or intercross mean was used to estimate its genetic variance.

The regression of the means and within-plot variances were calculated for sucrose percent using the method employed for weight of root. The regression value of —.3978 was significantly different from zero at the one percent level. The relationship differed from the regression value for root weight in that it was a negative value. The mean sucrose percent accounted for 58.2% of the sums of squares for variances, the remaining 41.8% was due to the interactions. This also prohibited the use of regression to estimate the environmental variances for sucrose percent.

The within-population variances for sucrose percent of the two single-cross populations differed only by 0.158 and were considered to be estimates of the same effect. The mean of the two environmental estimates, 2.098, was used to estimate the genetic variances associated with the varieties and hybrids for sucrose percent.

Genetic Variances for Root Weight

The genetic variances for root weight of the parental populations and their Fi intercrosses are included in Table 2. Brief descriptions of these varieties together with relative estimates of their genetic variability based on the knowledge of the breeding history have been included in a previous paper (3).

Variety 58-411 was considered to have a broad genetic base when compared to the other varieties under test. The estimate of the within-population genetic variance of 1.452 was the highest calculation for the five varieties which verifies this previous assumption. Varieties 58-412 and 57-807 were believed to moder-

Table 2.—Within population genetic variance for root weight of the parental populations (parentheses) and their F-. intercross together with the average genetic variance for each variety based on the intercross performance (320 beets per population except where noted).

Parents	57-807	SP 5832-0	58-412	SLC 24	58-411	Mean
57-807	(0.721)	0.902	1.048	0.312	1.226	0.872
SP 5832-0		(1.055)	0.795*	0.632*	1.124[1]	0.863
58-412			**(1.149)**	1.508	1.988[1]	1.335
SLC 24				(0.799)	1.111[1]	0.891
58-411					**(1.452)**	**1.362**

* 160 beets per population

σ^2 Variety $\overline{X} = 1.035$

σ^2 Cross $\overline{X} = 1.065$

ate genetic variability with varieties SP 5832-0 and SLC 24 having low variability. This assumption is apparently true for variety 58-412 because the genetic variance of **1.149** was the second highest of the five varieties. The genetic variance for variety 57-807 was the lowest of the five varieties disputing the assumption that this variety had a relative broad genetic base. Variety SP 5832-0 produced from eight monogerm progenies had the third largest genetic variance.

As shown in Table 1 the mean variances for varieties were approximately the same as the mean variances for crosses. In general the genetic variance for a specific intercross was higher when the genetic variances of the two parents were high than when their variances were low.

Genetic Variances for Sucrose Percent

The within-population genetic variances for percent sucrose are included in Table 3. Variety 58-411 considered to have a broad genetic base, had the largest genetic variance for percent sucrose when compared with the other varieties. The American #2 variety, 58-412, which had a high genetic variance for root weight was fourth for sucrose percent. The second highest variance was for variety SP 5832-0 which was considered to have a limited genetic base. The ranking of the genetic variances for varieties 57-807 and SLC 24 were third and fifth respectively.

The mean genetic variance for varieties was little different from the mean variance associated with crosses. This observation for percent sucrose was identical to the results obtained lor weight of root.

Table 3.—Within population genetic variances for percent sucrose of the parental populations (parentheses) and their F_1 intercrosses together with the average genetic variance for each variety based on the intercross performance. (320 beets per population except where noted.)

Parents	57-807	SP 5832-0	58-412	SLC 24	58-411	Mean
57-807	(1.829)	1.623[1]	2.269	0.701	1.669	1.566
SP 5832-0		(2.125)	1.731[1]	1.001[1]	2.584[1]	1.735
58-412			(1.336)	2.637	—.108'	1.632 2.212'
SLC 24				(1.141)	1.172[1]	1.378
58-411					(2.444)	1.329 1.808[s]

[1] 160 beets per population
[2] Negative variance deleted

$\bar{x}$ = Variety $\bar{x}$ = 1.775
$\bar{x}$ = Cross $\bar{x}$ = 1.740

The genetic variation of the cross 58-412 X 58-411 was a negative value. Examination of the frequency distribution of this cross revealed that the sucrose determinations for individual plants were grouped quite closely around the mean of 13.02%, verifying that the genetic variation was low. The heterotic response of this cross for sucrose percent was 106.2% of the mid-parent value indicating considerable heterosis for this intercross. Two theories may be advanced to explain the lack of genetic variance in this cross. Several assumptions are necessary to substantiate these theories. The first theory assumes that heterosis has increased the sucrose yield of the hybrid considerably but that unknown limiting factors have surpressed the sucrose of the better genotypes. This ceiling has tended to force the population variability into a rather narrow range. The second theory assumes complete dominance of the high sugar variety, 58-412, for sucrose percent when combined with variety 58-411. However, this dominance is not evident in the other intercrosses of 58-412.

Discussion

The most critical part of an experiment of this type is the estimation of the environmental variance. There appears to be a close association between the means and variances for root weight. Thus, the regression method of estimating the environmental variances for each entry would be highly accurate providing enough estimates were available to establish that a true relationship exists between the means and variances. Another possibility would be the development of a series of inbreds and single crosses which encompass a wide range for both root weight and sucrose percent. Such a series could be utilized to measure the environmental effects by using the means and variances of the inbred or single cross which closely correspond to the means of the entry.

The genetic variances of varieties and intercrosses for root weight were estimated using the total variance of the single cross whose mean was closest to the mean of that particular entry. The regression method was calculated but not used because 32.8% of the variation was due to interactions, i.e., means and variances were interacting. The genetic variances for root weight could be slightly bias for those entries whose means were diverging from the single cross means.

The estimates of the environmental variances for sucrose percent were nearly identical varying only by .158. The mean of the two estimates was considered to be a good measure of the

environmental variance, thus, resulting in a quite accurate and comparable estimate of the genetic variances of percent sucrose.

A variety is considered to be a randomly segregating population with a balanced genetic constitution. Intercrosses between varieties should respond as F_2 and be segregating for genotypes. The estimations of genetic variation are actually measurements of the degree of segregation. One would expect that the average genetic variation for the crosses would be greater than the average variation for the varieties. However, the genetic variation means for crosses versus varieties were approximately equal for both root weight and sucrose percent. Examination of the data and frequency distribution showed that the genetic variation of certain crosses were less than the variation of the parental populations involved in the cross. A reaction of this type is difficult if not impossible to justify genetically. Several theories to explain these data are plausible. Two of the crosses which resulted in low genetic variation estimates involve the variety SP 5832-0. This variety was considered to have a narrow genetic base, however, the estimates of the genetic variation were higher than expected. A previous paper (3) presented data from reciprocal crosses which indicated that variety SP 5832-0 was self-fertile. This population could be composed of two types of material; plants resulting from selling and plants resulting from crossing. A population of this type would tend to have a high genetic variation due to the two extreme breeding types. The frequency distribution for this variety indicates two populations exist, but the population boundaries are not as apparent as in the cases of reciprocal crosses (3).

Another explanation of the similarity in the genetic variation of crosses versus varieties would be the lack of equilibrium in the intercrosses following hybridization. The intercross populations are essentially the first generation following crossing. Perhaps the genetic variation in later generations would be greater after the populations reached their genetic equilibrium. The advisability of selecting in the first generation after a variety cross is certainly open to question.

The results of the 57-807 X SLC 24 intercross are difficult to explain. The genetic variation for this intercross was considerably below either of the parental populations for both root weight and sucrose percent. The actual yield was also below that of the parents (2). The type of gene action that would cause this reduction in yield and genetic variation defies logical explanation.

The data generally follow a logical pattern and were considered to be reliable. However, the genetic estimates are relative only to the varieties and intercrosses under test and are possibly subject to yearly interactions.

Summary

Estimates of the genetic variation among five diverse monogerm varieties and their diallel intercrosses are presented for root weight and sucrose percent. The average genetic variation of the intercrosses was approximately equal to the average genetic variation of the varieties for both root weight and sucrose percent.

Certain irregularities in the results were discussed along with the method of estimating environmental variance.

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Variety Crosses in Sugar Beets (Beta vulgaris L.) III. Estimating the Number and Proportion of Genetic Deviates by the Partitioning Method of Genetic Analysis

R. H. HELMERICK, R. E. FINKNER AND C. W. DOXTATOR[1]

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The partitioning method of genetic analysis (5)[2] provides a means of estimating the numbers and proportion of genetic deviates in random mating populations. The population geneticists may be interested in number or proportion of individuals falling in certain classes of the frequency distribution because of their genotypes, while the plant breeder is interested in those individuals which occur in the upper classes of the frequency distribution. The latter must be reasonably sure that such superior individuals are not chance fluctuations from the mean of the population but are in the upper portion of the frequency distribution because of their genotype. Previous papers (1, 2) have presented the data on heterosis, combining ability and genetic variation when five diverse varieties of sugar beets were crossed in a diallel series. The purpose of this paper is to present the number of superior genetic deviates found in the varieties and their intercrosses using the partitioning method of genetic analysis.

Materials and Methods

Details of the experimental design and variety descriptions are included in a previous paper (1). The statistical analyses of the data presented in this paper are essentially those described by Powers (3, 4, 5) for the partitioning method of genetic analysis.

Frequency distribution tables were prepared for both root weight and sucrose percent for each entry in the experiment. These included the 5 monogerm varieties, their 10 intercrosses, 2 single crosses and the multigerm check. The frequency interval for root weight was .4 pound with a range from .4 to 6.4 pounds. The range for percent sucrose was from 7.50% to 18.00% at .75% frequency intervals.

The expected number of beets in each class interval was calculated (5) by projecting a normal distribution based on the population mean and standard error of the single cross which

[1] Plant Breeder, Manager Research Station, and Plant Breeder, respectively, American Crystal Sugar Company, Rocky Ford, Colorado.

[2] Numbers in parentheses refer to literature cited.

estimated the environmental variance. For root weight, the standard error used to calculate the expected frequency distribution of any particular entry was the square root of the total variance for the single cross used to estimate its genetic variance. The mean of the two standard errors for the single crosses was used to calculate the expected frequency distribution for percent sucrose.

The frequency distributions were partitioned into three groups based on the change of the expected from the observed according to the partitioning method. Homogeneity chi square calculations indicate that the obtained populations for all entries except the nonsegregating populations vary from the expected. These calculations indicate that genetic deviates occur in all the populations except the nonsegregating populations. The plants located in the higher partition were considered genetically superior and the number of those plants in the population calculated.

Experimental Results

The partitioning method of genetic analysis enables the plant breeder to mathematically select the genetic deviates which fall into the upper classes of the frequency distribution. Based on the frequency data, Table 1 shows the number of genetic deviates for root weight that the plant breeder could expect of find per 10,000 plants. The table includes the five varieties and their ten intercrosses that were tested in this experiment. The percentage of these genetic deviates in the population vary from 5.9 to 17.5%. The average number of genetic deviates in 10,000

Table I.—Number of genetic deviates per 10,000 plants for weight of root for five varieties and their intercrosses (parental populations in parentheses).

Populations	57-807	SP 5832-0	58-412	SLC 24	58-411
57-807	(375)	1375	1406	594	1169
SP 5832-0		(1187)	1000	1250	1750
58-412			(1594)	1719	1625
SLC 24				(656)	1312
58-411					(1094)

Table 2.—Number of genetic deviates per 10,000 plants for percent sucrose for five varieties and their intercrosses (parental populations in parentheses).

Populations	57-807	SP 5832-0	58-412	SLC 24	58-411
57-807	(125)	425	750	188	531
SP 5832-0		(62)	750	688	188
58-412			(375)	938	625
SLC 24				(219)	375
58-411					(312)

beets for the varieties is 981.2 while the average for the intercrosses is 1,350.0.

Table 2 presents the number of genetic deviates per 10,000 plants for percent sucrose. The percentage of the genetic deviates in the five populations and their intercrosses range from 0.62 to 9.38%. The average number of deviates for the varieties is 218.6 plants while the average for the intercrosses is 545.8 plants.

The number of genetic deviates per 10,000 plants for both root weight and sucrose percent are included in Table 3. The percent of plants superior in both weight and sucrose percent ranged from .03 to 5.67%. The average number of tliese plants in the varieties is 24 while 122.2 is the average of their intercrosses.

The obtained and estimated number of plants occurring above the .01% level of probability for each population are included in Table 4. These genetically-superior individuals are of particular interest to the plant breeder. Based on the calculations he can be reasonably sure that these plants were not in the higher frequency classes due to chance but are genetically superior.

Twelve genetically-superior plants were obtained, all found in the intercross populations, with 42 expected from the combined populations.

Table 3.—Number of genetic deviates expected per 10,000 plants based on the percentage of genetic deviates identified for root weight and percent :

Populations	**57-807**	**SP** 5832-0	**58-412**	**SLC 24**	**58-411**
57-807	*(5)*	567	105	11	78
SP 5832-0		(7)	75	86	**33**
58-412			(60)	116	102
SLC 24				(14)	**49**
58-411					**(34)**

Table 4.—Obtained and estimated number of genetically superior individuals for both high root weight and sucrose percent.

Populations		**57-807**	**SP 5832-0**	**58-412**	**SLC 24**	**58-411**
57-807	O	(0)	4	2	0	0
	E	(0)	18	4	0	2
SP 5832-0	O		(0)	0	0	0
	E		(0)	2	3	1
58-412	O			(0)	5	0
	E			(2)	4	3
SLC 24	O				(O)	1
	E				(2)	2
58-411	O					(0)
	E					(1)

Discussion

A greater number of genetic deviates were found for root weight than for sucrose percent. This data would indicate that if the plant breeder were mass selecting for tonnage, fewer plants would be necessary than if he were selecting for sucrose percent. Selection for both sucrose percent and weight of root would require an even greater number of plants than for either factor separately. The selection of 50 genetically-superior plants would require even a greater number of individuals. Considering all the populations, 3,840 plants were observed with 12 or 0.31% being genetically superior. If the plant breeder considered that 50 beets were sufficient to maintain genetic variability in the selected population, approximately 16,130 beets would be needed to insure the selection of this 50-beet sample.

No genetically-superior individuals were identified in the parental varieties. The 12 superior individuals for both root weight and sucrose percent were all found in the intercross populations. These data would suggest that the genetically-superior individuals are the result of hybridization and represent superior heterozygotes. Due to segregation during advanced generations, the original mean yield of the mother beets would be difficult to maintain. However, if the gametes which produced these genetically-superior individuals could be isolated and maintained by inbreeding, the original yield could be reproduced by hybridization.

Several of the populations studied appeared to have good genetic variability with a high probability of selecting genetically superior individuals. However, the mean yield and genetic variability of the populations *need* to be considered before subjecting them to selection. A population with a low mean yield and high genetic variability may be greatly improved by mass selection. However, the resulting increase in the mean yield may not equal a population with a high mean yield and low genetic variability before the variability in the population under selection is drastic-, ally reduced.

In our study population, 58-412, would be the best variety for advanced breeding: either mass selection or recurrent selection. The mean yield for both sucrose percent and root weight was high with good genetic variability present in the populations. No genetically superior individuals were identified in this population, however, two were expected.

The intercross 58-411 X 58-412 appears to be the best varietal hybrid to select within following several generations of open-

pollination. Based on the mean yield, heterotic responses and genetic variability, these two varieties, 58-411 and 58-412, would also be the two best varieties to incorporate into a reciprocal recurrent selection program. Inbreeding with the synthetic populations produced by this cycling process should isolate gametes (inbred lines) that when hybridized would result in a population of genetically superior individuals.

Summary

The proportion of genetic deviates and number of genetically superior individuals were studied in five open-pollinated populations and their intercrosses by the partitioning method of genetic analysis. The number of genetic deviates identified in the higher classes of the frequency distribution was greater for root weight than for sucrose percent. Twelve genetically superior individuals were isolated for the populations with forty-two expected.

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Cultural and Environmental Requirements For Production of Zoospores by Aphanomyces cochlioides in Vitro

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Sugar beet strains developed at the Plant Industry Station, Beltsville, Maryland, have been screened in the greenhouse for resistance to the fungus, *Aphanomyces cochlioides.* In each test of 24 entries, large quantities of inoculum—approximately 100 million zoospores—are required (5)[2] . At the outset of the testing program, zoospore inoculum was obtained in accordance with a previously described method (4) whereby mycelial mats of the fungus are submerged in water at 20 to 25 C for about 16 hours. Wide variation in number of zoospores produced at different times indicated a need to determine the variables, besides temperature, that influence zoospore production.

It has been shown that zoospore production by a related fungus, *Aphanomyces euteiches,* is influenced by the type of medium which the mycelial mats are produced, temperature, age of culture, type of water, and aeration of water (2).

The experiments described herein were conducted to determine the degree to which the following variables influence zoospore production by *A. cochliodes:* age of culture, type, pH, and aeration of water; relative amounts of mycelium and water. An abstract of some of the results has been published (6).

Methods

Monosporous cultures, isolated from damped-off sugar beet seedlings and maintained on maize meal agar, were used. Mycelial mats were produced in flasks containing 0.3% peptone or 0.3% Soytone[3,4]. Previous studies showed that addition of dextrose, maltose, or sucrose to broth did not enhance zoospore production (6). The size of the flask and amount of broth in which mycelial mats were produced varied from one experiment to another.

Zoospore production was induced by rinsing the mycelial mats and transferring them to flasks containing water at 20 to 25 C. Approximately 16 hours later, spore counts of 10 ml samples from each flask were made with a brightline counting chamber.

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[2] Numbers in parentheses refer to literature cited.

Zoospores were immobilized by the addition of 0.1 ml Roccal[4 5] solution (800 ppm) to each sample in order to facilitate counting.

Results

Age of culture

Zoospore production was compared between cultures of different age. Flasks containing 125 ml nutrient broth were inoculated 5, 7, 11, 14, and 20 days before each subsequent mycelial mat, incubated at 20 C, was transferred to 250 ml tap water. Zoospore production was greatly influenced by age of culture. Average number of zoospores produced by 3 myclelial mats in each age group was as follows:

Age of culture (days)	Zoospores/ml (thousands)
5	41.4
7	65.6
11	10.4
14	4.8
20	0
LSD (P = .05) =	7.1

The decline in zoospore production, noticeable by the 11th day, can be delayed by refrigeration. On numerous occasions, broth cultures placed in the refrigerator at 5 C, on the fourth day after inoculation, produced over 80,000 zoospores/ml when transferred to water 14 days later.

Type of water

Zoospore production was compared *in* 3 types of water (tap, distilled and demineralized) alone and with NaCl (120 mg per liter) added. The water in which each mat was rinsed was of the same tvpe as that in which it was subsequently submerged. The greatest number of zoospores was produced in tap water (Table 1). Zoospore production was increased by the addition of NaCl, especially in distilled and in demineralized water.

Table 1.—Zoospore production by mycelial mats of *Aphanomyces cochlioides* in 3 types of water, with and without NaCl.

Type of water	Zoospores/ml (thousands)[1]	
	+ NaCl (120 mg/liter)	Control
Tap	113.8	94.7
Distilled	94.0	33.5
Demineralized	70.9	30.6
LSD (P = .05)	18.9	

[1] Results expressed as average of 2 experiments, each with 4 replicates per treatment. Each replicate comprised one mycelial mat, produced in 30 ml broth, in 90 ml water.

[3] Trade name of an enzymatic hydrolysate of soybean meal prepared by Difco Laboratories, Detroit, Michigan.

[4] Mention of material and company name is for identification only and does not imply endorsement by U. S. Department of Agriculture.

[5] Trade name of a germicide containing benzalkonium chloride, prepared by Winthrop Laboratories, N.Y.

pH of water

Zoospore production was compared in demineralized water and in tap water adjusted to several pH values from 5.6 to 8.1 with M/3 KH_2PO_4 and M/3 Na_2HP0_4 buffer solutions. In both types of water, abundant zoospores were produced at pH 5.6 - 7.5 (Table 2). Zoospore production decreased noticeably at pH 7.8 and beyond.

Table 2.—Zoospore production by mycelial mats of *Aphanomyces cochlioides* in water of different pH.

Experiment No.	Type of water	Zoospores/ml (thousands) in water of indicated pH[1]								LSD (P = .05)
		5.6-5.7	5.8-5.9	6.0-6.1	6.4-6.5	7.0-7.1	7.4-7.5	7.8-7.9	8.0-8.1	
1	Demineralizcd	56.3	62.7		64.3		68.3	42.5	32.3	24.2
2	Tap	97.7	.....	118.0	142.2	127.5	123.5	70.7		21.1

[1] Results expressed as mean of 3 replicates, each comprising one mycelial mat, produced in 30 ml broth, in 100 ml water.

Aeration of zvater

Tap water was aerated by bubbling air through it during the 16 hours that the mycelial mats were submerged. Air was introduced through glass tubing (4 mm diameter) at the approximate rate of 250 ml/min. In 4 experiments, zoospore production in aerated flasks was increased approximately 2 to 3 times over that in control flasks.

Table 3.—Zoospore production by mycelial mats of *Aphanomyces cochlioides* in aerated and nonaerated water.

Experiment number	Amount of water (ml)	Zoospores/ml (thousands)[1] produced in water treated as indicated.	
		Aerated	Control
1	150	105.5	32.5
2	300	48.9	18.8
3	1000	31.5	J0.5
4	1000	102.2	48.8

[1] Results based on one re plicate per treatment in each experiment.

Relative amounts of mycelium and zvater

Zoospore production was compared between flasks containing equal amounts of mycelium in different amounts of water and between flasks containing different amounts of mycelium in equal amounts of water. The mycelial mats when fully grown appear to occupy all of the volume of the broth, hence the approximate amount of mycelium constituting a mat can be designated by the volume of broth in which it was produced. The relative amounts of mycelium and water can thereby be expressed as the ratio of broth (ml) in which mats are produced and of water (ml) in which they are submerged.

With quantity of mycelium constant, zoospore production increased with added amounts of water, then leveled off after a mycelium-water ratio of 1:3 was attained (Table 4). With quantity of water constant, zoospore production increased with added amounts of mycelium when the mycelium-water ratio was 1:8 but not when the ratio was 1:4 and less (Table 5). The average number of zoospores produced per mycelial mat was greatest when the mycelium-water ratio was 1:3 to 4.

Table 4.—Zoospore production by mycelial mats of *Aphanomyces cochlioides* in different amounts of tap water.

Amount of water (ml)	Ratio[1] mycelium: water	Zoospores/ml[2] (thousands)	Total zoospores/ flask[2] (millions)
50	1 1	33.4	1.67
100	1 2	40.3	4.03
150	1 3	35.3	5.29
200	I 4	25.3	5.07
LSD (P = .05)			1.85

[1] Ratio between ml broth in which each mycelial mat was produced and ml water in which it was submerged.
[2] Results expressed as mean of 3 replicates; each comprising one mycelial mat, produced in 50 ml broth, in indicated amount of water.

Table 5.—Zoospore production by different numbers of mycelial mats of *Aphanornyces cochlioides in* lal amounts of tap water.

Number of ycelial mats	Experiment 1			Experiment 2		
	Ratio[1] mycelium:water	Zoospores/ml[3] (thousands)	Zoospores[2] produced per mycelial mat (millions)	Ratio[1] mycelium:water	Zoospores/ml[3] (thousands)	Zoospores[3] produced per mycelial mat (millions)
1	1 : 3.3	82.8	8.28	1 : 8	59.0	14.75
2	1 : 1.7	92.6	1.63	1 : 4	119.9	14.99
3	1 : 1.1	83.1	2.77	1 : 2.7	96.9	8.16
4	1 : 0.8	92.2	2.31	.. .		
D (P = .05)		NS			35.8	

[1] Ratio between amount of water in which mycelial mats were produced and amount of water in which they were subsequently submerged.
[2] Results expressed as mean of 4 replicates; each comprising designated number of mycelial mats. duced in 30 ml broth, in 100 ml water.
[3] Results expressed as mean of 3 replicates; each comprising designated number of mycelial mats. duced **in** 30 ml broth, **in** 240 ml water.

Discussion

On the basis of the studies described, an improved methodology for production of zoospores in quantity by *A. cochlioides* has been established. Adequate quantities of zoospore inoculum have been consistently obtained from mycelial mats, not over

7 days old, submerged in a quantity of aerated tap water, with NaCl (120 mg per liter) added, equal to approximately 3 times the quantity of broth in which the mycelial mats were produced.

It is not fully known why more zoospores are produced in tap water than in distilled or in demineralized water. The presence of injurious elements or lack of essential ones has been cited as a possible cause of suppressed zoospore production by certain *Saprolegniaceae* in distilled water (3). The reason for increased zoospore production in water to which NaCl has been added, noted also by Sherwood (7) with *A. euteiches,* is, as yet, unknown.

In the laboratory where the preceding experiments were conducted, pH of tap water has been near optimum for zoospore production. Where pH of water is near 7.8 or above, reduced sporulation would be expected.

Increasing the amount of water in which mycelial mats are submerged would reduce the concentration of nutrients that may be carried from the broth by the mats. Increased zoospore production with increased amounts of water might therefore be associated with reduced concentration of nutrients in the water. Klebs (1) noted suppression of zoospore production by the water mold, *Saprolegnia mixta* with low concentrations of organic nutrients in the water.

Summary

Mycelial mats of *Aphanomyces cochlioides,* 5 to 7 days old, produced more zoospores than those 11 days old and older. More zoospores were produced in tap water than in distilled or demineralized water. NaCl (120 mg per liter) added to water in which mycelial mats were submerged enhanced zoospore production. Zoospore production at pH 7.8 and above was considerably less than at pH 5.6 to 7.5. The greatest number of zoospores per mycelial mat was produced when the quantity of water in which mycelium was submerged equaled 3 to 4 times the quantity of broth in which it was grown.

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The Effect of Method and Rate of Phosphate Application On Yield and Quality of Sugar Beets[1]

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The frequent need for phosphate fertilizer for sugar beet production in Colorado is recognized, and the annual rate of application has been about 100 lb P_2O_5 (44 lb P) per acre (l)[3]. Efficient methods of application of phosphate fertilizer are required to insure high yields and high quality of roots. The objective of this study was to determine the influence of method of application of phosphate fertilizer on yield and quality of sugar beets. Two rates of phosphate were applied to determine if a method X rate interaction would appear.

Experimental Procedure

The experiment was conducted on a calcareous Larimer fine sandy loam which contained 37 lb of available P_2O_5 (16 lb P) per acre by the $NaHCO_3$ test (4). The site had been in irrigated grass pasture for 17 to 18 years, then planted to alfalfa for one year with a barley nurse crop before the sugar beet experiment. There was no record of previous fertilizer application. Concentrated superphosphate was applied at two rates, 50 and 200 lb P_2O_5 (22 and 88 lb P) per acre by; 1) broadcasting the fertilizer on the surface and plowing under with the legume and grain stubble, 2) a broadcast application after plowing mixed 3 to 4 inches into the surface by disking, 3) a split application with one half the fertilizer plowed under and the remainder disked into the surface, 4) banding the phosphate $1^{1/2}$ to 2 inches below the seed and 5) plowing under 150 lb and banding 50 lb P_2O_5 below the seed. The treatments were replicated four times. Nitrogen at the rate of 150 lb N per acre was broadcast uniformly over the experimental area and mixed 3 to 4 inches into the surface with the disking operation.

The crop was planted April 15, 1958. Stands were good on all treatments and growing conditions were generally good throughout the season. Petioles were taken at three sampling dates and analyzed for acetic acid-soluble phosphorus (2). The beets were harvested October 22. Root weights and sucrose

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[3] Numbers in parentheses refer to literature cited.

content were determined. The data were analyzed statistically. A "significant" effect, as used in the text, indicates that the odds are 19 to 1, or greater, that the observed result was caused by the imposed treatment rather than by chance.

Results and Discussion

Beet and sugar yields and percentage sucrose are presented in Table 1. The method of application of phosphate had little influence on final yield of roots, sucrose content or sucrose production; nor was there a significant interaction for rate X method-of-application. The yield of beets was increased nearly 4 tons per acre by the application of 50 lb P_2O_5. Applying an additional 150 lb P_2O_5 increased the yield another $1^{1/2}$ tons above that of the 50-lb rate. Sucrose percentage did not appear to be influenced by either method or rate of phosphate application. Quality of beets, using sucrose percentage as the index, was maintained with the application of 150 lb N applied uniformly to the area. Yield of sugar was a reflection of beet yield.

Table 1.—The effect of method and rate of application of phosphate fertilizer en yield of beets, percent sucrose and sugar production.

Method of application of phosphate	50 lb P_2O_5 per acre			200 lb P_2O_5 per acre		
	Roots tons/A	Sucrose %	Sucrose tons/A	Roots tons/A	Sucrose %	Sucrose tons/A
Plow under	17.3	20.0	3.46	18.5	20.4	3.77
Disk	17.4	19.8	3.44	19.5	19.6	3.82
Split (plow &disk)	16.8	20.1	3.38	19.4	19.6	3.80
Band below seed	17.7	19.9	3.52	18.4	19.5	3.59
Plow 150 lb, band 50 lb				18.3	20.0	3.66
Avg.[1]	17.3	20.0	3.45	19.0**	19.8	3.73*
Significance—method of appl.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.

•Greater than 50 lb P_2O_5 rate at 0.05 level of significance.
* * Greater than 50 lb P_2O_5 rate at 0.01 level of significance.
[1] No phosphorus treatment: 13.5 tons roots, 20.2% sucrose, 2.73 tons sucrose.

The effect of the phosphate treatment on acid-soluble phosphorus in the petioles is shown in Table 2. Application of phosphate fertilizer increased petiole phosphorus over the no-phosphorus treatment in every case except for the first sampling where 50-lb P_2O_5 had been disked into the soil. Early in the season (June 17) petiole phosphorus was significantly higher for the band-applied phosphate than for the other methods of application at the 50-lb rate of fertilization. With the 200-lb rate there was little difference between band, plow-down and split (plow-disk) applications but petiole phosphorus was significantly

Table 2.—The effect of method and rate of application of phosphate fertilizer on acetic acid soluble phosphorus in beet petioles at different stages of growth.

Method of application	50 lb P_2O_5[1] Sampling date			200 lb P_2O_5 Sampling date		
	June 17	July 22	Au-f. 13	June 17	July 22	Aug. 13
	ppm P					
Plow under	2100	1750	1200	2600	2450	1500
Disk	1550**	1300**	950	1850**	2050**	1300
Split (plow-disk)	1950	1700	1000	2500	2050	1650
Band below seed	2400**	1600	1200	2450	1900**	1250
Plow 150 lb band 50 lb				2650	2300	1400

**Significantly different at the 0.01 level from plow-under method of application at the same rate of phosphate and the same sampling date.

[1] Acid soluble phosphate in the no-phosphorus treatment was 1600, 1050, and 600 ppm P for the June, July and August samplings, respectively.

lower for the disk application. For the July sampling, band, disk and split (plow-disk) methods of application were less effective, as indicated by petiole phosphorus, than the plow method of application. The sampling on August 13 showed little influence of method of application on petiole phosphorus. Petiole phosphorus for the combination of plow-under and banded phosphate was about the same as 200 lb P_2O_5, plowed under.

The interaction rate X method-of-application was significant for petiole phosphorus for the first and second sampling dates. This was caused by a relatively greater effectiveness of the plow-down method of application, as shown by petiole phosphorus, at the 200 lb rate than at the 50 lb P_2O_5 rate.

At all sampling dates and particularly for the plow and disk methods of application, increasing the rate of phosphate significantly increased the acid-soluble phosphorus content of the petiole. An early stimulation in top growth from phosphate fertilizer was observed, but the early visual effects were caused largely by phosphate rate rather than method of application.

The results of the experiment show that phosphorus fertilizer increased both root and sugar yields of beets grown in this phosphate-deficient soil. Early and midseason petiole measurements of acid-soluble phosphorus, however, were not a reliable index of the influence of method of fertilizer application on yield or quality of the crop at harvest. On the other hand, the late season sampling appeared to be more closely associated with yield. The seasonal change in compostion of the petiole would tend to support the thesis that the bulk of the absorptive root tissue of the sugar beet does not remain in the vicinity of a concentrated band of fertilizer but for a short time early in the

season. Other work in Colorado (5,8) has shown the influence of a band or concentrated placement of fertilizer is dependent upon the relative positions of fertilizer and seed and will change as the season progresses. The small effect of method of application of phosphate on crop yields suggest that the sugar beet plant has great ability to adapt to the environment.

Results of research in Montana (3), Colorado (8) and Wyoming (6) have shown that plowing down an application of phosphate fertilizer was generally as good or better, as indicated by crop yields, than band or surface applications. Incorporation of phosphate by disking the light-textured soil of this experiment was possibly more effective than with a heavier-textured soil. Since the soil was light in texture at the surface, more frequent early irrigations were required. This would have promoted more root growth in the top soil and could have caused relatively better results from the disked applications than often observed. At the same time, the band application should have benefited from the irrigation management.

The application of phosphate had no significant effect on the sucrose content of the root. Other results in Colorado (8) and in Nebraska (7) would suggest that applications of phosphate may increase the sucrose content of the root when applied to soils very low in available phosphate but would have little influence when applied to soils intermediate to high in available soil phosphorus.

Summary

A field experiment was conducted to study the yield and quality of sugar beets as affected by method and rate of application of phosphate fertilizer on a phosphate-deficient, calcareous soil.

1. The phosphate applications increased yield of beets and sugar production.
2. There were no significant differences in yield or sucrose content among methods of phosphate application for either 50 or 200-lb P_2O_5 rates; nor was there a method X rate interaction.
3. The early growth response to band-applied phosphate as shown by visual observations and chemical composition of the petioles did not continue through the season or result in enhanced yields for this treatment.
4. Yield responses to phosphorus fertilizer were more closely associated with late rather than with early season petiole analyses.

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Influence of Inhibitors in Sugar Beet Fruits on Speed of Germination at 50 and 70 Degrees Fahrenheit[1]

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In most areas where sugar beets are grown, planting as early as possible has been most profitable. Thus, the seeds (fruits) are planted in cold, moist soil. Soil temperatures below 50 F may delay germination and emergence a number of days during which diffusion of inhibitors may take place. Therefore at this low temperature, rapid water absorption and presence of inhibitors in the fruit may not be significant factors *in* the speed of germination. On the other hand, at 70 degrees the germination processes are initiated so rapidly that slow water absorption and inhibitors in the fruit may exert a considerable delaying action on germination.

Smith (2)[3] demonstrated that seeds of sugar beet varieties differ in their ability to germinate at 43 F and that this ability is a heritable trait. Sedlmayr (1) also demonstrated that speed of germination at room temperature is heritable. Snyder (3) and Sedlmayr (1) observed that speed of germination at room temperature is largely controlled by the fruit (maternal tissues) which surrounds the true seed. Speed of germination at room temperature has been causally related to the concentration of inhibitory substances in the fruit of the sugar beet[4]. Chemical inhibitors in the fruits of commercial varieties seem to control speed of germination more than does the physical nature of the fruit (3). Miyamoto and Dexter[5] removed the inhibitors by washing in water and inactivated them by soaking fruits in a solution containing mercury ions; however, emergence from cold soil was accelerated only slightly in comparison with untreated fruits.

This investigation was undertaken to determine whether (a) sugar beet strains could be selected that would emerge rapidly from soil at 70 and at 50 F, (b) the retarding effect of inhibitors

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[4] Unpublished data of F. W. Snyder, J. M. Sebeson, and J. L. Fairley.

[5] Unpublished data of T. Miyamoto, and S. T. Dexter, Michigan Agricultural Experimen! Station, East Lansing, Michigan.

in the fruits on germination and emergence would be completely dissipated in moist soil at 50 F, and (c) the relative growth activity of the embryos of strains of US 401 differed at 50 and 70 F.

Methods and Materials

Samples of open-pollinated seed (fruits containing the seeds) harvested from 538 plants of sugar beet variety US 401, previously indexed for speed of germination, were available for this study. Fifty whole seedballs of the 13 most rapid and the 13 slowest samples were again germinated by the blotter method. The 10 most rapid and the 10 slowest samples were chosen for the tests in soil.

Sandy loam was steam-sterilized, air-dried, and then moistened uniformly to contain approximately 17.5% moisture at planting time. Plastic dishes ($10^{3/4}$ x $7^{1/2}$ X $2^{1/2}$ inches) were filled to a depth of $1^{7/8}$ inches with soil, which was leveled and then compacted with a board ($10^{1/2}$ X $7^{1/2}$ inches) having 10 cleats 1/2 inch in depth to form the rows. A pressure of 150 pounds was applied to the board for approximately 15 seconds.

The seedballs (previously treated with a fungicide) were planted in a randomized block design. Each row contained 5 seedballs of a given entry. Thus with 10 rows, each dish contained 50 seedballs. Two dishes were required for one replication. Ten replications or 50 seedballs per entry were planted. After the seedballs were placed, they were coverd with 5/8 inch of loose soil of the same moisture content. The dishes were immediately placed in plastic bags to minimize evaporation. The dishes for the higher temperature phase of the experiment had been placed at approximately 70 F about 5 hours before planting. Those for the lower temperature were maintained at approximately 60 F until planted and then were placed at a mean temperature of 53 for the first 2 days, 49 for the next 6 days, and then maintained at 50 from the eighth day until the experiment was concluded.

The percentages for termination and emergence were corrected for germination failures whenever a seedball contained all defective seeds. Only the first seedling from a seedball was counted, since the seedball was considered as a single unit. Each entry was ranked according to an accumulated score based on speed of germination or emergence.

Results

Speed of germination by the blotter method and speeds of emergence from soil at approximately 50 F and 70 for the rapid

Table 1.—Speed of germination and emergence percentages of 10 rapid and 10 slow entries of US 401 sugar beets.

Method and length of test period (days)	Averages for		Ranges for	
	Rapid	Slow	Rapid	Slow
Blotter germination at approx. 70 F				
2	40.2	0.0	22-71	
2 1/2	82.2	7.9	62-94	0-20
3	95.6	28.4	86-100	4-67
3 1/2	98.6	50.8	94-100	12-90
4	99.8	69.7	98-100	32-98
5	100.	87.8		52-100
10	100.	96.4		
Emergence from moist soil at approx. 70 F				
3	5.4	0.8	0-14	0-6
3 1/2	54.4	12.7	34-88	0-33
4	96.8	51.2	90-100	4-90
4 1/2	99.4	**83.3**	92-100	14-100
7	99.4	98.4		
Emergence from moist soil at approx. 50 F				
11	19.0	8.8	6-36	2-18
12	53.4	25.3	30-84	2-44
13	87.2	60.3	70-100	20-84
14	99.0	86.2	96-100	60-98
19	99.8	97.0		

Table 2.—Time requiired to attain 75 percent germination and emergence for the 20 entries of US 401.

Test	Temp. F	Number of days required		Range in time
		Minimum	Maximum	
Blotter germination	70	2	10 +	8 +
Emergence from soil	70	3 1/2	5 1/2	2
Emergence from soil	50	12	15	3

and slow entries are given in Table 1. The time required to attain 75% germination or emergence was determined for each test (Table 2).

The data revealed the following: 1) Percentage of emergence from either 50 F or 70 soil was as good as percentage of germination on the blotter at 70. 2) The germination-time and emergence-time curves were essentially the same in shape, whether the entry was "fast" or "slow"; the difference was largely in the initial delay to first sprout. 3) In soil at 70, the variation in time to attain 75% emergence between "fast" and "slow" was about 57%, but at 50, only about 25% (based on minima). 4) The greatest range in speed of germination was on the blotters. In soil, the performance of the entries was more uniform.

The rank order of the 20 entries for performance in each of the 3 tests (Table 3) revealed three patterns of performance: 1) Maintained about the same relative rank in all three tests (entries 1,7,8,11, etc.); 2) improved rank in emergence from soil at 70 F and a further improvement in soil at 50 as compared with blotter germination (entries 12 and 19); and 3) maintained relative rank in the tests at 70, but had a slower relative speed of emergence at 50 (entry 3).

Correlation coefficients calculated from the accumulated scores for the speeds of germination and emergence were as follows: Blotter versus soil at 70 F, 0.765**[6]; blotter versus soil at 50 F, 0.522*; and soil at 70 versus soil at 50, 0.787**, while correlation coefficients calculated from the rank order data (Table 3) were 0.872**, 0.519*, and 0.462* respectively.

Table 3.—Ranking of 20 entries of sugar beet variety US 401 for speed of germination and speed of emergence from soil at 2 temperatures (F).

	Blotter germination	Emergence from moist soil	
Ranking	70	70	50
Most rapid	1	6	
	2	1	
	3	2	12
	4	4	10
	5	3	8
	6	7	7
	7	8	2
	8	10	19
	9	12	4
	10	11	14
	11	5	11
	12	9	5
	13	13	15
	14	19	9
	15	20	18
	16	14	13
	17	15	3
	18	18	17
	19	17	20
Slowest	20	16	16

Discussion

From the literature on sugar beets, the principal parameters of speed of germination on a blotter or speed of emergence from soil appear to be the relative concentration of inhibitors in the fruit and the relative growth activity of the embryo over a range of temperatures. The growth activity at 70 F often may be masked by the high concentration of inhibitors. At lower temperatures the interrelations are more uncertain.

[6] Significance: ** at the 1% level, * at the 5% level.

The three patterns of performance can be accounted for on the basis of these parameters. Entry 1, and to a degree, entry 6 have a low concentration of inhibitors and both have unusually active embryos at 50 F. Apparently both of these desirable characteristics can be found in a single strain. Both entries 1 and 6 emerged rapidly from soil at 70 and 50 F, and would be superior agricultural varieties on the basis of germination performance. Entries 2, 3, 4, 12, and 19 are conspicuously out of rank in speed of emergence from soil at 50 as compared with their rank at 70. The performance of 2, 3, and 4 at 70 F, presumably because of low concentration of inhibitors, would not be improved by a longer period of diffusion at 50. Their relatively slower emergence at 50 may be attributed to low embryo activity at that temperature. Entries 12 and 19 emerged relatively faster when their inhibitors were permitted to dissipate in soil at 70 or 50. However, since neither equalled the speed of entry 1 at the low temperature, they apparently had a lower embryo activity in addition to the greater concentration of inhibitors.

The performance of the entries in the three tests seems to delineate the parameters and indicate the characteristics of each entry. The various combinations of characteristics exhibited by entries used in this study are illustrated (Table 4).

Table 4.—Combinations of the two parameters which affect speed of germination and emergence of selected strains of US 401.

Entry	Concentration of inhibitors in fruit	Growth activity of embryo at	
		70 F	50 F
1, 6	Low	Fast	Fast
5	**Low**	Intermediate	Intermediate
3	**Low**	Fast	Slow
10, 12	**Intermediate**	Intermediate	Fast
11	**Intermediate**	Intermediate	Intermediate
19	**High**	Intermediate	Intermediate
16, 17, 18	**High**	Slow	Slow

The correlation coefficients calculated from the accumulated scores for speed of germination or emergence indicate a reasonably good relation between blotter germination and emergence from soil at 70 F and between emergence from soil at 70 and 50 F, but a low correlation between blotter at 70 and soil at 50. However, a sufficient number of exceptions to the general performance were noted that interpretations and extrapolations of results must be made with considerable caution. The exceptional performance of an entry, e.g. entry 1, may be of much greater value for improving germination than average performance of a number of entries.

The data (Table 3) indicate clearly that the performance of seeds cannot be predicted with certainty from a single test. However, the consistent relative performance of a number of the entries in the three tests suggests that results of germination and emergence experiments conducted at room temperature may be applicable over a wider range of temperatures than suspected. While the blotter method is the simplest and quickest germination test, emergence from moist soil at approximately 50 F appears to be a more reliable test to indicate potential performance under field conditions. The latter method may be especially useful in selecting the best germination characteristics for variety improvement.

Summary

Samples of open-pollinated "seed" of 538 plants of sugar beet variety US 401 were available for study. The 10 most rapid and 10 slowest germinators, as determined by the blotter method at approximately 70 F, were chosen. Fifty seedballs of each entry were planted in sandy loam at approximately 17.5% moisture at 2 soil temperatures, 70 and 50.

The 20 entries were ranked from the most rapid to the slowest in each test (blotter germination, emergence from soil at 70 F, and emergence from soil at 50). Three patterns of performance were found: 1) Some entries maintained the same relative rapidity in all 3 tests; 2) others germinated relatively slowly on blotters as compared to emergence from soil at 70 or 50; and 3) still others maintained relative rapidity in the 2 tests at 70, but had a slower relative speed of emergence at 50.

Speed of emergence from moist soil at approximately 50 F appears to be a more reliable test to indicate potential performance under field conditions than speed of germination on blotters at room temperature.

Strains of sugar beets which emerge rapidly at 70 as well as at 50 F can be selected.

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A Technique for Obtaining Identical Pairs of Seedling Beets[1]

A. M. HARPER[2] AND J. B. TENNANT

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There is considerable genetic variation between sugar beet plants because they are cross-pollinated. A technique of splitting large beets has been used to obtain genetically identical plants (4)[3]. Cuttings from crown buds and semi-vegetative seed-stalks have also been used in asexual propagation of beets (2). Although these techniques will remove genetic variation they are not satisfactory when young plants are being studied. Pawlowski (3) obtained identical pairs of sunflowers by splitting seedlings. In 1962 identical pairs of seedling beets were obtained by a modification of Pawlowski's method.

The method used to obtain the paired plants was as follows: Beets in the 2-leaved stage were equally bisected between the leaves. The 2 halves of each plant were placed in vermiculite in compartments of small plastic trays, were watered with nutrient solution (1), and enclosed in plastic bags for several days to maintain high humidity. The trays were placed in either a greenhouse or plant growth chamber until established. They were then transplanted into soil in 8-inch pots.

The roots of the beets were sometimes shorter than normal with profuse growth of secondary roots at the tip. Figure 1 shows paired beets in an early stage of growth while Figure 2 shows larger paired beets with almost normal development of the tap root and secondary roots. These beets had the zone of small secondary roots along one side of the tap root only, rather

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Captions for figures on next page.

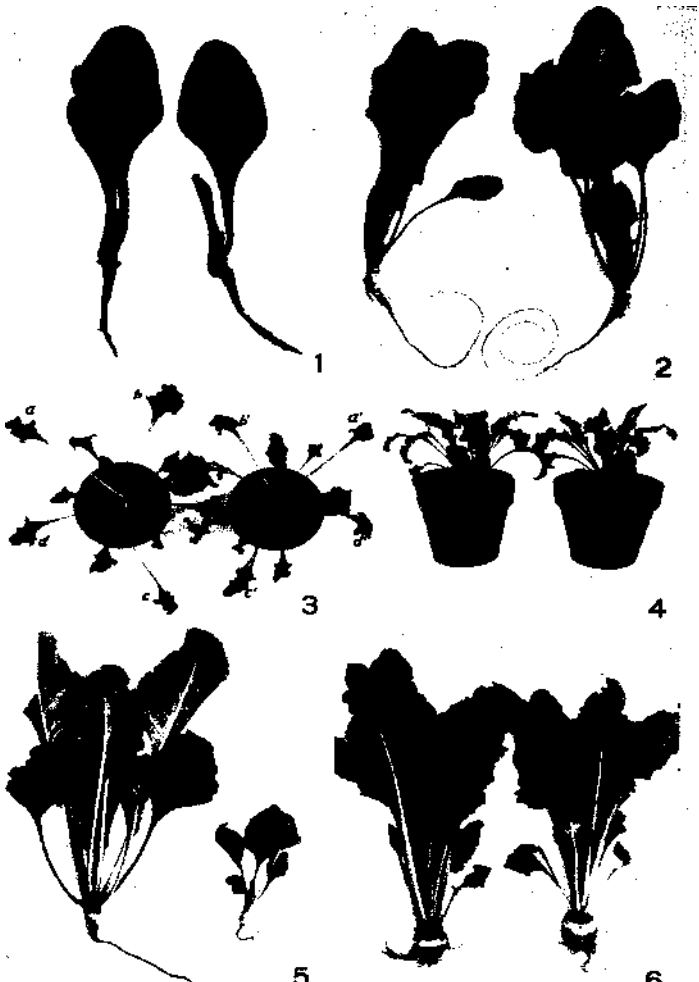
Figure 1.—Genetically identical plants from a seedling that was evenly bisected.

Figure 2.—Genetically identical beets with near normal root development.

Figures 3 and 4.—Two pairs of beets from bisected seedlings. The leaf development appears to be normal and almost identical in the paired plants. The plant on the left is the mirror image of the one on the right and leaves with the same letter are nearly identical.

Figure 5.—Genetically identical plants from a seedling that was not evenly bisected.

Figure 6.—Genetically identical plants showing the effect of peat moss containers on the development of the tap root.


1
2
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5
6

than along both sides. Most of the paired beets had almost identical leaf shape, leaf position, and longevity of leaves, and one small plant was the mirror image of the other (Figures 3 and 4).

The degree of success achieved in establishing the identical pairs varied from 5 to 20%. As some fungicides cause abnormal root development, the divided plants were not treated with fungicides and a few seedlings were lost due to root rot. If the seedlings were not split equally, the smaller plant died or developed more slowly (Figure 5).

In early trials beets were transplanted from the plastic trays to small peat moss pots containing soil. Then, when well established, the beets, still in the peat moss pots, were placed in soil in 8-inch pots. As the peat moss pots did not disintegrate as expected and affected the subsequent growth of the beet root (Figure 6), the plants in later trials were transplanted directly from the trays to soil in 8-inch pots.

The main advantage of this technique is that valid paired comparisons can be made with seedling beets. Thus, greater uniformity of results should be obtained and the replication necessary for detecting differences due to the treatment applied could be reduced. The identical beets should be useful for studying the effect of host plants on the development of insect populations under various conditions, the physiological changes during growth of sugar beets, and the influence of temperature, soil moisture, or soil fertility on young sugar beets.

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Selection for Speed of Germination in Sugar Beet[1]

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Rapid germination and uniform emergence of a field crop permit more timely cultural operations and may contribute to greater yields. Commercial varieties of sugar beets lack these traits in varying degrees. Within a given variety, seeds from individual plants vary markedly in speed of germination. This observation and Smith's (2)[3] report that the ability to germinate at low temperature is inherited have suggested that speed of germination may also be inherited. This paper reports a study of rapidity of germination within the broadbase variety of sugar beet US 401 initiated in 1955.

Methods and Materials

Twelve hundred sized seedballs, obtained from a mixed sample of seed, were used. Eighty seedballs were placed on a wire screen in contact with the surface of approximately 180 milliliters of a balanced nutrient solution (3) of 10.1-atmospheres osmotic pressure in each closed plastic box. The seeds germinated at room temperature. This method of germination will be referred to as the liquid-contact method. After 3 days, approximately 125 of the seedlings with the longest roots were transplanted to 3-inch pots. These plants were termed rapid germinators. At the end of 10 days, all the "ungerminated seedballs" were rinsed in tap water and transferred to blotters moistened with tap water. The last 125 seedlings which germinated were transplanted to 3-inch pots. These were termed slow germinators. Seedlings in this group germinated in a minimum of 16 days after the experiment was begun.

The seedlings were transplanted a second time to 6-inch pots and grown in the greenhouse; the rapid germinators remained there for about 12 weeks and the slow germinators for 6 to 10 weeks. All plants then were photothermally induced at approximately 48 F for 10 weeks. In early May, the rapid germinators were planted in an isolation plot approximately $1^{1/2}$ miles from a similar isolation plot of the slow germinators. Seedballs were harvested by individual plants in mid-August. The seed pro-

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[3] Numbers in parentheses refer to literature cited.

ductions resulting from open pollination were designated as polycross seed.

In September, polycross seed from 72 of the rapid germinators was compared with polycross seed horn 72 of the slow germinators. The seed from each plant was passed through a Boerner Sampler twice and a sample of 40 sized seedballs was prepared. With 4 exceptions, the whole seedballs ranged in diameter between 11/64 and 13/64 of an inch. The same method of germination was used as in the initial selection. Only the first seedling from a seedball was used in calculating germination percentages.

Samples of seeds from 6 plants, 2 rapid germinators, 2 with delayed germination, and 2 with both delayed and poor total germination, were chosen to determine the effect of the seedball on speed of germination. Eighty whole seedballs and 80 naked or true seeds of each were germinated by the liquid-contact method.

Root-elongation data for many of the samples were obtained by measuring the longest root from each seedball approximately 80 hours after the seedballs were placed in contact with the osmotic solution.

Repeated production of seed on clones having different seedball characteristics seemed to be the simplest method of determining the constancy of speed of germination in a given clone. For this test, seeds of a rapidly germinating polycross (Progeny 50232) were germinated by the liquid-contact method. The 18 most advanced seedlings were selected. Seedballs were harvested from each clone (mother root or stem cuttings) 4 times as follows: Greenhouse in 1956, greenhouse in 1957 with plants producing fruits at a mean temperature of *66* or 76 F, and in the field in 1957. Speed of germination of the seeds was determined by the liquid-contact method.

Results

Selection effect

The percentages of germination (Table 1) for the 3- and 5-day counts, expressed as means of 72 polycross progenies, were

Table 1.—Comparison of polycross seed from 72 plants of sugar beet variety US 401 selected for rapid germination and 72 plants selected for slow germination.

Speed of germination in initial selection	No. of parent plants	Percentage germination[1] in		
		2 days	3 days	5 days
Rapid	72	24.9	64.4	84.5
Slow	72	25.4	58.4	75.9
LSD 1%		N.S.	3.8	2.7

[1] Based on 2880 seedballs, 40 from each of 72 plants.

significantly higher for seeds produced by plants selected for rapid germination than for seeds produced by plants selected for slow germination. Under these experimental conditions, the total percentage of germination increases only a few percent when the period of germination is extended to 10 or more days. When the percentages of germination of seeds from the 144 plants were classified for 2- and 3-day counts, the progenies derived from the rapid-germinator polycrosses had a smaller number of slow germinators on the third day (Table 2).

Table 2.—Germination of seeds from individual plants of sugar beet variety *US* 401 selected for rapid germination compared with those selected for slow germination.

Percentage germination category	No. of parent plants in category of germination			
	2 days		3 days	
	Rapid[1]	Slow[1]	Rapid[1]	Slow[1]
80-100	2	3	14	21
60-79	4	4	38	20
40-59	9	14	12	13
20-39	22	15	6	9
0-19	35	36	2	9

[1] Initial selection.

Seedball effect

Although the data in Table 1 indicate that somewhat faster germination may be obtained by this method of selecting for speed of germination, the data in Table 2 suggest that it is relatively unsatisfactory without some modification. The selection procedure was based on the assumption that speed of germination is controlled by the embryo, but the data reveal that some slow germinators produced as high a percentage of rapid germinating seeds as those derived from the rapid germinators. The poor correlation between the speed of germination of a seed and the seeds produced on that plant at maturity suggested that the maternal tissues of the seedball might modify the germination response of the embryo. The data (Table 3) for the 6 samples reveal 3 facts, including the effect of the maternal tissues on germination. First, in 72 hours, the percentage of germination of seeds in whole seedballs ranged from 1.3 to 96.3. Second, in 72 hours, the naked seeds germinated equally well irrespective of how the seeds in the whole seedballs germinated. This would seem to be strong evidence for the profound influence that seedball characters may impose on the germination response of different progenies. Third, the germination percentage of naked seeds in 24 hours indicate an appreciable range in speed of germination among these progenies and suggest that the embryo may contain a heritable factor for rapid germination.

Table 3.—Effect of the seedball on germination of selected polycross progenies of sugar beet variety US 401 in nutrient solution of 10.1-atmospheres osmotic pressure.

Progeny	Percentage germination by hours[1]					
	Whole seedballs			Naked seeds[2]		
	48	72	120	24	48	72
50085	81.3	96.3	97.5	21.3	87.5	96.3
50216	92.5	96.3	97.5	17.5	65.0	88.8
50128	0.0	41.3	85.0 a	17.5	92.5	95.0
50182	2.5	21.3	85.0 a	33.8	93.8	96.3
50055	0.0	7.5	46.3 a	5.0	75.0	95.0
50174	0.0	1.3	12.5 a	32.5	88.8	96.3

[1] Based on 80 seedballs or naked seeds.
[2] Germination based solely on root elongation.
a Seedballs contained at least one mature seed, as determined by later examination.

Clone effect

The speed of germination performance of the 18 clones in the 4 cycles of seed production was as follows: Six were consistently slow, 5 were consistently fast, and 7 were intermediate in speed. Although the speed of germination of seeds produced by a given clone was not constant for the 4 cycles of production, almost without exception the clones did maintain their same relative rank. Apparently, the environment in which the seed developed and matured affected the absolute speed of germination, but essentially failed to alter the relative speed of germination among the clones.

Osmotic sensitivity

The progenies which germinated the earliest frequently had the longest roots, however, some progenies that germinated early had conspicuously shorter roots than others. Progenies of US 401 appear to be differentially sensitive to an osmotic pressure of **10.1** atmospheres.

Discussion

The great range in percentage germination in 2 days under an osmotic stress exhibited by seeds from individual plants of US 401 emphasizes the heterogeneity of this sugar beet variety. Germination performance of seeds of Foundation Inbred 169 which had been selfed for 5 generations also has indicated considerable variability in germination for this inbred.

The important role of the seedball in regulating speed of germination in sugar beet provides an explanation as to why a single selection for speed of germination which does not involve the female parent is ineffective. The initial selection procedure failed whenever the progeny had different seedball characteristics than the female parent. By employing reselection, Doxtator and

Finkner (1) were able to alter the speed of germination because the selection tended to select for a given type of seedball characteristic and eliminate the variants. The use of clones in the present study stabilized the seedball characteristics for the 4 cycles of seed production in different environments. The almost perfect relative rankings of the clones and the results of Doxtator and Finkner (1) suggest a genetic basis for speed of germination.

Since the seeds produced on a clone under different environments germinated at different rates, the role of environment in producing high-quality seed must be evaluated.

The liquid-contact method for germinating seeds, when used in conjunction with a solution of given osmotic stress, appears to apply selective pressure for two attributes. First, the ability to germinate against an osmotic stress, and second, the ability to germinate when the seedball is surrounded by a thin film of liquid. Under field conditions, seeds are forced to germinate under the osmotic stress of the surrounding soil solution.

If the germination response of the seeds from the 144 plants studied is assumed to be representative for US 401, then the potential progress through selection may be indicated. The average germination percentage in 2 days for seeds from the 144 plants was 25.1. In contrast, seeds from 5 of the plants germinated 90 percent in 2 days. If the plants producing the rapid-germinating seed were placed in isolation, permitted to polycross, and then subjected to reselection, a substantial improvement in speed of germination should be possible.

In 1960 at Michigan State University, T. E. Sedlmayr, in his doctoral thesis "Inheritance of Speed of Germination in Sugar Beets *(Beta vulgaris* I..)," reported that speed of germination is a heritable trait in sugarbeets.

Summary

Seedlings of sugar beet variety US 401 were selected on the basis of speed of germination by the liquid-contact method using a mineral nutrient solution of 10.1-atmospheres osmotic pressure. After a period of growth in the greenhouse and 10 weeks of photothermal induction, the rapid germinators were placed in an isolated seed plot and the slow germinators in a similar plot $1^{1/2}$ miles away. Seedballs were harvested by individual plants. Seeds from 72 rapid-germinator plants and 72 slow-germinator plants were compared for speed of germination by the liquid-contact method by using nutrient solution of 10.1-atmospheres osmotic pressure.

No consistent pattern of germination was obtained for either the rapid or the slow germinators. Seeds produced on the rapid-germinator plants averaged significantly faster; however, seeds obtained from some slow-germinator plants germinated as rapidly as seeds derived from the rapid-germinator plants.

Comparison of the germination of naked seeds with seeds in the whole seedballs confirmed that the differential speed of germination is largely regulated by the maternal tissues of the seedball itself.

Some differences in speed of germination were discernible between naked seeds derived from different plants.

The progenies exhibited a differential sensitivity to osmotic stress as measured by root elongation under 10.1-atmospheres osmotic pressure.

Acknowledgment

Valuable assistance and advice by my colleagues, G. J. Hogaboam, H. L,. Kohls and H. W. Bockstahler, are gratefully acknowledged.

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Comparison of Fluorescent and Incandescent Lamps for Promotion of Flowering in Sugar Beet Seedlings[1]

JOHN O. GASKILL[2]

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Incandescent-filament lamps are known to be superior to fluorescent lamps for use as a supplement to sunlight for the promotion of flowering in certain types of long-day plants including relatively mature sugar beets (2,3)[3]. Incandescent lamps have been used successfully in studies at Fort Collins, Colorado, as the sole source of light for very young sugar beet seedlings during the process of induction under low-temperature conditions—sometimes called vernalization—and as the source of supplemental light during the postinduction period (5,6,7). The seedling induction technique has been rather widely adopted by sugar beet breeders as a means of reducing the length of the life cycle and expediting the development of new varieties. Some of the induction installations are relatively large (1,4) and require considerable electricity for refrigeration as well as for illumination. A suitable light source producing less heat than the incandescent type would be desirable as a means of reducing refrigeration costs.

The investigations reported in this article were undertaken primarily to determine whether fluorescent lamps could be substituted, at least in part, for incandescent lamps during the induction treatment. Also in some experiments, the two types of lamps were compared during the postinduction period. The contrasting plant response to illumination by fluorescent lamps during the two developmental stages is of special significance and suggests a new concept with respect to the process previously referred to by the writer as photothermal induction. The 1959 results were presented at the 11th General Meeting of the American Society of Sugar Beet Technologists[4], but publication was postponed until additional data could be obtained.

[1] Report of investigations conducted by the Crops Research Division, Agricultural Research Service, U.S. Department of Agriculture, in cooperation with the Colorado Agricultural Experiment Station; publication approved by the Experiment Station Director as Scientific Series Article No. 860.

[2] Plant Pathologist, U.S. Department of Agriculture, Fort Collins, Colo. Assistance of Luther W. Lawson and Joseph A. Elder, Agricultural Research Technicians, in conducting the experimental work, is acknowledged.

[3] Numbers in parentheses refer to literature cited.

[4] An informal report, "Further studies on the use of artificial illumination during and after photothermal induction of sugar beet seedlings", presented by John O. Gaskill on February 4, 1960, Salt Lake City, Utah.

Material and Methods

Open-pollinated, relatively heterogeneous, biennial-type sugar beet varieties or strains with about average low-temperature induction requirements were used for all studies reported in this article. The commercial variety GW359 was used in experiments 1 and 2 and noncommercial material in experiments 3 and 4. Seed was planted in steamed soil in aluminum tubes 11/2 X 11/2 X 31/2 inches in size. The tubes were held initially in fiats on a bench in the greenhouse with moisture and temperature suitable for rapid growth. Incandescent lamps were used to furnish nightlong supplemental light throughout the preinduction growth period in all experiments except No. 1, in which no supplemental light was provided during that period. Soon after emergence, the plants were thinned to four per tube.

Between 9 and 14 days after planting, the tubes were transferred to a refrigerated induction chamber where continuous artificial illumination was provided in complete absence of sunlight. Temperature among the plants in each experiment, with the thermometer bulb about 1/2 inch above the soil and exposed to the direct rays of light, fluctuated between 6° and 9° C, approximately, and averaged about or slightly above 7°. The air was circulated continuously in the induction chamber by means of fans, and air flow among the seedlings of each group was regulated in such a manner as to counteract the differing heating effects of the experimental light units. By this means, the differences among the temperature averages recorded for the respective treatments, within any given experiment, were held to less than 0.5°. In some experiments, comparable, noninduced control plants Avere included. These plants were started in the greenhouse with timing such that they were about the same size as the induced plants when the latter were ready for removal from the induction chamber.

At the end of the induction treatment, the 4-plant cluster in each tube was transplanted in soil in one 6-inch pot. The pots were placed in the greenhouse or outdoors, with or without supplemental light, as described for the respective experiments. The greenhouse was cooled artificially during hot weather, and temperature conditions favorable for rapid plant growth were maintained. Plants located outdoors were covered with 1/4-inch-mesh wire screen for protection against hail. In certain instances the postinduction treatments were discontinued and the plants were returned to the greenhouse, a short time before the final counts were made, as a safeguard against freezing injury. Artificial fertilizers were applied as needed during the postinduction

period. The plants were examined periodically and a record kept of initial flowering. Ordinarily each plant was removed from the pot when recorded as flowering.

The Gro-lux fluorescent lamp was developed by Sylvania Lighting Products, and all other fluorescent lamps used were products of the General Electric Company. Only the Gro-lux, with energy emissions concentrated in the red and blue regions of the spectrum, had been designed especially for use in plant-growth chambers. Nonreflecting shields were used as needed in the induction chamber, field, and greenhouse to protect each treatment group from extraneous light. An ordinary foot-candle meter was employed for measuring light intensities at or near the level of the plant foliage. These measurements were made merely for reference purposes, in recognition of the fact that, for the various types of lamps used, the data do not accurately reflect either total radiant energy or relative biological effectiveness (3).

Experiment 1

Experiment 1 was designed primarily to compare types of light in the induction chamber. The following light units were used: (a) one ordinary, 100-W, 120-V, inside-frosted, incandescent lamp in a medium-depth reflector; and (b) two 20-w, 24-inch, starter-operated, deluxe warm white fluorescent lamps in a single reflector. Placement of the light units, light shields, and four groups of sugar beet seedlings was such that each group received about 62 ft-c at the center, with percentages of light from the fluorescent source as follows: 0, 10, 84, and 100 for treatments 1-1, 1-2, 1-3, and 1-4, respectively. The intermediate percentages, 10 and 84, are approximations. A set of noninduced control plants was assigned the treatment No. 1-5.

At the termination of the induction period (47 days, ending on July 21, 1959), each of the five sets of plants was subdivided into five comparable groups which were subjected to the following respective conditions throughout the postinduction period:

P-1: In greenhouse; nightlong supplemental light supplied by one 100-w incandescent unit (same as that used in the induction chamber) suspended 3 feet above the surface of the soil in the pots. The average light intensity at night, 9 inches above the soil, was approximately 37 ft-c.

P-2: In greenhouse; nightlong supplemental light supplied by one 40-w fluorescent unit (same as that used in the induction chamber) suspended 3 feet above the soil. The light intensity measurement comparable to that of treatment P-l was 30 ft-c

P-3: In greenhouse; no supplemental light.
P-4: Outdoors; supplemental light same as for P-l.
P-5: Outdoors; no supplemental light.

Experiment 2

The light units listed below were placed at a uniform distance above separate (shielded) groups of seedlings in the induction chamber with approximately 19 inches from lamps to foliage. Each light unit consisted of one reflector with one inside-frosted, incandescent lamp or two 24-inch, fluorescent lamps. Type of lamp, total wattage for the unit, and approximate light intensity at foliage level for the five respective treatments were as follows:

1-6: Incandescent, 75 w, 87 ft-c.
1-7: Incandescent, 40 w, 29 ft-c.
1-8: Fluorescent (deluxe warm white), 40 w, 61 ft-c.
1-9: Fluorescent (standard cool white), 40 w, 92 ft-c.
1-10: Fluorescent, purple (1 red lamp and 1 blue lamp), 40 w, 27 ft-c.

The chief purpose of this experiment was the comparison of light units of equal wattage without regard to the light intensities produced. The 75-w unit was included in order to obtain supplementary information. There were two lengths of induction—4 and 8 weeks—both ending on August 10, 1960. On that date, all plants were transferred to the greenhouse where nightlong supplemental illumination was provided by means of incandescent lamps.

Experiment 3

The Gro-lux fluorescent lamp was compared with the usual type of incandescent lamp, for induction purposes, with equal light intensities at foliage level. The Gro-lux light unit included two 20-w lamps, and one 60-w incandescent lamp was used in the other unit. Each light source was so placed as to provide approximately 68 ft-c to the intended group of plants. The induction treatment (10 weeks) ended on June 6, 1962. The plants subsequently were held in the greenhouse and supplied with incandescent light throughout each night. Noninduced control plants were not included in this experiment. However, the sugar beet strain used was known to require a low-temperature induction treatment in order to produce a high percentage of bolting.

Experiment 4

Evaluation of the Gro-lux fluorescent lamp as the source of supplemental lisrht during the postinduction period was the principal objective of experiment 4. All seedlings were given 12 weeks' induction under Gro-lux lamps, ending on June 21,

Table 1.—Flowering of sugar beet seedlings as affected by type of artificial illumination during the induction and postinduction periods (experiment 1).

Postinduction			Induction		No. of plants	Elapsed time after induction and cumulative % of plants flowering[b]				
Treat. no.	Location	Supplemental light source[a]	Treat. no.	Light source[a]		5 wks.	6 wks.	8 wks.	10 wks.	14 wks.
P-1	Greenhouse	Incandescent	I-1	Incandescent, only	24	33	54	75	75	83
			I-2	Largely incandescent	24	21	54	88	88	92
			I-3	Largely fluorescent	24	33	58	71	79	88
			I-4	Fluorescent, only	24	4	46	67	67	79
			I-5	Control (not induced)	24	0	0	0	4	4
P-2	Greenhouse	Fluorescent	I-1	Incandescent, only	24	0	0	0	0	4
			I-2	Largely incandescent	24	0	0	0	0	0
			I-3	Largely fluorescent	25	0	0	0	0	0
			I-4	Fluorescent, only	24	0	0	0	0	0
			I-5	Control (not induced)	24	0	0	0	0	0
P-3	Greenhouse	None	I-1	Incandescent, only	24	0	0	0	4	4
			I-2	Largely incandescent	24	0	0	0	0	0
			I-3	Largely fluorescent	24	0	0	0	0	0
			I-4	Fluorescent, only	24	0	0	0	0	0
			I-5	Control (not induced)	24	0	0	0	0	0
P-4	Outdoors	Incandescent	I-1	Incandescent, only	24	25	63	79	83	88
			I-2	Largely incandescent	24	13	58	79	79	88
			I-3	Largely fluorescent	24	25	67	96	96	100
			I-4	Fluorescent, only	24	17	67	83	88	92
			I-5	Control (not induced)	24	0	8	13	13	17
P-5	Outdoors	None	I-1	Incandescent, only	24	0	4	8	8	8
			I-2	Largely incandescent	24	0	0	0	0	0
			I-3	Largely fluorescent	24	0	0	0	4	4
			I-4	Fluorescent, only	24	0	0	0	0	0
			I-5	Control (not induced)	24	0	0	0	0	0

[a] The flourescent lamps were deluxe warm white.
[b] Induction treatments (47 days) ended on July 21, 1959.

1962. On that date the plants were potted, divided into four identical sets of 10 pots (40 plants) each and placed outdoors, each set arranged in a compact group. Nightlong supplemental illumination was supplied to the respective groups by means of the following lamps in appropriate reflectors: (a) one 75-w, incandescent lamp; (b) two 20-w, Gro-lux fluorescent lamps; (c) two 20-w, Gro-lux lamps and one 15-w, incandescent lamp; and (d) none. The heights of the light units were adjusted at the beginning of the postinduction period to provide 42 ft-c light intensity at night, as measured at the center of each group, 2 inches above the surface of the soil in the pots—i.e. at the approximate level of the plant foliage. Iamp-to-soil distances were 32, 31, and 34 inches for groups a, b, and c, respectively. The arrangement of the light units remained unchanged, and consequently light intensity at foliage level increased as a result of plant growth. The incandescent lamp furnished about 11 percent of the light recorded for group c.

Results

Experiment 1

As shown in Tables 1 and 2, plants receiving- supplemental light from incandescent lamps during- the postinduction period responded about alike to the four respective induction light treatments. Flowering percentages after the 5th week of the postinduction period were practically identical for the incandescent and fluorescent induction light sources (I-1 and I-40) Corresponding percentages for the mixtures of the two types of light (I-2 and I-3) tended to be higher but the differences were relatively small.

The contrasting effects of supnlemental light from incandescent and fluorescent sources, supplied during the postinduction period, were particularly striking (Table 1, treatments P-l and P-2). Under the fluorescent source, only one plant flowered in

Table 2.—Flowering of sugar beet seedlings as affected by type of illumination during the induction period (experiment 1) [a].

Treat. no.	Light source	No. of plants	Elapsed time after induction and cumulative % of plants flowering				
			5 wks.	6 wks.	8 wks.	10 wks.	14 wks.
I-1	Incandescent, only	48	29	58	77	79	85
I-2	Largely incandescent	48	17	56	83	83	90
I-3	Largely fluorescent	48	29	63	88	88	94
I-4	Fluorescent, only	48	10	56	75	77	85
I-5	Control (not induced)	48	0	4	6	8	10

[a] Summary of results for treatments P-l and P-4, Tab'e I (plants receiving supplemental light from incandescent lamps during the postinduction period).

a population of 95 induced seedlings (induction treatments I-1 through I-4). In the corresponding population under the incandescent lamp, 85 percent of the plants flowered. As stated under Material and Methods, the light intensity was somewhat lower under the fluorescent source. However, on the basis of other evidence[5], the extreme contrast in flowering percentage cannot be attributed to differing intensity per se. It is of interest to note that, where no supplemental light was provided during the post-induction period in the greenhouse (P-3), the results were the same as in the plant group receiving fluorescent light (P-2).

Experiment 2

The summarized flowering results obtained from experiment 2 are presented in Table 3. Several LSD values are shown as an aid in appraising differences. LSD values are not given in those instances where variation was seriously restricted by the frequent occurrence of percentages of 0 or 100.

Table 3.—Flowering of sugar beet seedlings as affected by type of illumination during inductio periods of different lengths (experiment 2).

Induc.ª time (weeks)	Treat no.	Light source	No. of plants	Elapsed time after induction and cumulative % of plants flowering				
				4 wks.	5 wks.	6 wks.	9 wks.	12 wks
4	I-6	Incandescent (75 w)	63	21	41	56	63	67
	I-7	Incandescent (40 w)	64	5	20	44	53	58
	I-8	Fluores. (del. warm white, 40 w)	64	0	6	17	41	44
	I-9	Fluores. (Stand. cool white, 40 w)	63	2	6	17	29	37
	I-10	Fluores. (purple, 40 w)	60	0	5	13	28	32
		LSD (5-percent point)				16	16	17
8	I-6	Incandescent (75 w)	63	41	68	87	94	95
	I-7	Incandescent (40 w)	63	14	51	70	87	89
	I-8	Fluores. (del. warm white, 40 w)	63	5	41	62	89	94
	I-9	Fluores. (stand. cool white, 40 w)	64	6	47	66	91	95
	I-10	Fluores. (purple, 40 w)	64	3	27	48	73	81
		LSD (5-percent point)			17	19		NS
0	I-11	Control (not induced)	64	0	0	0	5	5

Induction treatments ended on August 10, 1960.

For the plants receiving 8 weeks' induction, a rather substantial lag in flowering may be observed for the purple fluorescent light source (treatment I-10) and a strong tendency toward earlier flowering is shown for the 75-w incandescent source (I-6), in comparison with the other three treatments. However, these differences narrowed with time and had largely disappeared when the final counts were made. The fact that a minimum of 84 percent flowering occurred among the five sets of plants (I-6 through I-10) receiving 8 weeks' induction, as

[5] Unpublished results.

contrasted with 5 percent flowering for the noninduced set, is of special interest. Obviously some form of low-temperature induction treatment was required for the initiation of flowering, but the type of light used appeared to be relatively unimportant except as related to earliness of flowering. In this connection, it seems possible that the early- and delayed-flowering tendencies shown for treatments I-6 and I-10, respectively, may have been associated with the vigor of the plants at the end of the induction treatment. At that time, the plants of I-6 were the most vigorous and those of I-10 were the least vigorous of the five treatment sets.

Flowering percentages no higher than those resulting from 4 weeks' induction in this experiment ordinarily would be considered unsatisfactory for sugar beet breeding purposes, but some of the results are of academic interest. The 75-w incandescent source was particularly outstanding, and even the 40-w incandescent lamp appeared to be more effective than the white fluorescent units, taken together, as judged on the basis of final flowering percentages as well as earliness of flowering.

Experiment 3

The results from experiment 3, a simple comparison between two types of light used in the induction chamber, are summarized in Table 4. Final flowering percentages for the two treatments were practically identical. The lag in flowering indicated for the plants receiving incandescent light is attributed largely to two pots which became temporarily water-logged, retarding the rate of plant development. Thus it was concluded that the difference between treatments in flowering response, if any, was negligible. With the exception of the water-logged pots, general plant vigor for the two treatments was about alike throughout the experiment.

Table 4.—Flowering of sugar beet seedlings as affected by type of illumination during the induction period (experiment 3).

Light source	No. of plants	Elapsed time after induction and cumulative % of plants flowering [a]					
		4 wks.	5 wks.	6 wks.	8 wks.	10 wks.	14 wks.
Incandescent	42	2	60	76	93	93	98
Fluorescent (Gro-lux)	45	0	51	91	100	100	100

[a] Induction treatments (10 weeks) ended on June 6, 1962.

Experiment 4

The results obtained for the fluorescent lamps in experiment 4 (Table 5 and Figure 1) agree with the results from experiment 1 in demonstrating very decisively the ineffectiveness of fluorescent lamps when used alone, as a supplement to sunlight,

Figure 1.—Response of induced sugar beet seedlings to two types of supplemental illumination during the postinduction period: top, incandescent; bottom, Gro-lux fluorescent. The photograph was taken on July 27, 1962, 36 days after transfer of the plants from the induction chamber into the open.

during the postinduction period. A light mixture consisting of approximately 89 percent from fluorescent lamps and 11 percent from an incandescent lamp was much more effective than light from a fluorescent unit alone, but was clearly inferior to light of equal intensity furnished solely by an incandescent lamp.

Table. 5.—Flowering of sugar beet seedlings as affected by type of supplemental illumination during the postinduction period (experiment 4).

Light source	No. of plants	Elapsed time after induction and cumulative % of plants flowering [a]				
		5 wks.	6 wks.	8 wks.	10 wks.	14 wks.
Incandescent (75 w)	40	38	70	95	98	98
Gro-lux fluorescent (40 w)	40	0	3	15	20	28
Gro-lux fluores. (40 w) + incand. (15 w)	40	5	20	53	60	70
None	40	3	10	18	20	23

[a] Induction treatment (12 weeks) ended on June 21, 1962.

Discussion and Conclusions

The ineffectiveness of fluorescent lamps, as used during the postinduction period in these studies, is in agreement with results reported by Borthwick and Parker (2) for relatively mature, thermally induced sugar beet roots. The superiority of incandescent lamps for the promotion of flowering in such sugar beets and in certain other long-day plants has been attributed by Downs et al. (3) to the far-red component of the radiation emitted by those lamps—a feature lacking in the emission of the fluorescent lamps studied. According to information furnished by the manufacturers, neither the deluxe warm white nor the Gro-lux fluorescent lamp emits appreciable far-red energy. Consequently, the ineffectiveness of those lamps during the postinduction period in the current studies was to be expected.

In view of these results it is especially significant that, for the process of induction with at least 47 days' exposure to low temperature, each of those two types of fluorescent lamps was essentially equal to incandescent lamps as measured by the flowering response. Furthermore, it is noteworthy that a third type of fluorescent lamp (standard cool white) also was about equal to those two, and that a fourth type (purple) was nearly so.

From these results it is apparent that the function of artificial light during the induction treatment is quite different from its function during the postinduction period with respect to the mechanisms involved in the flowering process. It is postulated that the physiological or other changes conducive to flowering, occurring during the induction period, are basically due to low temperature, and that light serves primarily as the source of energy for photosynthesis and growth. If this is true, any type of light that is suitable for photosynthesis and growth should be relatively satisfactory for use in the induction treatment. In this connection it is pertinent that field-grown sugar beet roots commonly are induced by prolonged cold storage in complete

darkness—a thermal-induction process. Seedling induction, as described in this article, probably involves the same mechanisms with the carbohydrate utilized in growth and other processes being derived largely from the leaves directly instead of from a fleshy tap root. Accordingly, it appears that the term "photothermal", formerly used with reference to seedling induction, is not strictly accurate.

It should be emphasized that the tentative conclusions as stated in the preceding paragraph pertain to sugar beet material of the type used in these studies—i.e., varieties or strains having about average thermal induction requirements. These conclusions may not necessarily apply with equal force to bolting-resistant sugar beet types.

Summary

A series of experiments was conducted for the purpose of comparing the effects of fluorescent and incandescent-filament lamps on flowering of sugar beet seedlings. One phase of these studies involved comparisons between types of lamps used during a prolonged period of low temperature, the induction treatment. In the other phase, comparisons were made between types of lamps used to provide light at night, supplementing sunlight, throughout the postinduction period.

After 9 to 14 days' growth in the greenhouse, seedlings were held in a refrigerated room for varying time intervals, usually ranging from about 7 to 12 weeks, at a temperature of approximately 7° C. Four types of fluorescent lamps were compared with incandescent lamps, and certain mixtures of light from fluorescent and incandescent sources also were included. The lamps were operated continuously, and sunlight was completely excluded.

At the end of the induction period, the seedlings were transferred to the greenhouse or outdoors where the percentage of plants flowering in each population was recorded periodically. Nightlong illumination by incandescent lamps throughout the postinduction period was standard procedure in all experiments. In certain instances, comparisons were made between such supplemental light and that provided by fluorescent lamps.

The results clearly showed that the two types of fluorescent lamps, used as sources of supplemental light during the postinduction period, were wholly ineffective in promoting flowering. Incandescent lamps were highly effective. On the basis of reports by other investigators, these contrasting responses were attributed to the presence of a far-red component in the radiation emitted by incandescent lamps and its absence in the radiation from fluorescent lamps.

A rather wide range of types of fluorescent lamps, as well as the incandescent type serving as the standard, were highly effective in the low-temperature induction process. That is to say, a high percentage of the seedlings, subjected to such conditions for a sufficient length of time and given proper environment thereafter, flowered within a relatively short period after the end of the induction treatment.

From these contrasting results, it was concluded that the function of artificial light, with respect to the promotion of flowering in young sugar beet seedlings having average low-temperature induction requirements, is distinctly different in the induction and postinduction periods. It is postulated that flowerage-promoting changes, occurring in such seedlings during the induction process, are primarily due to low temperature and that the principal function of light at that time is to provide energy for photosynthesis and growth.

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Application of the Shortcut Method for Estimating the Standard Deviation

Statistics have helped the plant breeder to better understand the biological material with which he works. Often times simple statistics such as the means or standard errors are sufficient to obtain an insight into certain creeding materials. In many sugar beet breeding programs several thousand mother roots are selected each year. Each beet usually is given a number, weighed and tested for percent sugar. The resulting data are recorded and the general mean calculated. The mass selection method was refined by Powers (2)[2] when he developed the unit-block method of selection. Powers' method utilized inbreds and/or F_1 hybrids within each unit-block to measure the environmental variation. A modification of this method would be to harvest several mother beets within a unit-block, obtain the block mean for the characters being studied and note the range.

By using the range and the number of observations an estimated standard deviation could then be calculated by utilizing the formula, s/Range = mean ratio, or s = range X mean ratio. This shortcut method is described in Snedecor's "Statistical Methods", Table 2.2.2, on pages 38-44 (3). This procedure was used by Finkner et al. (1) in selecting mother beets for high and low aspartic acid and similarly for glutamine content. Once the standard deviation has been determined, the degree of selection pressure can be applied by choosing the beets which are beyond the mean by one or two standard deviations. The variances also are readily calculated by squaring the standard deviation.

The reliability of this method is shown in Table 1 for aspartic acid where it is compared with the actual calculation of the standard deviation. Good agreement between the two methods was found.

An example of how this shortcut method was used is shown below. There were 22 beets selected from unit-block, A-l, and analyzed for aspartic acid. The mean of the 22 beets for aspartic acid content was 0.18. The known range was 0.34 -0.08 = 0.26. The range (0.26) was then multiplied by the "ratio mean" which was found in Snedecor's Table 2.2.2. The "ratio mean" for 20 beets was .268. Therefore, the equation becomes 0.26 X .268 = the standard deviation of .070. The usual method of calculating

Table I.—Comparison of two different methods for calculating the standard deviation.

Unit blocks	s/Range = Mean Ratio	$\sqrt{\frac{\Sigma(x)^2 - \frac{(\Sigma x)^2}{n}}{n-1}}$
A-1	0.070	0.063
A-2	0.085	0.078
A-3	0.066	0.074
A-4	0.146	0.136
A-5	0.085	0.088
A-6	0.088	0.087
A-7	0.103	0.124
A-8	0.072	0.096
A-9	0.145	0.131
A-10	0.090	0.098
A-11	0.104	0.116
A-12	0.113	0.118
Mean	0.097	0.101

the standard deviation was round to be .063. The 2 estimates are very close to each other.

This simple, quick, reliable method of estimating the standard deviation can be a valuable guide in applying selection pressure for any characteristics being studied.

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Beta Macrorhiza Stev.

R. K. OLDEMEYER[1]

Received for publication April 16, 1963

There is record of only one introduction of *Beta macrorhiza* Stev. to the United States prior to World War II. G. H. Coons (1)- reports that he received a sample of seed in 1936 from N. E. Vavilov, the famous Russian plant breeder. Plants were grown in the USDA greenhouse in 1937 and 1938, but this accession of *B. macrorhiza* failed to flower and was subsequently lost when the research work of the Sugar Plants Section of the USDA was transferred to Beltsville, Maryland.

Transfer of germ plasm to sugar beets from the section *Corollinae* Tr. of the genus *Beta* has not been successful using *B. lomatogona* F. & M.[3], *B. intermedia* Bunge or *B. trigyna* W. & K. (2). Apparently there is little homology of the chromosomes of species of *Corollinae* with the chromosomes of *B. vulgaris;* F_1 hybrids between them are highly sterile. Progeny resulting from backcrossing are of two classes: (a) those which resemble the recurrent parent and are fertile; and (b) those which are intermediate morphologically and sterile (2). This backcross behavior indicates, most probably, that the only viable gametes are ones containing a complete set of chromosomes from one or the other of the parental species.

There are no direct reports or descriptions of hybrids between *B. macrorhiza* and sugar beets. Zossimovitch (3) refers to such hybrids and indicates that their genetic behavior confirms his belief that *B. macrorhiza* of the *Corollinae* species is phylogenetically most closely related to *B. vulgaris.* This information indicates that if germ plasm of a *Corollinae* species is to be transferred to *B. vulgaris,* hybrids with the diploid (2n = 18) *B. macrorhiza* should offer the best chance of success.

Unsuccessful attempts to transfer germ plasm from *B. trigyna, B. lomatogoma* and *B. intermedia* prompted a search by plant breeders of Great Western Sugar Company for an accession of *B. macrorhiza.* In April, 1956, the Great Western Sugar Company received a few seeds of an accession of seed from the USDA which was indicated to be *B. macrorhiza.* These seeds came from Russia through Dr. Henrik Bögh of Børkoy, Denmark. Plants grown from this seed did not resemble the taxonomic description of

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[2] Numbers in parentheses refer to literature cited.

[3] Personal communication from Dr. Helen Savitsky. USDA, Sal inns, Calif.

B. macrorhiza, and after Dr. Gerald Coe[4] of the USDA indicated the plants had 36 chromosomes, further study of this accession was discontinued.

In March 1957, H. E. Brewbaker[5] wrote to the Director of the Tiflis Botanical Gardens, U.S.S.R., requesting seed of *B. macrorhiza.* The reply (in Russian) indicated that the species was not cultivated there.

The reply to another contact in June, 1957, to The Sugar Research Station at Keiv, U.S.S.R., requesting *B. macrorhiza,* (in English) from Dr. I. A. Sizov, Director of the Institute of Plant Industry, Leningrad, indicated that seed was being sent. Fifty grams of seed was subsequently received in December of that year. Translation of the shipping slip (in Russian) indicated that this race of *B. macrorhiza* had been originally collected in the region of the Karhas Mountains. (In the letter of transmittal, Dr. Sizov indicated they would be glad to receive seed of the *Patellares* species; seed of the three *Patellares* species was subsequently sent to Russia via diplomatic channels.) Small quantities of the *B. macrorhiza* seed were distributed to sugar beet breeders throughout the United States.

The seed was large, very horny and had the characteristics of the taxonomic description. Plants were grown from the seed, but it was soon evident that the parental plants had been badly outcrossed with other species; however, a few seedlings were typical of the description given for *B. macrorhiza.* The leaves are large, broad and obtusely ovate with the heartshaped base and the lobes curved upward. The left plant of figure 2 is characteristic. The petioles in the center of the rosette are intensely red.

Plants were grown in pots in the greenhouse continuously from the fall of 1957 through the winter of 1958-59. The plants remained vegetative although conditions at times were proper for vernalization of *B. vulgaris.* Remnant seed from the USDA accession from Denmark was germinated and a few plants typical of *B. macrorhiza* were selected. All plants suspected of being *B. macrorhiza* and others were transplanted out of doors in the summer of 1959. In January 1960, the plants were chopped from the icy soil and were forced, in pots, in the greenhouse.

The plants flowered and the identity of the pure species was confirmed. After the pure species was positively identified, it

[4] Personal communication from Dr. Gerald Coe, USDA, Beltsville, Md.
[5] Former director of the Great Western Sugar Company Agricultural Experiment Station, Longmont, Colo.

Figure 1.—A typical *Beta macrorhiza* plant in flower, growing in an 8-inch pot.

was obvious that *B. macrorhiza* would not easily be mistaken for other *Corollinae* species at any stage of development. Only the typical plants (see Figure 1) were fertile. Reciprocal pollinations were made between sugar beets and *B. macrorhiza* by introducing pollen into bags covering the flowering branches. Many seeds were harvested from mother plants of both species. A few seedlings were produced by the seeds from sugar beet but these were subsequently proven to have resulted from selfing. Scarification of seedballs from *B. macrorhiza* with a razor blade revealed that few embryos developed to normal seeds. The few plants produced had resulted from self-pollination.

Zossimovitch stated the roots of *B. macrorhiza* were white and not red as observed by Stevens in his original description. To check the descriptions, several seedlings which had resulted from sib or self-pollination were sacrificed and the pencil sized roots were split longitudinally. The roots are very dark pink fleshed from the crown down about two inches where there is an abrupt transition to white.

The *B. macrorhiza* plants were again transplanted out-of-doors and were brought back to the greenhouse in January 1962. To allow freer pollination, four individual plants of *B.*

Figure 2.—*Beta macrorhiza* seedling, left, and seedling suspected of being a *B. macrorhiza* X sugar beet hybrid, right, both 4 months old and growing in 6-inch pots.

macrorhiza in flower were placed in four different greenhouses in which sugar beet plants were blooming. Seed was collected from each wild plant individually. After the seed was scarified, about 30 plants resulted, of which only one may be a hybrid (right pot, Figure 2).

The one seedling suspected of being a hybrid was not originally thrifty but became more vigorous with age. At four months it resembled a sugar beet plant grown under the same conditions. However, the leaves of the hybrid were shorter and perhaps thicker and much lighter green than the *B. macrorhiza* plants the same age.

Hybridization between *B. macrorhiza* and *B. vulgaris* appears difficult. It remains to be seen if germ plasm can be transferred to sugar beets even after hybrids are produced.

Summary

Seed of *Beta macrorhiza* was received directly from Russia by The Great Western Sugar Company. After a number of attempts at hybridization, only one plant was produced which is suspected of being a hybrid.

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The Influence of Factors Other than Soluble Phosphorus in the Nutrient Medium on the Phosphorus Content of Sugar Beet Plants[1]

JAY L. HADDOCK AND BARREL M. STUART[2]

Received for publication May 8, 1963

In spite of extensive literature reviews (10) and investigations of problems of soil phosphorus availability to plants, no widely accepted theory exists which adequately accounts for the many reported observations. Arnon (1)[3] cited the influence of ammonium and nitrate ions on phosphate absorption by barley plants to support his general theory that the absorption of phosphate would be expected to be depressed by the presence of high concentration of rapidly absorbable anions and enhanced by an increast in the concentration of rapidly absorbable cations. Pratt and Thorne (12) concluded that availability of phosphates is entirely a function of their solubility from pH 4.0 to 7.0 and that from pH 7.0 to 8.0 the dominant factor in availability is solubility, with physiological availability of minor importance.

Nightingale (8) stated that while specific information has not been found on how plant nutrients behave, ample nitrate supplies invariably repress phosphorus uptake. Pirson (11) observed that it has not yet been clarified whether an excess of an element causes disturbances which represent direct and specific consequences of this particular excess within the cell or whether the results are dependent upon the exclusion of another element, according to the pattern of competitive inhibition.

McGeorge (6) believes that pH of the soil solution, or more specifically of the root-solution contact interface, is an important factor in phosphorus uptake in calcareous soils. He emphasizes that at pH 6.0 more than 80% of the ionized soluble phosphorus is in the form H_2PO_4 and 17% is in the HPO_1 form. At pH 7.0 only 30% of the soluble phosphorus is in H_2PO_4 and 70% in the HPO_4 form. He shows that when the same concentration of phosphorus is present in the growth medium, the rate of phosphorus uptake is much greater at pH 6.0 than at pH 7.0. He also recognizes that solid $CaCO_2$ plays an important role in both solubility of phosphate and its assimilation by the plant.

[1] Contribution from Southwest Branch, Soil and Water Conservation Research Division, Agricultural Research Service, USDA, in cooperation with the Utah Agricultural Experiment Station.

[2] Research Soil Scientist and Soil Scientist respectively, USDA, Logan, Utah.

[3] Numbers in parentheses refer to literature cited.

Students concerned with availability of phosphorus in the soil have tried to relate the concentration of soluble phosphorus in soil solution to plant growth. Whitney and Cameron (15), by centrifugal displacement of soil solution, found that phosphorus concentration based on dry soil ranged from 0.46 to 0.53 ppm. Morgan (7) using oil displacement found similar results, 0.36 to 1.5 ppm of phosphorus on a dry basis. Parker and Pierre (9) observed that corn achieved maximum growth when culture solutions contained 0.13 ppm of phosphorus. Growth was very good at concentrations as low as 0.05 and good at 0.03 ppm of phosphorus. Dean and Fried (3) stated that studies have shown growth of plants to be impeded when 0.1 ppm or less of phosphorus is in solution, but that soils with displaced solutions containing less than 0.1 ppm support normal crop growth.

The experiment considered here was designed to determine the nutrient concentration and balance most suitable for the growth of sugar beet plants. In the course of this study it was observed that sugar beet plants growing in nutrient cultures similar in phosphorus concentration, produced plant tissue varying widely in phosphorus composition. The data in this paper are given to show the extent of these variations and to attempt an explanation as to some of the probable causes for variation in phosphorus content of plant tissue.

Methods and Procedure

Ten different nutrient cultures were studied. These were largely modifications of Hoagland's nutrient solution number 1 at one-half strength (4). Commercial monogerm variety SL 126 sugar beets were grown in cans of ten-gallon capacity filled with No. 2 vermiculite. Five small holes were punched in the bottom of each can to provide adequate drainage. The cans were then buried in field soil to within 1 inch of the top rim in order to maintain growing root temperatures comparable to field conditions. From 2 to 3 inches of coarse gravel had been nlaced below the cans to facilitate drainage and spread of water. In no instance did the plant rootlets protrude throueh the holes in the bottom of the cans. Twenty-five seeds were planted April 15, 1960, in each can. On Tnne 29 these were thinned to leave a final stand of three plants per can.

The nutrient concentrations in each of the ten nutrient cultures as prepared, and the mean seasonal pH and phosphorus concentrations in the drainage solutions from each treatment are shown in Table 1. While no potassium Avas added to Low K treatment, the vermiculite provided about 15 ppm K in solution.

Table 1.—Nutrient concentration in various nutrient solutions, 1960.

No.	Description	Parts per million of various nutrients*						P	pH	
		NO_3-N	NH_4-N	K	Ca	Mg	Na	Nut. sol.	** Drain.	Nut. sol.	** Drain.
1	Check (1/2 Hoagland's)	105	..	110	145	50	18	15	5.4	7.6	8.0
2	1/2 K	105	..	55	165	50	18	15	4.1	7.7	8.0
3	Low K	105	...	15	205	50	18	15	3.4	7.6	8.1
4	1/2 N	70	...	110	145	50	18	15	5.0	7.8	8.1
5	1/4 N	30	.	110	145	50	18	15	5.5	7.7	8.0
6	NO_3 + NH_4	105	25	110	145	50	18	15	8.0	7.3	7.6
7	1/2 P	105	..	110	145	50	18	7.5	2.4	7.9	7.9
8	NH_4	.	75	110	145	50	18	15	2.9	7.4	6.0
9	1/2 Ca + 1/2 Mg	105	.	110	100	30	115	15	4.1	7.7	8.0
10	Check (Same as No. 1 except 1/2-N 9/1 to 10/1 No-N 10/1 to 10/15)	105	.	110	145	50	18	15	4.5	7.6	8.0
	L5D 6i .01								1.4		

* Minor elements were provided in all nutrient solutions at the following concentrations: B=0.25, Nu = 0.25, Zn = 0.028, Cu=.01, Mo—0.004, and Fe = 4.5 ppm.

* *Mean of 22 samplings of leachings.

One gallon of nutrient solution was applied to each can daily except during mid-July and August when one and one-half gallons were used. In the latter instance three quarts of solution were applied in early morning and three quarts about 2 PM daily.

Leaf blade and petiole samples were obtained, at two week intervals beginning July 1, one from each plant on eight sampling dates. These plant tissues were rinsed in distilled water and dried rapidly at 70° C, ground to pass a 40-mesh screen, and examined chemically by standard procedures. Phosphorus was determined by the Barton (2) procedure.

Experimental Results

Yield of sugar beets as affected by nutrient environment

The data on the yield of sugar beet roots are given in Figure 1. These show a considerable range in yield among treatments, varying from 1500 to more than 2900 grams of roots per square foot of can surface.

Changes in composition of nutrient solution and drainage

A slight change occurred in pH between the nutrient solution and that of the drainage solutions as shown in Table 1. In general the pH tended to increase. A striking exception is the ammonium solution treatment, which dropped 1.4 pH units.

Preliminary studies showed a drop in phosphorus concentration from 15 ppm in the nutrient solution to 4 or 5 ppm in the drainage waters when no plants were growing in the medium.

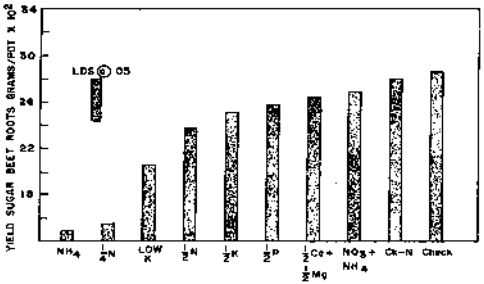

Figure 1.—Yield of sugar beet roots as affected by nutritional environment, 1960.

An equilibrium between phosphorus in solution and phosphorus absorbed on the surface of vermiculite or chemically precipitated is established almost immediately upon solution contact. Additional slight changes in phosphorus concentration were noted between nutrient and drainage solutions with all treatments. The concentrations of phosphorus in drainage solutions after equilibrium with vermiculite and plant absorption were well above the levels considered adequate in soil solution for crops such as corn (3,7,9,15). Nevertheless there were significant differences in phosphorus concentration of drainage solutions among the various treatments as shown on Table 1.

Influence of variation in nutrient cultures on the concentration of phosphorus in plant tissue.

The mean seasonal phosphorus compositions of sugar beet petioles, blades, and pulp as affected by nutrient cultures are

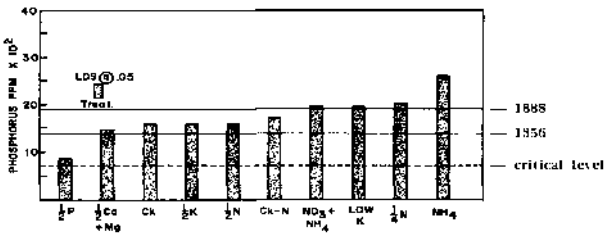

Figure 2.—Soluble phosphorus concentration in sugar beet petioles as affected by nutritional environment, mean for season 1960.

shown in Figures 2, 3, and 4. All nutrient cultures produced plants above the phosphorus "Critical Level" suggested by Ulrich (14) for well-nourished sugar beet plants (Figure 2). His proposed critical level is shown in Figures 2 and 3 as a horizontal dotted line. Wide variations occurred in phosphorus composition among petioles obtained from plants growing in solutions similar in phosphorus content. The cause of the high phosphorus content in petioles from treatments NO_3 + NH_4, Low K, 1/4 N, and NH_4 is not known with certainty. The last three of these treatments gave the lowest yields of roots (Figure 1). The solid horizontal lines are placed in Figures 2, 3, and 4 for comparative purposes.

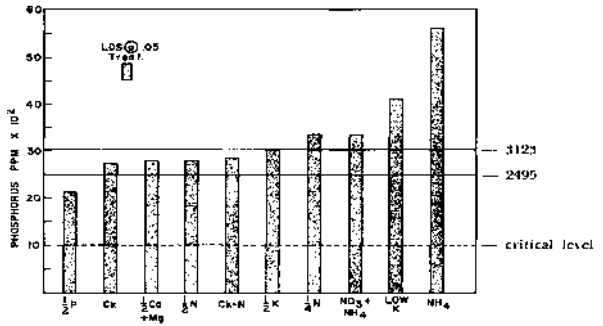

Figure 3.—Total phosphorus concentration in sugar beet leaf blades as influenced by nutritional environment, mean for season 1960.

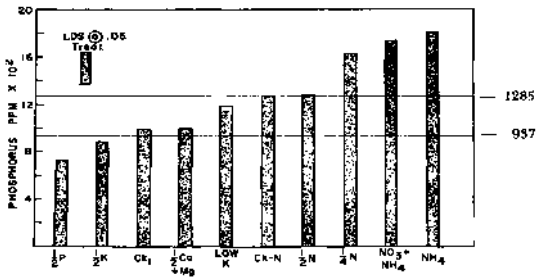

Figure 4.—Concentration of total phosphorus in sugar beet pulp as affected by nutritional environment, mean for season 1960.

These lines were obtained by taking the mean phosphorus concentration in plant tissue, obtained from the two highest yielding treatments, Check and Check—N, as shown in Figure 1. From this value, fiducial limits at .05 probability level were calculated and are used in these figures as points of reference.

Simple correlations were calculated for the purpose of identifying closely associated factors among yield of roots, phosphorus composition of plant tissue and phosphorus composition of nutrient solutions. The factors associated in a significant way are listed in Table 2. Reasoning from the conclusions of Pratt and Thorne (12) one rightly may expect to find a significant positive correlation between the phosphorus concentration in the nutrient solution and the phosphorus composition of sugar beet petiole and pulp tissue. On the other hand one may be surprised to note the negative correlation between yield of roots and phosphorus content of sugar beet leaves and petioles. If one recalls McGeorge's (6) finding, concerning the influence of pH in nurtient solutions, on the uptake of phosphorus, he will not be surprised to observe the significant negative correlations in Table 2 between the pH of the plant growing medium and the phosphorus concentration of sugar beet petioles, leaf blades and pulp produced from that medium.

Table 2.—Simple correlations among yield of roots, sugar beet tissue composition and nutrient solution composition from ten nutrient cultures, 1960.

No.	Factors of Correlation	r values*
1	Yield of roots vs P content of petioles	— .68
2	Yield of roots vs P content of leaf blades	— .74
3	Phosphorus content of petioles vs P in original solutions	+ .67
4	Phosphorus content of petioles vs pH in original solutions	— .75
5	Phosphorus content of petioles vs pH in drainage	— .69
6	Phosphorus content of leaf blades vs pH original solutions	— .63
7	Phosphorus content of leaf blades vs pH in drainage	— .81
8	Phosphorus content of pulp vs P content solutions	+ .63
9	Phosphorus content of pulp vs pH original solutions	— .60

* Required for significance at .05 with 9 d.f. = 0.60

Discussion

Published observations and conclusions dealing with the influence of various environmental factors on phosphorus uptake by plants suggest that a number of factors, whether related or unrelated, operate to favor or hinder phosphorus absorption. Which of the various theories advanced to explain phosphorus availability to plants can be invoked to account for the high phosphorus concentration in treatment NH_4? Arnon's (1) general conclusion that phosphorus absorption would be enhanced by

the presence of rapidly absorbable cations in the rooting medium is in harmony with the observed facts. McGeorge's (6) explanation that at pH 6.0 more than 80% of ionized soluble phosphorus is in the readily absorbable form, H_2PO_4 is not in conflict with the observed facts. The conclusions of Pratt and Thorne (12) that phosphate availability is a function of its solubility offer no help whatever. Why, in view of the favorable phosphorus uptake by plants in the NH_4 treatment, is the root yield so low? Possibly the nutrient balance concepts suggested by Shear and Crane (13) and Pirson (11) are near the mark. Nutrient elements can be as harmful to plant growth when taken up by plants in great excess as when in deficient supply. While the NH_4^+ ion may favor phosphorus absorption high concentrations in the plant may interfere with other vital physiological functions. The secondary effect, that of high phosphorus concentration within the plant, may produce unfavorable reactions with the small quantities of zinc or iron in the plant and thus limit essential enzymatic physiological functions. Whatever the cause of the harmful effect on growth of beet roots it was not inadequate phosphorus.

What theories can be offered to explain the high phosphorus concentration in beet tissue grown in solution l/4 N? The theories of McGeorge (6) and Arnon (1) offer no help. Aerain the explanation of Pratt and Thorne (12) is not enlightening. Nightingale's (8) observations that ample nitrate supplies repress phosphorus uptake provides a back door approach. The 1/4 N treatment obviously provided a much lower concentration of nitrate ions than in any other treatment with the exception of the NH, treatment. P>ased on this theory alone one might expect the phosphorus concentration of plant tissue grown in the 1/4 N treatment to be high in phosphorus.

The authors do not have a satisfactory explanation as to whv Low K treatment should result in high phosphorus concentration in beet tissue. Tissue from this treatment contained hisrh concentrations of calcium, masrnesium, and sodium but it is not obvious why these cations should greatly favor phosphorus absorption.

The fourth treatment which favored high phosphorus uptake was NO_3- + NH_4. The high phosohorus concentration in beet tissue from this treatment may result from a slightly favorable pH in the nutrient and drainage solutions according: to the theory of McGeorge (6). Arnon's (1) explanation for the favorable influence of NH_4^+ and the unfavorable effects of NO_3- on phosphorus uptake, hardly satisfies one in this case because large

quantities of both ions are present. If it could be demonstrated that, apart from the influence of NH_4^+ ions on lowering the pH of absorbing root mediums, the NH_4^+ ions and H_2PO_4- ions were unusually congenial traveling companions, this would offer an explanation. Without this, the favorable influence of solution pH appears to offer the only justification for favorable phosphorus uptake.

The only treatment which resulted in low phosphorus concentration in beet tissue was 1/2 P. The obvious explanation for this is the relatively low phosphorus concentration in the nutrient solutions and in the drainage solutions from this treatment.

The five treatments which are not discussed individually, 1/2 Ca + Mg, Check, 1/2 K, 1/2 N and Check—N, appear to be adequately but not excessively provided with phosphorus. Nutrient solutions from these treatments have similar pH values and contain similar phosphorus concentrations. Of these treatments 1/2 N and Check—N tend toward a build up of phosphorus, particularly in the pulp tissue. This supports the conclusion of Nightingale (8) who observed that ample nitrate supplies repress phosphorus uptake. Treatments 1/2 N and Check—N were provided with a lower nitrate supply than was provided in the other three treatments.

The relatively high but uniform concentration of soluble phosphorus in nutrient culture solutions (Table 1) and the frequent renewals (once to twice a day) give little justification for assuming that high concentrations of phosphorus in specific beet tissue is a consequence of low yields.

The positive correlation between phosphorus content of leaf petioles and pulp and nutrient solutions (Table *2)* must result largely from the relations between plant composition and solutions from treatment 1/2 P. All other treatments contained the same phosphorus concentration in solution but widely variable concentrations in plant tissue.

The high negative correlation shown between yield of roots and phosphorus concentration in leaf blades and petioles can be accounted for by the three low yielding treatments NH_4, 1\2 N, and Low K. These three treatments, for reasons previously indicated, produced plant tissue high in phosphorus.

The high negative correlations between phosphorus concentration in petiole, blade, and pulp tissue, and between pH nutrient and drainage solutions are accounted for largely bv the relations found in two treatments, NH_4 and NO_3 + NH_1, Solutions from other treatments show relatively uniform pH values.

Conclusions

Many factors influence phosphorus uptake by sugar beet plants. No single theory of phosphorus availability accounts for all conditions of phosphorus absorption.

The mechanisms of nutrient absorption of anions and cations may well differ as Lundegardh (5) contends. Nevertheless an interdependence seems to exist among them for absorption by plant roots. There is an indication that the ammonium ion and the monovalent phosphate ion are congenial plant absorption companions.

At a given pH value of nutrient solution in the rooting medium, the rate of phosphorus absorption by sugar beets depends upon the quantity of soluble phosphorus present, as stated by Pratt and Thorne (12). The pH of the nutrient medium appears to be one of the important factors controlling the rate of phosphorus absorption, frequently over-riding the influence of solution concentration, McGeorge (6).

High concentration of nitrates in the solution medium tends to repress the uptake of phosphorus and low concentration is conducive to high phosphorus absorption by sugar beets.

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Classification of Sugar Beet Strains for Resistance to Aphanomyces Cochlioides in Greenhouse Tests

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Increased resistance to the beet water mold, *Aphanomyces cochlioides,* is a major objective in the development of sugar beet cultivars for the Great Lakes region of the United States. Differences in degree of resistance to pure cultures of *A. cochlioides* in the greenhouse between sugar beet cultivars have been demonstrated (1,2,3)[2]. Resistance to *A. cochlioides* in the greenhouse was shown to be indicative of resistance in the field (2,3).

In 1957, a program of testing breeders' strains of sugar beets in the greenhouse for resistance to *A. cochlioides* was initiated. In this paper are presented methods employed and results obtained in testing over 2,900 strains from 1957 to 1961.

Methods

Seeds of the breeders' strains included in the tests were furnished by G. E. Coe[3] and G. J. Hogaboam[3]. Most of the strains were derived from plants selected for resistance to black root pathogens, including *A. cochlioides,* in field trials. Multigerm, monogerm, and monogerm-multigerm hybrid types were included.

The tests were conducted in greenhouses at the Plant Industry Station, Beltsville, Maryland. Seedlings to be inoculated were grown in steam-sterilized loam in well-drained clay saucers of 15 cm diameter and 3.5 cm depth. Twenty-five seed balls per saucer were planted uniformly spaced and at uniform depth. In each test were 24 to 36 entries arranged in 4 to 6 randomized blocks. A semiresistant variety was included in each test as a standard for comparison. In 1957-58 tests, variety US 400 was the standard; in 1959-61 tests, US 401 was the standard.

Zoospore inoculum was used because large quantities can be readily produced in the laboratory and expeditiously applied in regulated amounts in the greenhouse. Zoospores were obtained in accordance with previously described methods (3,4) from monosporous cultures previously isolated from blighted sugar beet seedlings and maintained on maize meal agar slants. Con-

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[2] Numbers in parentheses refer to literature cited.

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centrations of zoospores produced by mycelial mats of the fungus submerged in water were determined with a haemocytometer.

About 2 weeks after planting and after seedlings had been thinned to a maximum of 25 per saucer, each saucer was flooded with 50 ml tap water containing a known number of zoospores. In order to increase the likelihood of attaining the degree of disease intensity that would best distinguish resistant and susceptible host strains, several concentrations of inoculum were employed in each test. Usually the concentration varied with each randomized block of saucers. Concentrations of .2 to .5 million zoospores/saucer were employed in summer when high greenhouse temperatures increase the proclivity of sugar beet seedlings to black root. Higher concentrations, .5 to 1.5 million zoospores/saucer, were employed during cooler months when greenhouse temperatures rarely exceeded 25° C.

Early symptoms of black root—discoloration of the hypocotyl, damping-off—generally began to appear by the sixth day after application of inoculum. About 30 days later, the number of plants surviving and severity of above-ground symptoms displayed by survivors were recorded. Symptoms ranged in severity from a slight darkening at the base of the hypocotyl to a severe necrosis of the hypocotyl which appeared as a black thread.

An index of disease severity was computed for each entry. Each plant was assigned a numerical value according to severity of above-ground symptoms as follows: 0 (no symptoms); 1 (light); 2 (intermediate); 4 (severe); 5 (dead). The quotient of the assigned numerals summated and divided by the total number of plants inoculated equals the disease index.

An opportunity was afforded to compare greenhouse and field determinations of resistance to *A. cochlioides.* Forty-one of the entries had been grown at Waseca, Minnesota, in 1956 by H. L. Bissonnette[4] in field plots naturally infested with *A. cochlioides.* Following a relatively severe black root epiphytotic, harvest root weights of the 41 strains ranged from 75 to 144 percent of check variety US 400. After the 41 strains had been tested for resistance in the greenhouse, a correlation coefficient was calculated from paired greenhouse (disease index) and field (harvest root weight) data for each strain.

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Table 1.—Distribution of sugar beet strains according to disease rating in greenhouse tests for resistance to *Aphanomyces cochlioides*.

Type of sugar beet and year tested	Number of entries	Percent of entries in indicated disease rating class[1]								Average[2] disease rating
		65-74	75-84	85-94	95-104	105-114	115-124	125-134	135-144	
Multigerm, diploid										
1957-58	140	....	5.7	30.7	37.1	24.3	2.2	....	..	98.6
1959	307	2.6	7.5	26.5	40.6	19.2	3.8	...	....	97.7
1960	654	.2	7.6	41.6	39.1	9.8	1.5	.2	...	95.6
1961	114	.9	32.4	50.0	14.9	1.8	....	....	....	88.4
Multigerm, tetraploid										
1961	239	...	.5	12.1	38.4	43.9	4.6	.5	..	104.1
Monogerm, diploid										
1957-58	268	..	2.2	16.8	39.6	20.5	14.2	5.3	.4	104.8
1959	381	....	3.7	31.3	32.3	25.8	6.4	.5	...	100.1
1961	374	1.4	13.9	49.0	26.9	6.4	2.4	....	....	93.0
Monogerm-multigerm hybrid, diploid										
1957-58	61	..	1.7	18.0	67.1	11.5	1.7	....	....	99.4
1959	73	1.5	6.8	21.9	43.9	23.2	2.7	....	...	98.8
1960	249	5.6	25.3	37.3	25.3	5.3	2.0	.4	.8	91.3

[1] Disease ratings expressed in percent of that of commercial check variety US 400 (1957-58 tests) and US 401 (1959-61 tests). The higher the rating, the greater the amount of disease.

[2] Weighted average based on the number of entries in the several disease classes.

Results

The disease index of the commercial check variety included in each test ranged from 2.8 to 4.8 and averaged 4.2. Disease indices of the breeders' strains were converted to percentages of that of the check variety in order to facilitate comparison of strains included in different tests.

The results of the tests are summarized in Table 1. There were differences in degree of resistance among each type of sugar beet tested: multigerm, monogerm, and monogerm-multigerm hybrid. Among all types, disease severity ranged from 65 to 144 percent of that of the check variety. Most of the entries were equal to or exceeded the check variety in degree of resistance (Figure 1). The results of these tests are in sharp contrast to

Figure 1.—Sugar beet strains in soil infested with zoospores of *Aphanomyces cochlioides* in the greenhouse. Row A, monogerm-multigerm hybrid SP59485-1; row B, monogerm-multigerm hybrid SP59495-1; row C, commercial check variety US 401.

previously reported results of similar tests of strains not derived from plants selected for black root resistance wherein most of the entries were less resistant than the check variety (5). The results also indicate a progressive improvement in resistance to *A. cochlioides* among breeders' strains developed during the period in which the tests were conducted. In 1957-58 tests a minority of entries were more resistant than the check variety; in 1961 tests, a majority were more resistant.

As in previously reported studies (2,3), resistance to *A. cochlioides* in the greenhouse was indicative of resistance in the field. A correlation coefficient of —.555 indicates a significant negative association between greenhouse disease indices and harvest root weights of 41 strains exposed to *A. cochlioides* in greenhouse and in field (Table 2).

Table 2.—Classification of 41 sugar beet strains according to greenhouse and field determinations of resistance to *Aphanomyces cochlioides*.

Greenhouse disease rating[1] (y)	Entries in indicated disease rating class in the field (x)[2,3]							
	75-84	85-94	95-104	105-114	115-124	125-134	135-144	Total
75-84	0	0	0	0	0	8	0	8
85-94	0	1	2	2	4	3	3	15
95-104	0	3	5	6	4	1	0	19
105-114	1	0	2	1	0	0	0	4
Total	1	4	9	9	8	7	3	41

Correlation coefficient (r_{xy}) = —.555**

[1] Disease index in percent of check variety US 400. The higher the rating the greater the amount of disease.

[2] Root yield in percent of check variety US 400 in field plots naturally infested with *A. cochlioides*. Data based on 2 single-row plots, each 25 ft. long.

[3] Yield data furnished by H. L. Bissonnette.

Summary

A method of testing sugar beet seedlings in the greenhouse for resistance to the water mold, *Aphanomyces cochlioides*, is described. Greenhouse tests of over 2,900 breeders' strains included in the program of developing sugar beet cultivars for the Great Lakes area showed the majority to be more resistant than commercial check varieties US 400 and US 401. A progressive improvement in resistance to *A. cochlioides* was noted among the breeders strains tested during the period 1957-61. Additional evidence was obtained that resistance to *A. cochlioides* in the greenhouse is indicative of resistance in the field.

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Restitution of Growth In Nitrogen Deficient Sugar Beet Plants[1]

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In agricultural, as well as natural environments, wide fluctuations in the levels of individual environmental factors are common. Plant growth may be restricted by an unsuitable level of a particular factor, e.g., deficiencies of water or nutrients, while all other factors are optimal for growth. Under these circumstances, it is useful to know how the plants react as the deficiency develops and when the restriction is alleviated.

A renewal of normal leaf development commonly is observed after nitrogen deficient sugar beet plants are supplied with nitrogen. Associated with this is a decline in sucrose concentration in the roots. Root growth is also renewed, but there is conflicting evidence on the manner in which it occurs. Loomis and Nevins (3)[3] found considerable lag between the time nitrogen was resupplied to deficient plants growing in nutrient culture and the time root growth was renewed. In contrast, Ulrich (7) found that supplying nitrogen to plants grown in pots with soil shortly after they became deficient resulted in a rapid renewal of root growth.

Both from an ecological and from an economic standpoint, it is of interest whether growth occurs at an above normal rate during restitution. Such phenomena have been studied intensively with higher animals and have been termed "compensatory growth" (9). It appears appropriate to employ this same terminology in discussing plant growth. Compensatory growth has been observed in several plant species during recovery from moisture stress (literature reviewed by Stocker, 5). Owen (4) reported that this phenomenon occurred with sugar beet but his data appear inconclusive. Ulrich (8) observed compensation to the effects of temperature in that sucrose yields from plants which had experienced a period of growth in a hot climate and were then transferred to a cold climate, exceeded yields from plants which remained continuously in either hot or cold climates.

Less information is available on recovery from nutrient deficiencies. Compensatory growth did not occur in the nitrogen experiments cited above (3,7) although it might be expected

[1] This work was supported in part by a grant from the beet sugar companies operating in California and the California Beet Growers Association, Ltd.

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[3] Numbers in parentheses refer to literature cited.

if a substrate, normally limiting, were to accumulate during the period of stress. With sugar beet, the accumulation of sucrose in the storage roots is promoted by nitrogen deficiency. This sucrose is available for growth and might contribute to an above-normal growth rate when nitrogen again becomes available. This was not observed in the pot experiments but it may be that compensatory growth relationships are different for plants Grown in competitive stands than for plants grown in pots. In the present experiment, the influence of a period of nitrogen deficiency on subsequent growth in a high-nitrogen environment was studied under field conditions.

Methods

The crop was grown on Holtville clay loam soil at the University of California Imnerial Vallev Field Station. This soil releases large amounts of nitrogen but at a rate too low for maximum growth of sugar beet and a luxury level of nitrogen nutrition is maintained only by applying 200 to 400 pounds nitrogen per acre. The seed (Holly HH-3) was planted October 9 on double row beds (14-26 inch spacing). Thirty-five pounds phosphorus as treble superphosphate, and 100 pounds nitrogen as ammonium sulfate were applied per acre in the shoulders of the beds at planting. An additional 50 pounds of nitrogen per acre were applied to all plots in November. Nitrogen was the only limiting nutrient during the growth of the crop. Furrow irrigations kept the plants well supplied with water.

The experiment consisted of four nitrogen treatments arranged in a randomized block design with six replications. The treatments were designed so that the growth of high-nitrogen plants could be compared to that of low-nitrogen plants with or without fertilization. The treatments were established beginning February 23, when the plants approached a nitrogen-deficient condition, by sidedressing ammonium nitrate to appropriate plots as shown in the following table:

Treatment		Pounds of nitrogen applied per acre on:			
		Feb. 23	Mar. 13	Apr. 4	Apr. 23
A	High nitrogen	200	0	0	200
B	Low nitrogen Fertilized after 3 weeks deficiency	0	200	0	0
C	Low nitrogen Fertilized after 6 weeks deficiency	0	0	200	0
D	Low nitrogen	0	0	0	0

The plots were irrigated on the same day that nitrogen was applied.

The differences among the treatments may be seen from the tissue analysis (1) data presented in Figure 1. The plants which were not fertilized on February 23 became deficient about March 1 (NO_3-N in petioles of recently matured leaves dropped below 1000 ppm dry weight). With this soil there may be a 1-week delay following application of ammonium nitrate before nitrate appears in the plants (2). Thus, the refertilized low-nitrogen plants (B and C) were deficient for about 3 and 6 weeks, respectively. The high-nitrogen plots (A) approached a deficient level on April 23 at which time they were fertilized with an additional 200 pounds nitrogen per acre. During May, treatment B, the first to be refertilized, and then treatment C, became nitrogen deficient again; no more nitrogen was applied to these plots.

Harvests were made at 3-week intervals beginning February 20 and extending to June 26. On each date the beets from 60 feet of row in each plot were harvested. Fresh and dry weights of roots and tops (including crowns) were measured; sucrose concentration was determined on samples of roots[4].

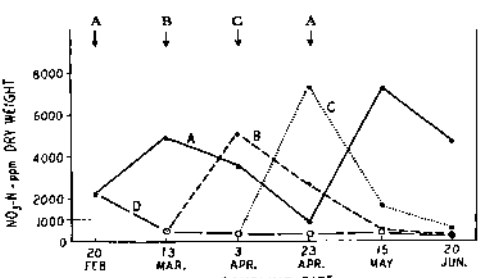

Figure 1.—The concentration of nitrate-nitrogen in recently mature petioles from plants receiving various experimental treatments. Letters refer to treatments and vertical arrows indicate dates when 200 lb. nitrogen/acre was applied to various treatments.

Results

As shown in Figure 2, the yield of fresh tops from the high nitrogen plants (A) increased rapidly between February 20 and

[4] The Holly Sugar Corporation generously conducted these determinations as well as having supplied the seed.

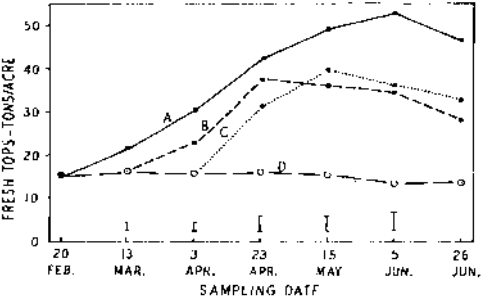

Figure 2.—Yields of fresh tops from sugar beet plants as affected by various nitrogen regimes. The vertical lines correspond to the LSD_{05}.

Table 1.—Absolute and relative rates of top growth between April 3 and April 23. (Means within a column followed by the same letter are not significantly different front each other, P = .95.)

Treatment	Absolute growth rate Lb/acre day		Growth relative to mean of April 3 and April 24 yields Lb/acre day	
	Fresh wt	Dry wt	Fresh wt	Dry wt
A	1140a	192a	0.016a	0.026a
B	1420b	160a	0.024b	0.025a
C	1480b	159a	0.032c	0.028a
D	76c	60b	0.002d	0.013b

June 5 and then declined. Yields of tops from low-nitrogen plants (D) remained approximately constant near 15 tons per acre; with refertilization (B and C), top growth was greatly stimulated. In an analysis of variance for treatments A, B, and C for April 3 and April 23, a significant date X nitrogen interaction was obtained indicating significant differences in the growth rates of these treatments. This is considered in detail in Table 1.

On a fresh basis, but not on a dry basis, the growth of the refertilized plants exceeded that of the high nitrogen plants. Considering that there was some delay after April 3 before the plants in treatment C obtained appreciable nitrogen from the soil, their peak growth rate was undoubtedly greater than the mean value shown. In the present case, there is no evidence of exponential growth and relative growth has been calculated as the ratio of daily growth to mean weight. The relative growth rates shown in Table 1 are lower than commonly reported for plants due to the large size of the plants on April 3. On a fresh

basis, the refertilized plants showed the highest relative growth rates and on a dry basis they equalled that of the high-nitrogen plants.

Root yields are summarized in Figure 3. The degree of nitrogen deficiency obtained may be ascertained by comparing growth rates of high- (A) and low-nitrogen (D) plants. Between March 13 and April 23 the yield of roots from low-nitrogen plants increased 460 lb/acre day or only 70% as rapidly as the high-nitrogen rate of 650 lb/acre day. Refertilized plants quickly recovered the same absolute rate of growth as the high-nitrogen

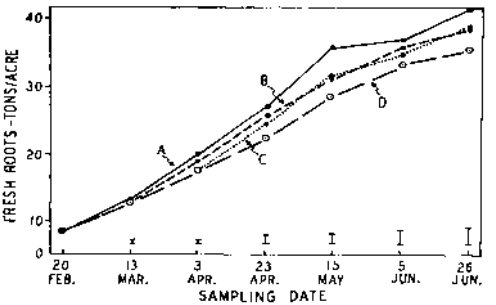

Figure 3.—Yields of fresh roots from sugar beet plants as affected by various nitrogen regimes. The vertical lines correspond to the LSD_{05}.

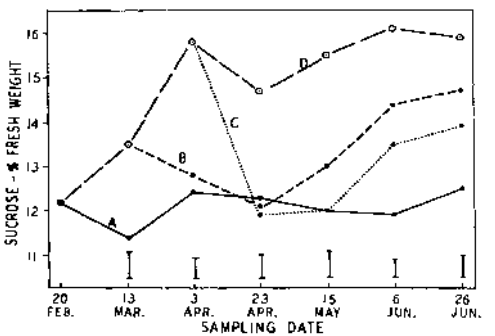

Figure 4.—The concentration of sucrose in sugar beet storage roots as affected by various nitrogen regimes. The vertical lines correspond to the LSD_{05}.

plants, i.e., between April 3 and April 23, treatments A, B, and C all increased 660 lb/acre day and compensatory growth did not occur. Treatments B and C ultimately produced the same root yield and were intermediate between treatments A and D.

After February 20, sucrose concentration in the low-nitrogen (D) plants increased rapidly to near 16% while that in the high-nitrogen (A) plants remained near 12% (Figure 4). The low-nitrogen plants returned to the lower value within 3 weeks after refertilization. With treatment D, the increase in sucrose concentration offset, for a period of time, the lower rate of root growth and on March 13 and April 3, sucrose yield was higher from this treatment than from treatment A (Figure 5). When the low-nitrogen plants were refertilized, the rate of sucrose accumulation slowed and the plants yielded less sucrose after April 23 than either the high- or low-nitrogen plants.

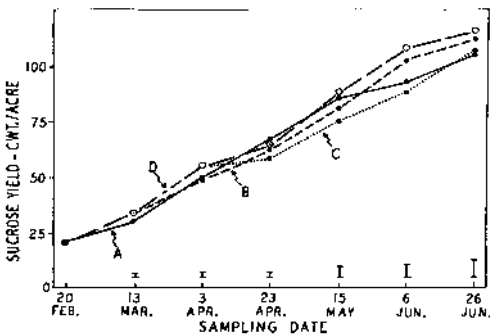

Figure 5.—Sucrose yields in sugar beet storage roots as affected by various nitrogen regimes. The vertical lines correspond to the LSD_{05}.

Discussion

In this experiment, nitrogen-deficient sugar beet plants renewed growth rapidly when nitrogen was resupplied after 3 or 6 weeks of deficiency. The patterns of response were similar to those obtained by Ulrich (7) with plants grown in soil in pots, i.e., root and top growth were stimulated quickly by applications of the limiting nutrient. These results contrast with those obtained previously with plants grown in vermiculite and watered with nutrient solution (3) where the transition from high to low nitrogen occurs rapidly and the degree of deficiency is more severe than with plants grown in soil. It appears that the growth

of refertilized plants may be more dependent upon the degree than upon the length of the deficiency. This could be studied by conducting the experiment on several soils having a wide range of nitrogen supplying power.

Compensatory growth may be defined as greater than normal absolute or relative growth over the same interval of time or at the same stage of development. Tn this study the plants were all in a vegetative phase of development and the only usable measure of stage of growth is plant size. Since similar plant sizes occurred at different times and under different environments, specific comparisons in plant growth, as shown in Table 1, were made only for the April 3-April 24 interval of time. Under the conditions of the experiment, this was the only period during which all treatments could be compared on a proper basis and the only period during which either of the refertilized treatments showed what might be termed compensatory growth for either roots or tops. Compensatory growth (on absolute and relative bases) occurred with fresh tops but not with dry tops or other characters.

The relative growth rates presented in Table 1 were calculated on mean weight of tops which correlates well with leaf area, rather than on total plant weight. Leaf areas were not measured but can be estimated for treatments A and D from the performance of similar plants in an adiacent experiment; on April 3, leaf areas for these treatments equalled aoproximately 8 and 4 acres leaves per acre land, respectivelv. From this it appears that the relative regrowth of fresh tops was inversely related to the initial leaf area. This is the reverse of what is observed with seedling stands or after defoliating a pasture but is expected with high leaf area where there is considerable mutual shading of leaves. However, the inverse relationship between too weight and resrrowth is not apparent in the relative growth of drv tops. Thus the comoensatory growth of fresh tops in the refertilized plants was in the enlargement of the young leaves and was evident as an increase in succulence. Evidently the nitrogen-deficient crop had a greater potential for leaf growth than did the high-nitrogen crop.

The use of relative growth rates implies a dependence of growth upon size of plant. i.e.. upon the "capital" for growth. The present results indicate that the relative growth of a nlant communitv with closed canopy may have little meaning since the community has passed the logarithmic phase of growth and light or some other environmental factor rather than the size of plants is limiting. The individual plants in the community

have a potential for much higher growth rates than is possible under competitive conditions and this apparently was expressed during restitution after the intensity of competition had been reduced by the period of nitrogen deficiency. It is not possible from the present data to determine whether the higher rate of leaf growth was "normal" or "above normal" for that environment and initial leaf area.

Total crop growth rates (dry matter increase per unit land area per day) were measured. Unfortunately, the dry matter determinations on roots were variable and the data have not been presented here. However, it was possible to conclude that the refertilized plants had lower growth rates than the high-nitrogen plants and there was no evidence, from this index, of compensatory growth.

Most of the carbohydrates used during regrowth presumably were supplied from current production while a lesser portion may have come from sucrose which had accumulated previously in the roots. Since sucrose continued to accumulate in the roots of the refertilized plants, current production of carbohydrates apparently exceeded use during this period. And, since the accumulation was at a low rate, it seems probable that under other conditions, more favorable for growth or less favorable for photosynthesis, a net loss of sucrose from the roots would have been observed.

From a practical point of view, these results are helpful in interpretating situations where nitrogen-deficient sugar beet plants experience an increase in the supply of available nitrogen. This may result from the growth of roots into unexplored volumes of soil, from an increase in nitrification, from leaching of surface accumulations of nitrate into the root zone (6), or from fertilization. An important conclusion from this experiment is that an increase in nitrogen supply did not cause a compensatory increase in sucrose yield but, instead, reduced the ultimate yield of sucrose below that of plants which remained at either high or at low nitrogen. Evidently, sugar beet should not be allowed to become nitrogen-deficient in midseason before applying supplemental nitrogen and care should be taken to avoid increases in nitrogen supply during the preharvest period.

Summary

The effects of a period of moderate nitrogen deficiency on the subsequent growth of plants in a high-nitrogen environment was investigated with sugar beet grown under field conditions. Growth of storage roots and of tops increased very soon after nitrogen was applied. During the restitution phase, fresh weight

of tops of the refertilized plants increased at an above normal, "compensatory", rate. However, the absolute increase of total dry matter and of dry matter in tops was less than with the high-nitrogen plants.

Sucrose accumulated more slowly in the storage roots of refertilized plants than in the roots of plants that were maintained at either continuous high or continuous low nitrogen. A net loss of sucrose did not occur indicating that the renewed growth of tops was supported by current photosynthesis and by carbohydrates which had accumulated in the leaves.

Allowing sugar beet to become nitrogen deficient before applying supplemental nitrogen appears to be a poor practice in the commercial production of sucrose.

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The Distribution of Airborne Mesophilic Bacteria, Yeasts and Molds in Beet Sugar Factories[1]

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The control of bacteria, yeasts and molds in finished crystalline sucrose is a problem faced by every producer of this very marketable chemical. Due to the ubiquitous nature of these microorganisms, their sources in finished sugar may be manifold, and the air in and about the factories has always been suspect. For this reason most, if not all, sugar manufacturing concerns go to great lengths to filter the air which contacts the finished product. Such filtering mechanisms may include banks of treated fiber glass filament furnace filters and Precipitrons or the furnace filters alone. Drying the wet granules immediately after washing requires a large volume of clean, warmed air. Forced circulation of air over sugar in bulk storage is apparently necessary to minimize moisture condensation in the silos. The air in both cases must be filtered to maintain as low a microflora contamination as possible.

Very little quantitative data are available which describe the contribution of air to the contamination of finished granulated sugar. This paper is a report of a preliminary study of the air in and about two beet sugar factories. Both were showing occasional high microorganism counts in finished sugar.

Material and Methods

Air Sampling

The air was sampled by the use of the Andersen Sampler (l)[3]. A diagramatic sketch of this instrument is shown in Figure 1. The sampler is a unique cascade type air sampling instrument. Air drawn through the sampler at a given rate, i.e., 1 cu foot per minute, passes through the six stages of the sampler and at each stage is impinged onto the surface of a nutrient agar plate containing medium prepared to grow the type of specific organism sought. There are 400 holes in each stage cover but from stage to stage, proceeding from 1 through 6, the holes become smaller, thus serving to increase markedly the velocity at which the air and particles are impinged upon the agar plate immediately below each cover. This velocity increase at each stage separates the particles suspended in the air into different sizes according to the mass of each particle.

[1] Acknowledgement: Research supported by Amalgamated Sugar Company, Ogden, Utah.
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[3] Numbers in parentheses refer to literature cited.

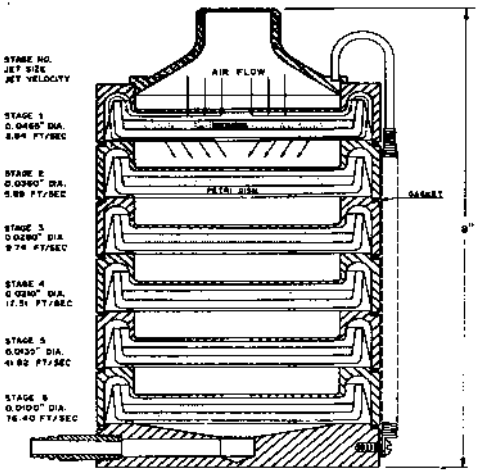

Figure 1.—Andersen Sampler.

Only the very large particles are deposited at stage 1 and at each stage the deposited particles are smaller until at the final stage, (No. 6) only particles of 1 micron or less impinge upon the surface of the growth media. Larger particles are distributed according to size among the intervening 5 plates. Should a particle be carrying a viable mold or yeast spore, or a mesophilic bacterium then a colony will grow to visible size in a few hours. Colonies may be counted and the number of microorganisms per cubic foot of air estimated.

Media

For the detection of yeasts and molds, BBL[4] mycophil agar plates were prepared with the pH of the agar adjusted to 4.5.

For detection of mesophilic bacteria, BBL nutrient agar adjusted to pH 7.0 was used in the plates.

Sampling procedure

At Factory A, 5 sampling stations were established as follows:

A. Silo air circulation—before filtration.
B. Silo air circulation—after filtration.
C. Tunnel under the silos. (Sugar was being moved on a conveyor belt at time of sampling.)

[4] **BBL - Baltimore Biological Laboratory**

D. Granulator air circulation—before filtration.
E. Granulator air circulation—after nitration.

Four samples were taken at each sampling station, a 5 min. and 10 min. sample each for yeasts and molds together and the same for the mesophilic bacteria. Yeasts and molds may be detected on the same plates and are distinguished by colonial morphology. After sampling all plates were returned to the laboratory where they were allowed to incubate for 72 hours; the mycophil agar plates for yeasts and molds at room temperature (22° C to 24° C), and the nutrient agar plates for mesophiles at 35° C.

At Factory B, 5-minute samples were taken at the following sampling stations:

A. Silo air circulation—before filtration.
B. Silo air circulation—after filtration.
C. Tunnel under silos. (Sugar was being moved on conveyor belt at time of sampling.)
D. Granulator air after passage through the granulator.
E. Silo air system. Air in area at top of silos.

The procedures followed were the same as for Factory A.

Factory A was again sampled late in the summer. At this time conditions were much different. The earlier samplings were taken during the manufacturing cycle. At the second sampling the plant was not in operation; many of the silos were empty and sugar movement was limited to transport by conveyor belt to bulk cars, to the classifying room, or recirculation to another of the large silos. Except for a slight change in the sampling stations, the procedures were exactly the same as described above:

A. Silo air circulation—before filtration.
B. Silo air circulation—after filtration.
C. Tunnel under silos. (Sugar was being moved on conveyor belt at time of sampling.
D. Classifying room. (Sugar dust was being blown about at the time of sampling.)

Results

First Sampling—Factory A. The results of the sampling of the air for molds, yeasts, and bacteria at the various sampling stations are presented in Table 1. The term "total" represents the total number of organisms from all six stages of the sampler for each sample. The totals of the 5-minute and 10-minute samples agreed well enough that the microorganisms in each class per cubic foot of air were calculated by taking the total number of organisms of both the 5- and 10-minute samplings and dividing by 15. A comparative summation of these results is presented in Table 1.

Table 1.—Microorganisms per cubic foot of air at each sampling station. Plant A.—First sampling.

		Mesophiles	Molds	Yeasts
A.	Silo air circulation before filters	5.2	13.6	4.9
B.	Silo air circulation after filters	1.0	3.5	0.5
C.	Silo—tunnel air	3.0	25.0	8.7
D.	Granulator air before filtration	22.2	5.2	5.7
E.	Granulator air after filtration and Precipitron	6.4	.3	.2

Table 2.—Microorganisms per cubic foot of air at each sampling station—Plant B.

		Mesophiles	Molds	Yeasts
A.	Silo air circulation before filters	56.6	6	.4
B.	Silo air circulation after filters	6.4	3.2	0
C.	Tunnel under silos	10.0	1.4	.4
D.	Granulator air after passage through the granulator	23.8	1.0	0
E.	Silo air system—air in area top of silo	4.6	.4	.6

Table 3.—Microorganisms per cubic foot of air at each sampling station. Plant A. Second sampling.

		Mesophiles	Molds	Yeasts
A.	Silo air circulation before filters	75.6	13	0.8
B.	Silo air circulation after filters	10.6	5	0.2
C.	Tunnel under silos	(Not sampled)	1.4	Neg.
D.	Classifying room	52.4	18	2

Factory B. A summary of the results of sampling of air in Factory B is presented in Table 2. Again the total colonies of all stages of the Andersen Sampler were utilized to arrive at the number of organisms per cubic foot of air.

Second Sampling—Factory A. Table 3 presents the results of air sampling of stations of Factory A late the following summer.

Discussion and Conclusions

Note that the highest concentration of mold spores in the first sampling, Factory A, was found in the tunnel air where the finished sugar was being transported on an endless belt to bulk cars. The belt had no protective cover. The contribution of this situation to the mold count of the sugar in the cars is unknown, but poses a potential addition of mold and yeast spores

to sugar on the belt. The tunnel air was laden with sugar dust and accounts for the large number of spores in the air.

The yeast count of 8.7 per cubic foot of air was surprisingly low. The same plates are used for the mold and yeast count, and mycophil agar adjusted to pH 4.5 was the medium used. Two conditions may have contributed to the low yeast count; i.e., the mold count was very high and it is suspected that the larger mold colonies not only hid many yeast colonies, but may have suppressed the growth of the yeasts and the low pH may have tended to inhibit the initiation of yeast growth.

The mesophile count was highest in the outside air as it was pumped into the granulator. Fortunately the bank of furnace filters and the Precipitron effectively remove these bacteria from the air supply to the granulator as indicated by the reduction of mesophilic bacteria from 22.2 to 6.4 per cu ft air. The few organisms showing on the plates probably were carried into the chamber between the Precipitron and the filter bank when the door was opened to gain entry with the sampler. There was a high negative pressure within the area between the two filters. The results of these tests certainly give one confidence in the Precipitron and fiber glass filtration system.

The fiber glass furnace filters used in the bulk silo circulation system effectively removed 3/4 of the mold spores, 9/10 of the yeasts, and 4/5 of the mesophilic bacteria. This is a fair reduction, but still leaves room for improvement of this filtering system.

The bulk storage air of Factory B was not nearly as contaminated with yeasts, 0.4 per cu ft, and molds, 6 per cu ft, as that of Factory A. In contrast, the mesophilic count was relatively high with 36.6 organisms per cu ft of air. Factory B was having a problem of a high mesophilic bacteria count in finished sugar at the time these air samples were taken. Also the number of bacteria per cubic foot of air sampled after passing through the granulator was shown to be high at 24 bacteria per cu ft. This air had been filtered and passed through a Precipitron before entering the granulator. Our experience at Plant A had indicated that air filtered in this manner was practically free of microorganisms, therefore the increase to 24 bacteria per cubic foot of air was a reflection of the high count in the newly produced sugar. It is also suggested that the high mesophilic count of the bulk silo air before filtration owes its origin to this same source. Thus a vicious cycle appears. The contaminated freshly produced sugar contaminates the air of the plant which in turn may reintroduce microorganisms into the manufacturing process, which then show up in the granulated sugar.

At the second sampling of Plant A, yeasts and molds were comparatively few in number, but the mesophilic bacteria counts were very high. In comparison with the first sampling carried out during the sugar campaign, the mold count in the silo air circulation system was about the same, but less in the tunnel beneath the silos. The number of mesophilic bacteria was much higher in the bulk silo circulation. The air in the classifying room was not sampled at the earlier sampling, but showed a high count at this time. It is interesting to note that the mesophilic counts routinely carried out on sugar being shipped from Factory A were high at the time of this sampling, and the count of mesophiles in the air circulation system before filtration reflected the count in the granulated sugar.

Summary

1. The mold count is usually high in air carrying large amounts of sugar dust as seen in the bulk silo tunnels. This is especially true where air is rapidly circulated by large fans, and the dust is "swept" off the top of the sugar. The circulation also tends to keep the dust stirred up throughout the bulk storage areas.
2. The spun glass filters (furnace filters) placed in the air circulation path remove a rather large portion of microorganisms from the air by removal of dust particles. However, the efficiency of these filters could and should be increased.
3. The Precipitron in conjunction with the spun glass filters, efficiently removes most bacteria, yeasts, and molds from air being forced through the granulator.
4. The outside air around Factory A carried a greater percentage of mesophilic bacteria than yeasts and molds. The yeasts and molds were found most often in sugar dust laden air.
5. It is apparent that a high mesophilic bacteria count in finished granulated sugar is reflected in the mesophilic count of air being circulated over that sugar.

Literature Cited

(1) ANDERSEN, ARIEL A. 1958. New sampler for the collection, sizing, and enumeration of viable airborne particles. J. of Bact. 76 (5) : 471-484.

The Control of Weeds in Sugar Beet By An Endothal / Propham Mixture Applied at Drilling

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Introduction

Endothal has been used in the United States commercially, for several years with considerable success. It was first introduced to Britain for experimental work in 1954 but Parker (6)[2] reported that it was not sufficiently effective against certain important British weeds. It was noted also by Parker that Endothal was influenced by both soil type and rainfall.

More recently Murant (4) described a combination of Endothal and Propham (IPC) which gave improved over-all kill of weeds. Endothal was poor against *Stellaria media, Sinapis arvensis, Kaphanus raphanistrum, Chenopodium alburn* and *Spergula arvensis,* and only moderate against *Avena fatua.* Propham, while itself not very good against *Matricaria maritima* gave with Endothal, a good control of most of the important weeds (with the exception of Brassica weeds, and *Chenopodium album).*

Murant also noted in this paper that while there was no evidence in the trials reported that the soil type affected Endothal and Propham, previous experience had shown inactivation of both chemicals in black Fen soils. No information was available on the effect of rainfall upon efficiency.

Subsequently it became clear that soils, other than fen soils, affected the efficiency of Endothal and Propham. Murant and Cussans (5) reported 7 trials conducted in 1959 from which it was clear that there was a relationship between activity and the amounts of organic matter and clay in the soils. A factor known as Relative Absorption was devised (= 5x organic matter % and clay %). In these same trials the rainfall was about 1 inch in all cases and no difference which could be attributed to this factor could be seen.

In the same paper 1960 trials are described. In that season an attempt was made to relate the Relative Absorption factor with efficiency but it was found that the rainfall variations overlaid the pattern of responses to such a degree that it was possible only to say that the effect of rainfall was greater than that of soil type.

[1] The Murphy Chemical Company Limited, Wheathampstead, Hertfordshire. England.

[2] Numbers in parentheses refer to literature cited.

The writer's colleagues, Bagnall, et al. (1,2) reported 21 trials with an Endothal/Propham mixture. The data presented showed that rainfall was an important factor in determining the efficiency of the mixture and also that the dose required was influenced by the soil type. The value of the Relative Absorption factor was not borne out, and it was suggested that the chief factors governing rate of use were the clay and coarse sand contents of the soils.

Three rates of the Endothal/Propham mixture were employed. In 13 trials out of 21, little or no rain fell and control was generally poor. In the remaining 8 trials a satisfactory weed control was obtained since a sufficient amount of rain fell upon the soil after application of the herbicide. In these 8 successful trials one or the other of the two lower rates used was adequate for control of weeds with safety to the beet. In the present paper the two rates have been designatd EIGHT Rate (L) and MEDIUM Rate (M) (see below) and in Table 1 and Figure 1 they have been related to the coarse sand and clay percentages in the soils treated. (The soil analysis method used was based on methods reported by Bouyoucos (3) and Tyner (7). It employed a 50 gram soil sample and was designed to record coarse sand at 178 microns and over and clay up to 2 microns. In this paper this method is called the "Standard LONG method").

In Figure 1, lines A-B C-D represent the approximate position on which most soil types lie if plotted in terms of coarse sand and clay. Although the data available to Bagnall, et al. was scanty, it was considered that the division of the main axis A-B in segments, by radii as shown, might well form a useful basis for dividing soils into dosage categories.

In 1961 the writer and his colleagues developed this aspect of the use of the Endothal Propham mixture.

Table 1.—Light and medium rates related to coarse sand and clay percentages in the soils treated.

Trial No.	Mechanical soil analysis				Organic matter	Satisfactory rate of Endothal/Propham
	Coarse sand	Clay	Fine Sand	Silt		
1	18.8	18.2	25.0	8.0	2.61	L
2	31.8	21.6	35.2	11.4	2.51	L - M
3	0.2	11.0	65.6	23.2	2.08	M
4	1.4	7.0	77.2	14.4	3.40	M
5	45.4	8.4	38.8	7.4	3.24	L
6	40.4	9.6	44.6	5.4	1.80	L
7	21.4	22.4	31.2	25.0	3.80	M
8	9.6	24.8	47.4	18.2	3.26	M

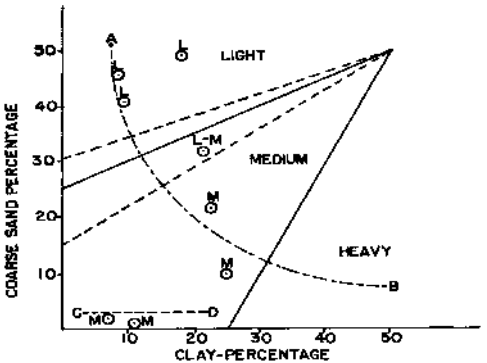

Figure 1.—1960 trials—shows relationship between rates of use of endothal/propham and soil types in terms of course sand and clay.

Materials and Methods

Forty-one trials were successfully carried out with a proprietary preparation of the herbicides, containing 11.4% w/v Endothal (acid equivalent), 8.55% w/v technical Propham. Six rates of use were tested, 3 rates out of the range being used at any one site, the choice depending on the nature of the soil. All soils were analyzed mechanically by the method referred to above, and these values were related to the correct rate of the herbicide (i.e., the rate giving nearest approach to 100% weed kill without adverse effect on the beet). The combined herbicide was applied by hollow cone nozzle in a band directly over the seed row immediately after drilling. Two types of machine were used: a commercial rig, comprising a band sprayer mounted on a precision drill (this method was used both by research staff and commercial growers); and a hand operated device incorporating the same type of nozzle which was pushed along the rows immediately after drilling in the normal way. The nozzles employed were specially developed to apply an even dose of spray across the 7-inch band used. For both machines the following operating data were appropriate: Speed 2 mph; nozzle output 8 fl. oz/minute; pressure 16 lb sq inch; application adjusted to give 7-inch band; and output of 7 gallons per acre, 21-inch row spacing. The formulation described above at dilutions of 1 in 16, 1 in 12, 1 in 8, 1 in 6, 1 in 5, and 1 in 4 give the following rates of use per acre in terms of over-all spraying:

Endothal (Acid equivalent) lb/acre	Propham (technical) lb/acre	Title of rate
1½	1⅛	Extra Light (X/L)
2	1½	Light (L)
3	2¼	Light/Medium (L/M)
4	3	Medium (M)
5	3¾	Medium/Heavy (M/H)
6	4½	Heavy (H)

The titles described have been used for convenience in practice and as they have been found self descriptive they are used here. The titles relate also approximately to the soil type to which they are appropriate.

In addition to the trials, observations on the efficiency and safety of the combined herbicide were made in the 61 cases where it was used commercially, and in these cases also results were related to soil analyses.

Full details of the methods of application, weed and beet counts in the trials will be given in a paper by Caldicott J. J. B. now submitted to 'Weed Research'. The present paper will be confined to the direct relationship between rate of use of the herbicide and the soil type.

Results

Table 2A shows the relationship between the mechanical soil analysis by the Standard Long method and the correct rates of use of the herbicide in the 41 experiments conducted.

Long method results

The results of the Standard LONG method of analysis are plotted in Figure 2. A circular form of graph has been found most appropriate because a) it lengthens the coarse sand axis and thereby tends to separate for pictorial demonstration the different samples and b) because it is found that the curves limiting the different sections are smooth and easily defined when plotted in this form.

It is readily seen that the different rates of use fall smoothly into categories indicating that these rates and the coarse sand and clay factors are clearly and directly related. There tends to be some uncertainty at the higher clay levels, as indicated by trials nos. 37 to 41 where the clay percentages by the Long method are all over 26%. These trials are plotted as problem soils. Field observations indicate that one of the explanations of this situation is the fact that such soils commonly do not form a good seed bed. In England this was particularly common in the spring

Table 2.—Mechanical soil analysis data and herbicide rates for 41 experimental sites.

Trial No.	A. Standard long method: Coarse sand %	Clay %	Fine sand %	Silt %	Organic matter %	Theoretical dose	B. Short method: Coarse sand %	Clay %	Theoretical dose	Dose by result
					(1) Soils in which Short and Standard method forecasts agree					
1	0.2	15.6	67.0	15.2	3.91	M	6.5	17.0	M	M
2	0.6	15.8	66.0	17.6	3.52	M	0.8	18.0	M	M
3	1.4	25.4	51.6	21.6	5.25	M	5.9	24.0	M	M
4	2.4	21.0	58.6	18.0	3.09	M	2.3	20.0	M	M
5	2.4	21.4	48.2	28.0	5.61	M	2.6	10.0	M	M
6	7.6	18.8	52.0	22.0	2.75	M	9.2	15.0	M	M
7	8.6	23.8	59.0	8.0	4.5	M	19.2	20.0	M	M
8	12.4	29.0	43.0	15.6	2.3	M	11.0	25.0	M	M
9	15.0	28.4	40.6	16.0	2.17	M	14.7	33.0	M	M
10	18.0	21.0	43.0	18.0	6.9	M	18.2	18.0	M	M
11	20.2	14.4	47.4	18.0	5.4	L/M - M[1]	18.4	15.0	L/M - M[1]	L/M - M[3]
12	25.6	21.3	32.6	20.0	3.6	L/M	28.6	14.0	L/M	L/M
13	27.0	15.2	45.8	14.0	.	L/M	25.3	15.0	L/M	L/M
14	28.0	19.2	41.6	11.2	3.8	L/M	28.2	11.0	L/M	L/M
15	29.0	11.0	31.2	18.0	3.8	L - L/M[1]	28.8	20.0	L/M	L - L/M[3]
16	32.0	19.6	36.4	12.0	2.9	L/M	33.3	29.0	L/M	L/M
17	32.2	15.6	39.4	12.8	2.3	L - L/M[1]	40.0	17.0	L	L - L/M[3]
18	32.4	20.0	30.6	17.0	2.2	L	37.7	24.0	L/M	L/M
19	33.2	8.6	51.4	6.8	2.5	L	96.6	15.0	L	L
20	33.2	13.2	28.4	25.2	3.2	L	31.5	12.0	L	L
21	36.9	14.2	40.1	8.8	2.2	L	41.4	18.0	L	L
22	37.0	12.0	41.0	10.0	1.2	L	45.0	11.0	L	L
23	38.0	8.6	46.2	7.2	2.6	L/M	49.0	16.0	L	L
24	38.0	21.8	28.2	12.0	4.4	L	40.3	28.0	L/M	L/M
25	39.4	14.2	32.2	14.2	3.5	L	45.6	17.0	L	L
26	39.8	11.6	45.0	3.6	2.0	L	47.0	7.0	L	L
27	40.6	11.2	44.2	4.0	4.5	L	54.8	5.6	L	L
28	41.8	9.8	44.4	4.0	2.4	L	44.6	18.0	L	L
29	47.4	9.4	31.2	18.0	3.8	XL - L[1]	23.9	3.5	L	L
30	47.4	14.6	30.0	8.0	4.1	L	46.3	12.0	L	L
31	54.4	9.8	33.6	2.0	1.7	XL/L	47.0	11.0	L	L
32	60.0	5.8	26.2	8.0	2.0	X/L	63.0	10.0	X/L	X/L
					(2) Soils in which Short and Standard methods disagree					
33	28.0	17.8	46.2	8.0	2.4	L/M	36.3	11.0	L	L - L/M[3]
34	43.0	18.0	29.0	10.0	4.3	L	47.5	24.0	L/M	L - L/M[3]
35	52.2	13.0	28.8	6.0	1.4	L	48.0	25.0	L/M	L
36	50.2	8.2	35.4	6.2	3.1	L	67.2	8.0	X/L	L
					(3) Heavy Soils which do not respond in usual fashion					
37	11.8	34.4	33.8	20.0	3.7	M	14.9	23.0	M	M/H
38	13.4	27.2	54.0	5.4	3.0	M	15.7	32.0	M	?
39	13.8	26.0	40.2	20.0	3.4	M	14.4	23.0	M	? ?
40	15.6	29.6	35.8	19.0	4.0	M	17.1	25.0	M	?
41	15.6	35.6	32.2	16.6	7.9	M	18.7	41.0	M	?

[1] Borderline case

[2] All poor at M, M/H and H rates

[3] Both rates described were used and found satisfactory

of 1961 since little or no frost was experienced over the previous winter, with the result that there was no frost mulch. Lack of frost does not affect soils with lower clay and higher sand contents to the same degree. With the exception of these soils therefore the pattern is clear, and the 1961 results confirmed a decision taken previously to restrict commercial usage of the herbicide to medium or lighter than medium soils and to prohibit its use on heavy land and on poor, rough seed beds.

This pattern of relationship was clearly of paramount importance to the future commercial promotion of the herbicide since it offered a means of forecasting correct rates. General practice in the same season (1961) had shown that to be one rate out of true was not serious either from the control or safety point of view. To be two rates out was likely to be serious.

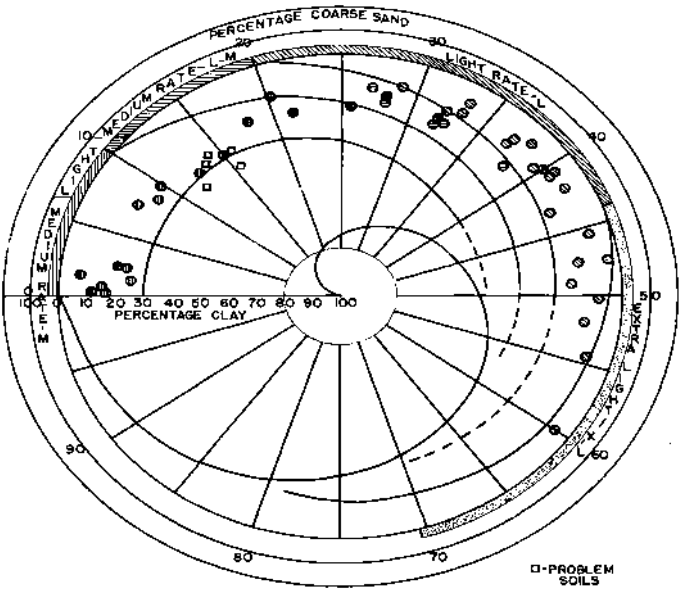

Figure 2.—Graph shows relationship between satisfactory rates of endothal/propham and course sand/clay percentages by standard "long" method.

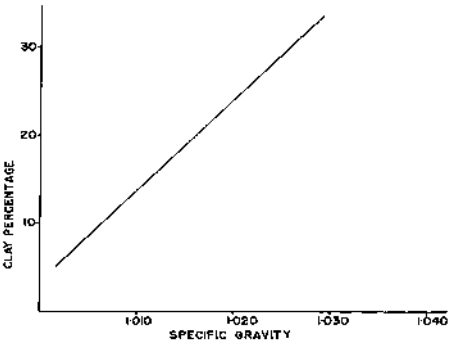

Figure 3.—Conversion graph—gravity to clay percentage.

Unfortunately the use of the Standard Long method of soil examination could not be considered from the commercial angle as it was too time consuming. Therefore, attempts were made to produce a quick easy method suitable for handling hundreds, perhaps thousands of soil samples from prospective commercial users of the herbicide.

Such a method was devised in the summer of 1961 and has since been put into current use. Briefly a special apparatus was developed suitable for handling a small but representative sample of the soil (5.0 grams) and permitting its sedimentation without too much handling. This is shown in Figure 6. The specific gravity of the supernatant liquid was taken by hydrometer after 5 hours. A series of readings on representative soils previously examined by the Standard Long method permitted the plotting of a graph whereby clay content could be read from specific gravity (Figure 3). Coarse sand was determined by sieving the sediment in the tube through a BSS 85 mesh sieve (178 ± 4 microns). The major advantages of this SHORT method are that initial drying of the soil, handling of the soil in the apparatus, and drying of the sand after sieving are all much easier and quicker than in the Standard Long method (a full report on this Short method is in an appendix to this paper).

Short Method Results

The coarse sand and clay values obtained by this Short method are listed in Table 23 alongside the results for the Long method. These values have been plotted on a graph (Figure 4) with the rates of use divisions as determined in Figure 2 (using

Long method data). It will be seen that the fit is almost as complete as it is in Figure 2. Four exceptions are listed which show a shift of one rate. In two of these, 33 and 34, both Light and Light/Medium rates were found to be satisfactory in practice and this means both Short and Long method forecasts would be accurate. In the cases of 35 and 36 an error of one rate would be made. The Short method forecast rate for trial 35 was in fact used and found to be safe. No data are available on the Short method forecast for trial 36.

The problem clay soils remain in the Medium category, near the upper limit. In practice the probability is that these soils would have responded normally if the seed bed had been good.

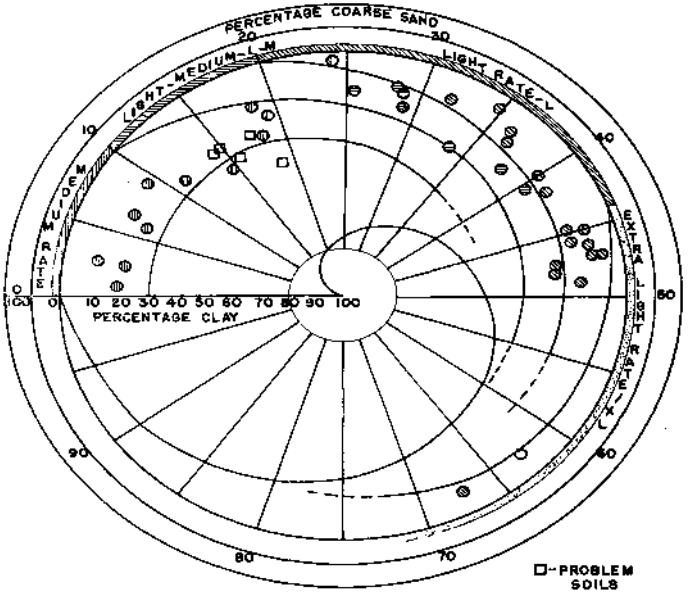

Figure 4.—Graph shows relationship between rates of use of endothal/propham and coarse sand/clay percentages by "short" method.

Table 3.—Soil mechanical analysis data and herbicide rates from 61 commercial sites.

No.	Coarse sand %	Clay %	Theoretical rate	Rate used in practice
A. Theoretical & practical rates identical Satisfactory use of Herbicide				
1	0.8[1]	18.0	M	M
2	1.0	14.0	M	M
3	1.0	10.0	M	M
4	1.09	9.0	M	M
5	1.1	11.0	M	M
6	1.8	10.0	M	M
7	1.9	8.0	M	M
8	2.8	4.0	M	M
9	18.4	11.0	M	M
10	21.6	37.0	L/M	L/M
11	21.7	37.0	M	M
12	22.4	17.0	L/M	L/M
13	22.6	11.0	L/M	L/M
14	22.9	15.0	L/M	L/M
15	23.5	11.0	L/M	L/M
16	23.9	12.0	L/M	L/M
17	25.6	9.0	L/M	L/M
18	25.8	8.0	L/M	L/M
19	25.9	9.0	L/M	L/M
20	26.5	11.0	L/M	L/M
21	26.6	7.0	L	L
22	26.9	10.0	L/M	L/M
23	28.0	18.0	L/M	L/M
24	28.0	14.0	L/M	L/M
25	28.0	9.0	L	L
26	28.8	9.0	L	L
27	29.0	16.0	L/M	L/M
28	30.9	9.0	L	L
29	31.2	4.0	L	L
30	31.5	10.0	L	L
31	31.7	18.0	L/M	L/M
32	32.0	6.0	L	L
33	33.2	6.0	L	L
34	33.9	10.0	L	L
35	34.1	15.0	L - L/M[3]	L/M
36	35.2	11.0	L	L
37	36.9	17.0	L - L/M[3]	L
38	39.0	8.0	L	L
39	39.4	4.0	L	L
40	40.3	13.0	L	L
41	43.6	18.0	L	L
42	44.5	17.0	L	L
43	47.0	7.0	L	L
44	51.9	12.0	L	L
B. Dose applied higher than theoretical				
i) Weed control good—no check to Beet				
45	31.5	10.0	L	L/M
46	36.8	14.0	L	L/M
47	39.5	14.0	L	L/M
48	54.2	10.0	L	L/M
ii) Weed control OK slight check to Beet[1]				
49	30.2	6.0	L	L/M
50	33.8	7.0	L	L/M
51	34.8	10.0	L	L/M
52	38.1	17.0	L	L/M
53	54.9	15.0	L	L/M
54	58.1	7.0	X/L	L
55[2]	67.0	12.0	L	L/M
C. Dose applied Lower than theoretical				
i) Weed control good				
56	0.4	19.0	M	L/M
57	21.6	6.0	L/M	L
58	33.2	17.0	L/M	L
ii) Weed control moderate to poor				
59	1.6	8.0	M	L/M
60	1.8	13.0	M	L
61	23.0	11.0	L/M	X/L

[1] Check only slight and grown out in 7-14 days.
[2] Overdosed by nearly x2 and check on beet severe.
[3] Borderline case.

Commercial Results

The pattern o£ relationship so far described was very promising and clearly opened the way for the development of a forecasting scheme. A further step was therefore taken. Sixty-one farms where the herbicide had been used commercially in 1961 were visited. In all cases the dosage of herbicide used was decided by the farmer on his own assessment of his soil type. A mechanical analysis was not done in advance of the treatment in any instance. The only requirement here was the assurance that the application had been accurate such that the rate of use to soil type relationship was a true one. After the season the soil from these farms was examined by the Short method and results are listed in Table III. They are plotted on a circular graph in Figure 5. In this instance the clay axis is logarithmic since we are interested only in the lower clay values (below 40%).

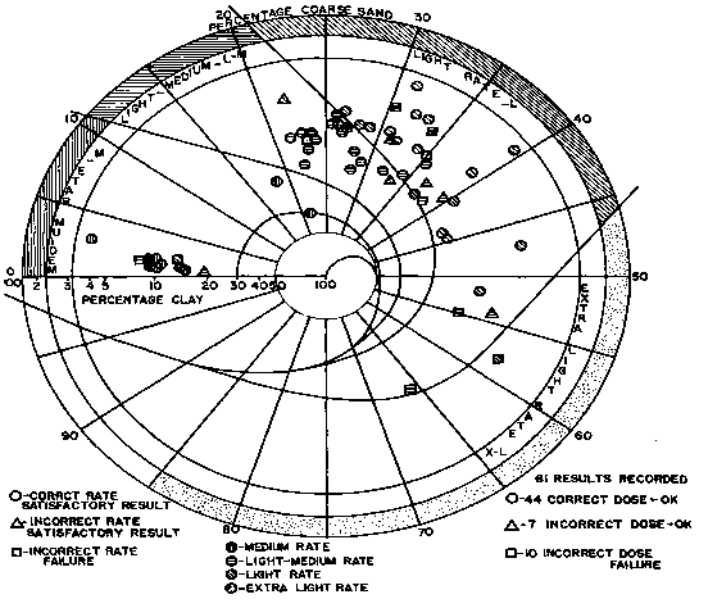

Figure 5.—Graph shows relationship between commercial results and "short" method of soil analysis—1961 results recorded.

Study of Figure 5 will show that the fit here is also good. In most of the instances of use the correct dosage for the soil (by consideration of coarse sand and clay fractions) was chosen and results were satisfactory both in terms of control of weeds, and safety to beet. In several instances however a wrong dosage was employed. In some of these results nevertheless were still satisfactory. In others results were unsatisfactory either as a result of damage, (where too high a rate was used) or through a failure to control the weeds, where too low a dose was used.

It should be pointed out that all the soils studied have a relatively low organic matter content, the highest recorded here being 6.9%. Higher organic contents tend to inactivate the herbicide and nullify the scheme described above. Further there is

Figure 6.—Upper left, apparatus for long method; upper right, apparatus for short method; lower left and right, apparatus for short method (general view).

an indication that the unusual soils of the Dutch Polders do not respond in the expected pattern. These soils comprise very large proportions of fine sand, and while the theoretical dose by the coarse sand/clay method is often the Medium dose, in practice a Light Medium or even a Light rate is adequate. This situation has not been found with British soils.

Summary

The results presented show that a reliable means of relating the required rate of use of the herbicide to the soil type (in terms of coarse sand and clay) has been evolved. The method is essentially arbitrary in that a range of suitable rates was adopted and the soils have been categorised in terms of this range. Nevertheless a high degree of accuracy in forecasting the required rate is obtained. This, coupled with the fact that an error of one dose from the true is not a serious error suggests that the technique can be used with confidence on a commercial scale. This in fact is being done at present by the writer's Company in preparation for the 1962 growing season.

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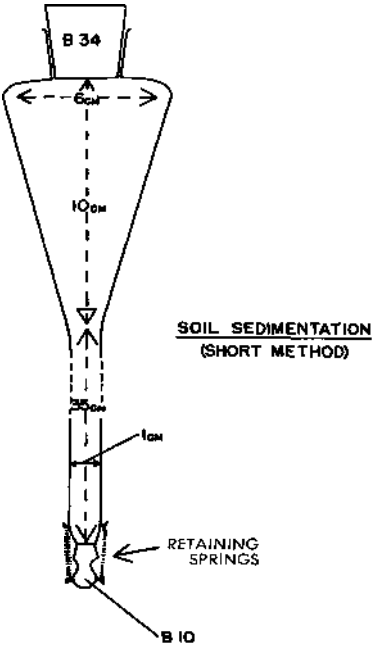
Appendix

The "Short" Method

The sample as received in the laboratory is re-sampled to give a weight of approximately 50 g. Preliminary partial drying of the field sample may be necessary before a true laboratory sample can be taken. This is carried out by infra-red heating. The 50 g. laboratory sample is dried at a temperature not exceeding 130°C. The time varies from 0.5 to 2 hours according to the initial water content of the samples.

The dried sample is broken down with a pestle and mortar in such a way as to disrupt all aggregates but not to reduce the actual particle size. It is then passed through a BSS 10 mesh



sieve (1676 ± 22 microns and from that part of the sample, which should be at least 95%, weigh 5.00 g into the apparatus (see diagram), via the smaller end, the larger end being stoppered. Add 25 ml of 0.125% Calgon (Sodium Hexametaphosphate) in the same manner, using the solution to wash the soil completely into the reservoir. Replace the smaller stopper and, keeping the slurry in the reservoir, shape with semi-rotary motion for three minutes. Place the apparatus in a clamp in an upright position with the reservoir at the top, and allow to stand for 5 hours ± 10 minutes.

Remove the apparatus from the stand and carefully decant the supernatant liquor without disturbing the silt layer into a 25 ml measuring cylinder and take the gravity at 20°C ± 1°C. Read from the prepared graph the percentage of clay.

Return the apparatus to the stand and wash the total sediment through a BSS 85 mesh sieve (178 ± 4 microns). Wash on the sieve with distilled water and dry at 110°C for 0.25 hours. Weigh and calculate the percentage of coarse sand.

Acknowledgments

The authors wish to acknowledge the assistance of their colleagues B. H. Bagnall, J. J. B. Caldicott and D. J. Minter who applied the herbicide and did the counts, and B. D. Owen and Mrs. M. Goodchild who assisted in the development of the short method of soil analysis.

Effect of Plant Spacing and Fertilizer on Yield, Purity, Chemical Constituents and Evapotranspiration of Sugar Beets in Kansas I. Yield of Roots, Purity, Percent Sucrose and Evapotranspiration[1]

G. M. HERRON, D. W. GRIMES AND R. E. FINKNER[2]

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Sugar beets have been grown for more than 50 years in southwest Kansas. Little research information has been published regarding the particular problems of this area. Many of the practices have been imported from other areas and adapted to local conditions. The success of sugar beet production has varied greatly, with low yields of inferior quality beets being a constant problem.

In recent years yields have been increasing, however, the quality has shown somewhat less improvement. Higher yields could be attributed to many factors such as mechanization, better varieties and culture practices, improved irrigation facilities and greater use of commercial fertilizers.

Because of the limited research information available, it seemed desirable to investigate some of the practices that might contribute to yield and quality of sugar beets. In the field studies, special emphasis was placed on soil moisture, plant population and fertilization. Soil moisture was not included as a variable in this phase of the study, however, it was deemed desirable to catalog the total moisture use and pattern of extraction from the soil on certain treatments. The importance of soil moisture management has been pointed out by Haddock (8,9)[3].

Plant, population studies have received much attention (2,3,4,8,18). Most studies have indicated high plant populations, e.g., 30,000 plants per acre, are desirable both from the standpoint of yield and quality. Some studies have shown narrower rows, 16 to 20 inches, would be more desirable than the current practice of 22 to 24 inch row width. The wider row width is more popular because of convenience in the use of farm machin-

[1] Contribution No. 68, Garden City Branch Experiment Station, Garden City, Kansas, a branch of Kansas State University, Manhattan, Kansas. Cooperative research between the Garden Citv Branch Experiment Station and the American Crystal Sugar Company, Rocky Ford, Colorado.

[2] In charge of irrigated soil fertility studies at Garden Citv Experiment Station: formerly in charge of irrigation studies at Garden City Experiment Station and now at Atnes. Iowa: and Manager Research Station, American Crystal Sugar Company, Rocky Ford. Colorado, respectively.

[3] Numbers in parentheses refer to literature cited.

ery. Uniform stands of beets have been stressed for yield and quality (4,14).

Many research papers have been published on the influence of fertilizer and manure on sugar beet yield and quality. Nuckols (12) summarized the crop rotation and manure studies in western Nebraska. Haddock (8) studied the influence of fertilizer and manure on sugar beets *in* Utah. Gardner and Robertson (6) studied nitrogen requirements of beets in Colorado. In Kansas, fertilizer studies were reported by Carlson and Herring (1) and Grimes (7).

In recent years, attention has been directed toward the problems associated with the use of "excessive" amounts of nitrogen fertilizer and the resulting lower beet quality (5,10,11,13,15,17). The "low quality beet" is an old problem that has been somewhat dormant. Present emphasis on quality can be attributed to two main factors: (1) production research is less demanding than formerly; and (2) the development of the scientific tools and personnel with which to solve the "low quality beet" problem. This stage of research achievement marks an important milestone in sugar beet technology.

Results reported here are concerned with plant spacing and fertilizers as related to yield, quality of beets, and soil moisture use in southwest Kansas.

Materials and Methods

Location and soil site: The field study was conducted in 1959 and 1960 on the Irrigation Project of the Garden City Branch Experiment Station located about 10 miles northwest of Garden City, Kansas. The soil is classified as Richfield silty clay loam (silted phase)[4]. It is calcareous to a depth of 8 to 12 inches while the second foot of depth is noncalcareous. For the surface layer, a pH value of 8.0 is typical; organic matter content of 1.8 to 2.2%; available phosphorus is medium to high; and potassium is very high. This soil is deep, moderately permeable and at field capacity it will retain about two inches of available moisture per foot of depth.

Wheat was the preceding crop each year. Conventional early seedbed preparation and tillage was practiced each year. The variety HH-1 was seeded at the rate of 5.5 pounds per acre in 22 inch rows April 4, 1959, and April 15, 1960. The experimental design was a split-plot in randomized blocks with four replications. Factorial fertilizer treatments were main plots (6 rows X 75 feet long) and spacing treatments were sub-plots (6 rows X 25 feet long).

[4] "Silted phase" is used to characterize soils in this area that have been modified in the surface layer by the accumulation of calcareous silt from ditch irrigation water.

Fertilizer: Each year 12 fertilizer treatments, consisting of three levels of nitrogen, two levels of phosphorus and two levels of potassium were used. In 1959, 80 pounds per acre of N as ammonium nitrate and 100 pounds per acre of K_20 was broadcast and worked in prior to seeding the beets. An additional 60 pounds of N as anhydrous ammonia was side dressed June 12, immediately preceding the second irrigation. In 1960 all of the N and K_20 was applied and worked in prior to seeding beets. During both years the phosphate was applied below and to the side of the seed with fertilizer attachments on the beet drill.

Spacing: Plant spacings of 8, 12, and 16 inches were accomplished by hand thinning as soon after emergence as possible. Plant counts during the season and at harvest indicated very close agreement to the planned populations of 35,640, 23,760, and 17,820 plants per acre.

Irrigation: The irrigation schedule was such that no moisture stress occurred. In 1959 the beets were irrigated six times and in 1960, five irrigations were necessary. Soil samples were taken periodically to a depth of 6 feet, by one foot increments, to determine the moisture use. Moisture was determined gravimetrically except for a few cases in 1960 when the neutron moisture meter was used. Rainfall was added to the moisture used from the surface foot. Approximately 8 inches of rainfall were received during the moisture use periods which is near normal for this area.

Sampling: The beets were harvested in the last week of October in 1959 and the first week of November in 1960.

Yields were determined by harvesting the four center rows by 23 feet in length. This allowed for two feet between sub-plots (spacing treatments) and two rows between adjacent fertilizer treatments. Two samples of the harvested beets from each subplot were taken to the American Crystal Research Laboratory, Rocky Ford, Colorado, for determination of percent sucrose, percent purity, various amino acids and mineral constituents. Laboratory procedures will be described in Part II of this paper.

Statistical methods of Snedecor (16) were employed in the analyses of the data[5].

Results and Discussion

Yields of Roots: In 1959 the root yields ranged from 26 to 31 tons per acre while in 1960 the yields varied from 29 to 32 tons per acre (Tables 1 and 2). This yield level is considerably above the area average for these years. Adequate and timely irrigation contributed to the high yields.

[5] Appreciation is expressed for the assistance provided by Dr. H. C. Fryer and associates, Statistical Laboratory, Kansas State University, Manhattan, Kansas,

Table 1.—The effect of fertilizer treatments and plant spacing of beets in 22 inch rows on yield of roots, % sucross, and % purity of the extract. 1959.

Fertilizer treatment			Yield of roots plant spacing, inches				Sucrose plant spacing, inches				Purity plant spacing, inches			
"N	*ftfh*	K^O	~8	12	16	Average	~~*	12"	16	Average	~8	12	16	Average
Lb/acre			Tons per acre				Percent				Percent			
0	0	0	26.46	27.34	26.86	26.88	16.1	15.8	15.3	15.8	91.1	91.4	91.0	91.1
80	0	0	28.39	29.09	27.45	28.31	15.5	14.6	15.1	15.0	89.0	89.2	90.4	89.5
140	0	0	30.00	29.08	30.43	29.84	14.7	13.9	14.0	14.2	87.1	88.6	87.2	87.6
0	120	0	27.22	26.36	27.76	27.11	16.4	15.6	15.4	15.8	90.7	91.1	90.5	90.8
80	120	0	27.59	29.97	28.89	28.82	15.9	14.8	14.6	15.1	89.1	88.8	90.6	89.5
140	120	0	30.70	30.55	30.39	30.54	15.2	14.5	13.8	14.5	87.8	88.6	86.4	87.5
0	0	100	27.42	27.36	27.63	27.47	15.8	15.8	14.8	15.6	89.1	91.7	90.7	90.5
80	0	100	26.24	28.38	30.03	28.22	15.2	15.3	14.2	14.9	91.2	89.8	90.0	90.4
140	0	100	27.97	30.34	29.63	29.31	15.2	15.6	14.9	15.2	89.2	89.8	89.2	89.4
0	120	100	27.40	28.08	26.81	27.43	16.0	15.4	15.9	15.8	91.2	88.3	89.8	89.8
80	120	100	29.73	31.28	28.86	29.96	15.1	14.9	14.2	14.7	89.6	89.6	88.7	89.3
140	120	100	29.23	29.62	29.86	29.57	14.5	14.4	14.6	14.5	87.9	87.9	87.2	87.7
Average			28.20	28.96	28.72	28.62	15.5	15.0	14.7	15.1	89.4	89.5	89.3	89.4

Statistically significant factors @ .05 level	Nitrogen **(Linear)** Spacing Spacing x N X P_2O_5 X K_2O	**Nitrogen (Linear)** N x K_2O Spacing Spacing **x** K_2O	Nitrogen **(Linear)** Nitrogen **x** K_2O

Table 2.—The effect of fertilizer treatments and plant spacing of beets in 22 inch rows on yield of roots, % sucrose, and % purity of the extract. 1960.

Fertilizer treatment			Yield of roots plant spacing, inches				Sucrose plant spacing, inches				Purity plant spacing, inches			
N	P_2O_5	K_2O	8	12	16	Average	8	12	16	Average	8	12	16	Average
Lb/acre			Tons per acre				Percent				Percent			
0	0	0	29.22	30.07	30.54	29.94	17.9	17.7	17.8	17.8	91.2	89.8	88.7	89.5
60	0	0	30.45	30.68	31.02	30.72	17.6	17.5	17.4	17.5	89.5	90.5	88.9	89.7
120	0	0	30.14	30.83	31.46	30.81	16.8	16.4	16.5	16.6	89.1	88.5	88.3	88.6
0	120	0	30.24	30.79	31.53	30.85	18.2	17.8	17.9	18.0	89.5	88.3	89.9	89.3
60	120	0	28.82	30.34	30.38	29.93	17.7	17.0	17.2	17.3	90.4	89.9	89.8	90.0
120	120	0	30.76	31.32	32.33	31.47	16.9	17.2	16.6	16.9	88.1	87.5	87.1	87.6
0	0	100	30.23	30.80	28.68	29.90	17.4	16.8	16.5	16.9	90.4	89.2	89.3	89.6
60	0	100	29.16	30.82	30.31	30.10	16.8	17.2	17.5	17.1	90.4	89.5	86.9	88.9
120	0	100	29.50	31.74	31.29	30.84	17.3	16.6	17.1	17.0	88.3	88.3	90.0	88.9
0	120	100	29.71	30.32	28.98	29.67	17.7	17.5	17.4	17.5	91.2	89.0	89.4	89.9
60	120	100	30.44	31.00	30.19	30.54	17.6	17.5	17.2	17.4	90.2	89.7	88.9	89.6
120	120	100	30.81	31.58	31.66	31.35	17.2	17.1	15.6	16.6	89.5	89.4	85.5	88.1
Average			29.96	30.86	30.70	30.51	17.4	17.2	17.0	17.2	89.8	89.1	88.6	89.2
Statistically significant factors @ .05 level				Nitrogen (Linear) Spacing				Nitrogen (Linear) Spacing				Nitrogen (Linear) Spacing		

Both spacing and fertilizer treatments resulted in statistically significant yield differences. Yield increase from fertilizer was in a linear relationship to the applied nitrogen. Neither phosphorus nor potassium significantly influenced yields. This soil is generally considered to be adequately supplied with available phosphorus and potassium. Nitrogen carry-over from previous crops and the short summer fallow period after the previous wheat crop would account for a high level of soil nitrogen.

A spacing interval of 12 inches produced the highest average yield both years. Lowest average yield resulted from the 8-inch spacing interval. Although mean differences due to spacing were less than one ton per acre, the differences were statistically significant at the 5% probability level. Under the conditions of these experiments, the highest yields occurred with approximately 25,000 plants per acre.

Percent Sucrose: Each year the percent sucrose was in an inverse and linear relationship to the amount of applied nitrogen. As an average, each 14 pounds per acre of fertilizer N decreased the sucrose content by 0.1%. Regression equations are given in Table 3. Phosphorus or potassium did not influence sucrose content.

Plant spacing interval of 8 inches resulted in the highest sucrose content each year. Decreasing the population resulted in lower sucrose content. Regression equations are given in Table 3. In 1959, each inch of space above 8 inches decreased the sucrose content by 0.1%, but, in 1960 about two inches of additional space above 8 inches were required for the same reduction in sucrose.

Coefficients of determination (Table 3) indicate 39%, of the variation in sucrose could be attributed to spacing and 47%, to nitrogen in 1959. The degree of association was much less for both nitrogen and spacing in 1960.

Percent Purity: Purity of the extract had an inverse and linear relationship to the amount of applied nitrogen each year. Phosphorus or potassium did not significantly influence purity either year.

Plant spacing had no effect on purity in 1959, but in 1960, purity was significantly lowered bv wider within-row plant-spacing intervals. Purity varied from 88% to 91% during both years of the study (Tables 1 and 2).

Gross Sugar Yield: Total sugar production was not significantly influenced by fertilizer treatments. Yield increases resulting from fertilizer were sufficient to compensate for the accompanying decrease in percent sucrose. Average gross sugar pro-

duction was 8,614 pounds per acre in 1959 and 10,503 pounds per acre in 1960 (Table 4).

Table 3.—Regression equations, relating the rate of nitrogen and within-row spacing to sucrose content and weight per beet-

Year	Regression equation	Correlation coefficient, r	Coefficient of determination r^2
	% Sucrose (Y) — N rate (X)		
1959	$\hat{Y} = 15.65 - 0.0078X$	0.683**	16-6%
1960	$\hat{Y} = 17.59 - 0.0066X$	0.604**	36.5%
	% Sucrose (Y) — plant spacing (X)		
1959	$\hat{Y} = 16.78 - .0906X$	0.628**	39-4%
1960	$\hat{Y} = 17.79 - .0469X$	0.364**	13.2%
	Beet size (Y) — plant spacing (X)		
1959	$\hat{Y} = .2889 + .1553X$	.973**	94.7%
1960	$\hat{Y} = .3694 + -1594X$	.975**	95.1%

* Significant at 0.05 n = 36
** Significant at 0.01

Table 4.—The mean calculated value for gross sugar production and extractable sugar as influenced by nitrogen, phosphorus, or potassium fertilizers and spacing intervals of plants.

Treatments	Gross sugar		Extractable sugar[1]	
	1959	1960	1959	1960
	cwt/A		cwt/A	
Nitrogen, lbs/A of N				
0	85.35	105.62	74.36	89.73
60- 80	86.11	105.18	74.09	90.97
120-140	87.01	104.31	72.48	86.70
LSD (.05)	NS	NS		
Phosphorus, lbs/A of P2O5				
0	85.40	106.12	73.52	88.67
120	86.90	103.96	73.77	89.60
LSD (.05)	NS	NS		
Potassium, lbs/A of K2O				
0	85.92	104.20	73.41	88.88
100	86.39	105.88	73.87	89.39
LSD (.05)	NS	NS		
Spacing, inches between plants				
8	87.06	104.48	74.57	89.37
12	87.04	106.01	74.52	89.32
16	84.36	104.62	71.86	88.73
LSD (.05)	2.45	NS		

[1] Extractable sugar was calculated on the means of four replications, using the formula developed for the Rocky Ford factory for the 1954 through 1958 campaigns.

In 1959, the 16-inch spacing resulted in significantly lower sugar production than other spacing treatments. In 1960, there were no differences resulting from spacing treatment. In 1959, there was a significant interaction between plant population and the potassium fertilizer treatment. This did not occur in 1960, therefore, it may have resulted from random variation.

Extractable Sugar Yield: The extractable sugar, i.e., the amount of sugar bagged, was decreased by nitrogen fertilization. Only the last increment of nitrogen fertilizer decreased the extractable sugar a substantial amount. These data would seem to confirm the observation that "excess" nitrogen results in "low quality beets". It will be noted that under the conditions of these experiments, rather liberal amounts of nitrogen could be applied (60 to 80 pounds per acre) before the "excess" N was instrumental in reducing extractable sugar. From an economical standpoint it would not have been profitable to supply the larger rates of nitrogen.

Phosphorus and potassium fertilizers produced no substantial difference in extractable sugar. In general, the same trend existed between gross sugar production and extractable sugar regardless of the application of phosphorus and potassium. Data for the average of the variables studied are reported in Table 4.

During both years, little differences were observed between 8- and 12-inch with-in row spacing, but 16-inch spacing decreased both gross and extractable sugar.

Size of Roofs: In 1959, applied N gave a significant linear relationship to beet size, but not in 1960 (Figure 1). Beet size was significantly increased both years by a wider within-row plant spacing. An additional inch of in-row space resulted in a 0.16 pound increase in the average weight per beet. Regression equations, correlation coefficients and coefficients of determination (Table 3) indicate that plant spacing was the principal factor determining average beet size in these experiments.

Large size roots generally are assumed to contain less sucrose and extractable sugar. It is very difficult to determine that beet size directly influences sucrose content or purity because the factors that alter the size of beets also change such things as the nutritional status of a plant. For example, increasing the spacing interval increases the amount of soil nitrogen and moisture available to an individual plant. Thus, if top growth isn't markedly increased, there may be sufficient "excessive" nitrogen to decrease sucrose content. In these experiments it was impossible to separate the effect of beet size on sucrose content or purity by covariance analyses, thus, large beets were not necessarily lower in

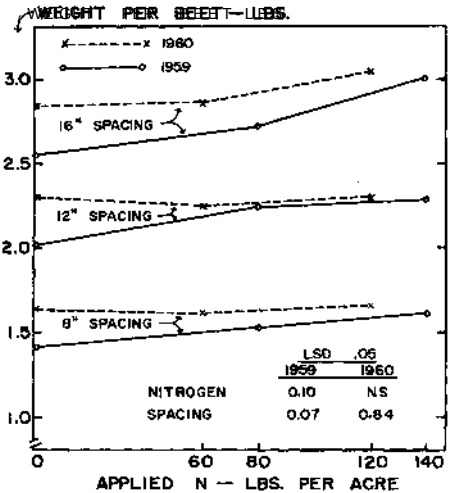

Figure 1.—The relationship between sugar beet size and applied nitrogen fertilizer.

quality. Loomis and Ulrich (10) found no direct relationship between beet size and percent sucrose.

Soil Moisture Use: Moisture use (evapotranspiration) was determined only for fertilizer treatments receiving the three nitrogen levels on plots receiving phosphorus and potassium. Data reported are averages of four replications.

Accumulative seasonal evapotranspiration for 1959 ranged from 32 to 40 acre-inches. Differences were not significant for fertilizer or spacing, however, the interaction was statistically significant. In 1960 evapotranspiration varied from 32 to 35 acre-inches, with differences not statistically significant (Table 5).

Maximum rate of moisture use of about 0.3 inch per day occurred in July. After the period of peak use, the moisture requirement declined until harvest when little more than 0.1 inch was used daily (Figures 2 and 3).

The extraction pattern (Figure 4) indicates most of the moisture use was from the surface three feet of soil. Moisture extraction from the 4th, 5th, and 6th feet of depth was mostly

Table 5.—Evapotranspiration of sugar beets as influenced by nitrogen fertilization and dthin-row plant spacing at Garden City, Kansas.

Applied N, lb/A	1959				1960			
	Plant spacing in 22 inch rows				Plant spacing in 22 inch rows			
	8	12	16	Average	8	12	16	Average
	Acre- inches per acre							
0	35.3	32.1	34.4	33.9	34.4	32.8	32.5	33.2
(50- 80	37.6	34.0	39.0	36.8	32.5	32.6	33.6	32.9
120-140	39.8	40.4	36.9	39.1	35.0	32.6	35.1	34.2
Average	37.6	35.5	36.8	36.6	33.9	32.7	33.7	33.5

DifTerences in evapotranspiration are not statistically different for nitrogen and spacing treatments, however, a significant interaction between spacing and nitrogen occurred in 1959.

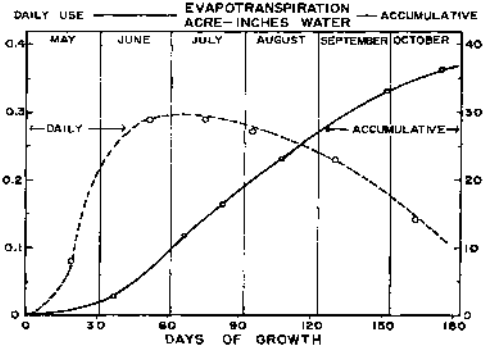

Figure *2*.—Daily and accumulative evapotranspiration for the 1959 sugar beet crop. Each sampling date is an average of 36 plots.

during July and August when the period of peak use occurred. Some moisture extraction could have occurred below the six-foot sampling depth but it would represent a small percent of the total.

Rainfall and irrigation water management drastically influences moisture extraction patterns. In Figure 4 all rainfall was included in the surface foot, although a heavy rain or rains on successive days may have influenced the moisture status at lower soil depths. A moisture extraction pattern in which a high percentage of total evapotranspiration is from near the surface would generally indicate the absence of moisture stress during the season.

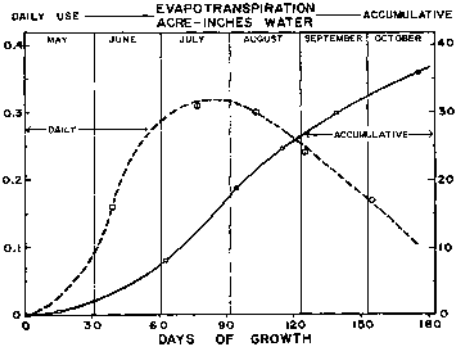

Figure 3.—Daily and accumulative evapotranspiration for the 1960 sugar beet crop. Each sampling date is an average of 36 plots.

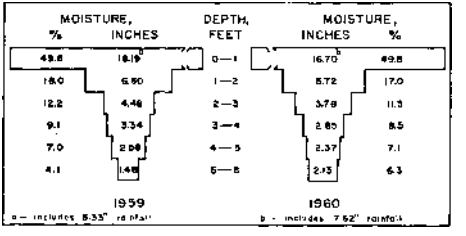

Figure 4.—The pattern of moisture extraction in a six-foot profile by sugar beets.

Summary

Yields of roots, percent sucrose, percent purity, gross sugar production and extractable sugar were used to evaluate the influence of applied fertilizer. Nitrogen exerted a major influence while phosphorus and potassium had little or no effect. The decrease in percent sucrose and purity associated with applied nitrogen was about equal to the gain in root yield. The 60 to 80 pounds per acre of N appeared not to be "excessive", however, 120 to 140 pounds per acre was definitely "excessive" and reduced quality. This soil was quite high in natural fertility.

Within-row plant spacing was important to sugar production. Wide spacing resulted in inferior beet quality, lower percent

sucrose and purity. The 8- and 12-inch spacing proved more desirable than the 16-inch spacing in all respects. Larger size beets were produced when the spacing interval was increased. The beet quality, i.e., percent sucrose or purity, could not be directly related to beet size through covariance analyses.

Moisture use ranged from 32 to 40 acre-inches per acre. July was the month of maximum daily evapotranspiration of near 0.3 acre-inch per day. Nitrogen fertilizer seemed to increase moisture requirements. The surface 3 feet of depth supplied most of the soil moisture, however, the lower horizions were important during the peak daily use period.

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Effect of Plant Spacing and Fertilizer on Yield, Purity, Chemical Constituents and Evapotranspiration of Sugar Beets in Kansas II. Chemical Constituents[1]

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The chemical constituents in sugar beets delivered to factories for processing have become a major concern in many producing areas of the United States. Their chemical make-up is determined by genetics, environment, and interactions between those two factors. Several investigators (3,5,19,22)[3] have demonstrated genetic control of many chemical characteristics. Field environment studies concerned with chemical composition have been conducted by many researchers most of whom varied moisture content of the soil and/or applied different fertilizers at different rates (4,5,6,7,8,9,10,12,14,16,17,18,19,20,21,23). Ogden et al. (16) and Herron et al. (12) reviewed many of these reports which have shown a close inverse relationship between nitrogen fertilization and sugar beet quality. Several experimenters (4,5,7,9,14, 17,19,20,23) have elucidated to some degree the effects of nitrogen fertilizer on nonsugars. All have shown that nitrogen constituents of sugar beet roots increase with increased nitrogen.

Complexing results are not surprising as soils are extremely variable and dynamic and are affected by micro and macro environments. Fertilizer results and recommendations, in general, are specifically applicable only to the general location in which tests were conducted. The investigation reported here was undertaken to:

1. Study effects of fertilizer treatments on several individual nonsugar constituents of sugar beets.

2. Study effects of varying plant populations on nonsugar constituents.

Materials and Methods

Experimental data were obtained from extensive field experiments at Garden City, Kansas, in 1959 and 1960, previously reported by Herron et al. (12). Sodium and potassium were de-

[1] Contribution No. 70, Garden City Branch, Kansas Agriculture Experiment Station, Garden City, Kansas, a branch of Kansas State University, Manhattan, Kansas. Cooperative research between the Garden City Branch Experiment Station and the American Crystal Sugar Company, Rocky Ford, Colorado.

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[3] Numbers in parentheses refer to literature cited.

termined on the Beckman DU Spectrophotometer, utilizing the method proposed by Bauserman and Olson (1), and are reported as percent. Phosphate was estimated colorimetrically as molybdenum blue (13, 15) and is reported in parts per million. Galactinol and raffinose evaluations were determined by paper chromatography similar to that described by Brown (2). Amino acids were determined by paper chromatographic procedures reported by Hanzas (11). The total amino acid content is the sum of the individual amino acids found by paper chromatography. All paper chromatographic determinations are reported as percent of dry substance. Total nitrogen was determined by a modified micro-Kjeldahl nesslerization procedure (17), and is reported as percent of dry substance. Sugar and purity were analyzed by standard sugar analysis procedures.

The amounts of nitrogen were applied in an arithmetical progression, treatments were subdivided into their linear and quadratic effects as shown in Table **1.**

Results and Discussion

Sixteen different chemical constituents were studied. Nitrogen produced the greatest effects on the constituents studied, as expected, because 11 of the 16 characters studied contained nitrogen atoms. Effects of nitrogen were not limited to compounds containing nitrogen atoms as it significantly affected 13 of the attributes studied in the 1959 test and 15 of those studied in the 1960 test (Tables 1 and 2). In all cases except for glutamic acid in the 1960 test, nitrogen effects were linear, i.e., as the rates of nitrogen fertilizer increased, the chemical constituents being studied increased proportionally. There were only three nitrogen quadratic effects and again only glutamic acid in the 1960 test snowed a greater quadratic than linear effect.

Adding phosphorus fertilizer produced a significant increase in P_2O_5 content of beet roots both years and a significant increase *in* the aspartic acid content in 1960 (Table 2). Potassium caused a significant increase in the glutamic acid content of beets in 1959 but no other significant effects.

The three different population levels produced significant differences in both years for the elements: sodium, potassium and phosphorus, and the amino acids: glycine, valine, leucine and total amino acid. Significant differences among populations also were detected in the 1960 data for glutamine, gamma amino butyric acid and alanine. In all cases (Table 2) decreased plants per acre caused an increase in the above mentioned elements and amino acids, or as beets were spaced closer together they contained less per plot of the elements and the amino acids studied.

Table 1.—Levels of significance obtained for nitrogen, phosphate, potassium and populations for 16 different characters in the Kansas fertility and spacing tests.

Source of variation	Na.	Phos.	K	Raff.	Gal.	Total nit.	Aspar. acid	Glu- tamic	Aspara- gine	Gluta- mine	Gly.	(G.A.B.A.	Alanine	Valine	Leucines	Total amino acids
								1959 Test								
Nit. L	···	NS	***	NS	NS	***	***	***	***	***	***	***	***	***	***	***
Nit. Q	NS	NS	NS	NS	NS	NS	NS	*	NS	NS	**	NS	NS	NS	NS	NS
P	NS	**	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
K	NS	NS	NS	NS	NS	NS	NS	**	NS	NS	NS	NS	NS	NS	NS	NS
Nit. L x P	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
Nit. Q x P	NS	NS	NS	NS	*	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
Nit. L x K	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	*	NS	NS	NS	NS	NS
Nit. Q x K	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
P x K	NS	NS	NS	NS	NS	NS	NS	··	NS	NS	*	NS	NS	NS	NS	NS
Nit. L x P x K	NS	NS	NS	NS	NS	NS	NS	**	NS	NS	NS	NS	NS	NS	NS	NS
Nit. Q x P x K	NS	NS	NS	NS	NS	NS	NS	*	NS	NS	NS	NS	NS	NS	NS	NS
Pop.	**	NS	***	NS	NS	*	NS	NS	NS	NS	*	NS	NS	***	**	*
Pop. x Fert.	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
Pop. x N	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	*	NS
Pop. x P	NS	NS	NS	NS	NS	NS	NS	NS	*	NS	NS	NS	NS	NS	NS	NS
Pop. x K	*	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
Pop. x N x P	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
Pop. x N x K	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
Pop. x P x K	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
Pop. x N x P x K	NS	NS	*	NS	NS	**	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS

Table 1 continued, next page.

Table 1.—Levels of significance obtained for nitrogen, phosphate, potassium and populations for 16 different characters in the Kansas fertility and spacing tests. *Continued*

1960 Test

Nit. **L**	***	• .	***	NS	*	***	***	NS	***	***	***	***	***	***	***	**
Nit. Q	NS	NS	NS	NS	NS	NS	NS	*	NS	NS	NS	NS	NS	NS	NS	NS
P	NS	*	NS	NS	NS	NS	*	NS	NS	NS	NS	NS	NS	NS	NS	NS
K	NS	NS	NS	NS	NS	NS	**NS**	NS	NS	NS	NS	NS	NS	NS	NS	NS
Nit. L x **P**	NS	NS	NS	NS	NS	NS	**NS**	NS	NS	NS	NS	NS	NS	NS	NS	NS
Nit. Q x **P**	NS	NS	NS	NS	NS	NS	*	NS	*	*	NS	NS	NS	NS	NS	*
Nit. L x **K**	NS	NS	*	NS	NS	NS	NS	*	*	NS	.*	*	..	*	NS	*.
Nit. Q x **K**	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
P x **K**	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
Nit. L x P x **K**	NS	NS	NS	NS	NS	*	NS	NS	*	*	*	NS	*	NS	NS	*
Nit. Q x P x K	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
Pop.	***	*	*	NS	NS	NS	NS	NS	NS	**	..	**	..	..	• *	..
Pop. x Pert.	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	*	NS	NS	NS	NS	NS
Pop. x N	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	*	NS	NS	NS	NS	NS
Pop. x P	• •	NS	NS	NS	NS	NS	NS	NS	NS	*	*	*	*	NS	NS	*
Pop. x K	NS	*	NS	NS	NS	NS	NS	NS	NS	**NS**	NS	NS	NS	NS	NS	NS
Pop. x N x P	NS	NS	NS	NS	*	NS	NS	NS	NS	**NS**	NS	NS	NS	NS	*	NS
Pop. x N x K	NS	NS	NS	NS	NS	NS	NS	NS	NS	**NS**	NS	NS	NS	NS	NS	NS
Pop. x P x K	NS	NS	**	NS	NS	NS	NS	NS	NS	**NS**	NS	NS	NS	NS	NS	NS
Pop. x N x P x K	NS	NS	NS	NS	NS	**NS**	NS	NS	NS	**NS**	NS	NS	NS	NS	NS	NS

NS = Nonsignificant
*** — Significant at the .1% level
** = Significant at the 1% level
* = Significant at the 5% level

Table 1 shows several significant first and second order interactions. The 1959 data also had two significant third order interactions. Although these interactions were statistically significant, their variances were relatively low, compared with those of the main effects and therefore are relatively unimportant. Several of the higher order interactions merely reflect significant interactions that occurred at lower levels. Data which produced the significant first order interactions are given in Table 3 and discussed below.

Effects of different fertilizers and populations are discussed under elements, carbohydrates, and nitrogenous constituents.

Chemical Elements

Effects of different fertilizers on sodium, potassium and phosphorus content of the beets varied (Table 2). As nitrogen fertilizers were increased, sodium and potassium contents of the beets significantly increased both years. Phosphorus content was significantly decreased by nitrogen in 1960. The trend was similar but not significant in 1959.

Phosphate fertilizer produced no significant change in the amount of sodium and potassium in the roots either year. However it significantly increased the amount of phosphorus content both years, which indicates that the more phosphorus applied, the more the beets absorb from the soil.

Potassium application did not significantly change the level of sodium, potassium or phosphorus in beets either year.

Different plant populations resulted in significant differences in sodium and potassium content of the beets both years, with lower populations and greater differences occurring together. The higher population (8-inch spacing compared with 16-inch spacing) lowered sodium and potassium content of beets indicating that the wider the spacing, the more sodium and potassium the plants absorbed from the soil. Phosphate in the 1960 test was significantly greater than in the 1959 test of greater population. This probably was a chance occurrence because it did not occur in 1959. The "F" value for 1960 barely reached significance.

Carbohydrates

Beets were analyzed for both raffinose and galactinol. A significant linear nitrogen response for 1960 is shown in Table 1. The galactinol content was lowest at the 0 nitrogen level and increased with increasing rates of nitrogen fertilizers. Raffinose was not significantly affected by changing rates of nitrogen. Potassium and phosphate applications did not significantly affect raffinose or galactinol contents either year. None of the three different spacings significantly influenced raffinose or galactinol content.

Table 2.—The average mean effects for nitrogen, phosphate, potassium and populations for 16 different characters in the Kansas fertility and spacing tests.

Nitrogen applied	Na.	Phos.	K	Raff.	Gal.	Total nit.	Aspar. acid	Glu-tamic	Aspara-gine	Gluta-mine	Gly.	G.A.B.A.	Alanine	Valine	Leucines	Total amino acids
								1959 Test								
0	.039	677	.214	.387	.253	.72	.127	.021	.116	.56	.083	.183	.053	.049	.084	1.25
80	.047	614	.227	.379	.244	.82	.151	.022	.151	.74	.113	.202	.070	.064	.105	1.60
140	.055	634	.245	.365	.266	.96	.182	.030	.184	1.00	.163	.242	.101	.085	.141	2.12
LSD (0.05)	.004		.008			.05	.015	.004	.024	.10	.013	.018	.015	.010	.015	.18
LSD (0.01)	.005		.011			.06	.021	.005	.033	.13	.018	.025	.020	.014	.020	.24
Degrees of freedom = 2 and 33																
Phosphate applied																
0	.046	587	.228	.385	.262	.81	.152	.023	.154	.76	.120	.211	.072	.065	.105	1.64
120	.048	696	.229	.369	.247	.85	.155	.026	.146	.78	.119	.206	.077	.067	.115	1.67
LSD (0.05)		75														
LSD (0.01)		101														
Degrees of freedom=1 and 33																
Potassium applied																
0	.046	657	.227	.383	.254	.82	.151	.022	.149	.76	.124	.205	.071	.066	.110	1.63
100	.048	627	.230	.372	.255	.84	.156	.027	.152	.78	.114	.213	.078	.067	.110	1.68
LSD (0.05)								.004								
LSD (0.01)								.005								
Degrees of freedom = 1 and 33																
Populations																
8" Spacing	.044	635	.223	.389	.253	.81	.148	.023	.139	.73	.110	.205	.070	.058	.100	1.57
12" Spacing	.047	667	.230	.364	.252	.84	.155	.023	.155	.77	.117	.205	.074	.069	.110	.165
16" Spacing	.050	623	.233	.378	.258	.86	.156	.026	.156	.81	.131	.216	.080	.071	.119	1.74
LSD (0.05)	.003		.005			.03					.014			.007	.011	.13
LSD (0.01)	.004		.007											.009	.014	.18

Nitrogen applied	Na.	Phos.	K	Raff.	Gal.	Total nit.	Aspar. acid	Glu-tamic	J^spara-gine	Gluta-mine	Gly.	G.A.B.A.	Alanine	Valine	Leucines	Total amino acids
								1960 Test								
0	.031	713	.251	.280	.185	.71	.068	.019	.082	.367	.087	.117	.035	.043	.069	.87
60	.039	676	.262	.280	.193	.79	.071	.017	.096	.417	.102	.123	.046	.049	.076	.99
120	.045	622	.277	.279	.201	.92	.081	.023	.115	.499	.134	.142	.060	.060	.091	1.18
LSD (0.05)	.007	55	.009	....	.016	.06	.004	.004	.014	.045	.012	.011	.007	.007	.008	.09
LSD (0.01)	.009	79	.013			.09	.006		.018	.061	.017	.014	.010	.009	.012	.12
Degrees of freedom = 2 and 33																
					.191	.80	.071	.021	.099	.429	.105	.127	.048	.052		
					.196	.81	.075	.019	.096	.426	.110	.128	.046	.050		
							.004									
					****	****										
Potassium applied																
0	.038	664	.261	.273	.194	.82	.072	.019	.099	.435	.109	.131	.048	.052	.080	1.02
100	.039	676	.265	.286	.193	.78	.074	.021	.096	.420	.106	.124	.046	.050	.077	1.00
LSD (0.05)																
LSD (0.01)																
Degrees of freedom = 1 and 33																
Populations																
8" Spacing	.036	699	.258	.284	.194	.79	.072	.020	.094	.392	.101	.123	.044	.047	**.074**	.96
12" Spacing	.039	651	.264	.276	.188	.80	.071	.019	.098	.431	.106	.124	.045	.051	.079	1.00
16" Spacing	.042	660	.268	.279	.198	.82	.076	.020	.101	.460	.117	.135	.052	.054	.083	1.08
LSD (0.05)	.003	37	.007							.040	.008	.008	.005	.004	.005	.07
LSD (0.01)	.004									.054	.011	.010	.007	.005	.007	.09
Degrees of freedom = 2 and 72																

Total Nitrogen and Amino Acids

The total nitrogen content of all of the amino acids significantly increased with increased nitrogen applications. In 1959 all nitrogenous compounds tested showed a linear significant increase at the 0.1% level or .001 level. The glutamic acid and the glycine acid contents also showed a significant quadratic response, but the linear response accounted for a greater portion of the variation. In 1960 all linear responses except those for glutamic acid and total amino acid were significant at the 0.1% level. The total amino acid was significant at the 1% level; glutamic acid quadratic interactions, at the 5% level. In both years glutamic acid showed a significant quadratic effect. Although the values are significant, the amounts of glutamic acid in the beets (Table 2) were so small compared with other amino acids, importance of glutamic acid effects seems doubtful.

Phosphate applications produced only one significant effect for aspartic acid in 1960. Otherwise phosphate failed to affect significantly any nitrogenous character studied. Potassium applications significantly affected only glutamic acid and only in 1959. That both phosphorus and potassium affected only one amino acid and were not consistent both years indicate that those two fertilizers were not affecting the nitrogen content of the beets.

Effects of different populations on the total nitrogen and amino acid contents are shown in Table 2. The amount of nitrogen in the beets increased as spacing of beets increased from 8 to 16 inches, in nearly all cases, although some were not significant. Total amino acid content was significantly increased both years as beets were spaced farther apart. Sparse populations provide less competition for minerals and other soil elements so individual plants would be expected to gather more nitrogen and other mineral elements. Test results verily that hypothesis.

Significant First Order Interactions

There were several first order interactions in both years (Tables 1, 3, and 4). In 1959 (Table 3) only 7 significant interactions were found but 20 were found in 1960 (Table 4).

The 1959 data showed a significant N X P interaction for galactinol, due primarily to nitrogen. At the 0 rate of phosphate, galactinol was fairly high, but it dropped with application of 80 pounds of nitrogen, only to increase significantly with the 140-pound nitrogen rate. At the 120-pound phosphate rate, galactinol values were not significantly changed for any nitrogen rate.

Table 3.—Seven significant first order interactions which occurred in the 1959 test.

Galactinol

P \ N	0	80	140	Mean	
0	.263	.236	.287	.262	
120	.245	.251	.246	.247	NS
Mean	.253	.244	.266		

NS

N × P LSD (0.05) = .036

Glycine

K \ P	0	120	Mean	
0	.130	.119	.124	
100	.109	.120	.114	NS
Mean	.120	.119		

NS

P × K LSD (0.05) = .018

Sodium

Pop \ K	0	100	Mean	
1	.041	.048	.044	LSD (0.05)
2	.048	.047	.047	.003
3	.050	.050	.050	
Mean	.046	.048		

NS

K × P = Difference between 2 population means at the same level of K = .004.
Difference between 2 K means for the same population = .005.

Glycine

K \ N	0	80	140	Mean	
0	.082	.114	.176	.124	
100	.082	.112	.149	.114	NS
Mean	.082	.113	.163		

LSD (0.05) = .013

N × K LSD (0.05) = .018

Leucines

Pop \ N	0	80	140	Mean	
1	.076	.101	.123	.100	LSD (0.05)
2	.089	.109	.134	.110	.011
3	.087	.106	.165	.119	
Mean	.084	.105	.141		

LSD (0.05) = .015

N × Pop = Difference between 2 population means at the same level of N = .019.
Difference between 2 nitrogen means for the same population = .022.

Glutamic acid

K \ P	0	120	Mean	
0	.023	.022	.022	LSD (0.05)
100	.023	.031	.027	.003
Mean	.023	.026		

NS

P × K LSD (0.05) = .006

Asparagine

Pop \ P	0	120	Mean	
1	.129	.150	.139	
2	.164	.148	.155	NS
3	.169	.144	.156	
Mean	.154	.146		

NS

P × Pop = Difference between 2 population means at the same level of P = .028.
Difference between 2 P means for the same population = .030.

The significant interaction of N X K for glycine was due to potash applications with a high nitrogen level. Glycine significantly increased with increased rates of nitrogen. Potash has no particular influence on glycine at the first two nitrogen levels, however at the 140-pound nitrogen rate, a 100-pound application of potash significantly restricted the build-up of glycine.

There were two significant interactions of P X K. Glutamic acid was not significantly affected by either P or K when used individually, but when used together glutamic acid content increased significantly. But glycine had the highest value at 0 rate of P and/or K, either of which seemed to reduce glycine content of beets. K, at 100 pounds per acre without P significantly reduced glycine content.

Population interacted significantly with all three fertilizers. Leucines showed a significant population X N interaction. Nitrogen significantly increased leucines in all three populations. However, the 12-inch population spacing showed more leucines at 0- and 80-pound nitrogen rates than the 16-inch spacing did. At the 140-pound nitrogen rate, greatest amount of glycine was in the widest spacing.

The significant interaction for asparagine due to population X P was primarily caused by the switch which took place at the 8-inch and 12-inch spacings and at 0- and 120-pound rates of P. Least asparagine occurred with 8-inch spacing and zero P rate. At the 12-inch spacing and 0-rate asparagine significantly increased and was higher than at the 120-pound P rate with 12-inch spacing. In general asparagine increased as population increased with no P applied. When P was applied, the trend was opposite although values differed only slightly and were not significant.

The significant interaction of population X K for sodium was due to the significant increase in sodium content from K applied to the 8-inch spacing; at the other two spacings no difference in sodium content was detected. Also sodium content increased significantly between 8-inch and the other two spacings when no K was applied. At the 100-pound K rate no significant differences were found in sodium content of the three populations.

Most of the interactions in 1959 were barely significant and may not be important biologically. The second and third order interactions appear to have little or no meaning and resulted primarily from first order interactions.

Table 4 shows the simple interactions detected from the 1960 data. There were four significant interactions of N X P for

amino acids. With the 0 nitrogen rate values were approximately the same for both P levels. At the 60-pound N rate, P seems to have stimulated uptake of nitrogen, and all amino acid values were higher with the 120-pound N rate over P at 0. N at 120 pounds per acre, P reduced uptake of N so P at the 0 rate resulted in higher amounts of amino acids. This was true of all four N X P interactions.

Eight N X K interactions were significant (Table 4). Seven were amino acids; one was potassium. The interaction was produced by the 100-pound rate of K stimulating uptake of nitrogen and potassium at the 0 nitrogen rate. While at the 120 N rate, the 100-K rate reduced uptake of nitrogen ions compared with uptake at the zero-K rate. This complete reversal in all cases was the primary cause of the significant interactions. At the medium-(60 lb) nitrogen rate, amino acid values were approximately equal for both levels of K. That so many amino acids showed significant interactions of N X K definitely may have biological meaning even though they appeared in only the 1960 test. These interactions indicate that with high amounts of nitrogen, applying potash likely would raise beet quality because potash under those conditions seems to reduce the amount of amino acids in the beets.

Populations showed eight significant interactions with fertilizers: one each with nitrogen and potassium and six with phosphorus (Table 4). The significant N X population interaction resulted from the 12-inch spacing producing more glycine at 0-nitrogen rate and less at the 60-pound N rate, compared with the other two spacings.

Six interactions of P X populations were significant; five with amino acids and one with sodium. In all 8-inch spacings studied, (attributes were higher at the zero-P rate than at the 120 P rate. The reverse was true with the 16-inch spacing; the 12-inch spacing gave highest interaction values at the 0 level. The reversals produced significant interactions, indicating that to obtain highest quality beets, one would plant high populations (8-inch spacing) and apply 120 pounds of phosphate fertilizer.

Most interactions in 1960 were somewhat like those in 1959, i.e., barely significant. But the 1960 interactions differed by following a definite trend. That there were 20 significant interactions in 1960 and that thev followed definite trends indicates strongly that fertilizer elements used did not act independently and that chemical composition of beets depends on interactions among fertilizer elements applied. The data also show that fertilizer applied should be governed somewhat by beet populations.

Table 4.—Twenty significant first order interactions which occurred in the 1960 test.

Potassium

K \ N	0	60	120	Mean	
0	.246	.256	.282	.261	
100	.256	.268	.271	.265	NS
Mean	.251	.262	.277		

LSD (0.05) = .009
N × K LSD (0.05) = .013

Glutamic acid

K \ N	0	60	120	Mean	
0	.015	.017	.024	.019	
100	.023	.018	.021	.021	NS
Mean	.019	.017	.023		

LSD (0.05) = .004
N × K LSD (0.05) = .006

Asparagine

K \ N	0	60	120	Mean	
0	.078	.095	.124	.099	
100	.086	.096	.104	.096	NS
Mean	.082	.096	.115		

LSD (0.05) = .014
N × K LSD (0.05) = .019

Glycine

K \ N	0	60	120	Mean	
0	.080	.101	.146	.109	
100	.094	.103	.122	.106	NS
Mean	.087	.102	.134		

LSD (0.05) = .012
N × K LSD (0.05) = .018

Gamma amino butyric acid

K \ N	0	60	120	Mean	
0	.116	.122	.154	.131	
100	.118	.125	.131	.124	NS
Mean	.117	.123	.142		

LSD (0.05) = .011
N × K LSD (0.05) = .015

Alanine

K \ N	0	60	120	Mean	
0	.030	.048	.067	.048	
100	.040	.044	.054	.046	NS
Mean	.035	.046	.060		

LSD (0.05) = .007
N × K LSD (0.05) = .010

Valine

K \ N	0	60	120	Mean	
0	.040	.050	.064	.052	
100	.047	.048	.056	.050	NS
Mean	.043	.049	.060		

LSD (0.05) = .007
N × K LSD (0.05) = .009

Total amino acids

K \ N	0	60	120	Mean	
0	.83	.97	1.27	1.02	
100	.90	1.01	1.09	1.00	NS
Mean	.87	.99	1.18		

LSD (0.05) = .09
N × K LSD (0.05) = .13

Table 4.—Twenty significant first order interactions which occurred in the 1960 test. *Contiiuied*

Sodium

Pop \ P	0	120	Mean	
1	.037	.033	.036	LSD (0.05)
2	.040	.037	.039	.003
3	.039	.044	.042	
Mean	.039	.038		

NS

P × Pop = Difference between 2 population means at the same level of P = .004.
Difference between 2 P means for the same population = .006.

Glutamine

Pop \ P	0	120	Mean	
1	.407	.376	.392	LSD (0.05)
2	.449	.414	.431	.040
3	.431	.490	.460	
Mean	.429	.426		

NS

P × Pop = Difference between 2 population means at the same level of P = .057.
Difference between 2 P means for the same population = .060.

Glycine

Pop \ P	0	120	Mean	
1	.104	.097	.101	LSD (0.05)
2	.102	.109	.106	.008
3	.109	.125	.117	
Mean	.105	.110		

NS

P × Pop = Difference between 2 population means at the same level of P = .012.
Difference between 2 P means for the same population = .014.

Gamma amino butyric acid

Pop \ P	0	120	Mean	
1	.124	.121	.123	LSD (0.05)
2	.128	.120	.124	.008
3	.128	.142	.135	
Mean	.127	.128		

NS

P × Pop = Difference between 2 population means at the same level of P = .010.
Difference between 2 P means for the same population = .012.

Alanine

Pop \ P	0	120	Mean	
1	.049	.040	.044	LSD (0.05)
2	.046	.044	.045	.005
3	.050	.054	.052	
Mean	.048	.046		

NS

P × Pop = Difference between 2 population means at the same level of P = .007.
Difference between 2 P means for the same population = .008.

Total amino acids

Pop \ P	0	120	Mean	
1	.98	.93	.96	LSD (0.05)
2	1.03	.97	1.00	.07
3	1.03	1.13	1.08	
Mean	1.01	1.01		

NS

P × Pop = Difference between 2 population means at the same level of P = .10.
Difference between 2 P means for the same population = .11.

Table 4.—Twenty significant first order interactions which occurred in the 1960 test. *Continued*

Asparatic acid

P \ N	0	60	120	Mean	
0	.066	.006	.081	.071	
120	.069	.075	.080	.075	NS
Mean	.068	.071	.081		

LSD (0.05) = .005
N × P LSD (0.05) = .006

Asparagine

P \ N	0	60	120	Mean	
0	.084	.088	.123	.099	
120	.079	.103	.106	.096	NS
Mean	.082	.096	.115		

LSD (0.05) = .014
N × P LSD (0.05) = .019

Glutamine

P \ N	0	60	120	Mean	
0	.372	.390	.524	.429	
120	.361	.444	.474	.426	NS
Mean	.367	.417	.499		

LSD (0.05) = .045
N × P LSD (0.05) = .064

Total amino acids

P \ N	0	60	120	Mean	
0	.89	.92	1.22	1.01	
120	.84	1.06	1.14	1.01	NS
Mean	.87	.99	1.18		

LSD (0.05) = .09
N × P LSD (0.05) = .13

Glycine

Pop \ N	0	60	120	Mean	
1	.079	.101	.122	.101	LSD (0.05)
2	.094	.091	.131	.106	.011
3	.088	.114	.149	.117	
Mean	.087	.102	.134		

LSD (0.05) = .012
N × Pop = Difference between 2 population means at the same level of nitrogen = .015.
Difference between 2 nitrogen means for the same population = .018.

Phosphate

Pop \ K	0	100	Mean	
1	719	680	699	LSD (0.05)
2	638	665	651	37
3	636	684	660	
Mean	664	676		

NS
K × Pop = Difference between 2 population means at the same level of K = 52.
Difference between 2 K means for the same population = 57.

High nitrogen rates affected some sugar beet constituents that influence sugar beet quality. Herron et al. (12), using the same experimental material, pointed out that nitrogen decreased sugar content and purity. In this test, several nonsugars, which are melassigenic, increased significantly with increased N. Our data confirm voluminous reports of nitrogen effects on sugar beet constituents and quality: mainly that excessive nitrogen definitely increases melassigenic components of beets.

Summary

Data presented show that the different fertilizers and different amounts of the same fertilizer drastically affect chemical composition of sugar beets. Effects of nitrogen fertilizers were most striking as N significantly increased nearly all characteristics studied. Phosphorus mainly affected the phosphate content of beets. Potassium showed no consistent main effects but both potassium and phosphorus showed significant interactions with nitrogen. Phosphorus and population also produced several significant interactions.

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