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Some changes during storage in McIntosh, Cortland and Wealthy varieties of apples.

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SOME CHANGES DURING STORAGE IN McINTOSH
CORTLAND AND WEALTHY VARIETIES OF APPLES



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SOME CHANGES DURING STORAGE IN
McINTOSH, CORTLAND AND WEALTHY
VARIETIES OF APPLES

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University of Massachusetts

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Some changes during storage in McIntosh, Cortland,
and Wealthy varieties of apples.

INTRODUCTION

One of the major problems of the fruit grower after he has spent money and effort in producing such fruits as apples is to be able to keep them as long as possible at their prime value.

The apple is formed of living cells which undergo a process of metabolism. If sound storage practices are desired it is important to become acquainted with the different phases of this process.

It is known that temperature and atmosphere are the main factors controlling the rate of ripening (result of metabolism). Though many studies have been made on the physiology of the apple, it was not possible to find in the literature a comparative study of the principal changes occurring during the storage of these common New England varieties (McIntosh, Cortland, and Wealthy) under temperatures and atmospheric conditions usually found in this region.

Firmness was tested at intervals. This is the best physical measure of the state of ripening in storage. Among chemical changes the following were studied during the storage period: Water content, soluble solids, total acidity, pH, ascorbic acid and pectin. These tests were run on the three varieties stored at 32° F. (0° C.), 40° F. (4.4° C.) with high relative humidity, 40° F. (4.4° C.) with low relative humidity and in a controlled atmosphere room at 40° F. (4.4° C.) with average atmospheric composition of 3 per cent oxygen and 6 per cent

carbon dioxide.

The results of these experiments are given in the form of graphs which are discussed.

REVIEW OF LITERATURE

In a great many studies pressure tests have been used as an empirical means of measuring the internal changes inside the fruit by calculating their overall effect on softness of flesh. It has been found practical when comparing different treatments and storage conditions.

Morris (33) of Washington Agricultural Experiment Station was the first to suggest the use of a mechanical device to test the firmness of fruit. Murneek (34) in 1921 presented a test for maturity of the pear which was based on the same principle. In 1925 Magness (28) introduced an improved type of pressure tester and the same apparatus that he described was used for this study.

As a rule all the different varieties tested by numerous workers: Kidd and West (24, 25) in England, Smock and VanDoren (47), Magness (28) and many others in the United States, have been found to show a decrease in firmness of flesh from picking to late storage. If some treatment retards the rate of ripening, it decreases the rate of softening. It has been found by Carre (9) that the disappearance of pectic materials (protopectose) from the middle lamellae may be correlated with a decrease in firmness of flesh of the fruit.

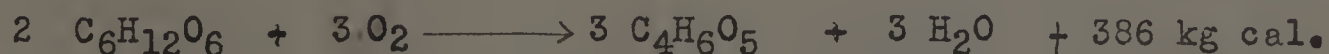
Loss of water from apples in storage has been studied by H. K. Archbold (1) who reported his results as changes in dry weight.

From his findings the dry weight of Bramley's Seedling does not change much from October to April and only fluctuates between 11.5 and 10.5 per cent. Smith (45) made investigations on the water loss from apples by evaporation. Bramley's Seedling, Cox's Orange Pippin apples and Beurre Clairgeau pears were used as material for these studies. A first rapid decrease in the rate of evaporation followed by a more stable condition was observed.

Haynes and Archbold (21) suggest that loss of water is due to transpiration which in turn is dependent on the size of the apple, the nature and condition of its skin, the size of the aperture at the calyx end and the humidity of the storage room.

Early work in carbohydrate changes in the apple was done by Bigelow and others in 1905 in the United States, followed by the researches of Magness (29), Neller (37), Pennington (39), and Plagge (40). All found that chemical changes did not differ greatly from variety to variety, and that the rate at which they take place is modified by such factors as temperature and humidity.

Sugars and acids are in close correlation. Indeed acid and especially malic acid in the apple can be synthesized from sugar by oxidation of fructose:



In turn malic acid upon oxidation gives:



Haynes and Archbold (21) suggest that acid is continually formed in the course of normal respiration of the apple and a correlation has been observed between the quantities of fructose and acid present in

different varieties. Plagge and Gerhardt (41), studying the acidity changes of Grimes and Jonathan apples, found some correlation between loss of total acid and soggy breakdown.

Some of the first work on changes in the acid content of stored apples was conducted by Haynes (20) in England with Bramley's Seedling. It was found that when very immature this variety contained 2.5 per cent total acid (as malic acid) and only 0.5 per cent after about 10 months of storage at 32° F. (0° C.). Other workers found many different concentrations of acid in different varieties of apples at different stages of ripeness.

As for the acid mixture found in apples, Franzen and Helvert quoted by Evans (13) found:

Oxalic acid	0.001 per cent
Succinic acid	4.22
Malic	69.57
Citric	24.95
Lactic	1.18

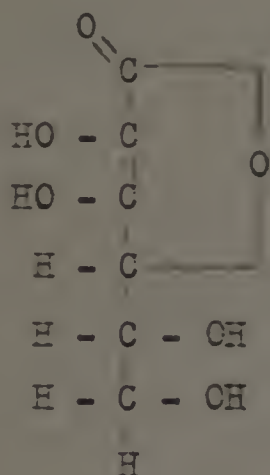
He points out also that the acid mixture may vary from apple to apple and from variety to variety. As far as the calculation of total acidity in malic acid is concerned, this author states "Assuming that 70 per cent of malic acid and 25 per cent of citric acid are present, the error by calculating citric acid as malic is about 3 percent, but as the total amount of acid present is small, usually less than 1 per cent, the error introduced by calculating the whole as malic acid is negligible."

Most of the soluble solids in the apple are sugars and from different analyses by Evans (13) and Kidd and West (25) about three times

as much fructose as glucose was found. Kidd and West (25) studying the influence of controlled atmosphere on Lane's Prince Albert, found in relation to carbohydrate that increasing concentration of carbon dioxide retards loss of carbohydrates and alcohol-insoluble materials but appears to accelerate loss of acids.

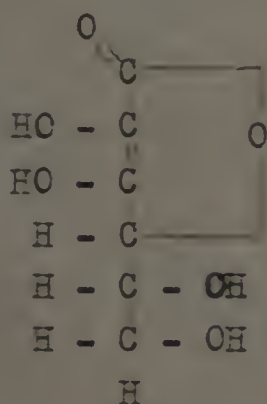
General Information About Ascorbic Acid

In the literature vitamin C is called "ascorbic acid". This chemical compound has an empirical formula of $C_6H_8O_6$, and a graphic formula of

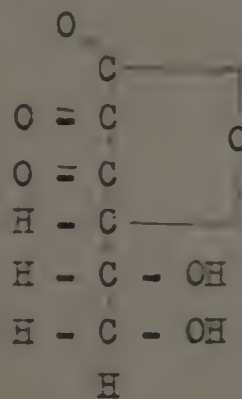


l - Ascorbic Acid

When ascorbic acid is oxidized it becomes dehydroascorbic acid and at that stage it loses part of its biological properties. It is supposed that this reaction occurs during the storage of apples.



L - Ascorbic Acid



Dehydro-1-Ascorbic Acid

In case of hydrolysis, there is a ring opening and a complete loss of vitamin activity occurs.

Ascorbic acid cannot be synthesized by the human body. For many people, fruits are the main source of that vitamin. Some fruits are richer than others: lemon, grapefruit and red pepper for instance are excellent sources for vitamin C. Other fruits such as apples may be considered as a fair source only.

Many studies have been made of the behavior of ascorbic acid under different conditions. The first studies were conducted on plants or fruits rich in ascorbic acid but research was also directed towards the other synthesizers of vitamin C.

Since its discovery by Holst and Frolich in 1907 many workers devoted their time to it and the work has been especially accelerated by the titration method developed by Tillmans and Hirsch (51) in 1932. Until that time only the biological assay method was used, giving much slower and less reliable results than the chemical method.

In 1931 Zilva, Kidd and West (55) found that among the varieties they had tested Bramley's Seedling contained the most ascorbic acid, followed by Lane's Prince Albert and Newton Wonder, which showed a little more antiscorbutic activity than average varieties.

Bracewell (5) in 1931 tested two varieties of apples. He found that Newton Wonder and Cox's Orange Pippin had approximately the same concentration and Lane's Prince Albert was between the latter and Bramley's Seedling. He found also that the same variety at different harvest seasons had about the same ascorbic acid concentration.

Smith and Fellers (46) in 1934 made a survey of 21 Massachusetts varieties of apples. They found that among them all Baldwin was the best source with a protective level of 4 g. At that time the biological assay was used to determine the amount of apple flesh necessary to protect a guinea pig against scurvy. Following Baldwin in order of protective level were: Ben Davis, 6 g.; Northern Spy, 5 g.; and Esopus, 7 g. For the varieties concerned in this study they found the following:

Cortland 12 g. after 2.5 months of storage

Wealthy 12 g. after 0.0 month of storage

McIntosh 25 g. after 0.0 month of storage

They found that the chromosome numbers of apples, diploid 34 and triploid 51, had little correlation with the vitamin C concentration, although Zilva suggested that it could be associated.

Kidson (26) found that Delicious had the lowest amount among common varieties. It showed a content of 1.9 - 5.1 mg. per 100 g.

Mockel, Wolf and Degen (32) in 1942 working in Germany on 80 German varieties found an average for the whole group tested of 13.4 mg. in 100 g. of apple including the peel. The apples considered best for their dessert value were not the best for their vitamin C content. They concluded that the apple may be regarded as a valuable source of ascorbic acid. In earlier work Wolf (31) found that the most commonly grown varieties in Germany, i.e. Ontario, Kaiser, Wilhelm, Goldparmäne, Shöner aus Boskoop, and Baumann's Reinette showed a vitamin C content from 10 to 20 mg. per 100 g. fresh weight.

Keys in New Zealand (23) in 1942 found that the variety Sturmer,

rich in vitamin C although not the richest, still contained after peeling the flesh 15 to 25 mg. of ascorbic acid per 100 g.

Literature on Changes in Ascorbic Acid Concentration in Apples During Storage

Bracewell (6) in 1930 reported that little loss occurred in ascorbic acid content of apples when stored in air at 1° C. (33°F.) or at 10° C. (50° F.) in a mixture of carbon dioxide, oxygen, and nitrogen for about three months. He pointed out, however, that the controlled atmosphere apple showed a greater loss of vitamin. In these experiments, only the flesh of the apple was tested; the core and peel were rejected.

Zilva and others (55) in 1930 found that apparently the vitamin is not affected in apples stored under frozen conditions. He notices that Bramley's Seedlings stored at 3° C. (37.4° F.) lost less vitamin than at 10° C. (50° F.). He concluded that the temperature rather than the composition of the atmosphere was the cause of the greater loss of ascorbic acid in controlled atmosphere storage.

Bracewell and others (5) stored Bramley's Seedling at -20° C. (-4° F.) during 4 months and found little loss of antiscorbutic activity. They found also that if the same variety was stored in air for 5 months at 3° C. (37.4° F.), there was no loss of antiscorbutic activity.

Fellers and others in 1933 (14), working with Baldwin apples, noticed that in a storage at 36° F. (2.22° C.) during 4 to 6 months there was a loss of about 20 per cent and after 8 months about 40 per cent loss. The juice obtained from freshly crushed apple was almost as rich as the flesh itself and little loss occurred in the first 24 hours.

Batchelder in 1934 (2) found a loss of approximately 17 per

cent during the storage of Delicious during 3 months at 7° C. (44.6°F.) and about 25 per cent after 6 months.

Zilva and others in 1935 (56) reported that apples kept at -5° C. (23° F.) for 3 months lost nearly all their ascorbic acid. If the apples were kept in air at -10° C. (14° F.) for the same period of time, the loss was about 70 per cent. After 6 months in vacuum at -20° C. (-4° F.) he found no reduction, but under the same conditions (vacuum) at -10° C. (14° F.) and 5° C. (41° F.) he noted a loss of 25 per cent.

Kroker (27) in 1939 found that in acid vegetables and fruits even at room temperatures the concentration of ascorbic acid remained about constant. On the other hand if products with a high pH are exposed to the action of oxygen, the loss may be very great. If the latter products are stored at low temperatures such as -18° C. (-0.4° F.), the losses are much reduced but the products must be immediately consumed after thawing to avoid losses which then occur rapidly.

Working with German varieties, Kessler (22) reported rapid loss in common storage and advised a temperature below 5° C. (41° F.) for the best conservation of ascorbic acid content. In 1941 (16) some tests done at Leipzig for the research station of Magdeburg showed that if there is loss of ascorbic acid in apple when kept in common storage, there is an increase in cool storage. The temperature of the room is not stated.

Also in Germany, about 37 varieties of apples from the south part of the country were tested by Wolf (54) who found that after a number of months at about 2.5° C. (36° F.), the loss of ascorbic acid

ranged from 0 to 50 per cent and at 0.5° C. (33° F.) it ranged from 3 to 30 per cent. The average concentration of the 37 varieties was 16.5 mg. per 100 g.

In 1942 Scupin (44) at Leipzig reported that an increase in ascorbic acid content of apple occurred in storage at -0.5° C. (31° F.) to -1° C. (30.2° F.) and a gain of 50 per cent was obtained when the apples were stored from December until July at these temperatures. On the other hand there was a little loss if the apples were kept at temperature of 1° C. (33.8° F.) to 5° C. (41° F.) from December until March 15th. When apples at a temperature of 4° C. (39.2° F.) to 12.5° C. (54° F.) were stored from March 15th to July 15th a loss of about 50 per cent occurred.

In 1943 Fish (15) determined the vitamin content of 8 different varieties grown in West Virginia which range from 1.7 to 4.5 mg. per 100 g. including the peel. After 2 months at 0° C (32° F.) to 2° C. (35.6° F.) the loss in ascorbic acid was 25.7 per cent and after 4 months 27.5 per cent. It was also found that during storage the loss was more rapid in the outside part of the apple.

In 1944 West and Zilva (53) conducted experiments to determine whether or not ascorbic acid was synthesized during storage as some other observations had deemed to indicate. From these experiments it was established that in the case of young Bramley's Seedling vitamin C was synthesized during storage at 3° C. (37.4° F.) and the rate of synthesis was found to slow down as the ratio of dehydroascorbic acid decreased.

Literature on the Influence of Carbon Dioxide Upon
Ascorbic Acid Concentration

Zilva (55) found that if there was more loss of vitamin C in controlled atmosphere than in cold storage at 3° C. (37.4° F.), it was due more to the higher temperature than to the composition of the atmosphere. He pointed out that further research was needed. Bracewell (6) also found a difference in the loss of vitamin C between 1° C. (33.8° F.) in air and 10° C. (50° F.) in controlled atmosphere after a storage period of 3 months.

Thornton (50) in 1927 found that the exposure of Northern Spy, Baldwin and Russet to various concentrations of carbon dioxide for as long as 10 days at various temperatures resulted in no detectable change in ascorbic acid content.

More work has been done on vegetables than on apples relative to the ascorbic acid changes in different concentrations of carbon dioxide. Some of the results obtained may give an idea of what might happen to ascorbic acid in apples under such conditions. In 1940 Sugawara (48b) working with spinach found that the highest vitamin C concentration was obtained in the sample which had been kept in the lowest concentration of carbon dioxide regardless of the temperature.

Different observations suggest that ascorbic acid may be recovered in part after extraction of the tissue with hydrogen sulfide, proving that during storage a part of the ascorbic acid becomes dehydroascorbic acid.

Thornton (49) reported in 1943 that for bananas removal from carbon dioxide storage resulted in a rapid increase in ascorbic acid as compared with fruit kept in common storage.

Effect of External and Internal Factors on the
Ascorbic Acid Concentration

The change in vitamin C concentration during storage is related to the relative amount in the product tested. The extent to which the amount may change with varying external and internal factors, such as climate, fertilization, spraying, genetic factors, will have some influence on the further discussion, therefore, the literature on this subject is interesting.

Bracewell (6) reported that the composition of the soil or the age of the tree had no practical effect on the vitamin C content of apples. Bramley's Seedling picked 14 days earlier than the main crop differs little from the main crop. On the other hand he reported that imported apples were richer in ascorbic acid content.

Zilva (55) confirmed that immature Bramley's Seedling picked in July had an activity per gram of tissue almost equal to that of the apple picked at the normal time.

Crane and Zilva (12) thought that the chromosome number of the apple could have a significance on the antiscorbutic value. They had noticed that Bramley's Seedling, a triploid variety, (51 chromosomes) was one of the richest in vitamin C. Therefore, they tested other triploid varieties and obtained results which were not very significant. In further experiment (11) they concluded that only a comparison of the triploid forms with the diploid forms from which they arose could give more definite indication. Indeed Gravenstein, a triploid variety, has about the same antiscorbutic potency as diploids, and Prince Albert, a diploid, has the same level as many triploid forms.

Potter and others (43a) studying the influence of fertilizers concluded that well manured trees synthesized more vitamin C than unmanured trees.

Fellers (14) found little difference in fruit from sprayed and unsprayed Baldwin trees.

Sansome and Zilva (43) favoring the theory that antiscorbutic potency is associated with triploidy decided to check their opinion on species of plants in which polyploidy can be easily produced. They chose tetraploid and diploid strains of tomato and found that all the tetraploids were about twice as potent as the diploids.

In further studies Fellers (14) found no apparent correlation between vitamin C content and chromosome number of apples. He noticed also that seasonal or other variations except storage had little effect on the antiscorbutic activity of the twenty-one Massachusetts-grown varieties tested.

Todhunter (52) reported no difference in ascorbic acid content in Rome Beauty apples receiving two different irrigation treatments; but Winesaps grown on land irrigated with 60 inches of water had higher ascorbic acid content than those from a location which received only 30 inches. He concluded that some plants seem to be influenced by external factors while others show little or no effects.

Kessler (22) found that vitamin C was influenced to a certain extent by light, condition of growth, place of origin, size of crop and manuring.

Sagawara (48) considered that the vitamin C content of the same variety may vary from place to place but in general rich varieties

will keep this character wherever they are grown.

Review of Literature on Pectin in Apple Fruits.

In 1824 Payen (38) and Braconnot (7) discovered pectin.

Since that time a considerable amount of work has been done to determine its chemical composition and structure, its physical properties, and its origin and metabolism in the living cell. Many facts have been brought to light but still there is much confusion in the explanation of them.

Very briefly, but step by step, the following covers the major advances in the chemistry of pectin. In 1848 Fremy (17) considered that during the ripening of fruits, the pectose is changed to soluble pectin under the action of acids in the cell sap. He found the compound that he called "pectose" or "protopectin" as insoluble material in the cell wall. In 1891-3 Mangin (30) located the pectic substances in the middle lamella of the cell walls, where they acted as a cement; he suggested that this material was calcium pectate. Since that time it has been found that protopectin occurs on the sides of the middle lamella. Buston (7b) suggests that protopectin may change to hemicellulose.

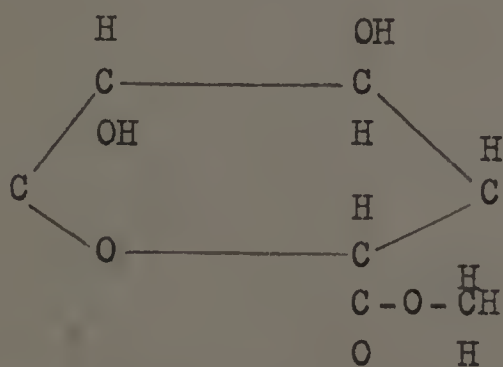
Fremy (17) observed that "pectose" or "protopectin" could be hydrolysed with boiling water or dilute acid. According to Bourquelot and Herissey (4) in 1898-99, the conversion of pectose into pectin is a result of enzyme activity. Fremy (17) found also what he called "pectic acid" compound obtained by heating of pectin in the presence of dilute alkali.

The chief constituent of pectic substances was discovered by

Ehrlich (12a) in 1917 to be galacturonic acid. According to this investigator the polygalacturonide nucleus present in all pectic compounds contains 4 molecules of galacturonic acid; Myers and Baker (29a) found that this nucleus is most likely octogalacturonic acid, formed by the union of two molecules of tetragalacturonic acid with the elimination of one molecule of water.

Bonner (3) pointed out that pectic acid does not occur as such in nature; the COOH groups are always combined. The most interesting combination is the methyl ester. All of the carboxyl groups may be esterified with methyl alcohol (then pure pectin) and there can be eight different states of esterification leading to the proposition of Baker (29a) that there are 8 molecules of galacturonic acid in the polygalacturonide nucleus.

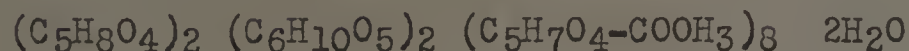
Bauer and Link (2a) found that by removing the methyl group combined with the carboxyl group with dilute alkali, he could obtain 8 and in some cases 10 galacturonic units.



Methylated Galacturonic Acid

Beside the polygalacturonic nucleus of the pectic compounds, they are thought by Ehrlich (12a), Nelson (37a), Bonner (3), etc., to contain, in the chain forming them, some galactose and arabinose molecules and also some acetyl and methoxyl groups.

The formula by Fellenberg (13a) is in accordance with this theory; to represent the neutral octomethoxy form of pectin he gave:



Bonner (3) states also that the major constituent of pectic acid is galacturonic acid anhydride in different proportion (65 to 95 per cent) the balance being made of two substances, galactose and arabinose in variable ratios.

According to some other investigators Schneider and Henglein (47a) arabinose, galactose, and sometimes acetic acid could be impurities or decomposition products.

According to Myers and Baker, pectin in the unhydrolysed condition is monoarabino-monogalacto-diacetyl-heptamethoxyl-octagalacturonic acid.

Bonner (3) summarizing the different types of pectic compounds presents the following:

1. Protopectin, (called pectose by some authors), insoluble in water found only in tissues.
2. Pectin soluble in water and obtained from protopectin by hydrolysis (not precipitated by calcium).
3. Pectic acid obtained from protopectin and pectin by alkaline hydrolysis (precipitated by calcium).

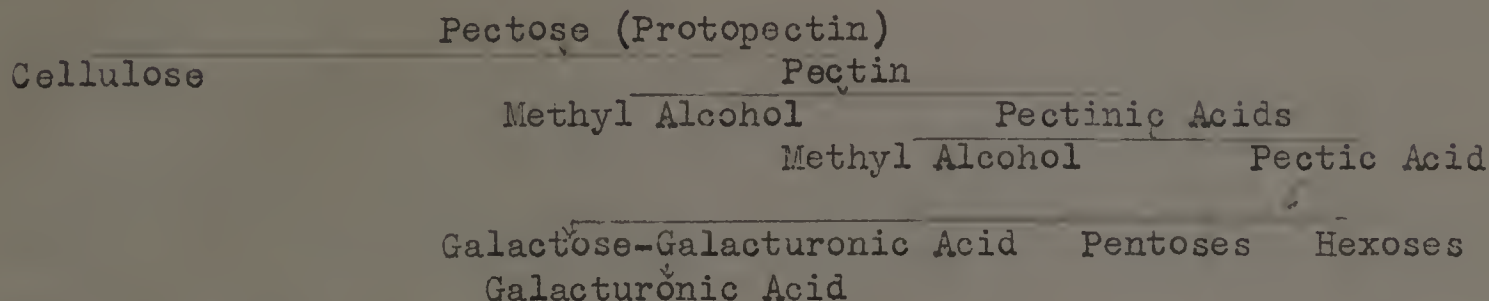
On account of their methyl groups the pectic substances may be classified as:

- pectin completely methylated---no free COOH groups
- pectinic acids---some free COOH groups
- pectic acid---all COOH group free

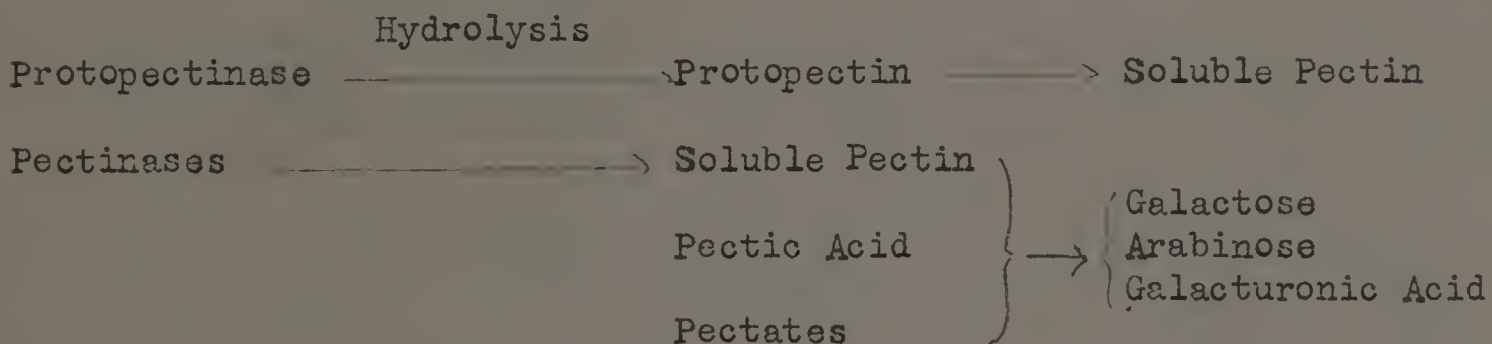
Carre in 1922 (9) and Carre and Horne (10) in 1927 made the

first intensive studies on the pectic changes in apples during storage.

A diagram given by Carre suggests what might happen in the apple to the pectic substances:



These subsequent changes are suggested to be due to enzyme activity. Bonner (3) give the following diagram showing the effect of the enzymes on pectic substances.



Protopectinase was discovered by Bourquelot (4) in 1898 in Gentian and by Carre (9) in 1922 in apple.

Carre (9a) in 1925 made determinations for protopectin, pectin, and total pectic substances. It was found that pectin increases in storage and begins to decrease after about 6 months. As for the protopectin and the total pectic substances, they change little early in the storage period but decrease markedly near the end.

PRESENTATION OF DATA AND DISCUSSION

The apples used for this study were grown in the University orchard (Wethersfield loam, brown phase. Scored 89 in "Rating of Massachusetts soil for orchards purpose"). Upon arrival at the cold storage

they were immediately placed in the 32° F. (0° C.) room.

In order to have samples as comparable as possible the fruit was graded according to U.S. Apple Grades. Apples from 2 to 2-1/2 inches in diameter, the U.S. No. 1, were retained for the experiment.

The grading process was done mechanically with the grading machine at an average room temperature of 60° F. (15.6° C.) to 64° F. (17.8° C.) and a relative humidity of 70-73 per cent. After grading the apples were divided in four lots which were placed in the following storage rooms:

North Cellar - 32° F. (0° C.)

Middle Cellar - 40° F. (4.4° C.) relative humidity 80-95 per cent

Room 2 - 40° F. (4.4° C.) relative humidity 70-85 per cent

Controlled Atmosphere Storage - 40° F. (4.4° C.) with about 3 per cent oxygen and 6 per cent carbon dioxide.

On October 16 a lot of each variety kept till then at 32° F. (0° C.) was placed in the controlled atmosphere storage.

Samples were taken monthly from each room beginning October 15, 1946 and ending on March 11, 1947.

SAMPLING

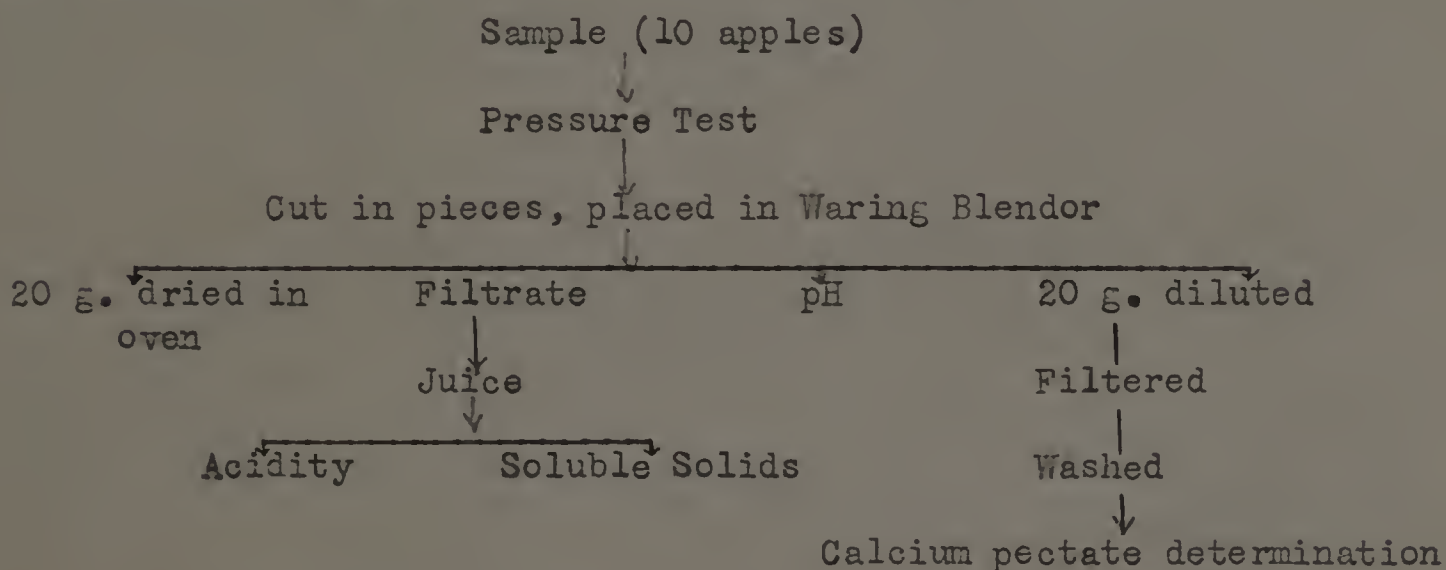
For most of the tests 10 representative apples were chosen. This size of sample has been taken by various workers as Plagge and Gerhardt (41). However, Gourley and Hopkins (19) took 15, and Haynes and Archbold (21) pointed out that a twenty or thirty apple sample should be preferred. The crushing of apples is an extremely slow operation and that is the major factor which limits the size of sample.

After a pressure test, the apples were sliced into thin pieces

and placed in a Waring Blendor, where they were crushed. An easier method was found later in which the apples were first run through a food chopper then through the Waring Blendor.

From the orchard storage, each month, samples were brought to the Food Technology Laboratory, where they were analysed. In the Food Technology Laboratory, the apples were kept during the time of testing in the cold room at an average temperature of 36° to 38° F.

The following diagram will illustrate how samples were handled for the different tests.



PRESSURE TEST

The Magness pressure tester was used to determine the resistance of the flesh to puncture. Before this test was made the skin was removed at the point where the resistance was to be measured. Samples of 10 apples from each experimental lot were taken and tests were made at three different points on each individual apple. The mean of the readings for each sample with the standard error, the coefficient of variation and its standard error, and the standard deviation and its standard error are given in Table I. The difference between two readings is in general greater

Table I.

Pressure test in pounds obtained on pared fruits kept under different storage conditions.

Variety tested	Date of test	Storage Condition	Average pressure in pounds	Standard deviation	Coefficient of variation %
Wealthy	10/15/46	After grading	10.54 ± .11	.53 ± .07	5.01 ± .72
	11/7/46	32°F.	9.43 ± .12	.66 ± .08	7.05 ± .91
		40°F. (B)*	9.28 ± .08	.42 ± .05	4.61 ± .59
	11/11/46	40°F. (A)*	8.33 ± .09	.48 ± .06	5.73 ± .74
	11/28/46	C.A.*	10.38 ± .10	.57 ± .07	5.46 ± .70
	12/11/46	32°F	10.23 ± .08	.45 ± .06	4.38 ± .56
		40°F. (A)*	9.96 ± .07	.37 ± .05	3.69 ± .48
		40°F. (B)*	9.71 ± .08	.43 ± .05	4.40 ± .57
	1/11/47	32°F.	9.83 ± .07	.38 ± .05	3.84 ± .50
		40°F. (A)*	9.70 ± .07	.38 ± .05	3.95 ± .51
		40°F. (B)*	9.40 ± .07	.40 ± .05	4.27 ± .55
		C.A.*	9.15 ± .08	.44 ± .06	4.78 ± .61
	2/14/47	32°F.	9.31 ± .10	.58 ± .07	6.20 ± .80
		40°F. (A)*	9.33 ± .11	.62 ± .08	6.65 ± .86
		40°F. (B)*	9.28 ± .11	.58 ± .07	6.27 ± .81
		C.A.*	9.08 ± .08	.42 ± .05	4.58 ± .60
	3/15/47	32°F.	9.18 ± .07	.38 ± .05	4.16 ± .54
		40°F. (A)*	9.26 ± .12	.68 ± .08	7.32 ± .95
		40°F. (B)*	9.21 ± .06	.31 ± .04	3.39 ± .44
		C.A.*	8.76 ± .10	.54 ± .07	6.11 ± .79

* 40°F. (A) = 40°F. with 80-95% relative humidity.

40°F. (B) = 40°F. with 70-85% " "

C.A. = controlled atmosphere storage - 3% oxygen, 6% CO₂.

Table I (Continued)

Variety tested	Date of test	Storage condition	Average pressure in pounds	Standard deviation	Coefficient of variation %
McIntosh	10/15/46	After grading	11.24 ± .09	.51 ± .07	4.56 ± .58
	11/7/46	32°F.	10.70 ± .10	.55 ± .07	5.14 ± .66
		40°F.(B)*	10.13 ± .09	.47 ± .06	4.65 ± .60
	11/11/46	40°F.(A)*	10.36 ± .09	.47 ± .06	4.55 ± .60
	11/28/46	C.A.*	10.65 ± .09	.48 ± .06	4.46 ± .58
	12/11/46	32°F.	10.41 ± .08	.47 ± .06	4.38 ± .57
		40°F.(A)*	9.96 ± .09	.51 ± .07	5.09 ± .66
		40°F.(B)*	9.56 ± .09	.52 ± .07	5.43 ± .70
	1/11/47	32°F.	11.21 ± .07	.36 ± .05	3.23 ± .42
		40°F.(A)*	9.30 ± .07	.38 ± .05	4.12 ± .53
		40°F.(B)*	9.18 ± .06	.31 ± .04	3.33 ± .43
		C.A.*	10.50 ± .07	.39 ± .05	3.74 ± .48
	2/14/47	32°F.	9.41 ± .09	.47 ± .06	5.03 ± .6
		40°F.(A)*	8.91 ± .09	.50 ± .06	5.51 ± .7
		40°F.(B)*	8.75 ± .09	.52 ± .06	5.94 ± .8
		C.A.	9.93 ± .06	.34 ± .04	3.42 ± .4
	3/15/47	32°F.	9.20 ± .08	.44 ± .05	4.75 ± .59
		40°F.(A)*	8.63 ± .10	.55 ± .07	6.31 ± .82
		40°F.(B)*	8.56 ± .09	.49 ± .06	5.66 ± .73
		C.A.*	9.26 ± .08	.45 ± .06	4.84 ± .63

* 40°F.(A) = 40°F. with 80-95% relative humidity.

40°F.(B) = 40°F. with 70-85% " "

C.A. = controlled atmosphere storage - 3% oxygen, 6% CO₂

Table I (Continued)

Variety tested	Date of test	Storage condition	Average pressure in pounds	Standard deviation	Coefficient of variation %
Cortland	10/15/46	After grading	11.71 ± .07	.41 ± .05	3.47 ± .45
	11/7/46	32°F.	10.66 ± .09	.48 ± .06	4.48 ± .58
		40°F.(B)*	9.25 ± .08	.45 ± .06	4.85 ± .63
	11/11/46	40°F.(A)*	9.18 ± .09	.50 ± .06	5.43 ± .70
	11/28/46	C.A.*	10.58 ± .10	.56 ± .07	5.26 ± .68
	12/11/46	32°F.	10.51 ± .08	.46 ± .06	4.40 ± .57
		40°F.(A)*	9.38 ± .07	.41 ± .05	4.33 ± .56
		40°F.(B)*	9.28 ± .08	.43 ± .06	4.61 ± .60
	1/11/47	32°F.	10.65 ± .08	.44 ± .06	4.11 ± .53
		40°F.(A)*	9.43 ± .08	.43 ± .06	4.55 ± .59
		40°F.(B)*	9.33 ± .07	.39 ± .05	4.15 ± .51
		C.A.*	11.05 ± .08	.46 ± .06	4.17 ± .54
	2/14/47	32°F.	9.55 ± .09	.48 ± .06	5.01 ± .65
		40°F.(A)*	9.11 ± .08	.43 ± .05	4.70 ± .61
		40°F.(B)*	8.40 ± .09	.48 ± .06	5.71 ± .74
		C.A.*	10.13 ± .07	.40 ± .05	3.86 ± .50
	3/15/47	32°F.	9.13 ± .08	.43 ± .06	4.73 ± .61
		40°F.(A)*	8.93 ± .09	.50 ± .06	5.63 ± .73
		40°F.(B)*	8.05 ± .09	.51 ± .07	6.37 ± .82
		C.A.*	10.13 ± .08	.42 ± .05	4.17 ± .54

* 40°F.(A) = 40°F. with 80-95% relative humidity.

40°F.(B) = 40°F. with 70-85% " "

C.A. = controlled atmosphere storage - 3% oxygen, 6% CO₂.

12 lbs.

11 lbs.

10

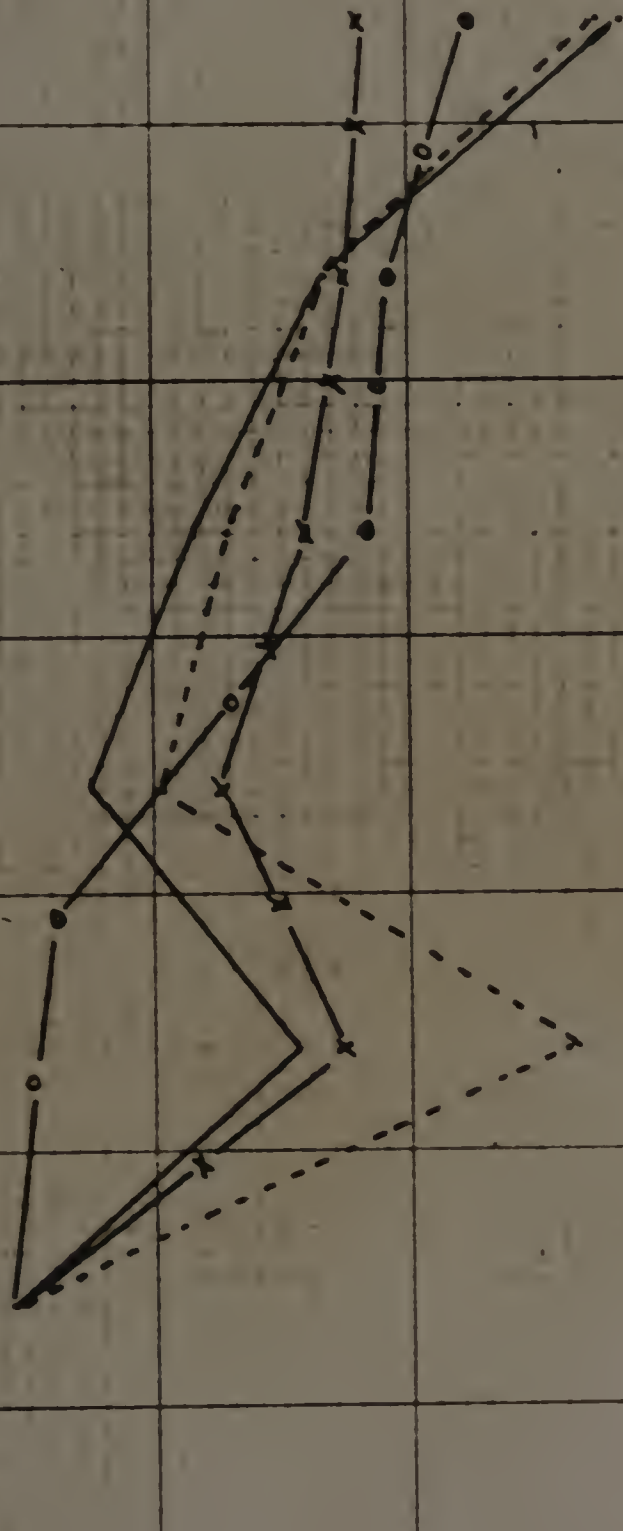
9

8

7

6

Figure 1. Pressure test in pounds obtained on pared Wealthy variety of apple kept under different conditions of storage.



—●— 32° F.
 - - - - 40° F. (80-95% rel. hum.)
 —x— 40° F. (70-85% " ")
 —●— Controlled Atmosphere
 40° F. 5% O₂, 6% CO₂

October

November

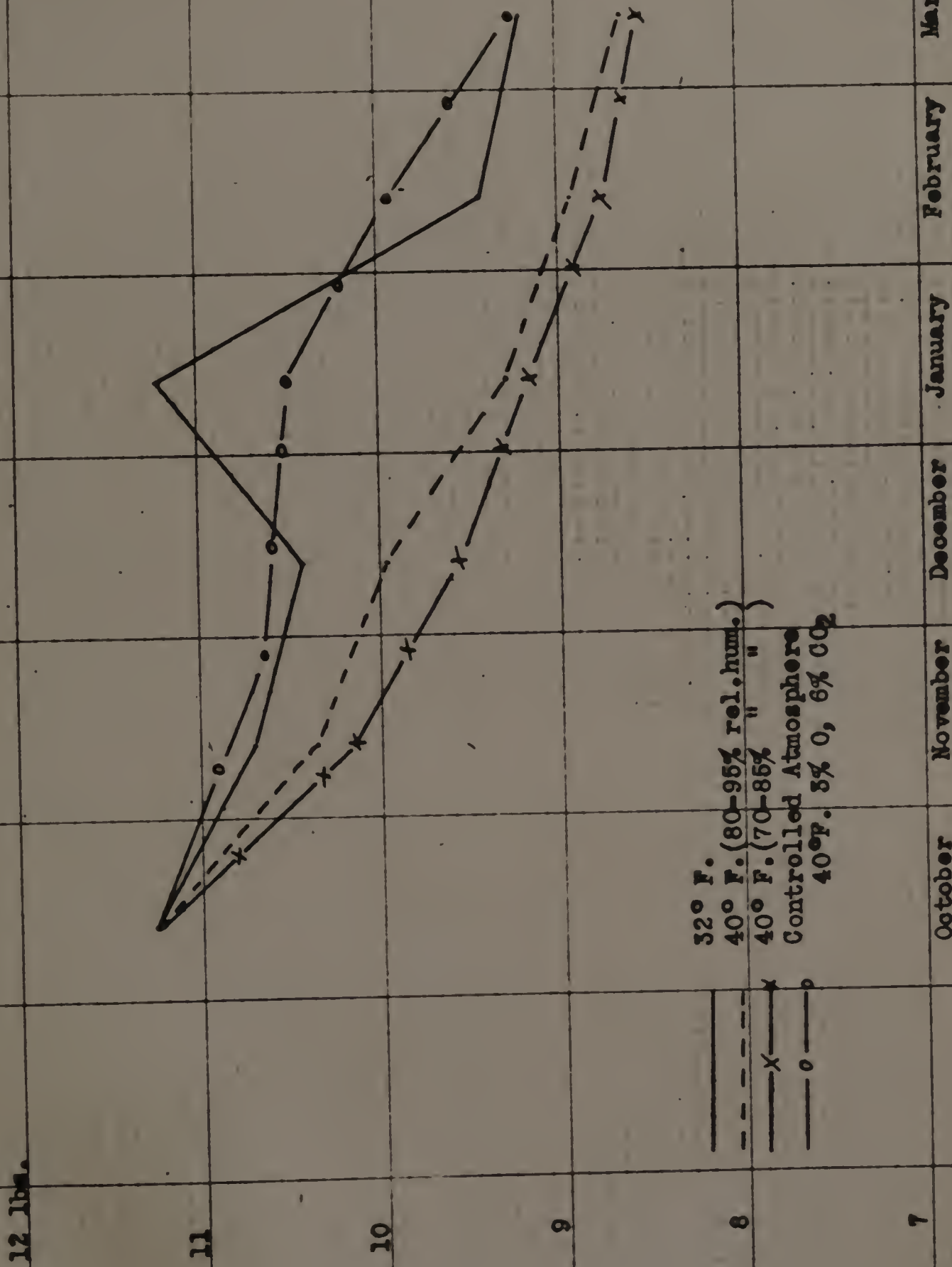
December

January

February

March

Figure 2. Pressure test in pounds obtained on pared McIntosh variety of apple kept under different conditions of storage.



32° F.
 40° F. (80-95% rel. hum.)
 40° F. (70-85% " "
 Controlled Atmosphere
 40° F., 5% O₂, 6% CO₂

12 lbs.

11

10

9

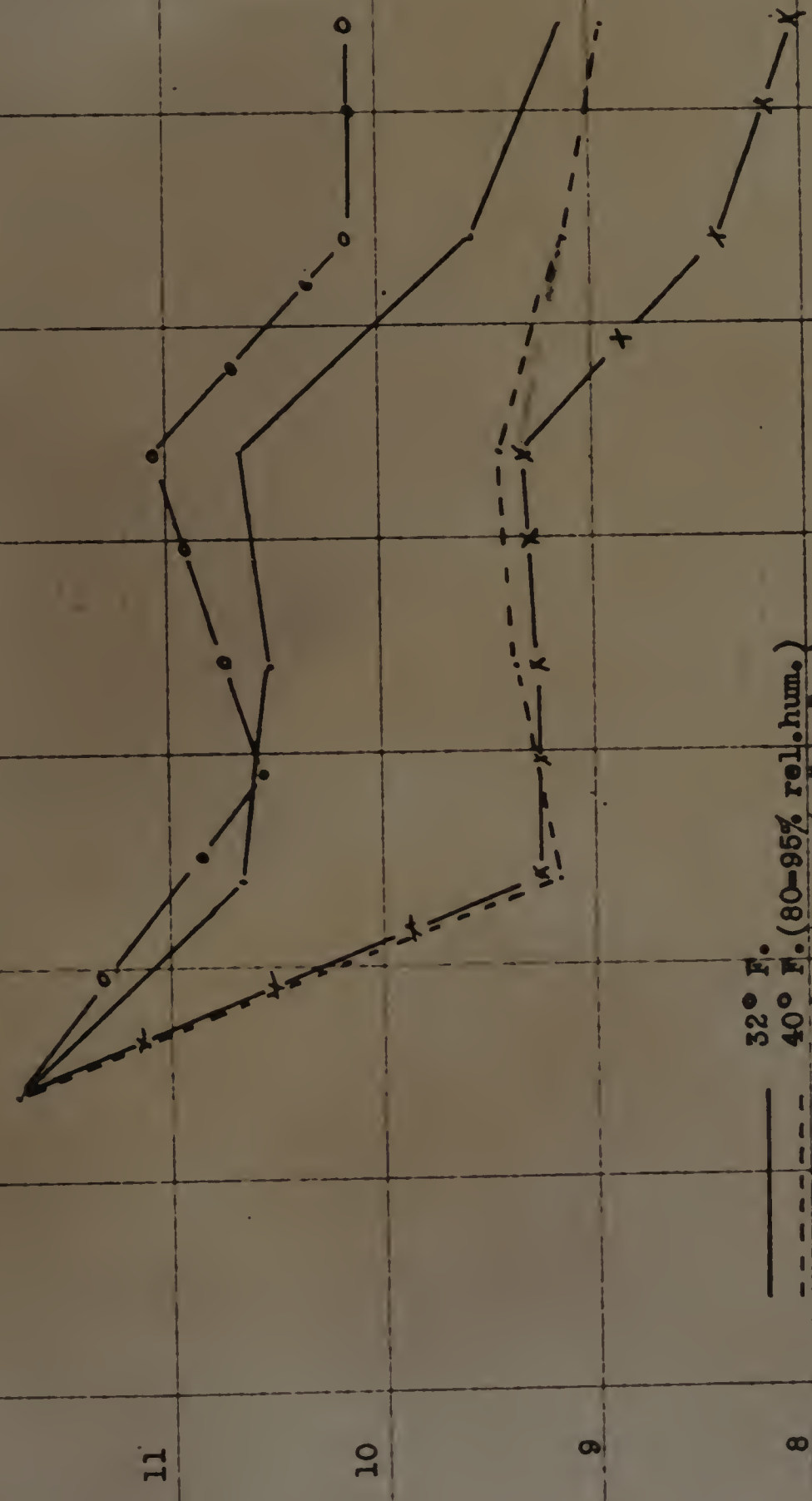
8

7

October November December January February March

Figure 3. Pressure test in pounds obtained on pared Cortland variety of apple kept under different conditions of storage.

12 lbs.



11

10

9

8

7

32° F. (80-95% rel. hum.)
 40° F. (70-85%)
 Controlled Atmosphere
 40°F, 3% O, 6% CO2

October November December January February March

than twice the standard error which in statistical work is considered as being significant.

We should expect a smooth curve. However, the graph shows in the case of Wealthy a marked decrease in November and in the case of McIntosh a marked increase in January as shown in Figures 1 and 2. The reasons for these deviations might be due to difference in the sample itself or to errors in taking the pressure tests. The tester, however, was checked against a scale before starting the determinations.

These tests indicate a clear influence of temperature on the rate of softening. Samples kept at 40° F. (4.4° C.) under different conditions of relative humidity gave about the same result, but the sample coming from the higher relative humidity (80 - 95 per cent) was slightly harder. This is illustrated in Figures 1, 2, and 3.

For Cortland, controlled atmosphere storage gave the best results in maintaining firmness. Storage at 32° F. (0° C.) and at 40° F. (4.4° C.) with the higher relative humidity (80 - 95 percent) seemed to accelerate the rate of softening over the rate in controlled atmosphere storage. Those stored at 40° F. (4.4° C.) in air with the lower relative humidity (70 - 85 per cent) showed a much faster rate of softening especially in the first month and also during the fifth and sixth month of storage.

For Wealthy, the results are not so clear. It seems that controlled atmosphere was not satisfactory in maintaining firmness, but in all cases the differences were small. Storage at 32° F. (0° C.) and 40° F. (4.4° C.) made little difference in the firmness of Wealthy. These results are shown in Table I and represented graphically in Figures 1, 2, and 3.

MOISTURE CONTENT

The water content was at first determined by the Bidwell-Sterling Method (Winton "Analysis of Food") but did not prove to be very reliable. For the following tests a modification of the official method for total solids in fruits given by the "Association of Official Agricultural Chemists" was used. Since it was not possible to dry in vacuo or under low pressure, a higher temperature was used; 90° C. for 12 hours. 100° C. was not used to avoid an undesirable amount of caramelization. Samples of 20 g. of the finely crushed apple were used. According to Archbold (1) losses due to oxidation or caramelization even at 100° C. are not important for the first 36 hours.

An arbitrary time of drying was used because Archbold (1) found that samples drying in an oven at 100° C. do not reach a constant weight even after 65 days.

The results are given in per cent of moisture as shown in Table II and presented graphically in Figures 4, 5, and 6. From the results obtained only small differences in moisture content were found between the beginning and the end of the storage period. Archbold found that "the existence of some substances gradually decrease in amount during storage life" and from his results the dry weight slightly decreased from October to April. At the same time the apples are losing water by evaporation, the carbohydrates are oxidizing and decreasing in weight. If the loss of water is greater than the loss in dry weight the loss of moisture is greater than it appears to be. It is reasonable to assume that when the same weight of sample (20 g.) was used for each experiment the volume of the sample changed from month to month.

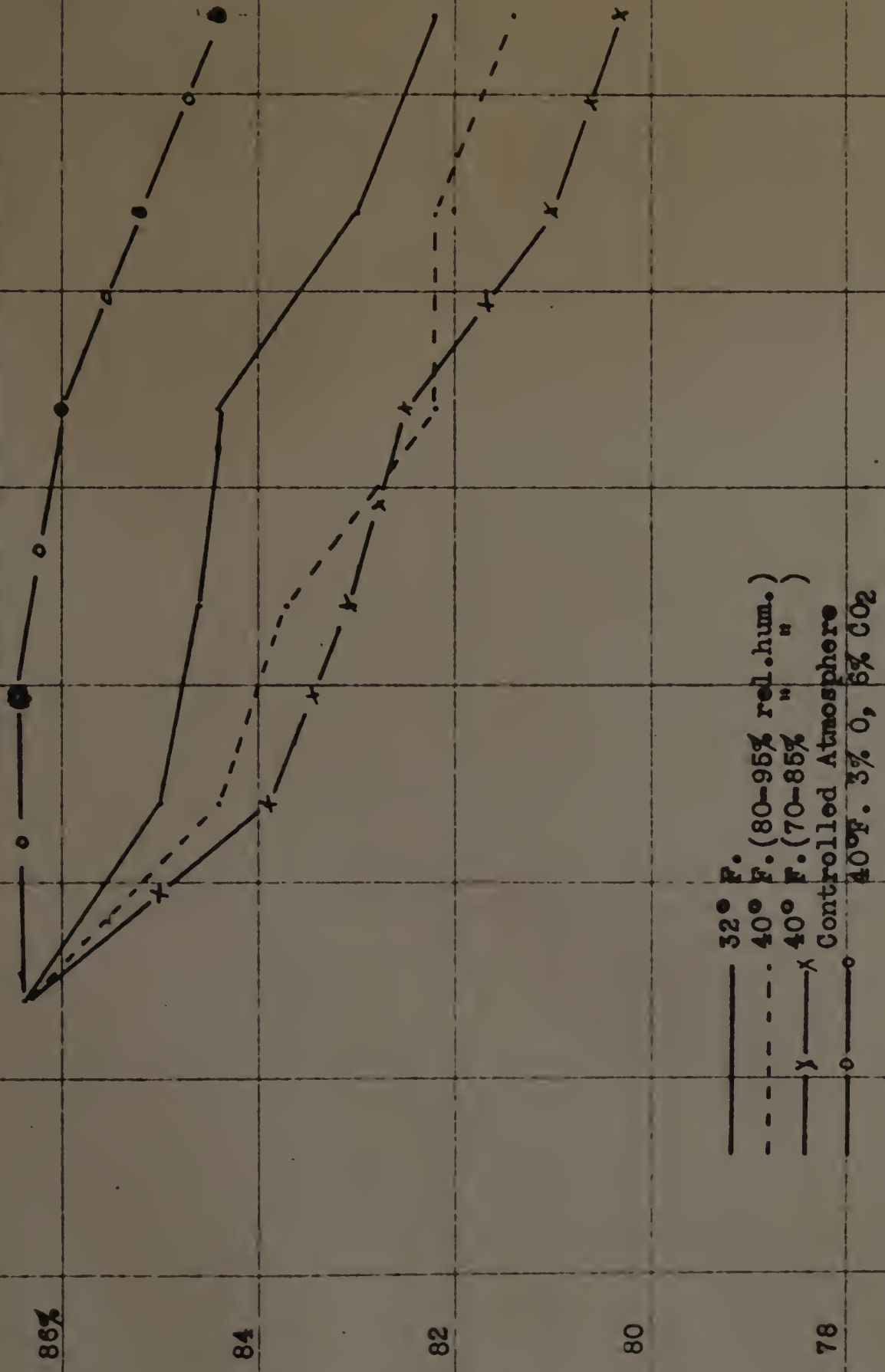
The loss of moisture is related to the relative humidity of

Table II.

Moisture content in per cent of fresh weight.

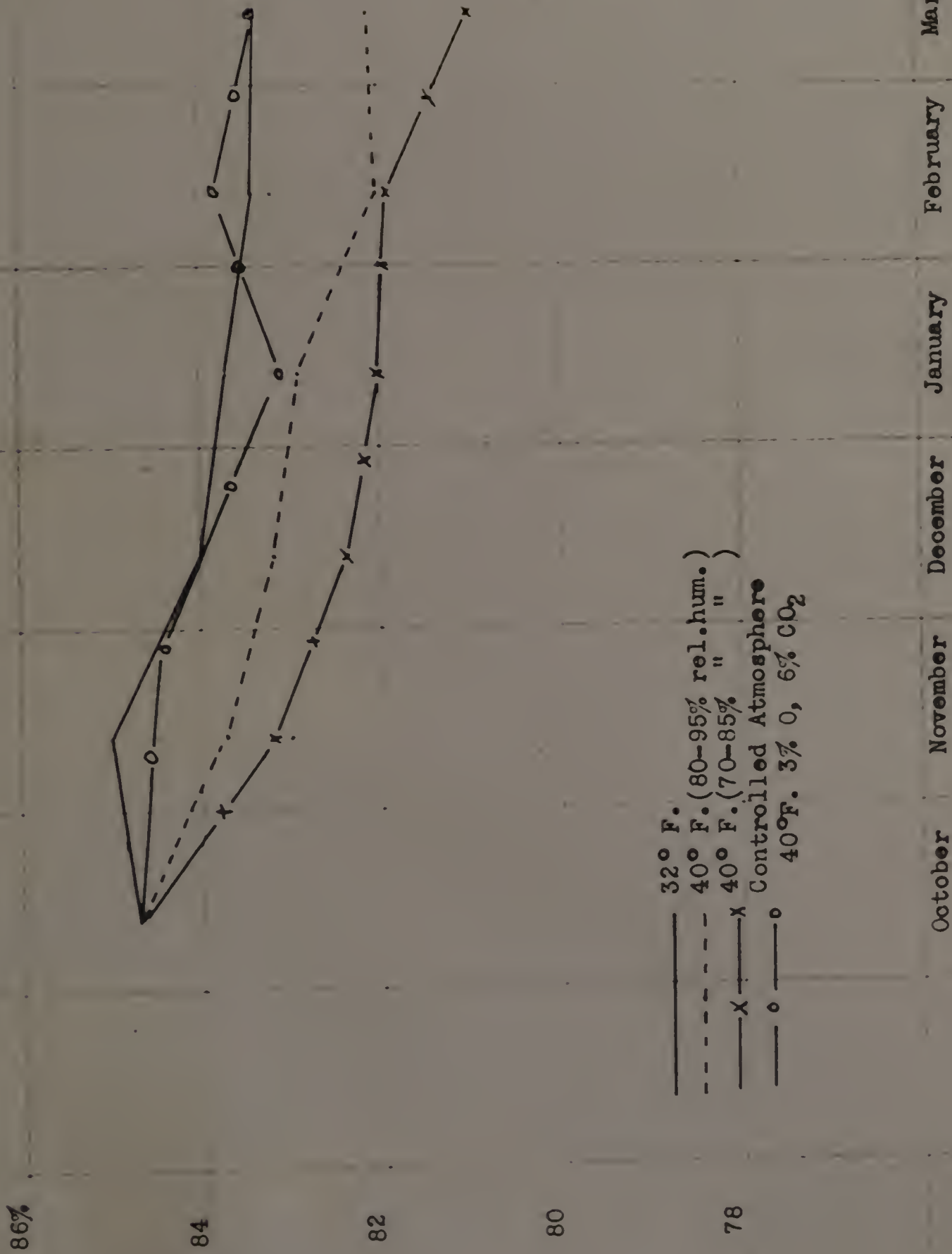
Variety tested	Storage condition	10/15/46	11/7/46	12/11/46	1/11/47	2/11/47	3/11/47
Wealthy	32°F.	86.2	85.0	84.6	84.4	83.0	82.2
	40°F. (80-95% rel.hum.)	86.2	84.4	83.7	82.2	82.2	81.4
	40°F. (70-85% rel.hum.)	86.2	83.9	83.1	82.5	81.0	80.3
	Controlled atmosphere	86.2	86.4		86.0	85.2	84.4
McIntosh	32°F.	84.7	85.0	84.0	83.7	83.2	83.4
	40°F. (80-95% rel.hum.)	84.7	83.7	83.2	82.9	82.0	82.1
	40°F. (70-85% rel.hum.)	84.7	83.2	82.4	82.0	81.9	80.5
	Controlled atmosphere	84.7	84.4		83.1	83.8	83.4
Cortland	32°F.	83.0	82.4	83.1	82.6	81.9	80.2
	40°F. (80-95% rel.hum.)	83.0	82.8	82.5	81.4	81.0	81.4
	40°F. (70-85% rel.hum.)	83.0	83.2	81.3	80.2	79.6	79.3
	Controlled atmosphere	83.0	83.0		82.7	82.3	81.4

Figure 4. Moisture content in per cent of fresh weight of Wealthy variety of apple kept under different conditions of storage.



October November December January February March

Figure 5. Moisture content in per cent of fresh weight of McIntosh variety of apple kept under different conditions of storage.



86%

84

82

80

78

——— 32° F.
 - - - - 40° F. (80-95% rel. hum.)
 —x— 40° F. (70-85% rel. hum.)
 —o— Controlled Atmosphere
 40° F., 3% O₂, 6% CO₂

October

November

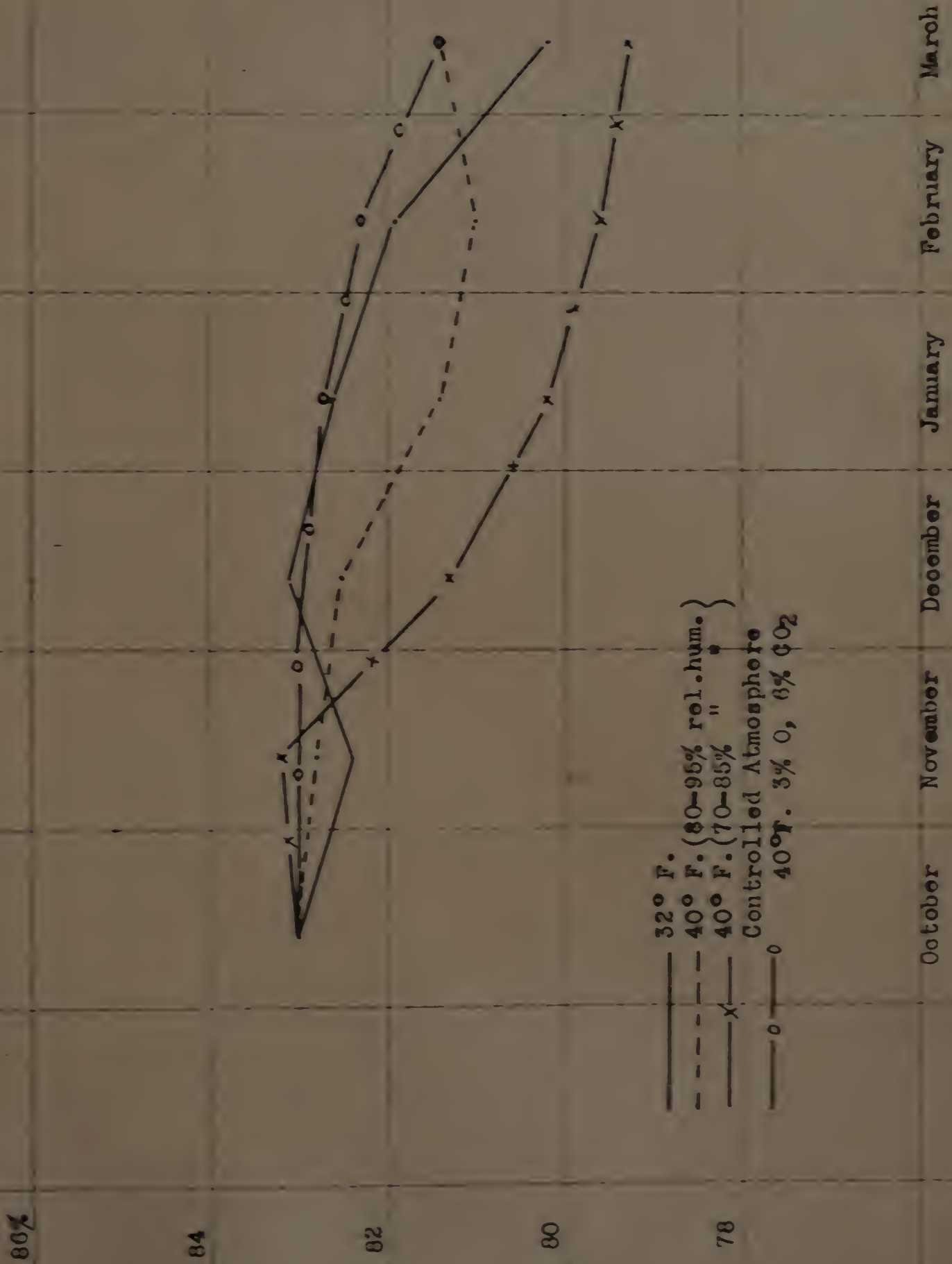
December

January

February

March

Figure 6. Moisture content in per cent of fresh weight of Cortland variety of apple kept under different conditions of storage.



the storage room. For the three varieties the room with the lowest humidity resulted in the lowest moisture content in apples at the end of the six months' storage period. Apples kept in the controlled atmosphere room, especially Wealthy, lost less than in the other rooms, where the relative humidity was high.

The greatest moisture loss was 5 per cent in six months by Wealthy at 40° F. (4.4° C.) with the lower relative humidity; the least was by McIntosh at 32° F. (0° C.) and in controlled atmosphere storage it lost there only 1.3 per cent.

The differences, however, are not very significant because they are too small. Differences in the samples themselves may have been the cause of the variations.

SOLUBLE SOLIDS

In February and March a Zeiss refractometer was used to determine the soluble solids in the juice extracted from the pulped flesh. The results of these tests are given in Table III. below:

Table III.

Variety tested	Date of test	Soluble solids tests in per cent	
		Storage Condition	Soluble solids of the juice in per cent
Wealthy	2/14/47	32° F.	10.0
		40° F. (80-90% relative humidity)	9.7
		40° F. (70-85% - -)	9.4
		controlled atmosphere	9.4
	3/11/47	32° F.	9.4
		40° F. (80-95% relative humidity)	9.0
		40° F. (70-85% - -)	8.2
		controlled atmosphere	8.0
McIntosh	2/14/47	32° F.	11.0
		40° F. ((0-95% relative humidity)	11.6
		40° F. (70-85% - -)	10.8
		controlled atmosphere	11.4

Table III. (Continuation)

Variety tested	Date of test	Storage condition	Soluble solids of the juice in per cent
McIntosh	3/11/47	32° F.	9.8
		40° F. (80-95% relative humidity)	10.0
		40° F. (70-85% - -)	11.2
		controlled atmosphere	10.2
Cortland	2/14/47	32° F.	11.0
		40° F. (80-95% relative humidity)	10.8
		40° F. (70-85% - -)	10.4
		controlled atmosphere	12.1
	3/11/47	32° F.	10.7
		40° F. (80-95% relative humidity)	10.7
		40° F. (70-85% - -)	10.4
		controlled atmosphere	10.8

The amounts of soluble solids given in Table III were obtained from apples which had been kept 4 and 5 months in different storage conditions. The percentage of soluble solids in the juice of the apples differed according to the varieties; Cortland showed the greatest percentage and Wealthy the least. The three varieties have responded quite differently to different conditions of storage, Cortland, for example, after 5 months' storage showed more soluble solids when kept in controlled atmosphere and for Wealthy the opposite is true. For McIntosh the difference in soluble solids under different storage condition was slight.

TOTAL ACIDITY

The official method of the Association of Official Agricultural Chemists was followed for the determination of total acidity. Ten gram samples of juice were used, obtained by filtering the ground flesh. The juice obtained in this way was rather colored and to have a more accurate

Table IV

Total Acidity as per cent Malic Acid of the juice obtained from apples kept under different conditions of storage.

Variety tested	Storage condition	Total acidity obtained after the following months of storage:					
		0	1	2	3	4	5
Wealthy	32°F.	1.38	1.31	1.30	.93	.89	.80
	40°F. (80-95% rel. hum.)	1.38	.83	.84	.82	.67	.63
	40°F. (70-85% rel. hum.)	1.38	1.15	.93	.81	.63	.60
	Controlled atmosphere	1.38	1.12		.92	.85	.82
McIntosh	32°F.	1.30	1.11	.79	.61	.56	.52
	40°F. (80-95% rel. hum.)	1.30	1.23	1.11	.64	.53	.47
	40°F. (70-85% rel. hum.)	1.30	1.11	.94	.74	.51	.51
	Controlled atmosphere	1.30	.77		.81	.59	.56
Cortland	32°F.	1.14	.91	.76	.52	.50	.49
	40°F. (80-95% rel. hum.)	1.14	.76	.66	.58	.47	.40
	40°F. (70-85% rel. hum.)	1.14	.82	.71	.62	.56	.43
	Controlled atmosphere	1.14	.81		.74	.59	.51

Figure 7. Total acidity as per cent Malic Acid of the juice obtained from Wealthy variety of apples kept under different conditions of storage.

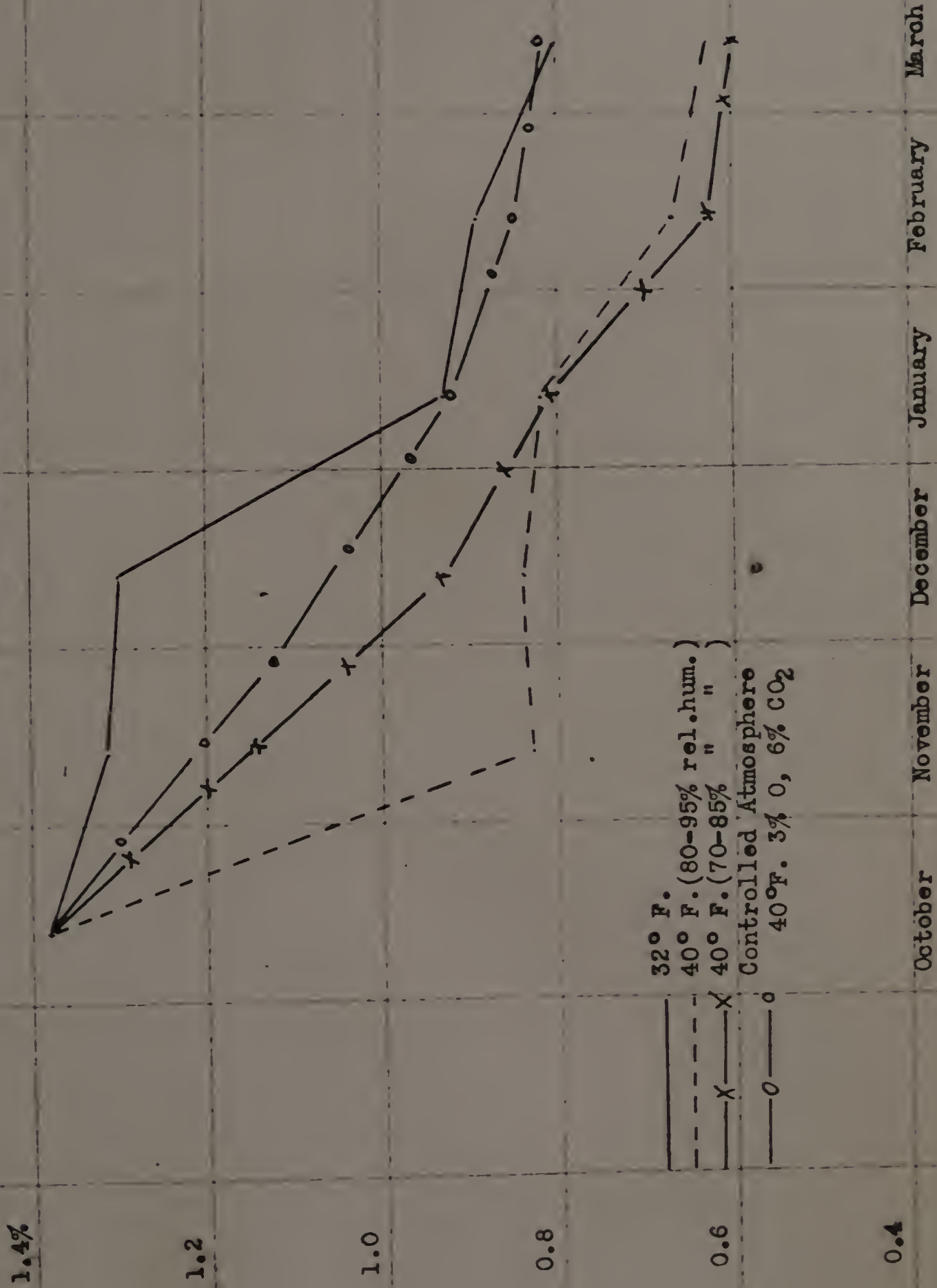


Figure 8. Total acidity as per cent Malic Acid of the juice obtained from McIntosh variety of apples kept under different conditions of storage.

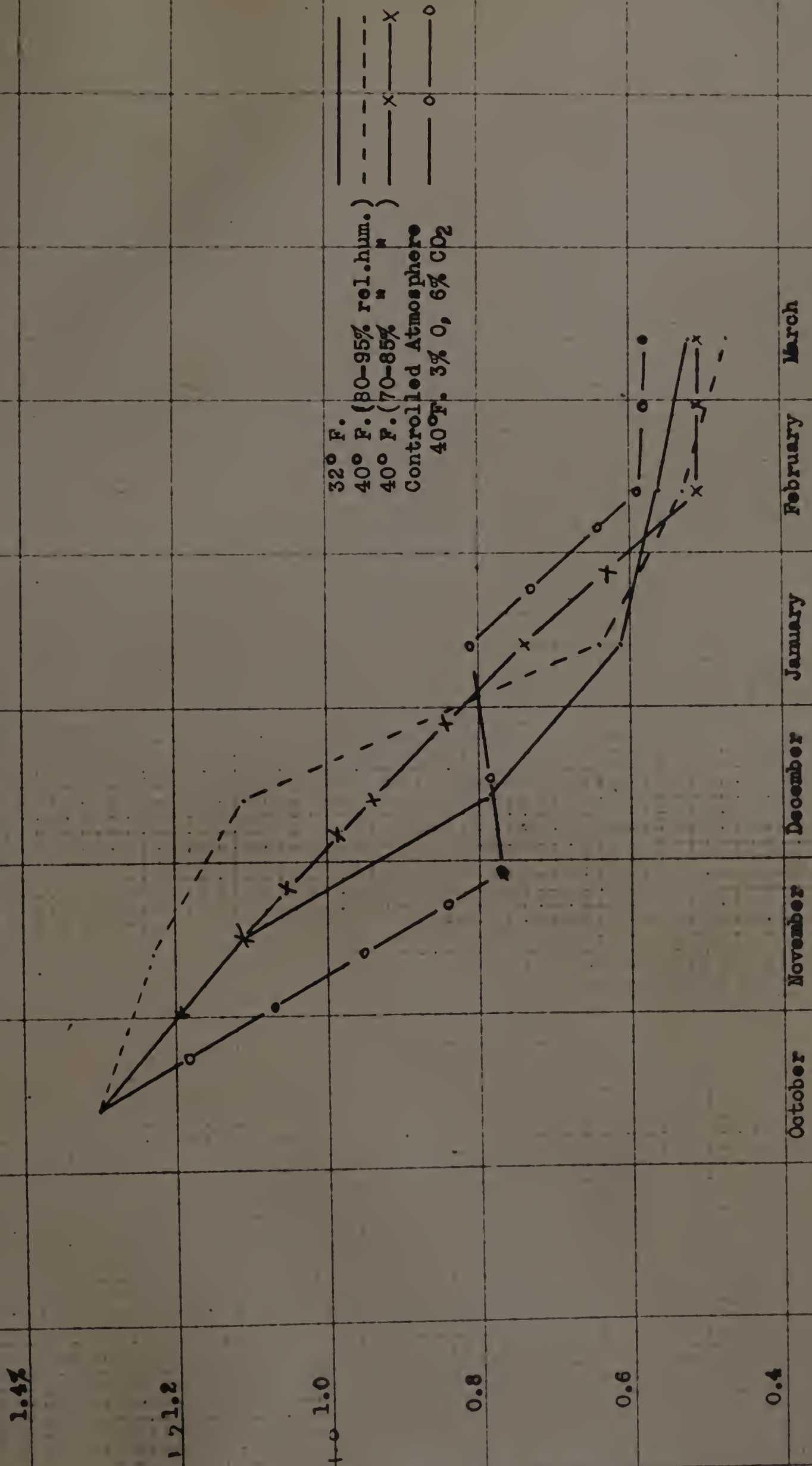
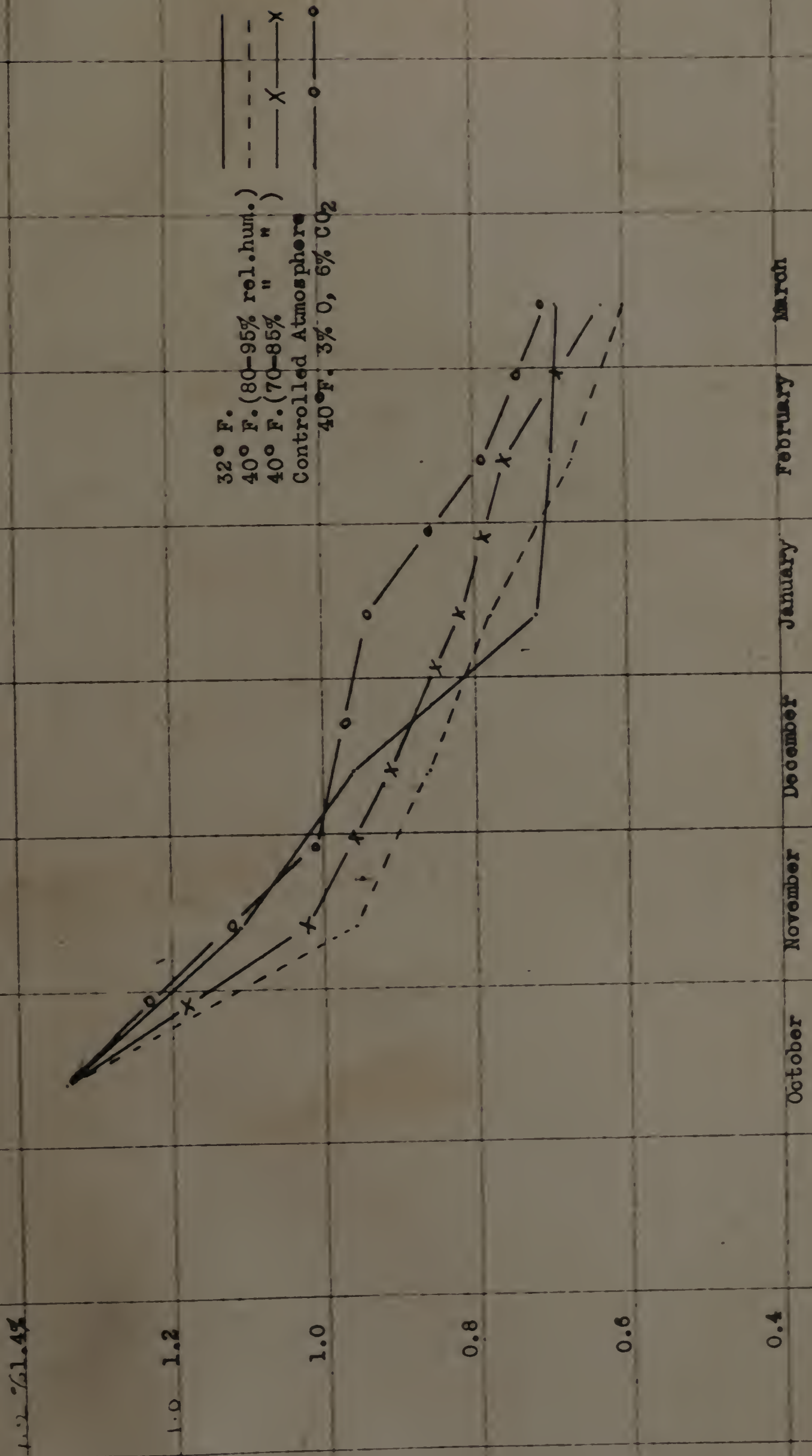


Figure 9. Total acidity as per cent Malic Acid of the juice obtained from Cortland variety of apples kept under different conditions of storage.



end point with phenolphthalein indicator, it was diluted to about 250 ml. with distilled water. Four titrations were made for each determination and the end point was determined by comparison with the other solutions. The results are given in Table IV. and represented graphically in Figures 7, 8 and 9.

The general tendency of all the curves was to decrease rather rapidly for the two first months of storage under all conditions of the experiment, and to become more flat at the end of the storage period.

Cortland shows a gradual decrease in all cases and the behavior of its total acidity is not much influenced by differences in storage conditions.

McIntosh lost acidity faster during the first 3 months, losing more acid when stored at 32° F. (0° C.) than at 40° F. (4.4° C.). In the following months the situation is reversed and the amount of acid has a tendency to be the same at the different conditions of storage.

Wealthy is the variety which shows most striking differences. It is interesting to note that for this variety the apples kept at 32° F. (0° C.) and in controlled atmosphere storage maintained a higher level of total acidity.

Haynes and Archbold (21) have suggested that apples which keep well in storage are those which lose acid slowly. However, Wealthy, the variety which kept a high total acidity level in modified atmosphere, was affected while in storage by some physiological disorder which reduced its keeping quality.

From these data it is clear that loss of acidity characterizes the metabolism of apples and it could be used as an index of storage condition. It has been observed that apples with less acidity taste sweeter.

CHANGE IN pH

The discussion on the behavior of the hydrogen-ion concentration must be related to the one on total acidity, since pH is a measure of the "active acidity". There was a marked loss in total acidity in the flesh of all apples concerned but the variation in hydrogen-ion concentration for all varieties was slight.

Plagge and Gerhardt (41) found that "there is a general tendency for the active acidity to increase in the early part of the cold storage period" of Grimes apples. He suggested that a possible rearrangement of the buffer system within the apple might occur, being produced by a temporary cutrailment of the respiratory processes at the lower temperature of storage.

The pH of the pulped flesh was obtained with a Beckman pH meter.

It is interesting to note here that Gourley and Hopkins (19) found that for Stayman, Wealthy and Jonathan, the hydrogen-ion concentration of the juice did not appear to be affected by applications of nitrate during the growing period in which the true acidity of the apple is being established. The tendency of the pH to change was small for all varieties under all storage conditions as can be seen in Table V. and graphically in Figures 10, 11, and 12.

Table V.

pH of the ground flesh of apples kept under different conditions of storage.

Variety tested	Storage condition	pH reading obtained after the following months of storage:					
		0	1	2	3	4	5
Wealthy	32°F.	3.4	3.4	3.4	3.4	3.4	3.5
	40°F.(80-95% rel.hum.)	3.4	3.4	3.4	3.3	3.6	3.8
	40°F.(70-85% rel.hum.)	3.4	3.4	3.4	3.3	3.6	3.7
	Controlled atmosphere	3.4	3.3		3.3	3.5	3.4
McIntosh	32°F.	3.5	3.5	3.4	3.4	3.7	3.8
	40°F.(80-95% rel.hum.)	3.5	3.6	3.4	3.4	3.7	3.9
	40°F.(70-85% rel.hum.)	3.5	3.6	3.4	3.4	3.7	3.8
	Controlled atmosphere	3.5	3.5		3.3	3.5	3.5
Cortland	32°F.	3.5	3.6	3.4	3.4	3.6	3.7
	40°F.(80-95% rel.hum.)	3.5	3.6	3.6	3.6	3.6	3.7
	40°F.(70-85% rel.hum.)	3.5	3.6	3.6	3.6	3.7	3.8
	Controlled atmosphere	3.5	3.5		3.4	3.5	3.6

Figure 10. pH of the ground flesh of Wealthy variety of apple kept under different conditions of storage.

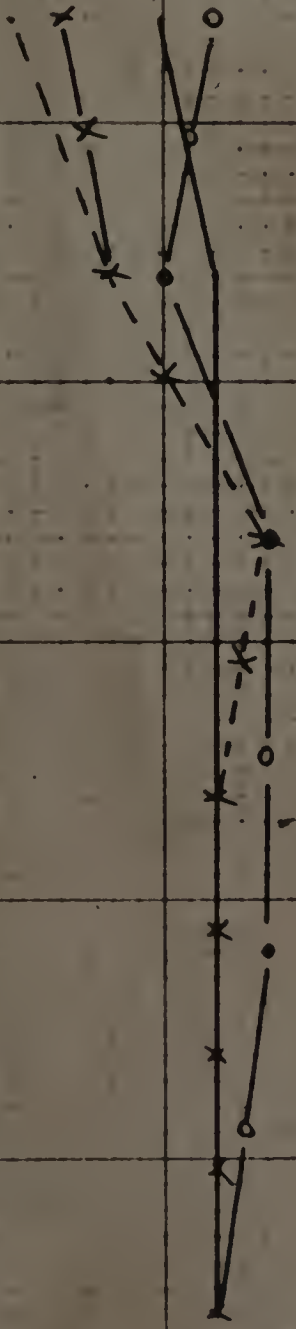
4.5

4.0

3.5

3.0

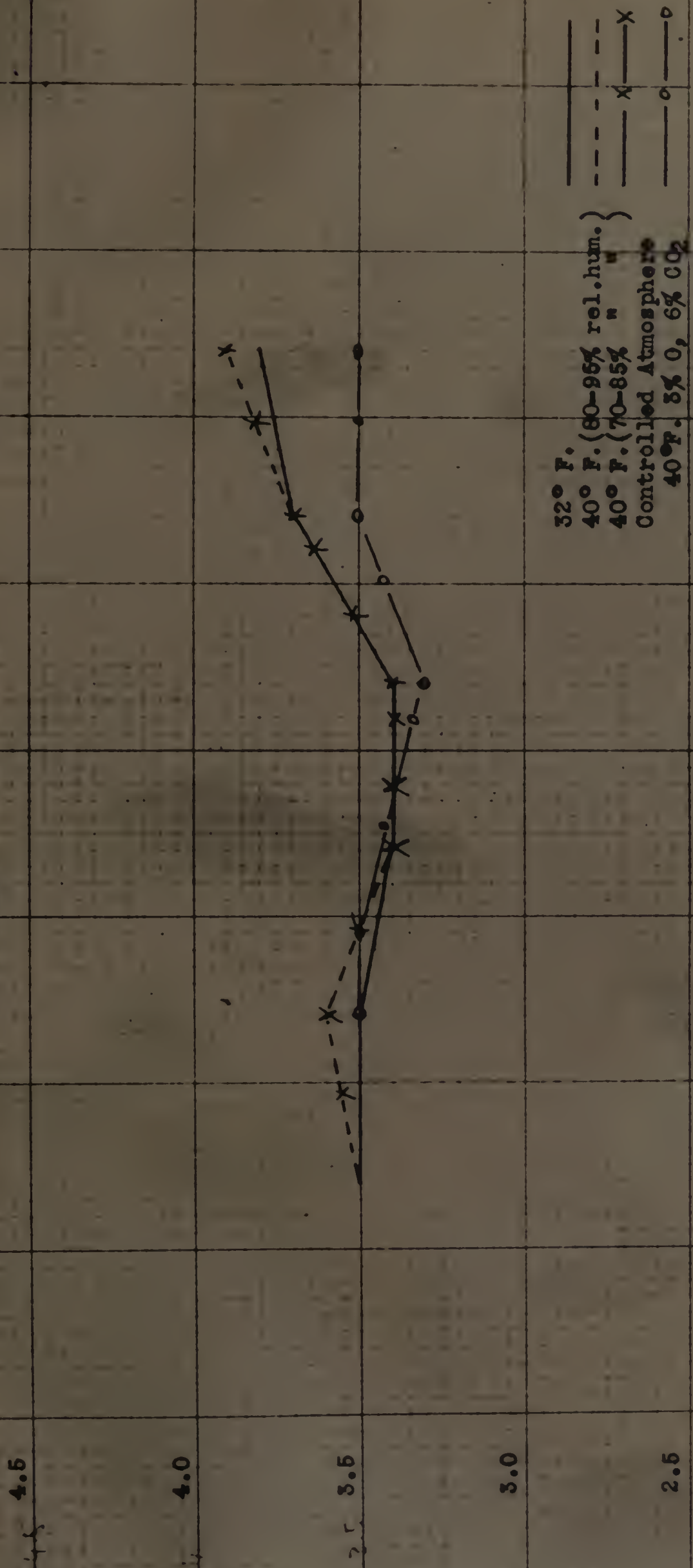
2.5



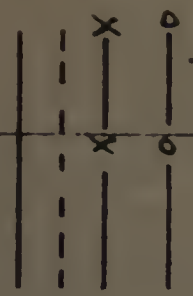
32° F. ———
 40° F. (80-95% rel. hum.) - - -
 40° F. (70-85% rel. hum.) —X—
 Controlled Atmosphere
 40° F., 3% O₂, 6% CO₂ —O—

October November December January February March

Figure 11. pH of the ground flesh of McIntosh variety of apple kept under different conditions of storage.

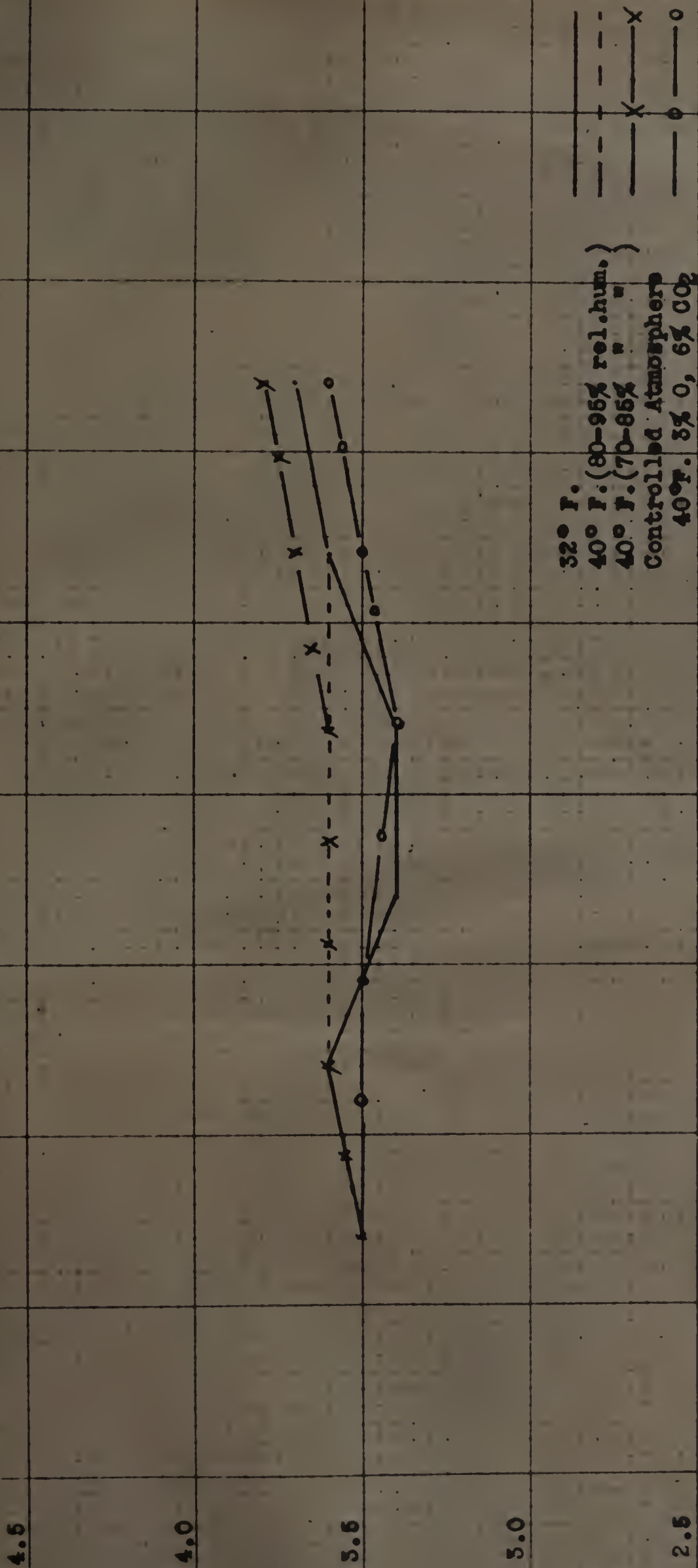


32° F. (80-96% rel. hum.)
 40° F. (70-85% rel. hum.)
 Controlled Atmosphere
 40° F. 3% O₂, 6% CO₂



October November December January February March

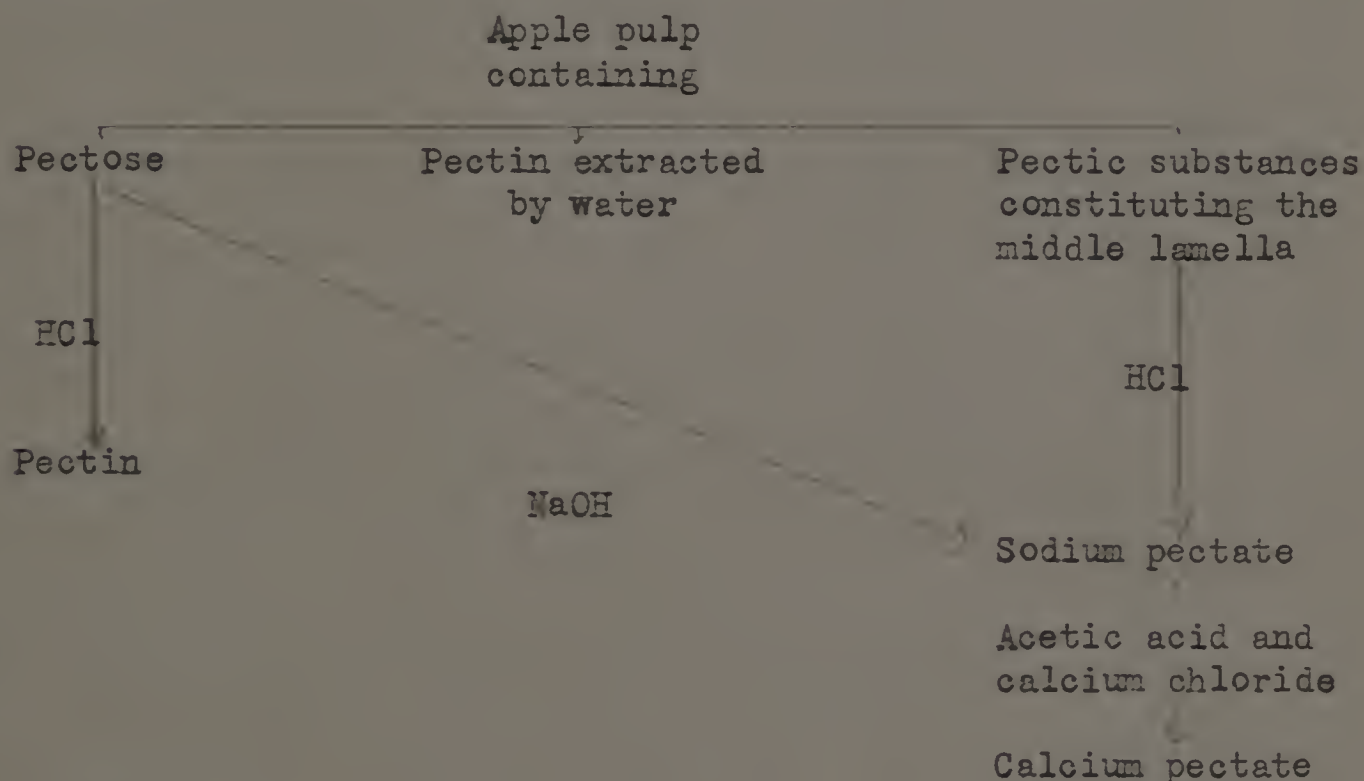
Figure 12. pH of the ground flesh of Cortland variety of apple kept under different conditions of storage.



However, it must not be overlooked that in the first month a general tendency for the pH to decrease occurred while the total acidity was undergoing a definite decline. Near the end of the storage period the increase in pH is more definite and is related to the decrease in total acidity. As a specific response to modified atmosphere storage, the three varieties showed almost no change in pH.

PECTIN

A chart given by Carre and Horne (10) shows the behavior of pectic substances during their extraction:



The tentative method given by the Association of Official Agricultural Chemists for the determination of pectin is based on the precipitation of pectic substances in the presence of ethyl alcohol. This method has been often criticized and it is very hard indeed not to lose any of the gel during the different washings of the precipitate, and to get rid of chloride ion in the alcohol precipitate. If by this method most of the proteins are eliminated, many of the polysacharrides

as starches and gums remain in the precipitate.

For these reasons, a method to determine the calcium salts of the pectic substances present was chosen. This method was devised by Carre and Haynes in 1922: upon saponification by an alkali, pectin gives a salt which is precipitated when calcium chloride is added; this precipitate is dried and weighed as "calcium pectate".

Bonner (3) explained that the precipitation of calcium pectate when a polyvalent cation (say CaCl_2) is added, is due to an increase of "effective electrostatic attraction". The maximal attraction is obtained when there is one calcium ion for every two COOH groups and if pectic acid is precipitated from a solution containing an excess of calcium, such a "calcium pectate" is formed. Pectic substances are very strongly negative colloids, which explains their precipitation by electrolytes.

It is interesting to note that pectic substances are easily stained by ruthenium red and can be detected and microscopically located in the flesh of the apple.

To determine pectin, a 20 g. sample of pulped flesh with ca 250 ml of cold distilled water was mixed for 2 minutes in a Waring blender. This mixture was then filtered and the filtrate washed with cold water. The solution obtained was then treated according to the method for determination of calcium pectate.

In all cases the total amount of soluble pectin increased in the first 5 months of storage. The decrease near the end of storage noted by Carre and Horne (10) was not observed in any case; this decrease was indeed observed by Carre only after the sixth month.

Table VI.

Pectin as per cent Calcium Pectate of the ground flesh of apples kept under different conditions of storage.

Variety tested	Storage condition	% calcium pectate obtained after the following months of storage:					
		0	1	2	3	4	5
Wealthy	32°F.	.10	.13	.18	.18	.24	.26
	40°F.(80-95% rel.hum.)	.10	.15	.17	.21	.32	.34
	40°F.(70-85% rel.hum.)	.10	.14	.19	.20	.29	.40
	Controlled atmosphere	.10	.16		.21	.24	.36
McIntosh	32°F.	.15	.19	.20	.25	.32	.35
	40°F.(80-95% rel.hum.)	.15	.22	.24	.26	.40	.38
	40°F.(70-85% rel.hum.)	.15	.21	.26	.25	.32	.37
	Controlled atmosphere	.15	.21		.25	.28	.36
Cortland	32°F.	.06	.11	.15	.22	.27	.29
	40°F.(80-95% rel.hum.)	.06	.12	.20	.24	.30	.34
	40°F.(70-85% rel.hum.)	.06	.13	.18	.23	.29	.40
	Controlled atmosphere	.06	.17		.21	.25	.37

Figure 13. Pectin as per cent Calolum Pectate of the ground flesh of Wealthy variety of apple kept under different conditions of storage.

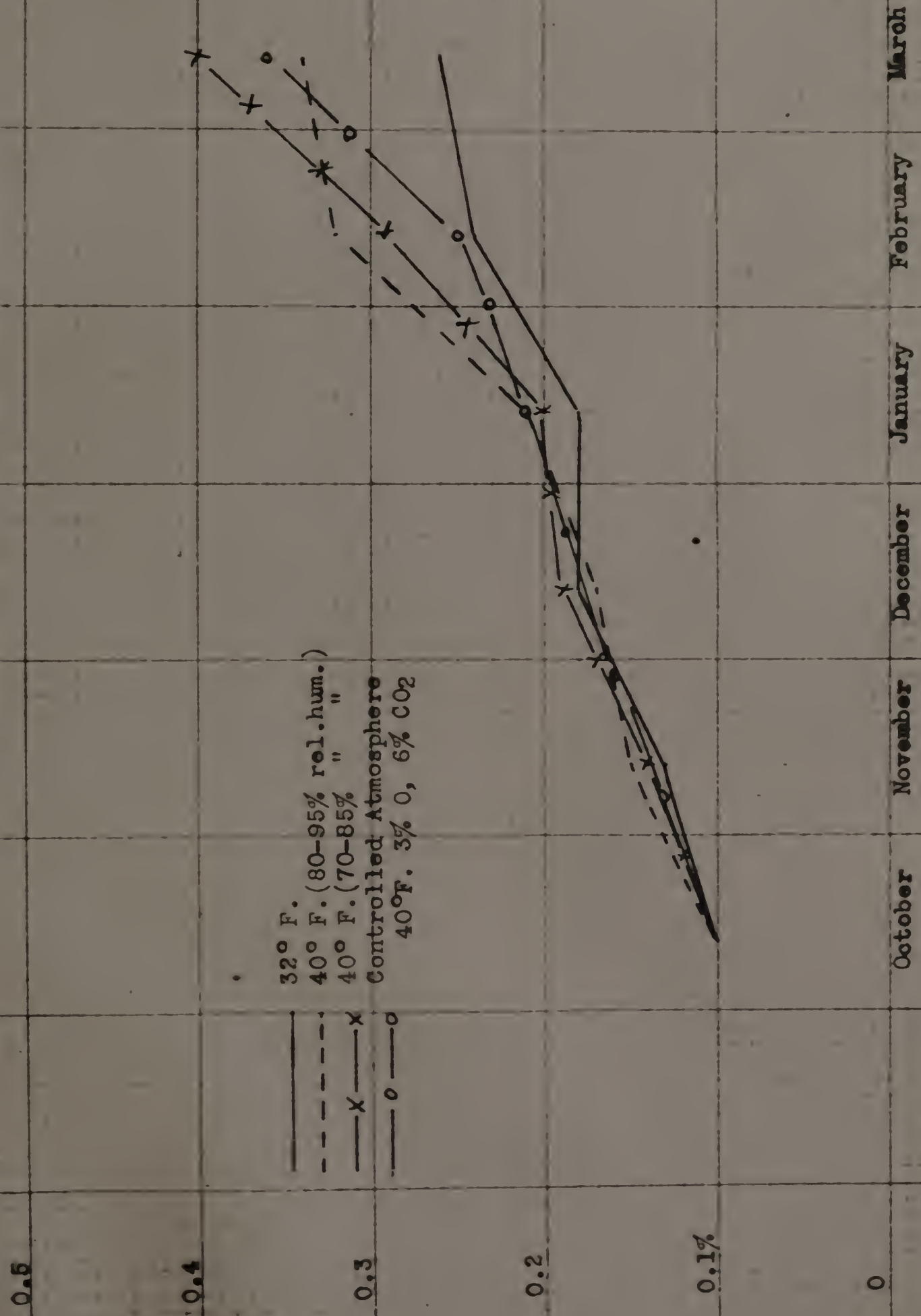


Figure 14. Pectin as per cent Calcium Pectate of the Ground flesh of McIntosh variety of apple kept under different conditions of storage.

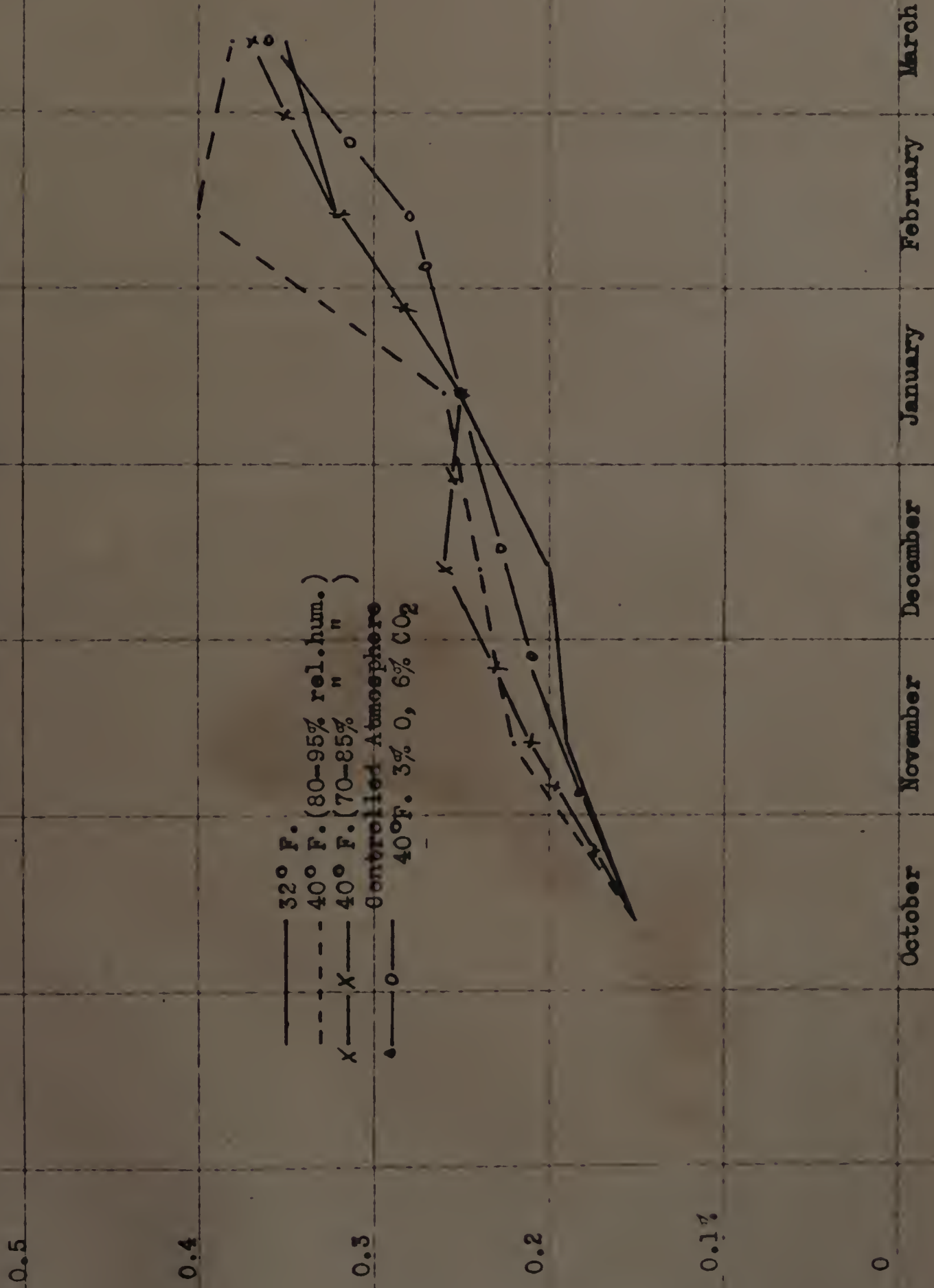


Figure 15.

Pectin as per cent Calcium Pectate of the
ground flesh of Cortland variety of apple
kept under different conditions of storage.

0.5

0.4

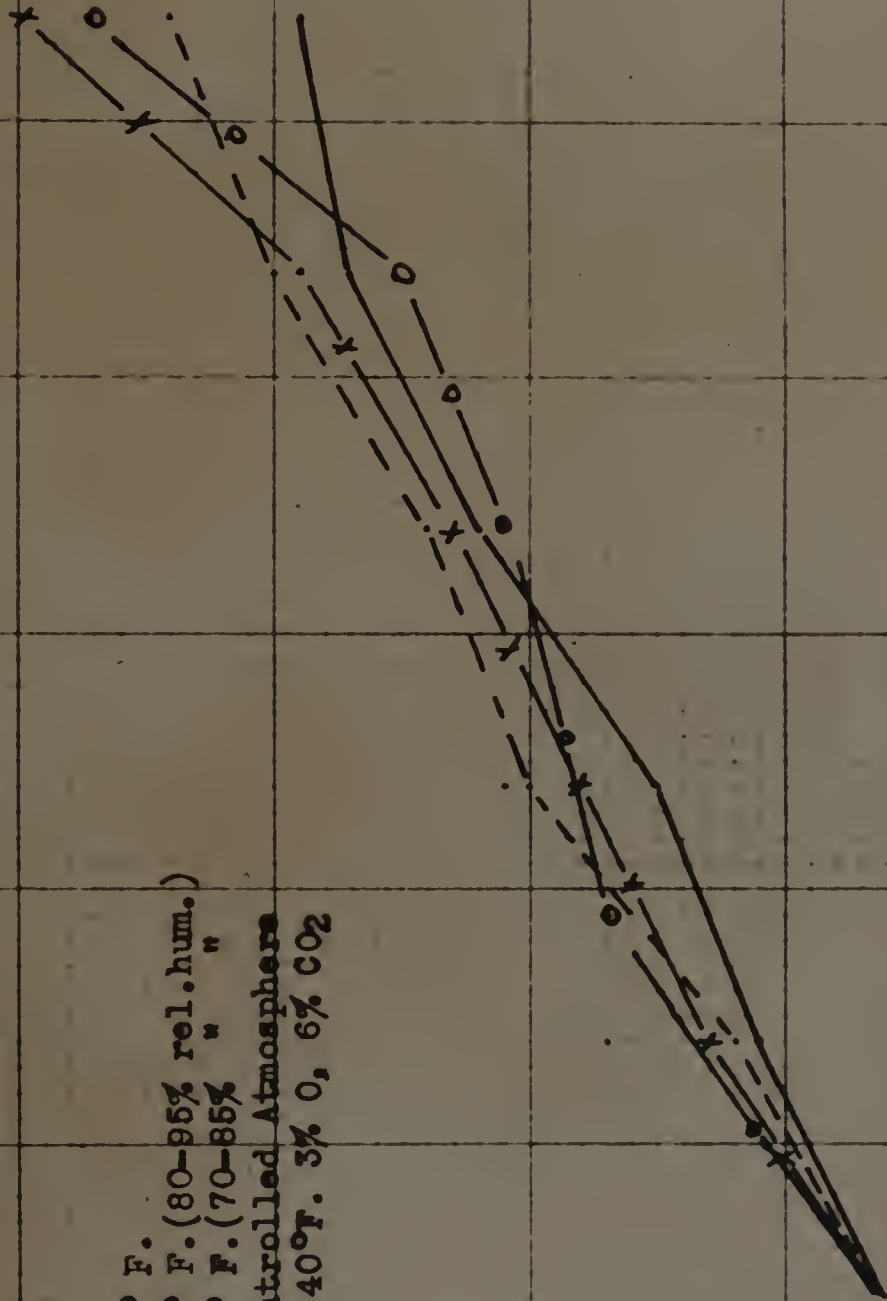
0.3

0.2

0.1%

0

32° F.
40° F. (80-95% rel. hum.)
40° F. (70-85% " "
Controlled Atmosphere
40°F. 3% O, 6% CO₂



October

November

December

January

February

March

Here again the effect of the various conditions of storage is only slight, as shown in Table VI. and graphically in Figures 13, 14, and 15. In general the rate of hydrolysis is retarded at 32° F. (0° C.) over that at 40° F. (4.4° C.). In Cortland for instance the amount of soluble pectin after 2 months of storage at 32° F. is comparable to the amount found 1 month at 40° F., thereafter the increase in the rate of hydrolysis is much retarded.

At the beginning of the storage period there was almost no soluble pectin. It should be noted that McIntosh had more pectin than Wealthy to start with even though it was picked later.

For Walthy and Cortland, controlled atmosphere storage did not influence much the rate of increase in soluble pectin which is about the same as that at 40° F. (4.4° C.) in air. For McIntosh a smaller accumulation of soluble pectin was observed in modified atmosphere than in the other rooms. A low relative humidity at 40° F. did not prove to have much influence on the rate of pectin increase.

ASCORBIC ACID

Fifteen years ago, the only method used for the determination of ascorbic acid was a biological assay devised in 1922 by Sherman, La Mer and Campbell. For this method guinea pigs were forced to derive all of their vitamin C from the product being tested. If the product fed did not contain enough ascorbic acid, the guinea pig lost weight and showed symptoms of scurvy in 15 to 30 days; a score was given to the animal after autopsy which by comparison could be interpreted in relative amount of vitamin C.

In 1932 Zilva (57) observed that penolindophenol was reduced

by decitrated lemon juice, but he thought that this reaction was not specific to ascorbic acid. The same year a chemical titration was devised by Tillmans and Hirsch (51) on the principle observed by Zilva: a quantitative reduction of 2,6 dichlorophenolindophenol by ascorbic acid. A modification of this method by Morell, given in "Food Analysis" by Winton, was used in this study of ascorbic change during storage.

To extract ascorbic acid from plant cells and to prevent its rapid oxidation Musulin and King (35) found that metaphosphoric acid gives the best results. At the same time to inactivate the enzyme ascorbase which occurs when extracting ascorbic acid and which catalyzes its oxidation, Musulin and King (35) used in addition to 2 per cent metaphosphoric acid, 6 per cent acetic acid or 3 per cent trichloroacetic.

In the case of apples, when the extracting solution is used, the red pigment of the skin and flesh dissolve in it. The solution then becomes pinkish and interferes with a sharp end point in titration, the dye being reddish-pink in acid solution.

It was found that an adsorbant such as charcoal, though it clears the solution from the pigments, cannot be used because it also adsorbes the ascorbic acid.

In titrating a colored solution, the use of a photoelectric colorimeter is advised by Morell.

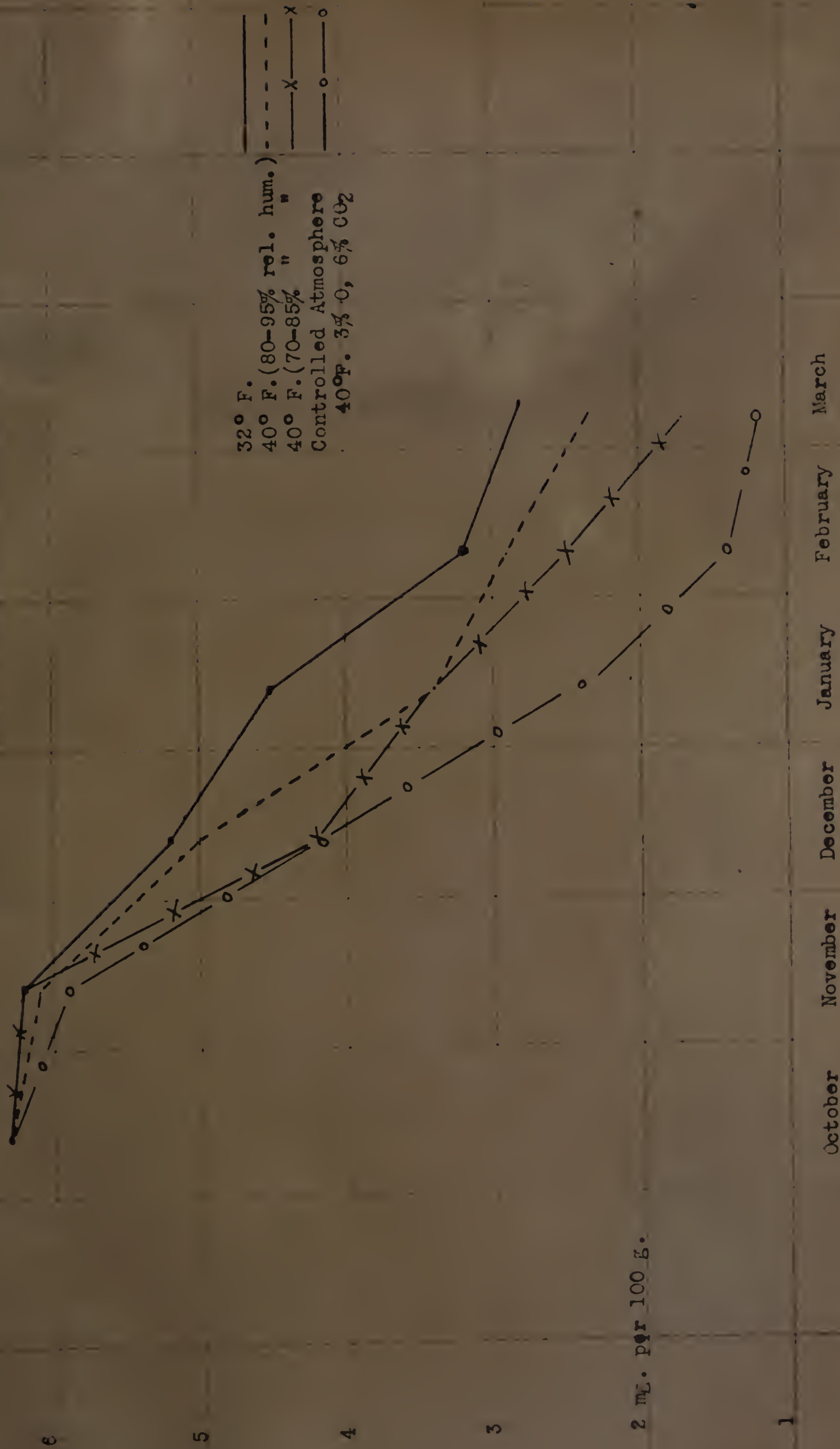
For this method an excess of indophenol is mixed with the solution so an excess of the dye will remain in the oxidized form. During the different determinations the same amount of dye is used and the depth of color is proportional to the amount of ascorbic acid being reduced. These degrees of coloration are measured by the intensity of

Table VII.

Ascorbic Acid mg. per 100 gm. of apple with peel.

Variety tested	Storage condition	Mg. ascorbic acid per 100 gm. after the following months of storage:					
		0	1	2	3	4	5
Wealthy	32°F.	6.3	6.2	5.2	4.5	3.2	2.8
	40°F.(80-95% rel.hum.)	6.3	6.1	5.0	3.4	2.9	2.3
	40°F.(70-85% rel.hum.)	6.3	6.2	4.2	3.4	2.5	1.7
	Controlled atmosphere	6.3	5.9		2.4	1.4	1.2
McIntosh	32°F.	3.1	2.9	2.5	1.6	0.9	0.8
	40°F.(80-95% rel.hum.)	3.1	2.5	1.2	0.9	0.0	0.4
	40°F.(70-85% rel.hum.)	3.1	2.3	1.5	1.2	0.8	0.4
	Controlled atmosphere	3.1	2.2		0.8	0.0	0.0
Cortland	32°F.	5.4	5.4	4.2	3.9	3.1	2.8
	40°F.(80-95% rel.hum.)	5.4	5.3	3.7	3.3	2.7	2.5
	40°F.(70-85% rel.hum.)	5.4	5.1	3.1	2.9	2.5	2.0
	Controlled atmosphere	5.4	5.2		1.5	0.8	0.0

Figure 16. Mg. of Ascorbic Acid per 100 g. of flesh with peel of Wealthy variety of apple kept under different conditions of storage.



2 mg. per 100 g.

Figure 18. Mg. of Ascorbic Acid per 100 g. of flesh with peel of Cortland variety of apple kept under different conditions of storage.

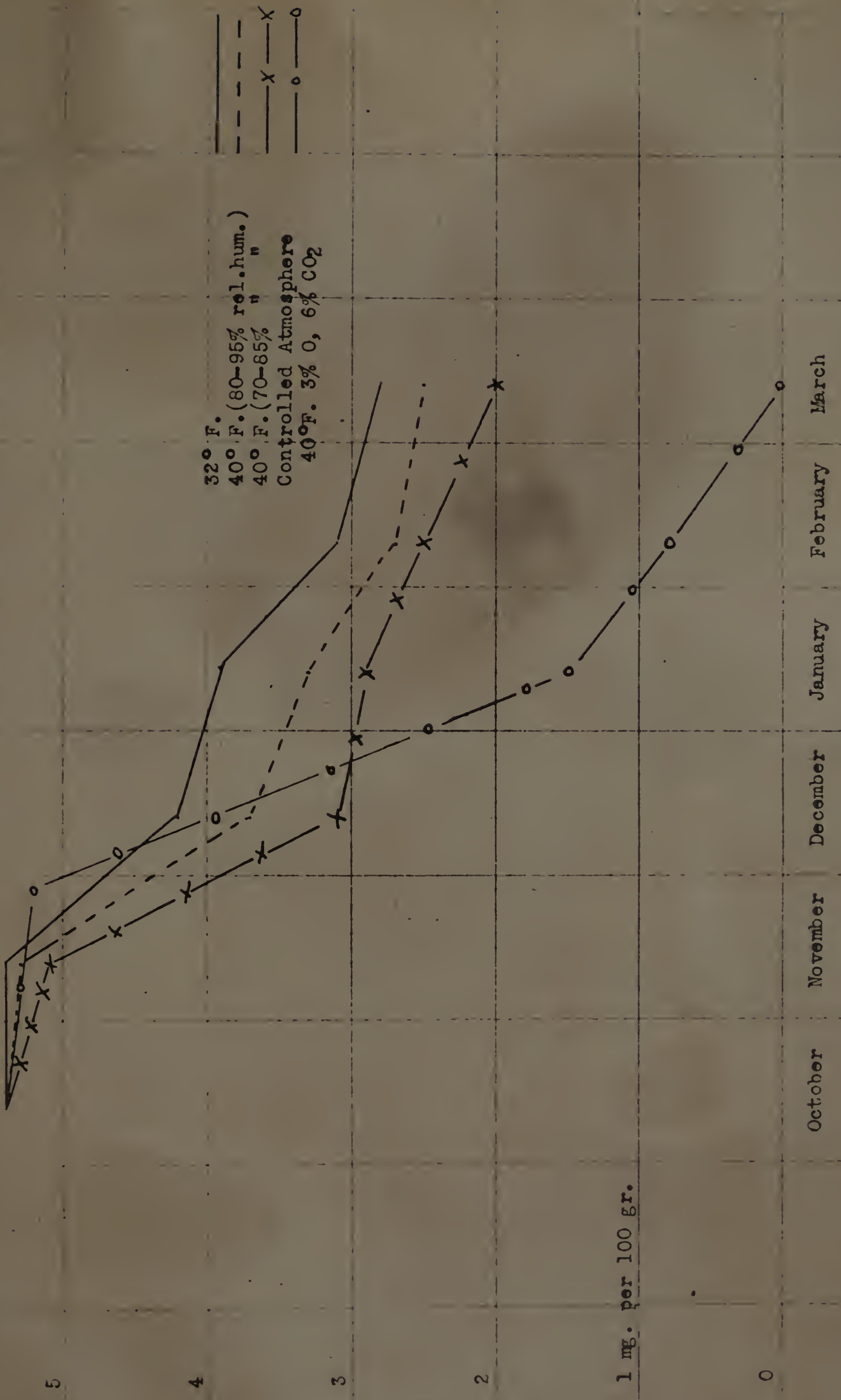


Table VIII

Percentage of loss of ascorbic acid in Wealthy, McIntosh and Cortland varieties of apples after 1, 3 and 5 months under different conditions of storage.

Variety tested	Storage condition	% ascorbic acid loss obtained after the following months of storage:		
		1	3	5
Wealthy	32°F.	1.6	28.6	55.5
	40°F.(80-95% rel.hum.)	3.2	46.0	63.5
	40°F.(70-85% rel.hum.)	1.6	46.0	73.0
	Controlled atmosphere	6.3	61.9	80.9
McIntosh	32°F.	6.2	48.4	74.2
	40°F.(80-95% rel.hum.)	19.4	71.0	87.1
	40°F.(70-85% rel.hum.)	25.8	61.3	87.1
	Controlled atmosphere	29.0	74.2	100.0
Cortland	32°F.	0.0	27.8	48.2
	40°F.(80-95% rel.hum.)	1.9	38.9	53.7
	40°F.(70-85% rel.hum.)	5.6	46.3	63.0
	Controlled atmosphere	3.7	72.2	100.0

mosphere storage generally had good flavor and were more crisp than the others. Storage at low relative humidity had a tendency to give a peculiar storage odor which may have been due to the room and not to the humidity.

SUMMARY

After harvest fruits undergo a series of changes which lead to ripening and later on to complete breakdown. In order to slow down the rate of these changes, cold storage and controlled atmosphere storage have become common practices.

In New England, the varieties McIntosh, Wealthy and Cortland are grown in great quantity and when refrigeration is available they are stored ordinarily at from 32° F. (0° C.) to 40° F. (4.4° C.). More recently the practice has been adopted by a few growers of keeping fruits in controlled atmosphere storage at low oxygen and high carbon dioxide concentration. Although many studies have been made on storage of apples, it was considered interesting to study the behavior of these New England varieties under conditions of storage which are most common in this part of the United States.

Samples of McIntosh, Wealthy, and Cortland were placed immediately after harvesting in rooms at 32° F. (0° C.), at 40° F. (4.4° C.) with 80 to 95 per cent relative humidity, at 40° F. with 70 to 85 per cent relative humidity and in a controlled atmosphere room with about 3 per cent oxygen and 6 per cent carbon dioxide.

At monthly intervals chemical and physical tests were made to follow the metabolism of these apples. The different tests were: Pressure, water content, total acidity, pH, pectin as calcium pectate,

and ascorbic acid content.

- (a) The rate of decrease in firmness varied during storage according to the varieties: the mean of the loss in firmness during five months' storage for the different varieties under the conditions tested was for McIntosh 20.7 per cent, Wealthy 10.4 per cent, and Cortland 22.6 per cent. This rate varied also with storage conditions. McIntosh and Cortland kept in controlled atmosphere maintained a greater firmness than in other conditions of storage. Wealthy, however, lost firmness in that room at a faster rate than under all the other conditions of the experiment. In air, the 32° F. temperature was more favorable to all varieties than 40° F. in maintaining firmness. Cortland and McIntosh softened more rapidly at a relative humidity from 70 to 85 per cent than under more humid conditions.
- (b) The water content of the three varieties remained about the same throughout the storage period. At 40° F., under relative humidity of 70 to 85 per cent the loss for the whole period of storage and for all varieties was about 1 per cent greater than at the same temperature with 80 to 95 per cent relative humidity. It should be noted, also, that fruits kept in controlled atmosphere showed the greater water content. This condition may have been due to the high relative humidity in that room.
- (c) Total acidity calculated as malic acid decreases significantly. During five months' storage the mean of the loss in total acidity under the conditions of the experiment was for McIntosh 60.1 per

cent, for Wealthy 48 per cent, and for Cortland 60 per cent. Temperature influenced the rate of loss of acidity for the first two months of storage only. After five months of storage only slight differences between varieties were noted. At the beginning of the storage period, the three varieties responded quite differently under the different conditions of storage: Wealthy and Cortland maintained high total acidity when stored at 32° F., McIntosh showed most total acidity when kept at 40° F. with high relative humidity.

- (d) pH showed almost no change during storage. A slight increase appeared at the end of the fourth and fifth months but no apparent relation was observed between total acidity and pH.
- (e) Pectin determined as calcium pectate increased during storage in all varieties studied. Storage at temperatures higher than 32° F. in air had a slight tendency to increase the rate of hydrolysis of pectic substances. Modified atmosphere had no particular effect on the hydrolysis of these substances.
- (f) Ascorbic acid content in all varieties decreased greatly during storage, especially in controlled atmosphere storage, where it had almost completely disappeared from McIntosh and Cortland after five months' storage. For all varieties 32° F. was found to maintain the highest level of ascorbic acid. Relative humidity did not influence significantly the rate of loss of ascorbic acid.

CONCLUSIONS

Decrease in firmness, decrease in total acidity and increase in pectin are the most important changes occurring in the apple during storage.

For McIntosh, controlled atmosphere storage or a temperature of 32° F. are advisable because they keep the apple firmer. High relative humidity maintains high total acidity.

Wealthy shows best keeping qualities at 32° F. At 40° F. with high relative humidity the rate of ripening is not much increased as far as loss in total acidity, loss in firmness and increase in pectin are concerned. It could also be concluded that if refrigeration is not available this variety could be kept successfully at 40° F. (4.4° C.) provided the storage was well humidified.

Cortland kept well under all conditions of the experiment but the results indicate that 32° F. (0° C.) or controlled atmosphere would be the most satisfactory conditions for prolonged storage.

Among the chemical tests which can be used to give an idea of the state of maturity of an apple, total acidity and calcium pectate tests are most reliable.

As a practical method of storage, controlled atmosphere should be considered for McIntosh and Cortland. For these varieties it reduced the rate of ripening well below what might be expected at the temperature involved. Wealthy on the other hand kept better at 40° F. without a modified atmosphere.

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