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Practical Seed Tests and Their Uses

C. E. Vaughan

PRACTICAL SEED TESTS AND THEIR USES

Charles E. Vaughan [1/]

One of the jobs of quality control is to minimize the amount of deterioration that occurs during seed production. This requires constant checking with sensitive tests that can detect small amounts of deterioration.

Many kinds of tests have been developed for measuring seed deterioration. Some of these are based on observations made during germination and seedling growth: seedling growth rate, cold test performance, emergence through a layer of crushed brick, respiration rate of germinated seedlings, speed of germination, and germination after subjecting the seeds to stress conditions. Other tests are based on detailed examinations of the seeds: determination of enzyme activity, the tetrazolium test, and measurement of electrical conductivity. Brief descriptions of some of the tests now in the forefront are as follows:

The cold test for corn determines how well seeds withstand seed rotting organisms under cold, wet soil conditions. It is the only vigor test now in routine use in this country. Seeds are planted in a mixture of sand and unsterilized soil, held at 50^{o}F for 7days, then transferred to warm temperatures to germinate. This test reflects the amount of mechanical damage in corn seed and the effectiveness of fungicide applications.

In the past, the cold test has been used almost exclusively on corn seed lots. Some of these uses were to: (a) determine the quality of carry-over seed; (b) evaluate seed treatments; (c) test the effects of processing methods on the seed; (d) evaluate vigor in parent stocks; (e) compare the resistance of inbreds and single crosses; (f) evaluate the adequacy of storage conditions with respect to seed deterioration; and (g) determine the extent of frost injury and immaturity and its effect on seed vigor.

Recently, however, we have found that the cold test is applicable to many other crops and shows great potential for predicting storability and field performance of various seed lots of the same kind.

There are, however, several factors which affect cold test performance. Among these are: (a) degree of seed maturation; (b) age of seed; (c) amount of mechanical damage; (d) seed treatments; (e) genetic resistance; and (f) degree of organism infestations.

[1/] Associate Agronomist, Mississippi Seed Technology Laboratory

Some other factors also make the cold test extremely hard to standardize among seed laboratories. These are:

1. Different soils used in cold tests. Soil used in various laboratories give different results due to variations in the amount of disease organisms present and differences in soil texture.

2. Different organisms in the soil exhibit various degrees of virulence.

3. Differences in cold test methods. Variations in soil moisture content and length of cold period and temperature cause varying results.

The lack of standardization, however, need not limit the usefulness of the cold test as a quality test. As long as each laboratory uses a consistent method, comparisons can be made among seed lots, varieties, etc. Procedures for conducting cold tests are given on page 76.

Growth rate tests will probably be well adapted and easily incorporated into a regular germination testing program.

Some advantages of growth tests are:

1. They are simple and easy to conduct. Any technician can conduct growth tests.

2. They can be incorporated into a germination testing program at little or no expense.

3. Most growth rate tests can be easily standardized.

First Count

This test can be incorporated into the standard germination test. The number of normal seedlings removed when the preliminary count of the germination tests is made is an indication of the quality of the seed lot. The higher percentage of normal seedlings removed at the preliminary count the better quality the seed lot.

If the first count is to be used to compare different lots of seed over a period of several months, all first counts must be made at the same time interval after planting.

Speed of Germination

If a more detailed test is desired, speed of germination may be used. This test can be incorporated into the standard germination test, but will require more time to evaluate than a regular, standard germination test. After the seed have begun to germinate they must be checked daily at approximately the same time each day. Normal seedlings are removed from the test when they reach a pre-determined size. This procedure is continued until all seed that are capable of producing a normal seedling have germinated.

An index is computed for each seed lot by dividing the number of normal seedlings removed each day by the day on which they were removed after planting. Thus quality indexes of 15 (lot A) and 23 (lot B) are obtained in the following manner:

$$\text{Lot A: } \frac{\text{no. seedlings removed daily}}{\text{day after plant.}} = \frac{0}{1}+\frac{0}{2}+\frac{0}{3}+\frac{8}{4}+\frac{10}{5}+\frac{24}{6}+\frac{28}{7}+\frac{24}{8} = (0)+(0)+(0)+(2)+(2)+(4)+(4)+(3) = 15$$

$$\text{Lot B: } \frac{\text{no. seedlings removed daily}}{\text{day after plant.}} = \frac{0}{1}+\frac{6}{2}+\frac{12}{3}+\frac{24}{4}+\frac{45}{5}+\frac{7}{6} = (0)+(3)+(4)+(6)+(9)+(1) = 23$$

Lot B would be considered the better quality of the two lots since the higher index indicates seed quality.

Root and/or Shoot Growth

Another growth rate test that may be easily employed is root length or shoot length or both.

This test involves the measuring of the length of root (or root and hypocotyl) or shoot at a specified number of days after planting in the germinator. When using this system of comparison, the lot making the most growth is considered to have the highest quality. Only normal seedlings should be measured.

Root and/or shootgrowth can be employed with grasses and grains. However, dicots will probably have to be limited to root or root and hypocotyl growth because of the slowness of epicotyl elongation in many dicot species.

The tests we have discussed thus far can be conducted in the laboratory with no additional equipment other than that required for standard germination tests.

In order to make valid comparisons of these tests both the conditions of the test and the conditions of germination must be very carefully controlled. If some replications become dry, for example, growth will be slower and the results of the test will be biased. If alternating temperatures are used, the time of day when the tests are placed in the germinator will affect the result. For example, corn is germinated at an alternating temperature of 20^{o} - $30^{o}C$. If tests are placed in the germinator at the beginning of the $30^{o}C$ cycle and are compared with tests placed in the germinator at the beginning of the $20^{o}C$ cycle, we would expect the test which was begun at $30^{o}C$ to make more total growth than the test which was begun at $20^{o}C$, when measurements are made the same number of hours after the beginning of the test. If laboratory growth tests are to be used it would be well to consider using a constant temperature to avoid this problem.

Speed of Seedling Growth and Seedling Weights

Several variations of laboratory growth rate tests may be used.

Total seedling growth in the greenhouse or field may be measured at a specified number of days after planting. The lot of seed producing the most growth per normal seedling is considered to be the best quality. Seedlings may be cut and weighed, fresh or dry, for additional information. Seedlings producing the most weight are considered to come from the better quality seed.

Emergence tests will require additional facilities, mostly field plots and greenhouses and other investments outside the laboratory. Conditions in the greenhouse or field are much more difficult to control than those in the laboratory; therefore, results can be expected to be more variable.

GADA (glutamic acid decarboxylase activity) is one of several tests of enzyme activity. Glutamic acid solution is added to finely ground seeds. The amount of carbon dioxide evolved from this mixture in 30 minutes is an index of the enzyme activity present in the seeds. Seeds with the highest rate of carbon dioxide evolution are the most vigorous.

GADA appears to be better related to seed quality of monocots rather than to quality of dicotyledonous species such as soybeans and cotton.

GADA is closely related to the storability, root growth and yield of corn. Grabe and Gill have shown yield decreases of 8% and 14% respectively between high and low GADA in corn. Relationships exist between GADA and seed quality in oats, wheat and rice.

The equipment needed for this test is relatively inexpensive. It consists of a water bath for controlling temperature, simple, easy to make manometers, a scale for measuring manometer fluid movement, small containers (1/2 pint jars) and a small grinder.

Some of the advantages of GADA test are:

1. It is simple and easy to conduct.

2. It does not require a large investment in equipment.

3. It can be completed in a short period of time.

Crop varieties differ in GADA; therefore, comparisons for quality must be made within varieties and not between varieties. Also, GADA is too sensitive to be used as an indicator for stand establishment. Procedures for conducting the GADA test are given on page 77.

The tetrazolium test is also based on enzyme activity. Vigor ratings are obtained by close observation of staining patterns and the physical condition of the embryo. Tests which measure the activity of enzymes are among the quickest vigor tests to make. 1/

Changes in permeability of certain kinds of seeds indicate decreasing vigor. Permeability can be measured by soaking seeds in distilled water and then measuring the electrical resistance of the water. Low resistance means that the seeds have deteriorated, allowing materials to leach from the seeds into the water.

Survival of seeds under Accelerated Aging conditions is an index of longevity of commercial storage. Seeds are placed in an atmosphere of 100% humidity and 42-42^{o} for 4 or 5 days. A germination test is then made to determine survival. Procedures for conducting the accelerated aging test are given on page 85.

Respiration rate of germinative seedlings can be used as an index for vigor. Seeds are germinated and the amount of carbon dioxide given off by the seedlings is measured. The more carbon dioxide evolved, the greater the vigor of the seedlings.

The brick gravel method is one if the older vigor tests used in European countries. Germinating seeds are covered with a layer of crushed brick and the ability of seedlings to penetrate this restructive layer is a general measure of seed vigor.

1/ Procedures explained in the Association of Official Seed Analysts "Tetrazolium Testing Handbook for Agricultural Seeds". Don Grabe ed. Contribution No. 29, 1970.

Microscopic examination for mechanical damage is a simple and effective test. Fast green or other stains may be used to make the cracks more visible.

These and numerous other tests are fully described in the literature. With this arsenal of tests available, the job now is to determine which are most suitable for use in quality control programs.

What Are The Best Quality Control Tests?

Several criteria need to be considered in selecting the tests to incorporate in a quality control program. These include (a) cost, (b) time involved, (c) personnel available, and (d) the particular aspect of quality to be tested.

Most of the tests that have been proposed do a reasonably good job of detecting differences in quality between seed lots. Most of these tests have been aimed at predicting difficulties in establishing stands under adverse field conditions and considerable experimental evidence is available on this point. We do not know if most of these tests can be used to measure losses in yield potential and storability of seed lots.

In order to predict potential performances, we must have more than arbitrary tests for "vigor". We must first of all know what levels of deterioration impair various aspects of performance. We must find out in what ways seeds deteriorate physiologically and then relate the condition of the seed to specific performance. The most useful vigor tests will be those that prove to be most closely related to crop performance.

Use of Vigor Tests in Quality Control

Different kinds of tests are probably needed to rate seed lots according to their potential for stands, yields, and storage life. For example, in one series of experiments, a lower yield of corn was not related to cold test performance. On the other hand the cold test was a better indicator of potential stand establishment than was enzyme activity or seedling growth rate. Enzyme activity shows great promise as an indicator of relative storage longevity of seed lots.

When the appropriate tests are perfected, seedsmen will be able to use quality control tests to monitor the quality of their seed production in much the same way that manufacturers control the quality of the goods they produce. Some of these tests have already proven themselves in the laboratory and are ready for field testing under commercial conditions.