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Seed Testing

Seed testing is the art and science of evaluating seed quality for agricultural purposes. Although initially developed for evaluating the planting quality of field and vegetable seeds, it is also valuable for determining the quality of lawn, flower, and tree seeds.

Even though humanity's use of seeds dates back to prehistoric times, the art and science of seed testing have only developed in the last century or so. Until about 300 years ago, our knowledge concerning seed morphology and physiology was limited. As a result, it was possible for unethical vendors to market seed of such crops as alfalfa mixed with sweet clover or other contaminants. Such practices became so widespread and of such serious concern that laws were passed and seed testing procedures established. Berne, Switzerland was the first city to enact seed legislation prohibiting the sale of adulterated clover seed in 1816. The first seed testing laboratory was established in Saxony, Germany, in 1869, and the first one in America was set up at the Connecticut Agricultural Experiment Station in 1876. Today, official seed testing laboratories are found in almost all 50 states and in nearly every country in the world, and many privately operated commercial laboratories are located throughout North America and Western Europe.

Other important milestones in seed testing include the publication of the first "Rules and Apparatus for Seed Testing" by the United States Department of Agriculture in 1897 and the first drawings of seeds in the publication "The Viability and Germination of Seeds" by F. A. Hillman in 1902. In 1915, E. G. Boerner developed the first divider for separating grain, seed and other material; in 1916, H. D. Hughes developed the first seed counter for preparing germination tests; and in 1917, G. N. Collins developed the first seed blower.

The expression seed quality is used loosely to reflect the overall value of seed for its intended purpose. Seed quality is usually a composite of several factors, all of which contribute to the desirability, or planting value, of the seed. The key question is "Why do we test seeds?" There are several reasons. First, and most obvious, is that the dry seed's potential to establish a seedling cannot be determined until the seed has been germinated. However, we also test seeds to determine the genetic (varietal) and mechanical (weed/other crop) components of the seed lot.

L. O. Copeland et al., *Principles of Seed Science and Technology*

Seed testing results provide important information to both the seed producer and purchaser. The seed producer wants to ensure that only a quality product is marketed so that consumers will return for their future seed needs. Seed purchasers need assurance that associated expenses such as field tillage, pesticide applications, and other costs are not lost due to stand failure as a result of planting poor quality seeds. Finally, both buyers and sellers recognize that seed laws have been enacted to aid in the orderly marketing of seed based on the principle of truth-in-labeling. In some cases, differences in reported values on the seed label exist and are litigated in court. The seed testing information and how it was acquired form the foundation of these legal cases and emphasize the need for proper conduct and interpretation of seed test results.

Because of the universal importance and value of seed, many organizations rely on the results of routine seed testing. These organizations vary from local or state agencies to national and international agencies. As a result, it is important that test procedures be standardized and that results be widely reproducible. This means that the tests must be conducted under the same conditions with uniform interpretations. The process toward standardization in the United States began in 1905 when the Annual Appropriations Act was passed which gave the U.S. Department of Agriculture authority to purchase seeds on the open market, to test and publish the results, and to identify individuals or organizations found to market mislabeled seed. The standardized testing of seeds required that a compendium of test procedures be developed. The Rules for Testing Seeds of the Association of Official Seed Analysts were developed to meet this objective. The following considerations discuss some of the specifications put forth in those rules. Individuals interested in a greater knowledge of the procedures for conducting a seed test should consult the AOSA Seed Analyst Training Manual (McDonald et al. 1992).

OBTAINING THE SAMPLE

No seed analysis, regardless of how carefully or accurately accomplished, is any better than the sample on which it is performed. The importance of a representative sample cannot be overemphasized; that is, the sample must truthfully represent the quality of the seed lot from which it is drawn. It is generally assumed that a seed lot is homogeneous. If this were the case, it would be satisfactory to extract a portion (sample) from any portion of the seed lot and presume that it represents the bulk of the seed. However, this seldom occurs. Seed lots are almost never completely homogeneous for at least four reasons. First, heavy and light seeds tend to segregate within the bulk or bag due to gravity, with heavier seeds being found predominantly at the bottom of the container. Second, harvesting of the crop in the field combines seed from differing locations, thus altering the composition of the seed as a result of variations in maturity, lodging, disease, or the occurrence of weeds. Third, failure to adequately blend two or more lots from differing locations at the time of bagging can result in seed lot heterogeneity. Fourth, lack of uniformity in harvesting, storage, and conditioning results in seed lot heterogeneity.

As a result of this heterogeneity in most seed lots, a lot must be sampled and the sample must be representative of the lot. Sampling is usually done in two steps. First, the sample to be submitted to a seed laboratory is drawn from the bulk seed lot and sent to the laboratory for analysis. This is known as the *submitted* sample. Second, when it reaches the laboratory, it must be divided further to a size that can be analyzed. This latter sample is used for the actual analysis and is called the *working* sample.

The Submitted Sample

The sample may be drawn at any time during seed conditioning or after the seed is offered for sale. If drawn during conditioning, it may be taken by hand or small container at frequent intervals during the conditioning operation or automatically drawn at specified intervals by a mechanical sampler. Either technique, if done properly, will give a representative sample. Seed is usually sampled for testing while still in storage or as it is offered for sale (Figure 15.1). Because of the variety of ways in which seeds are stored and offered for sale, they may be found in various types of containers, from small vegetable and flower seed packets, to boxes and cans of grass seed, to large bulk lots of cereal grain seed. Regardless of the container, the seed lot must be properly sampled so the sample is representative. Rules and procedures for sampling under various conditions have been established by the Association of Official Seed Analysts and the International Seed Testing Association. These rules provide for sampling by mechanical samplers, by use of standard sampling probes or triers (Figure 15.2), by hand, or by taking the entire container as the submitted sample.

The Sampling Process

Bulk Seed. A trier, or probe, is recommended for sampling bulk seed, although hand sampling may also be performed if handsful are carefully and randomly taken from well-distributed points throughout the bulk. Hand sampling is limited by the difficulty of reaching all portions of large bulk lots, whereas large probes up to 72 in. in length can be used to sample hard-to-reach locations within the seed lot.

Seed in Bags

When a seed lot consists of six bags or less, each bag should be sampled from well-distributed points throughout the bags. When lots consist of more than six bags, samples should be taken from five bags plus 10% of the remaining bags. Regardless of the lot size, however, it is not necessary to sample from more than 30 bags. Here are some examples:

No. of bags in lot	5	7	10	23	50	100	200	300	400
No. of bags to sample	5	6	6	7	10	15	25	30	30

Seed in Small Containers. Seed in small containers should be sampled by taking at random an entire unopened container from the supply in order to obtain the minimum amount required for the working sample.

Subdividing the Sample

The sample drawn by any of the various techniques may be too large for the submitted sample and should be divided further before submitting it to the laboratory. Further subdivision should be done by a mechanical halving device, such as the Boerner or Gamet dividers. Absolute care should be taken at this point to guard against introducing bias into the sample to be

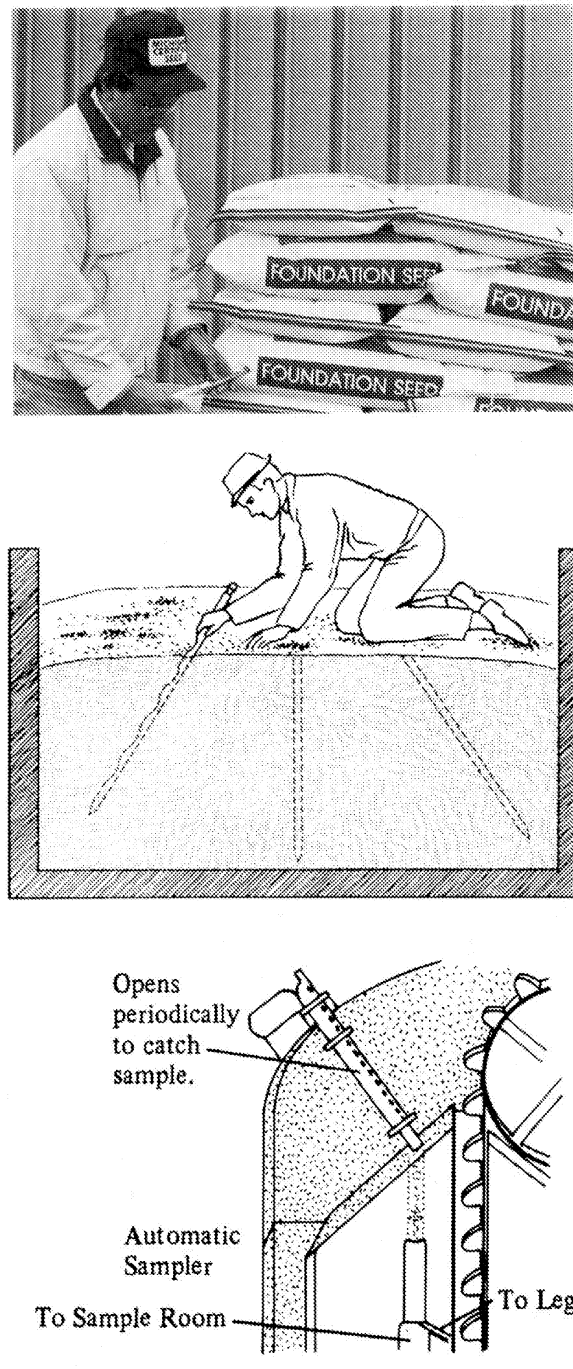

Figure 15.1. Seed sampling techniques: (A) bag sampling; (B) bulk sampling; and (C) mechanical sampling (A, courtesy of Bob Neumann).

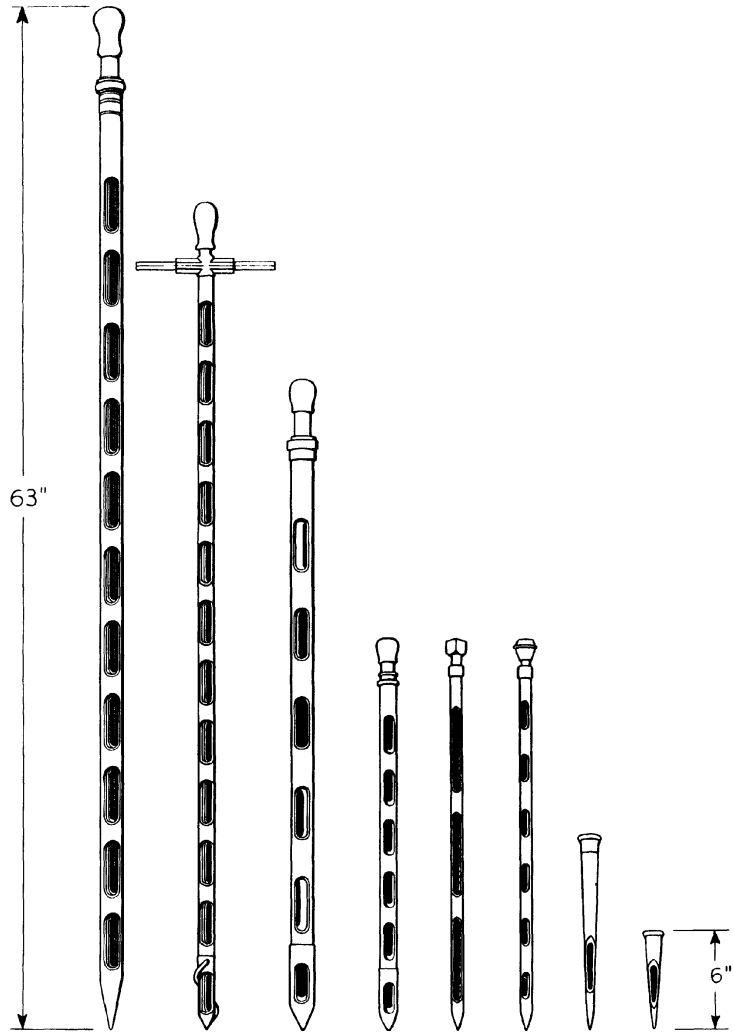

Figure 15.2. Examples of sampling probes. On the far right is a "thief probe."

submitted. During the subdividing process, there may be a tendency to unconsciously remove stones, stems, damaged seeds, or even noxious weed seeds. Such deviations from the correct sampling procedures can make all subsequent testing results meaningless.

Mailing the Sample

After the properly sized sample is obtained, it should be carefully labeled and placed in a container suitable for mailing to the seed laboratory. Cloth, plastic, or paper bags are acceptable; however, these should be placed inside a sturdy cardboard box that can be labeled and can withstand the rigors of mailing. Each sample should be labeled as follows: (1) name and address of owner, (2) crop kind and variety, (3) tests requested, and (4) lot number and number and weight of containers (bags) in the lot.

SUBSAMPLING

When the submitted sample arrives at the seed laboratory, it is entered in the official logbook, assigned a number, and the accompanying information is recorded. The sample then goes to the subsampling area of the laboratory where it is divided into working samples for the various tests that will be performed. Here, the working samples for the purity examination, noxious weed examination, germination, and other special tests are obtained. The remaining portion of the sample is retained as an official sample in case future tests are desired. The weight of the working sample for purity analysis is determined by the weight of seed required to comprise a minimum of 2500 seeds and will vary greatly from small- to large-seeded species.

The Importance of Subsampling

Dividing procedures must be absolutely precise and unbiased if the test results are to be meaningful. The working sample must accurately represent the sample submitted to the laboratory, which in turn represents the seed lot only if sampling procedures were properly followed. In contrast to sampling, sample dividing procedures are generally dependable, because this operation is performed in the laboratory by professional, trained personnel, while sampling from bulk seed lots is often done by persons who may not realize the importance of a representative sample.

Subsampling Techniques

The Rules for Testing Seeds state only that the working sample shall be taken from the submitted sample in such a manner that it will be representative. The actual procedure may be either by manual or mechanical methods. Several hand methods are used. One very simple method, often called the *pie method*, consists of spreading the sample on a clean, flat surface, and dividing it into sections as if cutting a pie. Any of the sections, if randomly selected, may be used alone or in combination with other sections as the working sample. Another hand technique, called the random cup method, consists of placing a number of uniformly sized thimbles or cups on a clean, flat surface and slowly pouring the sample so the seed is distributed evenly over the flat surface filling the cups as the seed is distributed. The working sample may then be obtained by randomly selecting several of the cups until sufficient seed is obtained.

Mechanical halving devices (Figure 15.3) are most often used for subdividing and are dependable for providing a representative sample. These are devices that divide the sample into two equal portions, both in size and content. The working sample is obtained by dividing the submitted sample one to several times until the proper weight of the working sample is obtained. Any of the divided portions may be combined and redivided to yield the proper-sized working sample.

Three mechanical dividers are commonly used for subsampling. Any of these dividers will yield a representative sample; however, each has certain advantages over the others. The Boerner divider is probably the most common; however, some chaffy grasses and other non-free-flowing seeds will not flow through it. A Boerner divider consists of a hopper, inverted cone, and a series of baffles directing the seed into two spouts. The baffles form alternate channels and spaces of equal width. They are arranged in a circle at their summit and are directed inward and downward, the channels leading to one spout and the spaces to an opposite

spout. A valve or gate at the base of the hopper retains the seed. When the valve is opened, the seed falls through the spouts into the seed pans. The Gamet Precision divider requires electrical power to operate and is more suitable for seeds that do not flow through the Boerner divider. The Gamet Precision divider makes use of centrifugal force to mix and scatter the seeds over the dividing surface. In this divider, the seed flows downward through a hopper onto a shallow rubber cup. Upon rotation of the rubber cup by an electric motor, the seeds are thrown out by centrifugal force and fall downward. The circle or area where the seed falls is equally divided into two parts by a stationary baffle so that one-half the seeds fall into one spout and one-half into the other. For non-free-flowing chaffy grass seeds, such as gramma grass and needle grass, the Hay-Bates or a similar divider should be used. For seeds of nondelinted cotton and certain other species, hand-dividing methods of subsampling may be necessary; however, extreme caution must be taken to obtain a representative sample.

PURITY TESTING

When individuals purchase seed, one of the primary decisions that they make is what kind of seed is needed. Their purchase is made with the understanding that the species and variety are the principal constituents of the seed lot. Yet, we know that harvesting and cleaning of seed are not exact sciences. Other types of seed and materials are almost always present. The type and

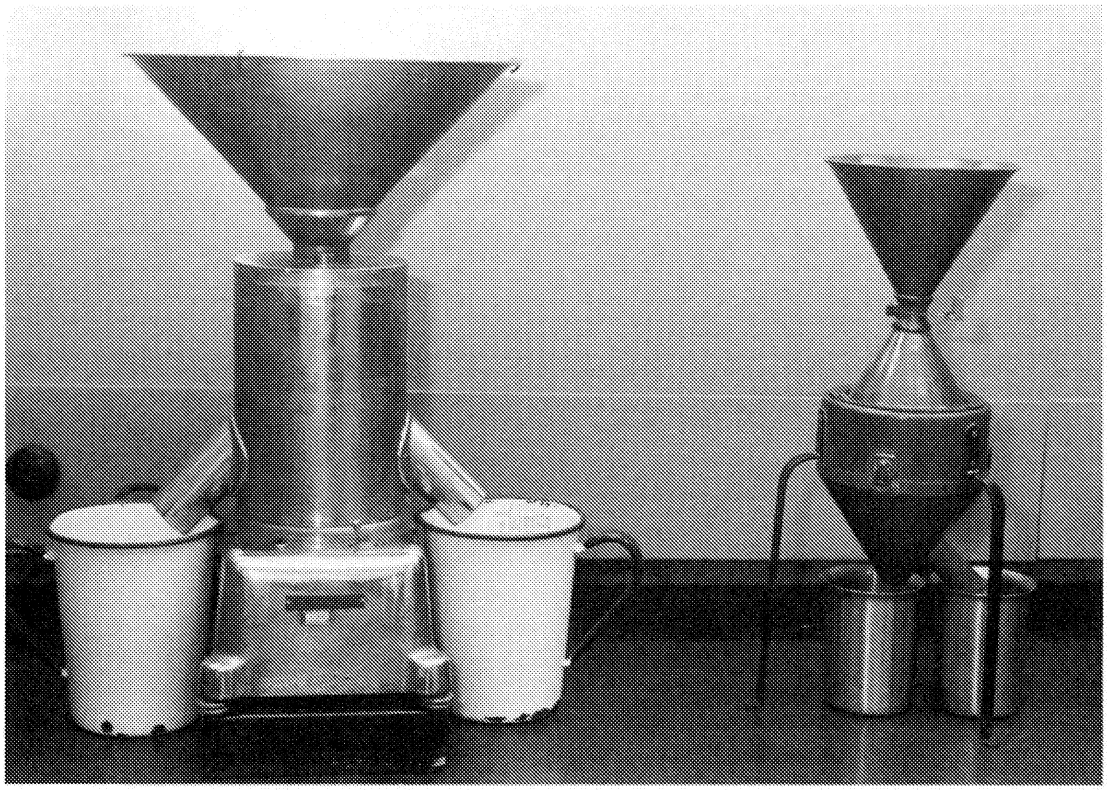

Figure 15.3. Subsampling dividers: on the left is a Gamet Precision divider and on the right is a Boerner divider (Courtesy of Bob Neumann).

level of contamination of these other components can significantly influence the value of the seed. The purity test, therefore, is designed to identify what these contaminants are and how much of them are present.

Seed purity denotes the composition of a particular seed lot. It is based on a physical determination of the components present and includes percentages by weight of: (1) pure seed, (2) other crop seed, (3) weed seed, and (4) inert matter. Pure seed is the portion of the working sample represented by the crop species for which the lot is being tested; in actual practice, it includes the percentage of each crop species present in levels of 5% or more. Other crop seed is the percentage of crop seeds, other than the species being tested, present in concentrations of less than 5%. Weed seed indicates the percentage of seeds present from plants considered as weeds. Sometimes this designation may be arbitrary, since a plant may be considered a crop in one state or country but a weed elsewhere. For any particular region, however, the analyst uses well-accepted guidelines for classifying as crops or weeds. Inert matter denotes the portion of the sample that is not seed. It usually consists of chaff, stems, and small stones, but may include pieces of broken, damaged, or immature crop or weed seeds that do not qualify as entire seeds. The criteria for this distinction are explicit and defined in the Rules for Testing Seeds (AOSA 2000).

The size of the sample on which the purity examination is performed is given in the Rules for Testing Seeds. The sample size (weight) is determined by the size of seed being tested; approximately 2500 seeds are considered acceptable to yield a statistically acceptable test result. The size of the working sample specified in the Rules is larger for large-seeded crops than for small-seeded crops; however, the actual number of seeds in the test is not greatly different.

Philosophy of Purity Testing

The philosophy of purity testing is to avoid judging whether seeds are capable of germinating when performing the test. Consequently, shriveled, immature, frosted, or otherwise damaged seeds are considered as pure seed. This may be called the quick method, in contrast to the strong method used earlier by European seed analysts, who classified crop seed as pure seed only if it appeared to be capable of germination. The quick method left the determination of germination capability to the germination analyst, but is no longer used.

Procedures for Purity Separations

The purity test is perhaps the most complex and exacting of all tests for seed quality. A seed analyst must have a comprehensive knowledge of seed structure and function and must be able to identify a wide array of differing species. For this reason, it is not uncommon to find that many seed analysts have their own working seed herbaria to assist in the identification of unknown samples. Seed herbaria are useful because published descriptions of seeds are rarely as comprehensive as those for plants and viewing actual specimens can be very helpful. An average seed herbarium contains 2,000-3,000 specimens, but varies in size depending on the range of seed materials typically encountered in the seed testing laboratory. The herbarium can be arranged alphabetically or by phylogeny. Phylogenetic arrangements are by plant families and then according to species within the family. This approach assures that specimens which are closely related are placed together. Its disadvantage is that many seed analysts are not familiar with taxonomic relationships which vary according to the authority

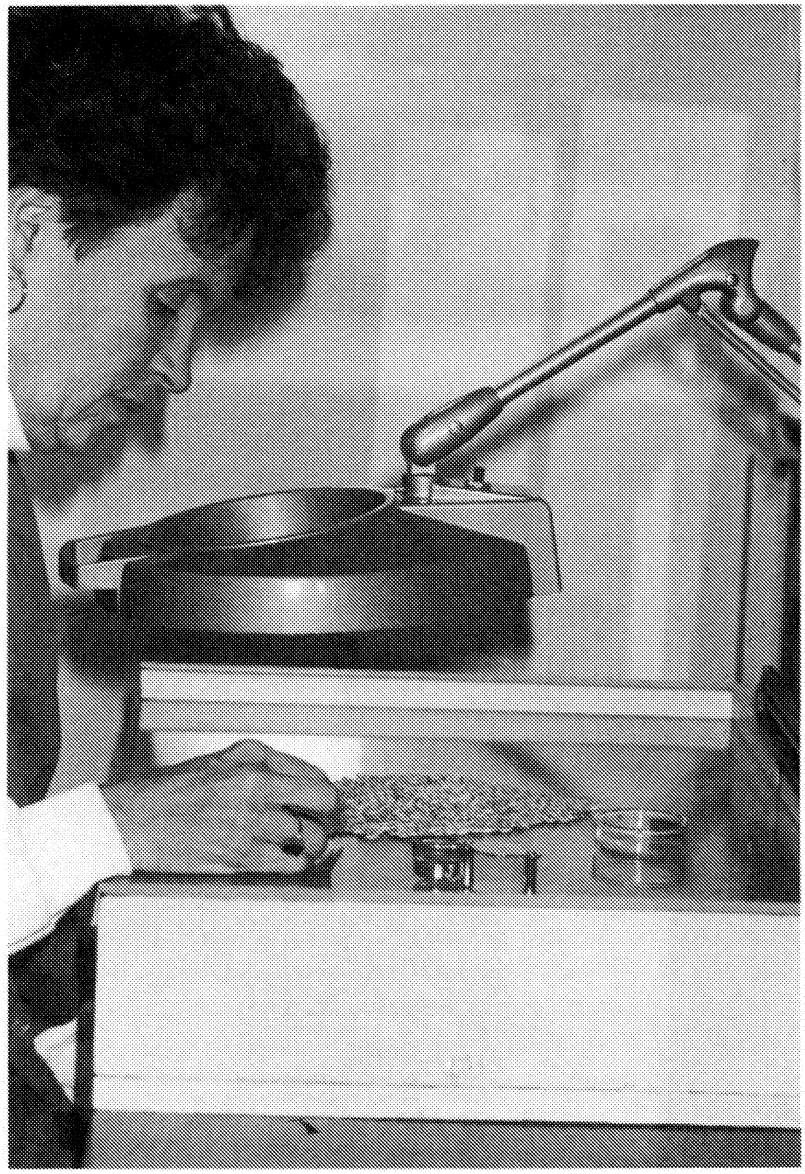

Figure 15.4. A purity testing station (Courtesy of Bob Neumann).

used to classify them. As a result, some analysts simply file specimens alphabetically according to family and then species. This approach offers the advantage of rapid retrieval of specimens but does not afford the opportunity for direct comparisons with related groups.

Purity separations are usually made by hand, although mechanical aids may be used to speed up the analysis or make it less tiring for the analyst. Purity analysis equipment usually includes a work board, adequate light, forceps, a hand lens, and a stereoscopic microscope for identifying small seeds.

Various mechanical devices are frequently used to aid in the purity analysis. The use of seed blowers (Figure 15.5) has contributed to speeding up and improving uniformity of purity analyses, especially for grass seeds, for use in separating empty florets, bits of stems and leaves, chaff, and other inert material. The Rules for Testing Seeds provide uniform blowing procedures for the purity determinations of small-seeded grass species in lieu of the more time-consuming hand separation procedures. The blowing procedure not only speeds up the test but reduces variability among seed laboratory results.

Another difficult area for purity testing is caused by the increased use of coated and pelleted seeds (see Chapter 13 on Seed Enhancements). The Rules for Testing Seeds specify coated seeds as a seed unit covered with any substance which changes the size, shape or weight of the original seed (seeds coated with ingredients such as, but not limited to, rhizobia, dyes and pesticides are excluded). This process alters the shape of flat seeds that are difficult to mechanically plant, for example, and makes them round by adding clay fillers so that the pelleted seed can easily roll into planters. While seed coatings are extremely valuable from a practical perspective, they make the purity analysis more difficult. The analyst must first separate the coating material (often 90% of the dry weight of the seed sample) to determine what is actually present in the clay pellet. This separated portion is weighed as other components of a purity test and recorded as "coating material." To aid in purity testing, seed scientists at

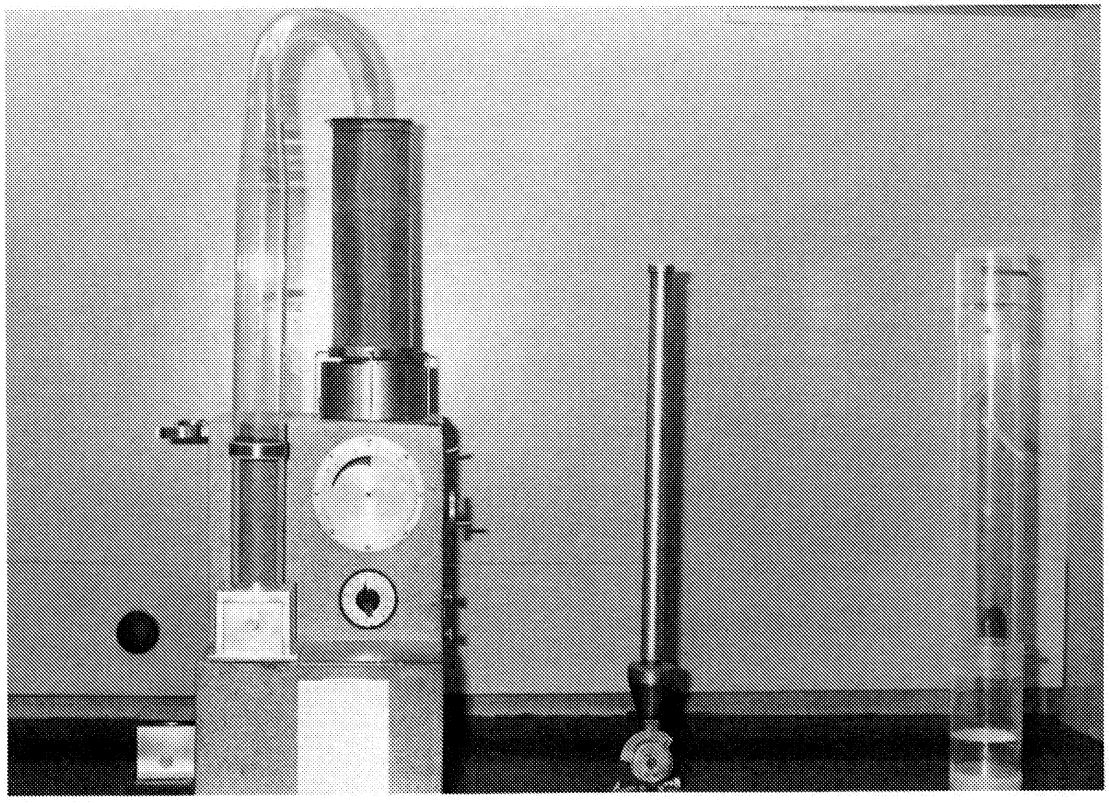

Figure 15.5. Seed blowers: (left) the General seed blower, (middle) the Ottawa blower, and (right) the South Dakota blower (Courtesy of Bob Neumann).

Oregon State University have developed a microscopic inspection station, as well as a vibrator-separator to help purity analysts in making their separations. This type of equipment is used in several North American seed laboratories.

NOXIOUS WEED SEED EXAMINATIONS

Each state has established an official list of noxious weed seeds. In general, the plants from these seeds are particularly troublesome and objectionable. Such lists are a part of the state seed law (or regulation) and are usually defined in two categories, *primary* (or *prohibited*) and *secondary* (or *restricted*) noxious weed seeds. Sale of seed lots containing primary (or prohibited) noxious weed seeds is prohibited), while the sale of lots containing secondary (or restricted) noxious weed seeds is permitted, but their number per pound (of crop seed) is limited. Since each state has its own seed law, the weed seeds listed as noxious are not necessarily the same from state to state.

The noxious weed seed examination is an attempt to provide special information about noxious weed seed content of seed lots. Because of the limited occurrence of noxious weed seeds, a large working sample is used for the noxious weed seed examination-often as much as 500 grams for large-seeded types down to 2.5 grams for small-seeded types. The occurrence of noxious weed seeds in the United States is reported in number of seeds per pound or ounce. Since state seed laws and noxious weed seed lists vary among states, the results of this examination may be reported for only one state, or it may be reported for two or more states. Crop seeds transported in interstate commerce should have an "all-state" noxious weed seed examination.

GERMINATION TESTING

Probably the single most convincing and acceptable index of seed quality is the ability to germinate. Seeds are tested for germination because a seed lot is composed of a population of individual seed units, each possessing its own distinct capability to grow and produce a mature plant. A seed germination test is an analytical procedure to evaluate seed viability and germination under standardized (favorable) conditions. It enables a seed vendor to determine and compare the quality of a seed lot before it is marketed to the consumer. Furthermore, the percent germination can be used to determine the planting value of a seed lot, its storage potential, and labeling information required to provide for standardized marketing of seed lots. Thus, germination testing is perhaps the single most important function of a seed testing laboratory. Since the process of seed germination is covered in Chapter 5, this discussion will cover only the laboratory techniques used for performing the analysis.

Procedures for Germination Testing

The germination test is ordinarily performed on the pure seed of the crop kinds that constitute 5% (or more) of the sample after all inert matter and other crop and weed seeds are removed. Each pure seed kind is germinated and reported separately. A minimum sample of 400 seeds is recommended for a statistically dependable germination test. These are usually planted in four replicates of 100 seeds each, although various other arrangements are sometimes used (Figure 15.6). Each replicate is evaluated separately, but the official germination report is an