

Native Seed Quality Bulletin



Native Seed Quality Task Force
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Preface

The Native Seed Quality Task Force assembled in June 2005 in response to requests from the seed industry and their concerns about the lack of understanding of native seed testing and lack of uniformity in the tests. The result was a determination that there is a need for a publication to explain differences between the natives and the crops classed as agricultural row crops.

The Task Force was selected from across the seed industry. The members are as follows:

Name	Representing Organization
Sue Alvarez	SCST
Dave Buckingham	AASCO (retired April 2010)
Chuck Dale	AASCO
Rod Fritz	ASTA
Tim Gutormson	SCST
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Brenda Watts	AOSCA
Morgan Webb	CSAAC
Garth Wruck	Consumer
Rich Wynia	NRCS

We hope that the readers will find this educational and a source of further information into their understanding of the complexities of the Native Seed Industry.

1

Understanding Native Seed Quality

RICHARD WYNIA AND VICTOR VANKUS

Landowners utilize native plants in many different ways. Some landowners are interested in restoring an area to its original native vegetative state. Others stewards of the land are interested in protecting their soil from erosive forces, feeding livestock, or providing cover and support for wildlife species. Whatever the landowner desires, native trees, grasses, and wildflowers can be used to help meet their goals.

Producing high quality native seed depends on a number of factors. Understanding the growth cycle of a native species, the origin or location of a harvested field, weed control, adequate environmental conditions for plant and seed development, proper harvest timing, drying of the seed post harvest, conditioning or cleaning of the seed, and seed storage are all important to producing high quality native seed.

Establishment Expectations

Establishment of native species is not an easy task. Germination of many native plant species and of species in a native seed mixture is not uniform. Many native plant species do not become established the year of planting. Perennial native species that do germinate tend to prioritize first year growth to the root system with reduced emphasis on the above ground stem and leaf production. Thus, slow or reduced shoot and leaf growth can be anticipated the first year of growth. These facts are discouraging and somewhat disheartening for novice native plant enthusiasts that expect to witness aggressive growth and flower proliferation the initial planting season. Patience is the first lesson learned from growing native plant species. Successful establishment of native plants requires good tillable land, a firm and weed-free seedbed, quality seed, good seed-to-soil contact, adequate rainfall or irrigation, good weed control, time, and a certain amount of luck.

Seed and plant producers and end-users have different requirements, expectations, and preferences for seed germination and plant establishment. Some end-users desire uniform germination in a short period of time; others want germination over an extended period of time. Many native seed are planted as part of mixtures and establishment and species composition may change over time.

Seed Storage

Proper storage conditions are critical to maintaining viability of seed over time. Storing seed at a low temperature and at low moisture content will have a significant impact on maintaining seed viability. The storage life of a seed lot will double for every 5-10° F decrease in temperature above 32° F. and for each 1% reduction of moisture content above 6%. Ideal long-term storage conditions for most species are below 10% moisture content and below 32°F. Seed stored in sealed bags and containers are not exposed to the ambient air and are not subjected to fluctuating relative humidities. Storing dry seed in sealed containers will also increase the storage

life of a seed lot. At relative humidities of 60% and lower, most seed not stored in sealed containers will equilibrate with the environment at moisture content below 13%. Seed of many native species can safely be stored at this moisture content for up to a year.

Seed Testing and Pure Live Seed

Testing the quality of native seed is a challenge for seed testing laboratories. Seed of native plants often have variable degrees of dormancy, a natural adaptive characteristic that allows native plant seed to germinate over an extended period of time. This adaptation will not limit the native plant species to a single germination event that could lead to establishment failure if unfavorable environmental conditions exist at the time of germination. The degree of dormancy in a native seed lot is determined by genetics and is influenced by environmental conditions at the time of development and may change from year to year or from location to location across the range of the species. Determining test conditions and procedures for overcoming dormancy and testing germination is often a lengthy process that must account for these seasonal and geographic differences. Due to this complex process, and the fact that seed of so many native species have entered into the market at the same time, many native species do not have standardized germination testing procedures in the *AOSA Rules for Testing Seeds*.

Pure Live Seed (PLS) is a number used to report the quality of a native seed lot on a seed test report or label. The PLS is a product of two tests, purity and viability. The Pure Seed Unit (PSU) of some native species can be difficult to determine. This difficulty influences the viability test and increases the likelihood of variable purity test results and PLS among laboratories testing the same material. Another source of variation among laboratory PLS results is the type of viability test conducted. Some laboratories use the tetrazolium test (TZ) to determine viability, others use a germination test alone, and still others use a combination of germination and TZ. This variability in native seed testing methods and reporting between laboratories means test results can vary dramatically in some instances. This is a big problem for the native plant industry, because it makes it difficult to determine the value of a seed lot.

Nomenclature

Each year more native species are introduced into the marketplace to meet the growing demand for plants and seed. It has become increasingly important to ensure that each plant or seed lot is correctly identified by its scientific or Latin name. Many native species have more than one common name and common names may change from one part of North America to another. Using the correct scientific name in addition to a common name will eliminate any confusion that can arise when using a common name alone. The USDA Agricultural Research Service (ARS) Germplasm Resources Information Network (<http://www.ars-grin.gov>) and the USDA Natural Resources Conservation Service (NRCS) PLANTS (<http://www.plants.usda.gov>) both maintain databases with current taxonomic information for native North American plant species.

2

Seed Sampling

DAVID BUCKINGHAM

Sampling is very important to accurate analysis of any seed lot. The sample is the beginning of laboratory analysis. Native plant species can present unique challenges to the goal of obtaining a representative sample. It is important that a sampler observe the physical characteristics of the respective seed kinds when obtaining a sample as these characteristics will determine the proper sampling procedure.

Some native seed kinds have structures that resemble tufts of fine hair, multiple florets, and other physical qualities that limit the flowability of the seed. The process of “debearding” can remove these structures and greatly improve seed flowability. Sampling seed that has not been debearded often cannot be accomplished with traditional sampling equipment (i.e., a seed probe or trier) because the seed will not easily flow into the seed probe. These lots have to be sampled by hand.

It is also important that the sampler be aware of the analytical weight required for the seed kind being sampled. Some native seed are not as dense as traditional seed kinds and therefore require more sample volume to obtain an adequate weight for analysis. It is a good practice to weigh the sample to make sure the proper weight for analysis has been collected.

Another important consideration is relative component size in mixtures of native species. Mixes that contain components with extreme differences in shape and density can be sampled, but the sample is most usually not going to be a representative sample for physical purity determinations. Mixes comprised of dissimilar components easily separate with handling and are difficult to mix correctly.

It is important to keep the above considerations in mind when sampling seed lots of native plant species. The purpose of sampling is to obtain a representative sample of a definite, identified quantity of seed. This quantity of seed is commonly referred to as a seed lot. The accuracy of the laboratory analysis as representative of the seed lot is dependent upon the sample being representative of the seed lot.

Sampling procedures and information presented here are taken from the Association of American Seed Control Officials’ *AASCO Handbook on Seed Sampling* (2006). General procedural guidelines that apply to all sampling activities include: 1) use of appropriate sampling equipment and precisely following procedures; 2) taking equal portions from each container sampled; 3) randomly or systematically taking samples throughout the seed lot; 4) sampling the proper number of containers; and 5) gaining access to the entire lot.

Selecting and Using a Trier

A trier should be used for sampling free-flowing seed in bags, larger bulk containers, and bulk storage areas. The trier should be long enough to reach within portions of the container being sampled. The width of the slot openings should be at least two times the maximum diameter of the seed being sampled. Triers are available with single and double sleeves.

Single-sleeved triers used to sample bags should be inserted diagonally on the horizontal plane from corner to corner with the slot down, exercising care not to puncture the container on the opposite side. The trier should then be rotated so that the slot is directly up, and then withdrawn.

The same insertion procedure applies to double-sleeved triers used to sample bags. The sleeve should be inserted in the closed position aligned so that the slot is facing up. The inner sleeve is opened, allowing seed to fill the sleeve, and then closed and withdrawn from the container. Care has to be exercised when closing the sleeved trier as damage to the seed can result if the trier is forcefully closed.

Double-sleeved triers are better to use when sampling bulk seed because the trier can be closed, which ensures containment of the seed in the trier as it is withdrawn. The trier should be inserted in the closed position on the vertical plane at a slight tilt. The trier is then opened, allowing the seed to fill the trier, and then closed, exercising care to not damage the seed when closing, and withdrawn from the bulk.

Obtaining a Hand Sample

Non-free-flowing seed, such as chaffy grasses, uncleaned seed, large fragile seed, or screenings that are difficult to sample with a trier, may be sampled by hand. Sampling intensity (the number of hand samples taken) should be the same as the number of bags detailed in the bag sampling chart (Table 2-1). To obtain a hand sample, insert the hand flat, fingertips down, with the fingers pressed together until the desired depth is reached. Close the hand into a fist and withdraw the sample. Hand samples should be taken from various locations in bags or bulk to obtain a representative sample.

Primary Sample Size, Random Sampling, and Access

Each individual trier or hand sample obtained from a container in the seed lot being sampled is called a primary sample. Each primary sample should contain an equal amount of seed. All of the primary samples are usually emptied into one container. This single container then holds the composite sample of the seed lot. Each primary sample should be taken in such a way to ensure random sampling. A true, random sample can only be obtained using a statistical table, where all containers are numbered and selected for sampling based on the table. Following this procedure in a field situation is usually not practical. Thus, effort should be made to systematically obtain primary samples from evenly-spaced containers across the entire seed lot. This will sometimes require that pallets be moved to have access to the entire lot.

Sampling the Seed Lot

Before starting to sample, determine the lot identification and number of bags in the seed lot. Primary samples should be obtained from unopened bags. If the seed lot to be sampled is in bulk, estimate the equivalent number of bags if the seed were packaged in a customary size, then obtain the appropriate number of primary samples based on the estimated bag number. When sampling a bagged seed lot, it is **very important** to observe the lot identification on each bag sampled. It is also important to look for and note any differences in the appearance of the bags that might indicate the seed lot is mislabeled. The number of primary samples to obtain is as follows:

1. For lots of six bags or less, each bag shall be sampled and a total of at least five primary samples shall be taken. Remember that the same number of primary samples should be taken from each bag.
2. For lots of more than six bags, five bags plus 10% of the number of bags in the lot shall be sampled. Regardless of the lot size, it is not necessary to sample more than 30 bags. A minimum number of bags or containers must be sampled to ensure representation of a seed lot. Refer to Table 2-1 below.

Table 2-1. Minimum Number of Bags Which Must be Sampled.

Number of Bags in Lot	Minimum Number to Sample	Number of Bags in Lot	Minimum Number to Sample
1 to 6	*		
7 to 14	6	135 to 144	19
15 to 24	7	145 to 154	20
25 to 34	8	155 to 164	21
35 to 44	9	165 to 174	22
45 to 54	10	175 to 184	23
55 to 64	11	185 to 194	24
65 to 74	12	195 to 204	25
75 to 84	13	205 to 214	26
85 to 94	14	215 to 224	27
95 to 104	15	225 to 234	28
105 to 114	16	235 to 244	29
115 to 124	17	245 to 254	30
125 to 134	18	255 or more	30

*For lots of one to six containers, sample each container and take at least five primary samples.

Recommended Sample Sizes

Each seed kind sampled will have a minimum weight that must be obtained in order for a laboratory to perform the desired tests. Some native plant species exhibit low densities; this needs to be considered when obtaining a sample. Sampling intensity in Table 2-1 are minimums; it may be necessary to sample additional bags to obtain enough seed for testing purposes. Weighing the composite sample to ensure you have enough weight for analysis is suggested for lighter seed kinds. A complete listing of the amount of seed required for each seed kind that could be sampled is impossible to provide in this format. The information can, however, be easily obtained by contacting the seed laboratory to which you intend to submit the sample for testing. This information may be available in the AOSA *Rules for Testing Seeds*, if the seed kind you are submitting has an official testing procedure. Seed lots intended for shipment outside of the U.S. may require a larger amount of seed than that required by AOSA Rules and a greater sampling intensity than described in the sampling table presented in this guide. Your seed laboratory can advise you of the proper number of containers to sample and the correct amount of seed to obtain.

Sampling Seed Mixtures of Native Plant Species

Sampling seed mixtures of native plant species can be problematic. A representative sample can be obtained from mixtures where components are similar in shape, size and density. Mixtures with components that have a wide variety of shapes, sizes and densities usually cannot be sampled in a manner that results in a representative sample of all components. Dissimilar mixture components have a tendency to segregate, even during the mixing process. Segregation will also occur when containers are handled. A good analogy is mixing marbles with sand. Obtaining a representative sample of each individual component is the suggested method. Calculated percentages of each component in the mixture based on the weight added to the mix are then the basis of the component values to be stated on the seed analysis tag. Consumers should be advised to use equipment designed to mix the seed lot during planting to assure an even distribution of all components.

Summary

An accurate analysis of a seed lot begins with obtaining a representative sample. Deviations from the sampling guidelines described should be noted when the sample is presented to a seed laboratory for analysis. The methods employed to obtain a representative sample are as significant to the final result as are the methods used to analyze the sample in the laboratory. A seed laboratory following standardized procedures will provide an accurate analysis of the sample received. A properly sampled seed lot will allow the seed laboratory to correctly quantify the components of the seed lot. Some native plant species require more attention during sampling because of obvious differences in weight, flow characteristics, and different levels of processing. A person sampling native plant seed kinds should consider these differences when obtaining the sample.

3

Seed Testing

Purity Analysis

DEBORAH J. LIONAKIS MEYER AND JAMES EFFENBERGER

The purpose of a purity test is to quantify the amounts of pure seed, inert matter, and contaminating species in a lot of seed. This is accomplished by examining a representative sample of the seed lot in the laboratory approximately equal to 2,500 pure seed units (PSU). The weight of the working sample will vary depending on the species tested, but the examination of 2,500 seed units is a standard recognized within the Federal Seed Act (FSA) and the Association of Official Seed Analysts (AOSA) *Rules for Testing Seeds*. Results of the purity analysis are reported as percentages by weight of each component found in the working sample. Classification of other crop and weed seed contaminants follow the guidelines prescribed by AOSA (Meyer and Wiersema, 2010).

The following types of equipment may be used during the purity analysis: magnifying lens, microscope, diaphanoscope, sieves, seed blower, analytical balance and seed herbarium (Figure 3-1).

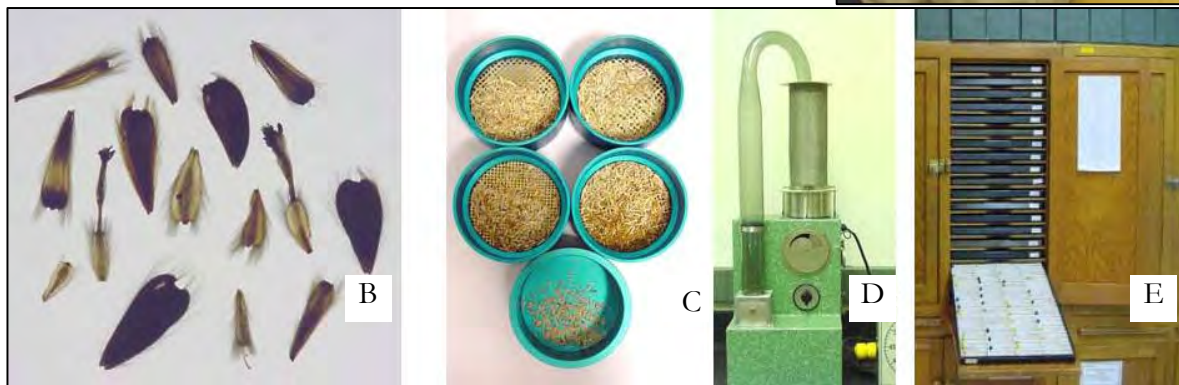


Figure 3-1. **A.** Purity examination on a standard purity station conducted under magnification lens. **B.** Achenes of *Encelia californica* viewed through a microscope over a diaphanoscope (illuminated from below the seed); immature achenes are transparent. **C.** Sieves are used to efficiently separate particles by size. **D.** General-type seed blowers are used to efficiently and uniformly separate pure seed from inert matter by means of air velocity. For species such as *Bouteloua gracilis*, *B. curtipendula* and *Puccinellia distans* the Uniform Blowing Procedure utilizing calibrated blower settings is required. For other species the blowing procedure is not required, but blowers can be used to speed up the purity separation. **E.** Seed herbaria are valuable reference tools used to identify contaminating species.

What constitutes a PSU will vary with the species being tested. In general, the PSU is described as “the structure usually regarded as a seed in planting practices and in commercial channels” (AOSA 2010a). Specific descriptions of 51 types of PSUs representing 749 agricultural, vegetable, tree, shrub, flower and range, or re-vegetation species can be found in the AOSA Rules. The simplest PSU is the true seed; however, for many species the true seed alone does not adequately represent the structures considered part of the natural planting unit. A PSU may also include the fruit, other floral parts (e.g., calyx) and accessory structures (e.g., bracts and stems). In some cases seed units may contain more than one seed or may contain no seed. The latter generally occurs in species where the fruit wall is thick and it cannot be determined if a seed is present without dissection or x-ray examination, neither option being practical nor cost effective in a standard purity analysis. Whether a PSU contains one seed, more than one seed or no seed at all, it is treated as a single unit both for purity testing, as well as for germination and viability testing. In addition, with few exceptions, seed units that are immature, shriveled, broken, diseased, insect-damaged, or have started to germinate are considered pure seed.

Whatever structures are included in the PSU of a particular species, it is important to remember the need for consistency within and among laboratories so that purity results can be compared. Additionally, whether a purity analysis is performed or not, PSU must be used for germination or TZ viability tests otherwise the results of these tests cannot be compared among laboratories or with label claims.

Confusing Types of Seed Units

In the grass family (Poaceae) the flower consists of the pistil, stamens, and lodicules (Figure 3-2). Members of this family have extra floral structures (e.g., lemma, palea, glumes) surrounding the flowers that may or may not remain attached to the fruit at maturity (Figure 3-3). Because of the variety of structures within the grass family it is difficult to have one general seed unit description for seed testing purposes.

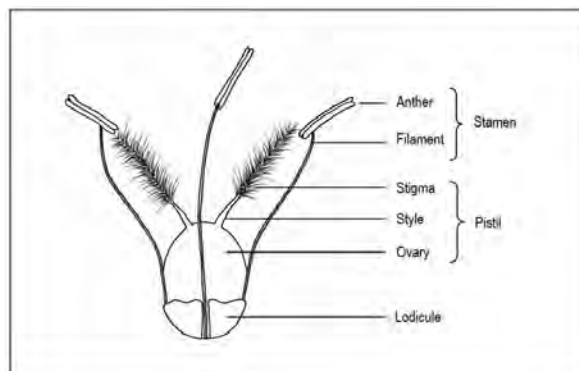


Figure 3-2. Basic grass flower (from Meyer, 2001).

In the AOSA Rules there are 16 separate definitions for members of the grass family (Table 3-1). These definitions provide a broad coverage of the grass family, such that if a species to be tested is not in the AOSA Rules an existing seed unit description for a similar species in the AOSA Rules can be, in most cases, successfully applied.

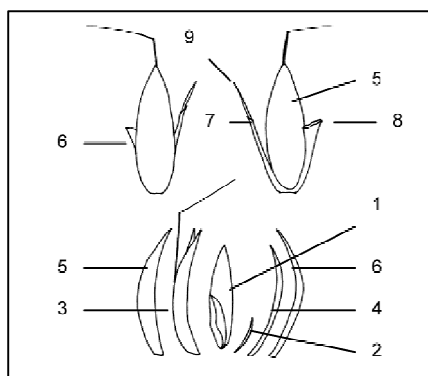


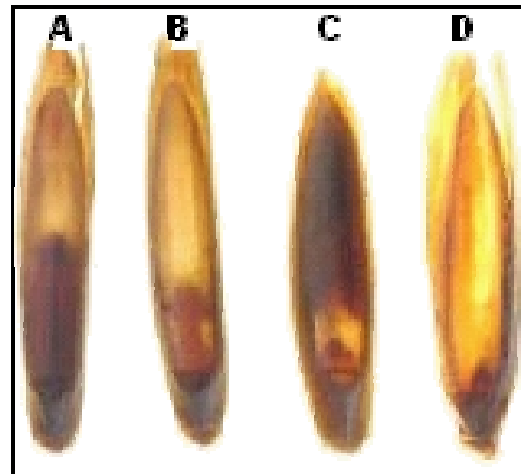
Figure 3-3. Spikelet structure in *Schizachyrium scoparium*. 1 – caryopsis, 2 – palea of fertile floret, 3 lemma of fertile floret, 4 – sterile lemma, 5 – 2nd glume, 6 – 1st glume, 7 – pedicel, 8 – rachis segment, and 9 – sterile spikelet (from Meyer, 1996)

Table 3-1. Comparison of structures in the grass family considered part of pure seed units (PSU) (AOSA, 2010a).

Species	AOSA PSU #	Caryopsis	Floret	Multiple floret	Spikelet	Spikelet group	Attached rachis and/or pedicel	Fascicle, involucre, bur or other
<i>Elymus trachycaulus</i>	12	X	X					
<i>Zoysia japonica</i>	13	X	X		X			
<i>Panicum virgatum</i>	14	X	X	X	X			
<i>Sorghastrum nutans</i>	15	X	X		X		X	
<i>Schizachyrium</i>	16	X	X		X	X	X	
<i>Elymus elymoides</i>	17	X	X	X	X	X	X	
<i>Pennisetum ciliare</i>	18	X			X			X
<i>Bouteloua dactyloides</i>	19	X						X
<i>Coix lacryma-jobi</i>	20	X						X
<i>Festuca arundinacea</i>	21	X	X					
<i>Pascopyrum smithii</i>	22	X	X	X	X		X	
<i>Puccinellia distans</i>	23	X	X	X	X		X	
<i>Dactylis glomerata</i>	24	X	X	X	X		X	
<i>Bouteloua curtipendula</i>	25	X						
<i>Poa bulbosa</i>	47	X						X

Most PSU definitions for grass species require a caryopsis be present, while the pure seed unit definitions for *Bouteloua dactyloides* and *Coix lacryma-jobi* do not because the nature of the seed unit structures makes it difficult to determine if one is present. *Poa bulbosa* reproduces via aerial bulblets and thus does not have the requirement for a caryopsis. For species requiring a caryopsis, there are three basic definitions: 1) a caryopsis with some degree of endosperm development (e.g., *Pennisetum ciliare*, *Elymus canadensis*, *E. elymoides*, *E. trachycaulus*, *Panicum virgatum*, *Psathyrostachys juncea*, *Pseudoroegneria spicata*, *Schizachyrium scoparium* and *Sorghastrum nutans*); 2) a caryopsis at least one-third the length of the palea measured from the base of the rachilla (e.g., *Agropyron cristatum*, *A. desertorum*, *Thinopyrum elongatum*, *T. intermedium*, and *Pascopyrum smithii*) (Figure 3-4); and 3) a caryopsis may or may not be present, however no analyst judgment required because the Uniform Blowing Procedure is applied (AOSA, 2010a, 2010b) (e.g., *Dactylis glomerata*, *Poa pratensis* and *Puccinellia distans*). For species covered by the first two definitions, care should be taken in the examination of the seed units because application of pressure to a seed unit (a recommended method in the AOSA Rules) to determine if a caryopsis is present or of a certain length can cause damage, particularly if the mature caryopses of the species being tested contain liquid, semi-liquid or soft endosperm (Terrell, 1971). The alternative to application of pressure is the examination over a diaphanoscope, during which the seed units are illuminated by high intensity light from below making the internal structures visible (Figure 3-4).

Figure 3-4. Comparison of caryopsis length in *Thinopyrum elongatum* florets illuminated by diaphanoscope: A and B – caryopses are greater than one-third the length of the palea measured from the base of the rachilla therefore these florets are classified as pure seed units; C and D – caryopses are less than one-third the length of the palea, therefore these florets are classified as inert matter. In contrast, if these florets were of *Elymus trachycaulus* all four (A to D) would be classified as pure seed units because there appears to be some degree of endosperm development.



In the sunflower family (Asteraceae) the typical seed unit consists of a one-seeded indehiscent fruit derived from an inferior ovary. This structure is generally referred to as an achene by many authors, although when the fruit bears attachments (e.g., awns or bristles) derived from accessory parts that extend beyond the apex of the fruit the correct term is cypsela (Spjut, 1994). The fruit wall is usually well developed while the seed coat and endosperm that surround the embryo are greatly reduced. The mature seed contains a linear embryo that usually fills the entire fruit. Presently there are three PSU definitions in the AOSA Rules covering approximately 90 members of the sunflower family. The main differences between these PSU definitions being the recognition of attached structures (e.g., beak, bristle, hairs, pappus, or wing) as part of the seed unit and whether or not the intact seed unit must contain a seed. Originally developed to cover highly milled cultivated crops (i.e., endive, lettuce, salsify, and sunflower), the PSU definitions for the sunflower family included intact achenes, whether or not a seed is present (AOSA, 1954). For the species originally covered by this definition, and for other species in the family with thick fruit walls subsequently added to the AOSA Rules, it would be impractical to determine whether a seed is present, as this would require dissection or x-ray analysis. However, for many other species in the sunflower family the fruit wall is thinner and detection of a seed within the fruit requires only simple visual examination with a diaphanoscope (Figure 3-1) or application of slight pressure to the fruit with forceps. For fragile non-free-flowing fruit, extensive milling to remove immature (empty) fruit is not an option because it could damage the filled fruit, potentially increasing the percentage of inert matter and decreasing the percentages of pure seed and germination. The temptation in the laboratory is to consider immature fruit as inert matter because it is obvious no seed is present; however, the current AOSA PSU definition require intact (undamaged) fruit, filled or not, to be classified as pure seed except for fruits of *Artemisia* and *Layia*. For these two genera, a seed with some degree of embryo development, as detected by applying slight pressure or by examination over light, must be present in the intact fruit (Figures 3-5, 3-6). The exceptions for *Artemisia* and *Layia* were recently added to the AOSA Rules, but this does not address situations with other species in the AOSA Rules with similar thin-walled fruits, nor does it address the potential for confusion when testing similar species that are not covered by the AOSA Rules. Alvarez (2005) provides a history of the sunflower family in the AOSA Rules and of recent research to improved PSU definitions addressing the issue of visibly empty intact fruit. Examples of other families in which empty intact fruit are considered pure seed include: Aceraceae, Apiaceae, Betulaceae, Boraginaceae, Brassicaceae, Chenopodiaceae (Figure 3-7), Cornaceae, Fabaceae, Fagaceae, Geraniaceae, Juglandaceae, Lamiaceae, Malvaceae, Nyctaginaceae, Oleaceae, Polygonaceae, Ranunculaceae, Rosaceae, Simaroubaceae, Ulmaceae, Valerianaceae, and Verbenaceae.

Figure 3-5: *Dimorphic* achenes of *Layia platyglossa*; the two forms of achenes are found in the same inflorescence head. Ray achenes are located around the perimeter of the head, while the disk achenes are centrally located. **A.** Portion of commercial sample prior to purity analysis; because of the potential for damaging the seed units further milling to remove inert matter is not an option. **B.** Ray achenes enclosed by phyllary bracts. **C.** Ray achenes with phyllary bracts removed – achene on left containing an embryo is classified as pure seed, while achenes at center and right do not contain embryos (determined by applied pressure) and are classified as inert matter. **D.** Disk achenes – the two on the left contain embryos and are classified as pure seed, while the two on the right do not contain embryos and are classified as inert matter.



Figure 3-6. *Artemisia tridentata* – portion of a commercial sample, prior to purity analysis, viewed through a microscope and illuminated by a diaphanoscope (inset shows PSU). The percentage of inert matter in commercial seed lots of this species is generally >85% (Alvarez, 2005). Empty fruit (i.e., lacking seed with some degree of embryo development) are classified as inert matter.



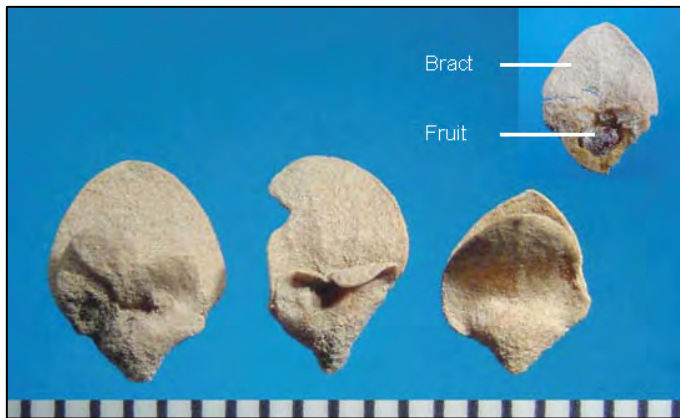


Figure 3-7. *Atriplex confertifolia* fruit surrounded by fruiting bracts. This species is not in the AOSA Rules, but for lab testing should be treated similarly to *A. canescens*, which is in the AOSA Rules. The PSU consists of the intact fruit with or without the fruiting bracts, whether or not a seed is present.

Conclusion

Correct classification of a pure seed unit will not only affect the outcome of a purity analysis but also will impact germination and TZ viability testing. It is critical that all testing laboratories utilize the same criteria for pure seed whether described in the AOSA Rules or determined by consensus among laboratories in order for test results to be comparable among laboratories.

Seed Viability

LOREN WIESNER

Germination

Native species are valuable resources for revegetation of disturbed ecosystems. The success of these plantings is dependent on the native species selected, quality of seed used, and the environmental conditions before and after planting. There are many factors that determine seed quality such as harvesting seed at physiological maturity, proper drying of seed after harvest, quality of conditioning, and the use of proper storage. Monitoring the quality of the seed with purity and germination or tetrazolium tests during harvesting and conditioning is critical in maintaining the quality of seed being produced.

Most lots of mature native seed contain dormant seed (see Glossary p. A-1), and this creates problems when conducting germination tests. Therefore, most seed lots need a combination of seed germination and tetrazolium tests to determine the amount of viable seed.

The environmental conditions during seed development will determine the quality of the seed and the degree of dormancy. Dry conditions during seed development will cause the seed to mature before it reaches physiological maturity resulting in small seed that lack vigor when planted. Generally immature seed will not have as high a degree of dormancy as mature seed. Limited nutrients during seed development can cause seed to be smaller and can cause seed to be more dormant. Low temperatures during the latter stages of seed development can also increase dormancy. These are general concepts concerning the causes of poor germination and the degree of dormancy; however, these concepts vary with species.

Most native seed are classified as orthodox, meaning that they can be dried to low moisture contents and stored at sub-zero temperatures. They have 28 to 50% moisture content at physiological maturity. Some of the native seed that are wetland species could be classified as recalcitrant. Recalcitrant seed cannot be dried below 30% moisture and cannot be stored at subzero temperatures. Some of the recalcitrant species that are found in temperate regions are citrus, willow, poplar, cottonwood, elm, river maple and wild rice. Recalcitrant seed are not dormant, will germinate as soon as they are shed from the plant, and have 50 to 70% moisture content at physiological maturity. There is a third classification of seed that is between the orthodox and recalcitrant called intermediate. Intermediate seed can be dried to moisture contents below 30%, but not to low moisture contents of 5 to 10%. Intermediate seed can be stored at low temperatures, but not subzero temperatures.

Seed germination is another phase in the life cycle of plants, and just as with all other developmental process of the plant, it is genetically programmed and environmentally controlled.

Seed of some species, such as smooth brome grass and ryegrass, are capable of germinating a few days after fertilization. Other seed such as soybeans will only germinate after they have matured and dried down. Still other seed will not germinate when they are mature due to dormancy factors such as chemical inhibitors or impermeable membranes.

There are two types of seed germination, epigeal and hypogeal. Epigeal germination is characteristic of beans, cucumbers, and many types of tree seed such as pine and ash. During this type of germination the cotyledons and plumule are pulled out of the soil by the elongation of the hypocotyl. The exposed cotyledons provide nutrients for the growing seedling through the stored food they contain and cotyledons of many species are capable of producing food through photosynthesis. Photosynthesis is the process of utilizing carbon dioxide, water, and light to produce nutrients for the plant. Hypogeal germination is found in peas and all grasses. During hypogeal germination the plant food storage organ remains beneath the soil and the plumule, protected by the coleoptile, pushes through the soil and emerges. The coleoptile, which protected the plumule during elongation through the soil, stops when it reaches the soil surface due to sunlight. The plumule then pushes through the tip of the coleoptile.

Germination is controlled by the environment in which the seed is placed. If the environment is not suitable for germination the seed will not germinate. There are four factors of the environment that control germination: water, temperature, gases, and light. Germination is also controlled by certain hormones. Water uptake (imbibition) is the first phase of seed germination. The process of imbibition depends on the composition of the seed such as oil, starch and protein content, permeability of the seed coat to water and gases, and the availability of water and temperature of the soil. Water uptake of the seed is phasic and is described as having three phases: imbibition phase, lag phase, and the radicle emergence phase. The imbibition phase is a physical process where the seed swells and water occupies the free capillary spaces and the intercellular space of storage materials. As tissues expand they create imbibitional pressure. Both viable and nonviable seed swell due to water uptake. Temperature will affect the rate of imbibition, but the amount of water imbibed will be the same. The salt content of the soil will also affect rate of imbibition — as the salt concentration increases, the rate of imbibition decreases.

Seed coat permeability greatly influences imbibition. Certain structures of the seed aid in water uptake, such as the micropylar area and the hilum. The hard seededness of certain species such as legumes has been attributed to small elongated pores in the seed coat, which do not allow for channels through the seed coat, and a thick layer of wax over the seed coat surface.

Each species has a range of temperatures in which they germinate – a minimum, maximum, and an optimum. Germination of a species at its optimum temperature usually results in the highest and most rapid germination. An increase in temperature does not necessarily cause an increase in either the rate of germination or the percentage of germination. Germination rate is not characterized by a simple temperature relationship. This can be understood if it is appreciated that germination is a complex process, and a change in temperature will affect each seed constituent differently.

The temperature at which different seed germinate and the range within which they germinate is determined by the source of the seed, genetic makeup of the seed, and the age of the seed. The rate of germination, regardless of the temperature, follows a sigmoid curve. First a slow initial rate followed by a rapid linear rate, and as germination is nearing completion the rate slows again. Many seed require a diurnal (alternating) temperature before they will germinate. Most forage grasses and weedy species usually require diurnal temperatures. The temperatures used for the diurnal effect is not as important as the need for alternation of temperatures as long as the temperatures used are within the normal range for germination of the species.

Alternating temperatures most frequently used are 68 to 77°F, 68 to 86°F, 68 to 95°F, and 50 to 77°F (15 to 25°C, 20 to 30°C, 20 to 35°C, and 10 to 25°C, respectively). The alternation of these temperatures occurs in a 24-hr period, with the low temperature period being utilized for 16 hours and the high temperature utilized for eight hours. Generally, seed of species that benefit from diurnal temperature are from species that have dormant seed (see Glossary p. A-1). The reason for the effects of alternating temperatures on germinating seed is unknown. Some scientists feel it is the differential influence on the sequential steps of germination. Another explanation is that alternating temperatures create a balance between inhibitor and promoter in dormant seed. The inhibitor decreases during the low-temperature cycle and the promoter increases during the high-temperature phase, which results in germination of dormant seed.

Prechilling of imbibed seed, also known as cold stratification is another method of overcoming seed dormancy. Seed are placed on a moist germination medium and placed in a cool chamber 41°F (5°C) for a given period of time (five days to six months). After the seed have been prechilled, they are placed in a germinator at the normal temperature used for germinating nondormant seed of the species.

All seed require moisture, oxygen and a favorable temperature for germination; however, several species also benefit from light or dark conditions for germination. Many wild species and some agricultural and horticultural species (lettuce and tomato) require light to germinate. Seed that require light for germination are called photoblastic. Both light intensity and light quality influence germination. The influence of light intensity on different species varies greatly. A light intensity of 100 to 200 foot-candles, which is found in most seed laboratories, is usually adequate for germination. Modern seed germinators usually provide adequate light for germination. The quality of light necessary for germination of light sensitive seed is the red light (660 nm) region of the light spectrum. Far-red light (730 nm) inhibits germination of light sensitive seed. The

photoreversible pigment which controls germination of photoblastic seed is known as phytochrome. It is a bluish colored protein pigment under far-red light and fades to a colorless form in the presence of red light. The far-red absorbing form, often abbreviated as Pfr, is induced by exposure to red light and is the biologically active form which promotes germination. Short flashes of red or far-red light will convert the phytochrome to the biologically inactive, red-absorbing form, often abbreviated as Pr. Prolonged periods of darkness will cause the beneficial far-red absorbing form to be converted to the phytochrome red absorbing form which will result in dormancy. High temperatures or exposure of the seed to far-red light will also result in the conversion of the beneficial far-red absorbing form to the red absorbing form and result in dormancy. Light sensitivity of germination is influenced by species, cultivar, imbibition period, imbibition temperature, seed age, and germination temperature.

Energy requiring processes in living cells are sustained by processes of oxidation. Respiration increases rapidly during seed germination. Since respiration is an oxidative process, an adequate supply of oxygen must be present for germination of most species. However, the amount of oxygen present in air (20%) is usually adequate for germination. Several species such as rice, aquatic plants, and barnyardgrass germinate under water where oxygen is present in limited supply. Rice seed can germinate even in complete absence of oxygen, but the seedlings are weak and abnormal. There are some seed that will germinate more rapidly if the oxygen concentration is increased above 20%. Some of these species are beardless wildrye, cereals, curly dock, and wild carrot. Carbon dioxide usually is detrimental to germination. Seed dormancy can be attributed to the prevention of free exchange of gases due to impermeable seed coats or to insufficient gas availability due to extremely wet conditions.

Respiration in the dry seed is low, but when water is imbibed there is an immediate release of gas from the colloids and free capillary spaces called the "wetting burst". This release of gas is not due to respiration, but to water filling the intercellular spaces within the seed. Enzyme activation begins during Phase I and II of imbibition. During Phase II, the seed undergoes many processes essential for germination. The seed begins to lose weight due to increased respiration and leakage of nutrients from the seed. In Phase III, radicle emergence occurs and an increase in water uptake occurs as the radicle becomes functional. Oxygen utilization in the seed increases as rapid oxidation of the stored food reserve occurs and seedling growth begins. In monocots (e.g., grasses), the breakdown of the stored food reserves is triggered by the activity of gibberellic acid on the hydrolytic enzymes present in the aleurone layer of the seed. These enzymes move from the aleurone layer to the endosperm and break down the starch, protein, lipid, and energy containing compounds to provide nutrients for seedling growth.

Emergence of the radicle from the seed is the first sign of germination and generally is the result of cell elongation and cell division. Cell elongation occurs first and then cell division. In most seed, cell elongation initiates the protrusion of the radicle through the seed coat and then cell division causes continued growth of the radicle. The first stage of radicle growth is slow and occurs without significant increase in dry weight. The second stage of radicle growth is marked by rapid increases in both dry and fresh weight and by a rapid mobilization of nutrients into the radicle. Once the radicle has emerged, germination is completed and the seedling gets its nutrients from the remaining stored food reserves. As reserves are depleted seedlings rely on photosynthesis for continued growth.

Tetrazolium Testing

LARRY PRENTICE

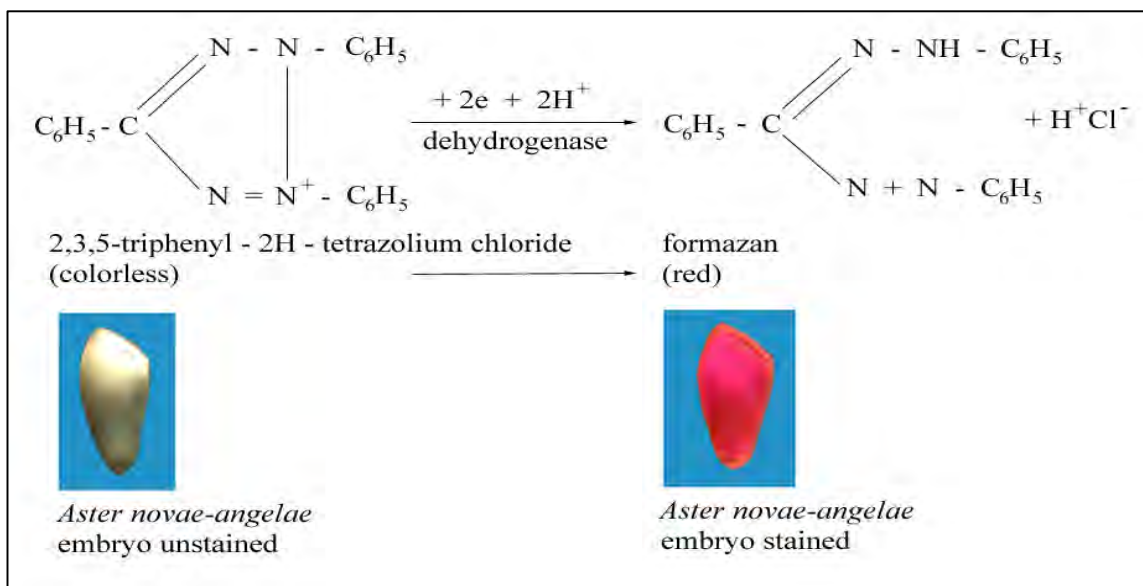
Seed testing of native species is a relatively new science and growing technology. The understanding of the diversity of seed and their many complex embryonic types, growth requirements, and dormancy mechanisms has driven the need for a quick biochemical assessment of the number of viable seed that is independent of the germination test.

Germination tests have the inherent problems of being long in duration and subjecting seed to an unnatural environment and various stresses. The duration of these stresses causes some of the weaker seed to be classified as nonviable because these seed die or become colonized with fungi during the test period. These are variables that cannot be controlled in a germination test, and thus the overall value of the seed lot may be determined to be less based upon the results of the germination test.

The tetrazolium (TZ) test is one of the tools used to determine seed value. Tetrazolium testing has been well documented for determining the potential number of viable seed (seed that theoretically can produce a normal seedling under appropriate conditions) at the point in time the test is performed. Tetrazolium testing does not give a “germination” score for a given lot of seed because “germination” is also only snapshot of the number of seed ready to sprout under the conditions that are provided in the field or laboratory. With most native species, the desire is to learn how many viable seed there are in a given quantity of seed. The relative speed of germination and the diverse environmental influences imposed upon those seed all will dictate the establishment and survivability of the species. Since TZ testing can be completed in 24 to 48 hours, seed are not subjected to long periods of stress and fungi development is not a problem. In addition, commercial labs can market the TZ test as viability test that can completed in a much shorter time than a germination test.

Tetrazolium is the common name for 2,3,5-triphenyl tetrazolium chloride (TTC), a colorless chemical when in solution. Respiring seed cells release hydrogen ions as a result of dehydrogenase enzymatic activity, with the hydrogen ions reacting with TZ to form an insoluble red compound called formazan (Figure 3-8). Thus formazan stains living cells pink to red resulting in a pattern of living and dead tissues. The analyst evaluates the stained and unstained embryonic tissues to determine if all the necessary parts of the embryo are alive and can germinate into a normal seedling. Provided the seed analyst is familiar with the topographical staining patterns of the various species, the use of TZ tests to determine the value of a given lot of seed is efficient and accurate.

Assessing native seed viability by TZ testing has the advantage of not depending on determining suitable germination testing conditions (light, temperature, and chemicals to break dormancy or stimulate germination) to evaluate whether seed are viable. Protocols that maximize germination can vary among seed lots of species because dormancy can vary (see *Germination, Dormancy-Breaking, Reporting Seed Dormancy*). Moreover, optimal germination protocols do not exist for many native species. Hence, the TZ test provides a method of determining seed viability independent of dormancy, and without the expense of determining optimum germination conditions.

Figure 3-8. The tetrazolium staining reaction.

The procedure for conducting a TZ test is to obtain pure seed from the completed purity sample or by conducting a purity test on one fourth the normal purity sample size. The seed are then hydrated to trigger the metabolic activity. Hydration, which may need to be done overnight, also softens the seed and allows the analyst to prepare the seed for staining by cutting through the embryo, clipping the seed, or piercing the seed (Figure 3-9). Some seed are put directly into the TZ solution without further preparation. The method used depends upon the species, the position of the embryo, and the size of the seed. After cutting the seed, it is placed into the TZ solution. The solution strength ranges from 0.1 to 1% depending upon the method of seed preparation. The time allowed for staining is temperature dependent and will be accelerated with every 5°F rise in temperature of the solution, up to a maximum of 113°F (45°C). Some species can withstand high temperatures while others are more sensitive to temperature, and temperature should not be raised above 104°F (40°C). Staining of the seed requires one to nine hours.

Figure 3-9. Bisecting a "seed" of big bluestem (*Andropogon gerardii*) to prepare it for TZ staining.

A trained, experienced analyst examines the staining patterns on the embryo of each seed under magnification (Figure 3-10). The analyst looks for nonstained areas, staining intensity, abrupt changes in the staining pattern, dark purplish stained areas (indicating bruising, heating, frost or microbial infestation), and also visible indicators of viability such as bisected embryo tissues curling outward.

Figure 3-10. Seed units of buffalograss (*Bouteloua dactyloides*) are cut laterally to evaluate TZ staining.

Dormancy-Breaking

TIMOTHY J. GUTORMSON

In a harvested native species seed population, seed dormancy is common, desirable, and has evolved as a species survival mechanism. Primary dormancy commonly exists at seed maturation, and duration of dormancy varies by species, production environment, and seed storage environment. After-ripening or loss of primary dormancy can start immediately after harvest or may take years in many families, such as the Poaceae (grasses). *Panicum virgatum* commonly can have 50% dormant seed within the seed population immediately after harvest; however, after-ripening can reduce dormant seed levels by 30 to 40% within six months. Other species such as *Nassella viridula* or *Orzya hymenoides* can have over 50% of the seed population dormant for years, requiring special planting (dormant seedlings) to produce successful field stands.

Dormancy mechanisms include seed coat (exogenous or externally imposed) and embryo (endogenous or internally imposed) inhibitors. The seed coat or seed membrane are physical inhibitors of water uptake or gas (O₂ and CO₂) exchange. Some species may have rudimentary embryos requiring special conditions, such as stratification for embryo maturation of ginseng. Considering all those interrelated factors it is clear that seed dormancy is more than a simple mechanism, with some species having multiple dormancy mechanisms influencing the seed population. Dormancy-breaking methods used within seed testing laboratories may not address all of the dormancy mechanisms present and can stress the nondormant and/or the deeply dormant portions of the total seed population.

With the increasing number of new native species entering commercial seed channels, and the lack of basic research to determine a species dormancy mechanism, seed analysts are beginning to question the use of dormancy-breaking methods within the laboratory. However, seed dormancy-breaking has been a common part of seed testing for over 50 years and existed well before the advent of TZ testing. Hence, seed analysts are often reluctant to rely on TZ testing to determine dormancy of seed populations when dormancy-breaking methods are provided for a specific species. Some of the common dormancy-breaking methods used in laboratories are outlined below.

Prechilling – A period of low temperature treatment with the seed in a moist environment, e.g., in between moistened rolled towels or on top of moistened blotters. The temperature may range from 37.4 to 50°F (3 to 10°C). The prechilling period can range from 24 hours to six months depending upon the species.

Alternation of temperature – The germination temperature is set to change from a low temperature period of 16 hours up to a high temperature period of eight hours. Typical alternating temperatures in northern hemisphere seed laboratories are 68 to 86° F (20 to 30°C) and 59 to 86°F (15 to 30°C). Other combinations of temperatures used in laboratories depend upon the species that they test. Normally, during the high temperature period the lights will be on in the germinator. Variations in the temperature and light regime can facilitate the breaking of the dormancy.

Light – The use of high intensity light to stimulate germination is common. Light exposure assists in converting the red-absorbing phytochrome, which inhibits germination, into the far red-absorbing phytochrome that promotes germination.

Chemicals – KNO_3 , GA_3 , and ethephon commonly are used in the water that is applied to the germination media to aid in reducing dormancy.

Reporting Seed Dormancy

Reporting of seed dormancy varies greatly among seed testing laboratories and may be a root cause for differences among seed analysis reports of the same seed lot. Reasons for differences may exist in the traditions of seed testing, depth of dormancy within a species, and lack of understanding of the importance of truthfully conveying the dormancy status of the seed lot. Traditionally, prior to the advent of TZ testing in the 1960s, seed testing laboratories used dormancy-breaking methods to promote germination since no objective method existed to determine if an ungerminated seed was dormant or dead. Remaining ungerminated seed on seed blotters that are flaccid are normally nonviable; however, firm seed may be either dormant or dead. Using dormancy-breaking techniques reduced the number of remaining firm ungerminated seed thus making dormancy determination less of an issue. However, some species with deep dormancy do not respond to dormancy-breaking methods, and laboratories often extend testing until seed decay or germinate as a method to determine dormancy status. This is not necessarily an accurate viability testing method.

In the last 20 years, the length of germination tests in some species has been reduced and the reliance on TZ staining of ungerminated firm seed has been accepted for dormancy determination. However, many laboratories are just now rethinking about their reporting accuracy. If paired testing is conducted (with and without dormancy-breaking), the true dormancy status of the seed lot can be reported; however, paired testing increases test costs, requires more lab space and analyst time, and thus is not a popular practice. So when looking at a seed analysis report a concerned party may not be able to determine the true dormancy status of the seed lot in question. If the laboratory reports its testing methods, days tested, temperatures and dormancy-breaking methods, an interested party may be able to determine if dormancy was broken in the seed lot.

Genetic Purity

KALYN BRIX-DAVIS

Genetic Variation

Evidence of genetic variation within a species has been observed for over 200 years (Kerner von Marilaun, 1902., Langlot, 1971) and clearly long before that as evidenced by selective breeding of crop species over millennia by human societies around the globe. Most wide-ranging species are a series of subpopulations, each with their own combination of characteristics (Curtis, 1959). These differing characteristics may be morphological (i.e., form) and/or physiological. Variation of form within a species includes such characteristics as leaf size, height, and flower color. Physiological differences include timing of growth and reproduction, resistance to disease, and breeding mechanisms.

Traditional plant breeding for natural resource conservation purposes was first developed by federal nurseries (USDA-NRCS Plant Material Centers) as a response to the need for re-vegetating plowed rangeland during the Dust Bowl era, beginning in the 1930's. The immediate focus was to reduce soil loss and degradation. Traits such as rapid germination and vigorous growth, and often a concomitant increase in seed size and seed production, were desired to aid in rapid establishment and to improve forage characteristics for livestock. Traditionally, seed or plants were collected from native stands, established in common gardens at the nursery, and evaluated for specific traits. These cultivated varieties, or cultivars, were then selectively bred and propagated based on performance, including establishment, forage production, winter hardiness, height, and vigor (Alderson and Sharp, 1994; Jacobson, Tober, Haas, and Darris 1986). Some varieties developed in this manner may even represent an extreme phenotype of the species (Olson, 1986). Gustafson (1997) reported that the cultivars of big bluestem, namely 'Kaw' (Kansas), 'Rountree' (Iowa), and 'Pawnee' (Nebraska) were genetically more similar to one another than any were to wild populations, presumably due to selective breeding for similar traits. Although original parent stock is collected from naturally occurring populations, the potential exists for genotypes from a single source population to be planted over a large geographical area (Knapp and Rice, 1996). In the case of 'Blackwell' switchgrass, a single plant was the progenitor of the variety (Alderson and Sharp, 1994).

A more recent approach in USDA-NRCS cultivar development has been to collect several populations over a broad region and pool them (or a subset of these populations) for evaluation and increase. This is in part a response to greater interest in and demand for native plant materials for ecosystem restoration. This approach is similar to other efforts that use provenance zones to define regions for collecting and bulking populations as foundation seed for restoration (Houseal and Smith, 2000; Knapp and Rice, 1996). Evaluation and intentional selection of some populations over others because of measurable traits may still be utilized. The 'Badlands' ecotype of little bluestem, for example, is a composite of 68 accessions selected from an initial evaluation of 588 accessions collected vegetatively from throughout North and South Dakota and Minnesota. Accessions were selected for resistance to a leaf spot disease which was affecting the nursery planting site at Mandan, North Dakota.

Verifying the identity of native plant cultivars has been questioned and needs to be confirmed to ensure that these native cultivars are marketed and used appropriately. With cultivars being developed and released and genetic information being used for their identification, the need for laboratory identification has increased.

Laboratory Methods for Genetic Identification

Determining genetic identification of native species is challenging. Seed characteristics vary greatly within a species due to seed maturation environment, harvesting conditions, and post harvest processing. Some cultivars can be identified by a seedling grow-out, such as 'Blaze' and 'Camper' little bluestem. This seedling grow-out is most definitive when little bluestem is near maturity.

Protein characterization of native seed species is a testing method used to differentiate cultivars. Proteins have a net charge (amphoteric), either positive or negative, and will move through a gel when an electrical current is applied. Isoelectric focusing (IEF) electrophoresis separates seed proteins by their pI value due to a pH gradient that is created in the gel prior to the proteins being separated (Figure 3-11). The pI is the isoelectric point, the pH at which the protein does not carry a charge, that is, $pI = 0$)

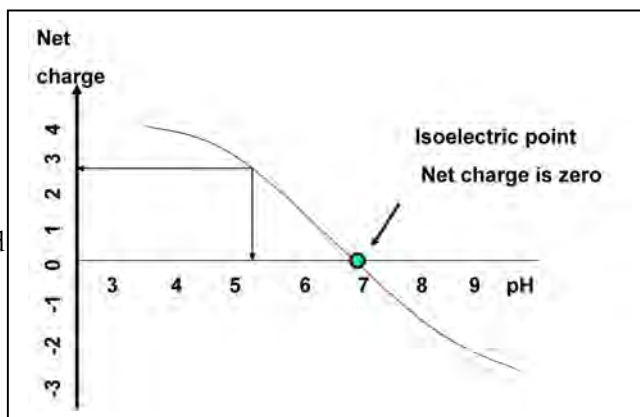


Figure 3-11. Diagram of isoelectric focusing with pH on the x-axis and net charge on the y-axis.

Seed storage proteins are extracted in an alcohol based buffer whereas seed enzymes are extracted with a phosphate or glycine solution. Proteins or enzymes extracted from the seed are placed on a gel that has a pH gradient across the gel (Figure 3-12). Since amino acids in the proteins have amphoteric properties, negatively charged ampholytes move towards the anode while positively charged ampholytes move toward the cathode within an electrical field. After the proteins have moved to their pI point (approximately 60 to 80 min), the gel is stained to visualize the proteins or enzymes (Figure 3-13) and then dried. Either a total protein stain can be used or different enzymes may be stained to detect polymorphism for a particular species. After the gel dries, the stain and banding pattern is stable and can be retained for documentation.



Figure 3-12. IEF gel being loaded with protein extracted from seeds.

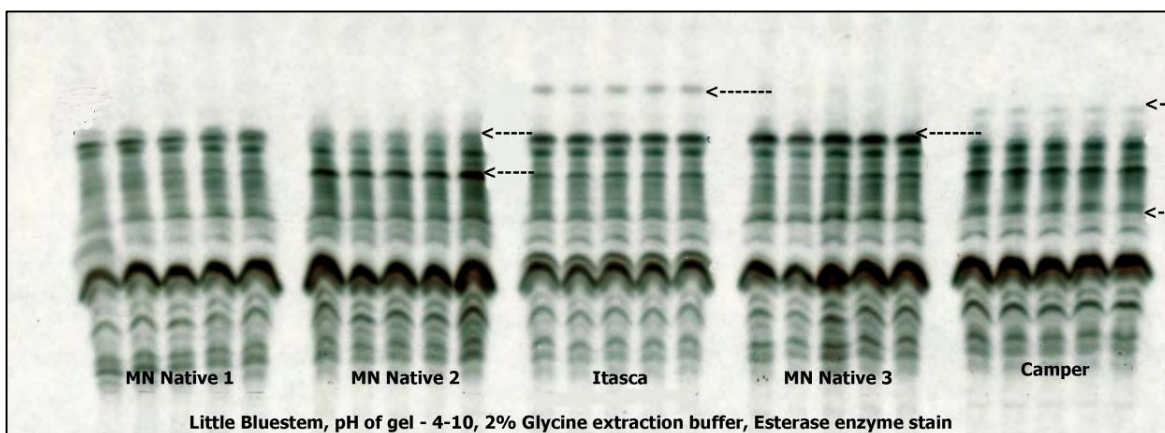
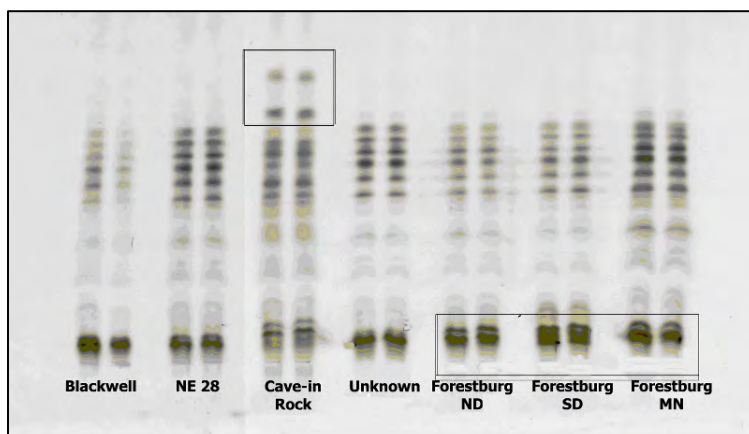


Figure 3-13. Example of little bluestem (*Schizachyrium scoparium*) cultivars and differences seen on an IEF gel. Arrows denote differences in banding patterns.

Different ranges of pH gradient, extraction solutions, and enzymes can be used to detect cultivar differences. Proteins extracted from foundation seed lots of specific cultivars are used to compare to unknown samples for cultivar classification. Band darkness is influenced by the number of bands in one area and/or amount of protein present, either in the seed storage or structural proteins.

With different native species, cultivars exhibit different proteins and are able to be differentiated (Figure 3-14). ‘Cave-in-Rock’ switchgrass foundation seed has some distinct bands on the upper portion of the gel that differentiates this cultivar from other switchgrass cultivars. ‘Forestburg’ switchgrass foundation seed from North Dakota, South Dakota and Nebraska are similar in banding patterns but on the lower portion of the gel slight differentiation occurs between

Figure 3-14. Example of switchgrass foundation seed samples and unknown.



foundation seed locations. This protein shift may be due to the environment/location in which the seed has been growing, thereby slightly changing the protein profile. The pattern unknown of the sample in this example matched the NE28 foundation seed stock pattern.

For IEF analysis, a representative sample (50 to 100 g) should be sent to the lab with information regarding the species and possible cultivars that you wish for the sample to be compared with. Approximately 30 seed are ground to present a “population” protein representation for the sample. The IEF analysis can be completed in two days. The report will compare the banding pattern of the sample to the banding patterns of the foundation seed lots that were run on the same gel as the sample.

Genetic variation occurs in native species; isoelectric focusing is a laboratory tool that may assist in cultivar identification. As more native species are characterized with IEF, information regarding cultivars will improve and cultivar identification may be more technical than subjective.

Seed Analysis Report

TIMOTHY J. GUTORMSON


The items listed below should be highlighted in a Seed Analysis Report. An example of a report is below (Figure 3-15).

1. Dates – received, complete, date of report and days tested
2. Sender information – customer provides genus and species
3. Viability
 - a. Germination – percent nondormant seed (note germination methods)
 - b. Dormant – percent dormant seed within the seed population
 - c. Total Viable seed – percent germination + percent dormant
 - d. TZ – tetrazolium test; estimate of maximum percentage of viable seed
4. Pure seed – percentage of pure seed units
5. Other crop and weed – number found and genus and/or species
6. Dormancy-breaking methods – list method or state “none”

Figure 3-15. Example of a seed analysis report.

Native Seed Testing

110 Seed Way
Germination, SD 57777
www.nativegermination.com

REPORT OF SEED ANALYSIS											
Names and Addresses		Date Received 1/1/2007	Date Completed 1/15/2007	Date of Report 1/15/2007	Test No. 10995						
SEED PRODUCER 53535 343RD AVE. BROOKINGS, SD 57006 Acct. Num: 0 MS		SENDER'S INFORMATION * Kind/Brand Name: SWITCHGRASS Variety: NEBRASKA 28 Genus/Species: PANICUM VIRGATUM Lot Number: LOT 1									
*The information provided here is that of the sender and not of the laboratory.											
Varietal purity guaranteed by labeler. The analysis report shown below is accurate only for the sample received at the laboratory. Whoever makes use of this information for labeling purposes is guaranteeing that the sample is representative of the seed lot from which it was drawn. The characters "--" mean the test is not complete or not reported and the letter 'N' means the test was not requested.											
Purity Analysis					Viability Analysis						
(4.214 Grams Analyzed)		Germination %	abn%	Dormant	Hard Seed %	Total Viable %	Number of Seeds	Days Tested	TFL %	TZ %	
Pure Seed Component(s):		47	2	40	--	87	400	14	N	97	
SWITCHGRASS (<i>PANICUM VIRGATUM</i>) 97.22%		Comments: PURE LIVE SEED: 84.58%									
OTHER CROP SEED: 0.12%											
INERT MATTER 2.59%											
WEED SEED 0.07%											
OTHER CROP SEED: 5 CLOVER-PURPLE PRAIRIE (<i>DALEA PURPUREA</i>) 538/lb		A 41.20 g sample was examined for weed seed classified as noxious in the 48 contiguous states (except for Undesirable Grass Seeds)									
INERT MATTER: No Soil Peds Found. No Sclerotia/Ergot Bodies Found. Chaff and Stems Found.		(1) Noxious weed seed species found. Number of species found must equal number of species listed below for an authentic report.									
WEED SEED: 1 GREEN FOXTAIL (<i>SETARIA VIRIDIS</i>) 108/lb		23		GIANT FOXTAIL (<i>SETARIA FABERI</i>)					253/lb		
TEST CODE AND FEES: Fee: --		OTHER DETERMINATIONS:									
Germination, Purity, TZ, USA Noxious											
TO: Fax:		<div style="display: flex; justify-content: space-around; align-items: center;"> <div style="text-align: center;"> <p><i>Signature</i> <i>Here</i></p> </div> <div style="text-align: center;">  </div> </div>									
CC: Germination, Purity and Noxious Weed Examination tested in accordance with AOSA Rules. DORMANCY BREAKING METHOD: NONE. GERM METHOD: 15-30°C, BLOTTER, 7d, 14d Ungerminated pure seed units were evaluated for dormancy by Tetrazolium staining.		COMMENTS:									

Seed Package Labeling

DAVID BUCKINGHAM

The seed analysis label or seed tag provides information that quantifies and describes required labeling components. The information present on the seed analysis label is used to establish the value of the seed lot. A standard approach to labeling provides a consistent approach to determining the value of the seed lot.

The definition of terms used for labeling components required by state and federal laws are familiar to everyone involved in the commerce of traditional seed kinds. Additional terms used to quantify or describe seed of native species, including pure live seed (See Glossary p. A-1), dormant seed, and TZ, are not defined or used in state and federal law in a context that relates to labeling. However, these terms are commonly used, singly or in combination, in the commerce of native species. Incorporating these terms on the seed analysis label of native seed kinds will provide a consistent approach to labeling.

Suggested labeling formats for both single seed kinds and mixtures of native species that incorporates additional components are illustrated. Definitions of the additional component terms also are addressed to assure a consistent approach to their use within the context of labeling.

Figure 3-16. Suggested labeling format for native seed sold as single seed kinds that have AOSA approved or suggested germination and dormancy testing procedures.

Variety Kind	Bulk lbs.	Lot No.	Pure Seed %	Inert %	Crop Seed %	Weed Seed %	Germ. %	Hard/Dorm. Seed %	Total Germ. + Hard/Dorm. Seed %	Pure Live Seed lbs.	Origin	Test Date
Chieftain Indiangrass	32.04	IG06	89.70	10.30	0.00	0.00	35	52	87	25	KY	12-09

Noxious Weeds-*Name and number per pound of each*

NET WEIGHT 32.04 Lbs

Distributed by
 IMA SEEDSMAN
 418 Seedy Lane
 Anywhere, USA

Figure 3-17. Suggested labeling format for mixtures of native seeds that have AOSA approved or suggested germination and dormancy testing procedures.

Quail Farm Native Mixture
Lot QF123

Variety Kind	Bulk lbs.	Pure Seed % *	Seed Lot Used in Mix	Component Pure Seed % **	Inert %	Crop Seed %	Weed Seed %	Germ. %	Hard/Dorm. Seed %	Total Germ. + Hard/Dorm. Seed %	Pure Live Seed lbs.	Origin
Chieftain Indiangrass	32.04	18.98	IG1	89.70	10.30	0.00	0.00	35	52	87	25	KY
Roy Big Bluestem	27.53	17.02	BB06	93.62	6.06	0.05	0.27	13	84	97	25	MN
VNS Little Bluestem	28.49	17.02	LB01	90.45	9.42	0.13	0.00	22	75	97	25	WI
Lark Partridge Pea	23.16	15.29	PP06	99.95	0.03	0.02	0.00	42	38	80	18.52	AL
Black-eyed Susan	3.60	2.37	BS33	99.74	0.26	0.00	0.00	87	0	87	3.12	OR
Illinois Bundleflower	10.17	6.60	IB24	98.39	0.33	0.00	1.28	94	0	94	9.41	OK
Purple Coneflower	26.42	13.18	PC71	75.51	24.47	0.00	0.02	94	0	94	18.75	CO

Noxious Weeds - *Name and number per pound of each*

Inert Matter - 9.36%*

Crop Seed - 0.04%*

Weed Seed - 0.14%*

Test Date: 6/05***

NET WEIGHT 151.41 Lbs

Distributed by
IMA SEEDSMAN
418 SEEDY LANE
ANYWHERE, USA

*Please note that these components must be calculated from the contents of each individual seed lot used to make the mixture.

**Component pure seed percentage is the labeled pure seed percentage of each ingredient prior to addition to the mixture. It is necessary to include this value to calculate the PLS of each labeled seed kind.

***For mixtures, the earliest test date for seed lot components shall be used.

These examples represent typical labeling for single seed kinds and mixtures that either have approved testing methods or suggested methods for determining germination and dormancy. The example for the mixture label demonstrates the complexity of providing all of the necessary information. The lot number column could be eliminated as it is not used for calculation purposes. Pure live seed is a mathematical function of the pure seed, the total of the germination plus hard/dormant seed, and the bulk weight of the seed kind component. Testing methods used to determine the germination and dormant seed for those seed kinds that do not have approved methods should be stated on the seed analysis report to assure consistency of analysis among testing laboratories.

There are some native seed kinds in commerce that do not fit into the labeling scheme presented in the first two examples. Those seed kinds do not lend themselves to timely tests for germination and dormant seed or they have not been evaluated by enough testing laboratories to arrive at a suggested or approved testing method. For these seed kinds a TZ test is the most commonly used approach to quantify potential seed emergence. It is important to note that the examples that follow do not have columns for germination, dormant seed or pure live seed. As stated above, the pure live seed figure is a function of the germination plus hard/dormant seed. Since the TZ test is a measure of respiration in the essential embryonic seed structures, TZ test results should not be stated as germination when labeling seed lots.

Figure 3-18. Suggested labeling format for native seed sold as single seed kinds that do not have AOSA approved or suggested germination and dormancy testing procedures.

Variety Kind	Bulk lbs.	Lot No.	Pure Seed %	Inert Matter %	Crop Seed %	Weed Seed %	Origin	Test Date	TZ %

Noxious Weeds-Name and number per pound of each

NET WEIGHT _____

Distributed by
 IMA SEEDSMAN
 418 SEEDY LANE
 ANYWHERE, USA

Figure 3-19. Suggested labeling format for mixtures of native seeds that do not have AOSA approved or suggested germination and dormancy testing procedures.

Native Seed Mixture
Lot NSM123

Kind Variety	Bulk lbs.	Component Lot No.	Pure Seed %	Inert Matter %	Crop Seed %	Weed Seed %	Origin	TZ %

Noxious Weeds-Name and number per pound of each

NET WEIGHT _____

% Inert Matter-*

Distributed by:

% Crop Seed-*

IMA SEEDSMAN

% Weed Seed- *

418 SEEDY LANE

Test Date**

ANYWHERE, USA

**Please note that these components must be calculated from the contents of each individual seed lot that is used to make the mixture.*

***For mixtures, the earliest seed lot component test date shall be used.*

Summary

The seed analysis label is used to quantify components present in a seed lot. Native plant species do not conform well to labeling schemes typically used for traditional seed because additional information is used in the commerce of native species. State seed laws vary, so shippers should check with individual states to verify that the labeling formats suggested will meet their respective labeling requirements. Much of the information presented in these labeling formats is extra or supplemental label information. In the context of both the Federal Seed Act that governs interstate commerce and state seed laws that govern intrastate commerce, additional information can appear on a label, but must be truthful. This attempt to identify labeling formats for native plant species is challenging because of differences that exist in seed laws. It is hoped that the suggested formats will provide a basic labeling pattern that, if needed, can be adjusted to comply with the majority of labeling situations.

For more information:

Federal Seed Act – <http://www.ams.usda.gov/AMSV1.0/getfile?dDocName=STELPRD3317283>. (Accessed 12 November 2010.)

Federal Seed Act Regulations – http://ecfr.gpoaccess.gov/cgi/t/text/text-idx?c=ecfr&tpl=/ecfrbrowse/Title07/7cfr201_main_02.tpl. (Accessed 12 November 2010.)

Association of American Seed Control Officials Recommended Uniform State Seed Law (RUSSL) – <http://www.seedcontrol.org/pdf/russl.pdf>. (Accessed 12 November 2010.)

4

Seed Certification

Overview

STANFORD YOUNG

How can you be assured that the seed in the bag is the variety or germplasm as claimed? This assurance is necessary for successful restoration and reclamation projects, as the genetic capabilities of the seed must be adapted to the environmental conditions of the planting site. The answer is to use certified seed. The third-party seed certification process is based on verification of planting stock germplasm identity. Strict requirements for field/site inspections, harvesting, seed conditioning, and seed sampling and analysis are then followed to assure that high standards for germplasm purity and seed quality are met.

Information on an analysis label (as covered elsewhere in this bulletin) is usually based on a seed analysis report from an accredited seed laboratory. Such reports, however, do not normally verify that the identity of a certain variety or germplasm, as listed by the sender of a seed sample, is correct as claimed. Thus there is no easy way to confirm that a bag of Indian ricegrass seed is actually ‘Nezpar’ or ‘Paloma’ or ‘Rimrock’, or from a specific wildland collection site. Likewise, seed laboratories normally do not distinguish between subspecies such as Wyoming, mountain, or basin big sagebrush, or even between species of many wildland plants such as the penstemons or globemallows. Seed certification is the most efficient and cost effective way to verify this information.

For released varieties, the most common official certification tag encountered will be a light blue tag with “Certified Class Seed” printed on the top of the tag. Other variety tags, intended for stock seed use, are white “Foundation Class” and purple “Registered Class”. Variety tags list the crop or species, the specific variety, and certification and/or lot numbers as appropriate.



For Pre-Variety Germplasm (PVG) of field produced or wildland collected native or naturalized species, the most common certification tag encountered will be a yellow tag with “Source Identified Seed” printed on the top of the tag; in this case the geographic site of collection is the only information known. Other pre-variety tags, denoting more advanced stages of germplasm study and development, are green “Selected Class” and dark blue “Tested Class”. PVG tags list common and scientific species name, generation, and site (state, county and elevation) where the germplasm was collected, developed and/or grown. Other pertinent information can include whether a germplasm is indigenous (if known) to the site where it was collected.



PVG entities can also be identified on the certification tag as to their genetic status, e.g., natural track or manipulated track. Natural track means that the germplasm originated as an unrestricted representation of the intact wildland plant population on the original site, and that care has been taken to avoid contamination from other populations of the same species during field increase. On the other hand, germplasm that is purposefully or inadvertently hybridized with other populations or has been selected to concentrate distinctive traits within the population (whether on the original site or in succeeding field or nursery generations) is routed to the manipulated track.

Germplasm can be developed utilizing either track, leading to natural or manipulated track releases of Selected Germplasms, Tested Germplasms, or Varieties. All of these materials are appropriate for end use according to objectives for the planting site.

Decisions as to how much germplasm “development” is desired can depend on the size, location, and ecological status of the geographic area to be treated, and the amount of lead time available for genetic studies (to quantify traits such as seed production, disease resistance, or seed zone adaptation) and the associated seed market potential. As a rule of thumb seed availability is lower and prices higher for restoration projects specifying “local” geographic services of seed; larger reclamation projects such as fire rehabilitation often utilize widely adapted varieties that are usually more available and cheaper.

The “AOSCA Native Plant Connection” bulletin gives more details of the certification program for reclamation plants and is available on the AOSCA website (<http://www.aosca.org>). Click on “Programs/Services” and then “Native Plant Connection”.

AOSCA Process for Certified Production of Native Plant Seeds

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The Association of Official Seed Certifying Agencies (AOSCA) has implemented certification requirements and standards that accommodate plant germplasm of native grasses, forbs and woody plants. AOSCA seed certification agencies (acting as a third party) require seed collectors/producers to follow established requirements, procedures and standards to assure seed native plant restoration, revegetation and stabilization identity and purity for the consumer.

Long-term success in restoring a species to a given site is dependent upon obtaining adapted plant materials. Adapted plant materials are most likely to originate from the same site or nearby sites with similar physical and biological environments. AOSCA has implemented certification requirements and standards that accommodate plant germplasm of native grasses, forbs, and woody plants.

Certification Procedure

AOSCA seed certification agencies (acting as a third-party) require seed collectors / producers to follow established requirements, procedures, and standards to assure germplasm identity and purity for the seed consumer. Stock seed or plants for establishing certified field/nursery production must be of an eligible generation and have appropriate labeling. Certification procedures for field/nursery production include:

- Application for Certification
- Verification of origin and generation of planting stock
- Seedling inspection
- Field inspection before harvest to check compliance with species requirements for isolation and genetic purity (control of prohibited and other specified weeds or other species may be required)
- Tagging of the seed lot after compliance with applicable requirements and standards; seed purity and viability analysis may be required.

Wildland collected seed can be used for direct sales to end users, for establishment of field/nursery production, or for entry into plant germplasm development programs. Certification procedures wildland collected seed include:

- Pre-Collection Application filed before harvest
- Proper permitting and/or permission for collecting on public and private lands
- Site Identification Log filled out during and after harvest
- Verification of the collection site and identification and evaluation of plant and seed samples before, during, and/or post harvest
- Tagging of the seed lot after compliance with applicable requirements and standards; seed purity and viability analysis may be required
- Germplasm accessions acquired within established protocols of recognized public or private agencies are normally eligible to enter the certification process as planting stock.

Contact the official seed certifying agency in your country, state or province for further information, application forms, or help in following wildland plant materials certification procedures. A list of certifying agencies is available from the AOSCA Office:

Association of Official Seed Certifying Agencies
1601 52nd Ave., Suite 1
Moline, IL 61265
309-736-0120
Website: <http://www.aosca.org>

5

Production and Marketing Expectations

Overview

GARTH KASTE

A critical issue of late for the native seed industry has been the continued controversy over native seed testing methods. Far too often, significant differences in testing methods have led to conflicts between producers, marketers, and end users. It appears that the use of native seed will continue to increase, so this problem has the potential to increase as well. Therefore, continued research, communication, and compromise by all industry stakeholders are needed to reduce the controversies that have existed. Consumers and regulators need to be aware that:

- Testing differences directly affect value. Since native seed is marketed on a pure live seed per pound basis, testing differences have a greater influence on value than for seed traded on a bulk per pound basis.
- Seed producers and marketers desire consistent results, within recognized tolerances, that represent the maximum potential of their seed. In addition, they desire prompt service to satisfy today's "just in time" customer.
- Seed producers and marketers are not seed scientists; therefore they should not be expected to make judgments on technical seed testing methods between laboratories. This is a job for seed analysts and researchers.

The native seed industry needs to:

- Define "high quality" native seed and educate consumers about this definition.
- Fund research projects to improve testing methods.
- Work with regulators to eliminate fraud.

6

Consumer Expectations and Requirements

Natural Resources Conservation Service and Farm Service Agency

RICHARD L. WYNIA

From correspondence originating from the Natural Resources Conservation Service (NRCS), Farm Service Agency (FSA), and agencies of US Department of Agriculture, it was stated that their main requirement was that there is a germination percentage based on the germination test and that germination plus hard seed numbers be expressed. A test of seed purity is also required in order to calculate the seed lots Pure Live Seed (PLS) percentage. No noxious weed seed is tolerated. Tetrazolium (TZ) tests are only allowed on certain recalcitrant species such as green needlegrass and western wheatgrass in the state of South Dakota. Different states allow for the use of seed based on TZ tests, with Nebraska allowing some 15 native species to be handled in this manner. A check with the local offices of NRCS or FSA should identify if your state allows the TZ test to be used to determine germination percentage. FSA is satisfied with the current seed tag as long as purity and germination numbers are spelled out, and information is provided to complete agency paperwork. Neither FSA nor NRCS could recommend additional data or information on the seed tag that would be useful for them. They also had no requirements or expectations that are not being fully met by information that is currently listed on seed tags. Further clarification and information may be located at the following web site: <http://www.fsa.usda.gov>.

Bureau of Land Management

SCOTT M. LAMBERT

The Bureau of Land Management (BLM) has set a minimum purity, germination (or TZ), and PLS for most of the species or types of seed we utilize in our seed projects on the western US public lands. The minimum PLS usually is based on the standards for the seed types as set by AOSCA. For example, bluebunch wheatgrass minimums are 0.90 purity, 0.85 germination (or TZ), and 0.8075 PLS.

Prior to seed procurement, BLM must have the seed lot sampled upon arrival at our facility by our trained staff or an official seed sampler, who then sends the seed sample to an official seed testing laboratory for analysis by a certified seed technologist (AOSA) or registered seed technologist (SCST), as approved by BLM. Most often when obtaining a seed lab analysis, BLM will request a TZ for seed viability instead of germination. A TZ test is preferred due to the shorter time needed to complete it, and also dormancy issues for many native forb and shrub lead to some problems with germination tests.

Per our multiple seed contracts, BLM does not accept any seed lot with prohibited noxious weed seed per State Law(s) and the Federal Seed Act. Restricted noxious weed seed amounts must also comply with the State Law(s) in the respective states. The seed lab analysis will show no more than 0.5% by weight of “other weed seeds”; the seed may contain up to 2.0% of “other crop seeds”.

Labeling / Seed Certification

Every bag of seed BLM obtains must have a vendor’s seed tag and a certification tag (if seed is Certified). The vendor’s tag must include lot number, the vendor’s name, address of shipper, species/cultivar, purity, germination/TZ, crop seed, weed seed, inert, noxious weed seed and states tested for, origin, test date, bag weight, referenced Certified lot number as needed, and the BLM Contract number.

In the past four years, over 75% of the seed purchased on the commercial market for BLM has been blue-tag certified on cultivars or yellow-tag source identified on native seed. BLM’s 10-year average amount of conservation and native seed procured per year is more than 2.5 million pounds.

BLM only procures single species or cultivars. No seed mixes are purchased; BLM custom mixes seed as necessary to suit project needs.

State Departments of Transportation

JOY WILLIAMS

State Departments of Transportation (DOTs) purchase native seed in different ways. Most DOTs specify quality standards, and the seed is then purchased on the market by seeding contractors, while other DOTs purchase native seed directly from seed vendors and provide the seed to the seeding contractors. Some DOTs preapprove seed vendors to provide specified mixes. In all systems, the analyses that determine PLS are extremely important. Pure live seed provides a basis to compare value of seed lots and is often used as the primary unit of commerce. It is also used to estimate potential performance and calibrate seeding rates. And purity analyses are critical for avoiding weed introductions.

The Iowa DOT purchases large quantities of native grass and wildflower seed through a competitive bid process administered by the department. Certified source-identified seed (generation 0 [G0]) is specified, but other seed is accepted when source-identified G0 seed is unavailable. Each species is a separate bid item, so it is possible that seed in a final mix came from numerous vendors. Bid awards are strictly by price per pound PLS (within source G0 selection groups.) There is an expectation that the analyses that determine PLS are uniform – i.e., seed tested at one lab would give the same result as the same seed tested at another lab and one vendor’s seed can be compared against another through PLS. While differences in the testing methods may be explained in the Seed Analysis Report, it would be difficult to weigh the differences in a competitive bid system.

One commonality among all state DOTs is an interest in seed purity. Highways have extensive borders with adjacent property owners, and highways are corridors in which invasive weeds can spread. Weed contaminants in seed, or even a perception by adjacent property owners

of weed contaminants in seed, can cause big problems. This includes weed species not restricted by state seed laws. Iowa DOT specifications are more stringent than state seed law, requiring native forb seed to be tested and labeled the same as agricultural seed, with corresponding tolerances for noxious weed seed. But that does not encompass all the weeds of concern. For this reason more than any, DOTs need the eyes and expertise of seed analysts and the documentation they provide in easy-to-read analysis reports.

Quality Standards in Canada

GARTH WRUCK AND MORGAN WEBB

Native seed quality in Canada is based only on purity. It is an examination of weed, other crop and other contaminants but not a percentage by weight test as the term is used in the US. Native seed quality is governed only at the federal level by the Canadian Food Inspection Agency through the *Canada Seeds Act, Weeds Order 2005*, and there is no current provincial or municipal legislation directly governing the commodity trade of native seed or quality identification. Some provincial noxious weed acts have sections that would cover the transportation of noxious weed materials, which includes seed, but these are only enforceable by weed inspectors who mainly monitor feed and fill material for transportation. Few guidelines and recommendations exist from various government departments, conservation industries and natural resource industries that stipulate expectations for native seed quality. Most of these are extremely general and recommend the use of “Certified Seed”, which is a grade under the Canadian Methods and Procedures for Testing Seed.

Only six native grasses are designated with crop “kind” status within the Grade Tables of Canadian Methods and Procedures for Testing Seed. These six are only a small percentage of the native plant species traded in Canada and between Canada and the US.

- Northern Wheatgrass/Thickspike (*Elymus lanceolatus* ssp. *lanceolatus* var. *lanceolatus*)
- Western Wheatgrass (*Pascopyrum smithii*)
- Slender Wheatgrass (*Elymus trachycaulum*)
- Streambank Wheatgrass (*Elymus lanceolatus* ssp. *lanceolatus* var. *riparius*)
- Fowl Bluegrass (*Poa palustris*)
- Creeping Bent Grass (*Agrostis stolonifera*)

It is very difficult to identify quality by grades. Grades cannot be applied to noncrop “kinds”, and purity testing can only state that a seed lot meets the minimum purity standards of table XI, XII, XIII for grasses or XV wildflowers when it is not a crop “kind” of the Canadian Methods and Procedures for Testing Seed.

The *Canada Seeds Act and Regulations, Weeds Order 2005* and the Canadian Method and Procedures for Testing Seed do not adequately address native seed purity requirements.

- For a contaminant to be designated an “Other Crop” in Canada it must be listed in the grade tables, which is Schedule I of the Seeds Regulations. All species not listed in the tables and not listed as noxious weed are designated “Other Weed”.
- Native seed not classed as “kinds” (i.e., crop kinds) are identified as “Other Weed Seed” on the seed certificate of analysis. This can cause a seed lot to be rejected for entry/use in Canada because of a nonnoxious “contaminant” which is likely desired by the end user.
- Invasive species detrimental to native restoration, such as crested wheatgrass (*Agropyron cristatum*, *A. desertorum*) or smooth brome (*Bromus inermis* ssp. *inermis*), are identified as “other crop seeds” in native seed lot analysis reports because they are crop kinds. As such, they only may be reported on a percentage weight basis and not an actual seed count, which is misleading in regards to the contaminant’s quantity.
- Weed species like downy brome (*Bromus tectorum*) have the same weed status as noncrop kind native species, meaning it does not fall under the noxious weed category on a certificate of analysis when seed is tested using the Canadian Methods and Procedures for Testing Seed. In Canada, contaminant searches may be done using sequential analysis, which breaks down the analysis into portions. If a weed like downy brome is not found in the first quantity but found in following quantities it may not be reported due to its “Other Weed Seeds” status.
- Germination, tetrazolium, and percentage pure seed are almost always used to buy and sell native seed in Canada. However, the Canadian Government does not require these tests and therefore the Canadian Methods and Procedures for testing seed are inadequate for most native species. Many Canadian labs use AOSA and ISTA rules where rules exist.

Canada has an excellent network in seed testing. Lab and analyst accreditation is well established and the Commercial Seed Analysts Association of Canada fosters communication and education. Much work needs to be done to use this system to ensure that seed testing meets the needs of producers, processors, and consumers of native species.

The Canadian Seed Growers Association is an AOSCA (Association of Official Seed Certification Agencies) member and is the registered administrator of *The AOSCA Native Plant Connection* (native plant genetic source information certification system).

Procedures to identify and label the value of genetically diverse seed, whether it is an individual species lot or a diverse mixture of highly desirable native grasses for use in ecologically sensitive revegetation projects, remains a huge barrier to the native seed industry’s producers, suppliers and consumers. Current systems (purity analysis and source identified certification) require a high degree of interpretation by the consumer and therefore facilitate errors. The real issue with the Canadian system is that although it does not directly reject or adversely identify native seed (all native species that use to be included as Prohibited Noxious Weeds in the weeds order were removed in 2005), it does not properly represent or support native seed products thereby leaving non-crop “kinds” identified as “Other Weed Seeds” or the commodity market as a whole.

Appendix

Glossary

Dormant seed A viable seed that fails to germinate when provided the specified germination conditions for the kind of seed in question.

Fresh A term used to describe seed but whose meaning varies widely. Hence the use of this term seems to have little to no value. “Fresh Seed” generally are considered to be those that are freshly harvested, that is, **seed** that have not been stored for the winter or are less than four to six months old. After the end of the first sales season for a particular crop, seed is then considered to be carryover seed. However, according to ISTA's *International Rules for Testing Seeds* (January 2008; section 5.2.7.2 “*Fresh Seeds*”) “Fresh Seeds are able to imbibe water when provided with the conditions set out in Table 5A, but the germination process is blocked.” (*editors' note: in other words, dormant*).

Germination A process involving water uptake, metabolic changes and cell elongation resulting in radicle emergence from the seed. In seed testing, the emergence and development from the seed embryo those essential structures which, for the kind of seed in question, are indicative of its ability to produce a normal plant under favorable conditions.

Hard seed A seed that remains hard at the end of the prescribed test period because it has not absorbed water due to a water-impermeable seed coat. Hard seed are presumed to have physical dormancy, and hence are deemed viable.

PLS (pure live seed) Many native species are sold on the basis of pure live seed pounds. The pure live seed percentage is based on pure seed and germination (and in some cases, germination and dormancy) of a seed lot. In this context the term germination is actually the germination plus dormant/hard seed. The pure live seed percentage is determined by multiplying the pure seed percentage by the germination percentage (% germination + % hard/dormant) and dividing by 100 ($[97\% \times 80\%] \div 100 = 77.6\%$).

Postgermination TZ test A tetrazolium test (see **Tetrazolium**) conducted at the conclusion of the prescribed germination period to determine viability of remaining firm, ungerminated seed.

Pregermination TZ test (independent TZ test) A TZ (see **Tetrazolium**) test run concurrently (paired) with a germination test; 200 seed are tested to determine dormancy when a separate germination test is requested.

Tetrazolium A shortened term for tetrazolium chloride and often referred to as TZ. Tetrazolium is a water soluble colorless chemical used to determine viability of seed. In respiring tissues, dehydrogenase enzymes reduce TZ to form the water insoluble, reddish compound formazan. TZ also is commonly used to refer to the percentage of viable seed based on the **tetrazolium test**. TZ tests are not considered legal tests to determine germination by the Federal

Seed Act and in some states. TZ tests are generally used as indicators of potential germination for seed kinds that exhibit high dormancy.

Total germination Number of seed that have produced seedlings classified as normal under the conditions and within the period specified.

Total viable seeds Sum of germinated and dormant seed.

TZ – See Tetrazolium.

Viable A term used to describe a seed that contains structures and substances, including enzyme systems, that give it the capacity to germinate under favorable conditions in the absence of dormancy. Viability is usually determined by germination and TZ tests. In a commercial seed testing lab, a seed classified as viable by a germination test must have developed into a normal seedling within the specified time of the germination test. Abnormal seedlings are classified as **nonviable** as are seed that do not germinate and fail a postgermination TZ test.

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