

ABSTRACTS

*from oral and poster presentations
given at the 99th
Association of Official Seed Analysts
and the 86th
Society of Commercial Seed Technologists
(AOSA/SCST)
Annual Meeting
held in Ft. Collins, Colorado on
May 31st–June 5th, 2009*

— ORAL PRESENTATIONS —

Comparison of Two Accelerated Aging Test Methods

S.G. Elias¹, R. Balbaaki², M.B. McDonald³ and J.M. Filho⁴

The accelerated aging test (AAT) has been successfully used to evaluate seed vigor in a wide range of crop species for decades. Extensive studies with soybean [*Glycine max* (L.) Merr.] have standardized the laboratory variables that influence AAT. However, no published data are available to compare whether aging the seeds by placing them as one layer on the AA screen without weighing versus weighing the seeds before placing them on the screen would affect final germination results of the test. The objective of this study was to evaluate the effect of each method, i.e., aging the seeds as one layer vs. weighing seeds before the aging process has on final germination results. High (approximately 95% standard germination) and borderline quality (using the seed industry standard of approximately 85% standard germination) seed lots of soybean, sorghum (*Sorghum bicolor* L.) and tomato (*Lycopersicon esculentum* L.) were included in the study. Twelve commercial and public laboratories conducted the AAT in 2009 using both the one layer and weight methods for soybean, and six laboratories for each of the sorghum and tomato seed lots. The parameters, i.e., temperature, duration of aging, initial moisture content and seed weight listed in the AOSA Seed Vigor Testing Handbook, 1983, were followed for both methods. The only exception was placing a single layer from the crop to be tested on the surface of the AA screen in one test and weighing the seed sample to be placed in each AA box as specified in Table 2 of the AOSA Seed Vigor Testing Handbook in the other. The International Seed Testing Association tolerance Table 15.5 was employed to determine whether the AAT germination results of the two methods were within tolerance in each laboratory. No significant differences in final AAT germination results were detected whether laboratories placed the seeds without weight in one layer on the AA screen or weighed the seeds as directed in the AOSA Seed Vigor Testing Handbook for all seed lots and crops used in the study. The only exception was in one laboratory for one soybean high quality sample where the variation between the two methods was significant. Variation among laboratories using the same method was found to be greater than variation between methods within the same laboratory. Based on these findings, either aging the seeds by placing them in one layer on the AA screen or weighing the seeds can be used without significant difference in final AAT germination results. These findings provide flexibility to laboratories in using either method depending on each laboratory's preferences and circumstances.

¹Oregon State University Seed Laboratory, 3291 SW Campus Way, Corvallis, OR 97331-3801, E-mail: sabry.elias@oscs.orst.edu; ²California Department of Food and Agriculture Seed Laboratory Plant Pest Diagnostics Center 3294 Meadowview Road Sacramento, CA 95832-1448, E-mail: rbaalbaki@cdfa.ca.gov; and ³Dept. of Horticulture and Crop Science, The Ohio State University, 202 Kottman Hall, 2021 Coffey Rd, Columbus, OH USA 43210-1086, E-mail: mcdonald.2@osu.edu. ⁴Escola Superior de Agricultura Luiz de Queiroz. E-mail: jmarcos@esalq.usp.br.

Comparison of Inert Matter Content Separated from Tall Fescue Samples Using the AOSA Manual Method and a Uniform Blowing Procedure

Adriel Garay, Sabry Elias and Heather Nott

The current method for separation of light inert matter in tall fescue (TF; *Festuca arundinaceae* Schreb.) is lengthy and depends primarily on individual's visual interpretation to the one-third caryopsis development in the AOSA Rules. The Uniform Blowing Procedure (UBP) has been successfully used in inert matter assessment of Kentucky bluegrass and orchardgrass for decades. Similarly, a UBP has been developed for TF. The objective of this research was to determine whether the UBP for TF would produce comparable results (within tolerance) to the current AOSA visual/manual method. Two studies including commercial and certified samples with different varieties and inert matter levels from various years, 2003 (a drought year), 2004 and 2005 (normal years) were conducted to separate inert matter using both the UBP and the current AOSA method. The results showed that regardless of the variety, year, environment or the inert matter content, both methods produced comparable results, i.e., within tolerance, in every case. A national referee was conducted in 2007 to study the correlation between the current AOSA method and the UBP in assessing the inert matter of TF samples and compare the time savings between the two methods. The results showed that all participating laboratories separated comparable amount of inert matter (i.e., within tolerance) according to the AOSA Tolerance Table 13A, using both the UBP and the current AOSA method. In 2008, a Northwest referee was conducted on eight TF samples that contained up to an average of 12% inert matter, and were considered problematic. The hypothesis was that the current AOSA method and the proposed UBP would give different results. Four laboratories from Oregon submitted the samples as they came from customers. Six labs from Oregon, Washington, Idaho, and California participated in the referee. The results indicated that the inert matter separated by both the current AOSA method and the UBP were within tolerance for all samples by all laboratories with the exception of one sample with high multiple florets, which was within tolerance in three laboratories. Results indicated that variation among laboratories within the same method was greater than the variation between the two methods. The results of all the studies showed that both methods produce comparable results; hence, the UBP can be used as an alternative to the current visual/manual method. Because of its mechanical nature, the UBP will contribute to simplicity, efficiency, standardization and consistency of test results within and among laboratories.

How to Produce and Assure Uniformity in Master Calibration Samples

Adriel Garay, Sabry Elias and Heather Nott

Calibration samples are used to calibrate seed blowers that are used to separate light inert matter in purity tests. These samples make it possible to find comparable optimum blowing points across blowers regardless of physical variation. This contributes to uniform blowing across blowers and laboratories. As more grasses are entering national and international markets, such procedures are needed. However, there is no established procedure on how to develop uniform calibration samples and how to ensure uniformity is maintained during their life span. The objective of this study was to develop a step-wise procedure to prepare uniform master calibration samples, using tall fescue as a model. The overall procedure requires several critical steps, including: finding the optimum blowing point for the species of interest; validating that point across samples; preparing calibration samples that can find that point; and verifying that all calibration samples are comparable (uniform). The optimum blowing point was identified in tall fescue and was validated across samples representing different varieties, years and growing conditions. The validation studies used visual examination of florets according to the one-third caryopsis rule and by germinating the light and heavy fractions of samples. Preparation of Master Calibration Samples followed making sure the overlap of pure seed (colored red) and the light portion (colored green) would coincide with the desired optimum blowing point. The master calibration samples developed in this study found the blowing point accurately, consistently and efficiently. Finally, a set of calibration samples that are proven to be uniform can be called "Master Calibration Samples". The importance of maintaining uniformity throughout the life of the calibration samples as well as the potential applications and implications of this method for other temperate and tropical grass species is discussed.

Oregon State University Seed Laboratory, 3291 SW Campus Way, Corvallis, OR 97331-3801. E-mail: adriel.garay@oscs.orst.edu; sabry.elias@oscs.orst.edu.

— POSTER PRESENTATIONS —

Bonafide BDI[®] - Ryegrass,
A Novel, DNA-based Diagnostic Tool for
Adventitious Presence Test in Perennial Ryegrass

A.C. Chandra-Shekara, Michael Thompson and Pegadaraju Venkatramana*

Adventitious presence (unintended presence) of annual ryegrass in perennial ryegrass seed lots causes significant economic losses to the grass seed industry. International Seed Testing Association (ISTA) recommends the usage of SRF (Seedling Root Fluorescence) and/or grow-out test procedures to estimate the levels of annual ryegrass presence in perennial seed lots. However, both SRF and grow-out tests are labor intensive and time consuming. Furthermore, the SRF test produces inaccurate results and is environmentally influenced. Increasing numbers of perennial seed lots are rejected each year due to the inaccuracy of the SRF test. Thus, there is a clear need for a better testing procedure that could meet the diagnostic needs for ryegrass in an efficacious, rapid and cost effective manner. Towards this end we have developed a high throughput quantitative PCR (Q-PCR) based diagnostic tool that effectively detects the presence of annual ryegrass seed contamination in perennial ryegrass lots. The DNA test is designed using an insertion/deletion (In-Del) site in a ryegrass gene involved in regulating the vernalization response of ryegrass. This new DNA test is more sensitive, accurate and cost effective in detecting annual and intermediate type contamination in perennial ryegrass with a high sensitivity of 0.04% in a sample size of 2500 seeds. We have currently validated this method on 68 perennial, 26 annual and 14 intermediate ryegrass varieties with consistent results.

BioDiagnostics Inc., 507 Highland Drive, River Falls, WI-54022. E-mail: Venki@biodiagnostics.net

Differential Scanning Calorimetry
as a Tool for Nondestructive Measurements
of Seed Deterioration in Lettuce
(*Lactuca sativa* ‘Black Seeded Simpson’)

Jennifer Crane and Christina Walters

This study was undertaken to determine if changes in lipid phase behavior could be used to detect lost viability in lettuce (*Lactuca sativa*) seeds. We used seeds from the cultivar Black Seeded Simpson that were purchased every 2–3 years since 1989 and stored in resealable plastic bags at constant 5 °C and relative humidity ranging from 30–60%. Viability of seeds from each harvest year was recently tested by germination assays carried out on 2 replicates of 50 seeds each, and seed lots were scored for percentage and rate of germination,

physiological necrosis and abnormal development. Seed lipids were extracted from an aliquot of seed from each harvest year and total lipid content and fatty acid composition were measured. The temperature and energy associated with lipid melting were measured using differential scanning calorimetry (DSC) on whole seed and extracted lipid samples from each harvest year. Reduction of normal germination was evident in seeds after 4 years of storage and germination was less than 20% after 9 years. However, 100% of seeds germinated (radicle emergence) until 13 years of storage and then dropped precipitously to 0% by year 18. Amount of extractable lipid appeared to decline in seeds with increasing time in storage. In addition, a significant decline in linoleic acid was noticed after 9 years of storage. The energy of the lipid melting transition of intact seed also declined with time in storage and is significantly correlated with reductions in normal germination. The DSC measurements required no special handling protocols and did not affect seed viability or vigor. Hence, it may be a useful, nondestructive tool for determining the progress of seed aging and help schedule actual germination assays for monitor testing.

USDA-ARS, National Center for Genetics Resource Preservation, Fort Collins, CO 80521. E-mail: Jennifer.Crane@ars.usda.gov; Christina.Walters@ars.usda.gov.

The NCRPIS — Providing Diverse Plant Genetic Resources for Worldwide Research and Development

**Maria Erickson¹, Lisa Pfiffner¹, Lisa Burke¹, David Kovach¹,
Mark P. Widrechner^{1, 2}, Candice Gardner^{1, 2}**

The North Central Regional Plant Introduction Station (NCRPIS) is an active plant genebank of the U.S. National Plant Germplasm System (NPGS). Dedicated to conserving and providing plant genetic resources and valuable information to researchers worldwide, the NPGS is a network of federal and state institutions and research units coordinated by the U.S. Department of Agriculture - Agricultural Research Service (USDA-ARS). The NCRPIS was established as the first Plant Introduction Station in 1948 in Ames, Iowa in cooperation with Iowa State University. Today, the NCRPIS collections contain over 1,400 different plant species and ca. 50,000 accessions of crop cultivars, elite lines, landraces, populations, and wild and weedy crop relatives (valuable sources of genetic diversity). Our poster will present an overview of these collections and the personnel who curate and conserve them. Our mission includes acquisition and conservation of genetically diverse crop germplasm and associated information, characterization, evaluation, distribution, enhancement and education. Plant germplasm is collected, regenerated, stored under controlled conditions, monitored for viability, and distributed to researchers and educators worldwide. Accessions with ample seed quantities are backed up at the National Center for Genetic Resources Preservation (NCGRP) in Ft. Collins, CO. Where applicable, NCRPIS collection viability is monitored according to

AOSA seed testing rules in our AOSA-certified laboratory under the supervision of an AOSA-certified seed analyst (CSA). The lack of standardized viability testing protocols for many species necessitates a prioritized, collaborative approach to germination research. We conduct research that supports conservation activities, crop improvement and germplasm utilization to meet a wide array of objectives. The expertise of our NCGRP research partners is important to achieving these objectives.

¹USDA-ARS Plant Introduction Research Unit, Ames, IA, ²Adjunct Asst. Professor, Agronomy Dept., Iowa State University, Ames, IA. E-mail: maria.erickson@ars.usda.gov; lisa.pfiffner@ars.usda.gov; lisa.burke@ars.usda.gov; david.kovach@ars.usda.gov; mark.widrechner@ars.usda.gov; candice.gardner@ars.usda.gov

Identification of Foxtail (*Setaria*) Impurities: Examination and Comparison of Four Species

Jennifer Neudorf and Ruoqing Wang

Green foxtail (*Setaria italica* subsp. *viridis*) and yellow foxtail (*Setaria pumila* subsp. *pumila*) are common impurities in a number of crops throughout Canada. Giant foxtail (*Setaria faberi*) is an Asian weed that currently grows in South-central Canada. It is currently on the Weed Seed Order (2005) as a Class 1: Prohibited Noxious Species. Knotroot bristlegrass (*Setaria parviflora*) is a weed found normally in the South-eastern United States (plus California) and may be found in import samples from the southern states and south into South America. These four species have some similar characters and may be difficult to distinguish. The staff at the National Seed Herbarium have examined these four species and found characters that may aid in distinguishing commonly encountered *Setaria* species. These characters are typically located on the fertile lemma, the fertile palea and the 2nd, or upper, glume. The general shape and size are also important to the correct identification of foxtails.

National Seed Herbarium, Canadian Food Inspection Agency, Seed Science and Technology Section, 301-421 Downey Road, Saskatoon, Saskatchewan S7N 4L8, Canada. E-mail: Jennifer.Neudorf@inspection.gc.ca; Ruoqing.Wang@inspection.gc.ca

The Effect of Seed Vigor on the Uniformity of Soybean Seedling Emergence

D.B. Egli and M. Rucker

By definition, high vigor seed is expected to exhibit "... rapid, uniform emergence... in a wide range of field conditions". The practical benefits of high-vigor seed in stress environments are well documented and they include higher levels of emergence that are reached sooner in comparison to low-vigor seed. Less is known about the effects of seed vigor on the uniformity of soybean (*Glycine max* (L.) Merr.) seedling emergence; consequently, our objective was to evaluate the effect of seed vigor on the timing of seedling emergence. Soybean seeds

from seed lots with high standard germination (86–99%) and a range in accelerated-aging germination (42–94%) were planted 3.8 cm (1.5 inches) deep in soil-filled cups (3 seeds per cup and 100 seeds in each of two replications) in a greenhouse. Emergence (cotyledons above the soil surface) counts were taken at 6-h intervals until emergence ceased. A four-term Gompertz model was used to describe emergence with time and to calculate a Uniformity Index (UI)—the hours from 10–90% of final emergence. Lowering soil temperature from about 22 °C to 18 °C increased the average time to 50% emergence from 106 to 174 h. The UI averaged 66.3 h at the lower temperatures and it decreased (seedling emergence was more uniform) to 38.0 h at the high temperature. Seed vigor had very little effect on UI when the seedlings emerged rapidly, but emergence of seedlings from high seed was more uniform (UI was smaller) than for low-vigor seed when emergence was delayed by low temperatures. Our results are consistent with the definition of seed vigor; high-vigor seed exhibited more uniform emergence than low-vigor seed, but only under stress conditions. Seed vigor had almost no effect on uniformity under ideal conditions.

Department of Plant and Soil Sciences, University of Kentucky Lexington, KY 40546-0312. E-mail: dennis.egli@uky.edu.

Preservation of Plant Genetic Resources at the National Center of Genetic Resources Preservation

David Ellis

The National Center for Genetic Resources Preservation (NCGRP) in Fort Collins, Colorado, with storage of over 750,000 collections of seed and vegetative propagules, safeguards one of the largest ex-situ collections of plant genetic resources in the world. This largest part of the collection is the base collection for the National Plant Germplasm System (NPGS) consisting of over 500,000 collections from 1,162 genera and 6,800 species (as of April 1, 2009). Almost 99% of these collections are stored as seed, usually consisting of 1000–3000 seed/collection. Seed collections are equilibrated (dried) to 6–10% moisture content and stored long-term (decades to centuries) at –18 °C or in the vapor phase of liquid nitrogen (–175 °C). Viability of the stored seed is initially assessed post-equilibration and then periodically as resources allow. All information on the collections is entered into the NPGS central database, the Germplasm Resources Information System (GRIN). Small research quantities of seed (25–100 seed) from all collections in the NPGS are distributed freely to qualified researchers throughout the world. In addition to the base collection at the NCGRP, the Center also provides duplicate back-up storage for important national and global collections. Examples of the duplicate storage include the global rice collection from the Philippines, the global wheat and maize collections from Mexico as well as collections from NGOs in the U.S.

The Plant Genetic Resources Preservation Program, National Center for Genetic Resources Preservation, 1111 South Mason Street, Fort Collins, CO, USA. E-mail: David.Ellis@ars.usda.gov.

Svalbard Global Seed Vault: Safeguarding the Future of Agriculture

David Ellis

The National Center for Genetic Resources Preservation in Fort Collins, Colorado coordinates the participation of the USDA-ARS National Plant Germplasm System (NPGS), along with 26 other genebanks from around the world, in a global effort to safeguard seeds of plants of importance to agriculture throughout the world by placing them in the Svalbard Global Seed Vault. Situated mid-way between the northern tip of Norway and the North Pole in a remote arctic island archipelago, the Svalbard Global Seed Vault marks its second year of operation in 2009. The Seed Vault currently contains over 400,000 seed collections from 220 countries, 31,000 of these samples from the NPGS. The vault entrance is ~430 feet above sea level, well above any predicted rise in sea level due to global warming. The vault consists of a 370-foot tunnel drilled into the solid rock permafrost in the side of a mountain leading to three seed storage rooms (~90' × 30' × 20'). The storage rooms and surrounding rocks are cooled to -18 °C, although the permafrost will ensure the seed remains frozen in the unlikely event of a long-term power outage. The vault has no full time personnel on site, but local authorities have been enlisted to assist with security and any mechanical problems. The yearly operating expenses (estimated to be ~\$200,000) are supported by the Global Crop Diversity Trust and the Government of Norway. The Seed Vault is managed by the Nordic Genetic Resource Center (NordGen) and continually monitored. Seed in the vault is stored free of charge under a deposit agreement. Ownership of the seed is retained by the donor and access is limited to the donor only. More information can be obtained at <http://www.nordgen.org/sgsv>.

The Plant Genetic Resources Preservation Program, National Center for Genetic Resources Preservation, 1111 South Mason Street, Fort Collins, CO, USA. E-mail: David.Ellis@ars.usda.gov.

Laboratory Methods to Break Dormancy in Eastern Gamagrass (*Tripsacum dactyloides* L.) Seeds

Cindy L.H. Finneseth¹ and Robert L. Geneve²

Eastern gamagrass (*Tripsacum dactyloides* L.) is a widely-distributed native warm-season perennial grass with considerable utility including erosion control, wildlife planting, ornamental, forage, and as a biofuel source. Many cultivars, selections and ecotypes are available commercially. Plantings are most commonly established from seed; however, dormancy is a barrier to stand establishment. Development of one or more practical treatments to reduce dormancy and improve germination is of immediate commercial value to producers. The objective of this project was a practical assessment of laboratory methods for overcoming dormancy in eastern gamagrass. A single 'Pete' seed lot harvested

in 2005 with an initial estimated viability of 74% based on tetrazolium (TZ) analysis was used for the experiments. Primary dormancy breaking treatments included: a) moist chilling, b) cupule removal, c) afterripening, d) predry, e) leaching, f) fire, g) charred wood extract, h) H₂O₂, i) GA₃, j) KNO₃, and k) scarification (chemical and physical). Seeds were exposed to treatments then germinated for 4 weeks at 20/30 °C. Fire and predry treatments were lethal; all other treatments except afterripening, leaching (48 h), and GA₃ (100 ppm), improved germination compared to the control (15%). Only moist chilling (6 weeks at 10 °C) was statistically superior at 52% germination, but 24% of the seed remained dormant after 4 weeks at 20/30 °C. An 18-h soak in 15% H₂O₂ solution improved germination, but the effect was not consistent across experiments. Although not commercially feasible, the treatment combination of cupule removal and caryopsis scarification hastened germination and completely eliminated dormancy. At this time, moist chilling remains the most simple, effective and consistent dormancy-breaking treatment for eastern gamma-grass, yet other dormancy-breaking chemicals and combinations of treatments may produce superior germination results. Additional research is necessary utilizing seed lots from other cultivars and harvest years.

¹Division of Regulatory Services Seed Testing Laboratory, 103 Regulatory Services Bldg., University of Kentucky, Lexington, KY, 40546 USA. E-mail: Cindy.Finneseth@uky.edu. ²Dept. of Horticulture, N-318 Ag. Science N., University of Kentucky, Lexington, KY 40546 USA. E-mail: rgeneve@uky.edu.

Identification and Characteristics of *Solanum, Physalis, Datura, and Quincula* Species

Patsy Jackson

The Solanaceae family consists of about 90 genera and 3000–4000 species and is among the most economically important genera worldwide. This family consists of both crops and weeds, and various species produce edible and poisonous fruits. Due to the global reliance on this family as a food source, it is important to understand seed identification characteristics. This poster will review common seed characteristics of various Solanaceae species and provide readers with proper tools for Solanaceae identification. Many morphological seed characteristics within the Solanaceae family are similar, but hilum, size, shape and seed coat texture can be used to determine differences between species. The seed shape may be oval, circular, C to D shaped and the diameter ranges from 1.5 to 2 mm. Seed color is usually uniform, but length of time in a mature fruit and aging can influence seed color. Although hilum, shape, texture, size and color are the most obvious indicators, analysts should also explore other options, such as knowledge of where the seed was found. This can often significantly narrow the list of species identification. Overall, Solanaceae seeds can be difficult to distinguish, but analysts can examine several seed characteristics for proper identification.

USDA-AMS-LS-SRTB, 801 Summit Crossing Place, Suite C, Gastonia, NC 28054. E-mail: patsy.jackson@ams.usda.gov.

A High-throughput System for Integrated Extraction and PCR Amplification of DNA from Seed and Leaf Tissue for Plant Genotyping Studies

Steve Michalik

The Extract-N-Amp™ Plant and Seed PCR Kits contain all the reagents necessary to rapidly extract genomic DNA from plant leaves or seeds, respectively, and amplify targets of interest by PCR. A novel Extraction Solution eliminates the need for conventional organic extraction of plant tissues, column purification, or precipitation of DNA. While mechanical disruption of seeds is necessary for successful extraction of DNA from seeds, we have found that our technology obviates the need for such processing of plant leaves. Not only do the kits provide all materials needed to effectively extract DNA, but they also include a PCR mix especially formulated for amplification directly from the extract. These PCR ReadyMix formulations contain dNTPs, buffer, MgCl₂, and use an antibody-based hot start for specific amplification. The PCR master mixes come in two formulations: Extract-N-Amp™ PCR Reaction Mix and REExtract-N-Amp™ Plant PCR Kit. The REExtract-N-Amp™ PCR mix contains a dye that acts as a tracking dye and allows for convenient direct loading of PCR reactions onto agarose gels for analysis. Genomic DNA is extracted from 0.5–0.7 cm plant leaf disks that have been cut with a standard paper punch or from ground seed material. The leaf disk or ground seed is simply incubated in Extraction Solution for 10 min and then an equal volume of Dilution Solution or Neutralization Solution is added to the extract to neutralize inhibitory substances prior to PCR. A portion of the DNA extract is then added to a PCR reaction containing primers and either the REExtract-N-Amp™ or Extract-N-Amp™ PCR.

Sigma Aldrich, 2909 Laclede Ave., St Louis, MO 63103. E-mail: Steve.Michalik@sial.com.

Annonaceae Seeds: Desiccation Tolerant with Unusual Physiologies

Gayle M. Volk*, Remi Bonnart, and Christina Walters

Pawpaw (*Asimina triloba*) is the only temperate species of the Annonaceae family. Wild pawpaw trees can be found along the river valleys of the eastern and central United States and produce the largest fruits of any species native to North America. Pawpaw seeds are reported to be classified as recalcitrant, with small embryos buried within a reticulate endosperm and a hard seed coat. We performed desiccation and low-temperature exposure experiments to determine if pawpaw seeds have the potential for storage in *ex situ* genebanks. Germination was determined for seeds desiccated to between 3 and

35% moisture content (fresh weight basis) after either 7 or 15 weeks of stratification at 4 °C. Seeds with 3% moisture content were also cooled to -18 °C. Our research revealed that pawpaw seeds had high germination levels after -18 °C exposure for 24 h when stratified either before or after the desiccation treatment. Desiccation tolerance was also investigated for several tropical members of the Annonaceae family. *Annona reticulata*, *A. glabra* and *A. muricata* seeds successfully germinated after desiccation, -18 °C exposure, and gibberellic acid treatments. Additional seed storage experiments will determine how long Annonaceae seeds can be successfully stored within genebanks.

USDA-ARS National Center for Genetic Resources Preservation, 1111 S. Mason St., Ft. Collins, CO 80521. E-mail: Gayle.Volk@ars.usda.gov; Christina.Walters@ars.usda.gov

Seed Storage Containers: Implications of Water Permeability Properties on Moisture Management

Christina Walters and Lisa Hill

Seed moisture must be controlled to maintain high seed quality. Moisture control is accomplished by adjusting or conditioning relative humidity and temperature surrounding the seed. Seeds are packaged in moisture-proof containers to maintain the desired moisture content. The effectiveness of containers as moisture barriers varies with the materials used, and water vapor permeation rates for most materials are known. Specifications when purchasing seed containers should be based on these known water vapor permeation rates as well as the outside environmental conditions and the time that the seed is expected to remain as inventory. While no package is completely moisture-proof, packaging that is highly impermeable to water will help to maintain a near constant seed water content. However, another problem may arise with moisture-proof containers if temperature is not also controlled. Using data loggers, we can demonstrate that relative humidity increases during warming of seed-filled containers and decreases during cooling. Thus, temperature fluctuations can cause fluctuations of relative humidity (RH) within moisture proof packaging. These fluctuations are predicted by water sorption isotherms, which describe temperature-RH-water content interactions within seeds. Elevated RH from warming seeds in sealed bags can cause them to deteriorate faster than expected and reduce the benefit gained from an expensive moisture barrier.

USDA-ARS National Center for Genetic Resources Preservation, 1111 South Mason Street, Fort Collins, CO, USA. E-mail: Christina.Walters@ars.usda.gov; Lisa.Hill@ars.usda.gov.

Cryogenic Storage of Cereal Grains: Results from a 20-Year Experiment

Christina Walters, Lana Wheeler, Phil Stanwood

This paper compares the viability of small grains stored under conventional ($-18\text{ }^{\circ}\text{C}$) or cryogenic conditions [vapor above liquid nitrogen(LN)] for 22–25 years at the National Center for Genetic Resources Preservation. Several accessions of different small grains crops were split in 1984–1987, stored at the two temperatures, and assessed for viability in 2003–2008 using standard testing protocols. While some grasses known to have poor shelf life (i.e., *Bromus inermis* and *Lolium multiflorum*) showed slightly higher germination percentages after storage in LN compared to $-18\text{ }^{\circ}\text{C}$, there were no significant differences between percent germination of most grain species stored under the two conditions. However, grains of *Hordeum vulgare*, *Sorghum bicolor*, *Triticum aestivum* and *Zea mays* showed lower germination following liquid nitrogen storage compared to $-18\text{ }^{\circ}\text{C}$. A more detailed study of *T. aestivum* grains to determine the basis for this surprising result revealed differences in initial moisture treatments, with $-18\text{ }^{\circ}\text{C}$ being stored at about 11% water while LN-stored seeds were dried to 7.5% water. This level of drying did not appear to reduce initial germination or cause imbibitional damage, but may have predisposed grains to damage by rapid cooling to LN or overdry storage. Older grains sealed in tubes and placed in vapor above LN ($\sim 40\text{ }^{\circ}\text{C}/\text{min}$ cooling) showed reduced germination compared to the same accessions cooled to vapor phase LN in an insulated container ($\sim 1\text{ }^{\circ}\text{C}/\text{min}$). Fresh grains showed no sensitivity to cooling rate. We conclude that several variables need consideration when placing cereal grains in LN storage and that overdrying and rapid cooling should be avoided.

USDA-ARS National Center for Genetic Resources Preservation, 1111 South Mason Street, Fort Collins, CO, USA. E-mail: Christina.Walters@ars.usda.gov; Lana.Wheeler@ars.usda.gov.

National Seed Herbarium

Ruojing Wang and Jennifer Neudorf

The National Seed Herbarium (NSH) of Canada contains approximately 20,000 specimens of 14,000 unique species, representing 239 families and 3,279 genera. These seed specimens were collected from international herbaria, government seed laboratories, and botanical gardens. The earliest seed specimen, a sample of larkspur seed (*Delphinium bicolor*), dates back to 1869. The mission of the NSH: support a fair and effective regulatory regime in consumer protection in compliance with the *Seed Act*, *Weed Seed Order*, *Plant Protection Act*, and *Canada Agricultural Products Act*. Protect the plant resource base by the detection and identification of agricultural weedy species, quarantine pest plant seeds, and invasive alien plant species associated with the *Invasive Alien Species Strategy for Canada*. Goals and Objectives of the NSH: serve as the national reference and resource centre for seed identification in seed trade certification

domestically, regionally and internationally by managing seed specimens, concentrating on economically and environmentally important species; identify and verify plant species for seed testing, seed trade certification, agricultural product importation, quarantine, and the detection of invasive alien plant species; and, develop material references, resources and diagnostic methods for seed identification of weedy species and invasive alien plant species using novel information technologies.

Canadian Food Inspection Agency, Seed Science and Technology Section, Saskatoon Laboratory, 301-421 Downey Road, Saskatoon, Saskatchewan S7N 4L8, Canada. E-mail: Ruojing.Wang@inspection.gc.ca; Jennifer.Neudorf@inspection.gc.ca.