

**National Plant Germplasm Coordinating Committee
and NRSP 6 Technical Advisory Committee
Joint Meeting Agenda**

**May 30, 2018
Sturgeon Bay, WI**

Minutes

Participants:

NPGCC - Peter Bretting, Tim Cupka, Larry Chandler, Ed Kaleikau, Eric Young, Greg Cuomo, Bob Stougaard, Gan-Yuan Zhong, Randi Johnson, Bill Tracey, David Baltensperger (by phone)

NRSP 6 Technical Advisory Committee - Max Martin, Cathleen McCluskey, Craig Yencho, Joyce Loper, Curzio Caruati, Jeff Endelman, Jesse Schartner, and John Bamberg, Ronald French, JL Willett, Josh Parrens, Phil Simon, Sager Sathmorali, Joseph Combs, Bill Barker

Time	Topic
Wednesday, May 30	
8:30 – 12:00	Joint NPGCC and NRSP 6 TAC Meeting
8:30 – 8:35	Welcome and introductions – Craig Yencho and Jan Nyrop
8:35 – 8:45	Welcome to the US Potato Genebank and PARS – John Bamberg and William Barker
8:45 – 9:30	<p>ARS and NPGS Update – Peter Bretting (PowerPoint)</p> <p>In meeting report:</p> <ul style="list-style-type: none"> • Number of accessions and distributions in 2017 are both up from 2016 • Budget essentially flat since 2014, with a minor increase in 2018 • PGR management capacity has been decreasing essentially since the mid-2000s • Maintenance of germplasm still highest priority, regeneration and data management are second highest • Many NPGS personnel have retired, but hiring freeze was just lifted. ARS will be hiring 700 personnel throughout the agency over next year, a few of which will be new NPGS staff. • Gayle Volk (ARS, Ft Collins) and Pat Byrne (CO State) organized a workshop in April 2018 at Colorado State to discuss need for curator training due to so many retirements (see further details below under PBCC report).

9:30 – 10:00	<p>PGOC and Regional Plant Introduction Stations Update – Gan-Yuan Zhong (PowerPoint)</p> <p>In meeting report:</p> <ul style="list-style-type: none"> • Due to the restricted budget, no PGOC meeting was held in 2017, only a conference call • Next GOC meeting June 19-21 2018 at Beltsville, MD and Washington, DC • Northeast Regional Plant Introduction Station NE-9, Geneva, NY – vegetables and apple, cold-hardy grape, and tart cherry clonal crops • Southern Regional Plant Introduction Station S-9, Griffin, GA – sorghum, peanut, pepper, other vegetables, and sweet potato • North Central Regional Plant Introduction Station, NC-7, Ames, IS – maize, grasses, cucurbits and other vegetables, brassica, sunflower, etc. • Western Regional Plant Introduction Station W-6. Pullman, WA– alfalfa, garlic, beets, beans, chickpeas, lentils, cool season grasses. • Unintended presence of transgenes in gene bank accessions is an ongoing issue <ul style="list-style-type: none"> ○ Testing for transgenes occurs routinely for maize, alfalfa and cotton, but everyone involved in managing germplasm for these and other crops must help prevent this ○ ARS now has specific best management practices to follow that are aimed at preventing this
10:00 – 10:30	<p>NIFA and AFRI Update and Discussion – Randi Johnson, Ed Kaleikau, and Liang-Shiou Lin (PowerPoint)</p> <p>In meeting report:</p> <ul style="list-style-type: none"> • Sonny Ramaswamy’s appointment has ended and an interim NIFA Director is in place, new permanent director should be in place early next year • Competitive grants related to breeding this year are ~ \$50 Million • Capacity Funds used for breeding related projects is ~ \$20 Million • All CAP grants now under the Sustainable Agriculture Systems RFA • New programs Food & Agriculture Cyber Informatics and Tools (FACT) and Microbiome
10:30 – 11:00	Joint Break
11:00 – 12:00	<p>Future Funding Considerations for NRSP 6 – John Bamberg and William Barker</p> <p>In meeting report:</p> <ul style="list-style-type: none"> • Some directors feel that the NIFA/SAES support for the U. S. Potato Genebank (USPG) through NRSP-6 should cease; so future funding from that source is at risk • Project needs to develop plan to increase USPG support from ARS and outside support from industry and other sources • One constraint is that there are only two or three private potato breeding programs in the US, who already contribute to the project. However, the potato processing industries are not currently providing support

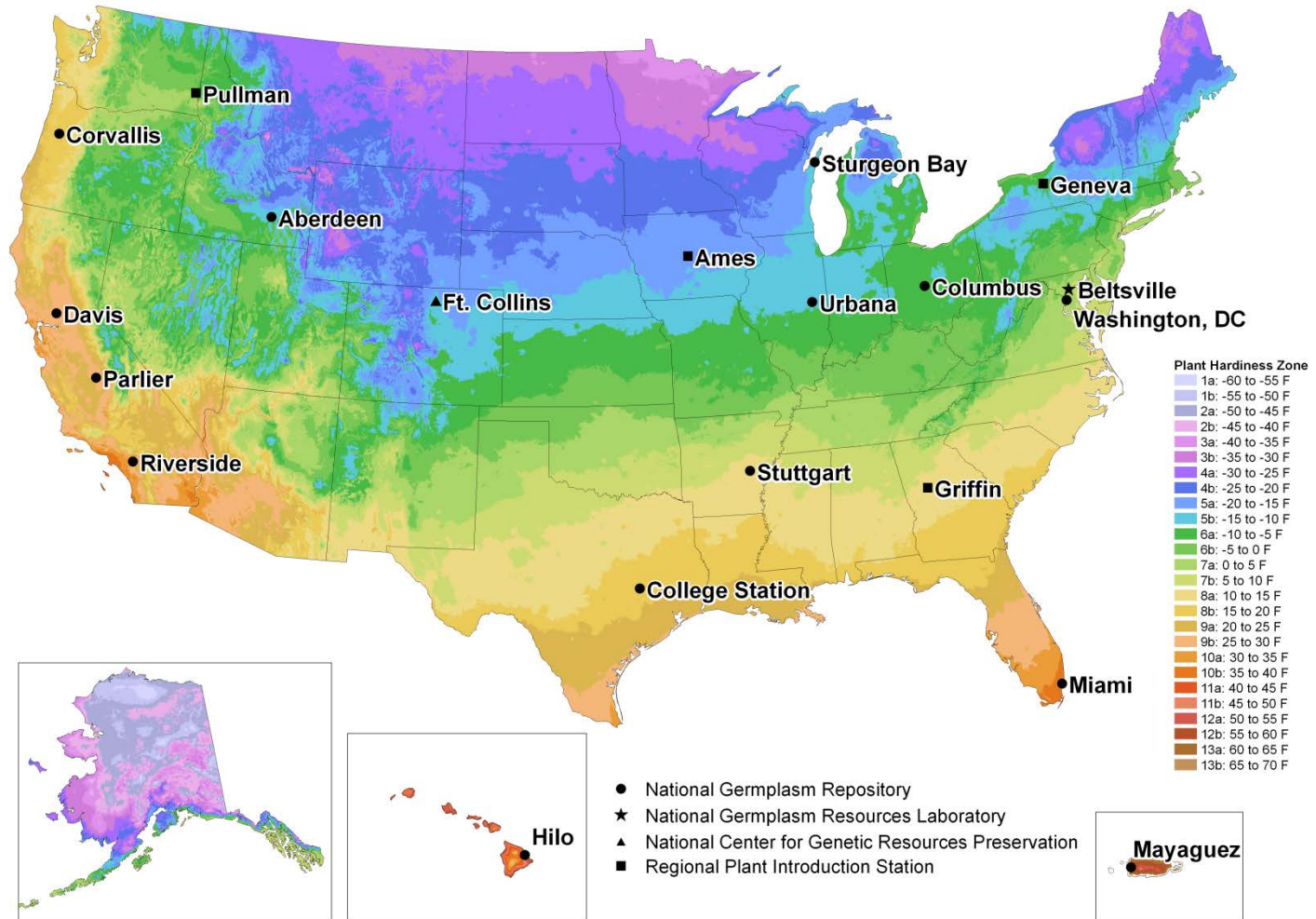
12:00 – 1:30	Joint Lunch
1:30 – 3:30	Separate NPGCC and NRSP 6 TAC Meetings
1:30 – 2:10	<p>Comments from Liaisons – AOSCA – Chet Boruff ASTA – Tim Cupka NAPB – David Baltensperger PBCC – Bill Tracy</p> <p>In meeting report:</p> <ul style="list-style-type: none"> • ASTA – Tim Cupka <ul style="list-style-type: none"> ○ Private seed industry currently in a depression, mergers are occurring to save money, not to expand operations • NAPB & PBCC – Bill Tracey (PowerPoint and handout) <ul style="list-style-type: none"> ○ NAPB meeting in Guelph in August ○ NAPB is ramping up advocacy effort ○ NPGS Workshop on Curator Training (PowerPoint) <ul style="list-style-type: none"> ▪ Also an international need for curator training ▪ Subsequent to the April 2018 workshop in Ft. Collins, a NIFA Higher Education Challenge Grant was submitted to continue developing the curriculum for the training ▪ Multiple delivery mechanisms will be used for this training, including on line, continuing education/in-service, and in person at universities
2:10 – 2:30	<p>National Genetics Resource Advisory Council Report – Peter Bretting (PowerPoint)</p> <p>In meeting report:</p> <ul style="list-style-type: none"> • Currently four vacancies on the Council, which should be filled within the year
2:30 – 2:45	<p>ARS Policies & Procedures for GE Plants – Peter Bretting (handout)</p> <p>In meeting report:</p> <ul style="list-style-type: none"> • BMPs are being implemented now in ARS • Asking Potato Genebank and breeders if a BMP is needed specifically for potato
2:45 – 3:00	<p>International Treaty on Plant Genetic Resources for Food and Agriculture – Peter Bretting (PowerPoint)</p> <p>In meeting report:</p> <ul style="list-style-type: none"> • US State Department diplomat is now serving as Chair of the International Treaty Governing Body • New international issue is that some countries want to regulate access to digital sequence information/genetic sequence data generated from plant genetic resources similar to access to the germplasm itself
3:00 – 4:30	Joint Tour of Potato Genebank

4:30	Adjourn
Thursday, May 31	
8:00 – 12:00	Optional tour of research activities at PARS

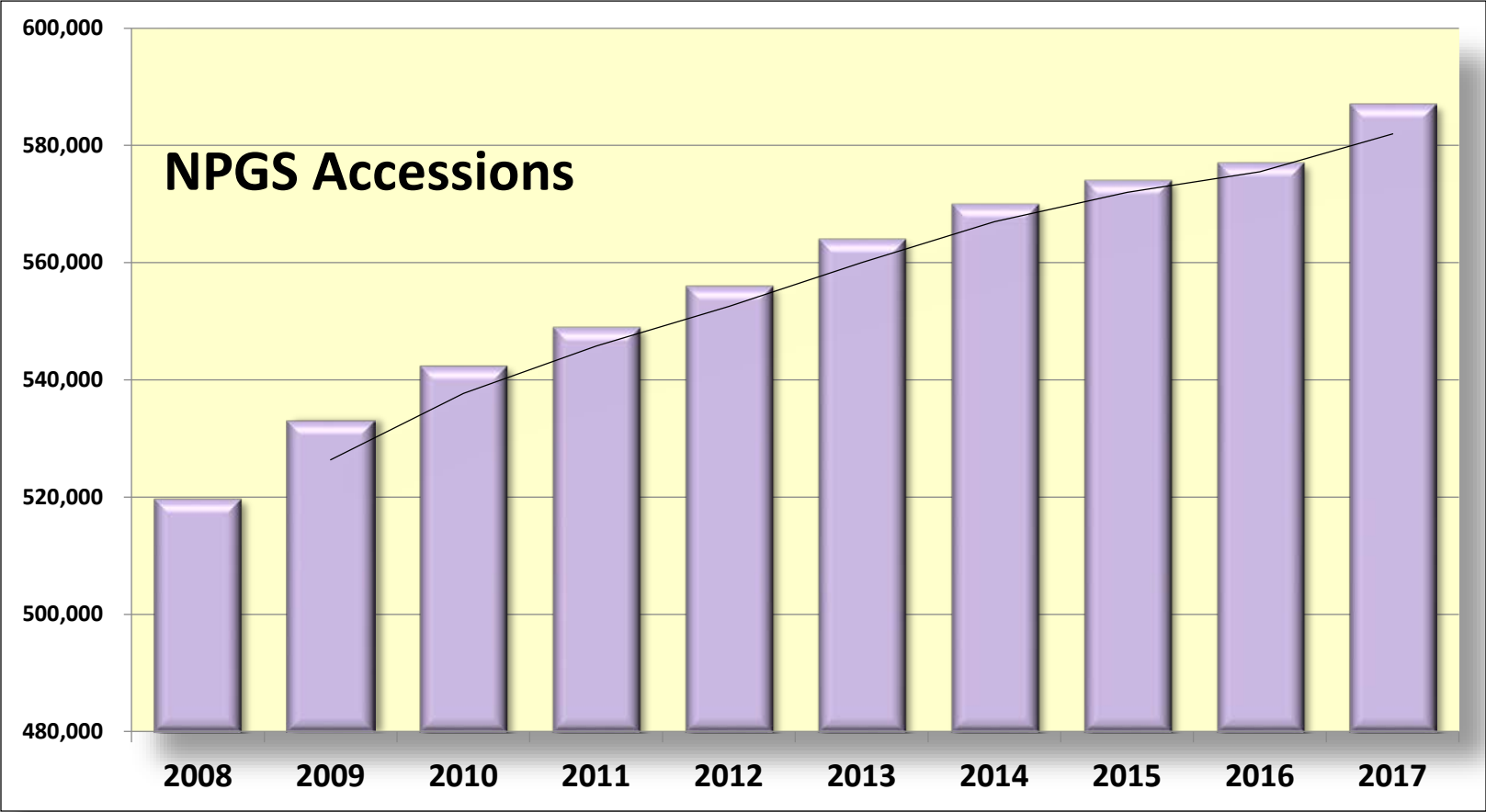
The National Plant Germplasm System: 2018 Status, Prospects, and Challenges

Peter Bretting
USDA/ARS Office of National Programs
Peter.bretting@ars.usda.gov
1.301.504.5541

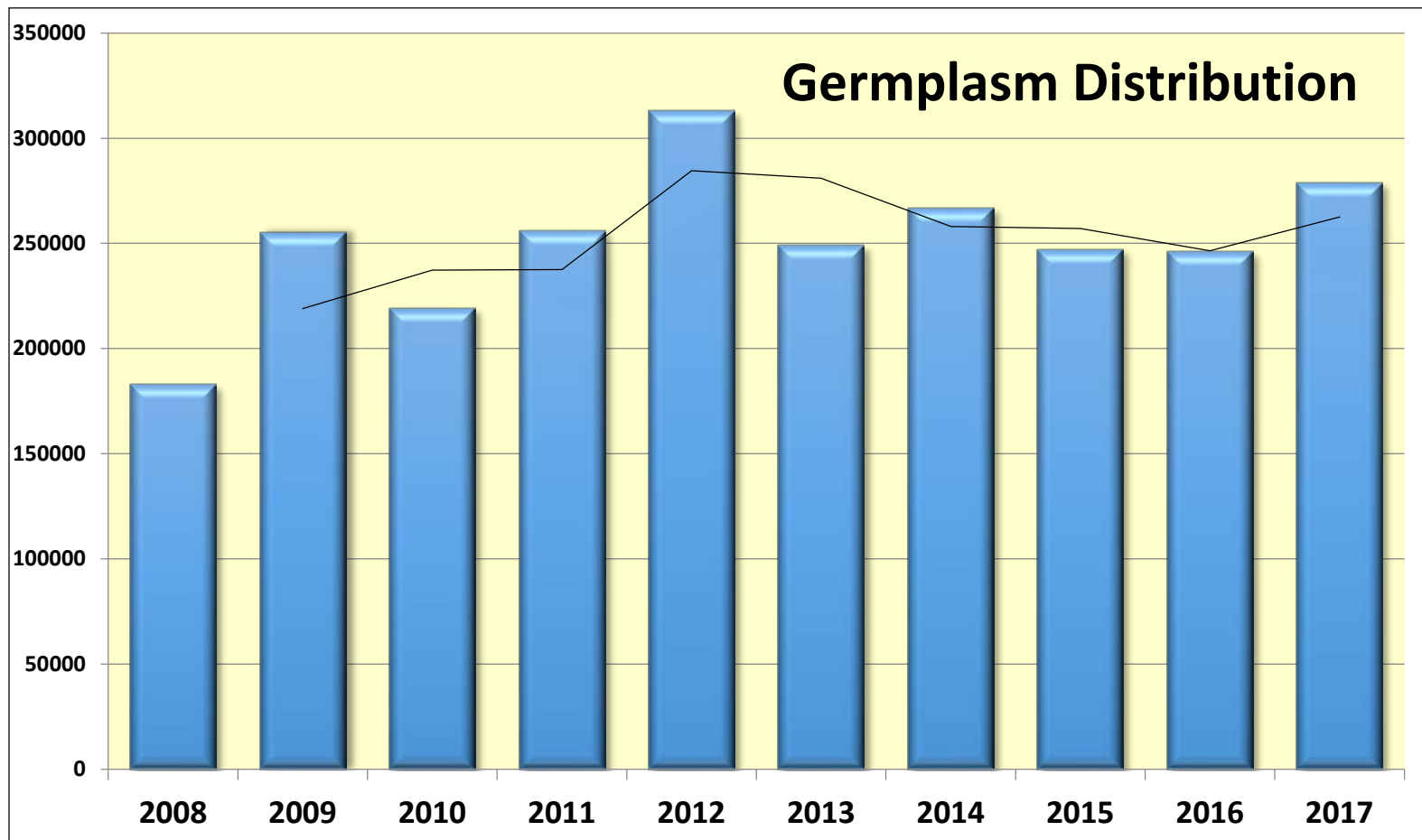
USDA National Plant Germplasm System (NPGS)



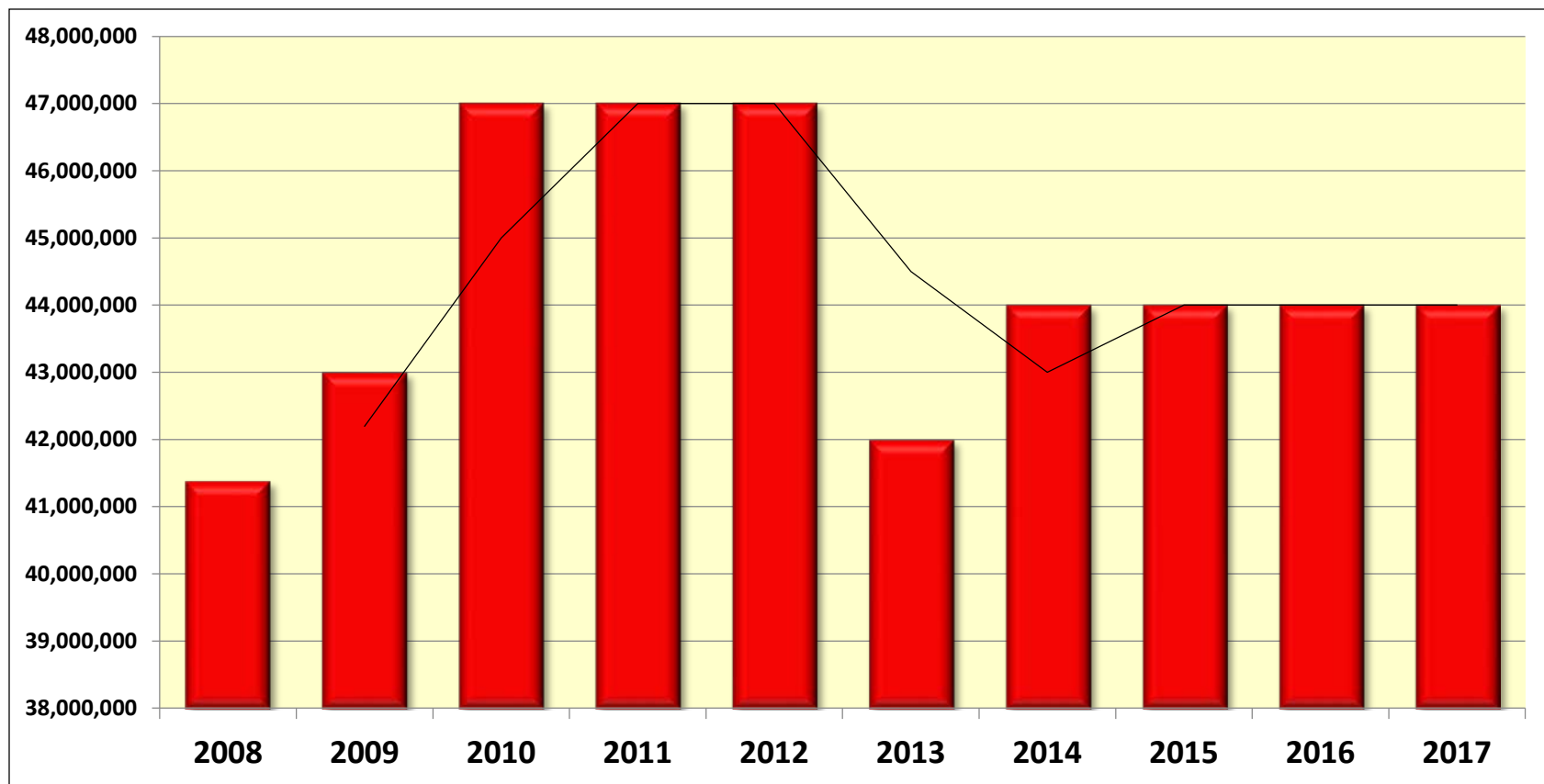
NUMBER OF NPGS ACCESSIONS 2008-2017



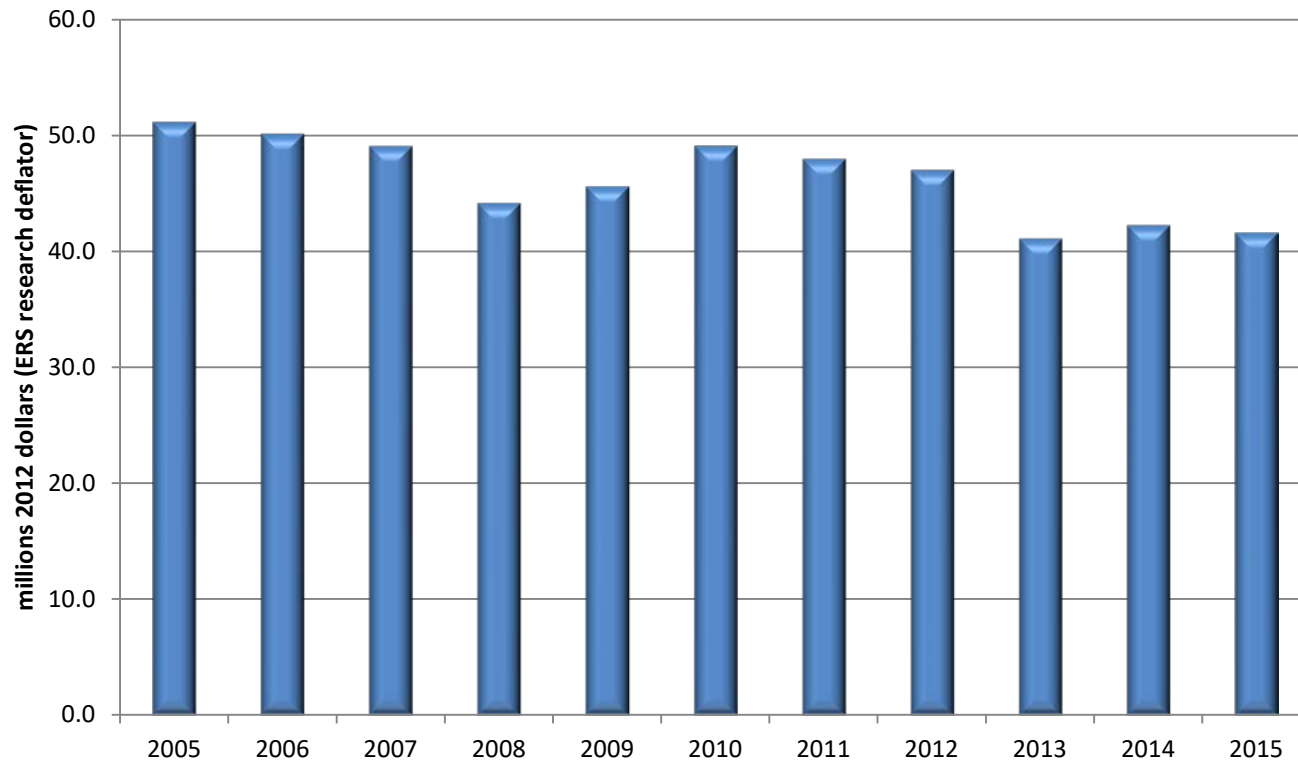
DEMAND FOR NPGS GERMPLASM 2008-2017



ARS NATIONAL PLANT GERMPLASM SYSTEM BUDGET 2008-2017



Real ARS National Plant Germplasm System Budget, 2005-2015, converted to 2012 dollars with ERS research deflator



Note: Deflator for 2015 is preliminary

Some key challenges for the NPGS

- **Managing and expanding the NPGS operational capacity and infrastructure to meet the increased demand for germplasm and associated information.**
- **Recent and upcoming NPGS personnel retirements.**
- **Developing and applying cryopreservation and/or in vitro conservation methods for clonal germplasm.**
- **BMPs and procedures for managing accessions (and breeding stocks) with GE traits and the occurrence of adventitious presence (AP).**
- **Acquiring and conserving additional germplasm, especially of crop wild relatives.**

Genetic Resource Management Priorities

- **Acquisition**
- **Maintenance**
- **Regeneration**
- **Documentation and Data Management**
- **Distribution**
- **Characterization**
- **Evaluation**
- **Enhancement**
- **Research in support of the preceding priorities**

Personnel Changes

- Farewell and best wishes to RLs Richard Percy (ARS-College Station), Randy Nelson (ARS-Urbana) and Gary Pederson (ARS-SRPIS, Griffin) for their retirements.
- Congratulations to Melanie Harrison (ARS-SRPIS, Griffin) for her promotion to RL.
- Farewell and best wishes to Merrelyn Spinks (ARS-SRPIS, Griffin; GRIN-Global, information management).
- Welcome and best wishes to Melanie Schori, new plant taxonomist for GRIN Taxonomy at NGRL, Beltsville.
- With hiring freeze lifted, we hope to hire additional staff in the near future.

Plant Genetic Resource (PGR) Management Training Initiative

- **At least 1/3 of NPGS PGR managers could (likely will) retire within 5 years.**
- **Currently, no formal, comprehensive program exists for training new PGR managers.**
- **G. Volk (ARS-Ft. Collins) and P. Byrne (CSU-Ft. C.) secured a USDA/NIFA grant for a workshop at Ft. C. 24-26 April 2018 that discussed designing & developing a training program for PGR management to be delivered primarily through distance-learning.**
- **The workshop generated numerous insights; another grant (Higher Education Challenge) was already submitted to extend concepts and ideas from the workshop.**



PGOC and Regional Plant Introduction Stations Update

Gan-Yuan Zhong

USDA, ARS, Plant Genetic Resources Unit Geneva, NY

May 24, 2018

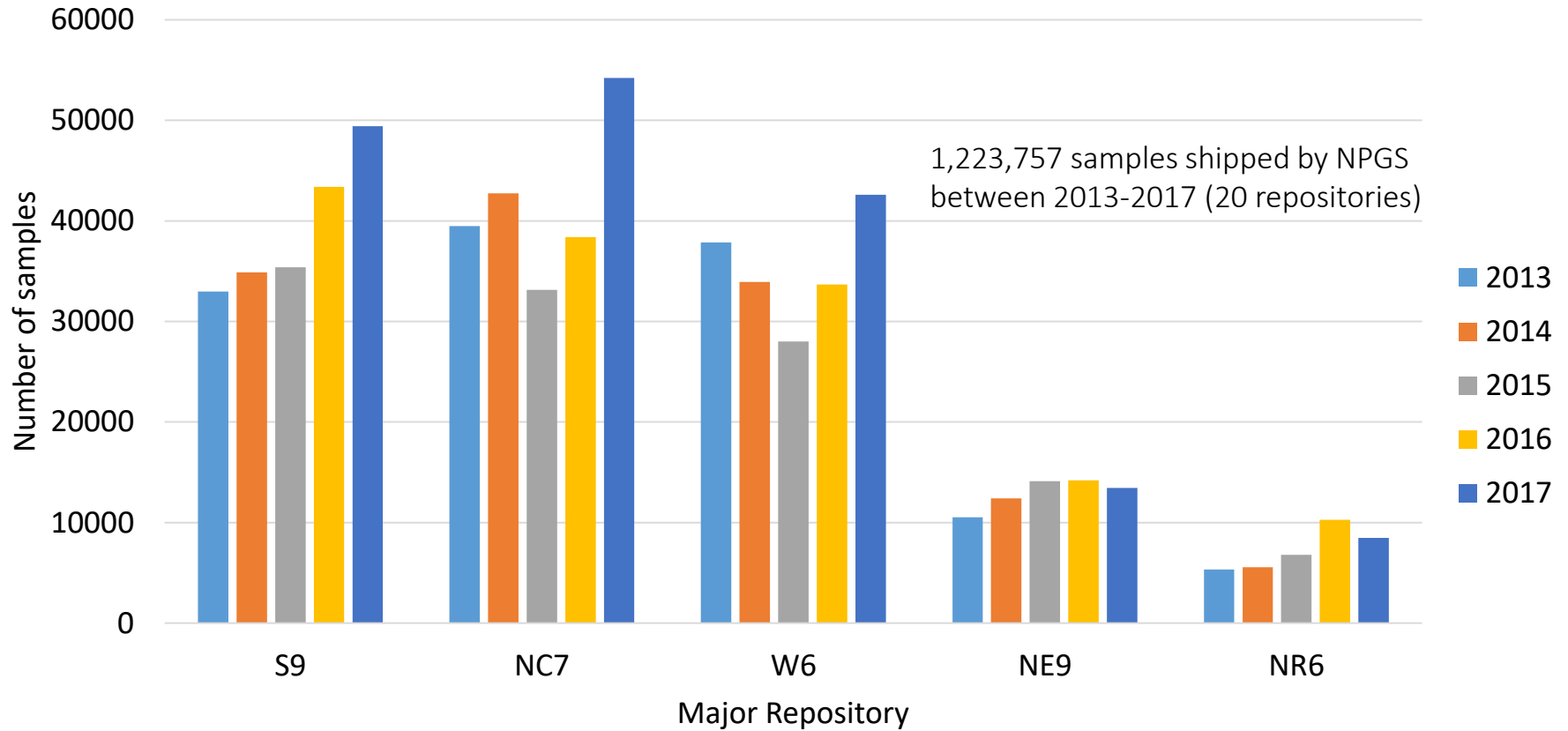
Plant Germplasm Operations Committee

- ✓ A 2hr. short PGOC meeting was convened via teleconference call on Oct. 17, 2017
- ✓ John Preece (PGCO Chair) and Stephanie Greene (Vice PGOC Chair) hosted the teleconference call.
- ✓ The 2018 PGOC meeting will take place at the USDA/ARS National Arboretum in Washington, DC on June 19-21, 2018.
- ✓ Peter Bretting: Current Status and Challenges Facing the National Genebanks.
- ✓ Gary Kinard: Update in GRIN-Global and the National Germplasm Resources Laboratory
- ✓ John Preece: Strategies to Help Solve Resource Limitation Challenges for the National Plant Germplasm System

US Regional Plant Introduction Station Functions

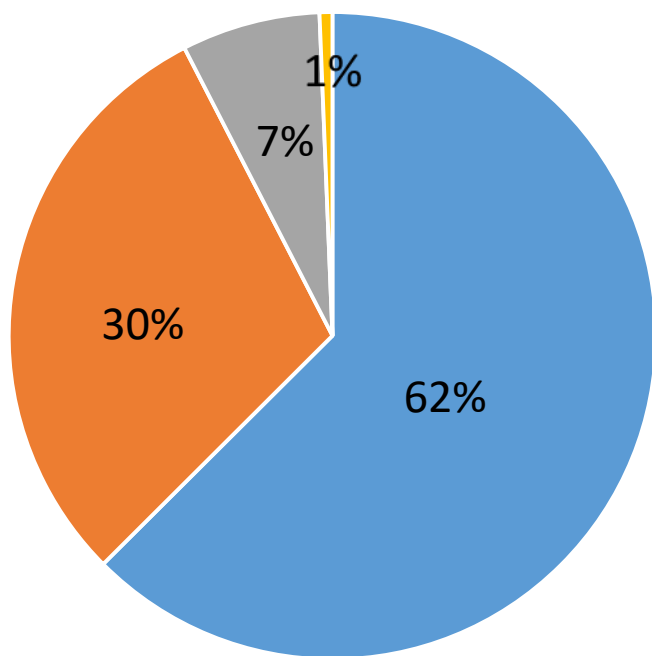
- Acquire, conserve and distribute plant genetic diversity and associated information
- Encourage use of germplasm (User-focused)
- Conduct research to improve genetic resource management programs
- Evaluate and characterize germplasm to facilitate targeted research objectives
- Conduct prebreeding activities to facilitate utilization

Germplasm Distributions for 5 Major Repositories



Northeast Regional PI Station (NE9)

Crops conserved



■ Vegetables	12690 accessions
■ Apples	6069 accessions
■ Grapes	1405 accessions
■ Tart cherries	130 accessions

Two curatorial programs:

- ❖ Vegetable Crops (Joanne Labate & one vacancy) tomato, onion, radish, winter squash, cabbage, cauliflower, broccoli, other cole crops, celery, tomatillo, asparagus, buckwheat and other vegetables
- ❖ Clonal Crops (Thomas Chao & Ben Gutierrez) apples, grapes and tart cherries

Northeast Regional PI Station (NE9)

Highlights:

- Distribution of 13,446 germplasm samples in 2017 and 64,706 samples from 2013 – 2017.
- Successful regeneration of 796 accessions from 2013 – 2016 (average 200/yr) 225 regenerations performed in 2017 for seed production of vegetable germplasm to distribute and replenish stocks.
- Clonal collection added 55 scion and 45 seed accessions to the *Malus* collection.

Wild Malus explorations



Northeast Regional PI Station (NE9)

Highlights:

- Vegetable collection continues to provide germplasm, training, workshops and demonstrations for small-scale seed production to the Northern Organic Vegetable Improvement Collaborative.
- Clonal collection hosted 90 groups/classes with 1,708+ students, growers, researchers, and general stakeholders at the collection from 2012-2017.



Organic Farming Conference outreach

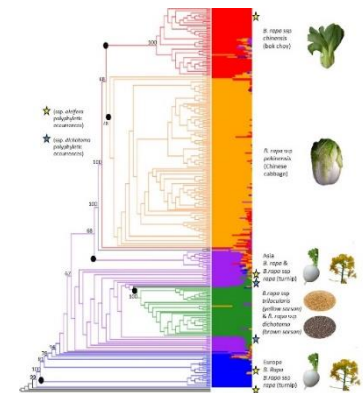


College classes, growers, and CBS clonal visits

Northeast Regional PI Station (NE9)

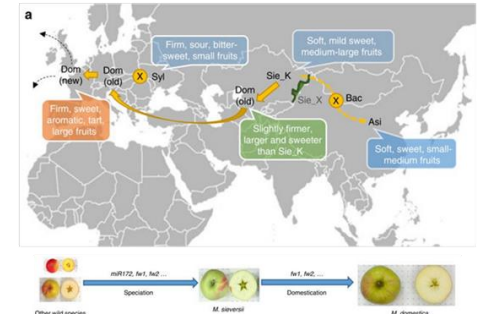
Highlights:

- Characterization data are collected during routine regenerations from vegetable crops; a backlog of more than 6,000 observations for seed crops were uploaded into GRIN-Global since 2017.
- Genetic relationships among of the diverse range of *Brassica rapa* edible types were elucidated using DNA sequencing.
- Tomato cation profile results will provide insight for development of new tomato varieties with favorable nutrient profiles in terms of human health and fruit quality.
- Genome analyses of *Malus* species revealed that the domestication of apple fruit size was a two-stage events and only the *M. sieversii* in Central Asia directly contributed to the domestication of modern day apples.
- A novel QTL controlling grape berry size was identified using a GWAS analysis



Brassica rapa subspecies diversity

Fig. 2a. Apple evolutionary map along the west and east bounds of the Silk Route with center of origin at Kazakhstan in central Asia



Northeast Regional PI Station (NE9)

Impacts of NE9 Collections:

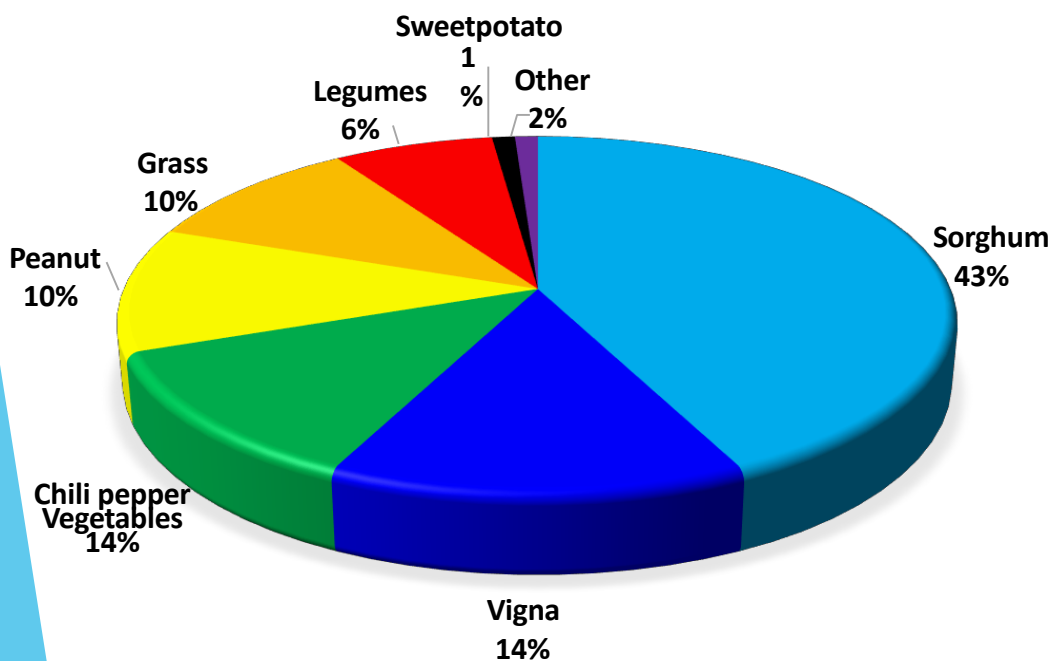
- The major crops managed by this project represent approximately 36% of the combined dollar value of fresh and processing vegetables in the USA in 2017.
- Genes from wild tomatoes have been used to increase ease of harvesting, stress and drought tolerance, and for resistance to pests and diseases. Since the 1960s, the value of fresh and processed tomato to the USA economy increased from \$347 million to its current value of \$1.7 billion.
- *Malus* collection is the largest and most diverse collection in the world.
- The collection contributes significantly to the breeding of grape and apple scions and rootstocks with superior disease, pest, and environmental stresses resistance.
- The collection contributes to the development of emerging cider apple industry



Novel blue mold resistance from wild *M. sieversii*

Southern Regional (S-009)

Crops managed



Curators and Scientists

- Sorghum & S-009 Millets (Melanie Harrison)
- Peanut & Vigna (Shyam Tallury)
- Pepper, Melons, Vegetables (Bob Jarret)
- Legumes, Warm-season Grasses, Clover (Brad Morris)
- Sweet Potatoes (Ming Li Wang)

Please note that several changes in curation responsibilities have taken place this year as reflected in the above list.

Southern Regional (S-009) Stats

- This project has grown from 811 accessions of 41 genera in 1949 to 99,414 accessions representing 263 genera and 1601 species in 2018.
- In 2017, over 49,000 accessions were distributed worldwide to stakeholders.
- Over 84% of S-009 accessions are available for distribution and over 95% are safely backed up at Ft. Collins, CO
- Approximately 88% of the accessions have inventories at -18°C to extend seed viability in storage.



Southern Regional (S-009)

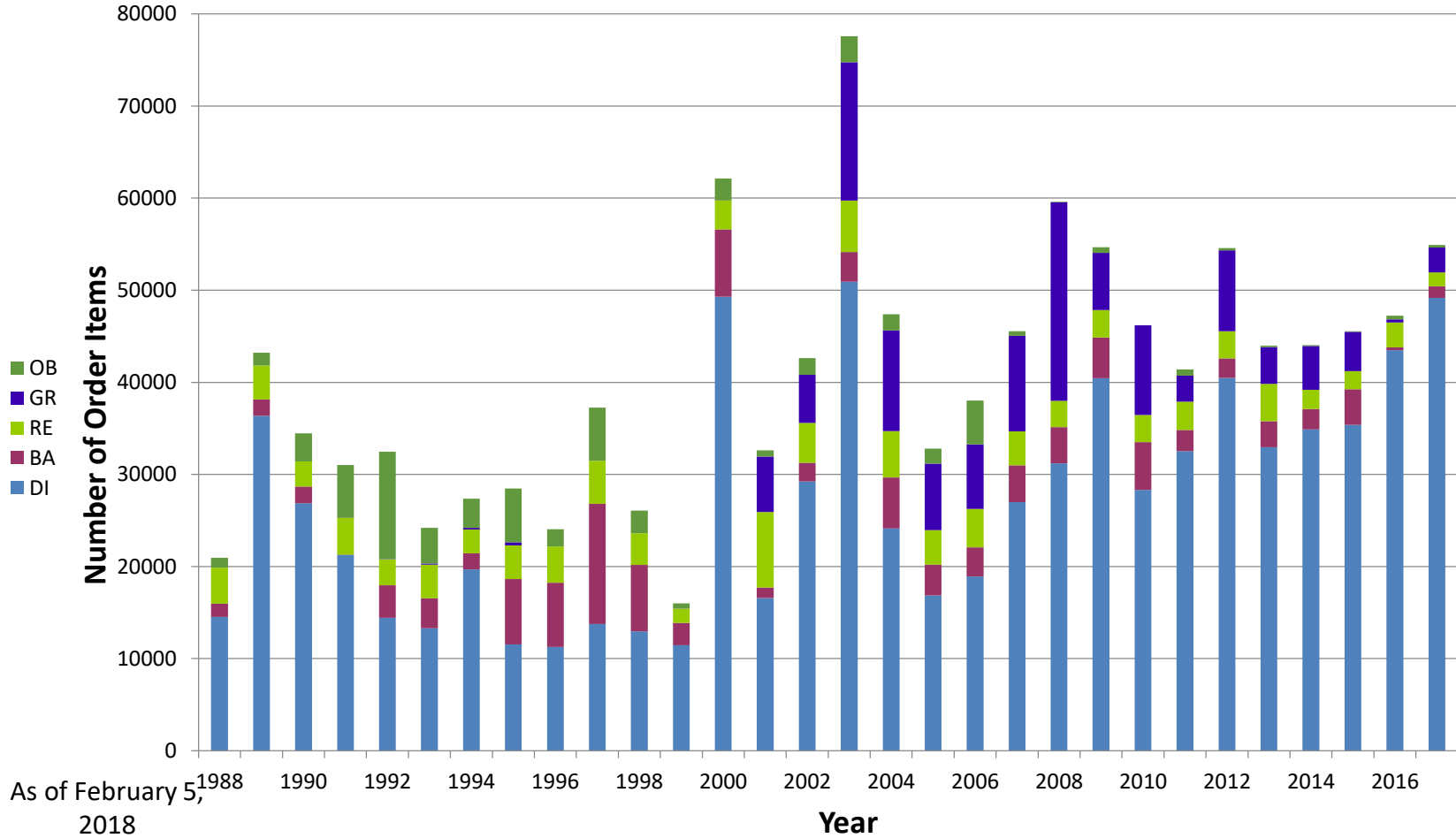
Personnel Changes:

- There were three retirements this past year: Lee Ann Chalkley (Seed Storage Manager), David Pinnow (Plant Pathologist), and Merrelyn Spinks (IT Specialist\GRIN-Global Database Manager).

- New Hires\Appointments:
 - Nick Stigura was hired in Feb 2018 as the IT Specialist\GRIN-Global Database Manager
 - Melanie Harrison was appointed permanent Research Leader effective October 1, 2017.
 - Mylee Mobley was hired in April 2018 as the new peanut technician (S-009 employee)

- Vacancies Approved and to be Advertised Soon:
 - Seed Storage Manager (Biol Sci Tech; GS-7/8/9)
 - Maintenance Mechanic (WG-9/10)

Southern Regional (S-009) Distributions



OB = Observation; GR = Germination; RE = Regeneration; BA = Back Up; DI = Distribution

Southern Regional (S-009)



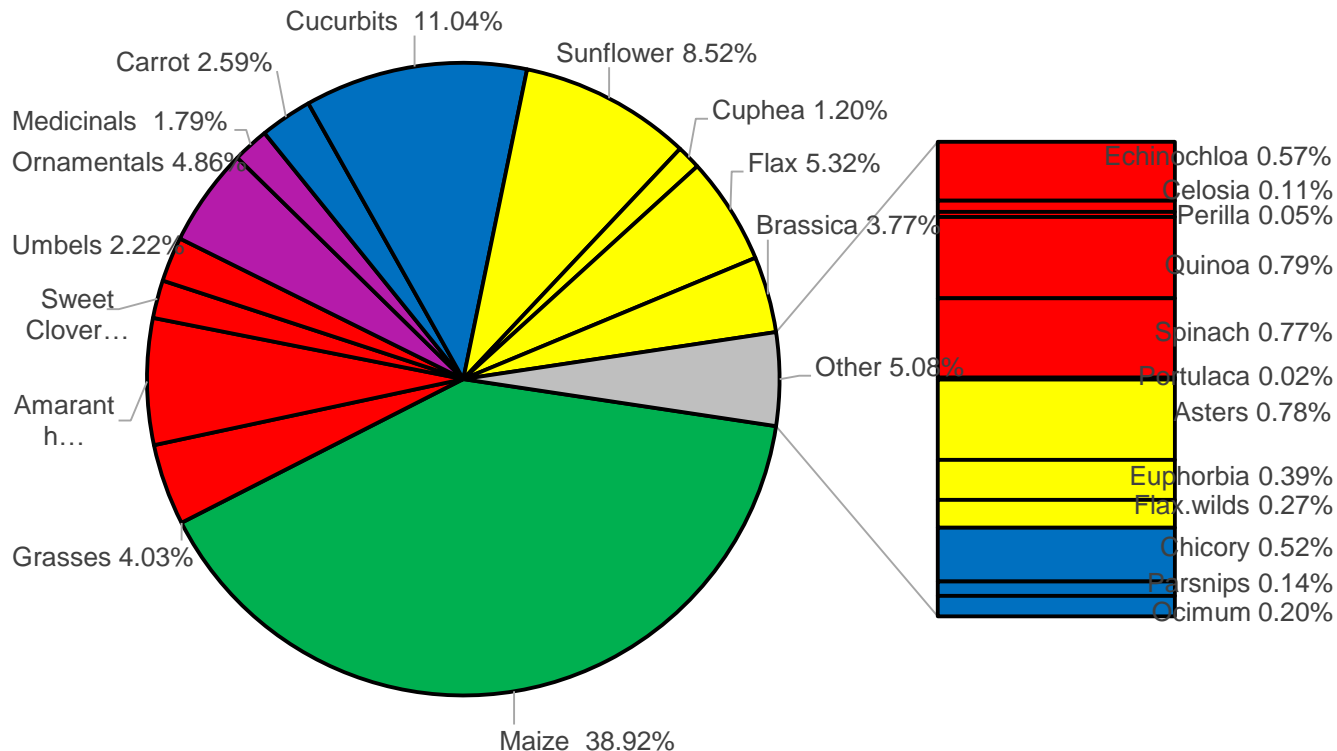
Characterization Efforts

- Seed from 325 watermelon accessions were evaluated for root growth characteristics in collaboration with ARS scientists at Charleston, South Carolina.
- A total of 769 and 1,846 cultivated peanut accessions were evaluated for oil content and fatty acid composition, respectively. Three cultivated peanut accessions were identified that contain a high concentration of oleic acid (averaging 80%), and molecular markers for the high oleic acid trait have been developed for peanut breeders to use.
- Collaborating with ARS scientists in Puerto Rico and Lubbock, Texas, 320 sweet sorghum accessions have been evaluated for early-spring cold tolerance. More than 255 sorghum mutants generated using ethyl methanesulfonate-mutagenesis were sequenced and resulted in the identification of six abscisic acid insensitive mutants for potential environmental stress tolerance.
- Genetic diversity for seed traits and protein content in 111 cowpea core accessions was determined using principal component and cluster analysis.
- A first year crude protein evaluation of seed from 26 functional vegetable accessions including *Vigna* species, sesame, guar, and jute showed crude protein content ranging from 14-26%.
- Seeds from eight field grown sesame accessions were evaluated for the nutritional compounds, sesamin and sesmolin with content ranging from 0.788-6.68 mg/g.

North Central Regional PI Station (NC-007)

Curation and Distribution of 54,412 Accessions

- Strategic collection development; 194 new accession in 2016; 2576 in past 6 years, including 360 pre-breeding sunflower lines containing introgressions from 11 different crop wild relative annual *Helianthus* taxa (below)



Collecting record blue ash tree seeds, *Fraxinus quadrangulata*

North Central Regional PI Station (NC-007)

Curation and Distribution, cont.

- Maintain and provide high quality, well-documented germplasm for research and educational objectives for primarily heterogeneous, heterozygous, outcrossing crops; more than 55,000 packets distributed in 2017
- Characterization and evaluation to increase collection utility



Completion of 4-yr
collaboration on
taxonomic revision of
Daucus and allied
Apiaceae



North Central Regional PI Station (NC-007)

Germplasm Maintenance

Provide six insect pollinator species on demand to support regenerations and software to help manage pollinator requests



Outreach event demonstrating pollinator technologies utilized



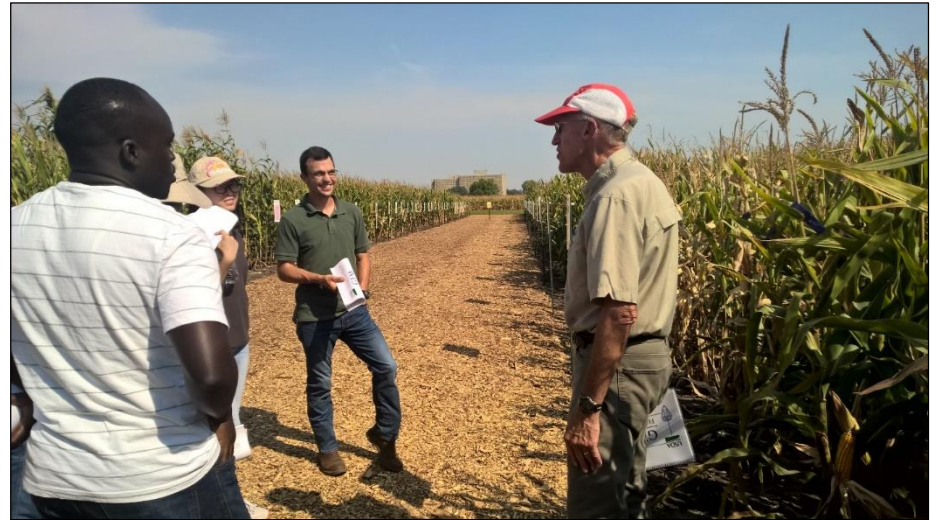
North Central Regional PI Station (NC-007)

2017 Information System Development in Ames

- New version of the GRIN-Global Curator Tool, v 1.9.8.14 released to genebank personnel
- New Viability Wizard for processing information from germination testing released
- New Attachment Wizard for attaching documents (PDFs, images, spreadsheets, text docs) to accession/inventory records released
- Enhancements to the Order Wizard for more efficient processing of germplasm orders released
- Developed Excel tools for better order management
- New Pollinator Tool compatible with GRIN-Global in use for creating, tracking, and closing requests for insect pollinators in regeneration cages

North Central Regional PI Station (NC-007)

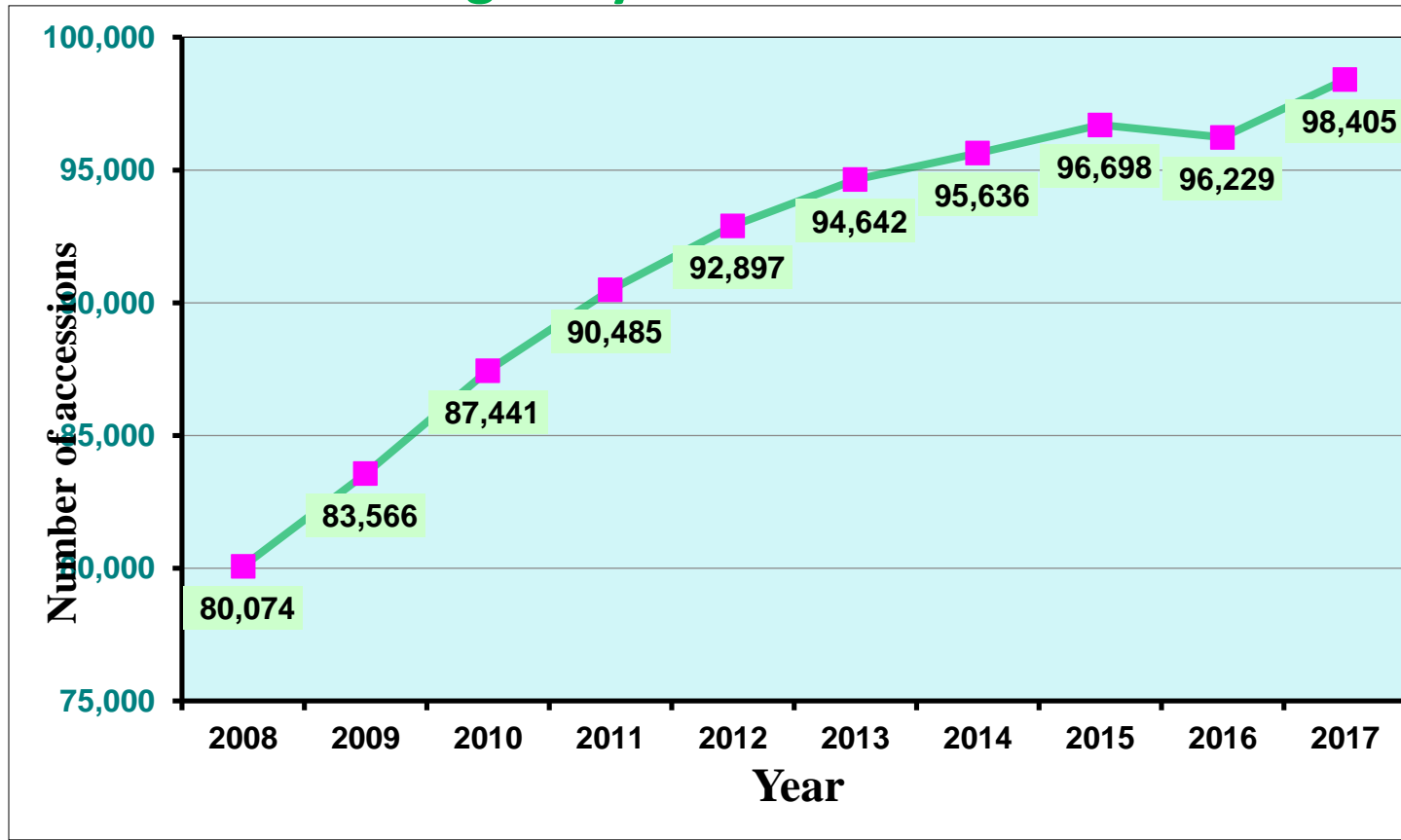
Field Days and Outreach



Sunflower prebreeding lines (top left);
Amaranthus (bottom left);
GEM Project Field Day (above)

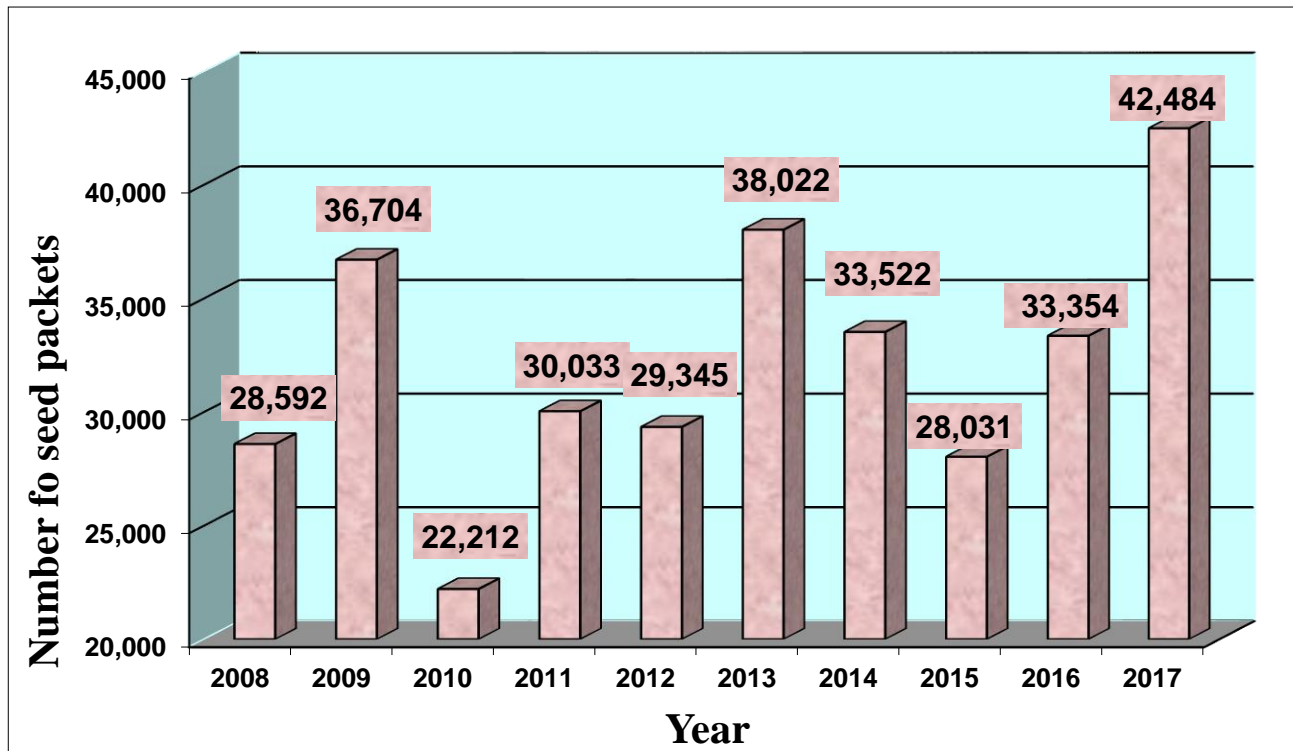
Western Regional PI Station (W-006)

Changes of total number of accessions managed by WRPIS since 2008



Western Regional PI Station (W-006)

Serving the global plant research community
by providing needed germplasm samples



Number of seed packets distributed by WRPIS each year since 2008

Western Regional PI Station (W-006)

Personnel changes:

New Hire:

Mr. **Bo Gao** started on July 20, 2017 as the IT Specialist of the W6 Station.

Retired:

Mr. **Dave Stout** retired in March 17, 2018 after working almost 48 years at the station. Dave started to work at the station when he was attending Washington State University (WSU), Pullman in 1970. He was hired by WSU as a Seed Curator to work at the station with a wide range of responsibilities from germplasm regeneration, characterization, seed cleaning, germination and seed storage and database management. Dave retired from WSU and joined UDSA-ARS in 2004 as the seed storage and database manager. Dave is one of the group members who started the Germplasm Resources Information Network (GRIN) database and contributed substantially to the recent upgrade of the GRIN to GRIN-Global. Dave continues to help us by working half-time after officially retiring from the federal government. Dave is currently serving as a member of the NPGS GRIN-Global Advisory Committee representing the Pullman Station. We greatly appreciate Dave's dedication to managing plant genetic resources.

Western Regional PI Station (W-006)

Maintaining genetic integrity of NPGS alfalfa germplasm

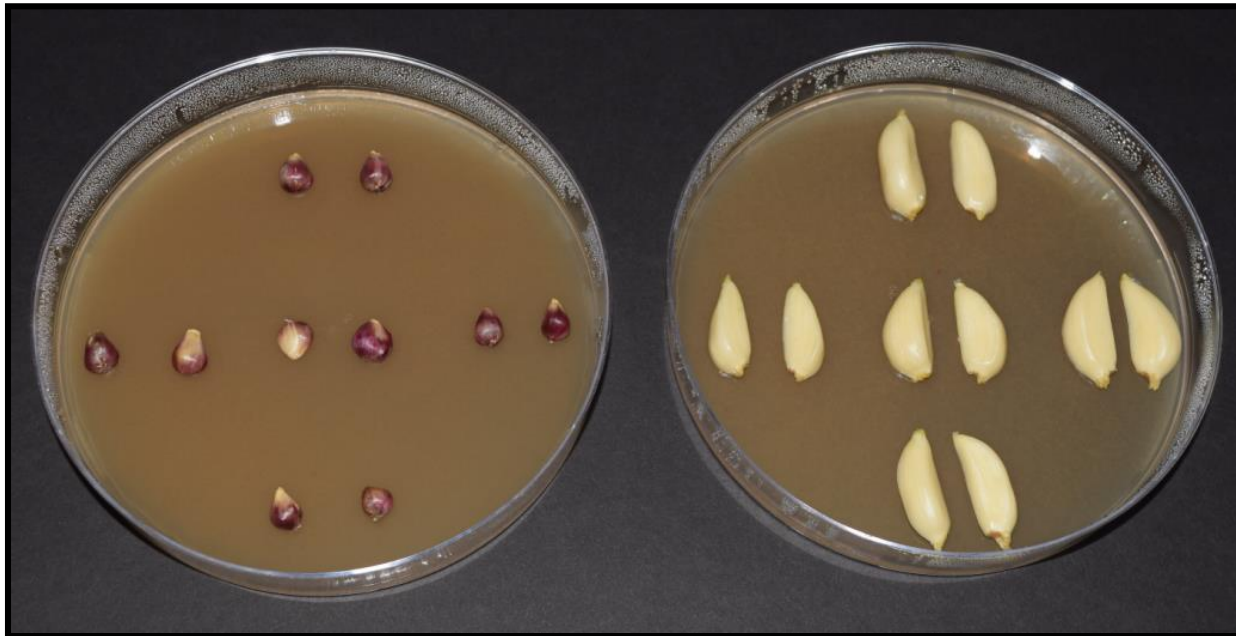


R1	R2	R3	R4	R5	R6
1	50	51	100	101	150
2	49	52	99	102	149
3	48	53	98	103	148
4	47	54	97	104	147
5	46	55	96	105	146
6	45	56	95	106	145
7	44	57	94	107	144
8	43	58	93	108	143
9	42	59	92	109	142
10	41	60	91	110	141
11	40	61	90	111	140
12	39	62	89	112	139
13	38	63	88	113	138
14	37	64	87	114	137
15	36	65	86	115	136
16	35	66	85	116	135
17	34	67	84	117	134
18	33	68	83	118	133
19	32	69	82	119	132
20	31	70	81	120	131
21	30	71	80	121	130
22	29	72	79	122	129
23	28	73	78	123	128
24	27	74	77	124	127
25	26	75	76	125	126

Genetically engineered feral alfalfa plants have been found in conventional hay fields on WSU Roza farm at Prosser, WA where our alfalfa germplasm is regenerated (left). Procedures of preventing transgene-flow have been implemented in our alfalfa regeneration to maintain the genetic integrity. No adventitious presence of transgene was detected from the seeds sampled from the red-circled sentinel plots (center) in our 2017 alfalfa regeneration plots.

Western Regional PI Station (W-006)

A new way to lower *Fusarium* infestation for garlic regeneration



We demonstrated that *Fusarium proliferatum* infestation can be dramatically lowered by planting bulbils instead of seed cloves for regeneration of garlic.

Western Regional PI Station (W-006)

W6 table beet collection diversity



Root flesh color

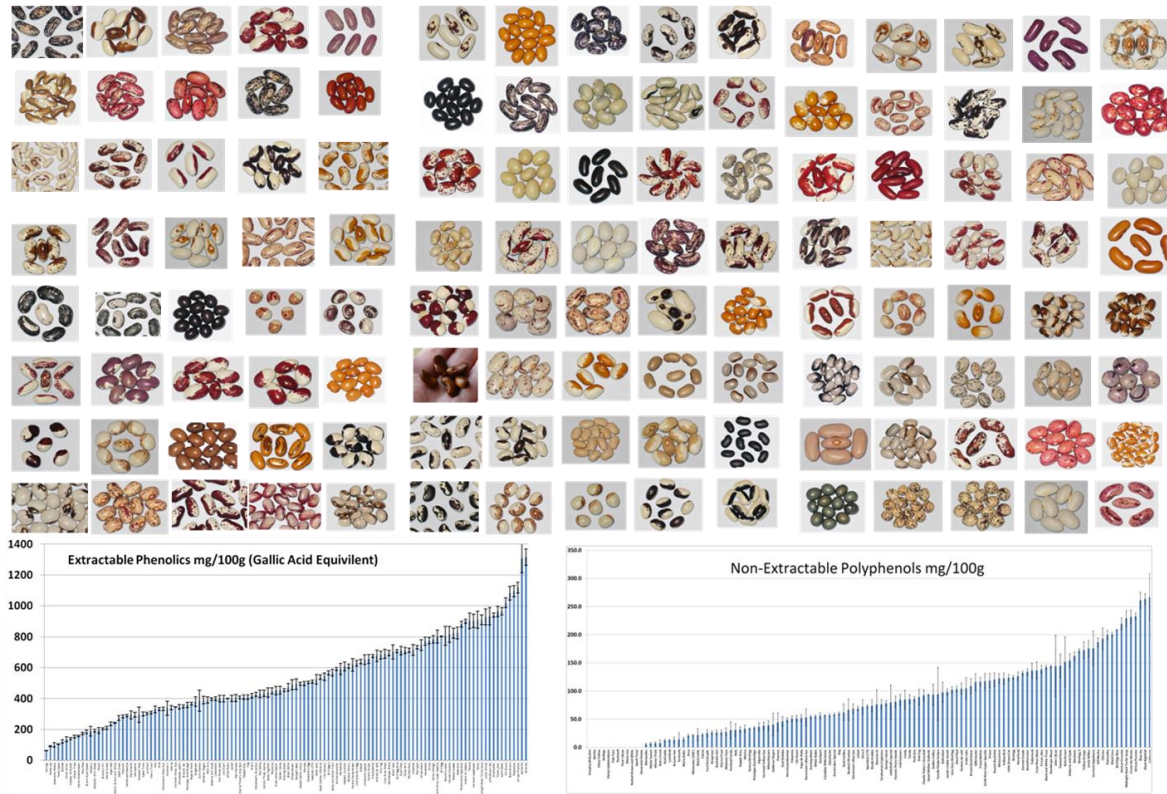


Root shape, leaf density and color



Western Regional PI Station (W-006)

New descriptors for phenolic content in bean accessions



We analyzed 120 Heirloom Beans for protein, extractable phenolics and non-extractable proanthocyanidins. There were 2 to 3-fold differences even within market classes of Red and Black beans. Some of the data has been entered into our GRIN Global database.

Western Regional PI Station (W-006)

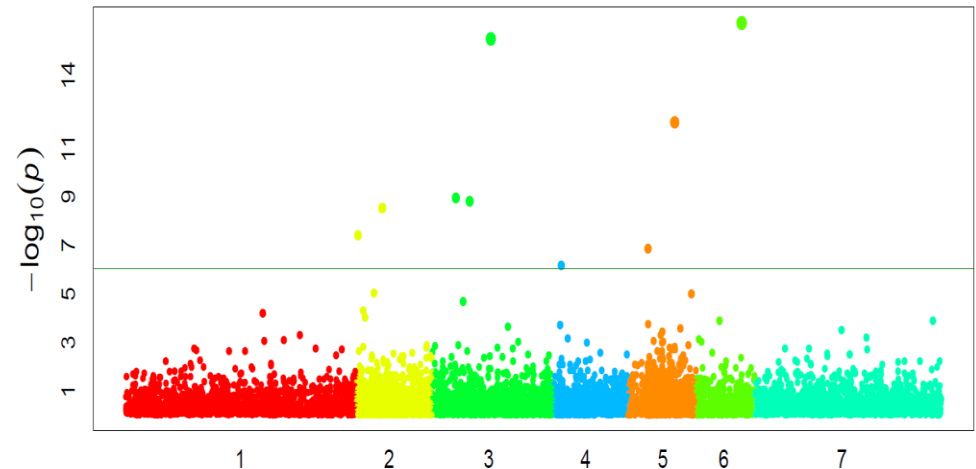
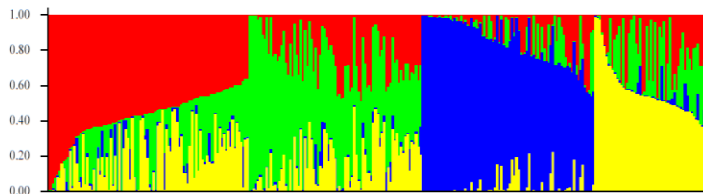
Completed a GWAS with the USDA lentil core collection

- 10,052 SNPs from GBS
- 2 years, 2 locations, RCBD
- Significant markers identified

As a part of the Ph.D. dissertation of Dr. Md Nurul Amin, native of Bangladesh, attending Washington State University.



Daystoflowering



Western Regional PI Station (W-006)

Using solar powered fans in beet pollen proof regeneration cages



Air circulation by the solar powered fan in beet pollen proof regeneration cages can alleviate flower stalk tip die-back.

We used this fan system that is a 10 watt, 12 v solar panel with a dc connection to power a 12 inch high velocity fan at our Pullman Farm for the 2017 field season and will continue to use it in the future.



Western Regional PI Station (W-006)

A new pathogen of stored sugar beet roots

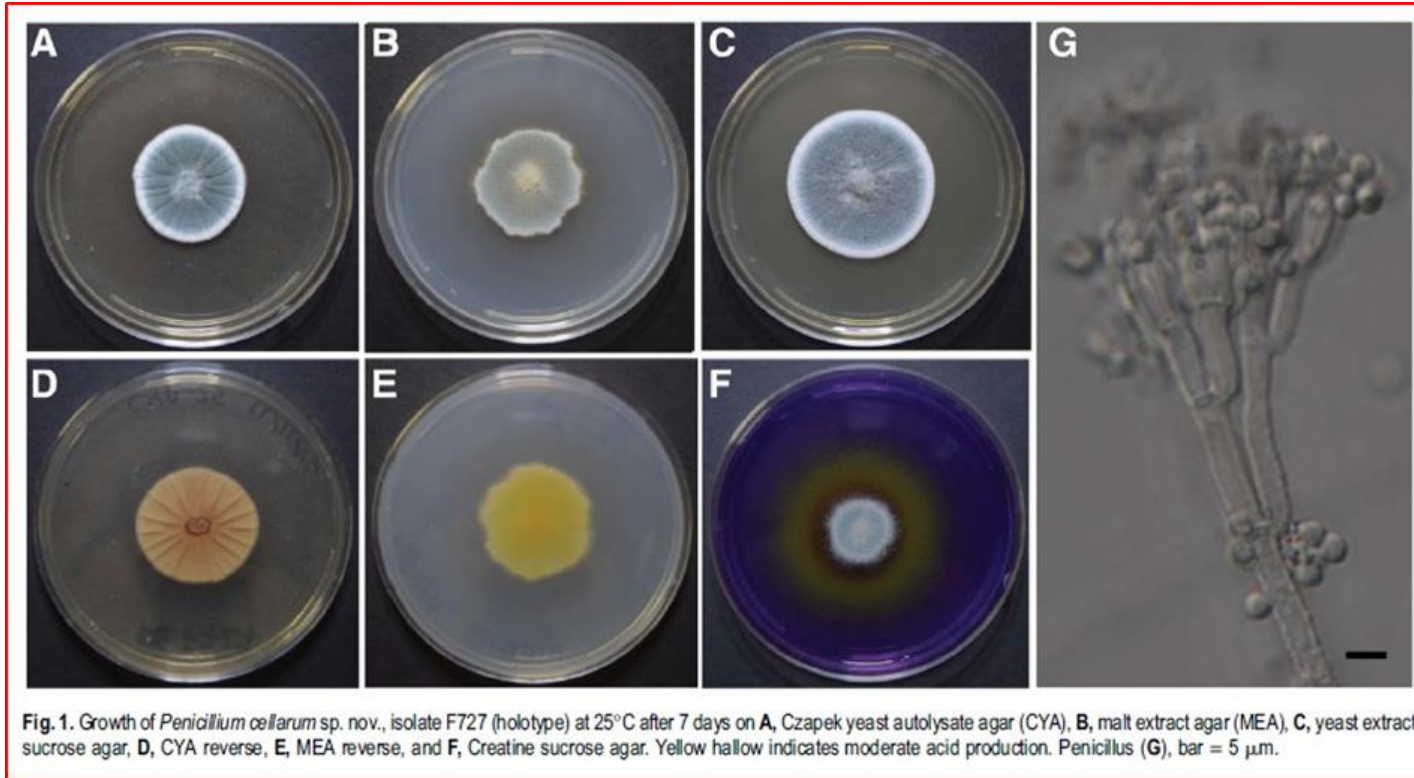


Fig. 1. Growth of *Penicillium cellarum* sp. nov., isolate F727 (holotype) at 25°C after 7 days on **A**, Czapek yeast autolysate agar (CYA), **B**, malt extract agar (MEA), **C**, yeast extract sucrose agar, **D**, CYA reverse, **E**, MEA reverse, and **F**, Creatine sucrose agar. Yellow hallow indicates moderate acid production. Penicillus (**G**), bar = 5 µm.

In collaborating with an ARS scientist at Kimberly ID, we identified a new pathogen of stored sugar beet roots and named it *Penicillium cellarum* sp. nov., together with demonstrating its pathogenicity and environmental preferences.

Western Regional PI Station (W-006)

VIP visit



ARS Associate Administrator Dr. Steven Kappes and ARS PWA Area Director Dr. Robert Matteri visited W6 Seed Storage Facility and the common bean greenhouse on April 20, 2018. We truly appreciate the support from our senior leaders!



NIFA and AFRI Updates NPGCC and NRSP6 meeting May 30, 2018

Drs. Randi Johnson, Ed Kaleikau & Liang-Shiou Lin

USDA NIFA

NIFA programs that support plant breeding/genetic research

- **Competitive programs** (typically \$50M associated with plant breeding)
 - **Agricultural and Food Research Initiative (AFRI)**, main competitive program under NIFA - \$400M (+\$25M)
 - **Non-AFRI Competitive Programs**
 - SCRI: Specialty Crop Research Initiative (\$80M)
 - OREI: Organic Agriculture Research and Extension Initiative (\$20M)
 - SARE: Sustainable Agriculture Research and Education – \$35M (+\$8M)
 - Commodity-specific:
 - Canola (\$0.825M)
 - Potato -\$2.5M (+\$0.25M)
 - Alfalfa (\$2.25M)
 - SBIR: Small Business Innovation Research - ~\$27M

NIFA programs that support plant breeding/genetic research

- **Capacity/formula programs**

(typically about \$20M associated with plant breeding)

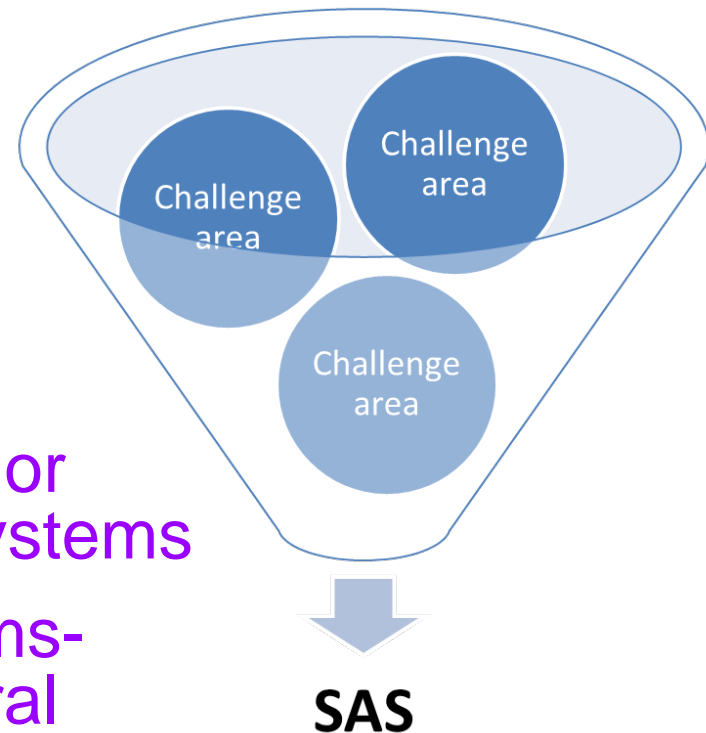
- Hatch (research) - \$244M
- McIntire-Stennis (forestry) - \$34M
- Evans-Allen (1890 institutions) - \$54M
- Smith-Lever (extension) - \$300M

Organization of AFRI

- Aligned with the 6 Farm Bill priorities
- Three Requests for Applications (RFAs)
 1. Sustainable Agricultural Systems
 2. Education and Workforce Development
 3. Foundational and Applied Science

Sustainable Agricultural Systems

- New RFA for 2018
- No more challenge areas – combined here
- Large integrated projects (research, education and extension) that address major challenges of agricultural systems
- Up to \$10 million per systems-level Coordinated Agricultural Project (CAP) grant



Sustainable Agricultural Systems

- 25-Year Goals

- Increase agricultural total factor productivity growth from 1.5% to 2% per year
- Improve water and nitrogen use efficiency by 50%
- Reduce crop losses due to environmental stress and pests, or diseases by 20%
- Produce 50 billion gallons of biofuels and biobased chemicals and bioproducts
- Reduce food-borne illnesses to 8.5 cases/100,000

Education and Workforce Development

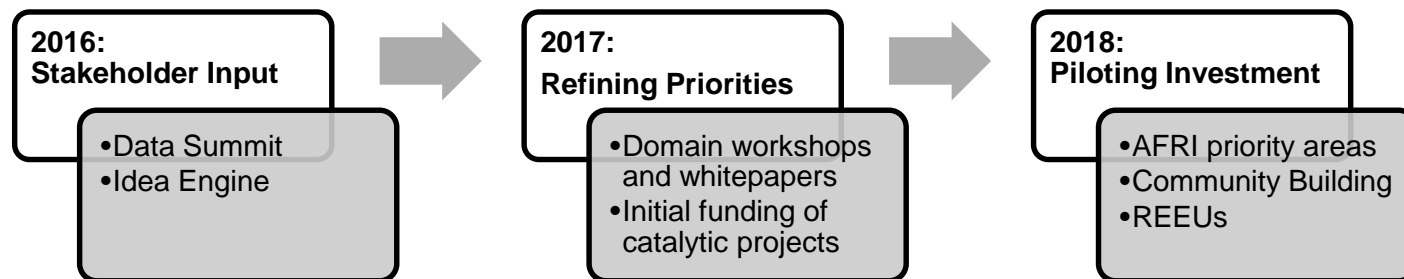
- Postdoctoral Fellowships and Predoctoral Fellowships – individual applies
- Research and Extension Experiences for Undergraduates (REEU) – institution applies for a group of fellowships
- Professional Development for Secondary School Teachers and Education Professionals – training and continued education of K-14 ag teachers

Foundational and Applied Science: Program Areas directly aligned with the Farm Bill Priorities for AFRI

- Plant health and production, plant products
- Animal health and production, animal products
- Food safety, nutrition, and health
- Bioenergy, natural resources, environment
- Agriculture systems and technology
- Agriculture economics and rural communities

What is **FACT**?

- Targeted investments in data science, infrastructure, and cross disciplinary data driven decision making
 - Data-driven advances in agriculture and the food production system;
 - Cross-sector advances in data applications;
 - Data-driven advances to address societal well-being and consumer demands;
 - Data management and application;
 - Developing a data literate workforce and end-user; and
 - Big data in communication, property rights, and communities



What is **Microbiome**?

Goal: Understand the multipartite **interactions** among the host, environment, and the microbiome to help improve and sustain agricultural productivity and quality in plant systems and associated natural resources.

Research will help fill major knowledge gaps in characterizing agricultural microbiomes and microbiome functions across agricultural production systems, and natural resources through **cross-cutting projects**.

- Sustainable Agroecosystems: Functions, Processes and Management
- Bioenergy Feedstock Logistics
- Agricultural Microbiomes in Plant Systems and Natural Resources
- Networks for Synthesis, Data Sharing, and Management

Plant Breeding for Agricultural Production FY 2018 program priorities

- Supports public breeding efforts to improve crop productivity, efficiency, quality, performance, and/or local adaptation. Both conventional/classical and genomics-enabled plant breeding will be supported.
- Applications must address one or more of the following priorities:
 - Pre-breeding and germplasm enhancement
 - Cultivar development
 - Selection theory
 - Applied quantitative genetics and phenomics
 - Participatory breeding

Plant Breeding for Agricultural Production FY 2018 priorities (cont)

- Commodity Board co-funding topics:
 - Kansas Wheat Commission:
 - (1) Develop innovative technologies/approaches to enable wheat breeders to address the prevalence and impact of viruses
 - (2) Utilize technologies/tools/methods to access and mobilize genes within collections of wild wheat species
 - National Peanut Board:
 - (1) Identify genetic markers to develop improved cultivars & breeding lines with superior biotic/abiotic and quality traits
- Conference:
 - Workshop that brings together experts in plant breeding, food safety and related sciences to identify the challenges/opportunities in breeding vegetable crops to decrease bacterial food borne illnesses and enhance food safety

Plant Breeding for Agricultural Production FY 2018 priorities (cont)

- FACT initiative: Proposals on big data analytics and tool development to support a plant breeding data network and cyberinfrastructure should submit a letter of intent to the FACT Program Area Priority.
- Additional information:
 - Release or distribution of germplasm: *Researchers must consult with the relevant National Plant Germplasm System (NPGS) curator to determine whether and how to deposit germplasm, transgenic plants, mutants, plant populations, etc., into the NPGS or stock center. Project directors must confer with the crop curators and crop germplasm committees early in the application development process regarding the desirability of depositing genetic stocks and experimental plant populations generated by NIFA funding in NPGS gene banks. More information is available at www.ars-grin.gov/npgs/index.html.*

AFRI Plant Health and Production and Plant Products (PHPPP)



Examples of projects funded by Plant Breeding for Agricultural Production Program

Project director	institution	Project title
Patrick Byrne	Colorado State University	PLANNING CONFERENCE TO DEVELOP STRATEGIES FOR TRAINING THE NEXT GENERATION OF U.S. PLANT GENE BANK MANAGERS
Peggy Ozias-Akins	University of Georgia	NATIONAL ASSOCIATION OF PLANT BREEDERS ANNUAL MEETING: "SUSTAINING AGRICULTURE THROUGH GENETIC ENHANCEMENT"
Bill Tracey	University of Wisconsin	IPR AND PUBLIC BREEDING: FACILITATING PUBLIC-PRIVATE PARTNERSHIPS
Ethalinda Cannon	Iowa State University	CHARACTERIZING THE USDA PEANUT CORE COLLECTION THROUGH GENOTYPE AND PHENOTYPE INFORMATION

Physiology of Agricultural Plants FY 2018 program priorities

- This program will support projects that use molecular, biochemical, whole-plant, agronomic or eco-physiological approaches to improve plant productivity or performance through studies on:
 - Plant growth and developmental processes
 - Mechanisms of plant response to abiotic stresses, including water use efficiency
 - Photosynthetic efficiency, carbon assimilation, and source-sink relationship
 - Primary and secondary metabolism
 - Nutrient uptake assimilation, accumulation and utilization
 - Harnessing plant biochemistry and biodiversity to develop bio-based plant products.
 - **Investigate potato starch to fill knowledge gaps in understanding the structure of potato starch and interactions associated with desired sensory attributes (National Potato Promotion Board priority)**

AFRI Plant Health and Production and Plant Products (PHPPP)



Examples of projects funded by Physiology of Agricultural Plants Program

Project director	Institution	Project title
Kimberly Novick	Indiana University	Drought impacts on species-specific carbon uptake and growth in Eastern U.S. hardwood forests
Kaiyu Guan	University of Illinois	Parsing Multiple Mechanisms of High Temperature Impacts on Soybean Yield Combining Infrared Heating Experiments and Process-Based Modeling
Jonathan Lynch	Penn State University	Optimizing root metaxylem phenotypes to improve drought tolerance in maize
Endang Septiningsih	Texas A&M University	Enhancement of flooding tolerance : Physiological and molecular characterization of qSUB8.1 and its interaction with SUB1

New Interagency Funding Opportunity



United States Department of Agriculture
National Institute of Food and Agriculture



- **Dear Colleague Letter: NSF-USDA-BBSRC Joint Funding Opportunity - Early Concept Grants for Exploratory Research (EAGERs) to Develop Breakthrough Ideas and Enabling Technologies to Advance Crop Breeding and Functional Genomics**
- **2-page summary due March 14, 2018, Proposals due July 16**

Potential Research Areas:

- Advancing **genome editing** to generate new phenotypes
- Achieving reliable and high throughput production of **doubled haploids** to accelerate breeding
- Controlling and understanding meiotic **recombination** to enable whole genome manipulation
- Modifying **epigenetic inheritance** to facilitate phenotypic changes
- Understanding mechanisms of **heterosis** to exploit hybrid vigor

Non-AFRI competitive programs:

- **Specialty Crop Research Initiative (SCRI)**
 - Research in **plant breeding**, genetics, genomics, and other methods to improve crop characteristics, such as: **environmental responses and tolerance**

- **Organic Agriculture Research and Extension Initiative (OREI)**
 - Facilitating the development and improvement of organic agriculture production, **breeding**, and processing methods.

Non-AFRI competitive programs (continued)

- **Potato Breeding Research**
 - supports potato research that focuses on the development and testing of superior commercial potato varieties using **classical breeding and advanced molecular and biotechnological approaches**
- **Supplemental and Alternative Crops (canola)**
 - **Testing germplasm and breeding** to develop superior performing canola varieties that increase productivity, profitability, and adaptation to an expanded range of U.S. growing environments
- **Alfalfa and Forage Research Program**
 - Increase alfalfa forage and seed yields and forage quality through improved management practices, **plant breeding**, and other strategies to reduce biotic and **abiotic stresses** and costs of production

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ILLUSTRATE

to the American people how research, education, and extension are improving lives.

REPORT

to Congress that NIFA-funded projects are making a difference.

INFORM

state and local governments about the ways their partnership betters their communities.

COMMUNICATE

to partners and stakeholders the opportunities available to educate the next generation.

ENABLE

USDA *Leadership* to make sound decisions regarding mission priorities.

Need for a Plant Genetic Resource Management Training Program

- Estimated 30% of NPGS staff could/will? retire during the next 5 years.
- National Plant Germplasm Coordinating Committee (NPGCC) discussed that during May 2017 meeting.
- Gayle Volk (USDA/ARS) and Pat Byrne (Colorado State) submitted a grant proposal to USDA/National Institute for Food and Agriculture (NIFA) to convene a workshop to discuss how to train the next generation of PGR managers, researchers, etc.



Planning Conference to Develop Strategies for Training the Next Generation of U.S. Plant Genebank Managers

Objectives

- 1) Host a conference to identify the pedagogical options, logistics, and curriculum topics for a U.S. plant genetic resource management training effort, with major emphasis on a distance-learning course.
- 2) Design a strategy to develop, deliver, and sustain a plant genetic resource management training program.

Planning Conference
Co-Hosted by NLGRP and Colorado State University
April 24-26, 2018, Fort Collins, Colorado

<http://genebanktraining.colostate.edu/index.html>

33 Attendees represented USDA-ARS, USDA-NIFA, land-grant universities, seed industry, Mexico's and Canada's genebanks, a botanic garden, and The Crop Trust

Presentations:

- NPGS operations
- Accessibility/utility to customers
- International perspectives
- Distance-learning
- Business models

Breakout sessions:

- NPGS genebank and information management
- Ensuring utility of germplasm and associated information

Proposed Training Program

Goals

- To build and sustain the human capacity to appreciate, maintain and promote utilization of plant genetic resources.
- To educate professionals in principles and practices of genetic resources management, utilization, and conservation.

What's next?

Submitted a Higher Education Challenge Program Planning Grant proposal to USDA-NIFA Planning Activity for a Multi-Institution Genebank Training Program (1 year)

PIs: Pat Byrne (Colorado State) and Silvia Cianzio (Iowa State)

Key Personnel:

USDA: Gayle Volk, Candice Gardner, Stephanie Greene, Christina Walters

Iowa State: Thomas Lubberstedt, Maria Salas Fernandez

Objectives

1. To explore existing PGR educational resources and determine subject areas that are lacking or need enhancement.
2. To investigate alternative administrative frameworks for coordinating training materials developed by multiple universities.
3. To identify the best options for delivering training on specific topics (e.g., online courses, a resource library of “learning objects”, webinars, hands-on workshops).
4. To convene a meeting in February 2019 to decide on an administrative framework, specific content and format of training materials, responsibilities, and budget.
5. To develop a 1-credit online training module on a priority PGR topic.

Plan to submit a multi-year Higher Education Challenge Program Grant in 2019 for implementation

What's next?

- Design a framework for making learning objects available through GRIN-Global
- Develop criteria for learning objects (resolution, quality, etc.)
- Seek learning objects from NPGS sites
- As resources permit, develop learning objects for site activities

Planning Activity for A Multi-Institution Genebank Training Program
Submitted to Higher Education - Institution Challenge Grants Program

PD: Byrne, Patrick F. Institution: Colorado State University
CO-PD: Cianzio, Silvia Institution: Iowa State University

The genetic diversity available through the USDA-ARS National Plant Germplasm System and similar facilities is an essential resource for crop improvement and food security. However, there is a lack of accessible, high-quality training materials on management and use of plant genetic resources (PGR). Our long-term goal is to expand the agricultural workforce that is well trained in subjects relevant to PGR acquisition, preservation, distribution, evaluation, and utilization. The specific objectives of this planning activity are (1) to explore existing PGR educational resources and determine subject areas that are lacking or need enhancement or improved accessibility; (2) to investigate alternative administrative frameworks for coordinating training materials developed by multiple universities; (3) to identify the best options for delivering training on specific topics (e.g., online courses, a resource library of “learning objects”, webinars, hands-on workshops); (4) to convene a meeting in early 2019 to decide on administrative framework, specific content/format of training materials, responsibilities, and budget; and (5) to develop a prototype 1-credit online module on a high-priority PGR topic. Leadership development will be addressed through the participation of two graduate students to aid in the development of engaging training materials. These efforts are in preparation for an HEC Collaborative 2 grant submission in 2019 to complete implementation of the training program. This proposal addresses HEC program goals by enhancing the quality and availability of instruction on PGR for bachelor’s and master’s degree students, and by developing learning materials that appeal to audiences beyond the standard student population in the agricultural sciences.

NAPB Report for NPGCC by David Baltensperger, Liaison with NAPB

<https://www.plantbreeding.org/>

NAPB began as an initiative of the Plant Breeding Coordinating Committee (PBCC). The PBCC (official committee SCC 080) is a forum for leadership, regarding issues, problems, and opportunities of long-term strategic importance to the contribution of plant breeding to national goals. The NAPB is the outreach group that represents plant breeders in federal, state, commercial and non-government organizations.

We strive to inform our members of events and opportunities, and educate the public on what plant breeding is and what plant breeders do.

Our Mission: The National Association of Plant Breeders strengthens plant breeding to promote food security, quality of life, and a sustainable future.

Our Vision: The NAPB works to help create a future in which 1) Strong public and private sectors work independently and together to deliver varieties and improved germplasm to society, 2) The value and importance of plant breeding to food security, quality of life, and a sustainable future are known and appreciated by the public, and 3) Plant breeding is viewed as dynamic, problem solving, and creative. The NAPB intends to become a recognized and valued advocate for plant breeding research and education, helping to guide and implement a cohesive national plant breeding agenda.

Publications: Communication with the public, policy makers, potential students, scientists and additional stakeholders is an important component of the NAPB and PBCC. This is done through publications and documents for communications. We hope to share the excitement, opportunities as well as challenges and needs of plant breeding with you.

NAPB Group Shot UC Davis 2017



The 2018 NAPB Annual Meeting will be held from August 7-10, 2018 at the University of Guelph in Guelph, Ontario, Canada. Please follow this link to the meeting [website](#).

The 2018 National Association of Plant Breeders (NAPB) and Plant Breeding Coordinating Committee (PBCC) Annual Meeting will be held from August 7-10, 2018 at the University of Guelph in Guelph, Ontario, Canada.

Important deadlines:

Early Bird Registration Cutoff: May 18, 2018

Abstract Submission Deadline: **Poster abstracts are still being accepted for the 2018 meetings**

Final Registration Deadline: July 27, 2018

2018 NAPB meeting highlights:

- Reception and banquet featuring local bands
- Pre-conference tour of the Vineland Research and Innovation Centre
- Field Tours at the Elora Research Station
- Poster sessions and 1-minute poster introductions
- ***We look*** Award talks and 2018 awards

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Dave Wolyn

Professor, Univ. of Guelph

dwolyn@uoguelph.ca

Current Membership

Terrence Tiersch (Chair), Louisiana State University Agricultural Center

Paul Gepts, University of California, Davis

Tim Johnson, Smith College Botanic Garden

Sarada Krishnan, Denver Botanic Gardens

Kevin McCluskey, Fungal Genetics Stock Center, Kansas State University

James McFerson, Washington State University Tree Fruit Research and Extension Center

Plus 4 upcoming appointments



THE NATIONAL GENETICS RESOURCES ADVISORY COUNCIL (NGRAC)

NPGCC Meeting
May 30, 2018



What We Do



- **Advise the Secretary of Agriculture and Director of the National Genetic Resources Program (NGRP) on the activities, policies, and operation of NGRP.**
- **Scope includes acquisition, preservation, access, evaluation, characterization, distribution, and exchange of genetic resources of life forms important to American agriculture; plants, forest species, animals, aquatics, insects, and microbes.**

What We Do



- **Currently focusing on crops, animals including aquatic species, tribal issues, and moving into microbial issues.**
- **NGRAC shall make recommendations to ensure that these essential resources are adequately conserved and appropriately accessible in order to address current and future agricultural needs.**
- **NGRAC is also to advise on research needs for genetic resources, on coordination of NGRP with similar domestic activities, and on policies—both international and domestic—regarding access and exchange of genetic resources for the public’s benefit.**



**1st meeting after reconstitution of NGRAC, March, 2013.
Monthly teleconferences and 1 or 2 in-person meetings annually**

- Identified major issues and needs in plant genetic resources
- Strongly encouraged the Secretary of Agriculture to seek ratification of the International Treaty for Plant Genetic Resources for Food and Agriculture by the U.S. Senate
- At the request of Secretary of Agriculture, identified the principal players, problems and solutions at different stages of the seed development process **for response to AC21 recommendations**

Systems Approach for response to AC 21 recommendations



We examined issues at each of the following stages of germplasm use:

- Uncharacterized germplasm for breeding
- Characterized germplasm for breeding
- New inbred lines and varieties in the appropriate form including Foundation seed, which is the first generation multiplication of breeder's seed
- Seed for farmers
- Harvested products for processors and consumers

Crop Focus



We focused on 8 major crops that currently have GE varieties available in the U.S.:

- Corn, Soybean, Cotton, Canola, Alfalfa, Sugar Beet, Squash, and Papaya
- Similar issues apply to future GE crops

Area 1. Ongoing evaluation of the pool of commercially available non-GE and organic seed varieties.

***Recommendation 1** – USDA should encourage and facilitate seed producers to provide information on the available pool of appropriate organic and non-GE seed.*

***Recommendation 2** – USDA should work with plant breeders and other seed providers to increase the availability of organic and non-GE germplasm.*



Area 1. Ongoing evaluation of the pool of commercially available non-GE and organic seed varieties.

Recommendation 3 – USDA should commission a study on the release and availability of inbred lines and varieties developed at public universities in order to determine the extent to which they deliver well adapted crop genetics for different agricultural systems. This should include an assessment of the unintended impacts of the Bayh-Dole Act on public sector capacity to serve all agriculture.



Area 2. Identify market needs for producers serving GE-sensitive markets.

Recommendation 4—USDA should conduct an ongoing economic assessment of non-GE and organic seed markets to allow stakeholders to better understand the value and plan investment opportunities in the seed sector. Market demands for organic and non-GE should be identified by crop for each of the crops affected by commercial GE trait adoption by region, acreage, maturity and adaptation.



Area 3. Ensure that a diverse and high quality commercial seed supply exists that meets the needs of all farmers.

***Recommendation 5**—USDA should convene regular roundtables with balanced representation by all stakeholders on extending GE trait stewardship to encompass prevention and mitigation of adventitious presence in non-GE breeding programs and gene banks.*

***Recommendation 6** – To facilitate coexistence and maintain stewardship, USDA should work with and encourage industry to develop and provide low cost assays of GE traits.*



Area 3. Ensure that a diverse and high quality commercial seed supply exists that meets the needs of all farmers.

***Recommendation 7**—The NGRAC encourages USDA to promote diversity in agriculture by devoting additional resources to genotyping, phenotyping, evaluation, breeding and/or pre-breeding. USDA should facilitate more public, private, and/or tribal partnerships in developing, characterizing, and evaluating genetic resources from the NPGS and non-U.S. sources adapted to U.S. growing conditions. Further assessment is needed for developing, characterizing, and evaluating tribal genetic resources.*



Area 3. Ensure that a diverse and high quality commercial seed supply exists that meets the needs of all farmers.

***Recommendation 8**—USDA should identify gaps in genetic diversity and/or passport information, including samples or accessions with known use restriction issues, and remedy those omissions by additional collection or documentation.*

***Recommendation 9**—USDA should communicate to State seed foundations and the American Seed Trade Association (ASTA) members the importance and need for inbred lines and foundation seeds that are not treated with chemicals prohibited by USDA National Organic Program.*



After response to AC21, the Council has been engaged in thinking broadly about supporting genetic resources infrastructure needs and benefits to users in the following major issues:

- Crop Genetic Vulnerability
- Animal Genetics Conservation
- Aquatic Species Genetic Resources
- Tribal Issues and Genetic Resources
- Microbial Resources



Summary: Actions for addressing crop vulnerability and supporting genetic resources infrastructure.

The NGRAC Crop Vulnerability Subcommittee was formed to recommend how to enhance the baseline crop genetic vulnerability data.

The subcommittee is soliciting input from the respective Crop Genetic Committees in the short-term form of Quad Charts, and in the long-term as traditional reports.



Crop Vulnerability Update for Potato

Vulnerabilities & Threats

- Susceptibility to many current, evolving and emerging pathogens and pests.
- Narrow US genetic base for commercial varieties.
- US production is input-intensive.
- Changing climates: hotter, drier—reducing yields.
- Some crop wild relatives (CWR) endangered in situ.
- Reduced budgetary support and operational capacity for breeding and plant genetic resource (PGR) management.

NPGS PGR Status & Impacts

- **Status:** Large collection (ca. 6000 accessions) with superior representation of CWR, managed as tubers and seeds in cold storage and greenhouses at Sturgeon Bay, WI. Accessions backed up at Ft. Collins and internationally.
- Rigorous disease and quarantine protocols in place, but add time and expense to germplasm import.
- **Impacts:** Protects and genetically improves top US vegetable with ca. \$4 Billion/yr production value, and up to 25 tons/acre state average yields.
- NPGS source of base germplasm for most new US potato varieties; host-plant resistance to many diseases and pests; base genetics for specialty potato varieties.

Genetic research & breeding capacities

- ARS breeding and genetics programs at Beltsville, MD/Orono, ME & Madison, WI. Prosser, WA & Aberdeen, ID collaborative breeding with CO, MI, MN TX, WI state projects; OR, ID, and WA state projects as NW Variety Development Program.
- Very high quality requirements for 30+ traits.
- Cultivar selection lengthy (10+ yr); need rapid reliable disease-free propagation methods.
- Pepsico-Frito and Michigan State provide genetic marker and genotyping-by-sequencing (GBS) data.
- Germplasm evaluations by public & private sectors.

Priority Issues

- Additional budgetary support crucial for expanded NPGS potato PGR management capacity, handling high PGR demand (80% of collection distributed/yr.), and additional PGR evaluations and genomic characterizations.
- Additional budgetary support crucial for expanded potato breeding capacity, especially for host-plant resistance to biotic stresses, tolerance to abiotic stresses, and input use efficiencies.
- Additional CWR should be acquired.
- See https://www.ars-grin.gov/nr6/tac/CGC_PotatoVuln2014.pdf for more info.

Summary: Actions for addressing other areas and supporting genetic resources infrastructure.

The Animal Genetics Subcommittee was formed to address a larger view of the program and its needs. The subcommittee has developed a national summary and set of recommendations addressing the needs and opportunities for animal and aquatic genetic resources. These are scheduled to be reviewed for approval by the NAREEE Board.

NEXT MEETING Proposed to be held in August 2018 to finalize recommendations of these and discuss other major emerging issues



Excellent support is provided by *Ex-Officio* members, additional experts and NAREEE Board:

Michele Esch, Executive Director,
NAREEE Advisory Board

Shirley Morgan-Jordan, NAREEE
Board Program Support Coordinator

Peter Bretting, National Program
Leader, USDA Agricultural Research
Service

Preston Hardison, Watershed
Resource Analyst, Office of Treaty
Rights, Tulalip Tribes

Harvey Blackburn, Coordinator,
National Animal Germplasm Program,
USDA Agricultural Research Service

Ann Marie Thro, Senior Advisor,
Office of the Chief Scientist



National Genetics Resource Advisory Council (NGRAC)



Thank you! Questions?

ARS procedures and best management practices for genetically engineered traits in plant germplasm and breeding lines

v. 73, 14 Feb. 2018

Introduction

In November 2012, the USDA Advisory Committee on Biotechnology and 21st Century Agriculture (AC 21; see the acronym glossary) issued its report “Enhancing Coexistence: A Report of the AC21 to the Secretary of Agriculture.” The AC 21 made three specific recommendations regarding the need for Best Management Practices (BMPs) to monitor and maintain the purity of publicly held plant germplasm:

“For every plant species with commercially available or new GE [genetically engineered] varieties on the market, the USDA should assure that a credible plan is implemented to monitor and maintain the purity of publicly held germplasm. Each plan should include BMPs for maintenance of purity, and should include measures to:

- Determine the presence of plants with the GE trait or traits in publicly held germplasm stocks;
- Conduct ongoing monitoring of unintended presence in germplasm stocks, sufficient to detect any significant increase in its frequency in germplasm and breeding lines;
- Address what to do when unintended GE presence is detected in such germplasm stocks.”

In response, USDA/ARS has updated and refined its procedures and BMPs for maintaining true-to-type plant germplasm samples (also termed “accessions”) and breeding stocks, and for assuring compliance with all relevant regulatory requirements associated with plants incorporating genetically engineered (GE) traits. Furthermore, it is particularly important that such an update occur now, because updated procedures and BMPs should be in place before USDA/ARS begins to incorporate currently proprietary--but soon to be public domain-- varieties or enhanced populations with GE traits into its crop breeding programs and to distribute them from its genebanks. USDA/ARS genebanks and breeders must continue to generate high-quality, true-to-type materials that ensure repeatable research, uninterrupted breeding progress, and maintain the Agency’s scientific reputation. These procedures and BMPs will be subject to periodic reviews and updates.

Background for the updated procedures and BMPs

The updated procedures and BMPs presented in this document were developed by a team of USDA/ARS researchers, germplasm curators, line managers, and National Program Leaders to provide Agency-wide guidance for handling adventitious presence (“AP”, the low frequency, unintentional, and incidental occurrence of unwanted genetic off-types) of GE traits in conventional USDA/ARS crop breeding stocks and germplasm accessions, and also management of USDA/ARS varieties that do incorporate GE traits. They draw on existing guidelines,

practices and procedures to manage material incorporating GE traits and to avoid AP of unintended GE traits, such as those described in BIO 2013; CropLife International 2014; Excellence in Stewardship 2008; and IPGRI 2004. They encompass five major **Elements** for guiding USDA/ARS stewardship of germplasm accessions and breeding stocks.

USDA/ARS germplasm curators, plant breeders, and researchers are responsible for implementing these updated procedures and BMPs. It cannot be over-emphasized that effective implementation of and adherence to BMPs are critical to maintaining the genetic integrity of USDA/ARS germplasm accessions and breeding stocks. Mitigation procedures (see **Element 4** below) are designed to correct inadvertent occurrences “after the fact” rather than to compensate for BMPs that are lacking or not followed scrupulously. Conversely, adhering to BMPs assiduously can enable quality problems to be diagnosed more effectively when they occur and simplify mitigation procedures, including diagnostic seed testing. Furthermore, consultation with local growers, experiment station personnel, and neighboring seed production operations can be critical for successful BMP implementation.

In the context of USDA/ARS genebanks and breeding programs, seed testing (see BMP 2 below) is complicated by numerous technical, logistical, financial and practical challenges. Although USDA/ARS personnel have always aspired to produce accessions and breeding lines as true-to-type as possible, new, sensitive and accurate detection methods for some GE traits necessitate a reconsideration of tolerance levels for “off-types.” This is especially the case for AP of GE traits.

In general, standard seed testing procedures (see **References** at the end of the document) are designed to detect low frequencies of offtypes in large volumes of relatively genetically homogeneous seeds. Doing so requires testing many seeds, e.g., more than 4,700 seeds to detect 0.01% or more AP with a 95% probability. Importantly, at certain stages of development, USDA/ARS germplasm accessions and breeding lines can be highly heterogeneous genetically but encompass few seeds (100 or less). Genebanks often first acquire accessions as samples of fewer than 100 seeds from field collections or donors. Several hundred or fewer seeds might be harvested from seed increases of germplasm accessions or some breeding stocks. Genebanks also distribute such samples to numerous requestors as batches of 100 or fewer seeds.

Such small quantities of seeds restrict the degree of statistical precision and confidence levels of quality tests. Those tests are also usually destructive, and therefore can deplete the already small supply for these genetically diverse and valuable (sometimes irreplaceable) materials. Furthermore, seed testing for AP of GE traits can involve substantial additional costs to the comparatively small operational budgets for USDA/ARS genebanks and breeding programs.

Therefore, weighing those factors, for most cases USDA/ARS proposes testing for a <1% AP tolerance level for GE traits in conventional germplasm accessions and breeding lines as a balance between aspiration and practicality (see additional details under **Elements 1-5** below).

Organization of this document

This document is organized into five major components--termed “**Elements**”—followed by a **Glossary (p. 32)**, **References (pp. 30-31)**, two **Appendices (pp. 33-36)**, and two **Figures (pp. 17-18)**.

Element 1: Well-documented, reviewed, and accessible best management practices (BMPs) for maintaining seed purity in both USDA/ARS breeding and genebank programs (pp. 5-19).

- **Element 1** includes three specific BMPs. More BMPs can be added, as needed, to future versions of the document.
 - BMP 1: Conduct risk analyses of AP in the USDA/ARS National Plant Germplasm System (NPGS) accessions or USDA/ARS breeding stocks. (pp. 5-15) The results of risk analyses for AP are presented for five of the most important major U. S. crops with substantial acreage of varieties with GE traits: alfalfa, cotton, maize, soybean, and sugarbeets. These results comprise the largest section of this document. Risk analyses for additional crops can be added to future versions of the document.
 - BMP 2: Assure genetic integrity. (pp.15-19) Based on the preceding risk analyses, procedures for assuring the genetic integrity of germplasm and breeding stocks are described, including tolerance levels for AP, and seed management practices.
 - BMP 3: Document requirements and procedures. (p. 19) The BMPs must be adequately documented to ensure their implementation and measure their effectiveness.

Element 2: Testing for purity at critical control points (pp.20-22).

- Testing for purity is such an important aspect for ensuring overall seed quality that it is highlighted here as a separate **Element 2**. **Figures 1 and 2** diagram the relationships among the critical control points and testing procedures. **Appendix 2 (pp. 35-36)** includes details for recommended testing and sampling techniques.

Element 3: Mandatory purity testing of new USDA/ARS varieties or enhanced germplasm prior to formal release (p. 23).

- Purity testing before the release of new USDA/ARS varieties or enhanced germplasm is actually a subset of **Element 2**, but it is presented here as a separate **Element 3** to highlight its importance. The procedures diagrammed in **Figures 1 and 2** are applicable to this **Element 3**.

Element 4: Guidelines for mitigating the effects of adventitious presence (AP) of GE traits in USDA/ARS breeding stocks and germplasm accessions (pp. 24-26).

- The recommended steps for mitigating the effect of AP are described under this **Element 4**, and are diagrammed in **Figures 1** and **2**.

Element 5: Communication strategies for disseminating information about USDA/ARS procedures and practices for handling future occurrences of adventitious presence (AP) of GE traits (pp. 27-28).

- The recommended strategies and steps for communicating information about these BMPs and practices, and for future occurrences of AP of GE traits are described in this **Element 5**. **Appendix 1 (pp. 33-34)** contains current contact information for U. S. governmental agencies, and industry and commodity group representatives whom might be consulted, depending on the specific situation.

Element 1: Well-documented, reviewed, and accessible best management practices (BMPs) for maintaining seed purity in both USDA/ARS breeding and genebank programs.

Note: Beyond the BMPs listed below, USDA/ARS personnel must follow any BMPs stipulated by the agencies with regulatory responsibilities for particular GE traits (see list of key regulatory agency personnel in Appendix 1).

The individual BMPs listed below are based generally on Hazard Analysis and Critical Control Point System of Food Safety (HACCP) Principles (see Excellence through Stewardship 2008; CAC 1997; IPGRI 2004. Note: there are no scientifically-documented food safety concerns for deregulated GE traits). In this document, “critical control points” refer to stages in processes or activities with the potential of introducing AP into germplasm accessions or breeding stocks. Current BMPs for AP detection and monitoring methods (Excellence Through Stewardship 2008; USDA/GIPSA 2000; Holden et al. 2010; Redmund et al. 2001), and for genebanks (IPGRI 2004; Kameswara Rao et al. 2006) were evaluated. Various BMPs for breeding major crops were also evaluated to determine applicability to USDA/ARS’s needs, and were adopted, modified, and/or strengthened as necessary to address those needs.

The first two BMPs essentially set out the standards to which USDA/ARS genebanks and breeding programs will adhere. The third BMP describes procedures for documenting that BMPs have been successfully implemented and are followed. These BMPs primarily address the challenge of AP of GE traits in conventional breeding stocks and germplasm accessions, but are also broadly applicable for maintaining all types of plant genetic materials true-to-type. Some of the practices for preventing, detecting and addressing AP in conventional germplasm accessions and breeding stocks differ from those designed for managing germplasm accessions and breeding stocks with GE traits and AP in the latter (see BMP 2 below).

Currently, widely-cultivated commercial varieties of five major crops in the U. S.—alfalfa, cotton, maize, soybean, and sugarbeet--incorporate deregulated GE traits. Consequently, the following risk analyses for AP in NPGS accessions and USDA/ARS breeding stocks focus on the reproductive systems and other biological attributes of these five crops. Notably, for the most part, NPGS germplasm accessions and USDA/ARS breeding stocks for these crops are not genetically engineered, and germplasm accessions were acquired by the NPGS before these crops ever began to be genetically engineered.

BMP 1: Conduct risk analyses of AP in the USDA/ARS National Plant Germplasm System (NPGS) accessions or USDA/ARS breeding stocks

Alfalfa

- Alfalfa is an insect-pollinated, outcrossing perennial crop with hard seeds that persist in the soil--characteristics that increase the risk of AP under certain conditions. The Council for Agricultural Science and Technology 2008 report on gene flow in alfalfa (CAST 2008) stated that pollen-mediated gene flow of GE traits can occur from alfalfa seed fields, hay fields and feral plants, with

rates influenced by physical distance between conventional populations and those with GE traits, synchrony of flowering, and density and species of insect pollinators.

- Gene flow from hay fields cut at 20% bloom (in contrast to the standard practice of cutting at 10% or less bloom) was 0.5% at distances closer than 50 m (the routine Association of Official Seed Certifying Agencies [AOSCA] certified isolation distance), and 0.01% at 100-180 m (Teuber et al. 2007). Because the experimental hay field was surrounded by seed fields, numerous honey bees were present. Actual gene flow will depend on the pollinator species and density, and environmental and topographical features that influence pollinator foraging. Although this study provides a general estimate of gene flow rates, the actual rates might be higher or lower depending on locale-specific factors.
- Gene flow from seed fields (which are usually smaller than hay production fields) was less than 0.5% at 300 m, less than 0.2% at 450 m, and could not be detected at 600 m when leaf cutter bees served as pollinators. Fields pollinated with leaf cutter bees and honey bees (which generally carry pollen farther) averaged gene flows of 2.3% at 50 m, 0.9% at 275 m, 0.6% at 1200 m, 0.2% at 1600 m, and 0.03% at 4800 m (Teuber, 2007). Again, this study provides a general estimate for gene flow rates; actual rates might be higher or lower depending on locale-specific factors. Gene flow from feral plants with GE traits is less likely, considering that feral populations are small and isolated and shed less pollen in total than conventional seed fields. Gene flow can also be seed mediated through seed admixture and volunteer seedlings that germinate from remnant or spilled seed, in fields or along roadsides.
- For USDA/ARS alfalfa breeding programs, the risk of AP from gene flow of GE traits during seed regeneration from hay or seed production fields or from feral plants can be high if seed is multiplied where hay with GE traits and or seed production of alfalfa with GE traits occurs. Standards and BMPs developed by the National Alfalfa and Forage Alliance (<https://www.alfalfa.org/CSCoexistenceDocs.html>) and AOSCA (http://ccia.ucdavis.edu/Certification_Programs/Identity_Preserved/Alfalfa_Seed_Stewardship_Program/) for producing AP-sensitive seeds should be followed in those cases.
- Most of the alfalfa seed grown in Washington State is located near Walla Walla, about 80 miles east of the WSU-IAREC station in Prosser, WA where the NPGS's alfalfa germplasm is regenerated. Orchards and vineyards surround Prosser, and alfalfa hay is also widely grown, including a substantial amount on the WSU-IAREC station. Although GE alfalfa hay is not grown on the station, AP was detected in seedlings harvested from WSU-IAREC hayfield edges. In 2011, AP was detected in two fields, but in 2012 AP was detected in 14 fields. These results indicated that the GE trait is present in

close proximity to the alfalfa germplasm collection regeneration site. Following routine BMP, such as insect-proof caging, will lower the risk of AP as a result of seed regeneration, but routine maintenance of germplasm requires cages to be periodically opened. To reduce the risk further, the alfalfa germplasm regeneration site could be relocated to the USDA/ARS genebank site at Central Ferry, Washington, which is surrounded by dry land wheat cultivation, with little alfalfa production.

- The NPGS's genebank collection of alfalfa does not include any varieties with GE traits at present. The greatest risk of AP to NPGS germplasm accessions and to USDA/ARS alfalfa breeding stocks is from incoming materials from the public-sector. AP has already been identified in a standard alfalfa check variety produced in California by a public-sector breeding program. Industry is now routinely testing standard check varieties for AP before they are released for distribution. Strict adherence to BMPs established for AP-sensitive seed should minimize the risk of AP in USDA/ARS germplasm accessions and breeding stocks.
- To summarize, the risk of AP in NPGS alfalfa germplasm accessions and USDA/ARS breeding stocks as they are currently curated is currently probably low. Prominent risk factors for AP in NPGS alfalfa germplasm accessions and breeding stocks include:
 - Failure to adhere to BMPs;
 - Incorporation of new sources of germplasm, especially from public-sector programs, with AP of GE traits into the genebank collection or breeding program;
 - Insufficient spatial isolation of breeding plots from commercial production fields of alfalfa with GE traits;
 - Inadequate testing for AP.

Cotton

- Cotton is primarily self-pollinated, but with variable rates of cross-pollination from insects. Because of its size and morphology, cotton pollen is not dispersed by wind. Gene flow in cotton via pollen has been extensively investigated, and appears to be dependent on environmental factors and the presence of pollinator insects. Pollen-mediated gene flow under field conditions in California has been reported to decrease exponentially from 7.7% at 0.3 m to less than 1% beyond 9 m (Van Deynze et.al, 2005). With fewer active pollinators, gene flow was less than 1% beyond 1 m. In another investigation of gene flow in cotton, outcrossing decreased from 5% to less than 1% as distance increased to 7 m (Umbeck et al., 1991). The preceding data agreed well with results from China where gene flow of the Bt transgene via pollen ranged from 8.16% at 1 m to 0.08% at 20 m from the plants (Zhang

et.al, 2005). Similarly, research in Arizona determined that field-to-field gene flow via pollen was relatively rare (0.23%) as compared to seed-mediated gene flow (Heuberger et.al, 2010). The preceding investigation found that fields of cotton with GE traits located within 750 m of a field of conventional cotton best explained observed outcrossing rates. Nonetheless, Heuberger et al. 2010 (p. 7) study also noted that “although seed-mediated gene flow has received less attention than pollen-mediated gene flow in the literature, it was clearly the most prominent source of cry1Ac transgene flow...”

- The indeterminate perennial flowering habit of cotton is a qualifying factor to the limitations on gene flow imposed by distance. With blooming and pollination occurring continuously over a period of 60-70 days, cotton is available to pollinators and the potential for out-crossing for that entire period. Although most cotton seeds are set during the first 30-50 flowering days, AP can occur over the entire 60-70 day period.
- Many public and private-sector cotton breeding programs utilize pedigree breeding procedures that create large segregating populations of open-pollinated plants for individual plant selection. Due to the indeterminate flowering habit of cotton, these generations of open-pollinated populations and their resulting progeny (if not tested for AP) are prime avenues for the inadvertent introduction of GE traits.
- Although the extended period of flowering in cotton does moderate the effectiveness of distance in reducing gene flow, the use of stringent spatial isolation is effective. State certification and regulatory entities have chosen to enforce stringent spatial isolation criteria for production of foundation and registered seed. In Arizona the minimum mandated isolation distance between fields containing different cotton species (i.e., Pima vs. Upland), or between varieties differing substantially in leaf type, is 1320 feet (Arizona Crop Improvement Association, 2015). In California, fields producing Foundation or Registered seed must be isolated by at least 1320 feet from any other variety of a similar cotton type (California Crop Improvement Association, 2000). Whenever possible, adoption of the 1320 foot (402 m) isolation distance would be prudent for the stages of breeding programs involving open-pollinated plants, and for increasing the quantity of seeds for cotton breeding stocks or genebank accessions when enforced self-pollination is not possible.
- BMPs for reducing the risk of AP are well-understood by USDA/ARS and university cotton breeders, but can be difficult to implement. Breeders often operate on university or state experiment station facilities that lack adequate isolation from commercial fields of cotton with GE traits. Experiment station facilities sometimes grow cotton with GE traits as a source of revenue, but more often cotton with GE traits is grown on stations in tests associated with cotton breeding (e.g., commercial variety tests) and tests associated with other

disciplines (e.g., entomology, plant pathology, weed science, etc.). The maintenance of bee populations on public research facilities for the purpose of pollinating other crops also increases the risk of AP in cotton.

- Under prevailing weather and soil conditions in the southern U. S. (especially in the Southwest), cotton seed can successfully survive and overwinter in the field. “Volunteer” plants with GE traits from prior cotton crops can lead to AP if BMPs for identifying and removing such plants are not followed. Phenotypically, many “volunteer plants” are indistinguishable from the current cotton crop and cannot be visually identified and removed. Currently the most feasible BMP for alleviating volunteer introduction of GE is the practice of field rotation, with cotton being planted in alternate years, i.e., a two year rotation.
- Plants with GE traits can rarely be visually identified and therefore traditional means of assuring trueness-to-type through removal of off-type phenotypes are ineffective. Testing for AP of plants with GE traits is expensive in general, and especially so for cotton because of the commercial availability of multiple GE traits requiring assays at critical control points throughout the breeding process (see “Testing”).
- To mitigate the possibility of AP, seed supplies for NPGS cotton germplasm accessions have been multiplied under strictly-controlled, mechanical forced self-pollination which minimizes the risk of AP. The primary location for seed multiplication in Tecoman, Mexico was geographically-isolated by several hundred miles from commercial production of cotton with GE traits, and had the capacity for forced self-pollination. Nonetheless, that location was closed recently and a comparable alternative site has not yet been identified.
- At present, no cotton germplasm accessions with GE traits are being multiplied or distributed from the NPGS genebank collection, obviating the current need for special isolation, mechanical cleanup procedures, or other practices.
- Under current conditions, the risk of AP in the NPGS cotton germplasm collection is almost entirely from germplasm acquisition: most frequently from improved germplasm donated to the collection, less often from germplasm exchanges, and rarely from collecting new materials. Accumulating evidence indicates that the risk of AP via germplasm acquisition is moderately high.
- To summarize, the risk of AP in seed stocks from USDA/ARS and other public-sector cotton breeding programs is moderate to high. At present, testing programs are detecting AP in 10-15% of the samples assayed. AP in NPGS cotton germplasm accessions is low to moderate, depending on the type and origin of the germplasm. Prominent risk factors for AP in cotton include:

- Failure to adhere to BMPs;
- Incorporation of germplasm containing AP, especially from public-sector sources, into breeding programs or genebank collections;
- Lack of sufficient spatial isolation of breeding stocks from commercial production fields of cotton with GE traits;
- Relatively frequent occurrence of cross-pollinating insects;
- Mechanical mixing caused by inadequate cleaning of pickers, harvest stacks, gins, seed cleaning and delinting.
- Relatively long flowering period for cotton plants;
- Lack of appropriate crop rotation in breeding nurseries leading to the occurrence of “volunteer” cotton plants with GE traits;
- Cotton breeding procedures that involve generations of open-pollination;
- Inadequate testing for AP.

Maize

- Maize is a cross-pollinated, monoecious plant with separate flower structures on the top (male-tassel) and middle (female-ear) of the plant, producing numerous pollen grains that are readily dispersed primarily by wind, but also by insects, or mechanical means. USDA/APHIS guidelines for spatial separation by about 660 ft (200 m) apparently are highly effective for preventing unwanted geneflow (Burriss, 2001a, b; Ma et al., 2007; cross-pollination frequency in maize is generally <1% beyond 30 m spatial separation; the results of research by Goggi et al. 2006 and Ireland et al. 2006 confirmed that those current practices maintain a purity level of 99% true-to-type, yet unwanted cross-pollination can still occur at longer distances). Nonetheless, the many acres planted to commercial maize hybrids near the USDA/ARS NPGS maize genebanks at Ames, IA and Urbana, IL present particular challenges with respect to AP because of the increased risks of unwanted cross-pollination.
- Maintaining maize true-to-type in NPGS genebanks and USDA/ARS maize breeding programs depends on well-defined controlled pollination methods that exclude unwanted pollen, and other best management practices. Maize populations are maintained by manually-performed sib-mating of plants, and inbreds are maintained by self-pollination. New breeding or testing materials are generated by cross-pollination, either through isolation plantings with designated rows of a “pollen parent” line and detasseled rows of “ear parent” lines, or through manual pollinations. Maize is sold commercially as a hybrid between two or more inbred lines, and the hybrid vigor is usually sufficient to identify off-types resulting from cross-pollination with commercial maize during seed increases of inbred lines. Off-type plants are removed prior to flowering to eliminate the risk of cross-pollination.
- Crop rotation is usually effective in eliminating the risk of AP caused by “volunteer” maize plants that persist in a field over years. Fortunately, maize

kernels readily germinate the following season, or very soon after contact with moist soil, unlike seeds of some other crops that might persist in the soil for several years.

- In commercial maize breeding programs, new lines with GE traits are commonly first derived by traditional breeding methods and then “converted” to a version with GE traits by backcrossing with a line with GE traits. The latter line with GE traits may also be used directly in breeding programs, a “forward breeding” method (Mumm 2007). Care is taken to preclude AP in developmental or propagation nurseries through temporal differences in flowering and/or spatial separation, because eliminating off-types based on visual differences between conventional versions of a line and versions with GE traits can be unreliable.
- Because maize is both a major crop and a “model species” for basic plant science research, regulated GE traits used only for research present a special risk for AP in NPGS maize germplasm accessions or USDA/ARS breeding stocks. Such GE traits are regulated by APHIS and must be grown with APHIS permits under conditions that minimize the possibility for gene transfer and persistence in the environment, and ensure that unwanted seeds are devitalized. USDA/ARS genebanks require the APHIS permit number from potential recipients before distributing such regulated materials.
- AP of regulated GE traits in maize is much less likely to occur than with deregulated traits because plants with those traits are grown on very small acreages. On the other hand, AP of regulated GE traits is potentially very difficult to detect because of tremendous number and variety of regulated GE traits, and the lack of readily available testing/detection procedures. Researchers should notify genebank personnel and other researchers whenever stocks with regulated GE traits are under cultivation.
- AP from deregulated traits can be readily detected by standard test procedures.
- To reduce the risk of AP, field evaluations of maize acquired from external sources should be conducted in locations with effective temporal or spatial isolation from seed propagation fields.
- To summarize, the risk of AP in maize seed stocks from USDA/ARS maize breeding programs is moderate to high, and probably low to moderate for NPGS maize germplasm accessions. Prominent risk factors for AP in maize include:
 - Failure to adhere to BMPs;
 - Initial incorporation, from external sources, of germplasm with AP into NPGS maize genebank collections or USDA/ARS breeding programs;
 - Maize “volunteer plants” with GE traits from the previous growing season;

- Insufficient spatial or temporal separation of seed increase or breeding plots from commercial maize;
- Possibility of AP or seed admixture of materials with regulated GE traits from genetic experiments;
- Inadequate testing for AP.

Soybean:

- Soybean is self-pollinated, with pollination often occurring before the flower opens, so cross-pollination is rare. Soybean pollen is not windborne, but cross-pollination can occur through insect vectors. Relatively few studies of soybean pollen movement have been published, and most of that research has focused on pollen movement to male-sterile plants where self-pollination is not possible. In central Illinois, when male-sterile plants were grown in commercial soybean fields within 15 cm of fertile plants, 75% of the male-sterile plants were barren and the average seed set per plant was fewer than 3 seeds. Under the same experimental design, experiments conducted in southern Illinois, resulted in only 5% barren plants and average seed set was 40 seeds per plant (Nelson and Bernard 1984).
- Those results highlight the importance of insect vectors to gene flow via pollen, with cross-pollination rare in the cultivated landscape of central Illinois but more frequent in southern Illinois where the insect vectors are more numerous. For soybean, pollen movement differs within and across rows and, as with other crops, distance from the pollen source does affect the frequency of cross pollination in soybean. In Georgia where insect vectors are common, pollen transfer to male-sterile plants from source plants located farther than 7 m between rows and farther than 12 m within rows averaged 0.4%. (Boerma and Moradshahi 1975).
- The experimental results from pollen-sterile soybean plants represent the worst case scenario for self-fertile germplasm accessions and breeding stock because they estimate pollen movement to flowers without pollen. The results indicate how far pollen is likely to move but with self-fertile plants, which are our focus, the stamens shedding pollen are about a millimeter away from the stigma and enclosed by petals. Research conducted in Japan detected no pollen movement from wild soybeans to nearby domesticated soybeans. But within wild soybean populations outcrossing rates averaged 2.2% with an average distance of 10.5 m for pollen movement (Kuroda et al. 2008).
- Accessions from the USDA/ARS soybean germplasm collection are increased at three locations depending on their zone of adaption. Those adapted to northern North America are increased at Urbana, IL in 4 row plots with seeds harvested only from the two center rows. No plants with GE traits are grown in the same field with germplasm accessions. Wild soybean accessions are grown in Urbana

inside insect-proof cages to protect the plants and also exclude any insect pollen vectors. Those soybean accessions adapted to the southern U.S. are increased at Stoneville, MS following the same procedures as in Illinois. Insect pests for wild soybean do not occur in Mississippi so the protection of a screen cage is not needed. Accessions adapted to tropical regions are grown in Costa Rica where no plants with GE traits are allowed.

- All accessions introduced from other nations into the USDA/ARS soybean germplasm collection are pure lined. Depending on the level of inbreeding within the acquired seed sample, each accession is largely homozygous and homogeneous. Approximately a dozen, mostly qualitative, visual traits are assayed to confirm genetic integrity for each accession every time it is increased. Plants or seeds that do not match all of the descriptors of an accession are discarded.
- Because cross-pollination is infrequent in conventional soybean breeding, the distances between breeding plots is not a significant factor for maintaining genetic integrity. When lines with GE traits are incorporated into breeding programs even very low levels of offtypes can be significant, so spatial separation of breeding plots is considered. Admixture between seed lots is also a major concern in this case, so managing the non-harvested areas around seed increase plots is standard procedure. Ensuring that all planting, harvesting, and seed cleaning equipment is free of seeds from one lot before a second lot is processed is also standard procedure. Differences in flower, pubescence and hilum color traits, and other morphological differences serve as markers for monitoring genetic integrity.
- With soybean, the greatest risk for AP results from incorporating germplasm with AP from other programs into the NPGS genebank collection or USDA/ARS breeding programs. Lines obtained from programs that are actively incorporating GE traits present the greatest risk, but lines from other programs that could be exposed to AP of plants with GE traits need to be carefully monitored as well. Accidental cross pollination from lines with GE traits that are not a part of USDA/ARS programs into USDA/ARS-held germplasm must be considered.
- To summarize, the risk of AP in soybean seed stocks from USDA/ARS soybean breeding programs is low to moderate, and probably low for NPGS soybean germplasm accessions. Prominent risk factors for AP in soybean include:
 - Failure to adhere to BMPs;
 - Incorporation, from external sources, of germplasm with AP into NPGS soybean genebank collections or USDA/ARS breeding programs;
 - Insufficient spatial or temporal separation of seed increase or breeding plots from commercial soybeans;
 - In the future, AP from admixtures of seeds with GE traits with conventional seeds will become an important risk when soybeans with GE

traits are incorporated into the USDA/ARS soybean breeding programs and the NPGS soybean genebank;

- Inadequate testing for AP.

Sugarbeet:

- Sugarbeet is a wind-pollinated crop with documented gene flow frequency of 0.15% between plants 1 km apart (Alibert et al 2005). The risk of AP for USDA/ARS germplasm accessions regenerated in Pullman, WA is negligible because sugarbeets are not grown commercially in eastern Washington State and commercial seed production is primarily in the Willamette Valley in Oregon several hundred miles away.
- The greatest risk of AP in the NPGS's germplasm accessions stems from recently acquired germplasm that has not been maintained according to BMPs.
- The risks of AP in USDA/ARS breeding stocks are small but not negligible, and are primarily associated with breeding stocks acquired from external sources, because no USDA/ARS breeding program currently incorporates deregulated GE traits. The vast majority of commercial seed sold in North America incorporates GE traits. Consequently, open-pollinated seed production fields within pollinating distance of commercial sugar crop production fields with "bolting beets" constitutes one possible risk for AP. Pollination of genebank samples and breeding stock is usually controlled by isolation chambers and/or isolation tents in the field that exclude or filter out pollen on the order of 10 μ m in diameter.
- Other risks include mixing roots with GE traits from "check varieties" with USDA/ARS breeding stocks before seed multiplication, or pollen flow in isolated plots where commercial seed producers multiply seeds for USDA/ARS breeding programs.
- The risk of AP to USDA/ARS germplasm collections from non-USDA/ARS public-sector breeding programs would likely stem from public-sector breeding programs for vegetable beet, because USDA/ARS currently conducts all U. S. public-sector sugarbeet breeding. Gene flow from vegetable beets is a lesser risk because vegetable beet roots to be used for seed production are morphologically distinct from those of sugarbeet so that hybrid off-types would be easily recognized. Another factor reducing AP risk is that vegetable beet seed production is often geographically isolated from sugarbeet seed production, although hundreds of acres of vegetable beet seed production do occur in the Willamette Valley, OR, and pollen from these beets, at times, can cross-pollinate sugarbeet seed production fields. Greater attention has been placed on minimizing gene flow of this sort following the recent deregulation of sugarbeets with GE traits for commercial production. (APHIS Final Impact Statement 2012 for glyphosate-tolerant sugarbeet, http://www.aphis.usda.gov/brs/aphisdocs/03_32301p_feis_std.pdf).

- To summarize, the risk of AP in sugarbeet seed stocks from USDA/ARS sugarbeet breeding programs is low to moderate, and probably low for NPGS germplasm accessions. Prominent risk factors for AP in sugarbeets include:
 - Failure to adhere to BMPs;
 - Initial incorporation, from external sources, of germplasm with AP into NPGS sugarbeet genebank collections or USDA/ARS breeding programs;
 - “Volunteer “ sugarbeet plants with GE traits from the previous growing season;
 - Insufficient spatial or temporal separation of seed increase or breeding plots from commercial plantings of sugarbeets and/or failure to control pollination via isolation chambers or tents;
 - Inadequate testing for AP.

BMP 2: Assure genetic integrity

The procedures for assuring the genetic integrity of the products of USDA/ARS’s breeding programs and germplasm accessions are based on numerous factors, including those considered in the previous risk analyses, and included in references such as BIO (2013). Such factors are summarized in the following bulleted list, which is not all inclusive; **Element 2** and **Figures 1** and **2** of this document includes information on procedures for sampling and testing for AP, especially at critical control points.

- The reproductive biology and breeding system of the crop (See BMP 1 above);
- Degree of genetic improvement (i.e., enhanced genetically segregating population vs. synthetic population vs. inbred line);
- When the accession or breeding stock was acquired by USDA/ARS, and its prior history;
- Certification standards for the particular crop;
- Whether GE traits are regulated or deregulated, proprietary or non-proprietary;
- Specific national regulations or phytosanitary requirements of international germplasm recipients;
- Technology and methods for testing (also see **Element 2**);
- Resources available for testing (also see **Element 2**);
- The different processes required to maintain true-to-type germplasm with GE traits versus conventional germplasm.

As mentioned above under “Introduction,” with respect to genetic integrity, USDA/ARS NPGS genebanks seek to deliver accessions that are true-to-type. Taking into account the preceding factors, in general NPGS genebanks aspire to zero tolerance of AP, but test for a <1% tolerance level of AP within the practical constraints described below (see **Element 2**) for alfalfa, cotton, maize, soybean, and sugarbeet. For USDA/ARS breeding stocks of these crops, the acceptable AP tolerance level is <1%, and testing is designed accordingly to detect that frequency within those practical constraints (also see **Elements**

2, 3, and 4). Notably, the terms “none” or “zero tolerance” do not imply that the material is completely free of GE traits, rather, that no GE traits were detected via standard testing protocols.

The <1% AP tolerance level was chosen to enable the genetic purity to be maintained in germplasm accessions and breeding lines without depleting, through testing, the typically small seed samples (often 100 seeds or fewer available for testing) from the seed population. In general, testing will be conducted on sample sizes that will detect at least 1% AP with 95% confidence. Seed populations that are smaller than usual would necessitate smaller sample sizes for testing (so as not to deplete the seed supply). When testing of pooled samples of seeds cannot detect at least 1% AP, an alternative sampling protocol will be devised, in consultation with statisticians and seed testing experts. Under some scenarios, pre-emptive mitigation steps might be required to increase the number of seeds available for testing. Conversely, for larger populations of seeds and for certain purity requirements (e.g., certification standards), sampling can sometimes be increased and the detection level lowered.

Individual NPGS genebanks and USDA/ARS plant breeding programs and/or projects will develop and adhere to detailed program-, project-and/or crop-specific BMPs for maintaining genetic integrity of their germplasm and breeding stocks. The BMPs will take into account the principles included in this document, especially the crop-specific risk factors (see BMP 1) and tolerance level listed above, and incorporate the following major aspects:

- Determining the genetic integrity of incoming germplasm by analyzing its accompanying documentation and/or by conducting tests for AP, when warranted (see **Figs. 1, 2, and Element 2**);
- Implementing procedures for avoiding seed admixture when handling, storing, and distributing germplasm, such as clear and durable labelling and packaging, accurate recording and transfer of information, and proper operation of seed cleaning and handling equipment;
- Conducting propagation and seed increases with crop rotation to avoid volunteer plants, optimal distances and buffer and/or sentinel strips between plants and between seed increase plots and other fields to avoid pollen flow, controlled pollinations (e.g., insect cages, hand-pollinations), and removing off-type plants before flowering;
- Breeding procedures that include tests for AP at key steps such as new parental lines prior to making initial crosses, materials destined for counter-season nurseries or for regional, multi-institutional performance trials.
- Harvest and seed processing methods that avoid seed admixture and retain identification of specific plots, rows, and/or fields to maintain trueness-to-type and facilitate testing for AP, when warranted (see **Figs. 1, 2, and Element 2**);
- Incorporating the correct labelling, testing, certification, and packaging procedures for shipping and distribution of germplasm internationally, in compliance with international regulations.

Figure 1: Decision tree and critical control points for assessing probability of and testing for AP in NPGS genebank accessions. Blue = processes for determining/confirming AP and maintaining true-to-type. Yellow = action needed. Orange = potential critical control points for AP testing. Red = processes associated with AP-positive findings and mitigation of AP. Purple = processes associated with AP-negative findings.

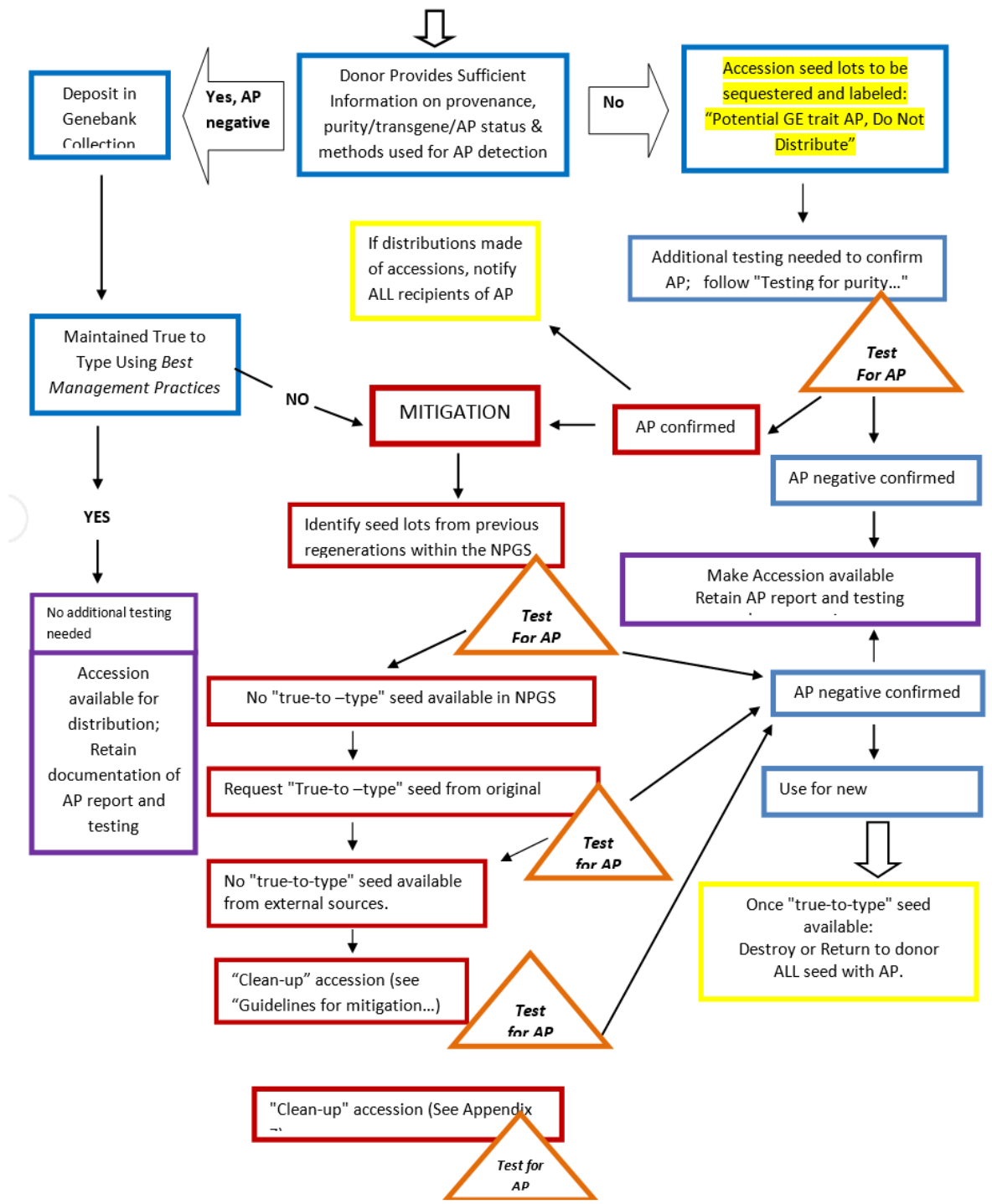
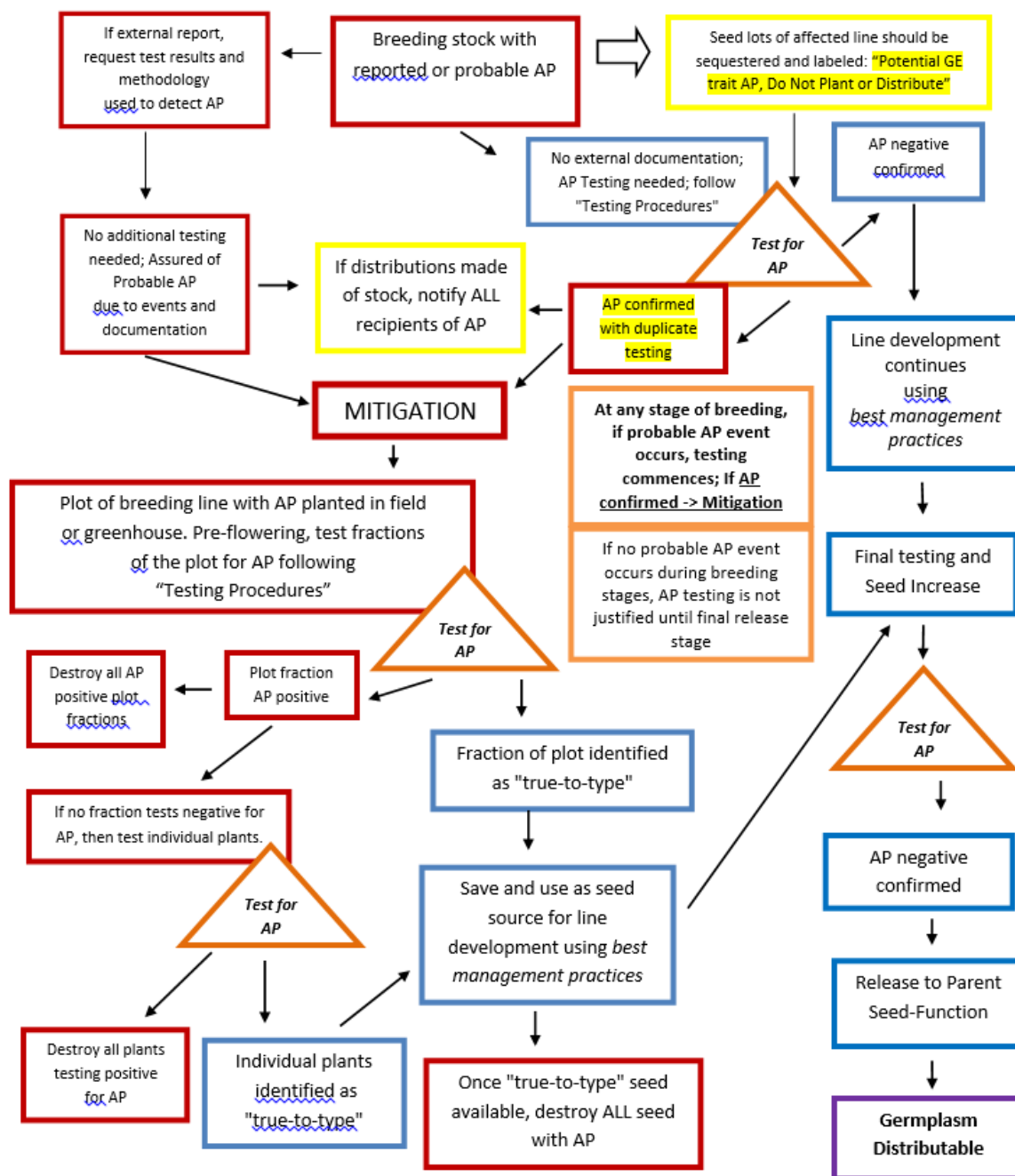


Figure 2: Decision tree and critical control points for assessing the probability of and testing for AP in USDA/ARS plant breeding programs. Blue = processes for determining/confirming AP and maintaining true-to-type. Yellow=action needed. Orange=potential critical control points for AP testing. Red= processes associated with AP-positive findings. Purple=process associated with AP negative germplasm.



Maintaining genetic integrity when NPGS genebank collections and USDA/ARS breeding programs incorporate both materials with GE traits and conventional accessions or breeding stocks requires additional procedures listed below, because the risk of AP through seed mixtures increases. Keeping plants with GE traits separate from conventional materials either spatially or temporally throughout the management process can be an important overall practice for maintaining genetic integrity.

- Clear, distinct labeling with a standard format that distinguishes material with GE traits from conventional material in databases and on all packets, containers, tags, etc., used for regeneration or research activity.
- Separate fields or growing environments. If possible, field borders would be planted with conventional materials to reduce the possibility of seed admixture.
- Separate planting, harvest, seed processing, and storage regimen for conventional materials, and materials with GE traits, which might include different cleaning and processing equipment and/or procedures.
- The more complicated the cleaning and processing equipment, the higher the possibility that seeds from one lot will remain in the equipment and contaminate a subsequent seed lot. Extra caution is needed when monitoring the equipment to ensure that all seeds are removed between processing each seed lot.
- Separate storage areas might be designated for materials with GE traits to avoid mistakes in identifying seed lots, especially in breeding programs where individual seed lots may be handled many times between harvesting and planting.

BMP 3: Documentation requirements and procedures

Documentation for the adoption of BMPs is key for verifying their successful implementation. To ensure that such BMPs are actually implemented, periodic external (to the USDA/ARS genebank or breeding projects) physical inventory checks should be conducted. Such checks could be conducted by non-USDA/ARS personnel, or staff from other USDA/ARS genebanks or breeding projects. The records, which would be maintained in GRIN and/or in local genebank databases, should include:

- Complete, thorough, written BMPs that address critical control points.
- Documentation that crop-specific BMPs are in place and followed for each breeding program and genebank, to ensure genetic integrity of accessions are maintained (also see **Elements 2 and 4**).
- Documentation that personnel have been trained in BMPs.
- Documentation that an external (to the particular genebank or breeding program) process is in place for determining if BMPs are followed.
- Documentation that BMPs have been evaluated for their effectiveness.
- Documentation for potential corrective actions when AP or misidentification of breeding stocks or germplasm accessions occurs (also see **Element 4**).
- Documentation of actions when recipients report AP (also see **Elements 4 and 5**).

Element 2: Testing for purity at critical control points.

Plant breeding programs and genebanks follow some different management practices because of the different nature of their products and the role that these products play in conservation, research and breeding. Considering the different management practices, testing for AP of genetically engineered traits must balance the precision desired for detection with resources available for testing. Levels of AP that are detected should be reported through approved, standard methods (see **Element 5**). Testing of new varieties and enhanced germplasm released by USDA/ARS is mandatory under certain conditions (see **Element 3**). The testing results will be documented in GRIN and in local records.

- The probabilities for AP occurring will vary depending on the crop/trait, the age and history of the germplasm accession or breeding line, and the degree of adherence to BMPs (see **Element 1**). Genebank and breeding program personnel should be able to identify instances of potentially increased probability for AP. Decisions for testing can be based on increased potential for AP (see **Element 1** and **Figs. 1** and **2** decision trees).
- As mentioned under **Element 1**, NPGS genebanks focus on generating and delivering accessions that are true-to-type, aspiring to zero tolerance of AP, but testing for <1% level of AP. Note that it is not feasible to provide assurance of 0% AP due to statistical limitations on testing results.
- For USDA/ARS plant breeding programs testing procedures also will test for a <1% level of AP. Optimal testing procedures are especially important for the continuing process of removing AP from breeding stocks (see **Element 4**).
- **Figures 1 and 2 illustrate decision trees and critical control points for testing for AP and for assessing the need for such testing for NPGS germplasm accessions and USDA/ARS plant breeding programs, respectively. Additional information appears in Elements 1 and 2.**
 - Decisions regarding testing occur at critical control points in the genebank management process, and are guided by decision trees (**Fig. 1**) which were developed to manage the risks identified in **Element 1**. The list of critical control points below is not intended to identify stages for mandatory testing but instead represents a menu of options to help genebank managers tailor superior strategies. The optimal approaches will vary according to the particular crop, the finite (sometimes limited) financial and/or material resources, and the operational capacities available for testing.
 - In general, testing is recommended after any seed increase in which cross-pollination and/or admixture with plants with GE traits could possibly

- have occurred. These points might include, but are not limited to, critical aspects of the following operations and workflows (also see **Fig. 1**):
- Incorporation of new germplasm with potential risk of AP into genebank collections. Such risks are exacerbated when documentation of such material is lacking or incomplete;
 - Whenever BMPs are not followed during germplasm storage, packaging, labelling, shipping, and other processing;
 - Propagation, especially when testing is needed to monitor the effectiveness of BMPs for regenerating accession true-to-type.
 - Harvest and post-harvest processing from seed increases; particularly seed incoming from external cooperators, where AP documentation might be lacking;
 - Reintroduction of seeds from an off-site germplasm increase where BMPs have not been assured.
- Decisions regarding testing occur at critical control points in the breeding process, and are guided by decision trees (**Fig. 2**), which were developed to manage the risks identified in **Element 1**. The list of critical control points below is not intended to identify stages for mandatory testing but represents a menu of options to help breeders tailor superior strategies in the most efficient manner; those strategies will vary according to the particular crop, the finite (sometimes limited) financial and/or material resources, and operational capacities available for testing.
- These control points include but are not limited to critical aspects of the following operations and workflows (also see **Fig. 2**):
- Introduction of new germplasm into breeding programs;
 - Parental materials used to develop breeding stocks;
 - Propagation, especially when testing is required to monitor the efficacy of BMPs;
 - Harvest and post-harvest processing from seed increases, particularly of seed received from external cooperators;
 - Reintroduction of germplasm from evaluation trials, counter-season nurseries and other external sources;
 - Prior to formal germplasm release, registration, and deposition in the NPGS (See “Mandatory purity testing of new varieties...”).
- Recommended molecular, biochemical, phenotypic and sampling procedures for AP testing:
 - Recommendations and guidelines for tests available to detect AP and recommended/approved laboratories with validated testing protocols are listed in **Appendix 2**, according to each crop/trait. This information will be updated periodically.
 - Positive results for AP should be confirmed with sensitive, reliable, and reproducible testing methods, preferably by an independent, accredited testing laboratory.

- Tests currently available for the detection of AP include:
 - Bioassay testing, e.g., survival after herbicide application;
 - Immunological testing for the expressed products of specific transgene or promoter antigens;
 - Phenotypic testing for specific GE traits not supposed to occur in the specific material;
 - DNA-based testing for particular gene promoters;
 - DNA-based testing for specific GE traits.

Element 3: Mandatory purity testing of new USDA/ARS varieties or enhanced germplasm prior to formal release. (see BIO 2012)

As mentioned earlier in this document, mandatory purity testing of new USDA/ARS crop varieties and enhanced germplasm prior to formal release is actually a subset of **Element 2** (Testing) but is presented separately to highlight its importance. The decision tree for testing USDA/ARS plant breeding stock, illustrated in **Fig. 2**, also covers mandatory purity testing of new USDA/ARS crop varieties and enhanced germplasm prior to release. Information about remedial actions to be taken with breeding stocks with AP appears in **Element 4**.

- The release notice and registration article for released conventional USDA/ARS varieties or enhanced germplasm should include a full description of the testing procedures applied. If feasible, it is desirable to identify the deregulated GE traits present, when AP is conclusively demonstrated. If deregulated GE traits or genetic-engineering constructs (e.g., promoters) are present and cannot be removed, an estimate of their frequency, and details regarding the extent and nature of testing process should be documented in GRIN and/or in specific local genebank records. The release notice might state: “USDA/ARS’s best management practices were followed to ensure genetic integrity and to avoid adventitious presence (AP) of GE traits. These best management practices include testing for AP of unintended GE traits at a <1% detection sensitivity. This statement does not imply that this material is completely free of GE traits but, rather, that it has been tested as described.”
- Prior to releasing varieties or enhanced germplasm with GE traits, USDA/ARS will follow procedures to ensure compliance with all relevant regulatory requirements. The release notice and registration article for released USDA/ARS varieties or enhanced germplasm with GE traits should include a full description of the transgenes or genetic-engineering constructs (e.g., promoters) present, sufficient to enable design of tests for AP. This includes reference to where the DNA sequence of the transgene can be found. Reference to the approved registration document for the GE trait should also be included. Guidelines relative to AP in conventional varieties or enhanced germplasm (preceding bullet) should be followed as well.
- Mandatory testing of specific germplasm accessions before distribution is triggered primarily when documentation is lacking that curatorial BMPs have been followed, or when a breach of such practices is documented. The extent of such mandatory testing is determined primarily by the interacting factors listed in the “Best Management Practices” and “Testing” sections. An estimate of the frequency of AP, plus details of the extent and nature of testing (types of tests, genes tested, sample sizes, etc.) should be documented, in GRIN and/or local genebank records, for curatorial and breeding records.

Element 4: Guidelines for mitigating the effects of adventitious presence (AP) of GE traits in USDA/ARS breeding stocks and germplasm accessions

As stated earlier, USDA/ARS genebanks and breeding programs aspire to 0% AP (or off-types of any kind), but test for AP at <1% detection level. The procedures applicable to mitigating the effects of AP of GE traits reported or determined for USDA/ARS NPGS germplasm accessions (see **Fig. 1**) differ in several aspects from those applicable for USDA/ARS breeding stocks (see **Fig. 2**), and so are treated separately in the preceding figures and in the following text.

- For NPGS accessions (Fig. 1), the first steps for mitigation, following reports of AP of GE traits, are:
 - Stop distributing the accessions, and categorize those accessions as “Potential AP of GE trait. Do not distribute” in GRIN-Global and in other records.
 - If a germplasm recipient or donor had reported AP in the accession, request their testing results and methodology/source, and determine if additional confirmatory testing is needed either “in house” or by an independent laboratory.
 - Subject the accessions to the testing procedures described in **Appendix 2** and **Element 2** if AP is confirmed, notify (see **Element 5**) any recipients of accessions produced from affected sources and/or seed lots.
 - Subsequently, if AP is confirmed for the accession, identify seed lots from previous propagations of that accession and then test for AP (**Appendix 2** and **Element 2**):
 - Seed lots that contain AP should be sequestered temporarily, with the intent of eventually destroying or returning them to the donor.
 - The most recently-increased seed lot that is AP-free would serve as the new seed source for repropagation of the accession, through standard BMP increase methods.
 - After propagation of a “true-to-type” seed source, all seed lots with AP should be destroyed by autoclaving.
 - If no “true-to-type” seed source can be located within the NPGS active collection or in seed lots in back-up storage:
 - A “true-to-type” source would be requested from the original germplasm donor.
 - If no “true-to-type” seed sources are available from the donor:
 - Determine whether “cleaning-up” the genebank accession with AP is a feasible mitigation strategy.
 - If “cleaning up” is feasible, on a case-by-case basis make arrangements to remove the GE trait from the germplasm accessions:

- Apply information about the frequency of the AP in the accession in question, and the statistical probabilities of achieving mitigation, given the detected level of AP, to design a “clean-up” process.
 - Examine opportunities to collaborate with breeders of that crop (either from USDA/ARS or cooperators) to “clean-up” the accession.
- For USDA/ARS breeding stocks (Fig. 2), the first steps for mitigation following reports of AP of GE traits are:
 - Stop distributing the breeding stocks, and categorize those breeding stocks as “Potential AP of GE trait. Do not distribute” in records.
 - If a germplasm recipient or donor had reported AP in the breeding stock, request their testing results and methodology/source, and determine if additional confirmatory testing is needed either “in house” or by an independent laboratory.
 - Subject the breeding stock to the testing procedures described in **Appendix 2** and **Element 2**. If AP is confirmed, notify any recipients of the breeding stock produced from affected sources and/or seed lots (see **Element 5**).
 - Test for AP in germplasm received from sources external to the USDA/ARS breeding program and at appropriate critical control points for the breeding program (**Appendix 2** and **Element 2**).
 - If AP is confirmed in germplasm received from sources external to the breeding program, inform the provider of those findings and return seed to them, or destroy them, if instructed by the provider. Determine if alternative, AP-free sources are available.
 - If AP is found in a seed lot at critical control points (see **Fig. 2**) in the USDA/ARS breeding program, do not plant the affected seed lot in a breeding block or increase nursery, and follow the mitigation procedures described below and in **Fig. 2**.
 - Mitigation options (**Fig. 2**) depend on the particular situation, and the most efficient and effective procedures will vary widely according to the breeding objectives and available resources. The following methods might be applicable to most ARS breeding programs.
 - Plant a plot or multiple pots of the seed lot with AP in the field or greenhouse, isolated from other materials in space or time to preclude outcrossing.
 - Divide the plot or the pots into quarters or some other appropriate fractions, and harvest fresh tissue from each “fraction,” bulking tissue by fraction, according to defined protocols.
 - Test the “fractions” for the previously-identified AP off-types according to defined protocols (see **Appendix 2** and **Element 2**).

- If the “fractions” with AP cannot be identified by testing and destroyed prior to plant flowering, then the plants in those “fractions” should be self-pollinated in isolation, and these procedures should be applied repeatedly until the “fractions” with AP can be identified.
- The “fractions” identified as containing AP should then be destroyed by autoclaving. Depending on the nature of the breeding program, the remaining fractions could be grown to maturity and harvested as new seed sources for the breeding program.
- If no “true-to-type fractions” are identified, then individual plants within a chosen fraction would be tested according to the procedures in **Appendix 2** and **Element 2**, and individual plants with GE traits would be destroyed. Depending on the nature of the breeding program, the remaining plants could be grown to maturity and harvested as new seed sources for the breeding program.

Element 5: Communication strategies for disseminating information about USDA/ARS procedures and practices for handling future occurrences of adventitious presence (AP) of GE traits.

General information about Agency procedures and practices will be posted on ARS and possibly other USDA web sites. It will be communicated to U. S. regulatory agencies, collaborators, partners, customers/stakeholders (see **Appendix 1**), and upon request. Communication strategies for specific situations are described below; these will be augmented and refined as experience with such topics accumulates.

Requests for information about the conventional/GE trait status of USDA/ARS NPGS germplasm accessions are ever more frequent. A standard information statement and/or response to such queries is provided below, which can be cited per se, or serve as the basis for more extensive statement or responses that address specific instances.

- “The seed supplies for the accessions you requested have been increased with standard genebank procedures (e.g., controlled pollination, individual plot harvesting, etc.) designed to minimize the chance of cross-pollination or seed admixture. Available information about trueness-to-type or purity for accessions of this crop is provided through GRIN-Global or by the crop curator.”

The procedures for communicating information about AP of deregulated GE traits in USDA/ARS germplasm accessions or breeding stocks focus on notifying the seed donor(s) and recipient(s) as soon as is practicable.

- Seed recipients and/or donors are notified, usually first by telephone, and then by e-mail messages. They are provided with as much relevant information as possible, preferably in a standard format.
- Depending on the particular situation, regulatory agencies, the owner(s) of proprietary GE traits, and/or officials of the Plant Variety Protection Office (PVPO) might be notified.
- Seed donors are asked for alternative, AP-free seed sources, if extant. Seed recipients are asked to destroy, or to return, remnant seeds to the USDA/ARS genebank or breeding project that distributed them.

AP of unauthorized and/or regulated GE traits in USDA/ARS germplasm accessions or breeding stocks is considered extremely unlikely. Nonetheless, it was considered prudent to prepare for that by developing procedures for communicating information to regulatory agencies, governmental officials, seed recipients and/or donors, collaborators, partners, and customers/stakeholders. These procedures, listed below in chronological order of execution, are more elaborate as compared to those described above for AP of deregulated GE traits.

- Following the initial report of the AP to USDA/ARS line management and Office of National Programs (ONP), specific USDA/ARS personnel will be designated by the

Agency to manage the communication of information within the Agency and the Department, and external to the latter.

- The telephone is usually the preferred medium for initial contacts regarding such AP incidents. The initial contacts are within USDA/ARS, and include the Administrator, Associate Administrators, relevant Area Directors, ONP, and Director of Information Staff.
- Briefing papers and/or talking points will likely then be prepared at USDA/ARS HQ for USDA/ARS and USDA audiences.
- ARS Administration and/or Information Staff will inform the USDA Office of Communications, the REE Office, and the Office of the Secretary.
- Key contacts for notifications of regulatory agencies, customer/stakeholders, and/or for further information dissemination are listed in **Appendix 1**.
- As soon as is practicable, seed recipients and/or donors are provided with as much relevant information as possible, preferably in a standard notification format, usually first by telephone, and then by e-mail message. Seed donors are asked for information about how the seeds were managed prior to their receipt by the USDA/ARS genebank or breeding program. Seed recipients are asked for information about how the seeds were used, and to destroy or to return remnant seeds to the USDA/ARS genebank or breeding project.

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Glossary

AC 21	USDA Advisory Committee on Biotechnology and 21st Century Agriculture
AOSCA	Association of Official Seed Certifying Agencies
AP	Adventitious Presence
ASTA	American Seed Trade Association
BIO	Biotechnology Industry Organization
BMP	Best Management Practices
GE	Genetically Engineered: genetic modification by recombinant DNA techniques
GIPSA	USDA Grain Inspection, Packers, and Stockyard Agency
GMO	Genetically Modified Organism
HACCP	Hazard Analysis and Critical Control Point
IPGRI	International Plant Genetic Resources Institute
NPGS	National Plant Germplasm System of the United States
ONP	USDA/ARS Office of National Programs
REE	USDA Research, Education, and Economics Mission Area
USDA/APHIS	United States Department of Agriculture/Animal and Plant Health Inspection Service
USDA/ARS	United States Department of Agriculture/Agricultural Research Service

Appendix 1: Contacts and Contact Information

US Government Agency Contacts and Contact Information

Institution	First Name	Last Name	E-mail address	Telephone
APHIS				
	John	Turner	John.T.Turner@aphis.usda.gov	301.851.3954
	Ed	Jhee	Edward.M.Jhee@aphis.usda.gov	301.851.3948
	Doug	Grant	Douglas.W.Grant@aphis.usda.gov	970.494.7513
	Compliance	Hotline	BRSCompliance@aphis.usda.gov	301.851.3935
EPA				
	John	Kough	Kough.John@epa.gov	703.308.8267
	Chris	Wozniak	Wozniak.Chris@epa.gov	703.308.4043
FDA				
	Kathleen	Jones	Kathleen.Jones@fda.hhs.gov	240.276.8243
	Jason	Dietz	Jason.Dietz@fda.hhs.gov	240.402.2282
USDA/FAS				
	Elizabeth	Jones	Elizabeth.jones@fas.usda.gov	202.690.3317
	Paul	Spencer	Paul.Spencer@fas.usda.gov	
	Melinda	Belisle	Melinda.belisle@fas.usda.gov	202.690.0292
USDA/GIPSA				
	Byron	Reilly	Byron.e.reilly@usda.gov	202.690.3368
	Tandace	Bell	Tandace.A.Bell@usda.gov	816.891.0459
USDA/Biotech				
Coordinator	Fan-Li	Chou	Fan-li.chou@ars.usda.gov	202.720.3817
2/14/18				

Industry Contacts (will vary according to crop and situation)

- ASTA: Andy LaVigne, Jane DeMarchi, Bernice Slutsky
- CropLife: Craig Richard
- BIO: Matt O'Mara, Kate Hall
- Organic Trade Association: Laura Batcha

Commodity Groups Contacts (will vary according to crop and situation)

- North American Export Grain Association: Gary Martin, Paul Green
- National Corn Growers Association: Nathan Fields, Jeff Mullen
- American Soybean Association: Ryan Findlay
- US Grain Council: Floyd Gaibler, Andrew Conner
- North American Wheat Growers Association: Steve Joehl
- National Cotton Council: Bill Norman
- Cotton Inc.: Kater Hake, Don Jones
- Beet Sugar Development Foundation: Paul Pfenninger
- National Alfalfa and Forage Alliance: Beth Nelson
- For future use:
 - U. S. Canola Association: Dale Thorenson

Appendix 2: Molecular, biochemical, phenotypic and sampling procedures for detecting AP of GE traits in USDA/ARS germplasm and breeding lines.

(Provided by Tandace Bell, USDA/GIPSA, with a contribution by Kathleen Yeater, USDA/ARS to the sample collection section)

Maintaining USDA/ARS germplasm and breeding lines “true-to-type” for crops with varieties incorporating deregulated GE traits involves testing for AP. Although every instance of AP is distinct, with a unique set of constraints, a general framework can be recommended for sampling and analytical procedures. Typically, the trait in question must be unequivocally identifiable by either protein or DNA-based detection method(s), with the latter of the two techniques offering increased specificity and sensitivity. Prior to analysis, the protein or DNA must be liberated from the other cellular components. The constituent of interest can then be directly measured, in the case of a protein, but DNA-based techniques require further purification and quantification prior to analysis. The following points outline a general procedure for detecting the AP of GE traits.

- Sample collection

A sampling plan should be designed that reliably minimizes seed stock depletion and analytical effort. But, that same sampling plan design should detect the AP in the sample within acceptable confidence levels. Advice of a statistician is sometimes required for anomalous sampling situations. The program *SeedCalc* is often consulted by industry to design seed testing protocols. This and other such statistical tools can be found online at: <http://seedtest.org/en/statistical-tools-content---1--1198.html>. Genebank personnel, breeders, and researchers should be aware that sampling from small population sizes, such as those regularly encountered with genebank samples and breeding stocks, might require applying the hypergeometric probability distribution to calculate the most accurate estimate of % AP in the population.

- Grinding of samples

Grinding is the first step required to liberate the analyte from its matrix. Prior to extraction, seeds must be ground in a manner that minimizes the risk of cross-contamination between samples. When individual seeds are bulked for analysis, it is important to take into account the validated limit of detection (LOD) for the detection assay(s). To avoid confusion, it is recommended that the amount of seeds to be ground for testing should be less than the LOD of the assay for the GE trait of interest. For example, if the LOD of a method is one off-type from a total of 1000 seeds, then the sample size for grinding should be equal to or less than 1000 seeds. The required particle size of the grind will determine the preferred type of grinding apparatus, ranging from mortar and pestle, to coffee grinders, to dedicated mills.

- DNA Extraction

Several basic DNA extraction methods are suitable for PCR-based GE trait detection protocols. Examples include: magnetic bead separation, silica column purification, and alcohol precipitation. Regardless of the method it should be validated by the laboratory performing the method. The links below provide information for common extraction products utilized by the USDA/GIPSA. Mention of a trade name or supplier does not constitute endorsement by USDA.

www.qiagen.com

www.promega.com

www.genetic-id.com

http://www.monsanto.com/products/documents/dna-detection/dna_im.pdf

- DNA-based identification and quantification

In many instances the GE trait of interest must be identified and/or quantified. Real-time quantitative polymerase chain reactions (RT-qPCR) require specific instrumentation, highly-trained staff, and dedicated laboratory instrumentation, but it is more sensitive and quantifiable as compared to protein-based methods. RT-qPCR requires primers and probes which are typically proprietary in nature and specific to the GE trait of interest. Sometimes information regarding the DNA-based detection method and corresponding primers and probes are publicly available, while often in the case of a regulated GE trait, this information must be requested from the trait manufacturer. Detection methods for many deregulated events in grains and commodities are available through a public database maintained by the Joint Research Centre.

<http://gmo-crl.jrc.ec.europa.eu/StatusOfDossiers.aspx>

- Protein-based identification

Protein-based detection methods are rapid and require minimal operator training and specialized equipment to conduct the assay. These methods often serve as rapid preliminary screening procedures. Disadvantages of the protein methods include decreased sensitivity and specificity because they only detect the protein of interest and not the genetic event. Several protein-based test kit manufacturers offer a wide array of both qualitative and quantitative rapid test kits suitable for detecting the AP of a deregulated event.

www.envirologix.com

www.agdia.com

www.neogen.com

- Repeated testing

Unrepeated protein or DNA-based detection are not necessarily reliable or conclusive, because false positive or false negative results are possible. For example, contaminants can be introduced during the protein testing or DNA extraction and reaction preparation processes. False positive or false negative results can be diagnosed through analyzing the same lot of seeds in duplicate by two independent analysts in different laboratories, or within the same laboratory with different kits on different days. When seed quantity is low, some of the seeds might need to be germinated first then vegetative tissue sample repeatedly for protein and/or DNA testing.

**The FAO International Treaty (IT) on Plant
Genetic Resources for Food and Agriculture,
the Convention on Biological Diversity (CBD),
and its Nagoya Protocol (NP)**

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The IT and the CBD

- The IT is a legally-binding Treaty under the UN Food and Agriculture Organization.
- The objectives of the IT are:
 - the conservation and sustainable use of PGRFA (Plant Genetic Resources for Food and Agriculture = plant germplasm) and
 - the fair and equitable sharing of the benefits arising out of their use.
 - The IT is in harmony with the CBD, and focused on sustainable agriculture and food security.
- The CBD is a legally-binding Convention.
- The objectives of the CBD are:
 - the conservation of biological diversity
 - the sustainable use of its components and
 - the fair and equitable sharing of the benefits arising out of the utilization of genetic resources.

The IT and the CBD

- **The IT:**
 - **140+ nations are Parties to it; in force since June 2004.**
 - **The US Senate approved ratification on 29 September 2016; US became a Party on 13 March 2017.**
- **The CBD:**
 - **190+ nations are Parties to it; in force since Dec. 1993.**
 - **The US signed (1993) but has not yet ratified it.**

The IT and the CBD

- **The IT:**
 - Recognizes nations' sovereign rights over "their" PGRFA but agree to:
 - Establish a multilateral system (MLS) for benefit-sharing and facilitated access to certain PGRFA for crop genetic improvement for food security.
 - Provisions for PGRFA in International Agricultural Research Centers (CIMMYT, IRRI).
- **The CBD:**
 - Recognizes nations' sovereign rights over "their" genetic resources.
 - Emphasizes contractual (bilateral) arrangements.
 - "Prior informed consent" can be required by a nation, and "benefit sharing" takes place via "mutually agreed terms" regarding access to germplasm.

The IT and the CBD

- **IT's scope includes all PGRFA. The MLS includes:**
 - **PGRFA of 64 food and feed crops key to food security; more crops may be included;**
 - **Held in ex situ collections by national governments (e.g., US National Plant Germplasm System), in the public domain; or held by IARCs; or**
 - **Donated by private entities.**
- **CBD's scope and coverage:**
 - **Most genetic resources under national jurisdiction exchanged internationally post-29 Dec. 1993.**
 - **US considers the IT to cover all PGRFA but some nations' IT interpretations might lead to their applying implementing legislation for the CBD Nagoya Protocol (NP) to crops not now covered by the IT MLS (e.g., soybean, tomato).**

The IT and the CBD

- **Benefit-sharing under the IT:**
 - In a broad sense, benefit-sharing under the IT will come from nations who are obliged to conserve PGRFA and make them available for research and breeding.
- **Benefit-sharing under the CBD:**
 - Negotiated by providers and recipients (e.g., in contracts for exchanging genetic resources); in some cases involve national governments.
 - In many nations, will be guided by the Nagoya Protocol (NP) on Access to Genetic Resources and the Fair and Equitable Sharing of the Benefits Arising from their Utilization: came into force October 2014.

The IT and the CBD

- **Benefit-sharing under the IT:**
 - In a narrow sense, monetary benefit-sharing will flow from individual PGRFA transactions via the Standard Material Transfer Agreement (SMTA) which details obligations of recipients and providers. Funds do not flow back directly to the PGRFA's country of origin: they are invested in projects supporting farmers in developing countries who conserve crop diversity and assisting farmers and breeders globally.
- **Benefit-sharing under the CBD:**
 - National implementation procedures are under development, and will be key to the NP's effects.
 - See the CBD Access and Benefit-Sharing Clearinghouse for more information:
<https://absch.cbd.int/>

The IT and the CBD

- **Germplasm access and exchange under the IT:**
 - **Via the MLS's SMTA, which includes conditions for end use (excludes non-food and non-feed), conservation, management of intellectual property rights (IPRs), and benefit-sharing upon commercialization.**
- **Germplasm access and exchange under the CBD:**
 - **Variable terms, negotiated by parties to individual contracts.**
 - **National implementation of the NP might affect those terms, and might not be tailored to the needs of agriculture, which can involve problematic requirements.**

Effects of IT and CBD on US germplasm users

- **IT:**
 - Has not affected use of PGRFA acquired pre-IT, nor of domestic US PGRFA.
 - Use of some PGRFA acquired internationally post-IT has been affected by terms and conditions of SMTA.
 - In some cases, the IT's SMTA has facilitated international access to PGRFA for U. S. public-sector researchers, genebanks, etc., but in other cases it has not.
- **CBD:**
 - Generally has not affected use of germplasm acquired pre-CBD, nor of domestic US germplasm.
 - Use of germplasm acquired internationally post-CBD affected by terms and conditions of exchange consistent with U. S. law.
 - Effects of the NP will be determined by its implementation.
 - Post-CBD and post-NP, access to germplasm internationally has become increasingly problematic.