Alfalfa Crop Germplasm Committee (CGC) Meeting Logan, UT - June 4, 2018 Report prepared by <u>Charles Brummer</u> (Chair)

Attendees: See list below

Meeting was called to order by Chair, Charles Brummer.

No previous meetings had been held in some time and therefore, no meeting minutes needed committee approval.

- 1. Introductions
 - Attendees introduced themselves (see attendee list below). The Alfalfa CGC meeting was held in conjunction with the NAAIC Standard Check/Standard Test committee as many attendees were interested in both meetings.
- 2. Peter Bretting and Brian Irish gave overviews of CGC roles and responsibilities
 - Urged the CGC to develop vulnerability statements for its crops
 - CGC to identify other descriptors and information if data are to be taken in Prosser
 - Collection explorations for more acquisitions could come from the vulnerability statement (submit proposals through CGC chair to be forwarded to ARS for consideration of funding)
 - A list of current descriptors, with associated data, exists for alfalfa from GRIN-Global and more descriptors could be developed through CGC consultation
 - Priority setting the CGC can help define priorities for the Curator and alfalfa collection
- 3. Brian Irish, Curator for the USDA-ARS National Plant Germplasm System Prosser, WAbased <u>Temperate-adapted Forage Legume</u> (TFL), presented his report on the previous year. The main topics included the following¹:
 - Limited staff to deal with 13K accessions!
 - Has a new Project Plan looking for collaborators on all aspects
 - Testing for adventitious presence (AP) of transgenes using sentinel plots did find AP in uncovered plots, but not in cages. This is very costly effort.
 - If accessions have low viability, plans are to use clonal propagation to get numbers up done only in emergencies
 - Few resources to get evaluation data but project is currently imaging flowers, pods, seeds, etc. The committee felt that this was a great idea and suggested getting pictures of the plants in the cages (all plants together) for general idea of growth type
 - Substantial distributions of the collection. Brian pointed out that requestors can ask the Curator; he can help target requests or provide other information that may not be obvious simply going through GRIN-Global and requesting through the shopping cart. Brian is involved in seeing/approving requests that come in. Feedback on distributions is welcome and encouraged.

¹ Full report bellow/attached

- Collecting new germplasm needs to be done carefully as the resources to handle more accessions is very limited. Long-Xi Yu has a possible collaborator in Kazakhstan who may have genetic resources to contribute. During collections/exchanges NPGS through a Plant Exchange Office exploration grant, could cover benefit sharing by possibly aiding researchers in originating country with evaluations of germplasm or via possible training Difficulty still lies in legal access to those genetic resources.
- 4. Proposal was made to join "Alfalfa CGC" and the "Clover and Special Purpose Legume CGC." There was no opposition in the audience, but conferral with Clover and Special Purpose Legume CGC Chair, <u>Ann Blount</u>, and that membership needs to be sought.
- 5. The CGC agreed to meet with the National Forage Conference (formerly NAAIC) and virtually by video conference in alternate years. Brian agreed to help in hosting the CGC Video Conference/Webinar.
- 6. The core collection in alfalfa is available, but the usefulness of the core could be questioned. The core could possibly be updated.
- 7. Germplasm evaluation funding still remains for horticultural crops, but not for others due to historical reasons....therefore, funding to evaluate the alfalfa (or other forage legume) collection is not available at this time from NPGS.
- 8. Brian's group oversees distributing seed of the check varieties used in NAAIC Standard Tests. He asked several questions how should they distribute the standard checks, to whom should they send the check seed, and with what frequency could people request seed? A further question is whether the standard checks are performing as they are supposed to, or perhaps related to this, whether the current standard check seed is genetically the same as the seed used in the standard test initially. All these issues merit further discussion.

At this point, CGC business was completed and the meeting transitioned to the NAAIC Standard Test Committee Meeting.

Attendee list:

Name	Affiliation	Email Address	
Baldwin, Brian	Mississippi State University	bsb2@msstate.edu	
Bhamidimarri, Suresh	Noble Research	sbhamidimarri@noble.org	
Blauer, Jake	Forage Genetics Institute	Blauer@lankolakes.com	
Bretting, Peter (ex officio)	USDA-ARS	peter.bretting@ars.usda.gov	
Brummer, Charlie	UC Davis	ecbrummer@ucdavis.edu	
Das, Modan	S&W Seed Co	modandas@swseedco.com	
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Irish, Brian (ex officio)	USDA-ARS	brian.irish@ars.usda.gov	
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Temperate-adapted Forage Legume Germplasm project report to the Alfalfa CGC Logan, UT - June 4, 2018 Report prepared by: Brian M. Irish

Summarized activities for the USDA-ARS National Plant Germplasm System Prosser, WAbased <u>Temperate-adapted Forage Legume</u> (TFL) germplasm collection.

- Personnel: A full-time Curator (Geneticist) is responsible for overall project implementation and for design of service and research activities. Project is also supported by a full-time USDA-ARS Biological Science Technician and one ½ time USDA/WSU Technician/Farmer as well part time Laborers throughout the year.
- Late 2017/early 2018 a 'new' Project Plan was developed and submitted for OSQR peerreview. This Project Plan is specific to the <u>Temperate-adapted Forage Legume</u> germplasm collection and outlines activities proposed for the next 5-year program cycle. Plan received a 'minor revisions' score and was certified on January 29, 2018. Project intends to focus resources on maintenance, documentation and distributions. As resources permit, possibilities exist to target germplasm for acquisition as well as several research objectives (see figure below for Project Plan specifics)².
- As part of the newly drafted project plan, collaborations were set up with several scientists at Arizona State University Polytechnic (Dr. Steele *Medicago* spp.) and Loyola University Chicago (Dr. Laten *Trifolium* spp.) and the University of Puerto Rico (Dr. Siritunga *Lotus* spp.) to correctly voucher species and to examine possible mislabeling and/or misidentification in TFL germplasm using DNA barcoding. Dr. Melanie Schori, USDA-ARS systematist, is also aiding this collaborative effort.
- During the 2017 summer regeneration period, alfalfa (*M. sativa* subsp. *sativa* 'Vernal') sentinel plots (½ covered and ½ uncovered) were established around germplasm regeneration field plots to monitor adventitious presence of genetically engineered (GE) glyphosate trait in alfalfa germplasm. Using a glyphosate-amended seedling germination assay and a seed grinding technique more than **140,000** seed was tested from 13 plots. No AP was detected from any covered (insect proof cage covers) sentinel plots, however <u>AP was detected</u> in five of the uncovered plots. AP detection in uncovered sentinel plots suggests transgenic gene flow to regeneration field site and extreme care must be taken to ensure timely placement of cage covers for alfalfa regenerations.
- Low seed number and low viability inventories continue to accumulate, especially for wild clover species and some accessions have been lost. These difficult to regenerate accessions are being targeted systematically for regeneration utilizing an in vitro germination procedure followed by clonal propagation in greenhouses. The idea is to increase the low number of plants germinating in the laboratory to the optimal (100) needed for field establishment. Personnel understands that genetic erosion might be occurring due to the less-than-ideal effective population size, but must do everything possible to avoid losing accessions.
- A recent systematic review of the *Lotus* genus has reclassified former native North American *Lotus* species into two new genera: *Acmispon* and *Hosackia*. Therefore, the TFL project gained **19** *Acmispon* species with **63** accessions and **6** *Hosackia* species with

² Anyone interested in more details regarding Project Plan is welcome to contact me

14 accessions. The number of *Lotus* species/accessions was adjusted correspondingly.

- Project coordinated the field planting and harvest (regeneration) of **184** *Medicago*, *Trifolium*, *Lotus* (*Acmispon/Hosackia*) germplasm accessions during the 2017 growing season. Additionally, **50** *Medicago*, *Trifolium* and *Lotus* species germplasm accessions were planted in the fall to be overwintered and regenerated CY 2018. The regeneration field site also included **10** sentinel plots and one faba bean (*Vicia faba*) accession.
- Project personnel were involved in the collection and scanning of 143 flower images for accessions being regenerated in 2017 as well as 173 seed and 109 pod voucher images for 2016 regenerated accessions. These images will be included in the GRIN-Global database and associated with corresponding accessions.
- During FY 2017 project coordinated the distribution of a total of 180 (174³) requests from 154 (142) cooperators (135 U.S./19 Int.). The number of unique accessions distributed was 3,222 (2,052) and total number of items distributed was 4,407 (3,243).
- Significant distributions during the FY were made to National and International requestors associated with Governmental, Private and Non-profit organizations. These genetic resources were used mostly in breeding and varietal development using traditional and genomic techniques, screening for disease resistance and abiotic stresses, screening for use as a cover crop, companion or in rotation, chemical analyses, phylogenetics, phytoremediation, class instruction and basic research (especially for model species e.g., *M. truncatula*).

Genus	Acc. ^a	Spe. ^a	Ava. ^a	Bac. ^a	Inc. ^b	Dis. ^b	Acq. ^b
Medicago	8,614	79	7,715	7,735	80	3,125	1
Trifolium	3,736	99	2,680	2,779	65	1,148	6
Lotus	920	40	767	736	39	134	3
Acmispon	63	18	51	48	-	-	1
Hosackia	14	6	9	11	-	-	1
Total	13,347	242	11,222	11,309	184	4,407	12

Table 1. Summary statistics for the Temperate-adapted Forage Legumes (TFL) collectionsconserved by the Western Regional Plant Introduction Station (WRPIS) of the NPGS.

Legend: Acc. = Accession; Spe. = Species; Ava. = Available; Bac. = Backup; Inc. = Increased; Dis. = Distributed; Acq. = Acquired; Rec. = Records (Observation records added to GRIN-Global). ^a Information verified on 10/16/2017. ^b Data summarized for the 2017 FY.

³ Numbers in parenthesis are for FY 2016 and used for comparison.



Figure 1. Project plan objectives, key products, and their interactions. To effectively and efficiently conserve germplasm and encourage its use (**Objective 1**), germplasm must be characterized/evaluated and its data be accessible (**Objective 2**) with routine revision of standard procedures and crop vulnerabilities to improve conservation strategies (**Objective 3**).



Monitoring for gene-flow and adventitious presence of genetically engineered (GE) glyphosate resistance in and around alfalfa regeneration plots in Prosser, WA. Left: Alfalfa hay field close to the regeneration site with feral glyphosate resistant plants between road and field (inset - plants testing positive for the glyphosate resistant GE trait with immuno-strips). **Right:** Field regeneration plot map indicating five alfalfa sentinel plots (red circle). Seed from sentinel plots is tested for gene-flow and adventitious presence at the end of the growing season.



Summer morning image of regeneration field site with isolation cages (**right**) and (**left**) example of leaves, pods and seed for a regenerated *M. polymopha* (alfalfa crop wild relative) accession.