Minutes from Tomato CGC meeting on September 15, 2014 at the Tomato Breeders Roundtable in Asheville, NC.

Meeting Attendees:

- **Jit Baral**
- **Roger Chelelat**
- **Mike Canady**
- **Majid Foolad**
- **David Francis**
- **Robert Heisey** – submitted resignation
- **Joanne Labate**
- **Richard Ozminkowski** (Chair)
- **Dilip Panthee**
- **Larry Robertson**
- **Felix Serquen**
- **John Snyder**
- Gary Kinard (via telephone)
- Sam Hutton
- Barbara Liedl
- Jasmine Lopez

(names in **boldface** = member or ex officio prior to meeting)

The meeting was called to order at 7 pm by Rich Ozminkowski, Chair.

1. **Crop Vulnerability Report** – (**Kinard and Ozminkowski):** The apple CGC has published their crop vulnerability report in the journal "Genetics Resources and Crop Evolution" and was discussed as an example. They included about 40 pages, with descriptors, in the supplements. Perhaps the journal would be interested in having similar documents for publications. The group discussed that we download the template and look at the options on what we would need to do for our crop. It was suggested that we also consider farmers market needs in this report. The new Xanthomonas problem is a good example of something to list in a vulnerability statement. Currently there is no way to fund this type of emerging issue in a vulnerable matter and grant funding doesn't necessarily lend itself to working on these threats. Gary thinks we did one in 2004 but no one remembers this. Four members volunteered to (Chetelat, Francis, Panthee, Hutton and Ozminkowski) form a committee to work on developing a draft in 4 months (January, 2015).

2. **NPGS report – (Kinard)**:
   a. NPGS staff changes: Retirement or resignations are a continuing issue at NPGS. Part of this is due to massive retirements 2 years ago and they are just now are able to rehire some of these positions.
   b. GRIN Global is not ready yet, continuing to develop and transition. USDA is getting supplemental staffing to support it.
   c. FAO International Treaty on Plant Genetic Resources for Food and Agriculture: No final action has been taken in the Senate since 2009. We have not had a vote since 2010. Universities are trying to rekindle this in the Senate. If State Experiment Station Directors have questions, then sent them to Peter Bretting for additional information which he has available.

3. **Membership**: Membership changes were discussed.

Member resignations (prior to the meeting):

- Angela Baldo: in absentia
- Mark Barineau: in absentia
- Randy Gardner: in absentia
- Robert Heisey
- John Prendergast: in absentia
- Jay Scott: in absentia

Membership additions during meeting:

- Sam Hutton
- Barbara Liedl
- Jasmine Lopez

Members incommunicado, to be removed:

- Rohilio Hernandez: in absentia
- Elaine Graham: in absentia
- Phyllis Himmel: in absentia

4. **Selecting a Vice Chair for Tomato CGC:** The committee selected Barbara Liedl as the new Vice Chair of the Tomato CGC.

5. **Recent tomato germplasm proposals funded**: Sam Hutton received the 2014 USDA NPGS Germplasm Evaluation project award. The group discussed how paltry the funds were available for evaluation. There have been no changes in the amount available in the last twenty years. Also it does not equate with the time that is taken to write and review these proposals even if our CGC has received funds for one project every year. Rich Ozminkowski appreciated the input from the group who evaluated the grant proposals this year. There was a discussion about whether data from these projects are getting into GRIN as is required with accepting the funds. The group was also reminded that when a publication arises from the research to let the NPGS know about this.

6. **PGRU Report – (Robertson)**: USDA germplasm report see attached. "Many challenges" would a good way to describe it overall. There have been a number of problems with water and late blight. Because of wet conditions, it was difficult to get in to spray. They also received 6" of hail on July 31 so they were only able to get seed from the second set of plants last year. Plans are in place to have Joanne Labate to take over as curator as Larry Robertson is here for only 3 years and 3 months. There have been problems not being able to hire staff to replace those who have left; there has been no one to handle seed distribution for the last two years. They have also lost another field person and the replacement is a term position because the administration is not willing to commit to replace the position. The other position they lost was due to a transfer.

   Most tomato lines are backed up at Fort Collins and the others are undergoing regeneration to get samples they will be able to send to Fort Collins. From 2012 through 2014, 499 accessions were regenerated, mostly from Fort Collins. Most had low seed counts. In last five years, the PGRU has distributed the full collection this was due to two large orders for screening. They have had to restrict who gets seed. For instance, they send home gardeners to other locations to get the seed. They are continuing to focus on regenerating those with low seed supply or when the viability drops. Currently they only do germination tests for tomato every twenty years or when feedback on a line suggests testing. Right now they have a backlog of 4 years of tests to do. The new person they hire would do both fieldwork as well has help in seed distribution. They do not have access to student help to alieve some of this labor shortage.

7. **TGRC Report – (Chetelat)**: The 2013 annual progress report was provided (attached). They have new populations of RILs, as well as lines from Rob Last and other mutants. They are doing more seed tests because they are stored in 5C frost free freezers which works well because they send out so much but they do acknowledge it would be good to have -20C hermetically sealed vaults for storage. The increased need for phytosanitary permits has cut back filling some requests. Wild species can be sent, but cultivated (*S. lycopersicum*) needs an import permit in addition to the phytosanitary permit to be sent to Europe. The TGRC database is okay as well as everything on the website. Thrips/TSWV has been a problem in the regeneration nursery. Staff turnover is another major problem. It is hard to keep staff at the pay and benefits they are available to provide. Instead they are using students, not a good thing for filling things such as seed requests.

8. **TGRC funding support – (Chetelat)**: TGRC received $25,000 in funding from NPGS ARS (Bretting) via the PGRU (Robertson); it was suggested the money go directly to TGRC from NPGS. This amount is low, in years past they sometime received $60,000. This comes in as a cooperative agreement from Peter Bretting via PGRU and the University of California at Davis, UCD does not take indirect overhead. PGRU has been keeping some $ on hand in case funds come in late. TGRC's other sources of funding include the endowment which generates $80,000 and the California Tomato Commission gives $15,000. There is a new initiative to raise another million for the endowment to give them an additional $40,000 yearly. The group discussed that it would be helpful to have a letter from the committee to help the companies see the importance to support the TGRC. The group was also asked to provide testimonials. There was a discussion of compiling contacts at companies so they are not contacted for this request for funds twice. Chetelat mentioned that TGRC would be talking with companies in November at the California tomato conference in Napa and so they hoped to get the ball rolling around mid October. David Francis suggested some additional ideas on funding which included working with the NAPB, writing letters to Bretting on the importance of the collections as well as our representatives, and check into how the other collections are being funded (OPGS for example). However he did caution that we need to be careful to not do multiple requests for funding support.

9. **Germplasm Priority Survey Results** – We are the only crop germplasm committee with a list. The group discussed that historically funding populations was important. The chair asked everyone to rate using 150 points total to develop an index. The updated priority list will be posted on the tomato CGC site.

10. **Other business** – The group discussed establishing semiannual conference call meetings to deal with annual business. However it was suggested this not be in September. The first test of the use of a conference call system will be in 4 months on the crop vulnerability report being developed.

The meeting was adjourned at 9:00 pm.

Respectfully submitted by B. E. Liedl

2014 Tomato Germplasm Evaluation Priorities			
Type	Description	Current (2014) ranking	Previous (2011) rating
Bacterial Diseases	Bacterial canker	High	High
Bacterial Diseases	Bacterial spot	High	High
Bacterial Diseases	Bacterial soft rot (post harvest)	Moderate	Medium
Bacterial Diseases	Bacterial speck (new race)	High	Medium
Bacterial Diseases	Bacterial wilt	High	Low
Bacterial Diseases	Candidatas liberbactes Solanacearum	Moderate	High
Fungal Diseases	Verticillium wilt race 2	High	High
Fungal Diseases	Target Spot	Moderate	High
Fungal Diseases	Corky root	Moderate	High
Fungal Diseases	Late blight	High	Medium
Fungal Diseases	Phytophthora root rot	Moderate	Medium
Fungal Diseases	Fruit rots	low	Medium
Fungal Diseases	Powdery mildew	Moderate	Low
Viral Disease	Pepino mosaic virus	High	High
Viral Disease	Non-spotted wilt tospo viruses	Moderate	High
Viral Disease	Marchites manchada syn Sinoloa necrosis	low	Medium
Viral Disease	gemini viruses	High	Medium
Viral Disease	Tomato Spotted Wilt Virus	High	Medium
Viral Disease	CMV	low	Medium
Viral Disease	Beet curly top virus	Moderate	Low
Viral Disease	PVY	low	Low
Viral Disease	Groundnut ringspot virus	low	High
Insect screening prot	Silverleaf whitefly	High	Medium
Insect screening prot	Nematodes, heat stable	High	High
Insect screening prot	Aphids	Moderate	Medium
Insect screening prot	Psylid insects	Moderate	Medium
Insect resistance	Tuta absoluta	Moderate	High
Stress	Cold tolerance	Moderate	High
Stress	Heat tolerance	High	High
Stress	Salinity tolerance	Moderate	Medium
Stress	Color disorders	Moderate	Medium
Stress	Graywall tolerance (finding markers)	High	High
Horticultural	Soluble solids	Moderate	High
Horticultural	Flavor (define components)	High	High
Horticultural	Antioxidants/nutritional content	Moderate	Medium
Horticultural	Color	High	Medium
Horticultural	Sugar type	low	Medium
Horticultural	Peelability/dicing	low	Medium
Horticultural	Viscosity	low	Medium
Horticultural	Blossom-end smoothness	low	Low
Horticultural	Fruit chilling tolerance	low	Low
Genetic Resources	Genotyping to define core collections	High	High
Genetic Resources	Phenotypic characterization of segregating populations	Moderate	High
New	Drought Tolerance	High	n/a
New	Flood Tolerance	low	n/a
New	Nutrient Use Efficiency	low	n/a
New	Short-duration Freeze Tolerant	low	n/a
New	Internal white tissue	low	n/a
New	PSTVd and viroids	low	n/a
New	Fusarium race 3 - alternate gene	low	n/a
New	Thrips	low	n/a

Tomato Germplasm Conservation at the
Plant Genetic Resources Unit – Geneva, NY

September, 2014
Asheville, North Carolina

Currently, there are 6,591 accessions of 13 *Solanum* species conserved at the Northeast Regional Plant Introduction Station located at the PGRU, Geneva, New York (Table 1). This includes 927 accessions which have been transferred to Geneva from the National Center for Germplasm Conservation to consolidate the tomato germplasm collection. 1423 accessions that previously had only G or NSSL accession numbers have been assigned PI accession numbers and the only G accession numbers are accessions that have been recently received and are in the process of being regenerated. Accessions that were considered duplicate accessions have been inactivated with the accession they are duplicates of marked in the germplasm database so that users can access the representative sample of the accession; this includes 239 PI accession numbers, 51 G accession numbers, and 225 NSL accession numbers. The new taxonomic classification system for tomato has been implemented in GRIN and is reflected in Table 1.

Table 1. Tomato germplasm conserved at the Plant Genetic Resources Unit located at Geneva, NY

Species	Accessions	Backup
arcanum	4	3
cheesmaniae	7	6
chilense	1	1
chmielewskii	7	7
corneliomulleri	13	12
galapagense	5	5
habrochaites	63	40
neorickii	1	1
pennellii	10	4
peruvianum	122	96
pimpinellifolium	231	227
lycopersicum	5955	5785
subsect. lycopersicon hybr.	158	158
subsect. lycopersicon spp.	14	3
TOTAL	**6591**	**6348**

Characterization for the minimal descriptor list and digital imaging of the germplasm accessions has been incorporated as part of the regeneration process. In 2013 58 and in 2014 60 accessions were characterized with digital images of fruit and foliage also taken. In addition, these accessions were evaluated using the Tomato Analyzer Program.

From 2010 through 2014 we have regenerated 499 cultivated tomato accessions; most of these were newly transferred accessions and accessions collected as part of the tomato core collection. Essentially all cultivated tomato accessions are now available for distribution. Funds were obtained to support the Assistance Cooperative Agreement with the Tomato Genetic Resources

Center at the University of California-Davis for one more year with $25,000 funds. Distribution of the tomato germplasm collection at PGRU for the past five years is given in Tables 2 and 3 (accessions and samples). There has been a significant increase in distribution of *Solanum* with an average of 4,000 accessions distributed per year. Over this time period almost all accessions were distributed at least one time. The majority of the *Solanum* accessions distributed have been *Solanum lycopersicum.*

Table 2. Distribution of tomato germplasm accessions at PGRU for 2010 through 2014*

Species	2010	2011	2012	2013	2014	Total
arcanum	3	3	4	2	2	4
cheesmaniae	3	6	5	2	2	6
chilense	1	1	1	1	1	1
chmielewskii	7	7	7	3	5	7
corneliomulleri	9	13	12	4	4	13
galapagense	4	4	4	3	4	4
habrochaites	13	46	52	9	15	53
neorickii	1	1	1	1	0	1
pennellii	5	5	5	2	3	5
peruvianum	76	94	112	10	11	115
pimpinellifolium	41	220	211	45	20	222
lycopersicum	922	5561	5148	1085	415	5655
subsect. lycopersicon hybr.	37	156	155	4	43	157
subsect. lycopersicon spp.	5	5	14	0	0	14
Total	1127	6122	5731	1171	525	6257

*2014 distributions are through August 1, 2014.

2012-2014 Tomato and tomatillo research highlights at PGRU

1) Labate, J.A., L.D. Robertson, S.R. Strickler and L.A. Mueller. 2014. **Genetic structure of the four wild tomato species in the *Solanum peruvianum* s.l. species complex.** Genome 57:169-180. National Center for Biotechnology Information Sequence Read Archive (SRA) Study SRP034922, BioProject PRJNA230524 and BioSample numbers SAMN02436034 - SAMN02436079. We used 14,043 SNPs genotyped by genotyping-by-sequencing in 46 plants to analyze genetic relationships within and among the four wild tomato species *S. peruvianum*, *S. arcanum*, *S. huaylasense* and *S. corneliomueller*i. Geographical origins of the sampled plants in Peru, Ecuador and Chile were mapped. We now understand genetic properties of the four species based on how plants were related to each other within the context of their geographic origins. Many thousands of DNA markers were identified that could potentially be used to distinguish among the four species by using simple lab tests. The markers also have potential to identify genes that were selected during adaptive divergence of the species.

2) Labate, J.A. and L.D. Robertson. 2014. **Nucleotide diversity estimates of tomatillo (*Physalis philadelphica*) accessions including nine new inbred lines.** Molecular Breeding (*in review*).
Genotyping by sequencing data have been analyzed for tomatillo (*Physalis philadelphica*) (125 accessions). Global Geographic Information Systems (GIS) data from Genetic Resources Information Network (GRIN) and from the International Rice Research Institute (IRRI) project

Table 3. Distribution of tomato germplasm samples at PGRU for 2010 through 2014*

Genus/Species	2010	2011	2012	2013	2014	Total
arcanum	7	3	4	2	2	18
cheesmaniae	4	8	14	3	2	31
chilense	5	2	1	1	1	10
chmielewskii	11	10	18	3	5	47
corneliomulleri	16	14	27	5	4	66
galapagense	11	5	10	4	6	36
habrochaites	24	64	72	13	18	191
neorickii	2	2	4	1	0	9
pennellii	17	15	10	4	3	49
peruvianum	146	118	225	20	15	524
pimpinellifolium	48	231	349	52	21	701
lycopersicum	1423	6985	6852	1612	533	17405
subsect. lycopersicon hybr.	46	200	183	5	47	481
subsect. lycopersicon spp.	5	8	16	0	0	29
TOTAL	1765	7665	7785	1725	657	19597

*2014 distributions are through August 1, 2014.

on geospatial references was verified using web and PC-based software tools. We found that multiple plants sampled per accession were closely related to each other, but there was no apparent pattern related to state in Mexico, latitude or isolation-by-distance. Average proportion of heterozygous sites was halved in nine inbred lines developed at PGRU relative to open-pollinated accessions (0.04 versus 0.08). Tests of population differentiation showed that 29% of pairs of accessions were significantly differentiated from each other.

3) We are in the process of publishing analyzing a study that

- characterized a diverse set of 52 historic U.S.A. tomato varieties for fruit quality (antioxidants, sugars, acids, Vitamin C and color), size and morphological (size and shape) variables using replicated trials across two environments
- evaluated the reproducibility of assays for organic acids, carbohydrates and antioxidants in cryopreserved samples of homogenized fruit
- estimated effects due to genotype, environment and their interaction on fruit quality and morphological variables
- provided insight into which traits distinguished the varieties and as to whether these traits were interrelated, correlated or predictive of each other
- discovered which traits are tractable for crop improvement based on their heritability, relationships and ease of assay

4) A core set of 190 PGRU tomato accessions was genotyped using GBS. These originated from 31 countries and included six different versions of the economically valuable variety San Marzano.

C. M. Rick

T G R C

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ANNUAL PROGRESS REPORT

2013

Figure 1. A male-sterile mutant in *S. chilense* LA2759. Accessions of the wild tomato species sometimes show segregation for apparently monogenic traits. In this accession of *S. chilense*, an obligate outcrossing species, we observed segregation for a male-sterile mutant with thin anthers. [photo by S. Peacock]

SUMMARY

Acquisitions. The TGRC acquired 182 new accessions of cultivated tomato. The new stocks include 148 recombinant inbred lines (RILs) developed by Dr. Majid Foolad at Penn State Univ. from a cross between cv. NC EBR-1 x *S. pimpinellifolium* LA2093. We acquired 15 mutant stocks with altered trichome biochemistry from Dr. Rob Last at Michigan State Univ. The mutant phenotypes include increased or decreased expression of several classes of phytochemicals related to plant defense against insects and wounding. In addition, 18 nearly isogenic lines (NILs) of developmental mutants in cv. 'Micro-Tom' were acquired from Dr. Lazaro Peres from the Univ. de Sao Paulo. We also regenerated a number of 'inactive' wild species accessions which had never been grown by the TGRC. The current total of number of active accessions is 3,839.

Maintenance and Evaluation. A total of 1,212 cultures were grown for various purposes, of which 555 were for seed increase (including 96 wild species stocks) and 353 for germination tests. Progeny tests were performed on 82 stocks of segregating mutants or various lines with unexpected phenotypes. Tests for transgenes (GMOs) in 18 stocks were all negative. Other stocks were grown to confirm wild species introgressions, or for research projects. Newly regenerated seed lots were split, with one sample stored at 5° C to use for filling seed requests, the other stored in sealed pouches at -18° C to preserve viability. As allowed by harvests, backup seed samples were also submitted to the USDA Natl. Center for Genetic Resources Preservation in Colorado, and to the Svalbard Global Seed Vault in Norway.

Distribution and Utilization. A total of 4,718 seed samples representing 1,675 unique accessions were distributed in response to 304 requests from 227 colleagues in 22 countries; over 33 purely informational requests were also answered. The overall utilization rate (i.e. number of samples distributed relative to the number of active accessions) exceeds 125%, showing that demand for our stocks remains high and that many accessions are requested at least once each year. Information provided by recipients indicates our stocks are being used to support a wide variety of research, breeding, and educational projects. Our annual literature search uncovered 95 publications mentioning use of our stocks.

Documentation. Our website was updated in various ways to add features and address security issues. Updates were made to our geographic mapping tools to maintain compatibility with the GoogleMap interface. Web pages related to seed requests were modified to enable charging for express shipping options and phytosanitary certificates. We revised our horticultural recommendations for growing wild species, and added guidelines on emasculating and pollinating tomato flowers. Descriptive data on new accessions were added and records on existing accessions were updated as needed. Our database was modified in various ways to improve internal record keeping related to seed requests, plant pedigrees, and seed lots. A revised list of wild species stocks was published in the Tomato Genetics Coop. Report (TGC).

Research. The TGRC continued research on the mechanisms of interspecific reproductive barriers that restrict crosses between cultivated tomato and its wild relatives. We published a paper on the role of a pollen factor, *ui6.1,* in self-incompatibility, and identified natural variation for two pollen factors in self-compatible biotypes and species. We received a new grant from the USDA-NIFA to develop a set of introgression lines representing the genome of *S. sitiens*, a wild tomato relative known for its tolerance to drought and salinity, but which has not been utilized in the past due to strong crossing barriers.

ACQUISITIONS

The TGRC expanded its collection of genetic stocks and wild species accessions through donations from external researchers and by rescuing inactive collections from seed storage. We acquired 182 new accessions of cultivated tomato in 2013. The new stocks include 148 recombinant inbred lines (RILs) developed by Dr. Majid Foolad at Penn State Univ. from a cross between cv. NC EBR-1 x *S. pimpinellifolium* LA2093, followed by multiple generations of single seed descent from the F2 (Ashrafi et al. 2009). NC EBR-1 is an early blight resistant breeding line developed by Randy Gardner at North Carolina State University. LA2093 is an accession of *S. pimpinellifolium* collected by Charley Rick and colleagues at La Union, Ecuador. This RIL population will be the first relatively large RIL library for tomato that is publically available through the TGRC. Another advantage is that the RILs are being genotyped to very high resolution using GBS (genotyping by sequencing) by Allen Van Deynze's group at UC-Davis as part of the SolCAP (Solanaceae Coordinated Agricultural Project) program. A high density molecular marker map has already been developed from these RILs (Ashrafi et al. 2009 *Genome* 52: 935), and it has been used to map QTLs for horticultural and fruit quality traits, such as fruit weight, lycopene content, soluble solids, and days to maturity (Ashrafi et al. 2012 *Mol. Breeding* 30: 549). We think this population will be a useful new genetic resource for various research and breeding purpose, particularly studies of yield, fruit size, fruit quality, abiotic stress tolerances, and possibly other traits.

Figure 2. Fruit of *S. lycopersicum-S. pimpinellifolium* recombinant inbred lines.

We also acquired 15 mutant stocks with altered trichome biochemistry from Dr. Rob Last at Michigan State Univ. The mutant phenotypes include increased or decreased expression of several classes of phytochemicals -- acyl sugars, tomatine, and methylmyricetin -- related to plant defense against insects and wounding. In addition, 18 nearly isogenic lines (NILs) of developmental mutants with hormone deficiencies and/or altered physiological responses were acquired from Dr. Lazaro Peres from the Univ. de Sao Paulo. These NILs were developed by backcrossing each mutant into the genetic background of 'Micro-Tom', a compact patio variety popular for experimental research purposes.

In addition, we regenerated several previously inactive wild species accessions by planting very old seed samples kept in storage for up to 30 years. None of these stocks had been grown before by the TGRC, thus, for the ones that germinated, we had our first opportunity to observe them. There were several noteworthy items among the newly rescued accessions. We grew a collection of *S. chilense* (LA2957) from Pozo, in the Camina drainage of Tarapaca Region, Chile, that turned out to be a mixture of *S. peruvianum* and *S. chilense* plants. We have only two other *S. peruvianum* collections from this river valley. They appear to represent the southern margin of the natural range for this species. In addition, we grew an accession of *S. chilense* (LA2881) from Socaire, Antofagasta, Chile that segregates for self-compatibility (SC) and self-incompatibility (SI). This species is almost exclusively SI, and as far as we know, the only other occurrence of SC is another mixed (SI/SC) collection also made at Socaire. We also revived a collection of *S. habrochaites* (LA2728) made near Las Juntas, Loja, Ecuador. Like most other collections from Ecuador, LA2728 has relatively small, pale colored flowers and appears to be SC. It has vigorous thick stems, darkly pigmented stems and fruit, and twisted leaflets. More detailed information on the new accessions can be found on our website at http://tgrc.ucdavis.edu/acq.aspx.

Obsolete or redundant accessions were dropped. The current total of number of accessions maintained by the TGRC is 3,839.

Table 1. Number of accessions of each species maintained by the TGRC. The totals include some accessions that are currently unavailable for distribution.

***Solanum* name**	***Lycopersicon* equivalent**	**No. of Accessions**
S. lycopersicum	*L. esculentum*, including var. *cerasiforme*	2,685
S. pimpinellifolium	*L. pimpinellifolium*	311
S. cheesmaniae	*L. cheesmanii*	41
S. galapagense	*L. cheesmanii* f. *minor*	29
S. chmielewskii	*L. chmielewskii*	29
S. neorickii	*L. parviflorum*	52
S. arcanum	*L. peruvianum*, including f. *humifusum*	46
S. peruvianum	*L. peruvianum*	75
S. huaylasense	*L. peruvianum*	19
S. corneliomulleri	*L. peruvianum*, including f. *glandulosum*	57
S. chilense	*L. chilense*	118
S. habrochaites	*L. hirsutum,* including f. *glabratum*	124
S. pennellii	*L. pennellii*, including var. *puberulum*	51
S. lycopersicoides	n/a	24
S. sitiens	n/a	13
S. juglandifolium	n/a	6
S. ochranthum	n/a	9
Interspecific hybrids, RILs	n/a	150
Total		**3,839**

MAINTENANCE

Led by Scott Peacock and his crew of undergraduate student assistants, the TGRC again managed large field and greenhouse plantings this year. A total of 1,212 families were grown for various purposes; 555 of these were for seed increase, including 96 of wild species accessions, most of which required greenhouse culture. The rest were grown for germination tests, evaluation, introgression of the *S. sitiens* genome, research on reproductive barriers, or other purposes.

Identifying accessions in need of regeneration begins with seed germination testing. Seed lots with a germination rate that fails to meet our threshold of 80% are normally regenerated in the same year. Other factors, such as available space, age of seed and supply on hand, are also taken into account. Newly acquired accessions are typically regenerated in the first year or so after acquisition because seed supplies are limited and of uncertain viability. This year, 353 seed lots were tested for germination responses. Average germination rates continued to be relatively high for most species (Table 2), indicating conditions in our seed vault are satisfactory. We observed unusually poor germination responses in seed lots of cultivated tomato and cherry tomato, possibly due to technical problems with those tests. We sometimes encounter lack of uniformity in seed bleaching or seed wetting. Use of cheesecloth ‘tea bags’ for bleaching multiple seed lots, and thicker germination paper (the blue blotter paper) for sprouting seeds give more consistent results than other methods we've tried.

Table 2. Results of seed germination tests. Values are based on samples of 50-100 seeds per accession, and represent the % germination after 14 days at 25°C. Seed lots with a low germination rate are defined as those with less than 80% germination.

***Solanum* Species**	**Date of Tested Lots**	**Avg % Germ.**	**# Tested**	**# Low Germ**	**# Grown[a]**
S. lycopersicum	2002-2003	49	158	154	378
S. pimpinellifolium	1998-2003	95	30	1	12
S. cheesmaniae, S. galapagense	2000-2003	69	16	7	5
S. chmielewskii, S. neorickii	1997-2001	99	10	0	12
S. peruvianum clade	1984-2003	89	27	4	5
S. chilense	1990-2003	78	38	15	14
S. habrochaites	1981-2003	91	14	2	4
S. pennellii	2003	93	3	0	1
S. lycopersicoides	1990-2002	69	12	9	6
S. sitiens	2002-2003	76	6	4	2
S. juglandifolium	2000	36	1	1	2
S. ochranthum	--	--	0	0	0

[a] Includes all accessions grown for seed increase in the 2013 pedigree year, whether for low germ or for other reasons.

For accessions grown in the field, the usual sequential plantings were made to spread out the work load. Seedlings were transplanted in the field on four separate dates, the first on April 19th and the last on July 10th. A total of 59 rows were planted. Early growth and establishment were favorable, except for outbreaks of curly top virus (CTV) and tomato spotted wilt virus (TSWV). Summer temperatures were again relatively mild this year, and generally favorable for fruit set, with only a few periods of excessive temperatures, during which manual pollinations were suspended.

For various reasons, many of the wild species, mutants and certain other genetic stocks require greenhouse culture. For the mutant stocks, we start the weakest lines first, and finish with lines of normal vigor. We now grow most of the introgression lines in the greenhouse, both to assure adequate seed set (some are partially sterile in the field) and to reduce the risk of outcrossing. For the wild species, plantings in the greenhouse are based on daylength response: those with the least sensitivity are planted first; next, those with intermediate reaction; last, the most sensitive (i.e. flower best under short days). Optimal planting dates for each species are listed on our website, at http://tgrc.ucdavis.edu/spprecommed.html.

Figure 3. Fruit from *S. ochranthum*.

Our greenhouse plantings were relatively trouble-free this year, except for persistent infestations of thrips. We had good success in reproducing *S. ochranthum*, a species that is normally reluctant to flower and set fruit under our conditions. The regime that worked well was to sow seeds in the fall, keep them root bound in speedling trays until early spring, then transplant to 1 gal. pots at 3 plants per pot. This prevented excessive vegetative growth and induced flowering early in the season while the daylength was still relatively favorable. Unusually heavy fruit set was obtained by repeated mass sib cross pollinations (Figure 3).

As in the past, we continue to store samples of all newly regenerated seed lots in our seed vault at 5-7°C; this is our 'working' collection, used for filling seed requests. In addition, we package samples of freshly harvested seed in sealed foil pouches for storage at -18°C. Samples of nearly all our wild species accessions have now been stored at -18 in foil pouches, which should extend longevity and limit the frequency of regeneration cycles, thereby reducing workload and better preserving diversity. As in the past, large samples of newly regenerated seed lots were sent to the USDA-NCGRP in Ft. Collins, Colorado, for long-term backup storage. This year, 34 accessions were sent to NCGRP, and 34 to the Svalbard Global Seed Vault in Norway.

EVALUATION

All stocks grown for seed increase or other purposes are systematically examined and observations recorded. Older accessions are checked to ensure that they have the correct phenotypes. New accessions are evaluated in greater detail, with the descriptors depending upon type of accession (wild species, cultivar, mutant, chromosomal stocks, etc.). In the case of new wild species accessions, plantings are reviewed at different growth stages to observe foliage, habit, flower morphology, mating system, and fruit morphology. We also record the extent of variation for morphological traits, and in some cases assay genetic variation with markers. Such observations may reveal traits that were not seen at the time of collection, either because plants were not flowering or were in such poor condition that not all traits were evident, or because certain traits were overlooked by the collector.

Many genetic stocks, including various sterilities, nutritional, and weak mutants, cannot be maintained in true-breeding condition, hence have to be transmitted from heterozygotes. Progeny tests must therefore be made to verify that individual seed lots segregate for the gene in question. We sowed 82 lines for progeny testing of male-steriles or other segregating mutants, as well as various other stocks with incorrect phenotypes. This year's progeny tests included the male-sterile mutants *ms-5, ms-6, ms-7, ms-23, Ms-48, ps* (*positional sterile*), *ses* (*semisterilis*), *sl* (*stamenless*), and *sl-2* (*stamenless-2*). Other tested stocks included the mutants *nv* (*netted virescent*, Figure 4), *pat* (*parthenocarpic*), *sha* (*short anthers*), a tetraploid stock of cv. San Marzano, cv. E-6203, and an unusual yellow-fruited *S. pimpinellifolium* from Vista Florida, Peru.

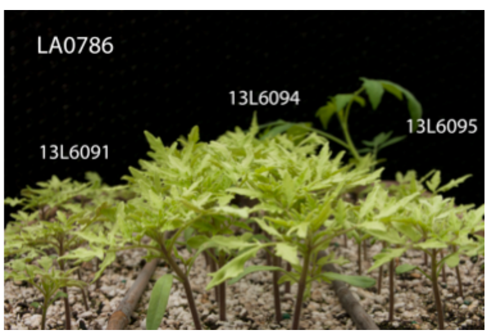

Figure 4. Progeny tests of *nv* stocks.

Tests for the presence of transgenes (GMOs) were performed on 18 stocks grown for seed increase, all of which were negative. We submitted 61 seed lots of various wild species accessions for testing to detect Potato Spindle Tuber Viroid, and all were negative.

DISTRIBUTION AND UTILIZATION

The TGRC again filled a very large number of seed requests this year. A total of 4,718 seed samples representing 1,675 different accessions were sent in response to 304 seed requests from 227 investigators in 22 countries. In addition, over 33 purely informational requests were answered. Relative to the size of the TGRC collection, the number of seed samples distributed was equivalent to a utilization rate of over 125% -- a high rate for any genebank, and a sign that demand for our stocks remains high.

The various steps involved in filling seed requests – selecting accessions, packaging seeds, entering the information into our database, providing cultural recommendations, obtaining phytosanitary certificates and import permits, etc – involve a large time commitment. Led by Jennifer Petersen, the TGRC crew did a splendid job filling requests promptly and accurately. The online payment system we implemented to recover the costs of phytosanitary certificates continues to function well, allowing us to keep up with the rising cost of phytos. We now recoup the cost of express mail shipping as well. Many countries are increasing the stringency of their import regulations, and obtaining the necessary phytosanitary certificates and/or import permits is becoming more onerous and time consuming. For instance, Japan now requires an import permit for some tomato species but not for others, so shipments need to be split, with different sets of documents accompanying each group of seed samples. We cannot ship seed of cultivated tomato lines to countries in the E.U. zone without a letter of authorization with the appropriate phytosanitary exemptions, however we can ship seeds of the wild relatives to the E.U.

Information provided by recipients regarding intended uses of our stocks is summarized in Table 3. A few trends are apparent in the data. There was noticeably less emphasis on breeding for resistance to various diseases and/or investigations of the molecular biology of host-pathogen interactions than in previous years. On the other hand, there was greater interest in abiotic stress responses, especially drought, salinity and high/low temperature stresses. There was less interest in carotenoids and antioxidants, and greater focus on fruit flavor than in the past. There continues to be increasing interest in the use of rootstocks for grafting. Many genetic studies mentioned diversity or natural variation, or gene expression. Other research topics accounting for many requests included studies of interspecific reproductive barriers, rhizosphere biology, metabalomites, trichome volatiles/exudates, and wound responses/signaling. We again received a significant number of requests for instructional uses. As in the past, the largest number of requests were for unspecified uses, either related to breeding or research, particularly in the private sector.

There continues to be high demand for introgression lines (ILs) -- stocks containing a defined wild species chromosome segment in the background of cultivated tomato -- as they offer many advantages for breeding and research. A total of 34 requests and 680 seed samples were processed for the *S. pennellii* ILs, 18 requests and 337 samples for the *S. habrochaites* ILs, and 12 requests and 167 samples for the *S. lycopersicoides* ILs.

Table 3. Intended uses of TGRC stocks as reported by requestors. Values represent the total number of requests in each category. Requests addressing multiple topics may be counted more than once.

Category	*# Requests*
Biotic Stresses	
Viruses:	
PepMV	1
ToMV	1
TSWV and other tospoviruses	2
TYLCV and other begomoviruses	2
Viroids	1
Unspecified viruses	1
Bacteria:	
Bacterial canker	1
Bacterial spot	4
Zebra complex	1
Fungi:	
Cercospora leaf mold	1
FORL	1
Fusarium wilt	1
Phytophthora fruit rot	1
Powdery mildew	1
Unspecified fungi	1
Nematodes:	

Category	*# Requests*
Root knot nematode	2
Unspecified nematodes	1
Unspecified diseases	15
Insects:	
Aphids	2
Biological insecticides	1
Plant insect interactions	4
Psyllids	1
Tuta absoluta	1
Unspecified insects	2
Parasitic plants	3
Unspecified biotic stresses	2
Abiotic Stresses	
Drought	14
Heavy metals	1
High temperatures	10
Low temperatures	9
Nutrient deficiency	2
Salinity	10
Shade or high light	2
Unspecified abiotic stresses	11
Fruit Traits	
Carotenoids	3
Chloroplast accumulation	4
Cuticle/wax properties	2
Development and ripening	4
Flavor, volatiles, aroma	4
Food safety	2
Postharvest and shelf life	5
Quality	2
Sugars, solids	2
Miscellaneous Breeding	
Doubled haploids	2
Glasshouse cultivars	1
Grafting, rootstocks	9
Home garden cultivars	1
Male sterility	4
Marker assisted selection	3
Marker development	9
Perenniality	1
Processing cultivars	2
Wide hybrids	1
Wild species introgressions	1
Unspecified breeding uses	29
Genetic Studies	
Association mapping	2
Biosystematics	1
Canalization	1

Category	*# Requests*
Comparative genetics	1
Cytogenetics	1
Diversity studies, natural variation	6
Epigenetics	2
Evolution and domestication	1
Gene cloning	1
Gene expression / transcriptomics	13
Gene silencing	1
Genotyping by sequencing	1
Mapping	2
Phenotyping	5
Population genetics	4
QTLs	4
Sequencing	4
SNP genotyping	1
Transformation	1
Transposable elements	1
Unspecified genetic, genomic studies	1
Physiology & Development	
Abscission	3
Acyl-sugars	1
Bioactive small molecules	1
Cell walls	3
Cytokinnins	1
Flower, inflorescence development	3
Gibberellin responses	1
Gravitropism	1
Hormone responses	2
Leaf shape, development, meristems	2
Leaf variegation	1
Metabolites, metabolomics	6
Modified seed set	1
Mycorrhizae, rhizosphere	5
Photomorphogenesis, photosynthesis	3
Plant habit	1
Pollen biology	3
Reproductive barriers, mating systems	14
Root biology, architecture, exudates	5
Seed development, ageing, germination	3
Stomata	1
Trichomes, volatiles, exudates	7
Wounding, defense signaling	8
Miscellaneous	
Horticultural studies	2
Genebank exchanges, backup storage	3
Instructional uses	4
Unspecified uses	37

Our survey of the 2013 literature (and unreviewed papers of previous years) again uncovered 96 journal articles, reports, abstracts, theses, and patents that mention use of TGRC stocks (see Bibliography, at end of this report). Many additional publications were undoubtedly missed, and cases of utilization by the private sector are generally not publicized. This publication record demonstrates the important role of the TGRC as a research resource, and its positive impact on many fields of investigation. The value of the collection for improving the tomato crop is shown by the many publications that address economic traits.

DOCUMENTATION

Our database and website were modified in various ways by Tom Starbuck to address security issues, improve usability and add content. On our website (http://tgrc.ucdavis.edu), Tom updated the mapping tools that allow plotting of wild species accessions to meet new specifications for the GoogleMap interface. The web pages involved in submitting seed requests were modified with the latest information on phytosanitary restrictions, and to enable recharging researchers for express shipping options as well as purchase of phytosanitary certificates. Web pages with horticultural recommendations for growing the wild species were updated with our latest guidelines, and we added an illustrated tutorial on emasculating and pollinating tomato flowers.

Our database was modified in various ways to improve internal record keeping related to seed requests, plant pedigrees, and seed lots. Descriptive data on new accessions were added and records on existing accessions were updated as needed. In addition, we uploaded or edited geographic coordinates for a number of wild species accessions with data obtained using GoogleEarth. As usual, our annual distribution records were provided to the USDA for incorporation into the GRIN database, and we issued a revised stock list, this year covering the wild species accessions, through the Tomato Genetics Coop. Report (TGC).

RESEARCH

In addition to the core genebank functions described above, the TGRC conducts research synergistic with the overall mission of the Center. One research project, funded by the National Science Foundation, focuses on the genetics of interspecific reproductive barriers that restrict crosses between cultivated tomato and its wild relatives. Wentao Li previously isolated a pollen factor, *ui6.1*, involved in interspecific pollen rejection. The *ui6.1* gene encodes a Cullin1 protein with homology to similar proteins implicated in self-incompatibility in other plant systems. Using SI and SC biotypes of *S. arcanum,* he found that *ui6.1* also functions in self-incompatibility. This work was recently published in the journal *GENETICS.* He also continues to work towards isolating *ui1.1,* a pollen factor that interacts with *ui6.1,* and to study other pollen genes involved in interspecific incompatibility. Jennifer Petersen is studying natural variation in *ui1.1* and *ui6.1* among several green-fruited tomato species. Her research has so far identified several populations of *S. habrochaites* with mutations in one or both pollen genes. She is using these mutant populations to draw inferences about how self-fertilization (inbreeding) evolves in a normally outcrossing species like *S. habrochaites*.

In another research project, the TGRC is seeking to develop a set of breeding lines representing the genome of *S. sitiens*, a wild tomato relative known for its tolerance to drought and salinity, but which has not been utilized in the past due to strong crossing barriers. The goal of this research is to develop a set of introgression lines – prebred stocks containing defined chromosome segments from the donor genome – that will provide the first breeder-friendly germplasm resources for this wild species. Low resolution DNA marker analysis using a sample of families from early backcross generations (BC2-BC3) showed that roughly 80% of the *S. sitiens* genome has been captured so far. However, these lines are still at a very early stage and more backcrosses and marker aided selection will be needed to produce a useful set of introgression lines. We received a grant from the USDA-NIFA's Plant Breeding program to complete development of this resource. The steps involved will include recovering the missing genomic regions, testing for overlap between adjacent chromosome segments, isolating recombinants with shorter donor segments, and genotyping the resulting introgression lines to high resolution by sequencing.

PUBLICATIONS

Barrios-Masias, F. H., R. T. Chetelat, N. E. Grulke, and L. E. Jackson (2014) Use of introgression lines to determine the physiological basis for changes in water use efficiency and yield in California processing tomatoes. *Function Pl. Biol*. 41: 119-132.

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SERVICE AND OUTREACH

Presentations. We gave presentations on the TGRC, our research projects, and related topics to: the Marin Garden Society, HM.Clause World Corporate convention, and the Plant Breeding Academy (Seed Biotechnology Center, UC-Davis).

Press Coverage. We provided interviews or background information to the Wall Street Journal for an article on tomato transplants, and to Sarah Phelan from the UC Berkeley School of Journalism for a film project on tomato diversity.

Visitors. Representatives of the following institutions visited the TGRC: Olter Seeds; NRI Agritech, India; Nunhems USA; Advanta India; Kagome Co.; AVRDC – The World Vegetable Center

PERSONNEL AND FACILITIES

Maintaining a large and diverse germplasm collection and an active research program involves contributions from many individuals. Scott Peacock oversaw our seed regeneration program, aided by undergraduate students Daniel Short, Christine Nguyen, Kristine Donahue, Adryanna Corral, and Angela Prada-Baez. Jennifer Petersen managed our seed distribution activities, assisted by the undergraduate students, and continued her research on natural diversity for two pollen factors involved in interspecific incompatibility. Marcus Tamura helped Jennifer in her research, and several undergraduate students, Jackie Lui, Jessica Tom, Hanna Casares, and Kathy Tran, did internships. Wentao Li continued his research on the molecular genetics of intra- and interspecific incompatibility. Angel Fernandez Marti joined our group as a

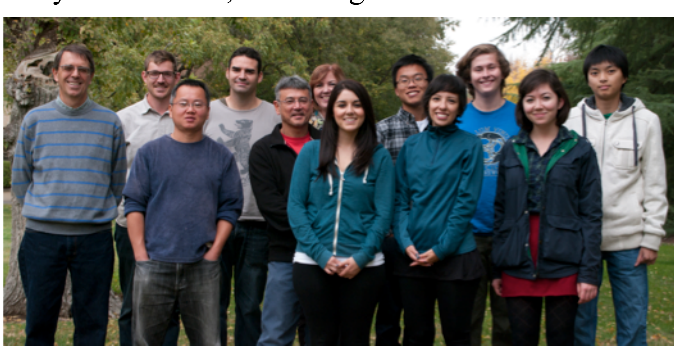

Figure 5. From left: Roger, Scott, Wentao, Angel, Tom, Jennifer, Adryanna, Marcus, Angela, Daniel, Kristine, Christine.

post-doctoral scholar to work on developing an introgression line resource for *S. sitiens.* Tom Starbuck continues to maintain our database and website. There were no significant changes in facilities.

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UC-Davis, College of Agricultural and Environmental Sciences
UC-Davis, Department of Plant Sciences
USDA – ARS, National Plant Germplasm System
USDA – NIFA (National Institute of Food and Agriculture)

TESTIMONIALS

"You have been doing great job with distributing the tomato seeds to all over the world."
-- Ozer Calis, Gaziosmanpasa University, Turkey

"We really appreciate the service. I really appreciate the TGRC site that let me focus on the genes we were interested in her working on, so I could get those lines."
-- Barbara Liedl, West Virginia State University

"I greatly appreciate the existence of the TGRC!"
-- Gregg Howe, Michigan State University

"We very much appreciate the work of the TGRC, it is a great contribution to the activities of many globally, and we will be very pleased to be acknowledging your contribution in future publications."
-- Mark Tester, King Abdullah University

"Thanks so much for the wonderful resource of the TGRC!"
-- Dan Chitwood, Danforth Plant Science Center

"…there are entire populations of people who will benefit from the work at TGRC for years to come."
-- Jennifer Ibarra, John Winthrop School

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