

SUMMARY REPORT
California Cotton Alliance Supported Project
2014 Project Activities to Date – September 2014

Prepared for review by the following groups: California Cotton Growers and Ginners Association,
California Cotton Alliance

TITLE: **Identification and development of cotton germplasm and potential breeding lines with improved Fusarium wilt (FOV) resistance, fiber quality, and yield**
(jointly funded project of the CA Cotton Growers Association and California Cotton Alliance)

For Support of Project in years: covering some 2013 and 2014 activities
Project operations planned for 2013 - 2016

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Budget: From one or multiple sources, project support needs by year – California operations support:

2013-2014: \$ 182,000 (1/2 of funds each year to be from CA Cotton Alliance, ½ from the research funds of the CA Cotton Growers Association)

2014-2015: \$ 232,000 (1/2 of funds each year to be from CA Cotton Alliance, ½ from the research funds of the CA Cotton Growers Association)

2015-2016: \$ 238,000 (1/2 of funds each year to be from CA Cotton Alliance, ½ from the research funds of the CA Cotton Growers Association)

Work status or initiated projects through September, 2014 are described in two components below:

- (1) Work in 2013 and early 2014 conducted in cooperation with a post-Doctoral fellow, Dr. Grant Poole (who was on staff from October, 2013 through mid-March, 2014; and
- (2) Work continued in 2014 that includes additional areas of work and that also builds on the work identified in #1.

OBJECTIVES (related to #(1) and #(2) above:

1. Develop cotton progeny and breeding populations segregating for FOV race 4 resistance. (approximately 2/3 of efforts will focus on Upland / Acala types of cotton, with remainder of effort on Pima germplasm).

For 2013 evaluations: Over 500 entries were sent from Mauricio Ulloa in Lubbock, TX representing populations derived from cooperative work with Bob Hutmacher and Steve Wright of UCCE in recent years. These entries were planted at one major field screening site for race 4 FOV in Tulare County, and evaluations of all entries were done in the summer and fall of 2013.

For 2014 evaluations: Over 410 entries were sent from Mauricio Ulloa of USDA-ARS in Lubbock, TX representing populations of interest. All of the entries were planted at the Tulare County FOV screening site in May, 2014 and about half of the entries were also planted at the Kern County FOV screening site that we have developed.

In addition, multiple entries (over 150 total) of Pima and Upland varieties were submitted for in field and greenhouse FOV-race 4 screenings at Kearney REC greenhouse location. All entries were evaluated for vascular staining, stunting and vigor index ratings, and survival of populations as influenced by race 4 fusarium at the sites.

2. Evaluate resulting progeny, breeding lines, and germplasm for FOV resistance, fiber quality traits.

Field screenings were conducted at each site (field in Kern Co., field in Tulare Co., greenhouse at Kearney REC) in summer and early fall, 2013. Progeny were grown in greenhouse for seed increase at Kearney REC, with plantings in mid-November. In addition, a small number of seed were collected from seed increase plantings made at the Kearney REC, with further seed increases done in the greenhouse and clean field site at Kearney REC.

In 2013, for selected entries, plant tissue samples were collected, sent to Lubbock, TX and frozen for later DNA molecular work related to molecular markers and screening approaches. Samples were collected by UCCE staff and submitted for cooperative evaluations to be done in the USDA ARS laboratories in Lubbock, TX. Improved laboratory equipment and setup were developed to some extent at Shafter to allow more detailed evaluations in cooperation with USDA-ARS in Lubbock, TX and UC Riverside cooperators.

3. Utilizing selected materials, maintain seed supplies through progeny propagation and breeding population increases either at field location(s) in California or in Tecoman, Mexico nursery.

Seed needed for selections to advance have been collected and will continue to be collected in the fall of 2013 from the Tulare Co fusarium screening site from: (1) selections made based on FOV resistance; (2) varieties for advancement based on desirable agronomic characteristics plus FOV resistance; and (3) materials of interest for molecular markers use in resistance screening efforts in CA or Texas.

4. As selected germplasm are advanced, also conduct trials to evaluate growth characteristics and yield performance (growth habit, growing season length requirements and yield performance) at West Side Research Center location.

Limited evaluations were initiated at Kearney for 9 lines we are hoping to advance in Pimas out of the existing race 4 resistant materials being developed. In addition, some select entries were also planted at the UC West Side REC for yield and fiber quality evaluations. Only evaluations to date will be for fiber quality, as the plot size and seed available not adequate for yield potential evaluations.

Progress with selections and increases (as of September, 2014):

September 2013 to early 2014 efforts (2013 FOV screening selections and continuing 2014 work with those entries, crosses):

- ✓ Mauricio Ulloa and Bob Hutmacher selected and provided around 512 entries from the above progeny: TM-1 x PS-6, PS-6 x TM-1, Shorty x PS-6, PS-6 x Shorty, Shorty x FBCX-2, FBCX-2 x Shorty, Phyto72 x STV474, and additional germplasm.
- ✓ Selected 50+ resistant/tolerant Upland and interspecific progeny – lines under infested FOV4 field on 2013.
- ✓ Seed were increased in the greenhouse of selected progeny – lines 2013-2014.
- ✓ Utilizing these 2013 selections initiated in the field, we are currently just starting in the 2nd Cycle of Selection under greenhouse high FOV4 fungus-concentration (inoculum concentrations up to 10⁻⁸ spores per ml of inoculation solution (about 100 times more concentrated than prior greenhouse FOV inoculation work we have done).
- ✓ Following additional selections done at the higher FOV inoculation levels in the greenhouse at Kearney, the next work with the best performing entries out of those tests will be to plant them as Advanced Breeding Lines at the 2015 FOV4 infested FOV4 field in Tulare County
- ✓ We will also use clean treated seed of these same entries for yield and fiber quality evaluations in trials done at the West Side REC to begin to collect more detailed yield and hvi data on the germplasm selected out of the FOV trials.

The first table shows the selections made in 2013 for which further evaluations were conducted in 2014 in the field and greenhouse, and these are entries for which seed increases are underway in 2014.

Selfed Seed Obtained for:	Entry #'s seed selection	Selections made and open pollinated seed obtained for:	Entry #'s seed selection
Shorty x PS-6	71,74,79,105	TM-1 x PS-6	5,35,37,51,55,46,54,38,40,60
PS-6 x TM-1	207,276	PS-6 x TM-1	182,207,211,252,276
PS-6 x Shorty	288	Shorty x PS-6	122,74,71,79,119,135,145,122,124,105,128
Shorty x FBCX-2	325,342,344,360,369,374,416,419,423,433, 434	PS-6 x Shorty	288
		Shorty x FBCX-2	439,391,429,435,422,438,424,400,426,437

Cultivar Selections and work initiated with new entries for 2014:

- ✓ New materials can be categorized into multiple experiments involving different sources of germplasm:

- Identified potential germplasm from 160 germplasm releases (1937 – 1999 yrs.) – these included materials out of the state of Mississippi (Stoneville and Delta and Pine Co., Delta Exp-Station, and USDA-ARS) – identified as experiment 1. These were multiple germplasm releases (1937 – 1999 yrs.) – Selected 13 cotton entries based on survival percentage and vascular stain rating primarily under FOV4 infested conditions in our Tulare County grower field.

With these materials, we used self-pollination and checked for transgenic contamination of selected entries for seed increases. The Pedigree/background of these entries was varied: Stoneville (7 lines) and DPL (1 line) companies, Delta Expt Station (1 line), and Mis (4 lines).

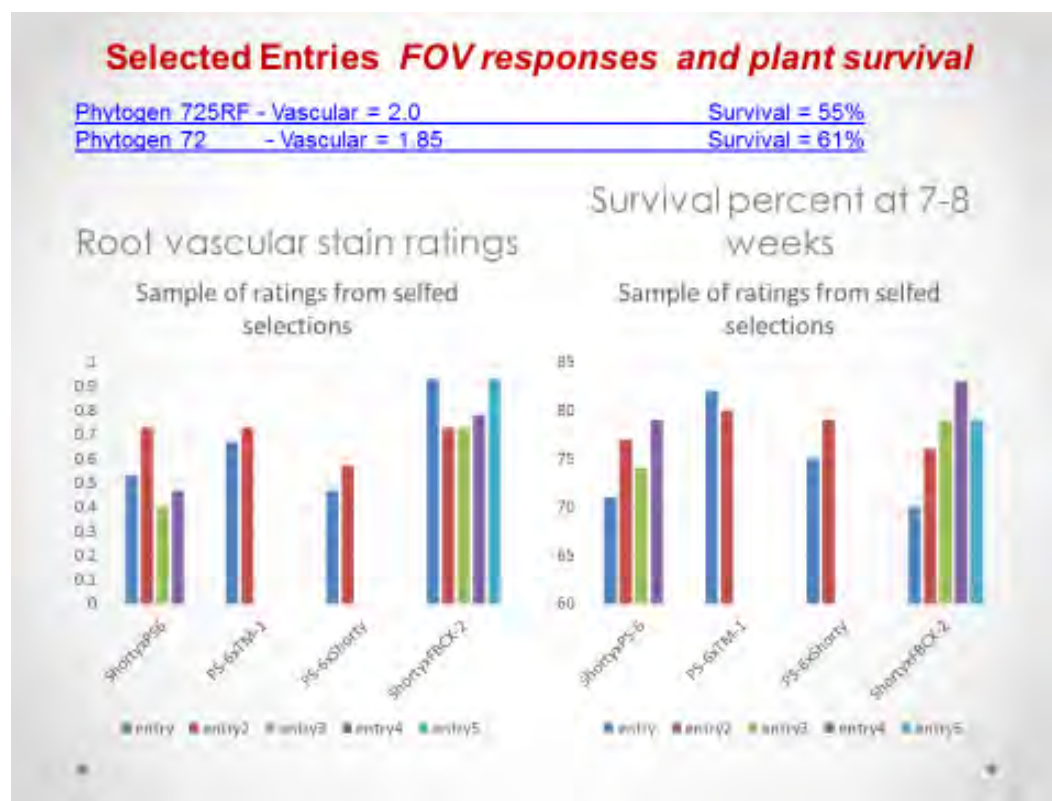
- An additional 270 entries were selected as potential germplasm of interest, and these were from the Cotton Germplasm Collection at the Tecoman, Mexico Winter Nursery by Mauricio Ulloa. This group is here identified as Experiment 2. This germplasm is from the Cotton Germplasm Collection maintained by the USDA. As in experiment #1 described above, we selected 11 entries based on survival percentage and vascular stain rating primarily under FOV4 infested conditions in our Tulare County grower field.

With these materials, we used self-pollination and checked for transgenic contamination of selected entries for seed increases. These 11 selections are candidates for advancement and will be moved into further high inoculum load testing in the greenhouse at Kearney REC to determine potential for further advancement and selections. The pedigree/background of these entries: MARS program, Uganda, Uzbekistan, and California old program donated to the collection in 1993.

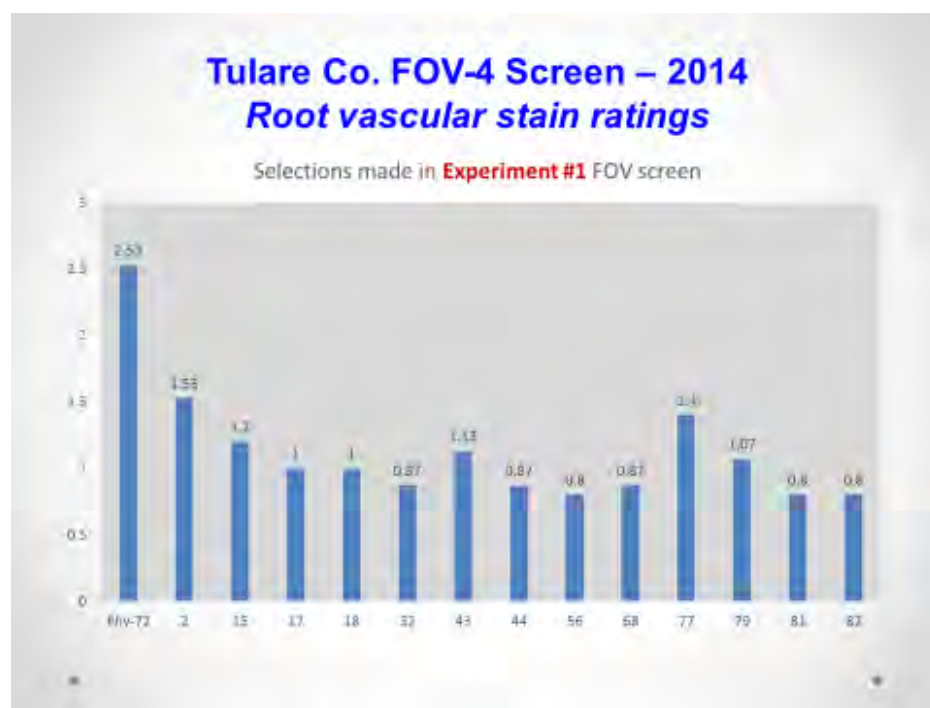
- In addition, USDA provided around 411 entries from the above Upland entries for testing FOV race 4 resistance in greenhouse and on infested field evaluations. Seed was increased in Lubbock, TX and also was provided by the cotton collection-curator.

All of the entries selected for advancement and more FOV-4 screening tests in both 2013 and 2014 were based on having better performance in terms of vascular staining (a relative indicator of infection level with race 4 FOV), and also plant survival in a severe FOV pressure locations.

Example data for specific UPLAND cotton entries that were selected for further FOV testing and possible advancement as improved resistance/tolerance materials is shown in the following figure, indicating very good levels of vascular staining (levels of 1.2 to 1.4 or less are target values for more tolerant cultivars) and better average stand survival under FOV-4 pressure. The values at the same site for the more moderately susceptible Acala cultivars Phytogen 725-RF and Phy-72 are also shown in the figure.



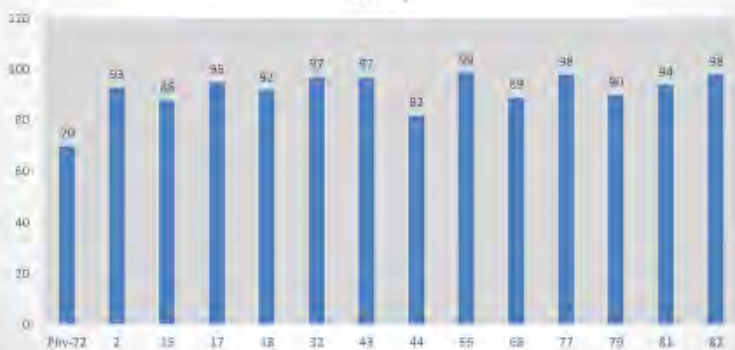
Similar selections were made in the experiments mentioned for different cultivars in 2014 in the field evaluations, and some limited selections made in experiment one at the FOV-race 4 test site in 2014 are shown in the two figures below (first one is vascular stain ratings, the second is stand survival values.)



Tulare Co. FOV-4 Screen – 2014

Survival percent at 7-8 weeks post emergence

Selections made in **Experiment #1** FOV screen (13 selections made from broad germplasm releases over decades from 1930's to 1990's)



These selections may in some cases be of direct interest based on cultivar performance in resistance/tolerance to race 4 FOV. However, it will in most cases be true that there may be value in trying to develop further improvements in agronomic characteristics, growth habit, yield or fiber quality. In those cases, in addition to the selections made in the various experiments, there are efforts being made to develop and evaluate crosses between cultivars / entries with the goal of further improvements in desired characteristics. Some of the crosses that were attempted in 2014 field trials are shown below (these are still out in the field as of the date of preparation of this report, so success with these crosses is unsure but looking good in multiple crosses at this time).

2014 Experiment Nos 1 and 2 - Selfing of flowers for seed increase of selected entries (selfed to reduce potential for cross pollination).

And

Made some specific crosses using selected entries as male and female parents in different **crosses-combinations** (table below)

CROSSES	2	56	68	90	714	715	716
2		X	X	X	x	x	X
56	x		x	x	x	x	X
68	x	x		x	x	x	X
90	x	x	X				
136	x	x	x	x			
307	x	x	x	x			
322	x	x	x	x			
323	x	x	x	x			

*As of 9/10/14, field evaluations suggest that so far we generally have over 20-30 bolls set from most of the entries selfed; and as of recent evaluations, we hope to have 3-5 bolls set from each of the crosses made.

For questions regarding any parts of this overall project, please contact Bob Hutmacher at rbhutmacher@ucdavis.edu and I will try to answer any questions or direct them as needed to Mauricio Ulloa or other cooperators. Thank you for support of this work.