Genomic Project Description:

Development and Phenotypic Evaluation of Recombinant Inbred Line (RIL) Populations for Mapping and Marker Development.

C. C. Holbrook et al.

Project Fiscal Year:

2011

Final Report and Executive Summary:

The primary objective of this work is to develop 16 structured mapping populations for the peanut genetic research community. These populations were advanced using summer nurseries on the mainland, and the peanut winter nursery in Puerto Rico. We have begun seed increase for some of these populations to provide the larger research community with material for phenotyping. Ultimately, these populations will be used to develop an improved genetic map for the cultivated peanut species. The parents were also selected to create populations that can be used to develop molecular markers for important traits that will improve the efficiency and effectiveness of U.S. peanut breeding programs. The information and populations from this project will also be valuable assets that the peanut research community can use to secure funding from additional sources for genomics research on peanut.

I. Abstract

Project Title: Development and Phenotypic Evaluation of Recombinant Inbred Line (RIL) Populations for Mapping and Marker Development

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Summary: The primary objective of this work is to develop 16 structured mapping populations for the peanut genetic research community. These populations were advanced using summer nurseries on the mainland, and the peanut winter nursery in Puerto Rico. We have begun seed increase for some of these populations to provide the larger research community with material for phenotyping. Ultimately, these populations will be used to develop an improved genetic map for the cultivated peanut species. The parents were also selected to create populations that can be used to develop molecular markers for important traits that will improve the efficiency and effectiveness of U.S. peanut breeding programs.

We also completed the development of a late generation RIL population from the cross Gregory x Tifguard. This population was planted in a replicated field study, and measurements were taken on plant, pod, and seed morphology, and on resistance to TSWV and leaf spot. These phenotypic data will be essential to the mapping and marker work that will be conducted by cooperating molecular geneticists.

II. Main Body of Report

Project Title: Development and Phenotypic Evaluation of Recombinant Inbred Line (RIL) Populations for Mapping and Marker Development

Project Investigator: C. Corley Holbrook
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Objectives:

Development and phenotypic evaluation of recombinant inbred line (RIL) populations, along with molecular genotyping, will be essential for marker development. The primary objective of this research will be to develop 16 structured RIL populations that can be used by the peanut research community to conduct genetic research for mapping and marker development. Additionally, we will continue phenotyping on a late generation RIL population that Dr. Peggy Ozias-Akins will be genotyping.

Procedures:

A large group of peanut breeders and geneticists meet in the summer of 2008 to begin planning a submission for a CAPS grant. Much of that meeting was devoted to the selection of the most appropriate parents to use for the development of the RIL populations that will be needed for mapping and marker development. Two common parents (Tifrunner and Florida-07) were selected to cross with eight unique parents (Bailey-HO, C76-16, NC3033, NM Valencia A, Olin, SSD6, SPT 06-6, and Florunner). The eight cross combinations with Tifrunner were completed in time to send to the 2008/09 winter nursery. DNA was sampled from these F_1 plants and frozen for future use. F_2 populations were grown in 2009 at Tifton, GA, and Raleigh, NC. Four-hundred F_2 plants were individually harvest from each of the eight populations, shelled, and seed were packaged and sent for generation advance in the 2009/10 winter nursery. F_1 seed for the eight cross combination with Florida-07 were also sent to the 2009/10 winter nursery. Dr. Tom Isleib has submitted a grant proposal for the land cost (and some of his labor costs) for these 3200 plots. These 3200 plots are in addition to the regular nursery. This will result in much greater harvest and shipping costs. In addition, much labor will be needed to process these samples to prepare them for planting in the summer of 2011. With the addition of the other eight cross combinations, an even greater effort will be needed to prepare material for the 2010/11 winter nursery.

We have completed the development of a late generation RIL population from the cross of Gregory x Tifguard. We planted $F_{6:8}$ plots of this population at Tifton in 2010. Measurements were taken on plant, pod, and seed morphology, and on resistance to TSWV and leaf spot. This study was repeated in 2011. These data will be essential to the mapping and marker work that will be coordinated by Peggy Ozias-Akins.

Results and Discussion:

The progress and status for development of the 16 structured RIL populations is presented in Figure 1 (Below). Each of the 16 populations are represented by 400 progeny.

Figure 1. Outline for the development of 16 structured recombinant inbred line (RIL) populations.

Year	Set A (Location)	Set B (Location)
2008	Produce hybrid seed (T & R)	
	F_1 plants (PR)	
2009	F_2 population (T & R)	Produce hybrid seed (T & M)
	F_3 population (PR)	F_1 plants (PR)
2010	F_4 population (T & R)	F_2 population (T & M)
	F_5 population (PR)	
2011	F_6 population (T & R)	F_3 population (M)
	Individual Plant Harvest	F_4 population (PR)
2012	F_7 line increase plots (T & R)	F_5 population (M)
	Begin phenotyping and genotyping	
2013	Continue seed increase as needed	F_6 population (M)
	Continue phenotyping	Individual Plant Harvest
2014	Continue seed increase as needed	F_7 line increase (M & T)
	Conclude phenotyping	Begin phenotyping and genotyping
2015		Continue seed increase as needed
		Continue phenotyping
2016		Continue seed increase as needed
		Conclude phenotyping

Set A = Tifrunner x Bailey High O/L; Tifrunner x SPT06-6; Tifrunner x C76-16; Tifrunner x NC 3033; Florida 07 x Bailey High O/L; Florida 07 x SPT06-6; Florida 07 x C76-16; and Florida 07 x NC 3033

Set B = Tifrunner x SSD6; Tifrunner x Olin; Tifrunner x New Mexico Valencia A; Tifrunner x Florunner; Florida 07 x SSD6; Florida 07 x Olin; Florida 07 x New Mexico Valencia A; Florida 07 x Florunner

Location: T = Tifton; R = Raleigh; M = Marianna; PR = Winter nursery.

A replicated field study was conducted to evaluate the Gregory x Tifguard RIL population. Field and greenhouse studies were also conducted to characterize resistance to late leaf spot (LLS), caused by *Cercosporidium personatum*. Gregory has previously been characterized as susceptible, and Tifguard as moderately resistance to LLS. This biparental population consisted of 78 $F_{7:8}$ lines and was grown with the parents in three replications in a non-sprayed field trial. The leaf spot intensity was assessed four times during the season using

the traditional Florida 1 to 10 severity scale and twice using the novel combination of lateral stem assay (LSA) and image analysis. Three lateral stems were randomly collected from each plot across the three replications in the field, and the leaves were then subjected to imaging by a flatbed scanner. The image analysis and manual phenotyping on these lateral stem samples yielded data on percent defoliation, percent area covered by disease, number of lesions per unit area etc. This RIL population was also evaluated in a detached leaf stugy with artificial epiphytotics and data on LLS resistance components was collected.

The development of the 16 structured RIL populations will be a valuable asset for the peanut genomics community. This should greatly strengthen any future proposal that the group submits for grant funding. Ultimately, these populations can be used to develop an improved map for the cultivated peanut species. The parents were also selected to maximize the probability of developing markers to important traits that will be useful to all U.S. breeding programs.

The Gregory x Tifguard RIL population is developed and ready to use for mapping and marker development. Phenotypic evaluation will be essential for the success of these efforts. The successful completion of this research will demonstrate the feasibility and value of further mapping and marker development using the 16 structured mapping populations. This can be used to strengthen any future proposal that the group submits for funding to support genomics research.