

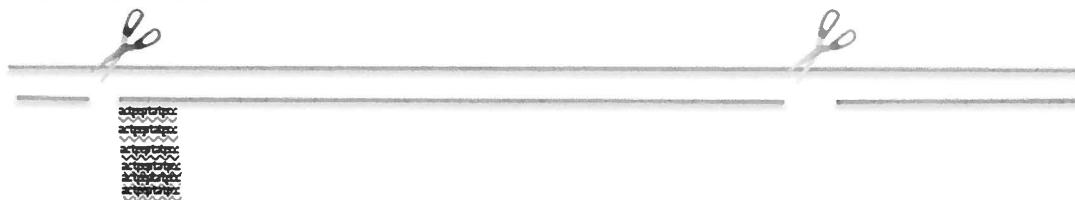
## Final Report: Genotyping by sequencing.

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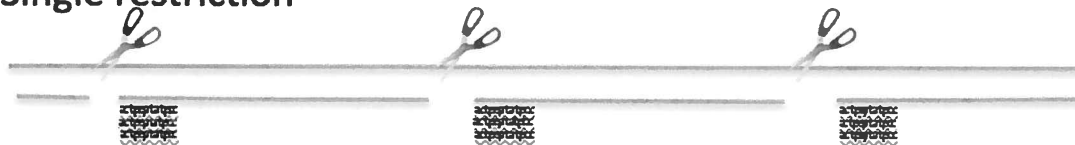
We were focused on populations segregating for early and late leaf spot, problems important to US peanut growers. Parents either resistant or susceptible to these pathogens were used to create populations (prior to beginning of grant) and were continuously phenotyped to provide marker-trait associations.

Two graduate students, both partially associated with this specific project, but both working on issues related to high throughput genotyping in peanut, participated in developing genotyping-by-sequencing (GBS) tools/technology using the parents of these populations. Due to the recent polyploidy, genotyping, by any method, is difficult in peanut. The two students (Carolina Chavarro and Josh Clevinger) used two different, but complementary approaches. 1) working with sequences derived from transcribed genes, Josh has worked on computational approaches to define genetic variants to specific peanut subgenomes. To do so, he has been using the emerging diploid ancestor genome sequences from the Peanut Genome Initiative. 2) Carolina has used a two-enzyme approach to sequence enzyme-associated fragments from the peanut genome. This approach is new and should reduce the complexity of the peanut genome (see figure below). Previous versions used single enzyme digestion which resulted in many more fragments to be sequenced and thus reducing the amount of coverage at any one site. This approach should lead to increased coverage and better variant calls for assigning to subgenomes in peanut. She is still working on computational analysis of the data (having just received it October 2013) and is also using data from the Peanut Genome Initiative to help refine her analyses. Once these approaches are confirmed and quality controlled, they will be applied more generally to the segregating populations to make and refine the marker-trait associations after which they will be tested in the Holbrook breeding program at Tifton. Thus, these results continue to feed into the goal of making good trait-marker associations and developing tools/technology to advance these associations to breeding programs for implementation into advanced cultivars.

### Double restriction



### Single restriction



Funding was used to support the students as well as the technicians necessary for the laboratory, field and especially the computational component.