**Research Project:** <u>Developing Strategies to Identify Useful Genes in Peanut and Breeding</u> <u>High Yielding Peanut Varieties and Germplasm</u>

## Location: Peanut Research

Title: Workflow to study genetic biodiversity of aflatoxigenic Aspergillus spp. in Georgia

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**Interpretive Summary:** As part of our Aspergillus biodiversity studies, we sequenced the genomes of 8 A. flavus isolates, three of which are nonaflatoxigenic, and one isolate of A. parasiticus. The data were uploaded to the public in a National Database, NCBI GenBank. The information and analyses obtained will allow studies on population structure to determine how the genetic diversity in section Flavi species is changing, and to recognize recombination events. Furthermore, it will allow the determination of evolutionary processes and the direction of this evolution.

**Technical Abstract:** Peanut seeds were sampled from the entire state of Georgia in 2014. More than 600 isolates of Aspergillus spp. were collected using modified-dichloran rose Bengal (MDRB) medium, 240 of those isolates were fingerprinted with 25 InDel markers within the aflatoxin-biosynthesis gene cluster (ABC). Cluster and Structure analyses were performed and genomic DNA of 10 isolates representing various clades were sequenced using illumina® Hiseq2500 at the UW-htSEQ, Seattle,WA. All analyses performed (Neighbor-Joining, 3D-Principal Coordinate Analysis, STRUCTURE) revealed that the Aspergillus isolates sampled in this study were grouped by their capacity to produce aflatoxin. Three main groups were obtained: Group I comprised of ten non-aflatoxin and non-cyclopiazonic acid producers, including one commonly used as biocontrol; Group II included all the aflatoxin B and G producers, A. parasiticus; and Group III, the largest, mostly included aflatoxigenic A. flavus except for three A. caelatus that conformed a sister cluster themselves. Here we propose a workflow to screen isolates for aflatoxin production and genotypic variations in ABC by fingerprinting with InDel markers using capillary electrophoresis. Cluster analysis allowed selecting few representatives within clades for whole-genome sequencing, which provided DNA information without sequencing all the individuals. Determining genetic diversity in section Flavi will provide valuable molecular information to select appropriate target genes to control aflatoxin accumulation in crops.