

TITLE: Physiological Responses and Functional Genomics of Cultivated Peanut for Improved Tomato Spotted Wilt Virus Resistance

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OBJECTIVE: To improve tomato spotted wilt virus (TSWV) resistance in peanut. We expect to achieve the objective of this proposal by pursuing the following *specific aims*:

- 1) Identify genes whose expression is associated with an increase in TSWV resistance.
- 2) Examine variation in genetic expression among peanut cultivars following TSWV infection using the genes identified in this project and previously cloned peanut genes.

RATIONALE AND SIGNIFICANCE: Our *long-term goal* is to develop peanut cultivars that are highly-resistant to TSWV, one of the most devastating economic threats to peanut production. Annual losses of \$40 million for Georgia alone have been attributed to TSWV. Because the disease is vectored by thrips, insecticide treatments are commonly used to control thrips populations. The insecticide phorate (Thimet 20G and Phorate 20G) has demonstrated suppression of TSWV, however the level of thrips control obtained with phorate is not greater than for other insecticides. Therefore, the actual mechanism of TSWV suppression by phorate treatment is unknown. Our *central hypothesis* is that in peanut, the oxidative stress resulting from phorate treatment elicits a defense response in the plant that allows it to better tolerate TSWV. If the genes involved in this defense response can be identified, they can be used as TSWV-resistance selection markers for peanut improvement.

In a previous two-year experiment and subsequent field tests examining different peanut cultivars under insecticide and control conditions, we have identified physiological traits that are altered following phorate treatment in peanut. The next step in this process is to develop specific genetic markers that can be used to select TSWV resistant genotypes that have superior physiological defenses. We are proposing to identify tissue-specific peanut expressed sequence tags (ESTs) related to the physiological disease responses we have quantified. The ESTs will provide numerous clues to the identities of genes and pathways crucial to TSWV resistance.

EXPERIMENTAL PROCEDURES: EST development has already been set as the number one national research priority by USDA and University peanut researchers (Peanut Genome Initiative). From a genotype whose physiological traits we have already well-characterized, we have been constructing cDNA libraries from leaves of control plants and phorate-treated plants, both with and without TSWV infection for a total of 4 treatments.

RESULTS AND CONCLUSIONS: cDNA libraries were made and sequenced. We obtained approximately 200,000 reads of an average length of 215 base pairs. The ESTs were analyzed by grouping them into unigene sets using appropriate software tools. The non-redundant sequence were searched against the UniProt reference database using the BLASTX program. The BLAST search results are being used to annotate the EST sequences using Gene Ontology terms. The EST sequences will also be analyzed for conserved protein domains using standard domain finding algorithms.