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INCREASING RESISTANCE TO ROOT-KNOT NEMATODE IN COMMERCIAL PEANUTS FOR THE SE

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Objective 1: To use the PCR marker to develop nematode resistant cultivars adapted to the southern US.

Results: Small-scale DNA extractions from frozen leaf material using a previously described method produced inconsistent PCR results, similar to previous larger-scale extracts, due to poor quality of starting material. An alternative DNA extraction method (partial nuclei purification) from frozen leaves to reduce potential carbohydrate contamination was tested. This method allowed extraction for DNA suitable for PCR from all samples. DNA extraction from peanut seed using a recently published method from our lab produced much more consistent quality DNA for PCR, although concentration varied and should be adjusted for each sample. The PCR marker was then used to screen samples from a NC94002/ NemaTAM cross and a AP3/ NemaTAM cross. Those samples that were positive were kept and carried forward in the breeding program.

Objective 2: To identify nematode resistance gene/s (introgressed portion of wild *Arachis cardenasii*) in NemaTAM that specifically confers resistance to root knot nematodes, and to identify differentially expressed genes that are specifically expressed in nematode resistant NemaTAM, but not in susceptible Florunner.

Results: To clone and characterize NemaTAM resistance genes (*R*-genes) that may confer resistance to the root-knot nematode, degenerate PCR primers were used to amplify the conserved nucleotide binding site (NBS) encoding regions of *R*-genes from NemaTAM. Analysis of 192 clones revealed that 80 sequences had homology to genes belonging to the section *Arachis*, 21 had significant sequence similarity to genes found in other plants, and eight were related to genes found in animals. Interestingly, two NemaTAM clones shared conserved motifs found in the well-characterized tomato nematode resistance gene, *Mi*, and an *Arachis cardenasii* *R*-gene. The cloned peanut *R*-genes will be further tested to determine linkage with nematode resistance. To identify differentially expressed genes in NemaTAM and Florunner in order to understand the nematode resistant and susceptible responses in peanut, suppression subtractive hybridization was used. First, a cDNA library enriched with differentially expressed sequences from nematode infested root tissues of NemaTAM and Florunner was constructed. Randomly picked clones (480) from tester NemaTAM-specific and driver Florunner-specific libraries were

used for differential screening. This process yielded 140 NemaTAM-specific clones, many involved directly in plant defense, and 123 Florunner-specific clones that were related to calcium and abscisic acid signaling. This work provides the foundation for the identification and study of genes involved in the molecular defense mechanism of nematode resistance in peanut.

Objective 3: To determine the comparative tolerance of commonly grown peanut varieties to root-knot nematodes.

Results: A field trial was conducted in 2006 to determine the comparative yield of seven peanut cultivars grown in a field naturally infested with root-knot nematodes (Meloidogyne arenaria). Varieties planted in the trial were Hull, AP3, GA Green, Carver, Andru II, C99R, and NemaTam. The test was arranged in a split plot design with Temik 15G nematicide treatment and no treatment as main plots and peanut varieties as subplots. Subplots were replicated 5 times. Plots were two rows wide and 15 feet long, and the test was planted on May 26, 2006. Temik 15G was applied at 10 lbs./A in the furrow at planting. Since Temik 15G suppresses thrips populations, Phorate 20G was applied at 5 lbs./A at planting to reduce thrips populations in the plots not treated with Temik. The peanut crop was maintained according to standard IFAS recommendations. Two digging dates, October 9 and October 23, were employed due to variety maturity differentials. Pod yields were determined from individual plots, and treatment averages converted to yield in lbs./A.

Table 1. Differential tolerance of seven peanut cultivars and lines to damage caused by the peanut root-knot nematode.

Variety & Treatment	Yield Differential[1]	Yield lbs./A
NemaTam + Temik	13%	2231
NemaTam - Temik		1974
Hull + Temik	1%	2862
Hull - Temik		2807
AP3 + Temik	8%	2768
AP3 - Temik		2565
GA Green + Temik	5%	2967
GA Green - Temik		2822
Carver + Temik	14%	2580
Carver - Temik		2260
Andru II + Temik	5%	2657
Andru II - Temik		2531

C99R + Temik	5%	2700
C99R - Temik		2575

[1]The lower percentage yield increase from Temik 15G application indicates a greater resistance or tolerance of the varieties to the peanut root-knot nematode.

The season was unusually dry and peanut yields were generally lower than in the previous 2005 trial. As a result, yield differentials within each variety between Temik and no Temik treatment. This trial, unlike that in 2005, showed little nematode tolerance differentials among varieties. A similar trial will be conducted in 2007, and results will be interpreted from three years average data. From both the 2005 trial data and grower field observations, Hull and AP3 are expected to continue to show good nematode tolerance compared to the other varieties.