Mississippi Peanut Growers Association
Title: Fungicide Suppression of Southern Stem Rot Infections - Final Report
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Mississippi State University Fund 345449-011900-037000

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Production of sclerotial inoculum
In December of 2013, sclerotia of 21 Sclerotium rolfsii, isolates were placed on Potato Dextrose Agar (PDA) in Petri plates. Germination percentage ranged from 50%-95%. Some of those not germinating seemed to die from fungal parasites. This is a problem reported in the literature. The published protocol to get around the problem is to grow the sclerotia on soil, but the growth process was not fully outlined. A small study was undertaken to find the best way to produce a lot of sclerotial inoculum.

Mycelia from the edge of vigorously growing, contamination free Sclerotium rolfsii isolates plated on PDA was placed on either oats, rye, or millet seed which had been amended 2:1 with water and autoclaved for 1 hour on two consecutive days. The bottles containing the grain were incubated in the dark at 30°C. The bottle contents were shaken once a day to distribute the growing culture in the bottle.

After 10 days of growth the grain culture was removed from the bottles and either dried on newspaper placed in either in a dark area or in a laboratory with windows or the grain was spread on sand contained in a food storage container to dry. The containers were wrapped in plastic and placed in a dark growth chamber at 30°C, in a 23°C room with low light or in growth chambers with no plastic wrap, low light and 23°C.

None of the cultures placed in the laboratory produced sclerotia. A few sclerotia were produced in the dark growth chambers. A moderate number of sclerotia were produced in the poorly lit room and growth chamber. Sclerotia were grew best on millet.

The health of the sclerotia were assessed by placing 5 sclerotia on each of three different Petri plates containing PDA. There was a 99% germination rate from sclerotia raised on millet. Very few colonies formed by the germinated sclerotia were contaminated with other fungi.

Growth chamber, soil and plant establishment
A source of washed sand was located and protected. A soil analysis was done on the sand and interpreted for the needs of peanut crop.

Five growth chambers were lamped as needed, and the temperature adjusted to 30°C using a thermometer with its bulb placed in a Erlenmeyer containing distilled water. Relamping consisted of
examining the fluorescent and incandescent lamps and replacing the burned out incandescent and fluorescent lamps with darkened ends. The end sockets on two lamps in one chamber had to be replaced due to electrical arcing. The photoperiod was adjusted to 16 hours, about equal to that near the end of July when stem rot starts to become a problem.

Plastic 8-in.-dia. plastic pots were filled with washed sand. Non-absorbant cotton balls were placed over the drainage holes in each pot to keep the sand from washing out. Each pot was amended with a measured amount of calcium, phosphorus and potassium according to the soil test recommendation.

Twenty pots were placed in each of five growth chambers. The soil temperature was allowed to adjust to the ambient chamber temperature.

On December 18, 2013 each pot was planted with Georgia 06 G Dynasty treated seed that had been purchased in the spring of 2013. The seed had been wrapped in three layers of plastic and stored at 4° C. Six seeds were planted in a star shape centered in each pot. Immediately after planting the pots were drenched with a 200 ml solution of Optimize lift and Abound at labeled rates.

Fots were watered as needed to keep the sand slightly moist. Germination took about 6 - 14 days. About 2/3 of the 600 seeds were lost to seedling disease (Aspergillus flavus, Aspergillus niger, Rhizoctonia) and were replanted. Another ca. 2/3 of those were lost to seedling disease and were replanted. This cycle continued until the disparity in plant ages became excessive. Pots were thinned to the healthiest three plants.

Two pots in each of five replications were inoculated with sclerotia of thirteen different isolates of Sclerotium rolfsii on April 8, 2014. One pot of each isolate in each replication was sprayed with the medium rate of the active ingredient tebuconazole (trade name Muscle). The thirteen isolates had been drawn from peanuts growing in four different production areas of Mississippi.

Plants were examined every third day for infection, and formal readings taken every 14 days.

Most inoculated but unsprayed plants became infected within 14 days of inoculation, but several plants required up to 20 days to become infected. With a single exception, inoculated but sprayed plants did not become infected.

On May 15, 19 of the 65 inoculated but untreated (sprayed with tebuconazole) plants remained alive, and were apparently uninfected. All but 7 of the inoculated and sprayed (with tebuconazole) plants were alive. One of these plants was killed by Sclerotium rolfsii. The others were killed by Aspergillus niger. There was no growth of sclerotia after the fungicide application.

Analysis of the data appears in Table 1. The overall model was significant, but R-square was moderate, especially for a growth chamber trial, and the coefficient of variation was high.
Table 1. Analysis of variance of the growth chamber data.

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| Corrected Total | 103 | 2011.759615

R-Square  Coeff Var  Root MSE  Rating Mean
0.486772  85.67631  3.896624  4.549077

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Rebuild of the field sprayer used in 2013
The field spray system used in 2013 was damaged by some vandalism and heavy rains and winds encountered during transportation and overnight sprays, Valves, nozzles and solenoids were replaced.
There was no difference among growth chambers or position of the pots inside the growth chambers. There was a significant difference due to application of a fungicide. Application of the fungicide reduced plant mortality. There was no germination of sclerotia after fungicide application.