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Title: "Determine level of resistance to fungicides in Georgia isolates of Aspergillus section Nigri associated with peanut seeds"

PI and Co-PIs: Renee S. Arias, Brian S. Jordan, Albert K. Culbreath, Valerie A. Orner, Victor S. Sobolev and Marshall C. Lamb

SUMMARY:

Initially, we had planned on using only 3 doses of each of six fungicides, and the goal was to evaluate either spore germination or mycelium growth as indicated in **Table 1**. However, once we had a protocol in place for the experiments, and having collected preliminary data, we decided to increase the number of fungicide concentrations to 6 in addition to the two controls [dimethyl sulfoxide (DMSO), and DMSO + salicylhydroxamic acid (SHAM)]. We also increased the number of observations to: quantify spore germination, quantify mycelium growth, and record sporulation in all fungicides at all concentrations.

Table 1. Fungicides and evaluations originally planned for the experiments

Mode of Action	Fungicide	Abbreviation	Evaluate	Evaluate
Strobilurin (QoI)	Azoxystrobin	AZO	spore germination	
Strobilurin (QoI)	Pyraclostrobin	PYR	spore germination	
Carboxamide SDHI	Fluopyram	FLU	spore germination	mycelium growth
Carboxamide SDHI	Penthiopyrad	PEN	spore germination	mycelium growth
Ergosterol Methylation inhibitor Triazole	Prothioconazole	PRO		mycelium growth
Ergosterol Methylation Inhibitor Triazole	Tebuconazole	TEB		mycelium growth

In order to give *Aspergillus* isolates similar incubation times for evaluations, we used 6-well sterile tissue culture plates; thus, at 16 h incubation we take photographs using a microscope for spore germination, after another 16 h we take photographs using a stereoscope for measuring colony size, and 72 h after that we examine spore formation using a stereoscope. Spore germination and mycelium growth require for quantification further processing of the pictures using the Leica software where the photograph conditions are tabulated. Whereas spore formation is recorded by visual assessment (on stereoscope) as presence/absence.

Give the current situation with Covid-19, and being unable to hire a student help, we are processing few isolates at the time. So far, we have obtained 2725 photographs, in addition to another 600 data points on sporulation. Photographs are currently being processed. The operation procedure is slow, however, we believe it reduces human error, gives similar incubation time to the isolates for comparison, and provides a permanent record of the results that can be re-analyzed if needed.