Analysis of late leaf spot fungi for resistance to leaf spot fungicides

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Group 11 fungicides, also known as QoIs, include azoxystrobin (Abound), pyraclostrobin (Headline) and several other active ingredients. These fungicides have provided excellent leaf spot control in peanut for more than a decade, but apparent control failures in 2017 and 2018 suggested that resistance to group 11 fungicides could be present in late leaf spot populations in North Carolina. Field research at Lewiston in both years showed very poor leaf spot control when group 11 fungicides were used for exclusively for disease control, providing further evidence for fungicide resistance there. We sought to develop a rapid assay for QoI resistance and use it to confirm the presence of QoI resistance and determine the relative frequency and geographic distribution of resistance in the late leaf spot fungus in North Carolina.

For many fungi, fungicide resistance can be assayed fairly rapidly by growing the fungus in culture at different doses of the fungicide and recording colony growth (Figure 1). The fungus is considered resistant if it grows at doses that would kill more sensitive types. The late leaf spot fungus is extremely slow growing and produces tiny colonies, making this typical assay in culture impractical.

Leaves were collected from infected peanut plants from 33 locations in eastern NC in 2018 and preserved by drying in a plant press. A rapid assay to verify the presence of known QoI resistance mutations was developed and tested. This assay allows extraction of DNA directly from lesions so that culturing the fungus is not necessary. DNA was extracted from 8 lesions per plant and PCR was performed to amplify regions of DNA around the QoI mutations. The resulting DNA pieces were sequenced and compared to sequences associated with known resistance mutations. A high frequency of QoI resistance was found among isolates collected across North Carolina. Thirty of 48 samples (62.5%) were resistant mutants, 12 (25%) were sensitive wild-type, and 6 (12.5%) were from mixed wild type and mutant lesions. Resistant samples were found dispersed across all areas sampled (Figure 2). Because resistance was so wide spread, it is unlikely that group 11 fungicides can be used successfully to control late leaf spot in North Carolina. This makes it even more critical to preserve efficacy of fungicides that currently are used for leaf spot control through wise resistance management.

Figure 1. Example of a typical fungicide resistance assay comparing fungus growth in culture at different fungicide concentrations. Resistant types grow at fungicide concentrations that inhibit sensitive types.

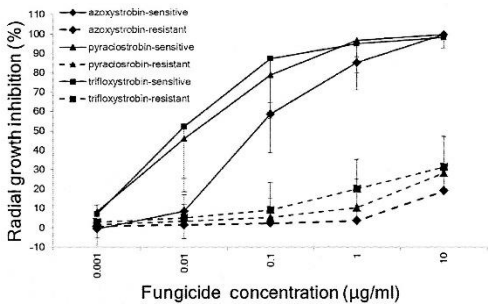

Figure 2. Distribution of QoI resistance in late leaf spot samples collected in North Carolina. Red points indicate QoI resistant samples, blue points indicate sensitive samples, and purple points indicate mixed resistant and sensitive lesions in the sample.