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Title: Management Strategies of a *Sclerotinia homoeocarpa* Population with Multiple Fungicide Resistance and Multidrug Resistance

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Objectives of the project:

- 1. To assess field efficacy of dicarboximide, DMI and SDHI fungicides in a dicarboximideresistant *S. homoeocarpa* population.
- 2. To develop the best fungicide options for controlling a *S. homoeocarpa* population with multiple fungicide resistance.
- 3. To understand how many applications of non-dicarboximides are required in order to revert a dicarboximide-resistant population into sensitive by monitoring the population shift.
- 4. To determine how persistent the reverted dicarboximide-sensitive population will be after reversion.

Start Date: 2016 Project Duration: Two years Total Funding: \$20,000

Summary Text:

Dollar spot, caused by *Sclerotinia homoeocarpa* F.T. Bennett, is one of the most significant diseases of cool-season turfgrass on golf courses. Resistance to the benzimidazole and dicarboximide classes and reduced sensitivity to the sterol demethylation inhibitor (DMI) fungicide class in *S. homoeocarpa* populations has been reported, moreover, a select number of golf courses also contain *S. homoeocarpa* populations with resistance to dicarboximide (iprodione and vinclozolin) sensitivity. In order to better understand the dynamics of multiple fungicide resistance (MFR) population and to develop fungicide options for dollar spot control, we conducted a field trial in 2015 and 2016 on a golf course fairway with a multiple fungicides-resistant *S. homoeocarpa* population.

During the 2015 and 2016 field season, fungicide efficacy was tested on two different fairway locations at Wethersfield Country Club (WCC) in Connecticut and the population exhibited a combination of four different isolate genotypes with differing resistance profiles to the benzimidazole, dicarboximide and DMI fungicide classes. Field efficacy data in 2015 and 2016 showed a fairly similar trend. Reduced field efficacy was observed using the following fungicides: iprodione (Chipco 26GT), vinclozolin (Curalan), and low rate of propiconazole (Banner MAXX II). On the other hand, good control was observed with high rate of boscalid (Emerald), fluxapyroxad (Xzemplar), fluazinam (Secure), and Enclave (Fig. 1).

The 2014-Initial *S. homoeocarpa* population displayed a higher proportion of DMI-R/Ben-R and DMI-R/Dicar-R/Ben-R isolates than the proportion of DMI-R and DMI-R/Dicar-R isolates. The 2016-Initial sampling had a higher proportion of DMI-R isolates than DMI-R/Dicar-R isolates (except for boscalid at 0.28 kg a.i. ha⁻¹ treated plots), and the DMI-R/Ben-R or DMI-R/Ben-R/Dicar-R isolate phenotypes were at the lowest proportion among all isolate phenotypes or absent. DMI-R/Ben-R/Dicar-R isolates disappeared over the growing season in 2014 and 2016 (Fig. 2).

The proportion of DMI-R/Ben-R isolates on the untreated plots decreased and the proportion of DMI-R isolates increased after 2014-2015 overwintering. The *S. homoeocarpa* population's phenotypic structure on the untreated plots in 2015 showed a similar proportion to the Initial population in 2016, despite these being different plot locations. Isolate phenotype proportion on the untreated plots did not significantly change during the 2016 growing season according to repeated measures MANOVA. This was the only treatment that did not significantly change from the Initial to Final sample times in either plot location (Fig. 2A).

After treatment of both boscalid rates, the proportion of DMI-R/Ben-R isolates increased in the 2014-7-DAT, 2015-Final, and 2016-7-DAT sample times. Boscalid provided excellent control of

dollar spot in 2015 and dollar spot lesions with active mycelia were not observed on either rate of boscalid treated plots at the 2015-7-DAT sample time. The proportion of DMI-R/Ben-R isolates selected by boscalid decreased after overwintering in 2014-2015 or 2015-2016. The proportion of DMI-R isolates increased after overwintering in 2014-2015 at Loc 1. The proportion of DMI-R/Dicar-R isolates decreased or was not detected by the both rates of boscalid treatments at the both locations (Fig. 2B and C).

On propiconazole (both rates) or iprodione treated plots, the proportion of DMI-R/Dicar-R isolates increased in all sample times. On the other hand, the proportion of DMI-R/Ben-R and DMI-R decreased or was absent by propiconazole (both rates) or iprodione treatments. After overwintering, the proportion of DMI-R/Dicar-R isolates decreased but the proportion of DMI-R isolates increased in every sample time except for 2016-Initial sample time on the propiconazole (0.5 kg a.i. ha⁻¹) treated plots at Loc 1 (Fig. 2D, E and F).

Summary points:

- Fluxapyroxad (low and high rates, succinate dehydrogenase inhibitor, SDHI), boscalid (high rate, SDHI), fluazinam (an uncoupler of phosphorylation), and Enclave[™] (a four-way mixture of chlorothalonil, tebuconazole, iprodione and thiophanate-methyl) provided better dollar spot control than the dicarboximides (iprodione and vinclozolin) or DMI (low rate of propiconazole).
- Propiconazole or iprodione application selected isolates with both DMI and dicarboximide resistance (DMI-R/Dicar-R), but controlled isolates with both DMI and benzimidazole resistance (DMI-R/Ben-R).
- Boscalid application selected DMI-R/Ben-R isolates but controlled DMI-R/Dicar-R isolates.
- Previously selected DMI-R/Dicar-R or DMI-R/Ben-R isolates decreased after overwintering, in the absence of selection pressure. In addition, the proportion of isolates with DMI, dicarboximide, and benzimidazole resistance (DMI-R/Dicar-R/Ben-R) decreased regardless of fungicide treatment.
- This is the first report of multiple fungicide resistant population dynamics in response to different fungicide classes and overwintering effects and will help develop effective strategies for managing multiple fungicide resistance and potentially delay the emergence of future resistant populations.



Fig. 1. Relative control percent (%) of dollar spot of fungicide treatments on two different fairway locations at Wethersfield Country Club, CT in 2015 (A) and 2016 (B). Relative control percentage (RC%) data were collected by counting number of individual infection centers and calculating area under (AUDPC) values for all rating dates among all treatments. Rating began on the first date of the first fungicide application and concluded 21 days after the final application. RC% was calculated with the following formula: [(untreated-fungicide treated)/untreated] \times 100 = RC%. Different letters on top of bar indicated significantly different (p<0.05) according to Fisher's protected least significant difference.



Fig. 2. Isolate phenotype proportions on untreated (A), boscalid (0.28 kg a.i. ha⁻¹) (B), boscalid (0.38 kg a.i. ha⁻¹) (C), propiconazole (0.5 kg a.i. ha⁻¹) (D), propiconazole (1.0 kg a.i. ha⁻¹ (E), and iprodione (3.05 kg a.i. ha⁻¹) (F) treated plots on Loc 1 (location 1) of fairway in 2014 and 2015, 2016-Initial and on Loc 2 (location 2) of fairway in 2016. Initial, 7-DAT, and Final refer to initial sampling before fungicide application, 7 days after treatment of fungicide, and 21 days after final treatment of fungicide, respectively. Isolate phenotypes: DMI insensitive (DMI-R), DMI and dicarboximide resistance (DMI-R/Dicar-R), DMI and benzimidazole resistance (DMI-R/Ben-R), and DMI, dicarboximide, and benzimidazole resistance (DMI-R/Ben-R).

Reference cited:

Hyunkyu Sang, James T. Popko Jr., and Geunhwa Jung. Evaluation of a *Sclerotinia homoeocarpa* population with multiple fungicide resistance phenotypes under differing selection pressures. (submitted to Plant Disease)