Fungicide Resistance in Turfgrass - Current Ideas and Research

By Paul Koch, *Turfgrass Diagnostic Lab, University of Wisconsin - Madison*

Author's note: This is the second in a three-part series looking at fungicide resistance in turfgrass.

In the May/June 2007 issue of *The Grass Roots*, I briefly reviewed the past history of fungicide resistance as it pertained to turfgrass, as well as the different selection pressures employed on fungal organisms by the benzimidazole, DMI, and strobilurin classes of fungicides (Koch, 2007). But in recent years, as researchers have continued to study fungicide resistance, conflicting results and ideas have failed to greatly further our understanding of fungicide resistance.

A golf course environment is a particularly confusing site to try to manage fungicide resistance. In traditional agriculture, great pains are taken to make the field as uniform an environment as possible. In turfgrass, and especially golf course maintenance, just the opposite is true. In accordance with how the game of golf is played the course is divided up into several distinct areas, each with a significantly different maintenance regime.

Putting greens are mowed daily during the growing season at heights around an eighth of an inch (3.2 mm). Daily mowing removes dew from the turfgrass leaf blade every morning, significantly reducing the duration of leaf wetness and reducing disease pressure (Williams *et* al., 1996). Due to the extreme value most golf courses put on their putting greens, fungicides are applied routinely to prevent any disease from blemishing their investment. These factors suggest a relatively low amount of disease pressure when compared to other areas of the golf course.

Fairways are mowed approximately three times per week at heights of approximately half an inch (13 mm). Mowing three times per week removes the dew off those leaf blades in the morning only on those days, significantly increasing the duration of leaf wetness and making the leaf surface more conducive to disease infection. The increased height of the fairway turf results in a denser canopy, trapping moisture and exudates from the turf at the ground level and increasing ground-level humidity (Giesler *et* al., 2000). Fungicides are expensive to purchase, and with the large acreages that golf course fairways can occupy, many clubs may opt to spray fairways less often with fungicides in an attempt to reduce costs. This suggests an increased level of disease pressure when compared to putting greens. Golf course tee boxes are often maintained in a similar fashion to golf course fairways.

The rough areas of a golf course are managed quite differently from either a putting green or a fairway. Golf courses in the temperate regions of the world usually have roughs that consist of Kentucky bluegrass (*Poa pratensis L.*), perennial ryegrass (*Lolium perenne L.*), and/or fine fescues (*Festuca* spp.). They are mowed two or three times per week at heights of approximately 2 inches (5 cm). Many golf courses do not irrigate their rough areas, and those which irrigate do not do so every day. Fungicide applications are seldom applied to roughs due to the extreme cost, even though dollar spot and other diseases can damage these areas. Disease pressure on roughs may be lower than a fairway or green due to its species composition and lack of irrigation, but significant disease infections can occur in optimum environmental conditions.

With such different environments it is likely that a significant variation in disease pressure would also be observed, and we do see this most noticeably with *Sclerotinia homoeocarpa*, the causal agent of dollar spot. With differences in disease pressure amongst sites within a single golf course, would it also be fair to assume that there is variation amongst the rate of fungicide resistance development at these same sites? This was the major objective of a study done by myself and Dr. Geunhwa Jung, former turfgrass pathologist at the University of Wisconsin - Madison who is now at the University of Massachusetts - Amherst.

Materials and Methods

The study was done during the summer of 2006. Six golf courses in Wisconsin and one golf course in Massachusetts were selected for *S. homoeocarpa* sampling to represent a range of course ages, fungicide history, and current fungicide application frequency (Table 1). Indicator areas or areas of high dollar spot disease pressure on one putting green, one fairway, and one rough area were chosen for isolate collection at each golf course. A 10 X 10 m grid was kept free of pesticide applications until our isolate collection was complete. Only courses A, B, and C did not apply fungicides to the putting green sampling grid throughout the season, allowing *S. homoeocarpa* isolates to be collected from all three course areas only from those golf courses. Isolates were collected from the fairway and rough sampling grids at the remaining four golf courses. Isolates were sampled throughout the summer of 2006 as symptoms became present at each site.

A total of 900 leaf blades showing lesions symptomatic of dollar spot were collected from courses A, B, and C with 100 collected from one putting green, one fairway, and one rough area at each course. A total of 800 symptomatic leaf blades were collected from courses D, E, F, and G with 100 samples collected from one fairway and one rough area at each course. Each sample collected was at least one meter apart. Samples were processed for pathogen isolation within 24 hr of collection.

These isolates were then subjected to an *in vitro* fungicide resistance assay, which in this particular case is a laboratory measurement of how well each isolate grows on fungicide-amended potato dextrose agar (PDA) media

Table 1. Golf course age, location, and fungicide history for each course used for *Sclerotinia homoeocarpa* isolate collection.

Golf Course	Location	Year Opened	Five year benzimidazole history[X]	Five year DMI history[X]
A	WI	2003	None	Moderate on greens Moderate on fairways
B	WI	1958	None	Intensive on greens Sporadic on fairways
C	WI	1967	Sporadic on greens None on fairways	Moderate on greens Sporadic on fairways
D	WI	1921	Moderate on greens Sporadic on fairways	Intensive on greens Intensive on fairways
E	WI	1991	None	Intensive on greens Moderate on fairways
F	WI	2001	Sporadic on greens None on fairways	Intensive on greens Moderate on fairways
G	MA	1916	None on greens Sporadic on fairways	Moderate on greens Moderate on fairways

[X]Intensive = >15 applications over the past 5 years, moderate = 5-15 applications over the past 5 years, and sporadic = < 5 applications over the past 5 years compared to un-amended PDA media. Resistance assays were done using two different active ingredients from two separate fungicide classes. Cleary's 3336 (thiophanate-methyl) was selected to represent the benzimidazole class of fungicides, while Banner MAXX (propiconazole) was selected to represent the demethlyation inhibitor (DMI) class. Because of the directional selection employed by propiconazole, growth of the fungus on the fungicide-amended media was measured with a ruler and taken as a percentage of the growth of the fungus on un-amended media (Figure 1). The disruptive selection of the thiophanate-methyl allowed a simpler rating system; if the fungus was growing at all on the media amended with thiophanate-methyl than it was rated with the number one, where if there was no growth than it was rated with a zero. Statistical analyses were performed on the data to determine significant relationships.

Results and Discussion

With over 1,400 isolates of *S. homoeocarpa* subjected to *in vitro* fungicide resistance assays, the data set was large and very complex. But there are three important observations that were made from this study that can be used as superintendents develop their fungicide program for 2008.

The first and most telling observation was that there were significant differences in the level of fungicide resistance of both DMI and benzimidazole fungi-

Figure 1. This is an example of the process used for rating the level of resistance to DMI fungicides. The plate in the middle is growing on media not amended with fungicide, while the outside four plates are growing on media amended with propiconazole. The lack of growth of the two on the right suggest it is sensitive to the fungicide, while the significant growth of the two on the left suggest a certain level of resistance to propiconazole.

cides based on if the isolates were collected from a putting green, fairway, or rough area. When dealing with resistance to propiconazole this roughly correlated with fungicide history, as those sites that tended to have more intensive fungicide application histories had higher levels of resistance. For example, the majority of the *S. homoeocarpa* population collected from the rough area of course "A" had very low levels of resistance to propiconazole. On the fairways of course "A", there was a shift in the *S. homoeocarpa* population to where the majority of the population now had higher levels of resistance to propiconazole, and on the greens the population shifted further towards increased resistance (Figure 2). This is not surprising in the sense that it agrees with the traditional model of fungicide resistance where the more fungicide applications made at a site equals a higher level of resistance. But with thiophanate-methyl the data seemed to suggest that higher proportions of the *S. homoeocarpa* population were resistant in the fairways regardless of the fungicide history, though the data was much more inconclusive on this point.

The second important observation from this study was that proportions of *S. homoeocarpa* populations resistant to both fungicides could generally be linked to their five year fungicide history. Those populations that received what I termed an intensive level of fungicide applications over the last five years generally had a higher proportion of resistant isolates than those populations with a moderate fungicide history, and always had a higher proportion of resistant isolates than those sites that received a sporadic level of fungicide applications. There was at least one major exception to this rule though. Isolates collected from the fairway at Course D received twice as many fungicide applications over the last five years as those collected from the fairway at course G, but the level of resistance in the population was significantly lower at course D than course G. After inspecting the fungicide records at these two courses, the only notable difference in the two spray schedules was the spray interval. Course D kept a fairly tight spray interval on their fairway of 14 days, while course G oftentimes let its interval go to 21 or even 28 days. This observation supports a traditional recommendation of fungicide resistance management that suggests that spray interval can affect the buildup of isolates with reduced sensitivities to fungicides.

The third important observation from this study was that to keep the proportion of the *S. homoeocarpa* population resistant to fungicides at or near baseline levels, an average of one fungicide application a year or less was required. The rough areas at five of the seven golf courses in this study were used as a baseline due to the very low and statistically similar resistance levels found at those courses. And the only two populations that had resistant proportions statistically similar to the baseline populations were the fairway populations from course B and C, where a total of five fungicide applications were made in the past five

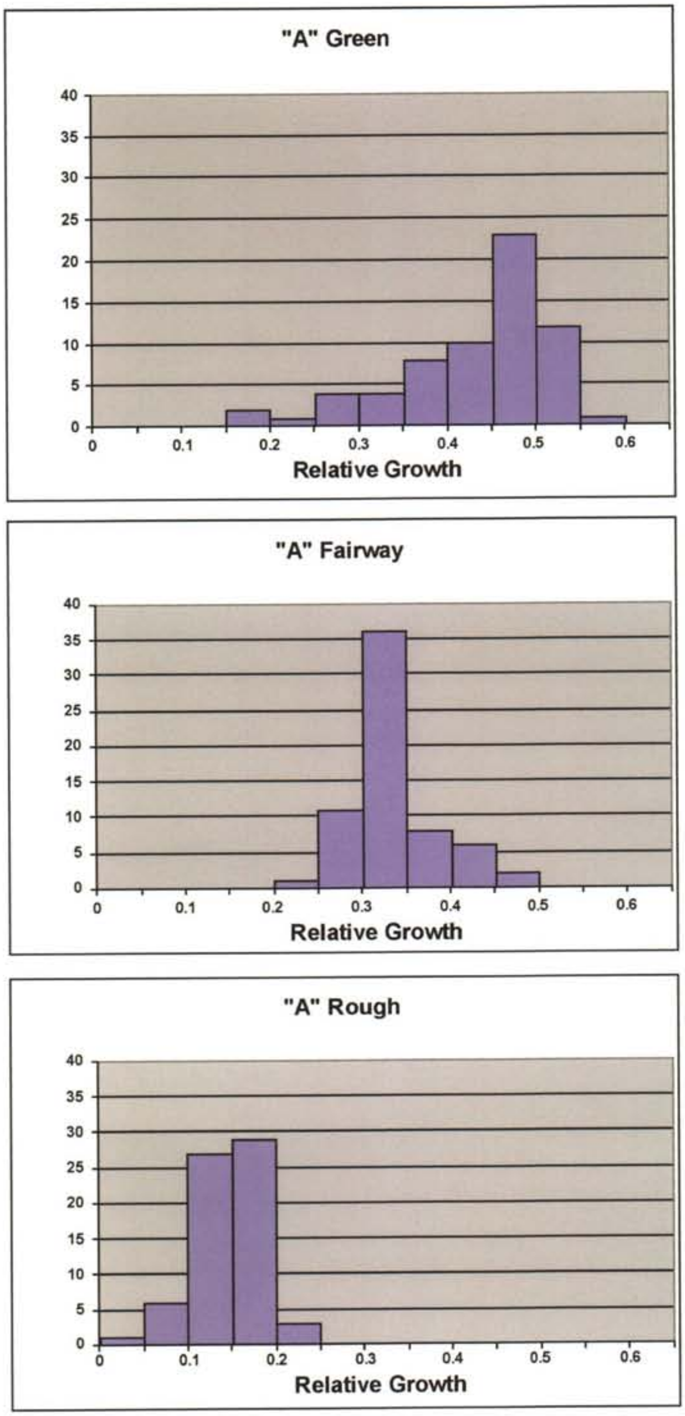

Figure 2. Histograms of *Sclerotinia homoeocarpa* populations obtained from a putting green, fairway, and rough area at course "A". Where applications of DMI fungicides were more frequent, the frequency of isolates with high mean relative mycelial growth values on propiconazole-amended media increased.

years. Course B and C are both several decades old and have been able to maintain this very low resistant proportion for years, while courses opened within the past decade already have levels of resistance far surpassing those at B and C. One fungicide application per year is clearly not a practical solution for managing fungicide resistance in most instances.

So how can we limit the development of fungicide resistance on our golf courses? While this study cannot give clear answers to that question, this study has shown that the issue of fungicide resistance may actually be even more complex than thought just recently and may have raised more questions than it answered. But it is clear that every golf course, no matter how old or how young, should have a plan in place to manage fungicide resistance. This plan should include implementing traditional strategies for managing fungicide resistance such as fungicide class rotation, inclusion of contact fungicides, adhering to the recommended spray interval, and applying at the correct label rate as recommended by the fungicide label. These traditional strategies do not appear to have the ability to completely stop the development of fungicide resistance, but should slow down the rate of resistance buildup significantly enough to maintain our most effective fungicides years into the future. New resistance management strategies without sufficient scientific data to support their validity should be looked at skeptically, though not completely discarded.

Fewer new fungicide chemistries are being produced by chemical companies due to the incredible cost, and many of our current fungicides are facing increasing regulation from federal and state governments. This leaves turfgrass managers with fewer options for controlling disease, and if we're not careful could lead to a reduction in efficacy due to resistance development in fungal populations. Much more research is needed to determine what strategies actually work to limit resistance development to keep our most successful disease control products effective years into the future.

References

Giesler, L. J., Yuen, G. Y., Horst, G. L. 2000. Canopy microenvironments and applied bacteria population dynamics in shaded tall fescue. *Crop Science*, 40(5), 1325-1332.

Koch, P. 2007. Fungicide Resistance in Turfgrass - An Introduction. *The Grass Roots*. May/June. 35(3). P. 32-35

Williams, D. W., Powell, A. J., Vincelli, P., Dougherty, C. T. (1996). Dollar spot on bentgrass influenced by displacement of leaf surface moisture, nitrogen, and clipping removal. *Crop Science*, 36(5), 1304-1309.