

Biological Control of Turfgrass Diseases

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Biological control is gaining popularity throughout the turfgrass industry. However, there have been very few if any successful biological control products introduced into the marketplace. This year, Wilbur-Ellis introduced BioTrek 22G, a trichoderma based biological control product. We are encouraging evaluation of this product by treating half greens with the biological control and comparing the treatments to normal management practices or comparison to the non-treated half green. As with any new technology, there will be some expected disappointments during the learning phase. BioTrek 22G will only control a limited range of diseases and it should not be expected to replace chemical control products. In this PACE Insights, you will find a critical review of the recent published results that supports the performance of the BioTrek 22G for control of pythium blight, brown patch and dollar spot. In addition, a background on biological control in general will be provided to aid in understanding how biological control of plant diseases works.

What is biological control? The definition of biological control has drifted over the years and a single definition is no longer accepted. Following years of discussion by the American Phytopathological Biological Control Committee, two distinct definitions were identified (Loper and Stowell, 1992):

1) Organism focused: Biological control is the reduction of the amount of inoculum or disease-producing activity of a pathogen accomplished by or through one or more organisms other than man (Cook and Baker, 1983).

2) Gene focused: Biological control is the use of natural or modified organisms, genes, or gene products to reduce the effects of undesirable organisms (pests) and to favor desirable organisms such as crops, trees, animals and beneficial insects and microorganisms (Cook, 1987)

In the organism focused approach, only the release a biological control organism is considered a biological control practice. For example, the release of ladybird beetles for control of aphids on roses would be considered biological control. Application of the fungus, *Trichoderma* (now available from Wilbur-Ellis as Bio-Trek 22G), for control of brown patch would also be considered biological control. However, spraying the *Bacillus thuringiensis*-produced (BT) endotoxin on plants to control lepidopteran insects does not necessarily fit the organism focused definition of biological control even though BT

applications have been the safest, most widely used, and most reliable "biological control" products.

The gene focused definition of biological control expands the range of biological control to include the BT endotoxin-based products and to include many other natural and genetically engineered organisms and their byproducts, for example antibiotics. The new fungicide, Heritage, is a modified byproduct isolated from a fungal fermentation. If a modified fungus byproduct is included in the definition in the future, Heritage would be considered a biological control product using the gene focused definition.

Mechanisms of biological control can be broken down into five major categories:

- Plant genetic resistance
- Plant induced resistance
- Competition
- Antibiosis
- Parasitism

Plant genetic resistance is the first line of defense against plant diseases. For example, *poa annua* is highly susceptible to a variety of diseases including summer patch caused by *Magnaporthe poae*. Bentgrass, however, is not susceptible to this pathogen and therefore use of bentgrass would prevent the heavy fungicide inputs that are needed to keep poa alive during the summer patch months between May and September. Unfortunately, implementing biological control using this mechanism would require a change in turfgrass varieties which is an expensive proposition. Moreover, the change from poa to bentgrass and maintenance of bentgrass can be difficult (see PACE Insights 2:2). Therefore, the use of genetic resistance is not always a realistic option except when greens are being constructed, rebuilt or resurfaced.

Plant induced resistance is an area of research that has become popular in recent years. Researchers have found that when a plant has been challenged by one microorganism, a resistance response is induced throughout the plant to other plant pathogens. The inducing organism does not need to be a pathogen of the plant being tested. This type of resistance was formerly termed "cross-resistance" because inoculation using one organism would result in plant resistance to a different pathogen. For example, Wei et. al found that cucumber seeds treated with a plant growth promoting rhizobacterium (PGR) resulted in induction of resistance to the anthracnose pathogen

Colletotrichum obiculare (Wei et. al., 1991). The mechanisms of induced resistance are not well understood but in some cases the plant will produce chitinase, an enzyme that dissolves chitin containing fungus cell walls. In other cases, lignification of the plant cell walls is stimulated which prevents fungal penetration into the plant.

Induced resistance has been taken to the field and has resulted in significant reduction in disease (Wei, G., Kloepper, J.W., and Tuzun, S. 1996). Although the results were statistically significant, the greatest increase in fruit yield was 19% over the non-treated diseased check. Neither a healthy check nor a chemical fungicide check were included in the trial to help the reader determine how effective the control is compared to commercial chemical control standards. The results are promising and induced resistance may play a role in turfgrass management sometime in the future.

Competition is a heavily studied mechanism for biological control. Competition may be for nutrients or for infection sites on the host leaf or root. One of the early organisms that was discovered to limit disease progress in the laboratory and also under some field conditions was *Pseudomonas fluorescens*, a naturally occurring soil bacterium. The *P. fluorescens* bacteria produce a compound that grabs or chelates iron from the environment and holds the iron so that the plant pathogens can not obtain the iron (Misaghi et.al. 1982, Stowell et.al. 1981, Kloepper, J.W., Leong, J., Teintze, M., and Schroth, M.N. 1980). Because iron is an essential component of all aerobic organisms respiratory electron transport systems, competition for this essential nutrient is intense in the soil. Without iron, fungal growth stops and plant infection can not occur. If excess iron is provided, however, the fungus will resume growth and infection will proceed. The problem encountered when exploiting competition between the biological control agent and the pathogen for nutrients or infection sites is the difficulty in delivering the biological control agent to all of the areas where the pathogen might attack the host plant. In addition, competition will be effective only under conditions where a nutrient is limiting to the extent that an antagonistic population of bacteria or fungi can capture the nutrients before a pathogen is able to grow and infect the host plant. The question arises; under normal turfgrass growing conditions are nutrients in levels that are so excessive that the biological control competitors will never be capable of capturing sufficient amounts of nutrients to result in effective competition and reduction in growth of the pathogen?

Competition for sites has mainly been applied to root infecting fungi. Competition for sites on the root requires the biological control organism to be what is termed "rhizosphere competent." This means the biological control fungus or bacterium must seek out

the root and multiply fast enough to keep up with the root as it grows through the soil in three dimensions. The infection sites may be captured by physically blocking the area to prevent infection by the pathogen, rapid consumption of any nutrients leaked around the infection site, or by production of chelators as mentioned above or antibiotics as mentioned below.

Antibiosis results from the production of antibiotics by the biological control organism that kills the pathogen. This mode of action differs from competition because antibiosis is not a reversible process - the pathogen has been killed. In addition to the iron chelators produced by *P. fluorescens* above, some strains of *P. fluorescens* also produce a phenazine antibiotic that inhibits the take-all pathogen *Gaeumannomyces graminis* (Thomashow and Weller, 1988). Under natural conditions, the competition between soil microorganisms for nutrients and space has resulted in evolution of many mechanisms of antibiosis and competition that might some day result in effective control of turfgrass diseases.

Parasitism is a sophisticated interaction between plant pathogens and the biological control agent that seeks out and consumes the pathogen. For example, *Trichoderma* is capable of attacking *Sclerotium rolfsii* sclerotia (resting structures that look like a dichondra seed) by producing enzymes that digest the sclerotia. (Benhamou and Chet, 1996). Unfortunately, plant pathogens do not always provide clues to the biological control agent that allow the agent to find the resting organism. Moreover, plant pathogens are frequently present in very low levels in the soil that make it difficult for the biological control agent to contact the pathogen. Although parasitism of growing mycelium is also possible, damage to the turfgrass has already occurred before the biological control agent can stop the pathogen. Cases where parasitism may be effective might be limited to organisms that produce large resting structures, for example *Sclerotium rolfsii* and *Rhizoctonia solani*.

Biological control of foliar and soil-borne diseases is fundamentally different. Foliar diseases, for example anthracnose caused by *Colletotrichum graminicola* or melting out caused by *Bipolaris sorokiniana* can cause rapidly spreading epidemics (Figure 1). Only one or a few spores of either fungus can initiate an infection that will result in production of millions of spores that can spread throughout the turfgrass stand. The stage of the disease cycle that is susceptible to biological control is primarily the short period, about 12 hours after the spore contacts the plant leaf. In this time the spore germinates and infects the leaf. If the biological control organism is not at the exact site where the spore lands on the leaf, the biological control will fail. Moreover, the foliar environment does not provide the moisture and nutrition needed by the biological control organism to

effectively colonize the infection site. For these reasons, foliar diseases are poor candidates for biological control by microorganisms and plant genetic or induced resistance are the biological control mechanisms that are most promising.

Soil-borne pathogens, for example brown patch caused by *Rhizoctonia solani* and dollar spot caused by *Sclerotinia homeocarpa*, are more easily targeted by a biological control agent such as *Trichoderma*. In these instances, the resting structures (sclerotia or hyphae in decayed plant tissue) are stationary in the soil and the biological control has a chance to colonize the soil and to destroy the pathogen before it becomes active (Figure 2). The biological control can also colonize the root in the moist, relatively nutrient rich root surface area. Combined, these factors make soil-borne diseases more likely candidates for biological control by microorganisms through competition and antibiosis. Plant genetic resistance and induced resistance are also possible mechanisms of biological control of soil borne diseases.

Figure 1. Disease cycle for spore forming foliar pathogen of turfgrass (anthracnose caused by *Colletotrichum graminicola*). 1) spore is delivered to the foliage. 2) conducive environmental conditions persist for about 12 hours and the spore germinates to infect the plant. 3) if the plant is susceptible the infection will proceed and the pathogen will reproduce itself. Foliar pathogens frequently produce many small spores during progress of an epidemic. Once the spore has been distributed to the infection site, germination and infection can take as little as 12 hours. Genetic resistance and induced resistance are probably the only biological control methods that will prevent the spread of the epidemic. It is unlikely that competition, antibiosis or parasitism will successfully stop the progress of this type of disease.

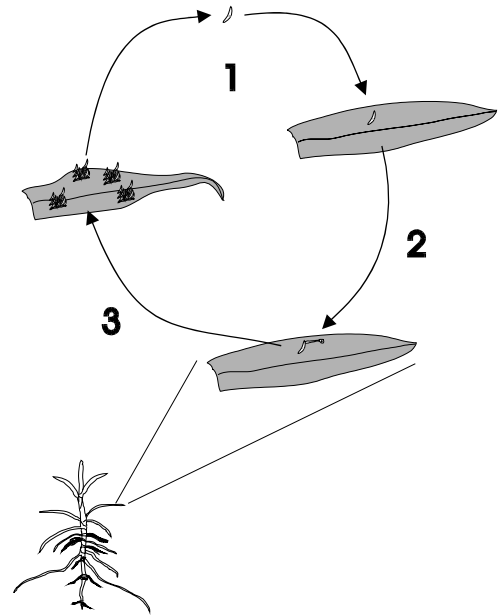
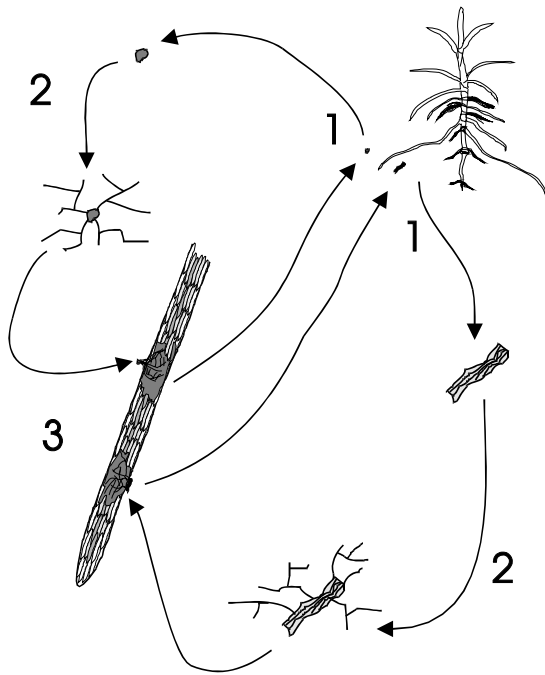


Figure 2. Disease cycle of a soil-borne pathogen (for example *Rhizoctonia solani* or *Pythium aphanidermatum*). 1) sclerotia or infected plant tissue survive periods of adverse environmental conditions in the soil. 2) the sclerotia, oospores in tissues, or mycelium in tissues germinate when stimulated by environment, nutrients, moisture or temperature. 3) The pathogens grow through soil in search of the plant and infects the plant if it is susceptible to disease. Soil-borne pathogens produce durable resting structures, for example sclerotia or oospores, that can survive through periods of adverse conditions. In some cases, thick-walled mycelium in colonized tissues act as the survival structure (e.g. *Magnaporthe poae*, summer patch). These resting structures can be present in the soil for months at a time without causing plant damage. During this time, the resting structures are waiting for a stimulus, nutrients, temperature, or moisture, before they become active. During this time, biological control organisms have a chance to locate and destroy the resting structures of the pathogen breaking the cycle of disease. Unfortunately, as few as 2 propagules per gram of soil can result in heavy disease while 1 to 10 million biocontrol spores may be needed to significantly reduce the disease (Lo, et. al. 1996). All four strategies of biological control, genetic resistance, induced resistance, competition, antibiosis, and parasitism may work to control soil borne diseases.



BioTrek 22G is a new biological control product introduced by Wilbur-Ellis in 1996. It represents the first EPA labeled biological control agent for turfgrass use. It was developed by Gary Harman and Eric Nelson both of Cornell University. A recent publication in Plant disease describes the scientific basis for promoting this organism as a biological control agent (Lo, et. al. 1996). The following is a critical review of the salient components of this publication.

Lo et. al. provided sufficient evidence to demonstrate that the *Trichoderma* strain tested could reduce the amount of disease compared to a non-treated check plot. The greatest level of disease reduction reported was 64% for dollar spot. The standard chemical treatment, propiconazole (Banner at 1 oz/1000 sq ft which should protect plants against dollar spot for 28 days), provided 100% control up to 20 days after treatment. When the biological control is compared to the chemical at 60 days after treatment, the biological control provided significantly better control (44%) compared to the propiconazole treatment (11%). The propiconazole and the non-treated check plots were not significantly different because propiconazole is only expected to last 28 days and ratings were taken at 60 days after treatment. These results combined with the other data provided in the report suggest that *Trichoderma* can provide some level of control of dollar spot but expectations should be set at about 50% control and then only for 30 - 60 days. Is *Trichoderma* a biological control agent? Yes. Is *Trichoderma* as effective as standard chemical treatments when they are used according to their label instructions? No. Is *Trichoderma* a product that

should be used at my golf course? This is the difficult question. If you and your golfers are satisfied with 50% control of dollar spot for a similar cost compared to a standard fungicide which will deliver 100% control, then maybe *Trichoderma* will fit into your disease control program.

Pace Turfgrass Research Institute's perspective: Biological disease management is a natural component of most turfgrass management practices whether we know it or not. It includes all processes that result in the reduction of damage caused by plant diseases that do not depend upon the use of synthetic agrochemical pesticides. This is a systems approach that attempts to redirect the immediate urge to reach for a jug, be it a biological control jug or a synthetic chemical jug. Balanced fertility that results in reduced plant stress and increased resistance to diseases is included under this umbrella of biological disease management. Aeration practices that modify the soil gaseous environment to stimulate growth of naturally occurring aerobic antagonists of plant pathogens is also a biological management practice. Of course, introducing a biological control agent, for example *Trichoderma* or BT are also included in this perspective of biological disease management. This broad perspective suggests that every process in a turfgrass management program will have an impact, be it positive or negative, on the turfgrass-plant-pathogen system. This is nothing new, just back to basics with a biological spin.

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