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Biological controls

Promising new tools for disease management

by Dr. Eric B. Nelson

DISEASE MANAGEMENT represents a significant challenge for turfgrass managers. The task is made particularly demanding by the perennial nature of turfgrass plantings, as well as that of the disease-causing organisms. Most, if not all, fungal pathogens of turfgrass are always present in turfgrass plantings.

As a result, the principal factors determining the incidence and severity of turfgrass diseases are environmental factors and plant stresses that influence not only the activity of pathogens, but the susceptibility of the plants. This is particularly true for some root pathogens that reside inside turfgrass plants year round. In many cases, these factors cannot be manipulated adequately to minimize losses from fungal diseases. So, to control fungal root diseases, turfgrass managers rely largely on fungicide applications.

Most of the materials currently used for turfgrass disease control are broad-spectrum systemic fungicides. Problems have arisen from the repeated and prolonged use of these chemicals:

- THE DEVELOPMENT of fungicide-resistant pathogen populations,
- DELETERIOUS EFFECTS on non-target organisms, particularly those involved in carbon and nitrogen cycling,
- ENHANCEMENT of non-target diseases,
- AND THE SELECTION OF FUNGICIDE-degrading microorganisms.

In an effort to reduce this fungicide dependency and to minimize the undesirable biological and environmental effects of excessive fungicide use, alternative management practices are being explored.

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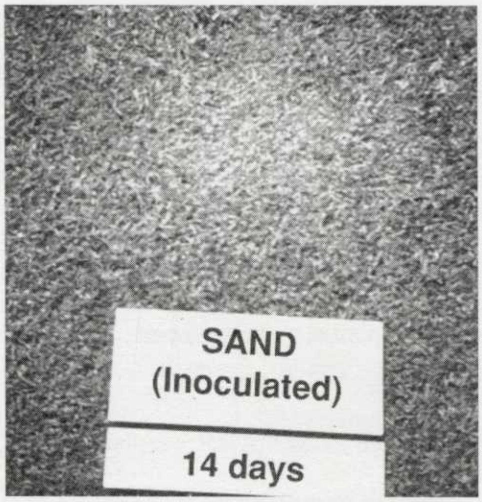

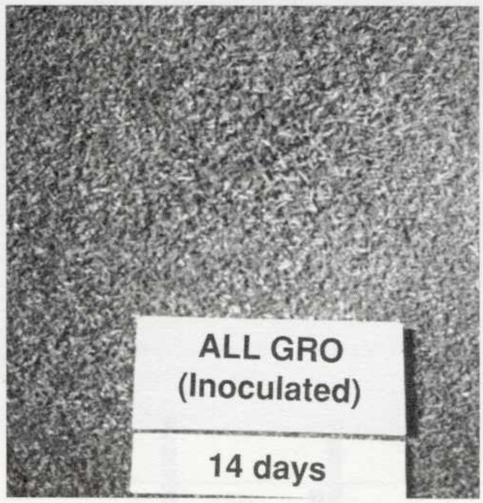

▲ Within two weeks the untreated part of a putting green innoculated with Pythium root rot fungi begins showing severe damage. Less damage is apparent in areas treated with All Gro, a commercial brewery waste compost (similar results obtained with Endicott sewage sludge compost).
Photo provided by Mary Thurn, Cornell University

" The principal factors determining the incidence and severity of turfgrass diseases are environmental factors and plant stresses that influence not only the activity of pathogens, but the susceptibility of the plants."

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One of the more exciting alternative management strategies being developed is the use of antagonistic microorganisms (also called 'antagonists') to reduce either the activities of pathogens or enhance the tolerance of plants to disease. This approach to disease control has been used successfully on an experimental as well as a commercial basis for the control of plant pathogens on several crop plant species and has recently seen applications in the turfgrass industry.

Biocontrol approaches

MOST TURFGRASS MANAGERS are familiar with the negative aspects of soil microorganisms, since some are pathogenic and can damage a turfgrass stand. However, in addition to pathogens, the soil harbors a variety of non-pathogenic microorganisms that actually improve plant health. These soil bacteria and fungi are responsible for

- INCREASING THE AVAILABILITY of plant nutrients,
- FORMING SYMBIOTIC ASSOCIATIONS with turfgrass roots,
- PRODUCING SUBSTANCES STIMULATORY to plant growth,
- AND PROTECTING PLANTS against infection from pathogenic fungi.

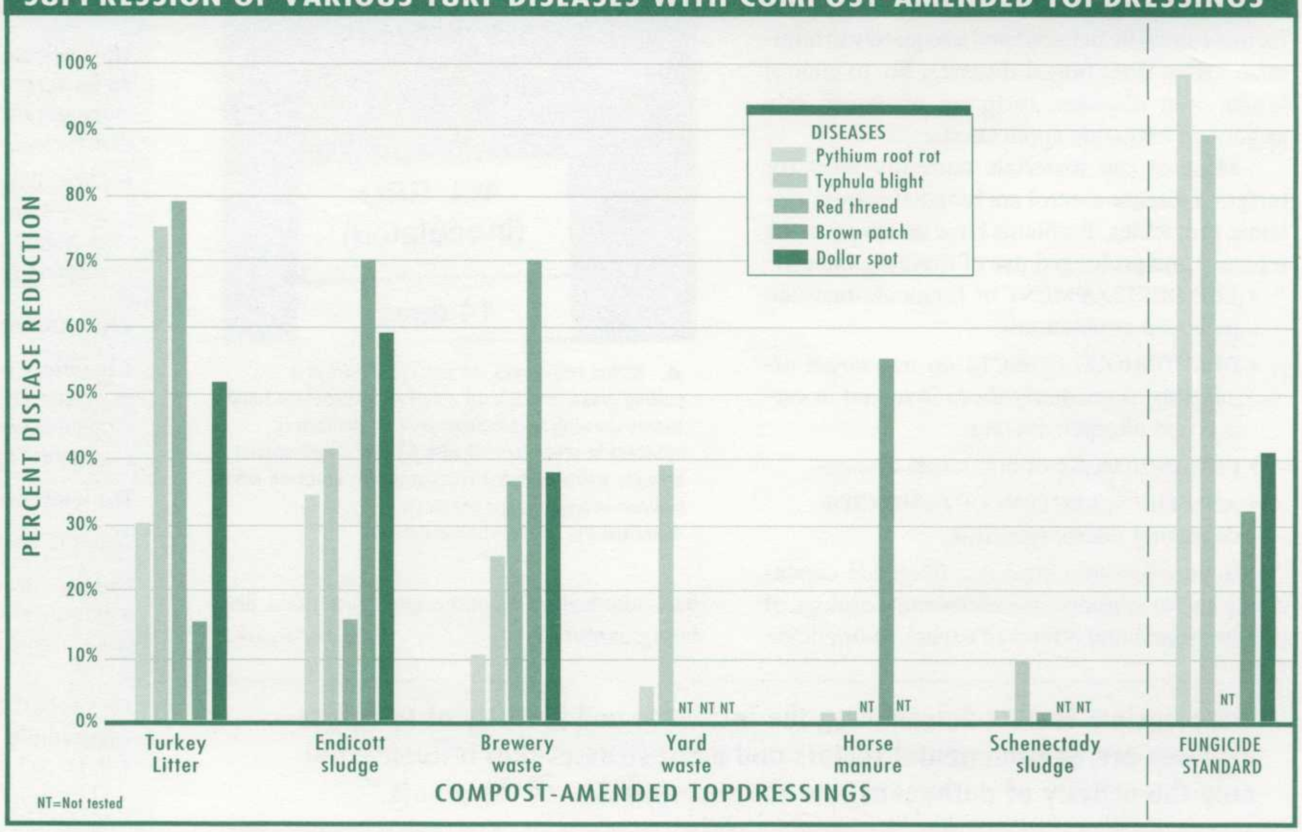
To minimize damage from plant pathogens, biological control attempts to take advantage of all the above-mentioned microbial attributes. For example, the application of composts, or other sources of organic matter, to turf may introduce large populations of antagonistic microorganisms that may reduce disease by interfering with the activities of pathogenic fungi. Similarly, cultural management techniques (such as core aeration, fertilization, or the application of pH-altering materials such as lime) may reduce disease development by altering the soil and thatch microbial communities within which pathogens must function. In such cases, cultural practices may indirectly affect disease severity by changing the environment to favor antagonistic microbial communities to the detriment of pathogen populations.

Biological control may be achieved either through the application of introduced microbes or through the manipulation of native microbes, present either on plant parts or in soils, that "naturally" suppress diseases. In either case, the goal is to reduce or eliminate pathogen activities by

- REDUCING PATHOGEN inoculum in soil,
- PROTECTING PLANT SURFACES from infection,
- OR TRIGGERING NATURAL DEFENSE mechanisms within the plants.

Biological control of pathogen inoculum is achieved by the microbial destruction of pathogen propagules and the prevention of inoculum formation—through the action of mycoparasites (fungi that are parasitic on other fungi). In addition, antibiotic-producing microbes may displace pathogens in decaying plant residues, such as thatch, and reduce their populations in soil. Some non-pathogenic soil

SUPPRESSION OF VARIOUS TURF DISEASES WITH COMPOST-AMENDED TOPDRESSINGS



KNOWN MICROBIAL ANTAGONISTS OF TURFGRASS PATHOGENS

DISEASE (PATHOGEN)	ANTAGONISTS	LOCATION*
■ BROWN PATCH	*Rhizoctonia spp.*	Ontario Canada
Rhizoctonia solani	*Laetisaria spp.*	North Carolina
	Compost microbes	New York, Maryland
■ DOLLAR SPOT	*Enterobacter cloacae*	New York
Sclerotinia homoeocarpa	*Fusarium heterosporum*	Ontario Canada
	Gliocladium virens	South Carolina
	Compost microbes	New York
■ NECROTIC RING SPOT	Native soil microbes	Michigan
Leptosphaeria korrae		
■ PYTHIUM BLIGHT	*Pseudomonas spp.*	Illinois, Ohio
Pythium aphanidermatum	*Trichoderma spp.*	Ohio
	Trichoderma hamatum	Colorado
	Enterobacter cloacae	New York
	Various bacteria	New York, Pennsylvania
	Compost microbes	Pennsylvania
■ PYTHIUM ROOT ROT	*Enterobacter cloacae*	New York
Pythium graminicola	Compost microbes	New York
■ RED THREAD	Compost microbes	New York
Laetisaria fuciformis		
■ SOUTHERN BLIGHT	*Trichoderma harzianum*	North Carolina
Sclerotium rolfsii		
■ SUMMER PATCH	various bacteria	New Jersey
Magnaporthe poae		
■ TAKE-ALL PATCH	*Pseudomonas spp.*	Colorado, France
Gaeumannomyces graminis	*Gaeumannomyces spp.*	Australia
var. avenae	*Phialophora radicicola*	Australia
	Microbial mixtures	Australia
■ TYPHULA BLIGHT	*Typhula phacorrhiza*	Ontario Canada
Typhula spp.	*Trichoderma spp.*	Massachusetts
Compost microbes	New York	

* The location indicates where experiments, demonstrating the effectiveness of the biocontrol agents on the indicated diseases, were conducted.

microorganisms are able to effectively colonize above-ground, as well as below-ground, plant parts and, in so doing, protect these tissues from infection by pathogens. It is also apparent that some biological control agents can induce natural defense mechanisms in plants. This phenomenon is called "cross protection" or "induced resistance."

The number and variety of potential antagonists is large and diverse. More commonly studied biological control agents include fungi in the genera: *Fusarium, Gliocladium, Laetisaria, Penicillium, Sporidesmium, Talaromyces, Trichoderma,* and *Verticillium* and bacteria in the genera: *Bacillus, Enterobacter, Erwinia,* and *Pseudomonas.*

Research has shown that these microorganisms can interfere with pathogen populations in a number of ways. Mycoparasites such as *Trichoderma* and *Sporidesmium* may parasitize pathogen propagules and mycelium. Other antagonists—particularly *Pseudomonas, Bacillus, Enterobacter, Erwinia* and *Gliocladium*—produce antibiotics that inhibit pathogen growth. Some strains of *Pseudomonas* and *Enterobacter* species are efficient competitors for essential nutrients and other growth factors, thereby reducing the amount of materials available for pathogen germination, growth, and plant infection.

Antagonists of turfgrass pathogens can be found in a variety of sites. They are particularly abundant in turfgrass soils and thatch, as well as in decaying organic substrates. Studies have shown that a greater percentage of antagonists of some pathogens are associated with thatch more commonly than with the underlying soil, both in low and in high maintenance sites. Also, these "thatch microbes" are generally more effective in suppressing diseases such as Pythium blight. In tests with various groups of soil bacteria, members of certain, less common groups showed significantly more biocontrol potential than other more abundant populations of bacteria.

To predictably and successfully manipulate biological control agents, turf managers must understand the biology and ecology of these micro-organisms in turfgrass ecosystems. (Unfortunately, we lack much of that knowledge.) The reason why this understanding is essential is simple: biocontrol agents differ fundamentally from chemical fungicides in that they must grow and proliferate to be effective. Therefore, effective antagonists must be able to become established in turfgrass plantings and remain suppressive to pathogens during periods favorable for plant infection.

The two factors most important in determining an-

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tagonist establishment and growth are:

- THE ENVIRONMENTAL CONDITIONS (particularly temperature, moisture, nutrients, and pH)
- AND THEIR ABILITY TO COMPETE with the existing soil and plant micro-organisms.

Just as some organisms are antagonists of pathogens, antagonists have their own antagonists as well.

Biocontrol agents also must be compatible with other management inputs. In particular, biological control agents must be tolerant of fungicides, insecticides, herbicides and fertilizers currently in use. Their activities must also not be discouraged by cultural practices used in turfgrass maintenance. Just as pathogens are influenced by environmental conditions, so too are biological control agents. Therefore, biological control strategies must be employed primarily to control pathogens, but at the same time, maintain the associated antagonistic microbial communities. Organisms isolated from many different environments might be suitable biocontrol agents, but composts are perhaps the best sources of complex mixtures of antagonistic microorganisms. Incorporation of organic matter into turfgrass soils is one of the better ways of maintaining antagonistic microbial communities.

Disease suppression with composts

A NUMBER OF DIFFERENT APPLICATION strategies have been tested with composts used for the purpose of disease control. These have included the use of composts as:

- TOPDRESSING AMENDMENTS,
- TURF COVERS,
- ROOT ZONE MIX AMENDMENTS,
- TEAS PREPARED BY EXTRACTING THE COMPOST with water for various periods of time. Another approach that has as yet to be tested is the use of composts as seed coating or pelleting material.

Perhaps the most exciting results have been obtained when composts have been used as a topdressing amendment. For example, monthly applications of topdressings composed of as little as 10 lbs of suppressive compost/ 1000 ft^2 have been shown to be effective in suppressing diseases such as dollar spot, brown patch, Pythium root rot, Typhula blight, pink snow mold, and red thread. Reductions in severity of Pythium blight, summer patch and necrotic ring spot have also been observed in sites receiving periodic compost applications.

Root zone amendments of various composts (20% compost: 80% sand; v:v) have produced excellent control of *Pythium graminicola*-incited root rot on creeping bentgrass putting greens. This technique involves incor-

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Process is the key to disease-suppressive composts

COMPOSTING CAN BE DEFINED as the "biological decomposition of organic constituents in wastes under controlled conditions". Since composting relies exclusively on microorganisms to decompose the organic matter, the process has biological as well as physical limitations. During composting, the environmental parameters (i.e. moisture, temperature, aeration) must be stringently controlled. This is necessary to maintain adequate rates of decomposition and to avoid the production of decomposition by-products that may be harmful to plant growth.

Compost "pile" design, construction, and maintenance play vital roles in the successful outcome of the process. For example:

- TO MAINTAIN PROPER TEMPERATURES, the composting mass must be large enough to be self-insulating, but not so large that compaction results in reduced air exchange.
- TO SUPPORT MICROBIAL ACTIVITY, the composting mass must be moist enough, but not excessively moist, so that the air exchange is limited.
- TO PROVIDE PROPER INSULATION, the particle size of the material must be small enough, but not too small—again to control air exchange.

When environmental and physical conditions are optimized, composting should proceed through three distinct phases (See the diagram of the composting process). The first stage of composting can last one or more days, depending on the type of starting materials used. During this phase, the temperature of the internal portions of the composting mass rise, as a result of the growth and activity of the indigenous mesophilic microorganisms associated with the starting organic material. During this self-heating phase, most of the soluble, readily degradable materials are broken down by these naturally-occurring microorganisms, precluding the

Biocontrols *continued from page 4*

porating the composts in the soil during construction of the greens.

Perhaps the most important benefit of compost use on established turfgrasses is its impact on root-rotting pathogens in the soil. Populations of soil-borne Pythium species are generally not suppressed following traditional chemical fungicide applications, but can be reduced on putting greens receiving continuous compost applications—in the absence of any chemical fungicide applications. Additionally, heavy applications of certain composts (~200 lbs/ 1000 ft^2) to putting greens in late fall can be effective, not only in suppressing winter diseases such as Typhula blight, but in protecting putting surfaces from winter ice and freezing damage.

One of the more practical uses of composts in turfgrass applications is in the preparation of compost teas. The preparation of these extracts consists of soaking compost in water at ratios of 1 part compost to 3–10 parts water. Extracts are allowed to incubate at ambient temperatures from 1 to 14 days, at which time the mixture is filtered—to remove large particulates prior to spraying with standard pesticide application equipment.

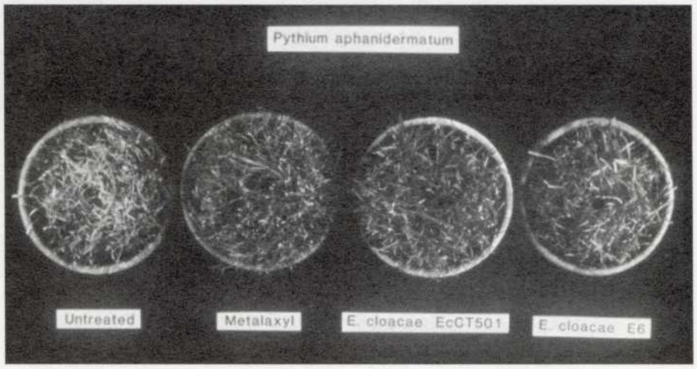
This method of disease control has proven extremely effective in laboratory experiments for control of Pythium species, but little is known about the activity of extracts under field conditions.

Microbiological variability in composts

COMPOSTS PREPARED from different starting materials, as well as those at different stages of decomposition, vary in the level of disease-suppression and in the spectrum of diseases that are controlled. This is primarily a result of the microbial variability among different composts and among the different qualities of organic matter present in any one compost at various stages of decomposition. Although microbial activity is necessary for the expression of disease-suppressive properties in most composts, the specific nature of disease suppressiveness is, in general, unknown.

The microbiology of disease-suppressive composts has not been extensively studied, but a limited number of studies have shed light on several important microbes in suppressive composts:

- FUNGAL AND BACTERIAL ANTAGONISTS suppressive to a number of plant pathogens have been recovered from hardwood bark and sewage sludge composts.
- RELATIONSHIPS BETWEEN microbial activity and Pythium suppression in bark composts have also been described. The levels of microbial activity have been used as a means of predicting Pythium suppression in composts.
- THE MOST IMPORTANT FUNGI in bark composts for the suppression of *R. solani,* the cause of brown patch were found to be *Trichoderma hamatum, T. harzianum* and *Gliocladium virens*.
- A NUMBER OF BACTERIAL SPECIES effective against *R. solani* and *Pythium spp.* have been discovered *(see photo below)*. Bacterial strains such as *Enterobacter cloacae, Flavobacterium balustinum, Xanthomonas maltophila* and various *Pseudomonas spp.* are more effective when combined with other fungal antagonists.



▲ This greenhouse test compared an untreated sample on the left, one using Metalaxyl (a chemical fungicide) and two strains of *E. cloacae* bacteria. The strain EcCT501 produced disease suppression comparable to the fungicide. In this test, all of the samples were inoculated with *Pythium aphanidermatum*.

- VARIOUS *PSEUDOMONAS* SPECIES from composts were found to be effective root colonists and antagonists of such root-rotting pathogens as *Pythium ultimum*.
- IN SOME SEWAGE SLUDGE COMPOSTS, strains of *Bacillus subtilis* have been shown to be effective in inducing suppression to a number of soilborne plant pathogens.

Although a wide variety of microbial antagonists can be found in composts, the predominant species, and their relative contributions to disease suppression, remain unknown. However, those microorganisms that are rapid and aggressive colonizers of organic matter or plant roots and crowns, are more likely to contribute the most to disease suppression in composts.

Predictable suppression is needed

TURFGRASS MANAGERS ARE ACCEPTING the use of composts as an attractive disease control alternative. In the few cases that have been examined, substantial reductions in fungicide use have accompanied the adoption of these strategies. Many composted materials and compost-based organic fertilizers are commercially available. Research has shown that the use of composts and organic fertilizers for turfgrass disease control is economically

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and technologically practical and, in some instances, can provide control equivalent to that currently attained with fungicides.

One of the principal problems with the use of composts for disease control is that a given compost may not be predictably suppressive from year to year, batch to batch, and from one site to the next. Turfgrass managers and compost producers agree that the future success of these materials depends upon the ability of producers to provide material with predictable levels of disease control. Gross variations cannot be tolerated.

Unfortunately, with our current level of understanding, it is not possible to predict the suppressive properties of certain composts without actually testing them in field situations. A number of tests have been developed to determine compost maturity and degree of stabilization for the purpose of reducing the variability in physical and chemical properties. However, none have been designed to directly assess microbiological aspects of maturity and disease suppressiveness.

In order to develop more effective biological control strategies with compost-based materials, several aspects of the turfgrass ecology of key compost-inhabiting antagonists will need to be understood. For example, the ability of antagonists to establish and survive in turfgrass ecosystems is necessary for biological control to occur. The interactions of antagonists with other soil organisms and the soil and plant factors affecting optimum biological control activity will be important in developing strategies with compost-based materials. In addition, these organisms may serve as indicators of how long to compost a material before it can be certified as disease-suppressive. Research aimed at understanding the fate of antagonistic organisms in soils and on plants following compost applications will aid in understanding why composts fail at certain times and in certain locations, but not at others. Such research should also help to predict the compatibility of composts and their resident antagonists with pesticides and other cultural practices now in use.

To make composts more predictably disease-suppressive, it may be possible to introduce antagonistic organisms, with known control properties, into composts at key stages in the curing process. This strategy has been used successfully to produce composts more predictably disease-suppressive and more highly suppressive to a number of plant pathogens. This approach should enable compost producers to ultimately produce predictably suppressive biological control materials.

Over the past five years, a large number of composts have become available for turfgrass applications. Some are properly composted and formulated and of high quality. Others are not. In the past, quality control was of less concern—when composts were used primarily as fertilizers. However, for disease management, quality control is important. When improperly composted, some organic materials can be extremely phytotoxic. Other improperly composted materials can even accentuate the development of some diseases.

Unlike traditional synthetic chemical fungicides, more careful consideration must be made of various aspects of the storage and application of microbial inoculants.

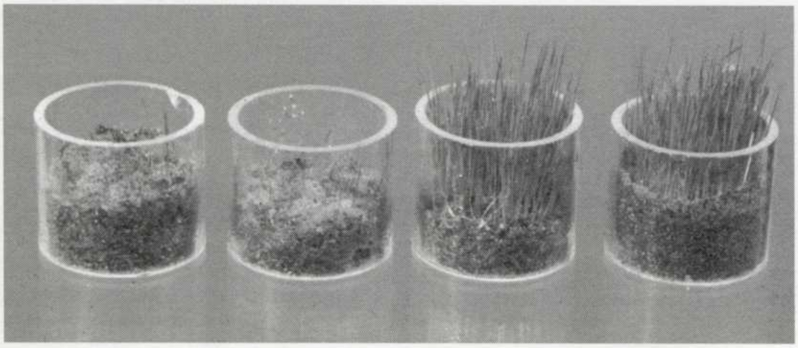
Use of microbial inoculants

MICROBIAL INOCULANTS ARE PREPARATIONS of living microorganisms that inhibit plant pathogens. In their development and use, beneficial microorganisms are isolated from the environment (usually from soils or plant tissues), and their populations are artificially increased. In some instances, they may be culturally or genetically improved in the laboratory. Then they are introduced back into the environment as an inoculant.

Unlike traditional synthetic chemical fungicides, more careful consideration must be made of various aspects of the storage and application of microbial inoculants. Of particular importance is the shelf life of microbial inoculants, since the organisms may not remain viable for extended periods of time. One also needs to consider that, for any microbial-based inoculant to be effective, the organism(s) must become established in turfgrass plantings and must remain active throughout the period when disease pressure is greatest. Additionally, the organisms in these products must be compatible with other agrichemicals in use. For example, whereas bacterial preparations should generally be tolerant of most chemical fungicides, fungal preparations may or may not be as suitable as bacterial preparations—depending on the characteristics of the species of fungus used.

The search for candidate strains of bacterial and fungal antagonists has been promising based on laboratory, greenhouse, and field tests, with many being effective against a wide range of turfgrass pathogens (see "Known Microbial Antagonists of Turfgrass Pathogens" table on page 3). Many of these antagonists, when applied at the proper time and manner, can establish high population levels in bentgrass putting greens and can be as effective as some of the newest chemical fungicides. The antagonist, *Enterobacter cloacae* is able to establish high populations in creeping bentgrass/annual bluegrass turf; levels between 100 million and 1 billion cells per gram of thatch. Although populations decline steadily through the season, nearly 1 million per gram remain after 13 weeks, and the following season, only about 1000 cells per gram can be recovered.

Through the past couple of decades, it has become apparent that the use of microbial inoculants is not without problems. This is primarily due to the lack of knowledge about how to adequately produce, formulate and handle living organisms. However, through continued evaluation in agronomic and horticultural systems, it has become evident that microbial inoculants may have an important place in commercial plant production and realistically offer important disease-control alternatives in plant health management. They can provide levels of disease control that, in many cases, facilitate reduced applications of fungicides and, in a few cases, may eliminate the need for fungicides altogether. In addition, microbial inoculants are a potentially important tool in managing fungicide resistance among pathogen populations. Furthermore, the success of sustainable plant production is largely dependent upon the integration of biological and other non- chemical means of control into disease management strategies. Recent developments in Integrated Pest Management (IPM) are a direct result of growing awareness of the importance of biological controls in holistic approaches to plant health management.



▲ This test of the suppressive effect of a particular group of microbial antagonists called Actinomycetes on *Pythium graminicola* showed dramatic results. The absence of the Actinomycetes resulted in total seeding failure, on the left. Their presence lead to successful establishment on the right. Photo by Christine Stockwell, Cornell University

Although the biological control of turfgrass diseases is still in the early development stages, long-term, the future of microbial inoculants for turf disease control is extremely bright. It is encouraging that a number of chemical pesticide companies are now funding biological control research and have made commitments to the development of microbial inoculants.

The future use of antagonists as microbial inoculants will come only from a better understanding of how antagonists function and how they interact with other turfgrass management inputs. Recent developments in molecular biology have tremendously increased our ability to answer some of these questions. These advances have been one of the principal reasons that biological control of fungal plant pathogens has become a more viable option for turfgrass disease management than it was just a few years ago.

The future of turfgrass biocontrols

BIOLOGICAL CONTROL OF TURFGRASS DISEASES is still in the developmental stage. Although there are a number of biological control products available for disease control on other commodities, none are currently available specifically for turfgrass disease control. Despite the past lack of emphasis on biological control research, the last five to 10 years have seen tremendous advances in our efforts to understand and develop biological control strategies for turfgrass diseases.

As the need to reduce fungicide dependency, and to provide more rigorous environmental stewardship become more critical, the greater the need will be to develop safe, effective and more environmentally sound control strategies.

The potential of composts to suppress turfgrass diseases is clear. At present, applications of these materials provide excellent alternatives to the use of fungicides on turf and may, in the long term, provide the only means of reducing soil populations of pathogens in turfgrass plantings. As we learn more about composting and the benefits of composted materials to plant health, there will be a greater demand from turfgrass managers for high quality disease-suppressive composts. Composted products for use on turfgrass are becoming increasingly available. In general, compost producers are committed to providing the highest quality materials at an equivalent cost of disease control far below that of traditional fungicides. In addition to providing effective disease control, the use of composts will help ease the burden on our nation's landfills and foster a commitment from turfgrass managers to sound environmental stewardship.

Because microbial inoculants used for disease control are relatively new to the marketplace, it is not yet clear, particularly in the United States, whether they will compete well with chemical fungicides and be acceptable to federal and state regulatory agencies. Although it is encouraging that more biological control products are becoming available, time will tell whether the beneficial properties of such materials can augment or replace traditional fungicides. It is critical that some of the initial biological control products consistently perform comparable to conventional fungicides if they are to find their way into the marketplace and gain widespread acceptance. As our search for more effective antagonists of turfgrass pathogens expands, suitable bacterial and fungal antagonists will provide a pool from which organisms can be developed into microbial inoculants. Biological control is on the verge of a new era of discovery and commercialization. The benefits of biological controls, once realized, may change the way in which disease control is approached. ■