Biological Control of Dollar Spot: A New Approach

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Concern related to the use of fungicides and the development of fungicide resistance has promoted the exploration for alternative means to manage fungal turfgrass pathogens. One area to receive much attention is the use of biological control, which is the use of beneficial organisms (bacteria, fungus, nematode, or insect) to manage population levels of a pathogen (through antibiosis, competition, or parasitism) or to reduce the incidence of plant diseases.

A bacterium isolated from a turfgrass sample was selected as a potential biological control agent based on its' ability to inhibit the growth of many fungal turfgrass pathogens in laboratory studies. Initial attempts to use this bacterium as a biological control agent in the field failed to yield significant disease reduction. In these studies the bacteria were applied on a 14 day schedule just as a fungicide would be applied. However, following application, the population of these bacteria would decrease rapidly over the following days. The most difficult obstacle to implementing a biological control program is to maintain high population levels of the biological control agent. Several ecological factors are responsible for the reduction in the viability of biological control agents following application. Just as ultra violet (UV) radiation from the sun is damaging to our skin, it is also damaging to organisms in the environment. Bacteria applied to the turf foliage experience high mortality from UV radiation causing genetic mutations and disrupting protein structure. Furthermore, in order for the bacterium to become established it must be able to colonize a particular niche. To do so it must be able to out compete the natural microbial populations present which are highly adapted to that particular environment..

To overcome the problem of maintaining the population levels of the biological control agent in the field, the frequency of application of the bacteria was increased to three times a week. The bacteria were cultured overnight, quantified, and applied at rates of $2*10^5$ and $2*10^7$ bacteria per cm^2 to field plots using a CO_2 hand sprayer. Control treatments included in this study consisted of an untreated control and a chemical control of chlorothalonil (Daconil 2787 WDG 3 oz. / 1000 ft^2 every 10 days). To account for possible fertility effects due to bacterial applications, an additional control treatment was included which consisted of an autoclaved culture (heat killed at 121 C and 15 lbs. pressure for 20 min) of bacteria. Ratings of dollar spot incidence were taken on a weekly basis throughout the season. Results of the field study are provided in Table 1 and Table 2. Application of the bacteria at the high rate provided disease reduction of 50% throughout the season, which was significantly better then the untreated control and autoclaved culture. No fertility effects by bacterial applications were evident as dollar spot incidence in the autoclaved culture treatment was the same as that in the untreated control plots.

The major question arising from this research is whether it is practical to make applications three times a week. This problem has recently been solved by the advent of the BioJect system (EcoSoil Systems, Inc., San Diego, CA) which couples a self contained bacterial fermentation system to an established irrigation system to which the bacteria are directly injected. It consists of a 155 gallon fermentation vessel within which the bacteria are cultured, automated temperature control and fermentation initiation, and self contained media and inoculum injection systems. Using this system it is possible to make daily bacterial applications. This system is well suited to night irrigation as it will apply the bacteria when foliar fungal pathogens are most active.

This study demonstrates that the bacterial strain is efficacious as a biological control agent for the management of dollar spot when applied frequently at $2*10^7$ CFU / cm^2. Such frequent applications are feasible with advent of the BioJect system. One handicap of this study is that the bacteria were often applied during mid-day during which time the bacteria are most susceptible to UV light. Bacterial applications made through an

irrigation system should be made at night when the dollar spot fungus is active and there is no UV light. The use of this bacterial biological control along with higher fertility rates (1 lb N/month) may well provide dollar spot management at levels provided by chemical fungicides.

Future research into biological control of dollar spot and other turfgrass diseases will focus on exploitation of the BioJect system. Future bacterial applications will be made in the evenings to simulate nightly irrigation. Duration of bacterial culture will also be examined. As bacteria are grown in a batch culture they go through different metabolic states depending on the availability of nutrients. By applying bacteria which are in these different metabolic states we can determine at which fermentation time the bacteria are most efficacious.

Table 1. Dollar spot ratings for September 6 and September 13.

		September 6, 1995		September 13, 1995	
Treatment	Rate[a]	Average # of Spots	Normalized[b] Data Analysis	Average # of Spots	Normalized Data Analysis
No Treatment	---	49.3	1.70 A	30.0	1.48 A
Bacteria Low Rate[c]	$2*10^5$ / cm^2	37.0	1.56 AB	22.3	1.34 AB
Bacteria High Rate	$2*10^7$ / cm^2	27.8	1.42 B	12.5	1.01 B
Autoclaved Culture[d]	Volume as Above	40.8	1.62 A	33.5	1.54 A
Chlorothalonil	0.15 g AI / m^2	6.3	0.85 C	1.5	0.35 C

[a] Application rates made on plots 0.9 m x 1.2 m.
[b] Data transformation performed as log (# of spots + 1) then analyzed with Tukeys Honestly Significant Test.
[c] Bacteria cultured on Trypticase Soy Broth 24 hours prior to application.
[d] Bacteria culture autoclaved after24 hours of growth and applied at equal volume to $2*10^7$ / cm^2 rate.

Table 2. Dollar spot ratings for September 30 and October 4.

		September 30, 1995		October 4, 1995	
Treatment	Rate[a]	Average # of Spots	Normalized[b] Data Analysis	Average # of Spots	Normalized Data Analysis
No Treatment	---	24.3	1.40 A	32.5	1.52 A
Bacteria Low Rate[c]	$2*10^5$ / cm^2	20.0	1.27 AB	21.8	1.31 AB
Bacteria High Rate	$2*10^7$ / cm^2	11.0	1.04 B	13.5	1.09 B
Autoclaved Culture[d]	Volume as Above	24.5	1.41 A	34.8	1.55 A
Chlorothalonil	0.15 g AI / m^2	0.3	0.08 C	0.8	0.19 C

[a] Application rates made on plots 0.9 m x 1.2 m.
[b] Data transformation performed as log (# of spots + 1) then analyzed with Tukeys Honestly Significant Test.
[c] Bacteria cultured on Trypticase Soy Broth 24 hours prior to application.
[d] Bacteria culture autoclaved after24 hours of growth and applied at equal volume to $2*10^7$ / cm^2 rate.