

BIOLOGICAL CONTROL OF DOLLAR SPOT: II
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Dollar spot, caused by *Sclerotinia homoeocarpa*, is one of the most common diseases of high maintenance turfgrasses. Although chemical fungicides can effectively manage this disease, both the development of resistance to certain classes of fungicides and public concerns over fungicide use have prompted research for alternative management strategies. Considerable progress has been made in recent years toward the development of effective biological control agents to managed dollar spot.

Research at Michigan State University toward this goal has focused on the application of the bacterium *Pseudomonas aureofaciens* strain Tx-1, a soilborne bacterium which produces antifungal compounds. Last year's field study showed that application of this bacterium three times a week could reduce dollar spot incidence by up to 60%. The high frequency of application is required to maintain high populations of this antagonistic bacterium on the turf foliage. If the bacteria are not applied frequently, the populations decline due to ultra-violet radiation, competition, and removal through mowing. The practicality of this approach has been made a reality through the development of on-site fermentor systems which allow the golf course superintendent to grow the bacteria overnight and make applications daily through the irrigation system.

This year's field study further examined the effectiveness of this bacterium to manage dollar spot. The frequency of application of the bacterium was increased from three to five times per week to better simulate daily applications. Unlike last year's study, the bacteria were applied late in the afternoon to reduce mortality due to ultra-violet light. A set of treatments were added in which the bacteria were grown for a period of two days and applied once and twice a week. These treatments were included to simulate application of the bacteria under conditions in which application through an irrigation system is not possible, and the bacteria must be downloaded into a spray unit to be applied manually.

In addition to testing the frequent application of *Pseudomonas aureofaciens* Tx-1, several other biological control treatments were tested, including two commercial biological products. A final treatment, glucose, was applied as a carbon source to stimulate the natural flora's ability to suppress populations of pathogenic fungi.

Materials and Methods:

The dollar spot control study was conducted at the Hancock Turfgrass Research Center on Penncross bentgrass maintained in greens condition. The study was conducted as randomized block design with 5 replications. Plots were 2 by 3 feet with 1 foot alley ways intervening. The study was initiated in the beginning of July and continued through mid-October. Variation in collected data was normalized as the log(number of spots + 1). Data was analyzed by Tukeys' Honestly Significant range test using the MSTATC statistical software.

Pseudomonas aureofaciens Tx-1 bacteria were prepared for daily application by culturing overnight in sterile media at 26°C while provided constant aeration. The bacteria were quantified and applied with a hand sprayer, using nitrogen gas, at rates of 2×10^5 and 2×10^7 colony forming units per square cm. Alternatively, bacteria were cultured for a period of 48 hours prior to quantification and application. A control treatment of boiled 24 hour bacterial culture was applied daily to assess for possible fertility effects. Commercial products were applied per manufacturers recommendations. Spray application were made with a hand sprayer. Actinovate was watered in with 1 gallon of water per plot.

Results and Discussion

Dollar spot disease pressure was most severe in the month of September. Data for the rating dates of September 11 and September 25 are provided on Table 1.

Further insight into biological control was also obtained in this study through the application of sugar glucose. It was believed that this treatment would stimulate bacterial populations, as is thought to happen with organic fertilizers, which could act in an antagonistic manner toward dollar spot. However, this treatment consistently exhibited the most disease. *Sclerotinia homoeocarpa* is known to be most aggressive when guttational water, which contains sugars and amino acids, is present. This study indicates that the application of glucose acted to increase the virulence of *Sclerotinia homoeocarpa* more so than it did to stimulate populations of antagonistic organisms.

The only treatments to provide significantly better dollar spot management than the untreated control were the daily application of *Pseudomonas aureofaciens* Tx-1 at the 2×10^7 rate and the application of the boiled Tx-1 broth. Both of these treatments consisted of equal amounts of the bacterial broth grown over a 24 hour period, and reduced dollar spot incidence by up to 80 %. The increased reduction in disease seen in this year's study when compared to last year's study may be attributed to the increased frequency of application of the bacteria at the high rate. The efficacy of these treatments is best explained by the production of the antibiotic phenazine-1 carboxylic acid (PCA) by the bacteria prior to application.

Treatment of the bacterial culture by boiling will kill the bacteria due to denaturing proteins, however, this heat treatment doesn't not destroy PCA. This suggests that it is most likely the PCA in the daily application of the bacteria and in the boiled culture that was responsible for reducing dollar spot incidence. Furthermore, application of autoclaved (120 C with 15 lbs. pressure) broth in the 1995 field study failed to reduce dollar spot pressure. Under the extreme condition of autoclaving, PCA will degrade, which explains why the autoclaved treatment of 1995 failed to provide disease reduction while the boiled treatment in 1996 was able to reduce dollar spot incidence. This study further demonstrates the efficacy of *Pseudomonas aureofaciens* Tx-1 and PCA as tools for the management of dollar spot.

Table 1. Biological control of dollar spot.

Treatment	Rate per 1000 ft ²	App. Interval	September 11, 1996		September 25, 1996		Tukey's Test ^b	Tukey's Test
			Avg. # of Spots	Normalized Data ^a	Avg. # of Spots	Normalized Data		
Control	—	—	48.2	1.62	15	1.16	A	ABC
Tx-1	2*10 ⁵ CFU/cm ²	daily	41.4	1.52	14.2	1.11	A	ABC
Tx-1	2*10 ⁷ CFU/cm ²	daily	8	0.84	3	0.46	BC	DE
Boiled Broth	As Above	daily	6.2	0.50	0.4	0.10	C	E
Daconil Ultrex	4 oz.	14 days	26.2	1.35	4.8	0.70	AB	CD
Tx-1	48 hr. growth	2x/week	34.8	1.36	7.6	0.74	AB	BCD
Tx-1	48 hr. growth	weekly	35.4	1.49	5.6	0.80	A	BCD
Companion	4 fl. oz.	14 days	49.2	1.69	14.6	1.03	A	ABCD
GB03	4 fl. oz.	14 days	55	1.70	22	1.34	A	AB
GB03 alt. Ultrex	4 fl. oz. alt. 4 oz.	14 days	50	1.66	34.8	1.52	A	A
Actinovate	3#/acre/11 wks.	14 days	50.6	1.65	15.8	1.15	A	ABC
Glucose	4 oz.	daily	65.6	1.8	55.6	1.63	A	A

^a Data normalized as log(n+1)

^b Data analyzed by Tukey's Test: Treatments followed by the same letter are not significantly different at p = 0.05.