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Application of *Pseudomonas aureofaciens* Tx-1 through Irrigation for Control of Dollar Spot and Brown Patch on Fairway-height Turf

Glenn A. Hardebeck,[1] Ronald F. Turco,[2] Richard Latin,[3] and Zachary J. Reicher[4]

Purdue University, 915 West State Street, West Lafayette, IN 47907-2054

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***Abstract*. *Pseudomonas aureofaciens* strain Tx-1 is suggested as a biological control for *Sclerotinia homoeocarpa* (F.T. Bennett) and brown patch (*Rhizoctonia solani* Kuhn) on golf courses. To overcome application difficulties, a field bioreactor is used to grow Tx-1 daily and then inject into nightly irrigation on the golf course. Though Tx-1 shows some promise for disease control in vitro, it is relatively untested under field conditions. We conducted three field experiments to 1) evaluate the efficacy Tx-1 when applied through an irrigation system for the control of dollar spot and brown patch; 2) determine if there is an interaction between nitrogen fertility or fungicides on efficacy of Tx-1; and 3) determine if Tx-1 can extend the duration of dollar spot control by a single application of fungicide. Nightly applications of Tx-1 through irrigation did not affect brown patch on 'Astoria' colonial bentgrass (*Agrostis capillaris* Sibth.) during the 2 years of our study. Tx-1 reduced dollar spot in 'Crenshaw' creeping bentgrass (*Agrostis palustris* Huds.) by 37% in 1998 compared to non-Tx-1 treatments, but Tx-1 had no effect on dollar spot in 1999. Under low disease pressure, Tx-1 increased the dollar spot control of fungicides by 32% and increased the duration of control by 2.6 days. However, Tx-1 had no effect on fungicide efficacy or duration of control later in the summer when dollar spot pressure was high. Fungicides did not negatively affect Tx-1's control of brown patch or dollar spot, nor did fertilizer regime affect brown patch or dollar spot control by Tx-1. Although delivery of Tx-1 in our studies was optimized, disease control was marginal and occurred only under low disease pressure. Therefore, we conclude Tx-1 has limited practical value for turfgrass disease control on golf courses.**

Dollar spot (caused by *Sclerotinia homoeocarpa* F.T. Bennett) and brown patch (caused by *Rhizoctonia solani* Kuhn) are destructive diseases of both cool- and warm-season turfgrasses (Burpee and Martin, 1992; Walsh et al., 1999). These diseases are controlled culturally and with fungicides (Fidanza, and Dernoeden, 1996a; Fidanza and Dernoeden, 1996b; Settle et al., 2001; Walsh et al., 1999). However, over-reliance on fungicides may lead to evolution of insensitive strains (Golembiewski et al., 1995). Biological disease control may help minimize reliance on fungicides by turfgrass managers.

Biological disease control is attempted by introducing disease suppressive antagonists or by manipulating the activity of antagonistic organisms already present in soils or on plant parts (Smiley et al., 1992). Disease suppressive organisms have been identified and isolated in a variety of crops including turfgrass (Cruz et al., 1992; Hodges et al., 1994; Lo and Nelson, 1996; Nelson and Craft, 1991; Qing and Shiping, 2000; Thompson et al,. 1996). *Trichoderma harzianum* is one biological control that has been identified and used for control of disease in turfgrass (Harmon, 2000). Although many disease-suppressive microorganisms have been isolated, their ability to suppress disease relates to colonization and persistence in sufficient numbers (Bull et al., 1991; Parke, 1990; Paulitz and Baker, 1987). Efficacy of an introduced organism suffers unless it can occupy a specific niche, proliferate, and produce an antagonistic response (Nelson et al., 1994). Colonization and persistence problems may be overcome by repeated applications of the antagonistic organism. This is the basis of the BioJect system (Eco Soil Systems Inc., San Diego, Calif.), which is a field bioreactor that grows a population of bacteria daily and then injects the culture into nightly irrigation. This delivery system reduces the labor demand of repeated sprays, supplies freshly grown bacteria, and allows nighttime application of the bacteria, which reduces viability losses due to desiccation and UV radiation. However, delivery and efficacy of this system is relatively untested.

Pseudomonas aureofaciens strain Tx-1 (Tx-1) is a commercially available biofungicide marketed under the trade name Spot-less Bio-Fungicide (Eco Soil Systems Inc.) and is used in conjunction with the BioJect system. Tx-1 is known to produce the phenazine antibiotic phenazine-1-carboxylic acid (PCA), which is primarily responsible for disease suppressive activity (Pierson and Thomashow, 1992; Powell and Vargas, 2000). PCA inhibits growth of several turfgrass pathogens *in vitro*, including *R. solani* and *S. homoeocarpa* and suppressed dollar spot development in greenhouse and field trials (Powell and Vargas, 2000).

Three separate experiments were initiated to 1) evaluate the efficacy of Tx-1 when applied through an irrigation system for the control of dollar spot and brown patch; 2) determine if there is an interaction between nitrogen fertility or fungicides on efficacy of Tx-1; and 3) determine if Tx-1 can extend dollar spot control by a single application of fungicide.

Materials and Methods

Tx-1, fertilization regime, and fungicides for control of brown patch or dollar spot. Field studies were conducted at the Wm. H. Daniel Turfgrass Research and Diagnostic Center at Purdue University, West Lafayette Ind., during 1998 and 1999. Soil type was Starks-Fincastle silt loam (fine-silty, mixed, mesic Aeric Ochraqualf) with pH of 7.0, 183 $kg \cdot ha^{-1}$ P, and 598 $kg \cdot ha^{-1}$ K. The area was established in Aug. 1997. The experimental area was mowed at 1.3 cm three times per week with clippings returned. A fresh culture of Tx-1 was grown in a field bioreactor (Eco Soil Systems, San Diego, Calif.) daily and injected into the irrigation system each evening beginning at 2100 HR during a two-minute watering cycle delivering 0.13 cm of water. The small-plot irrigation system was plumbed separately into 11 m × 11 m zones to eliminate cross contamination of Tx-1 into non-injected water. To further reduce contamination, plots not receiving Tx-1 were covered with vinyl tarps during irrigation of the plots receiving Tx-1-injected water. Tarps were immediately removed and plots not receiving Tx-1 were irrigated with fresh water. Additional morning irrigation was applied to all plots as needed to prevent drought stress. In 1998, Tx-1 was applied from 14 May through 20 Sept. with reactor production averaging 5.8×10^8 colony forming units (CFU)/mL and injected plot irrigation averaging 1.6×10^7 CFU/mL. In 1999, Tx-1 was applied from 15 May through 19 Sept. with reactor production averaging 6.9×10^8 CFU/mL and injected plot irrigation averaging 2.6×10^7 CFU/mL. Tx-1 concentrations were determined by sampling the field reactor and irrigation weekly. Samples were taken directly from the bioreactor immediately before the irrigation cycle whereas irrigation water was sampled from each main plot with an autoclaved catch pan on the turf surface during the entire irrigation cycle. Samples were stored at 2 ± 1 °C for 10 h before enumeration. Tx-1 concentrations were determined through plate counts with three replicate plates per sample using 1/10th trypticase soy agar as the plating medium supplemented with rifampicin and cycloheximide each at 50 µg ml-1. Dilution blanks consisted of 25 % Ringer solution (Weaver et al., 1994).

Brown patch. Colonial bentgrass (*Agrostis tenuis* Sibth.) 'Astoria' was used for this study. Treatments were arranged in a 2 × 2 × 4 factorial with two Tx-1 treatments (nightly and none),

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[1]Professional assistant, Dept. of Agronomy.

[2]Professor, Dept. of Agronomy.

[3]Professor, Dept. of Botany.

[4]Associate professor, Dept. of Agronomy. To whom correspondence should be sent; e-mail zreicher@purdue.edu.

two fertilizer regimes (high N and low N), and four fungicide programs (none, preventative, curative and *Trichoderma harzianum*). The low N regime included 24 kg·ha^{-1} N applied 14 May and 11 June and 49 kg·ha^{-1} N applied 3 Sept. and 12 Nov. each year. The high N regime included 49 kg N/ha applied 14 May, 11 June, 9 July, 3 Sept. and 12 Nov. in both years. Nitrogen was applied manually with shaker bottles as sulfur coated urea except in November when urea was applied. The preventative fungicide treatment began 25 June during both years and consisted of chlorothalonil (tetrachloroisophthalonitrile) at 9.1 kg·ha^{-1} alternated with propiconazole (1-{2-2,4-dichlorophenyl-4-propyl-1,3-dioxolan-2-yl}-1H-1,2,4-triazole) at 1.0 kg·ha^{-1} applied every two weeks. A post-symptom fungicide treatment (curative) consisted of chlorothalonil at 9.1 kg·ha^{-1} was applied when the disease severity rating reached 1.5 on the Horsfall-Barratt scale, which was equivalent to three percent infection (Campbell and Madden, 1990). Curative applications were made on 1 July, 15 July, 30 July, and 20 Aug. during 1998 and 20 July during 1999. All fungicide applications were made with a CO_2 backpack sprayer in 1630 L·ha^{-1} water. A commercial preparation of *Trichoderma harzianum* (Bio-Trek 22G, Wilbur-Ellis, Fresno, Calif.) was applied manually with shaker bottles as two May applications per year with 73 kg·ha^{-1} product per label recommendations.

Experimental design was a split plot with Tx-1 treatments as main plots while subplots were the combination of fertility rates and fungicide strategies. There were three replications of main plots and subplot size was 2 × 2 m. The site was inoculated with a local isolate of *Rhizoctonia solani* on 19 June 1998 according to Burpee and Goulty (1984). Brown patch disease severity was recorded weekly using the Horsfall-Barratt system (Campbell and Madden, 1990). The disease severity rating was converted to a percentage, and area under disease progress curve (AUDPC) was calculated and standardized. AUDPC data were transformed to $\log_{10}(Y)$ as suggested by Box-Cox evaluation (Box et al., 1978). Statistical analyses were performed using procedures by SAS (Statistical Analysis Systems Institute Inc., Cary, N.C.).

Dollar spot. Creeping bentgrass (*Agrostis palustris* Huds.) cv. Crenshaw was selected for the evaluation due to the cultivar's susceptibility to dollar spot (National Turfgrass Evaluation Program, 1999). Experimental design, general treatments, and Tx-1 application were similar to that described for brown patch, except for the fertility regime and fungicide programs. The low N regime included 24 kg·ha^{-1} N applied 14 May and 49 kg·ha^{-1} N applied 3 Sept. and 12 Nov. both years. The high N regime included 24 kg·ha^{-1} N applied 14 May, 11 June, and 9 July and 73 kg·ha^{-1} N applied 3 Sept. and 12 Nov. both years. The preventative fungicide treatment began 14 May during both years and consisted of chlorothalonil at 9.1 kg·ha^{-1} alternated with propiconazole at 1.0 kg·ha^{-1} applied every 2 or 3 weeks, respectively. A postsymptom fungicide treatment (curative) consisted of chlorothalonil at 9.1 kg·ha^{-1}. The curative treatment was applied when the average number of spots per plot was >15. The curative applications were made on 8 July, 30 July, 12 Aug. and 2 Sept. during 1998 and 20 June, 31 June and 17 Aug. during 1999.

The site was inoculated with a sorghum culture of a local isolate of *Sclerotinia homeocarpa* on 29 June 1998 as described for the brown patch experiments. Disease severity during 1998 was recorded weekly as the number of dollar spots per plot. Due to rapid increase of disease in 1999, dollar spot severity was recorded twice weekly as a visual percentage of affected turf. Disease severity data were used to calculate AUDPC and then standardized by dividing by the length of the epidemic in days (Campbell and Madden 1990). AUDPC data were square root transformed as suggested by Box-Cox evaluation (Box et al., 1978). Statistical analyses were performed using procedures by SAS.

Table 1. Effects of Tx-1, nitrogen, and fungicide application on brown patch in 'Astoria' colonial bentgrass.

	AUDPC[z]					
Fungicide	1998			1999		
program	+Tx-1	–Tx-1	Mean	+Tx-1	–Tx-1	Mean
Preventive[y]	2.7	2.2	2.4 a	1.9	1.9	1.9 a
Curative[x]	3.6	2.3	2.9 a	2.6	3.5	3.0 a
T. harzianum[y]	9.0	7.7	8.3 b	6.5	7.1	6.8 b
None	6.5	8.5	7.4 b	5.6	7.9	6.7 b
mean	4.9	4.3		3.7	4.4	
N treatment (kg·ha^{-1}·yr^{-1})						
146	5.5	3.5	4.4	4.2	5.2	4.7
244	4.3	6.2	4.7	3.2	3.7	3.4
Mean	4.9	4.3		3.7	4.4	
ANOVA						
Tx-1		NS			NS	
Nitrogen (N)		NS			NS	
Fungicide (F)		**			***	
N × F		NS			NS	
Tx-1 × N		NS			NS	
Tx-1 × F		NS			NS	
Tx-1 × N × F		NS			NS	

[z]AUDPC values were standardized by dividing by the duration of the disease epidemic in days and then transformed by $\log_{10}$(AUDPC). Backtransformed values are presented.

[y]The preventative fungicide treatment began 25 June during both years and consisted of chlorothalonil at 9.1 kg·ha^{-1} and propiconazole at 1.0 kg·ha^{-1} applied alternately on a 2-week schedule.

[x]The curative treatment consisted of chlorothalonil at 9.1 kg·ha^{-1} and was scheduled when the disease severity rating reached 1.5 on the Horsfall-Barratt scale.

[w]*Trichoderma harzianum* (Bio-Trek 22G, Wilbur-Ellis, Fresno, Calif.) at 73 kg·ha^{-1} product applied twice in May each year.

[NS,*,**,***]Nonsignificant or significant at $P = 0.05$, 0.01, or 0.001 not significant, respectively.

Table 2. Effects of Tx-1, nitrogen and fungicide application on dollar spot in 'Crenshaw' creeping bentgrass.

	AUDPC[z]					
Fungicide	1998			1999		
program	+Tx-1	–Tx-1	Mean	+Tx-1	–Tx-1	Mean
Preventive[y]	7.3 ab	3.2 a	5.1 a	0.0	0.0	0.0 a
Curative[x]	10.2 ab	16.2 b	13.0 a	15.4	19.6	17.5 b
T. harzianum[w]	56.0 c	92.7 d	73.3 b	61.9	60.5	61.2 d
None	53.6 c	109.2 d	78.9 b	52.6	54.0	53.3 c
mean	26.8	42.0		22.7	24.2	
N treatment (kg·ha^{-1}·yr^{-1})						
122	43.0	50.8	47.3	27.9	27.2	27.6
220	13.8	33.9	22.8	18.1	21.3	19.6
Mean	26.8	42.0		22.7	24.2	
ANOVA						
Tx-1		*			NS	
Nitrogen (N)		***			***	
Fungicide (F)		***			***	
N × F		NS			**	
Tx-1 × N		NS			NS	
Tx-1 × F		*			NS	
Tx-1 × N × F		NS			NS	

[z]AUDPC values were standardized by dividing by the duration of the disease epidemic in days and then transformed by sqrt(AUDPC). Backtransformed values are presented.

[y]The preventative fungicide treatment began 25 June during both years and consisted of chlorothalonil at 9.1 kg·ha^{-1} and propiconazole at 0.99 kg·ha^{-1} applied alternately on a 2 or 3 week schedule.

[x]The curative treatment consisted of chlorothalonil at 9.1 kg·ha^{-1} and was applied when the average number of spots per plot was greater than 15.

[w]*Trichoderma harzianum* (Bio-Trek 22G, Wilbur-Ellis, Fresno, Calif.) at 73 kg·ha^{-1} product applied twice in May each year.

[NS,*,**,***]Nonsignificant or significant at $P = 0.05$, 0.01, or 0.001 not significant, respectively.

Efficacy of Tx-1 for extending dollar spot control by various fungicides. The experimental site was adjacent to the previously described studies and was established with 'Penneagle' creeping bentgrass in August 1997. The area was maintained at 1.3 cm with clippings returned and fertilized with 24 kg·ha^{-1} N in May, 49 kg·ha^{-1} N in Sept., and 73 kg·ha^{-1} N in Nov. Treatments were in a 2 × 8 factorial with two levels of *P. aureofaciens* application (none and nightly) and eight fungicide treatments. Fungicide treatments were applied once with a CO_2 backpack sprayer in 1630 L·ha^{-1} H_2O. Treatments included an untreated check, propiconazole at 0.3 kg·ha^{-1}, iprodione (3-[3,5-dichloro-phenyl]-N-[1-methylethyl)-2,4-dioxo-1-imidazolidine-carboamide] at 2.3 kg·ha^{-1}, thiophanate-methyl (dimethyl[[1,2-phenylene]bis[iminocarbonothioyl]]biscarbamate) at 3.1 kg·ha^{-1}, mancozeb (Mn + Zn + ethylylene bisthiocarbamate) at 14.6 kg·ha^{-1}, chlorothalonil at 6.8 kg·ha^{-1}, trifloxystrobin ([E,E]-alpha-[methoxyimino]-2[[[[1-[3-trifluoromethyl]phenyl]ethylidene]aminooxy]methyl]-methyl ester) at 0.3 kg·ha^{-1} and a tank mix of propiconazole at 0.3 kg·ha^{-1} plus 0.3 kg·ha^{-1} trifloxystrobin .

Experimental design was a split block with Tx-1 treatment as main plots and fungicide treatments as subplots (1.5 m × 1.5 m). Main plots were replicated three times and the experiment was conducted four times during the summer of 1999 beginning on 2 June, 23 June, 14 July, and 4 Aug. All four trials were conducted on separate but adjacent areas, and dollar spot was minimized with chlorothalonil at 9.1 kg·ha^{-1} applied seven days before the start of each trial. Culture, application, and sampling of Tx-1 as well as plot size and irrigation zones were consistent with that described previously. The application of Tx-1 began on 2 June across all trail areas and continued throughout the study. Bioreactor production averaged 7.5 × 10^8 CFU·mL^{-1} and injected plot irrigation averaged 2.2 × 10^7 CFU/mL.

Dollar spot disease severity was recorded twice weekly as the number of dollar spots per plot up to 300. AUDPC data were calculated using eight ratings in all trials. Days to 50 infection centers/per plot thresholds were determined by interpolating disease severity data for each plot, and this threshold would be considered commercially unacceptable on fairway height turf. AUDPC data were transformed to $\log_{10}(Y + 1)$ for further analysis with one being added to the raw Y value to prevent the occurrence of $\log_{10}(0)$ (Box et al,. 1978). Analysis of variance was performed on the transformed AUDPC data with PROC ANOVA, and threshold data was analyzed using PROC GLM.

Results and Discussion

Tx-1, fertilization regime, and fungicides forcontrol of brown patch. Brown patch symptoms were present in 1998 from 13 July to 27 July and again from 17 Aug. through 31 Aug. During 1999, brown patch symptoms were present 20 July through 26 July and recurred on 30 Aug. Tx-1 did not significantly affect AUDPC in either year (Table 1). This was similar to our preliminary studies where Tx-1 at concentrations of 2 × 10^4, 2 × 10^6 and 2 × 10^8 CFU·mL^{-1} did not suppress brown patch development in a growth chamber (Hardebeck , 2001). Although Tx-1 produces PCA which inhibits *R. solani in vitro* (Powell and Vargas, 2000), our studies indicate Tx-1 has little effect on brown patch development in the field. Previous work in our laboratory had shown occurrence of PCA in the irrigation water (Sigler et al., 1999), thus the Tx-1 was active in our studies. Davis and Dernoeden (2001) also

Table 3. Effect of Tx-1 and fungicides on dollar spot in 'Penneagle' creeping bentgrass.

	AUDPC[z]											
	Early June			Late June			July			August		
Fungicide (F)	+Tx-1	–Tx-1	Mean	+Tx-1	–Tx-1	Mean	+Tx-1	–Tx-1	Mean	+Tx-1	–Tx-1	Mean
Untreated control	1036	1818	1372 d[y]	3009	3244	3124 c	5687	5703	5695 d	3446	3520	3483 d
Propiconazole	83	228	138 b	415	364	388 ab	532	712	615 b	1065	1361	1204 c
Iprodione	120	210	159 b	505	351	421 ab	1951	1618	1777 c	985	1084	1033 c
Thiophanate-methyl	10	19	14 a	558	108	246 a	197	241	218 a	295	373	332 a
Mancozeb	1054	1292	1167 cd	549	2603	1196 bc	5654	5093	5366 d	3190	3407	3297 d
Chlorothalonil	626	904	752 cd	1404	2663	1934 c	3611	4036	3817 d	2450	2499	2474 d
Trifloxystrobin	586	650	617 c	2879	2248	2544 c	4970	4693	4829 d	2273	3023	2622 d
Trifloxystrobin + propiconazole	83	106	94 b	344	335	339 a	996	2014	1416 c	594	660	626 b
Mean	208	330		848	835		1814	2047		1353	1548	
ANOVA												
Tx-1		*			NS			NS			ns	
Fungicide (F)		***			***			***			***	
Tx-1 × F		NS			NS			NS			NS	

[z]Means are backtransformed from $\log_{10}$(AUDPC + 1). Backtransformed values are presented.
[y]Means within each column followed by the same letter are not significantly different at $P = 0.05$.
[NS,*,**,***]Nonsignificant or significant at $P = 0.05$, 0.01, or 0.001 not significant, respectively.

Table 4. Effect of Tx-1 and fungicides on days to reach 50 dollar spot infection centers/plot in 'Penneagle' creeping bentgrass.

	Days to reach 50 spots/plot											
	Early June			Late June			July			August		
Fungicide (F)	+Tx-1	–Tx-1	Mean	+Tx-1	–Tx-1	Mean	+Tx-1	–Tx-1	Mean	+Tx-1	–Tx-1	Mean
Untreated control	16.3	13.7	15.0 a[z]	6.3	5.7	6.0 a	4.3	3.0	3.7 a	11.3	10.0	10.7 a
Propiconazole	NA[y]	NA	NA	23.3	23.3	23.3 c	20.7	18.7	19.7 c	19.7	18.7	19.2 c
Iprodione	30.3	25.3	27.8 c	22.3	23.7	23.0 c	14.7	16.0	15.3 c	20.3	20.0	20.2 cd
Thiophanate	NA	NA	NA	19.3	24.4	21.9 c	30.7	27.7	29.2 d	23.7	24.0	23.8 e
Mancozeb	17.0	18.3	17.7 ab	6.9	9.0	8.0 ab	5.0	5.7	5.3 ab	12.7	10.7	11.7 a
Chlorothalonil	21.0	19.7	20.3 b	14.7	9.6	12.2 b	9.3	7.7	8.5 b	15.3	14.3	14.8 b
Trifloxystrobin	19.3	19.7	19.5 b	10.3	11.0	10.7 b	6.0	6.7	6.3 ab	16.0	14.3	15.2 b
Trifloxystrobin + propiconazole	33.5	25.0	29.3 c	24.0	23.7	23.8 c	18.0	14.3	16.2 c	21.3	21.3	21.3 d
Mean	22.9	20.3		15.9	16.3		13.6	12.5		17.5	16.7	
ANOVA												
Tx-1		*			NS			NS			NS	
Fungicide (F)		***			***			***			***	
Tx-1 × F		NS			NS			NS			NS	

[z]Means within each column followed by the same letter are not significantly different at $P = 0.05$.
[y]NA = not applicable; 50 infection centers/plot threshold was not reached during study.
[NS,*,**,***]Nonsignificant or significant at $P = 0.05$, 0.01, or 0.001 not significant, respectively.

reported sporadic results with Tx-1 on brown patch in their field studies where Tx-1 reduced brown patch on two rating dates in the first year of their study under low disease pressure, only to increase brown patch the following year under high disease pressure.

Nitrogen main effects were not significant on AUDPC in either year of this study, even though higher N regimes reportedly increase brown patch severity (Smiley et al., 1992). Preventative and curative fungicide treatments reduced AUDPC in both years compared to both *Trichoderma harzianum* and no fungicides.

Tx-1, fertilization regime, and fungicides forcontrol of dollar spot. Dollar spot symptoms appeared on 22 June 1998 and developed steadily through 14 Sept. In 1999, symptoms appeared 20 July, progressed rapidly and remained through 6 Sept. The main effect of Tx-1 was significant in 1998 when AUDPC was reduced 37% in Tx-1-treated plots compared to Tx-1-non-treated plots (Table 2). However, Tx-1 had no effect on AUDPC in 1999. As expected, the preventative and curative fungicide treatments provided lowest AUDPC during both years, reducing AUDPC up to 94% compared to the no fungicide treatment. *Trichoderma harzianum* application resulted in 13% higher AUDPC than the no fungicide treatment in 1999. Tx-1 × fungicide interaction occurred only in 1998 where AUDPC was reduced up to 51% on plots receiving Tx-1 plus either *Trichoderma harzianum* or no fungicide compared to the same treatments without Tx-1.

As expected, higher N regime reduced AUDPC during both years of our study (Smiley et al., 1992; Williams et al,. 1996). However, there was no interaction between Tx-1 and nitrogen levels. This agrees with Davis and Dernoeden (2001) who found nutrient supplements did not increase dollar spot control with Tx-1. Han et al. (2000) reported that Tx-1 reduced dollar spot under 195 $kg \cdot ha^{-1} \cdot yr^{-1}$ N on only one of eleven rating periods, further suggesting that nitrogen regime has little effect on efficacy of Tx-1.

This was a severe test for Tx-1 because of high susceptibility of 'Crenshaw' creeping bentgrass to dollar spot, plus the experimental area was inoculated with the dollar spot pathogen immediately before the 1998 study. Though biological controls may have potential, our data suggests Tx-1 appears to be more effective under low dollar spot pressure as was also suggested by Davis and Dernoeden (2001).

Efficacy of Tx-1 for extending dollar spot control by various fungicides. AUDPC values varied among the fungicide treatments with propiconazole, trifloxystrobin /propiconazole, iprodione and thiophanate-methyl providing highest control throughout all four trials (Table 3). Tx-1 reduced AUDPC only during the early June experiment when disease pressure was lower than in the later trials. This again supports the idea that Tx-1 is effective and can improve control from fungicides only when disease pressure is low. Although concerns have been raised about fungicide decreasing efficacy of biocontrol agents (Burpee, 1990; Smiley et al., 1992), we found no negative interactions in dollar spot control between Tx-1 and any of the fungicides.

Acknowledging that Tx-1 only marginally controls dollar spot when pressure is low, it may still be beneficial in extending the duration of control from fungicides. When averaged over all fungicide treatments, Tx-1 increased the time to reach the 50 infection centers per plot threshold by 2.6 d in the early June experiment, but had no effect at later dates (Table 4). Other research results were mixed where Tx-1 extended dollar spot control by iprodione and propiconazole by 7 to 10 d during the first year of a Maryland study, but had no effect the second year (Davis and Dernoeden, 2001).

Our studies indicate Tx-1 applied via the BioJect through an irrigation system is capable of marginally reducing dollar spot severity and extending fungicidal control to a minimal extent only when disease pressure is low. Our field research was done under ideal Tx-1 delivery conditions. We irrigated less than 0.3 ha with a new, well-functioning irrigation system with irrigation heads a maximum of 70 m from the Tx-1 source. However, golf courses will irrigate 12 ha or more with irrigation systems of variable age and application uniformity, and with irrigation heads potentially 2 km or more from the Tx-1 source. This potentially complicates Tx-1 delivery, dilutes Tx-1 concentrations delivered to the turf, and likely decreases already marginal efficacy. Therefore, we conclude Tx-1 has limited practical value for disease control on golf course turf.

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