Curative Evaluation of Biological Control Agents and Synthetic Fungicides for *Clarireedia jacksonii*

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Abstract. *Clarireedia jacksonii* sp. nov. formerly *Sclerotinia homoeocarpa* F.T. Bennett, one of the causal agents of dollar spot, is the most widespread pathogen in turfgrass systems. Dollar spot (DS) affects both cool- and warm-season grasses, during a wide range of environmental conditions. Field studies were conducted at Clemson University, Clemson, SC, on a creeping bentgrass [*Agrostis stolonifera* L. var. *palustris* (Huds) cv. Crenshaw] putting green for 2 consecutive years from August to October in year 1 and July to September in year 2. The objective of the studies was to evaluate biological control agents (BCAs) and synthetic fungicides at reduced rates for their efficacy controlling dollar spot. Four replications of 1.5 × 1.5-m plots were used in the experimental design. Treatments included the following: *Bacillus subtilis* (BS); plant extract oils (EO) including clove oil + wintergreen oil + thyme oil; extract of *Reynoutria sachalinensis* (RS); *Bacillus licheniformis* (BL); chlorothalonil (CL); and azoxystrobin + propiconazole (AzP). Synthetic fungicides were used at reduced rates in combination with biological control agents, to evaluate curative control efficacy of various combinations. All reduced synthetic programs, except CL + EO, provided acceptable disease severity (≤15%) at the end of year 1 and acceptable (≥7) turfgrass visual quality. Azoxystrobin + propiconazole, CL, AzP + BL, AzP + EO, AzP + BS all provided ≤15% disease severity and ≥7 visual turfgrass quality 14 days after the last application in year 2.

C. jacksonii sp. nov. formerly *S. homoeocarpa* F.T. Bennett, the causal agent of DS, is the most widespread pathogen in turfgrass systems (Bishop et al., 2008; Salgado-Salazar et al., 2018). DS affects both cool- and warm-season grasses, during a wide range of envi-

ronmental conditions. Small tan lesions surrounded by a darker band and often presenting an hourglass appearance in circular patches 10 to 40 mm, and white mycelium characterize DS. Once blighted, turf areas often become necrotic and bare, often susceptible to weed invasion. During favorable conditions, spots may coalesce to form larger irregular shaped patches (Salgado-Salazar et al., 2018; Smiley et al., 2005). Management of DS includes regulating surface moisture levels, proper nitrogen management, and both preventive and curative applications of fungicides. Increased pressure to manage turf in a more ecologically friendly, greener man-

ner has reinvigorated the search for suitable biological fungicides. Aiding the drive are costs associated with synthetic fungicides and perceived risks to humans and the environment.

As an alternative to synthetic products, bacterial and fungal species are being evaluated for their ability to suppress DS. BCAs offer both advantages and disadvantages to turf managers. Potential biological species for use are native to most soils, and thus pose low toxicity risk to humans and wildlife. Synthetic products are subject to degradation, where active parent products are degraded into secondary metabolites that may pose certain risks (Harman, 2006). However, currently available BCAs typically show reduced acute efficacy and residual control. Application timing becomes much more critical, especially during high disease pressure conditions. Packaging and shelf life issues also pose challenges. Most of these products contain living organisms, requiring short-term storage under specific conditions (Kim et al., 1997). In this study, the goal was to investigate if quarter low label rates of the synthetic fungicides CL and AzP in combination with BCAs could provide similar control as full label rates of the synthetic fungicides.

Materials and Methods

Studies were conducted at Clemson University, Clemson, SC (lat. 34.67°N, long. 82.84°W) on a creeping bentgrass [*Agrostis stolonifera* L. var. *palustris* (Huds) cv. Crenshaw] putting green from Aug. to Oct. 2008 (year 1) and July to Sept. 2009 (year 2). The research site was an 11-year-old 85:15 U.S. Golf Association sand:peat-based green construction. Crenshaw creeping bentgrass was selected due to its high susceptibility to DS (McCarty, 2018).

Mowing height was ≈3 mm and occurred 5 to 7 times weekly. Irrigation was applied as needed to prevent moisture stress. Nitrogen was applied annually at 1.81 kg·ha⁻¹ N. Nutrients were supplied via organic-based products using granular and liquid products. Additional potassium (K₂O) and phosphorous (P₂O₅) was applied through separate products to provide a 1–1–2 ratio of N–P–K. Fertility treatments included EndoRoots 3–3–5 (LebanonTurf, Lebanon, PA) at 0.23 kg·ha⁻¹ N monthly, and Novozymes Turf Vigor 9–3–6 (LebanonTurf) was applied at 0.11 kg·ha⁻¹ N biweekly.

Four replications of 1.5 × 1.5-m plots were used in the experimental design. Spray applications were made using a pressurized CO₂ backpack boom sprayer, through 8003 flat-fan nozzles (Tee jet; Spraying Systems Co., Roswell, GA) with a water carrier volume of 374 L·ha⁻¹. Products were applied in cooler morning or evening hours to minimize turfgrass phytotoxicity. Treatments included BCAs and conventional control products (Table 1).

Reduced synthetics. Programs were based on applications of CL (Daconil Ultrex

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Table 1. Fungicide common name, trade name, and manufacturer of products used during a 2-year study on Crenshaw creeping bentgrass for curative control and suppression of dollar spot (*Clarireedia jacksonii*).

Common name	Trade name	Manufacturer
Azoxystrobin + propiconazole	Headway 1.39SC	Syngenta, Greensboro, NC
Chlorothalonil	Daconil Ultrex 82.5WP	Syngenta, Greensboro, NC
<i>Bacillus licheniformis</i> SB3086	EcoGuard 0.36SC	Novozymes, Salem, VA
<i>Reynoutria sachalinensis</i> extract	Regalia 0.48SC	Marrone Bio Innovations, Davis, CA
<i>Bacillus subtilis</i> QST 713	Rhapsody 0.13SC	AgraQuest, Davis, CA
Clove oil, thyme oil, and rosemary oil	Paradigm 3.61L	Parkway Research, Pleasant Plains, IL

Table 2. Treatments evaluated during a 2-year study for the efficacy of combinations of BCAs and synthetic products for dollar spot control on Crenshaw creeping bentgrass.

Treatment	Treatment abbreviation	Rate (L·ha ⁻¹)	Application	Total Applications
Azoxystrobin + propiconazole	AzP	4.77	Biweekly	6
Azoxystrobin + propiconazole + <i>Bacillus licheniformis</i>	AzP+BL	2.39 and 63.6	Biweekly	6
Azoxystrobin + propiconazole + <i>Reynoutria sachalinensis</i>	AzP+RS	2.39 and 5% v/v	Biweekly	6
Azoxystrobin + propiconazole + <i>Bacillus subtilis</i>	AzP+BS	2.39 and 31.8	Biweekly	6
Azoxystrobin + propiconazole + clove, thyme, and rosemary oil	AzP+EO	2.39 and 9.54	Biweekly	6
Chlorothalonil	CL	9.54	Biweekly	6
Chlorothalonil + <i>Bacillus licheniformis</i>	CL+BL	2.39 and 63.6	Biweekly	6
Chlorothalonil + <i>Reynoutria sachalinensis</i>	CL+RS	2.39 and 5% v/v	Biweekly	6
Chlorothalonil + <i>Bacillus subtilis</i>	CL+BS	2.39 and 31.8	Biweekly	6
Chlorothalonil + clove, thyme, and rosemary oil	CL+EO	2.39 and 9.54	Biweekly	6

Table 3. Visual quality on Crenshaw creeping bentgrass in year 1 from a curative dollar spot study. Study initiated 30 July 2008.

Treatment	Visual quality ^a					
	6 Aug.	20 Aug.	3 Sept.	17 Sept.	1 Oct.	15 Oct.
Azoxystrobin + propiconazole	3.0 a ^b	3.5 ab	6.2 a	7.2 a	7.7 ab	8.2 a
Azoxystrobin + propiconazole + <i>Bacillus licheniformis</i>	3.0 a	3.3 ab	4.5 bc	7 ab	7.8 a	8 ab
Azoxystrobin + propiconazole + <i>Reynoutria sachalinensis</i>	3.0 a	3.3 ab	4.3 bcd	6.2 cd	7 bc	7.3 de
Azoxystrobin + propiconazole + <i>Bacillus subtilis</i>	3.0 a	3 b	4.5 bc	6.8 abc	7.2 abc	7.5 cd
Azoxystrobin + propiconazole + clove, thyme, and rosemary oil	3.0 a	3.3 ab	4.8 b	5.8 d	7 bc	7.5 cd
Chlorothalonil	3.0 a	3.3 ab	6.5 a	6.8 abc	7.3 ab	7.8 abc
Chlorothalonil + <i>Bacillus licheniformis</i>	3.0 a	3.7 a	4.3 bcd	6.3 bcd	7.2 abc	7.5 cd
Chlorothalonil + <i>Reynoutria sachalinensis</i>	3.0 a	3.2 ab	4 bcd	5.7 d	6.2 d	7 e
Chlorothalonil + <i>Bacillus subtilis</i>	3.0 a	3 b	3.7 cd	5.7 d	6.5 cd	7.7 bcd
Chlorothalonil + clove, thyme, and rosemary oil	3.0 a	3.2 ab	3.5 d	4.2 e	5.2 e	5.5 f

^aVisual quality was rated on a scale of 1 to 9: 1 = dead turf, 9 = perfect turf, and 7 = minimum acceptable.

^bMeans with the same letter are not significantly different based on Fisher's least significant difference test ($\alpha = 0.05$).

82.5WP) or AzP (Headway 1.39SC) every 14 d at quarter label rates and tank mixed with the designated BCA. Chlorothalonil (2 kg a.i./ha) and AzP (0.4 kg a.i./ha) were considered quarter label rate. Biological control agents were BS formulated as Rhapsody SC; EO clove oil + wintergreen oil + thyme oil formulated as Paradigm L; extract of RS formulated as Regalia L; and BL formulated as EcoGuard SC (Table 2).

Synthetics. Synthetic products, CL (8.2 kg a.i./ha) and AzP (1.6 kg a.i./ha) were applied every 14 d at full label rate. Synthetic programs were the industry standards and BCA treatments were compared (Table 2).

Turfgrass plots were rated weekly for disease severity and visual turfgrass quality. Disease severity ratings used a line intersect grid rating method. Each plot was divided into 289 smaller subplots considered a grid, measuring 100 × 100 mm. Disease in any portion of a grid was considered a "hit." A percentage of disease severity was calculated by taking the number of hits and dividing by the total number of grids multiplied by 100. Severity ratings more aptly show the level of fungicidal activity provided by the various treatments. An acceptable disease severity level was set as ≤15%.

Turfgrass visual quality was rated on a scale of 1 to 9, where 1 = brown dead turf, 7 =

minimum accepted, and 9 = dark green dense turf (Morris and Shearman, 1999). Weekly ratings were averaged each month for statistical analysis.

Areas under disease progress curve (AUDPC) were calculated for each fungicide and rate using the following equation:

$$\text{AUDPC} = \sum [(Y_1 + Y_2)/2](T_2 - T_1), \quad [1]$$

where Y equals rating date and T equals days between rating date (Latin, 2008). The AUDPC calculates the disease epidemic over the course of the rating period.

Statistical analysis. The SAS statistical software package JMP Pro 9.1 (SAS Institute Inc., Cary, NC) was used for analysis of variance (ANOVA) and means separation on all data sets. The ANOVA was used to evaluate the main effects of experimental run per year, fungicide program, and days after initiation, as well as the interactions. When the main effects or interactions were significant, Fisher's least significant difference test ($\alpha = 0.05$) was used to separate means.

Results

Analysis of variance indicated an interaction of experimental run and year; therefore, data were not combined between years and is

presented separately. Experimental run per year was part of the statistical model.

Turfgrass visual quality. In year 1, no differences occurred on 6 Aug. or 20 Aug. (≈3) (Table 3). On 3 Sept., AzP and CL had greater turfgrass visual quality (>6), compared with the reduced synthetic programs (≈4.5) (Table 3). On 17 Sept., AzP had the highest turfgrass visual quality (≈7.2), followed by AzP + BL (≈7), AzP + BS and CL (≈6.8), CL + BL (≈6.3), AzP + RS (≈6.2), AzP + EO (≈5.8), CL + RS and CL + BS (≈5.7), and CL + EO (4.2) (Table 3). On Oct. 1, AzP, AzP + BL, AzP + RS, AzP + BS, AzP + EO, CL, and CL + BL experienced highest turfgrass visual quality (>≈7); compared with CL + RS, CL + BS (<≈6.5), and CL + EO (≈5.2) (Table 3). On Oct. 15, AzP, AzP + BL, CL experienced highest turfgrass visual quality (>≈7.8), followed by AzP + RS, AzP + BS, AzP + EO, CL + BS (≈7.5); CL + RS (≈7), and CL + EO (≈5.5) (Table 3).

In year 2, no differences occurred on 1 July or 14 July (≈3) (Table 4). On 4 Aug., AzP and CL had greater turfgrass visual quality (≈7), compared with the reduced synthetic programs (>≈4.7) (Table 4). On 18 Aug., AzP, AzP + BL and CL experienced highest turfgrass visual quality (≈7.3), followed by AzP + BS and AzP + EO (≈6.7),

Table 4. Visual quality on Crenshaw creeping bentgrass in year 2 from a curative dollar spot study. Study initiated 25 June 2009.

Treatment	Visual quality ^z					
	1 July	14 July	4 Aug.	18 Aug.	31 Aug.	11 Sept.
Azoxystrobin + propiconazole	3.0 a ^y	3.0 b	7.0 a	7.3 a	7.7 a	7.8 a
Azoxystrobin + propiconazole + <i>Bacillus licheniformis</i>	3.0 a	3.0 b	4.7 b	7.2 ab	7.6 a	7.3 a
Azoxystrobin + propiconazole + <i>Reynoutria sachalinensis</i>	3.0 a	3.0 b	4.2 bc	6.5 cd	6.8 b	5.3 b
Azoxystrobin + propiconazole + <i>Bacillus subtilis</i>	3.0 a	3.0 b	4.2 bc	6.7 bc	7.0 b	5.7 b
Azoxystrobin + propiconazole + clove, thyme, and rosemary oil	3.0 a	3.0 b	4.7 b	6.7 bc	6.8 b	5.8 b
Chlorothalonil	3.0 a	3.0 b	6.7 a	7.3 a	7.5 a	6.0 b
Chlorothalonil + <i>Bacillus licheniformis</i>	3.0 a	3.2 ab	4.2 bc	6.0 de	6.7 b	5.3 b
Chlorothalonil + <i>Reynoutria sachalinensis</i>	3.0 a	3.0 b	3.8 cd	6.5 cd	5.5 d	4.5 b
Chlorothalonil + <i>Bacillus subtilis</i>	3.0 a	3.0 b	3.5 cd	5.7 ef	6.0 c	3.5 d
Chlorothalonil + clove, thyme, and rosemary oil	3.0 a	3.0 b	3.3 d	5.2 f	6.8 b	3.5 d

^zVisual quality was rated on a scale of 1 to 9: 1 = dead turf, 9 = perfect turf, and 7 = minimum acceptable.^yMeans with the same letter are not significantly different based on Fisher's least significant difference test ($\alpha = 0.05$).

Table 5. Disease severity on Crenshaw creeping bentgrass in year 1 from a curative dollar spot study. Study initiated 30 July 2008.

Treatment	Disease severity ^z					
	6 Aug.	20 Aug.	3 Sept.	17 Sept.	1 Oct.	15 Oct.
Azoxystrobin + propiconazole	99.7 a ^y	92.4 bc	36.0 b	6.6 e	2.1 de	0.3 c
Azoxystrobin + propiconazole + <i>Bacillus licheniformis</i>	98.3 a	96.1 abc	71.5 a	10.6 de	1.4 e	0.7 c
Azoxystrobin + propiconazole + <i>Reynoutria sachalinensis</i>	98.2 a	95.9 abc	84.5 a	24.6 cbd	13.3 c	3.1 c
Azoxystrobin + propiconazole + <i>Bacillus subtilis</i>	99.6 a	94.6 abc	80.0 a	19.8 cde	8.6 cde	2.7 c
Azoxystrobin + propiconazole + clove, thyme, and rosemary oil	98.8 a	97.2 a	73.8 a	34.1 bc	15.2 bc	3.1 c
Chlorothalonil	97.7 a	92.3 bc	35.6 b	8.5 e	1.8 de	0.3 c
Chlorothalonil + <i>Bacillus licheniformis</i>	97.4 a	91.8 c	68.8 a	19.6 cde	11.8 cde	3.6 c
Chlorothalonil + <i>Reynoutria sachalinensis</i>	97.0 a	93.3 abc	83.3 a	35.5 b	25.7 b	12.1 b
Chlorothalonil + <i>Bacillus subtilis</i>	98.0 a	93.8 abc	88.2 a	34.9 bc	12.4 cd	4.4 c
Chlorothalonil + clove, thyme, and rosemary oil	97.9 a	96.7 ab	90.6 a	74.6 a	65.5 a	42.5 a

^zDisease severity was determined using a grid intersect on 3 m² plots, with 15% = maximum acceptable disease severity rating.^yMeans with the same letter are not significantly different based on Fisher's least significant difference test ($\alpha = 0.05$).

Table 6. Disease severity on Crenshaw creeping bentgrass in year 2 from a curative dollar spot study. Study initiated 25 June 2009.

Treatments	Disease severity ^z					
	1 July	14 July	4 Aug.	18 Aug.	31 Aug.	11 Sept.
Azoxystrobin + propiconazole	100 a ^y	94.7 c	5.8 e	1.5 d	1.0 e	1.0 f
Azoxystrobin + propiconazole + <i>Bacillus licheniformis</i>	100 a	96.9 abc	84.6 c	4.6 d	3.1 de	9.5 ef
Azoxystrobin + propiconazole + <i>Reynoutria sachalinensis</i>	100 a	96.7 abc	93.9 ab	20.2 c	17.5 bc	44.6 c
Azoxystrobin + propiconazole + <i>Bacillus subtilis</i>	100 a	95.4 bc	94.5 ab	13.9 c	10.4 cd	35.1 d
Azoxystrobin + propiconazole + clove, thyme, and rosemary oil	100 a	97.8 abc	79.8 c	18.4 c	14.7 c	53.5 bc
Chlorothalonil	100 a	95.6 abc	17.5 d	5.5 d	1.4 de	18.2 e
Chlorothalonil + <i>Bacillus licheniformis</i>	100 a	96.9 abc	88.7 bc	20.5 c	18.8 bc	58.7 b
Chlorothalonil + <i>Reynoutria sachalinensis</i>	100 a	99.7 a	94.0 ab	45.3 a	40.3 a	85.6 a
Chlorothalonil + <i>Bacillus subtilis</i>	100 a	98.9 abc	96.4 ab	32.3 b	24.2 b	90.9 a
Chlorothalonil + clove, thyme, and rosemary oil	100 a	97.8 abc	98.4 a	50.3 a	43.2 a	91.5 a

^zDisease severity was determined using a grid intersect on 3 m² plots, with 15% = maximum acceptable disease severity rating.^yMeans with the same letter are not significantly different based on Fisher's least significant difference test ($\alpha = 0.05$).

AzP + RS and CL + RS (≈ 6.5), CL + BL (≈ 6), CL + BS (≈ 5.7), and CL + EO (≈ 5.2) (Table 4). On 31 Aug., AzP, AzP + BL and CL experienced highest turfgrass visual quality ($> \approx 7.5$); followed by AzP + BS (≈ 7), AzP + RS, AzP + EO, and CL + EO (≈ 6.8), CL + BL (≈ 6.7), CL + BS (≈ 6), and CL + RS (≈ 5.5) (Table 4). On 11 Sept., AzP and AzP + BL experienced the highest turfgrass visual quality ($> \approx 7.3$); followed by CL (≈ 6), AzP + EO (≈ 5.8), AzP + BS (≈ 5.7), AzP + RS (≈ 5.3), CL + BS (≈ 4.5), CL + BS and CL + EO (≈ 3.5) (Table 4).

Disease severity. In year 1, no differences occurred on 5 Aug. or 20 Aug. ($> \approx 97\%$) (Table 5). On 3 Sept., AzP and CL experienced the lowest disease severity ($\approx 36\%$), compared with the reduced synthetic programs ($> \approx 68\%$) (Table 5). On 17 Sept., AzP, CL and AzP + BL (all $< \approx 11\%$) experienced the lowest disease severity; followed by CL +

BL, AzP + BS, AzP + RS, AzP + EO, CL + BS, CL + RS (all $< \approx 36\%$), and CL + EO ($\approx 75\%$) (Table 5). On 1 Oct., AzP + BL, CL, AzP, AzP + BS, CL + BL, CL + BS, AzP + RS, and AzP + EO experienced the lowest (all $< \approx 15.2\%$) disease severity; followed by CL + RS ($\approx 26\%$) and CL + EO ($\approx 66\%$) (Table 5). On 15 Oct., AzP and CL, AzP + BL, AzP + BS, AzP + RS, AzP + EO, CL + BL and CL + BS experienced the lowest (all $< \approx 5\%$) disease severity; followed by CL + RS ($\approx 12\%$) and CL + EO ($\approx 43\%$) (Table 5).

In year 2, no differences occurred in disease severity on 1 July or 14 July, with all treatments $> \approx 95\%$ (Table 6). On 4 Aug., AzP ($\approx 6\%$) and CL ($\approx 18\%$) experienced the lowest disease severity, compared with the reduced synthetic programs ($> \approx 79\%$) (Table 6). On 18 Aug., AzP + BL, CL, and AzP experienced lowest (all $< \approx 6\%$) disease severity; followed by AzP + BS ($\approx 14\%$),

AzP + EO ($\approx 18\%$), AzP + RS ($\approx 20\%$), CL + BL ($\approx 21\%$), CL + BS ($\approx 32\%$), CL + RS ($\approx 45\%$) and CL + EO ($\approx 50\%$) (Table 6). On 31 Aug., AzP, CL, and AzP + BL experienced lowest ($< \approx 3\%$) disease severity; followed by AzP + BS ($\approx 10\%$), AzP + EO ($\approx 15\%$), AzP + RS ($\approx 18\%$), CL + BL ($\approx 19\%$), CL + BS ($\approx 24\%$), CL + RS ($\approx 40\%$) and CL + EO ($\approx 43\%$) (Table 6). On 11 Sept., AzP and AzP + BL experienced lowest ($< 10\%$) disease severity; followed by CL ($\approx 18\%$), AzP + BS ($\approx 35\%$), AzP + RS ($\approx 45\%$), AzP + EO ($\approx 54\%$), CL + BL ($\approx 59\%$), CL + RS ($\approx 86\%$), CL + BS ($\approx 91\%$) and CL + EO ($\approx 92\%$) (Table 6).

Area under disease progress curve. In year 1, AzP (≈ 241), CL (≈ 245) and AzP + BL (289) experienced the lowest disease pressure; followed by CL + BL (≈ 308), AzP + BS (≈ 310), AzP + RS (≈ 328), AzP + EO (≈ 329), CL + BS (≈ 339), CL + RS

Table 7. Area under disease progress curves (AUDPC) for dollar spot on Crenshaw creeping bentgrass in years 1 and 2.

Treatments	AUDPC ^z	
	2008	2009
Azoxystrobin + propiconazole	240.6 gh ^y	211.3 h
Azoxystrobin + propiconazole + <i>Bacillus licheniformis</i>	289.0 efg	287.0 efg
Azoxystrobin + propiconazole + <i>Reynoutria sachalinensis</i>	327.6 cde	378.3 bc
Azoxystrobin + propiconazole + <i>Bacillus subtilis</i>	309.6 de	353.0 bcd
Azoxystrobin + propiconazole + clove, thyme, and rosemary oil	329.0 cde	348.6 bcd
Chlorothalonil	245.3 gh	256.0 fgh
Chlorothalonil + <i>Bacillus licheniformis</i>	308.0 def	384.6 b
Chlorothalonil + <i>Reynoutria sachalinensis</i>	353.3 bcd	450.0 a
Chlorothalonil + <i>Bacillus subtilis</i>	339.3 bcde	446.6 a
Chlorothalonil + clove, thyme, and rosemary oil	474.0 a	497.3 a

^zAUDPC was calculated using $\sum [(Y_1 + Y_2)/2] (T_2 - T_1)$. Lower values = better control.

^yMeans with the same letter are not significantly different based on Fisher's least significant difference test ($\alpha = 0.05$).

(≈ 353) and CL + EO (≈ 474) (Table 7). In year 2, AzP, CL and AzP + BL experienced lowest (≈ 210 to 287) disease pressure; followed by AzP + EO (≈ 349), AzP + BS (≈ 353), AzP + RS (≈ 378), CL + BL (≈ 385), CL + BS (≈ 447), CL + RS (≈ 450), and CL + EO (≈ 497) (Table 7).

Discussion

Compared with a previous trial, programs containing one-quarter rates of synthetic fungicides in combination with the BCAs, provided increased efficacy (Lo et al., 1997). Previously, synthetics + BCAs were used on a 30-day interval (Lo et al., 1997) compared with the 14-day interval in the present study, possibly indicating proper timing of synthetic products during increased disease pressure. Previous research indicates applications of BCAs provided both rhizosphere and foliar inoculation (Lo et al., 1997). Weekly applications of BCAs were able to provide control similar to that by synthetic fungicides. Monthly spray applications of BCAs were able to reduce DS; however, these did not achieve control similar to weekly applications. Research conducted at Purdue University during the spring indicated similar trends. Reduced rates of CL + BCAs achieved similar control as full labeled rates of CL (Latin, 2008). Treatments at the Purdue University study were applied on either a 7- or 14-day interval, whereas treatments during the present study were all applied on a 14-day interval. Bowers and Locke (2000) reported reductions in *Fusarium oxysporum* populations when clove oil was applied. Improved turfgrass visual quality was also noted with AzP and BL during both years of this study. Marvin et al. (2020) reported reduced label rates of CL and AzP in combination with BCAs improved DS control during in vitro and preventive field trials. Further greenhouse evaluations should be conducted to determine the possible physiological effects these fungicides are having on the plant, leading to enhanced color. Due to earlier disease pressure, the second study year was initiated earlier than the first year. Disease

pressure increased at the end of year 2. Treatments were stopped earlier than the first year to evaluate any "rebound effect" in disease pressure. Treatments were stopped on 17 Sept. in year 1 and 18 Aug. in year 2. From 31 Aug. to 11 Sept. in year 2, an increase in disease severity was noted. In general, continued applications may be required until environmental conditions are no longer conducive to DS outbreaks.

Previous research evaluating environmental impact quotient of BCAs revealed less impact to the environment than conventional synthetic programs (Grant and Rossi, 2006).

In conclusion, all reduced programs, except CL + EOs, provided acceptable disease severity ($\leq 15\%$) at the end of year 1 and acceptable (≥ 7) turfgrass visual quality. Results from year 1 are similar to Tomaso-Peterson (2006), where control was achieved using BCAs when disease pressure was not severe. In year 2, AzP, CL, AzP + BL, AzP + EO, and AzP + BS all provided $\leq 15\%$ disease severity and ≥ 7 visual turfgrass quality 14 d after the last application. At 28 d after the last application in year 2, an increase in disease severity occurred. The only treatments to maintain adequate DS control and visual turfgrass quality ratings were AzP and AzP + BL. The ability of AzP + BL to maintain acceptable turf quality is a promising step in developing programs with reduced amounts of synthetic products, while providing similar control. Future research should focus on understanding the competitive relationship between BCAs and turfgrass pathogens. Understanding the growth and development dynamics between the BCAs and turfgrass pathogens will lead to improved control efficacy.

Literature Cited

- Bishop, P., J. Sorochan, B.H. Ownley, T.J. Samples, A.S. Windham, M.T. Windham, and R.N. Trigiano. 2008. Resistance of *Sclerotinia homoeocarpa* to iprodione, propiconazole, and thiophanate-methyl in Tennessee and Northern Mississippi. *Crop Sci.* 48:1615–1620.
- Bowers, J.H. and J.C. Locke. 2000. Effect of botanical extracts on the population density of *Fusarium oxysporum* in soil and control of Fusarium wilt in the greenhouse. *Plant Dis.* 84(3): 300–305.
- Grant, J.A. and F.S. Rossi. 2006. Long-term evaluation and improvement of golf turf management systems with reduced chemical pesticide inputs: Preliminary Report. Ithaca, NY. Cornell University. <<https://hdl.handle.net/1813/43166>>.
- Harman, G.E. 2006. Overview of mechanisms and uses of *Trichoderma* spp. *Phytopathology* 96:190–194.
- Kim, D.S., R.J. Cook, and D.M. Weller. 1997. *Bacillus* sp. L324-92 for biological control of three root diseases of wheat grown with reduced tillage. *Phytopathology* 87:551–558.
- Latin, R. 2008. Interaction of biofungicides and chlorothalonil for control of dollar spot on creeping bentgrass. Report. Purdue University, West Lafayette, IN.
- Lo, C.T., E.B. Nelson, and G.E. Harman. 1997. Improved biocontrol efficacy of *Trichoderma harzianum* 1295-22 for foliar phases of turf diseases by use of spray applications. *Plant Dis.* 81:1132–1138.
- Marvin, J.W., R.A. Kerr, L.B. McCarty, W.C. Bridges, S.B. Martin, and C.E. Wells. 2020. *In vitro* and preventative field evaluations of potential biological control agents and synthetic fungicides for control of *Clariireedia jacksonii* sp. nov. *J. Plant Sci. Phytopathol.* 4:001–008.
- McCarty, L.B. 2018. Golf turf management. CRC Press, Boca Raton, FL.
- Morris, K.N. and R.C. Shearman. 1999. NTEP turfgrass evaluation guidelines. National Turfgrass Evaluation Program, Beltsville, MD.
- Salgado-Salazar, C., L.A. Beirn, A. Ismaiel, M.J. Boehm, I. Carbone, A.I. Putman, L.P. Tredway, B.B. Clarke, and J.A. Crouch. 2018. *Clariireedia*: A new fungal genus comprising four pathogenic species responsible for dollar spot disease of turfgrass. *Fungal Biol.* 122(8):761–773.
- Smiley, R.W., P.H. Dernoeden, and B.B. Clarke. 2005. Compendium of turfgrass diseases. 3rd ed. APS Press, Saint Paul, MN.
- Tomaso-Peterson, M. 2006. A demonstration trial of biofungicides with efficacy for controlling dollar spot in turfgrass. Mississippi Agricultural & Forestry Experiment Station. (23)17.