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Tolerance of Bentgrass Species and Cultivars to Methiozolin

A Thesis submitted in partial satisfaction
of the requirements for the degree of

Master of Science

in

Plant Biology

by

Nicholas Ryan Hoisington

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Dedication

This thesis is dedicated to my loving wife for all her support.

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Hoisington et al.: Bentgrass Tolerance

Tolerance of Bentgrass Species and Cultivars to Methiozolin

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Methiozolin is a new herbicide from South Korea currently under development for pre- (PRE) and postemergence (POST) control of annual bluegrass in bentgrass and most other cool- and warm-season turfgrasses. Greenhouse studies were conducted in 2012 at the University of California, Riverside, CA, and Auburn University, Auburn, AL, to evaluate the relative tolerance of nine creeping bentgrass (CRBG) cultivars, velvet bentgrass (VBG) and colonial bentgrass (COBG) to methiozolin at 0, 0.6, 1.1, 2.2, 4.5 and 9.0 kg ai ha⁻¹. Methiozolin was applied 7 weeks after seeding followed by a second application at 12 weeks. Methiozolin rates that produced 25% injury (TI₂₅) and 50% clipping dry weight reduction relative to an untreated control for each species or cultivar (GR₅₀) were calculated using four-parameter logistic regression. Results indicated that TI₂₅ rates at 56 days after initial treatment (DAIT) (plants had received two methiozolin treatments 35 days apart, with the second application 21 days before sampling) were the most accurate in describing relative tolerance among BG species. Overall, CRBG was more tolerant to methiozolin than VBG or COBG. Respective rates of methiozolin that caused TI₂₅ were 1.1, 0.2, and 0.3 kg ai ha⁻¹ for CRBG, VBG and COBG. Furthermore, VBG and COBG are unlikely to tolerate sequential applications necessary to control annual bluegrass with methiozolin. Herbicide rates that caused TI₂₅ and GR₅₀ decreased with the second application; therefore, suggesting methiozolin soil residual activity. Creeping bentgrass ‘Penn A-4’ was the most tolerant of methiozolin application; TI₂₅ required 4.5 kg ai ha⁻¹ at 28 DAIT but only 2.5 kg ai ha⁻¹ with two applications and longer exposure by 56 DAIT. All CRBG cultivars tested tolerated methiozolin at 0.5 kg ai ha⁻¹, the recommended sequential application rate.

Nomenclature: Methiozolin, MRC-01; 5-(2,6-difluoro-benzyloxymethyl)-5-methyl-3-(3-methyl-thiophen-2-yl)-4,5-dihydro-isoxazole; annual bluegrass, *Poa annua* L.; bentgrass, *Agrostis* spp.; creeping bentgrass, *Agrostis stolonifera* L.; colonial bentgrass, *Agrostis capillaris* L.; velvet bentgrass, *Agrostis canina* L.; herbicide.

Key words: Four parameter logistic regression, herbicide sensitivity, growth reduction, leaf necrosis.

Bentgrasses (BG) (*Agrostis* spp.) are utilized for intensively managed turf areas including tees, fairways, and putting greens due to their superior color, density, and texture when maintained at a low height of cut (Beard 1973). The dominate species in the U.S. is creeping bentgrass (CRBG) (*Agrostis stolonifera* L.), but velvet bentgrass (VBG) (*Agrostis canina* L.) and colonial bentgrass (COBG) (*Agrostis capillaries* L.) are also used in climates where they are best adapted. Annual bluegrass (AB) (*Poa annua* L.) can be a desirable turfgrass species, but is often considered a weed in stands of newly established or highly maintained BG where it is one of the biggest challenges faced by turfgrass managers (Beard 1973; Lycan 2005). Annual bluegrass control in BG putting greens is currently managed by plant growth regulators and cultural practices aimed at giving the competitive advantage to BG (Flessner et al. 2012; Johnson and Murphy 1996). Although several herbicides are available for postemergence (POST) control of AB, none are registered for use on BG putting greens in the U.S., and only bensulide is registered for preemergence (PRE) control (Anonymous 2009; Flessner et al. 2012).

Methiozolin is a new turfgrass herbicide in the isoxazoline chemical family. It was originally developed for herbicidal control of barnyardgrass (*Echinochloa galli* (L.) Gaertn.) in rice (Hwang et al. 2005). Methiozolin is registered in South Korea and is under development in Japan, Australia, and the U.S. for PRE and POST control of AB in BG turf including putting greens. Previous research has demonstrated that PRE and POST applications of methiozolin can effectively control AB in CRBG putting greens with little or no bentgrass injury (Brosnan et al. 2012; Flessner et al. 2012; Haguewood et

al. 2012; Han and Kaminski 2012; Hart et al. 2012; McCullough and Gómez de Barrea 2012; Hoyle et al. 2012; Trapp et al. 2012).

Methiozolin uptake is through foliage and roots with moderate acropetal translocation in both AB and CRBG (Brosnan et al. 2012; Flessner et al. 2012; McCullough and Gómez de Barrea 2012). Studies have also indicated that combined foliar and root applications or root applications alone have more success in controlling AB than foliar applications alone (Brosnan et al. 2012; Flessner et al. 2012). Methiozolin currently has two proposed modes of action. The first, less-studied mechanism, suggests that methiozolin inhibits cell wall biosynthesis by a mechanism different from other herbicides that inhibit cell wall biosynthesis (Flessner et al. 2012; Hwang et al. 2005; Lee et al 2007; Nam et al. 2012). Whether this is a primary or secondary effect of methiozolin remains unknown (Flessner et al. 2012; Hwang et al. 2005; Lee et al 2007; Nam et al. 2012). A second mode of action identified by Grossman et al. (2012) suggested that methiozolin was a tyrosine aminotransferase (TAT) inhibitor. Inhibition of TAT's involvement in the synthesis pathway of plastoquinone and tocopherols would have significant downstream effects; loss of plastoquinone function could ultimately lead to photo-oxidation and degradation of chlorophyll in growing tissues (Grossman et al. 2012).

Field studies have demonstrated that a single application of methiozolin is less effective in controlling AB in CRBG than sequential applications (Brosnan et al. 2012; Han and Kaminski 2012; Hoyle et al. 2012). Sequential applications of methiozolin at two-week intervals totaling 3.0 to 3.36 kg ai ha⁻¹ was the most successful in controlling

AB in putting green studies (Brosnan et al. 2012; Han and Kaminski 2012; McCullough et al. 2012). Field experiments previously conducted at University of California, Riverside (UCR) suggest there may be differential tolerance to methiozolin among BG species and cultivars (unpublished 2012). To our knowledge no research has been conducted to investigate this response to methiozolin; however, several studies documented differences in BG tolerance to other herbicides. Bentgrass species tolerance to herbicides such as fenoxaprop, metamifop, and bispyrabac-sodium was variable (Henry and Hart 2004; Kaminski 2005; McDonald et al. 2006; Straw et al. 2012). Tolerance of CRBG to bispyrabac-sodium was greater than that of COBG and VBG (Kaminski 2005; McDonald 2006). Moreover, some CRBG cultivars exhibited half the phytotoxicity of VBG when treated with metamifop (Straw et al. 2012). In contrast, VBG tolerated higher rates of fenoxaprop than CRBG (Henry and Hart 2004).

The objectives of this research were to determine responses of CRBG, COBG, and VBG and nine CRBG cultivars to methiozolin, and to identify tolerable use rates under greenhouse conditions.

Materials and Methods

Research was conducted in greenhouses at UCR and at Auburn University (AU), in Auburn, AL. The AU study was conducted from 16 February to 31 May 2012. When temperatures exceeded 23 °C, plants were cooled with an evaporative cooling panel system. Relative humidity averaged 68%. Plants received natural daylight only and photoperiods were 11 h and 14 h for February and May, respectively. The UCR study was conducted from 29 May to 11 September 2012. Average day/night air temperatures were 28/20 °C, and relative humidity averaged 53%. Plants received natural daylight only and photoperiods were approximately 14 h and 12.5 h for May and September, respectively.

Nine cultivars of CRBG were chosen based upon several criteria including widespread use, breeding background (i.e., genetic diversity), and observations in the field pertaining to tolerance or sensitivity to methiozolin. The CRBG cultivars were: Focus, T-1, Penn G-2, Bengal, 007, Tyee, Penn A-4, 96-2, and Penncross (Table 1). Velvet bentgrass and COBG cultivars were SR7200 and SR 7150, respectively (Seed Research of Oregon, Corvallis, OR).

Both locations utilized 10.5 x 10.5 x 10.2 cm pots in the research. Rootzone mix used at AU was a 90:10 v/v (sand: peat), pH 5.3. At UCR, the mix was a G-15S, 85:15 v/v (sand:organic matter), pH 6.8 (P.W. Gillibrand Co., Inc., CA). Bentgrasses were seeded at 48 kg ha⁻¹ in both locations and allowed to establish for 7 weeks. Overhead irrigation was applied as needed to prevent drought stress. During establishment, plants were fertilized every two weeks (Miracle-Gro Water Soluble All Purpose Plant Food; 28-

8-16; 5.8 kg N ha⁻¹). Grasses were maintained at a height of 1.3 cm using scissors prior to initial treatment application.

At both locations, methiozolin was applied at 0, 0.6, 1.1, 2.2, 4.5, and 9.0 kg ai ha⁻¹ at 7 and 12 weeks after seeding, which are 0 and 35 days after initial treatment (DAIT), respectively. Treatments at AU were applied using an enclosed research spray cabinet with an 8006VS nozzle (Spraying Systems Co., IL). At UCR, treatments were applied using a four-nozzle CO₂-powered backpack sprayer (8004VS, Spraying Systems Co., IL). Both spraying systems were set to deliver 1222 L ha⁻¹. Although the suggested application timing for methiozolin is every 2 weeks, a 5-week interval was utilized in these experiments to provide sufficient time to assess turf response following each application.

Turfgrass injury and clipping weights were determined 14, 28, 42, and 56 DAIT. Injury (INJ) was visually rated on a scale of 0% to 100%, where 0% = no turfgrass injury visible and 100% = complete necrosis of all the plants in the pot. Grasses were mowed to a ht of 1.27 cm, and clippings collected every 2 wk starting 14 DAIT. Dry weights were determined after drying clippings in a forced-air oven at 80 °C for 72 h. Clipping dry weight (DW) as a percent of the untreated control was calculated using Equation 1, allowing direct comparison of relative growth among bentgrass species and cultivars.

$$\text{Percent DW of mean control (DWPC)} = \left(1 - \frac{(\text{Average DW Control} - \text{DW Sample})}{\text{Average DW Control}}\right) \times 100 \quad [1]$$

The experimental design at both locations was a randomized complete-block with four replications in individual pots when comparing CRBG cultivars. When comparing BG species, the nine CRBG cultivars were averaged into four replicates to directly

compare CRBG to COBG and VBG species. Scatter plots of INJ vs. methiozolin rate and DWPC vs. methiozolin rate suggested nonlinear relationships between respective variables. A four-parameter logistic curve, as described by Seefeldt et al. (1995), best describes non-linear herbicidal dose response relationships. SigmaPlot software (version 12.3; Systat Software, Inc., CA) was used to conduct regression analyses fitting INJ and DWPC data to a four parameter logistic curve. The methiozolin rate that caused 25% turfgrass injury (TI_{25}) and that caused a 50% reduction in clipping DW as a percentage of the untreated control (GR_{50}) was calculated for each replicate. Twenty-five percent injury was chosen by the authors because it represented minimally acceptable turfgrass quality and plant health. Fifty percent growth reduction in DWPC is the most widely used value in four parameter logistic regression analysis for comparing plant sensitivities to herbicides and thus was chosen for comparison (Seefeldt et al. 1995). Methiozolin rate that resulted in TI_{25} and GR_{50} values were subjected to ANOVA using Statistix 8 software (version 8, Analytical Software, FL) followed by Fisher's protected least significant difference test at the 0.05 probability level when appropriate.

Results and Discussion

Data collected at 14 and 42 DAIT did not present adequate injury or growth reduction to accurately perform logistic regression analyses. This is likely due to the proximity of treatment application to sample collection combined with methiozolin's slow activity; therefore, data are not shown for these dates. For the purpose of this study, only data collected 28 and 56 DAIT will be discussed.

Differences Among Bentgrass Species. Four-parameter logistic regression closely estimated species injury and growth reduction caused by methiozolin at 28 and 56 DAIT. For DWPC, regression coefficients ranged from 0.83 to 0.99 at 28 DAIT and 0.84 to 0.99 at 56 DAIT (Figures 1 and 3). Regression coefficients for INJ ranged from 0.95 to 0.99 at 28 DAIT and 0.89 to 0.99 at 56 DAIT (Figures 2 and 4). A period of “post inhibition growth enhancement”, similar to effects of growth regulators like trinexapac-ethyl and paclobutrazol, was revealed for CRBG cultivars at 28 DAIT at both locations, and will be hereafter referred to as the “rebound effect” (Branham and Beasley 2007; Ervin and Zhang 2008; Fagerness and Yelverton 2000; Kreuser and Soldat 2011). Creeping bentgrass demonstrated a mean increase in growth of 153% at AU (Figure 2); the rebound effect was not observed for COBG or VBG at AU. At UCR, CRBG exhibited a rebound effect for the 0.56, 1.12 and 2.24 kg ai ha⁻¹ methiozolin treatments with respective increases in growth of 156, 172, and 106%; VBG treated with 0.56 kg ai ha⁻¹ methiozolin showed a 164% increase in growth (Figure 2). The rebound effect was not observed for COBG at UCR (Figure 2). There was no rebound effect observed at 56

DAIT at either location; this could be attributed to the proximity of the collection date to the second application, which occurred 35 DAIT (Figure 4).

Analysis of GR₅₀ at both 28 and 56 DAIT revealed a significant two-way interaction between species and location at the 0.05 probability level. Consequently, GR₅₀ data were subjected to Fisher's Protected LSD and presented separately for each combination of grass species and location (Table 2). At AU 28 DAIT, CRBG GR₅₀ was achieved with a methiozolin rate of 1.9 kg ai ha⁻¹ compared to 0.4 kg ai ha⁻¹ for COBG and VBG (Table 2). The methiozolin rates required to achieve GR₅₀ were higher for each grass species at UCR than at AU (Table 2); at UCR, the rates were 3.2, 0.9 and 1.1 kg ai ha⁻¹ for CRBG, COBG and VBG, respectively. Despite the difference in the magnitude of the rate for GR₅₀ between locations, similar trends were observed among grass species; the methiozolin rate for GR₅₀ of CRBG was significantly greater than that of COBG and VBG at both locations. In contrast, at 56 DAIT, a higher methiozolin rate was required for CRBG GR₅₀ at AU than at UCR: compare 1.1 and 0.8 kg ai ha⁻¹ at AU and UCR, respectively (Table 2). Differences in methiozolin rate for GR₅₀ between COBG (0.6 kg ai ha⁻¹) and VBG (0.3 kg ai ha⁻¹) were detected only at AU; this difference was not observed at UCR, where GR₅₀ was achieved at 0.4 kg ai ha⁻¹ for both species (Table 2).

Analysis of variance for TI₂₅ at 28 DAIT revealed a significant species effect and contemporary significant location effect at the 0.05 probability level; therefore, data were initially pooled over location and presented separately for grass species, and subsequently pooled over grass species and presented separately for each location. Creeping bentgrass

showed greater tolerance to methiozolin in comparison to the other species, with TI₂₅ at 2.4 kg ai ha⁻¹ (Table 3). When data were pooled across grass species, UCR resulted in less injury than AU, with TI₂₅ requiring a rate of 1.5 kg ai ha⁻¹ at UCR compared to only 0.8 kg ai ha⁻¹ at AU (data not shown). Brosnan et al. (2013) hypothesized that differences in soil type can affect methiozolin tolerance. Soil type may have caused the difference among locations observed in this study. At 56 DAIT, only species effect was observed for TI₂₅ (p = 0.05); therefore, data were pooled across location and presented separately for grass species. Creeping bentgrass was the least injured, 56 DAIT, from application of methiozolin; mean TI₂₅ observed for CRBG was attained with 1.1 kg ai ha⁻¹ when averaged across locations (Table 3). In contrast, COBG and VBG required only 0.3 and 0.2 kg ai ha⁻¹ methiozolin, respectively, to cause 25% INJ (Table 3).

Except for CRBG at AU 28 DAIT, methiozolin rates that caused 25% INJ (Table 3) were lower than methiozolin rates that caused 50% reduction in growth for respective species and rating dates (Table 2). These results suggest that methiozolin rates that cause 50% reduction in growth would cause unacceptable injury levels for turfgrass; consequently, in this study TI₂₅ rates were considered for species tolerance. Although, after one application of methiozolin, rates that caused TI₂₅ for COBG and VBG matched the recommended rate for methiozolin (0.5 kg ai ha⁻¹) on putting greens, control of AB requires multiple sequential applications of methiozolin (Brosnan et al. 2012; Han and Kaminski 2012; McCullough et al. 2012). Therefore, TI₂₅ rates 56 DAIT (two applications) are thought to be the most accurate indicator of species tolerance in our study. Results from this study suggest that CRBG is tolerant to two applications of

methiozolin, whereas COBG and VBG species are sensitive to sequential applications of methiozolin at rates that will effectively control AB.

Differences Among Creeping Bentgrass Cultivars. Four-parameter logistic regression analysis, for both DWPC and INJ data, was performed on each replicate for each of nine CRBG cultivars used in this study. Regression analysis fit closely to both DWPC and INJ data; regression coefficients for DWPC ranged from 0.57 to 1.00 at 28 DAIT and from 0.61 to 1.00 at 56 DAIT; regression coefficients for INJ data ranged from 0.92 to 1.00 at 28 DAIT and from 0.84 to 1.00 at 56 DAIT. Due to the large number of cultivars graphical representation of the regressions is not shown.

Analysis of variance of CRBG cultivar GR₅₀ data revealed no significant effects nor interactions among CRBG cultivars and location 28 DAIT; however, a cultivar by location interaction was detected 56 DAIT ($p = 0.05$). Rates of methiozolin resulting in GR₅₀ ranged from 0.5 ('Focus' and '007') to 2.6 kg ai ha⁻¹ ('Penn A-4') at AU and 0.7 ('T-1') to 1.0 kg ai ha⁻¹ ('Penn A-4') at UCR 56 DAIT (Table 4). Creeping bentgrass 'Penn A-4' demonstrated the greatest tolerance to the growth regulation effect of methiozolin at AU. This cultivar also required the highest rate for GR₅₀ at UCR, but the rate was not significantly different from that for other cultivars (Table 4).

Analysis of variance of mean TI₂₅ rates indicated location by cultivar interactions for both 28 and 56 DAIT at the 0.05 probability level. Methiozolin rates that caused TI₂₅ 28 DAIT ranged from 1.3 ('Penncross') to 4.5 kg ai ha⁻¹ ('Penn-A-4'; Table 5); these results are consistent with those of McNulty et al. 2011 who reported no injury to 'Penn

A-4' when methiozolin was applied at a rate as high as 4 kg ai ha⁻¹. At 56 DAIT, methiozolin rate for TI₂₅ ranged from 0.6 to 2.5 kg ai ha⁻¹ (Table 5). Across both locations and rating dates 'Penn A-4' exhibited the greatest tolerance to methiozolin with a TI₂₅ of 4.5 and 2.5 kg ai ha⁻¹ methiozolin at 28 and 56 DAIT, respectively (Table 5). At UCR 28 DAIT, only 'Bengal' (3.8 kg ai ha⁻¹) required the same rate of methiozolin as 'Penn A-4' (Table 5). Although, differences among cultivars were observed at UCR 28 DAIT, there were no significant differences in methiozolin rate for TI₂₅ among CRBG cultivars at UCR 56 DAIT (Table 5). Similar to the species effect, there was a decrease in magnitude of the methiozolin rate for TI₂₅ among cultivars between 28 and 56 DAIT (Table 5). This decrease could be attributed to methiozolin accumulation in soil. These results are in agreement with those of McCullough and Gomez de Barreda (2012) where soil residual activity was documented with methiozolin applications. Furthermore, research indicates methiozolin is a slow acting herbicide requiring multiple applications to effectively control AB, suggesting a possible correlation between number of applications and injury to turfgrass (Brosnan et al. 2012; Han and Kaminski 2012; McCullough et al. 2012).

From these studies, CRBG appears to be more tolerant than COBG and VBG to one or two applications of methiozolin. For CRBG cultivars tested in this study, GR₅₀ and TI₂₅ occurred only at methiozolin rates above the recommended rate. At both AU and UCR, 'Penn A-4' exhibited the least growth regulation and injury in response to methiozolin application when significant effects among CRBG cultivars were apparent. Colonial bentgrass and VBG are unlikely to tolerate sequential applications of

methiozolin at a rate that is adequate to effectively control AB. Further research is needed to accurately describe the relative tolerances among creeping bentgrass cultivars in the field.

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Table 1. Bentgrass species and cultivars treated with methiozolin to assess tolerance at Auburn University and University of California, Riverside.

Bentgrass Species	Cultivar	Source
	Focus	Seed Research of Oregon, Corvallis, OR
	T-1	J. R. Simplot Co., Boise, ID
	Penn G-2	Tee 2 Green, Hubbard, OR
	Bengal	Barenbrug USA, Tangent, OR
Creeping Bentgrass	007	Seed Research of Oregon, Corvallis, OR
	Tyee	Seed Research of Oregon, Corvallis, OR
	Penn A-4	Tee 2 Green, Hubbard, OR
	96-2	Pickseed International, Ontario, Canada
	Penncross	Tee 2 Green, Hubbard, OR
Colonial Bentgrass	SR7150	Seed Research of Oregon, Corvallis, OR
Velvet Bentgrass	SR7200	Seed Research of Oregon, Corvallis, OR

Table 2. Rate of methiozolin (kg ai ha^{-1}) that caused a 50% growth reduction (GR_{50}) in creeping, colonial and velvet bentgrass at Auburn University and at University of California Riverside 28 and 56 days after initial treatment (DAIT). Methiozolin treatment was initiated 7 weeks after seeding (0 DAIT), with a second application 35 DAIT.

Location	Bentgrass species	Mean GR_{50}	
		28 DAIT	56 DAIT
		kg ai ha^{-1} methiozolin	
AU	Creeping	1.9 b [†]	1.1 a
	Colonial	0.4 d	0.6 bc
	Velvet	0.4 d	0.3 d
UCR	Creeping	3.2 a	0.8 b
	Colonial	0.9 c	0.4 cd
	Velvet	1.1 c	0.4 cd

[†] Means separated by LSD followed by the same letter in a column are not significantly different ($\alpha = 0.05$).

Table 3. Kilograms of methiozolin ha⁻¹ that caused 25% Turfgrass Injury (TI₂₅) in creeping, colonial and velvet bentgrass 28 and 56 days after initial treatment (DAIT). Methiozolin treatment was initiated 7 weeks after seeding (0 DAIT) with a second application 35 DAIT. Values were pooled over two locations; Auburn University (AU) and University of California Riverside (UCR).

Bentgrass species	TI ₂₅ AU plus UCR			
	28 DAIT		56 DAIT	
	kg ai ha ⁻¹ methiozolin			
Creeping	2.4	a [†]	1.1	a
Colonial	0.5	b	0.3	b
Velvet	0.5	b	0.2	b

[†] Means separated by LSD followed by the same letter in a column are not significantly different ($\alpha = 0.05$).

Table 4. Kilograms of methiozolin ha⁻¹ that caused a 50% growth reduction (GR₅₀) in 8 different creeping bentgrass cultivars at Auburn University and at University of California Riverside 56 days after initial treatment (DAIT). Methiozolin treatment was initiated 7 weeks after seeding (0 DAIT) with a second application 35 DAIT.

Location	Cultivar	GR ₅₀
		kg ai ha ⁻¹ methiozolin
AU	Focus	0.5 c [†]
	T-1	0.8 bc
	Penn G-2	1.0 bc
	Bengal	1.4 b
	007	0.5 c
	Tyee	0.7 c
	Penn A-4	2.6 a
	96-2	1.0 bc
	Penncross	0.9 bc
UCR	Focus	0.8 c
	T-1	0.7 c
	Penn G-2	0.8 bc
	Bengal	1.0 bc
	007	0.9 bc
	Tyee	0.8 bc
	Penn A-4	0.9 bc
	96-2	0.8 c
	Penncross	0.8 bc

[†]Means separated by LSD followed by the same letter in a column are not significantly different ($\alpha = 0.05$).

Table 5. Kilograms of methiozolin ha⁻¹ that caused 25% Turfgrass Injury (TI₂₅) in 8 different creeping bentgrass cultivars at Auburn University and at University of California Riverside 28 and 56 days after initial treatment (DAIT). Methiozolin treatment was initiated 7 weeks after seeding (0 DAIT) with a second application 35 DAIT.

Location	Cultivar	28 DAIT		56 DAIT	
		kg ai ha ⁻¹ methiozolin			
AU	Focus	2.4	cdef [†]	0.7	d
	T-1	2.2	cdef	0.9	d
	Penn G-2	1.8	efg	1.4	bc
	Bengal	1.9	efg	1.5	b
	007	2.3	cdef	0.6	d
	Tyee	2.0	defg	0.8	cd
	Penn A-4	2.8	cd	2.5	a
	96-2	2.1	defg	1.0	bcd
	Penncross	1.3	g	1.0	bcd
UCR	Focus	2.7	cde	1.0	bcd
	T-1	2.5	cdef	0.9	bcd
	Penn G-2	2.6	cdef	0.9	bcd
	Bengal	3.8	ab	1.1	bcd
	007	2.1	defg	1.0	bcd
	Tyee	2.0	defg	0.9	bcd
	Penn A-4	4.5	a	0.8	bcd
	96-2	3.1	bc	1.0	bcd
	Penncross	2.0	defg	1.1	bcd

[†]Means separated by LSD followed by the same letter in a column are not significantly different ($\alpha = 0.05$).

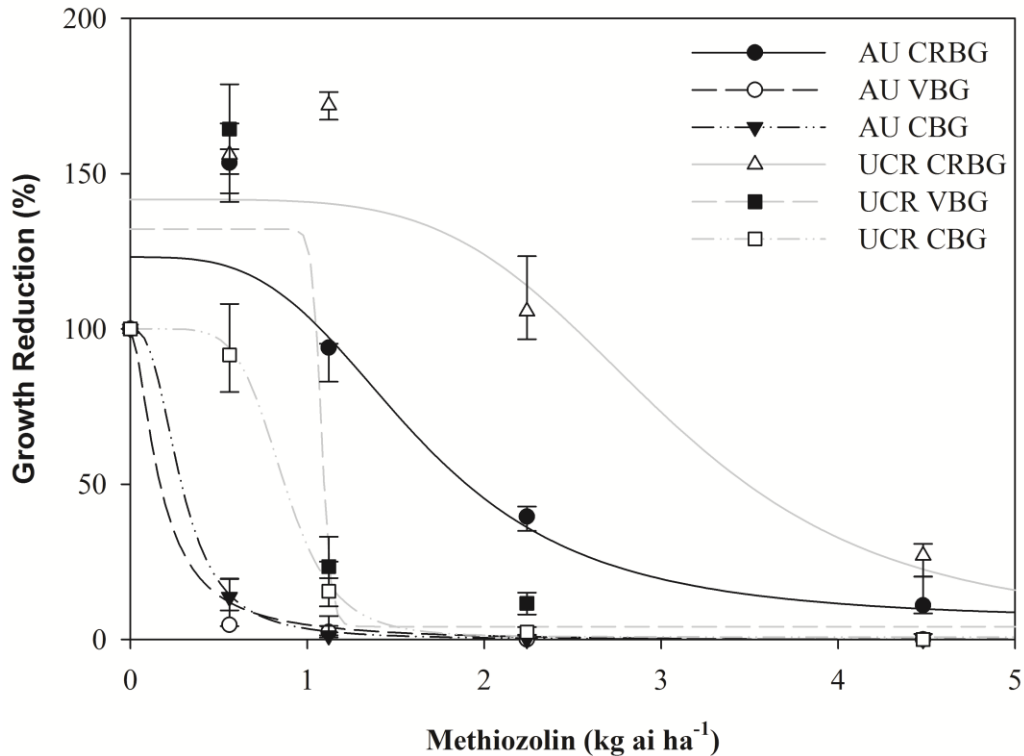


Figure 1. Percent growth reduction of creeping bentgrass (CRBG), velvet bentgrass (VBG) and colonial bentgrass (COBG) treated with methiozolin at 0, 0.6, 1.1, 2.2, 4.5, or 9.0 kg ai ha⁻¹ 7 weeks after seeding (0 days after initial treatment, DAIT). Data were collected 28 DAIT at Auburn University (AU) and University of California, Riverside (UCR). Data points represent an average of four replications. Error bars indicate Standard Errors. A four parameter logistic regression model of $y = y_0 + a/(1+(x/x_0)^b)$ was used to fit data. Regression parameters and coefficients of determination at AU were CRBG $a = 117.1$, $b = 3.4$, $x_0 = 1.6$, $y_0 = 6.1$, $R^2 = 0.88$; COBG $a = 99.7$, $b = 2.7$, $x_0 = 0.3$, $y_0 = 0.0$, $R^2 = 0.99$; VBG $a = 100.6$, $b = 1.6$, $x_0 = 0.2$, $y_0 = -0.6$, $R^2 = 0.98$. Regression parameters and coefficients of determination at UCR were CRBG $a = 137.4$, $b = 4.68$, $x_0 = 3.0$, $y_0 = 4.3$, $R^2 = 0.83$; COBG $a = 99.2$, $b = 6.2$, $x_0 = -0.9$, $y_0 = 0.8$, $R^2 = 0.94$; VBG $a = 128.0$, $b = 41.7$, $x_0 = 1.1$, $y_0 = 4.2$, $R^2 = 0.87$.

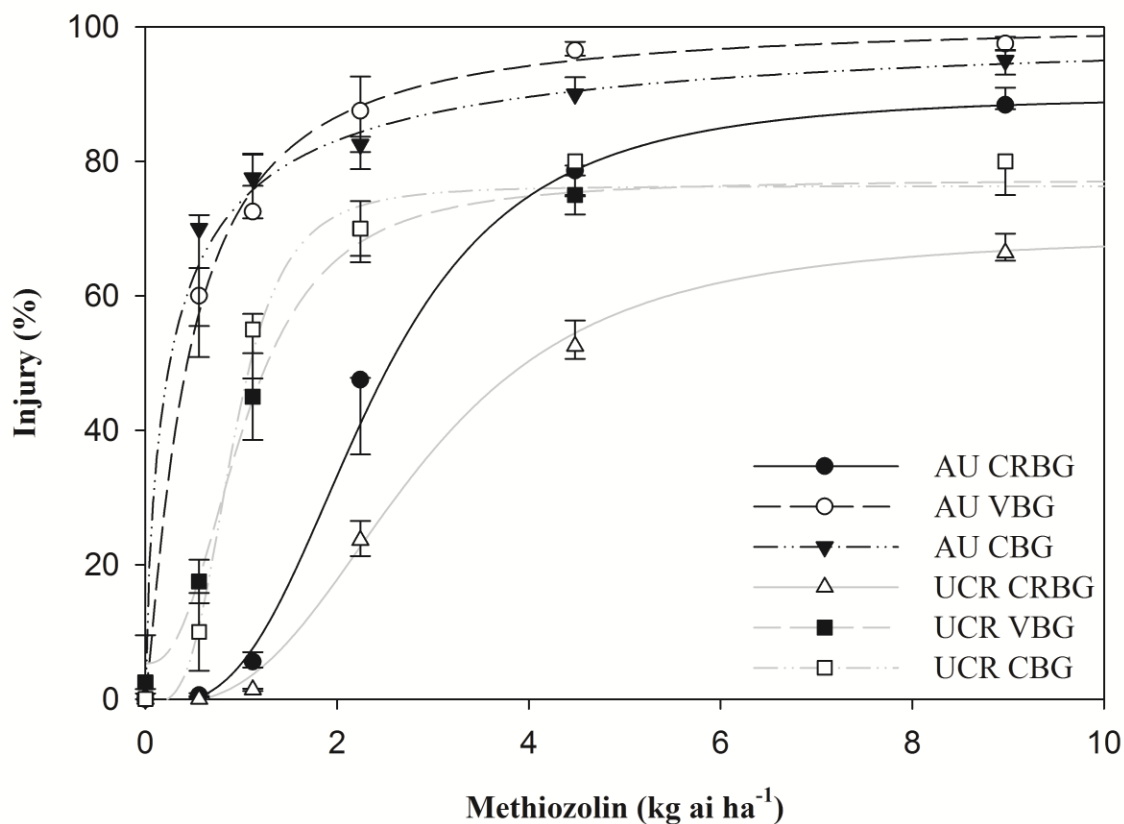


Figure 2. Percent injury of creeping bentgrass (CRBG), velvet bentgrass (VBG) and colonial bentgrass (COBG) treated with methiozolin at 0, 0.6, 1.1, 2.2, 4.5, or 9.0 kg ai ha⁻¹ 7 weeks after seeding (0 days after initial treatment, DAIT). Data were collected at 28 DAIT at Auburn University (AU) and University of California, Riverside. Data points represent an average of four replications. Error bars indicate Standard Errors. A four parameter logistic regression model of $y = y_0 + a / (1 + (x/x_0)^b)$ was used to fit data. Regression parameters and coefficients of determination at AU were CRBG $a = 90.7$, $b = -3.1$, $x_0 = 2.4$, $y_0 = -0.8$, $R^2 = 0.98$; COBG $a = 101.2$, $b = -0.7$, $x_0 = 0.3$, $y_0 = 0.0$, $R^2 = 0.96$; VBG $a = 101.8$, $b = -1.2$, $x_0 = 0.4$, $y_0 = 0.0$, $R^2 = 0.96$. Regression parameters and coefficients of determination at UCR were CRBG $a = 69.8$, $b = -2.9$, $x_0 = 2.8$, $y_0 = -0.3$, $R^2 = 0.98$; COBG $a = 76.9$, $b = -3.6$, $x_0 = -0.9$, $y_0 = -0.5$, $R^2 = 0.96$; VBG $a = 71.94$, $b = -2.5$, $x_0 = -1.0$, $y_0 = 5.3$, $R^2 = 0.94$.

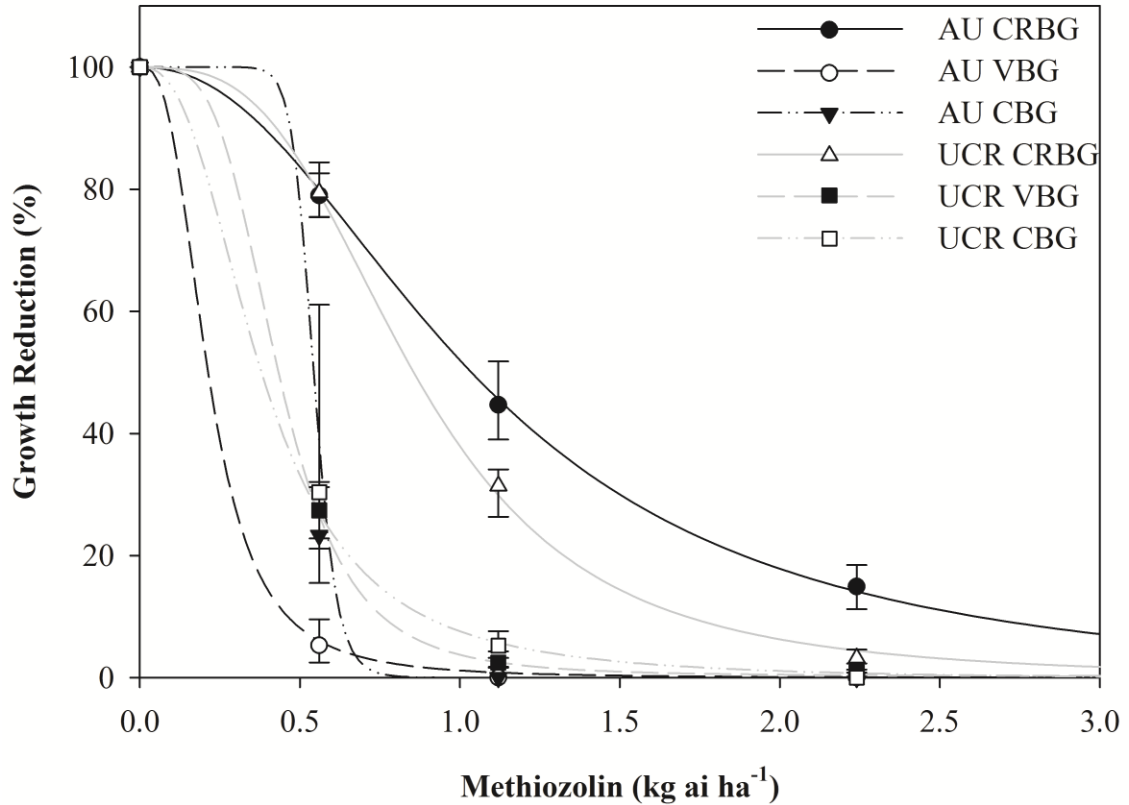


Figure 3. Percent growth reduction of creeping bentgrass (CRBG), velvet bentgrass (VBG) and colonial bentgrass (COBG) treated with either methiozolin at 0, 0.6, 1.1, 2.2, 4.5, or 9.0 kg ai ha⁻¹ 7 weeks after seeding (0 days after initial treatment, DAIT), with a second application at 35 DAIT. Data were collected 56 DAIT at Auburn University (AU) and University of California, Riverside (UCR). Data points represent an average of four replications. Error bars indicate Standard Errors. A four parameter logistic regression model of $y = y_0 + a/(1+(x/x_0)^b)$ was used to fit data. Regression parameters and coefficients of determination at AU were CRBG $a = 101.9$, $b = 2.2$, $x_0 = -1.1$, $y_0 = -1.9$, $R^2 = 0.97$; COBG $a = 100.0$, $b = 15.3$, $x_0 = 0.5$, $y_0 = 0.0$, $R^2 = 0.84$; VBG $a = 100.0$, $b = 2.8$, $x_0 = -0.2$, $y_0 = 0.0$, $R^2 = 0.99$. Regression parameters and coefficients of determination at UCR were CRBG $a = 100.0$, $b = 3.2$, $x_0 = 0.9$, $y_0 = -0.1$, $R^2 = 0.99$; COBG $a = 100.3$, $b = 2.5$, $x_0 = -0.4$, $y_0 = -0.3$, $R^2 = 0.99$; VBG $a = 99.7$, $b = 4.0$, $x_0 = -0.4$, $y_0 = 0.3$, $R^2 = 0.99$.

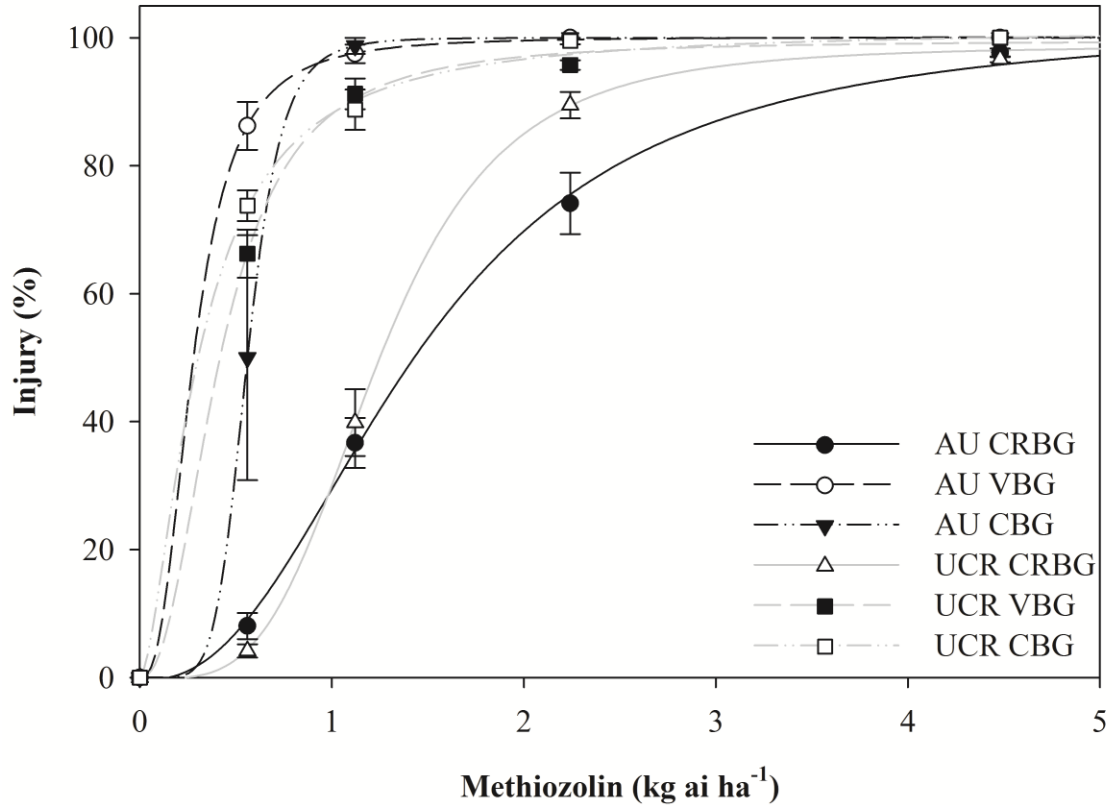


Figure 4. Percent injury of creeping bentgrass (CRBG), velvet bentgrass (VBG) and colonial bentgrass (COBG) treated with methiozolin at 0, 0.6, 1.1, 2.2, 4.5, or 9.0 kg ai ha⁻¹ 7 weeks after seeding (0 days after initial treatment, DAIT), with a second application at 35 DAIT. Data were collected 56 DAIT at Auburn University (AU) and University of California, Riverside. Data points represent an average of four replications. Error bars indicate Standard Errors. A four parameter logistic regression model of $y = y_0 + a/(1+(x/x_0)^b)$ was used to fit data. Regression parameters and coefficients of determination at AU were CRBG $a = 102.7$, $b = -2.34$, $x_0 = 1.4$, $y_0 = -0.5$, $R^2 = 0.99$; COBG $a = 100.0$, $b = -6.3$, $x_0 = 0.6$, $y_0 = 0.0$, $R^2 = 0.89$; VBG $a = 100.1$, $b = -2.6$, $x_0 = 0.3$, $y_0 = 0.0$, $R^2 = 0.99$. Regression parameters and coefficients of determination at UCR were CRBG $a = 98.9$, $b = -3.8$, $x_0 = 1.2$, $y_0 = -0.2$, $R^2 = 0.99$; COBG $a = 100.4$, $b = -1.6$, $x_0 = -0.3$, $y_0 = 0.0$, $R^2 = 0.99$; VBG $a = 99.6$, $b = -2.3$, $x_0 = 0.4$, $y_0 = 0.0$, $R^2 = 0.99$.