Chapter Two: Development and Validation of a Growing Degree Day Model for Trinexapac-ethyl Applications to Creeping Bentgrass Golf Course Putting Greens

ABSTRACT

The plant growth regulator trinexapac-ethyl (TE) is widely used in the turfgrass industry. However, loss of efficacy on cool-season turfgrasses has been observed during summer, and linked to more rapid metabolism by the plant. The purpose of this study was to determine if a growing degree day (GDD) model could be used to identify the optimum TE re-application interval on creeping bentgrass putting greens. This objective was accomplished in two stages 1) model development and 2) model validation. Model development was conducted on a creeping bentgrass (*Agrostis stolonifera* Hud.) golf putting green in Madison, WI during the summer of 2008. The treatments consisted five TE re-application intervals (100, 200, 400, 800 GDD, and four week) and a non-treated control. The GDD treatments were calculated in degrees Celsius with a base temperature of 0°C. Trinexapac-ethyl was applied at the rate of 0.05 kg a.i. ha^{-1}. Turfgrass clippings were collected daily, and turfgrass visual quality and color index were recorded weekly. The 100 and 200 GDD re-application intervals provided a consistent 10 to 20% reduction in clipping yield compared to the control plots throughout the summer. Other re-application intervals had mean clipping yield increases of 15% approximately 300 GDD after TE application. Model validation occurred on a different creeping bentgrass putting green during 2009 and 2010. The experiment was a completely randomized 3x2 factorial design with three TE application rates (0.00, 0.05, and 0.10 kg a.i. ha^{-1}) and two re-application frequencies (200 GDD and four week). The 200 GDD treatment consistently suppressed clipping yield during both years. Both clipping yield suppression and enhancement occurred with the four week re-application intervals. Application rate had no effect on the magnitude or duration of growth suppression. Season-long color index and visual quality enhancements occurred with the 200

GDD re-application interval. Re-applying TE every 200 GDD units provide more consistent growth regulation on creeping bentgrass putting greens than a calendar-based re-application schedule.

INTRODUCTION

The plant growth regulator trinexapac-ethyl (TE) is widely used to suppress clipping yield on all commonly managed turfgrass species (Table 2.1). TE suppresses clipping yields by disrupting the conversion of gibberellic acid$_{20}$ (GA) to GA_1, the bio-active gibberellin in cool season turfgrasses (Reid and Ross, 1991; Rademacher, 2000). As a result, cell elongation decreases while GA_{20} and total nonstructural carbohydrates (TNC) increase in concentration (Han et al., 1998, 2004; Tan and Qian, 2003). These alterations reduce leaf length and increase mesophyll cell density, chlorophyll concentration, tiller density and leaf area resulting in turfgrass color and visual quality enhancement (Ervin and Koski, 1998; Ervin and Koski, 2001; Stier and Rodgers, 2001; Bunnell et al., 2005; Beasley et al., 2007). Once TE is metabolized within the plant, increased GA_{20} and TNC concentrations result in a period of enhanced relative growth rate. Fagerness and Yelverton (2000) first described this period of yield enhancement following yield suppression as 'post-inhibition growth enhancement.' This effect will be hereafter called the rebound phase of growth regulation. The rebound phase has since been observed in other grass species including Kentucky bluegrass (*Poa pratensis* L.) and creeping bentgrass (*Agrostis stolonifera* Hud.) (Beasley and Branham, 2005).

A majority of the turfgrass species summarized in Table 2.1 had 50% relative yield suppression for a period of four to six weeks following TE application. One notable exception however, is creeping bentgrass golf putting greens. McCullough et al. (2006b) showed that TE-treated creeping bentgrass managed as a putting green in South Carolina had a 20% decrease in relative clipping yield which lasted for two weeks at the labeled application rate of 0.05 kg a.i. ha^{-1} during summer.

Table 2.1. The influence of TE application rate and re-application frequency on magnitude and duration of growth suppression in various turfgrass species.

Turfgrass Species and Mowing Height	Application Rate	Re-application Frequency	Growth Suppression	Approximate Duration of Growth Suppression	Investigator
Common name; mm	kg a.i. ha^{-1}	Weeks	% of control	Weeks	
Creeping bentgrass; 3.2	0.05	4	20%	2	McCullough et al., 2006b
Creeping bentgrass; 3.2	0.02, 0.03, 0.05	1, 2, 3	20-40%	3	McCullough et al., 2007
Kentucky bluegrass; 30	0.05	4-6	20%	4-6	Stier and Rodgers, 2001
Kentucky bluegrass, 35	0.05	4	50%	4	Tan and Qian, 2003
Kentucky bluegrass; 32	0.14, 0.29, 0.58	none	44-73%	4-5†	Beasley et al., 2007
Rough bluegrass; 80	0.29	6	55-80%	6	Gardner and Wherley, 2005
Sheep fescue; 80	0.29	6	35-50%	6	Gardner and Wherley, 2005
St. Augustinegrass; 75	0.14, 0.29	2, 4	50%	4	McCarty et al., 2004
Supina bluegrass; 30	0.05	4-6	60%	4-6	Stier and Rodgers, 2001
Tall fescue; 38	0.29	none	44-77%	4	Richie et al., 2001
Tall fescue; 80	0.29	6	58-76%	6	Gardner and Wherley, 2005
'TifEagle' Bermudagrass; 3.2	0.05	4	60%	3	McCullough et al., 2007
'Tifway' Bermudagrass; 16	0.07, 0.11	4	60%	4	Fagerness and Yelverton, 2000
'Tifway' Bermudagrass; 25	0.11	4	50%	4	Fagerness et al., 2004
Zoysiagrass; 12	0.05, 0.10, 0.19	4, 8, 12	25, 27, 0%	4-6	Qian and Engelke, 1999

† Duration dependent on summer or fall season

McCullough et al. (2007) investigated the effect of TE application interval on creeping bentgrass yield during the spring in South Carolina. In that study, total TE application rate was constant for all treatments. They found that TE reduced relative clipping yield by 20-40% and more frequent applications provided more uniform yield suppression. However, the magnitude and duration of yield suppression on creeping bentgrass putting greens was still low relative to other turfgrass species.

Lickfeldt et al. (2001) and Beasley et al. (2007) found TE efficacy decreased as air temperatures increased during summer. A similar effect was observed in hybrid bermudagrass during fall (Fagerness et al., 2002). As the daily average air temperature decreased, the duration and magnitude of the suppression period increased. In 2005, Beasley and Branham showed that TE half life was directly related to air temperature and turfgrass species. The TE half lives for creeping bentgrass and Kentucky bluegrass were 6.4 and 5.3 days at 18°C and 3.1 and 3.4 days at 30°C, respectively. Additionally, Fagerness and Penner (1998) reported that TE is not subject to rapid UV degradation and it further supports plant metabolism as the primary pathway of TE degradation.

A logical step forward was to develop a model that uses air temperature to predict when TE should be re-applied. Growing degree day (GDD) systems are useful models that use air temperature as a means of estimating plant growth and development (McMaster and Wilhelm, 1997; Ritchie and NeSmith, 1991). In a typical GDD model, daily average air temperature is recorded, subtracted from a base temperature where metabolism is minimal, and added with previous day's temperature to calculate cumulative GDD (Ritchie and NeSmith, 1991). Cumulative GDD is then correlated with plant observations and used to predict plant development. It is likely that such a model could be developed to predict relative clipping yield following TE and aid in scheduling of TE re-applications.

The objective of this study was to develop and validate a GDD model as a means to estimate TE metabolism in creeping bentgrass putting greens. The hypothesis of this study is that a GDD model can accurately predict the rate of TE metabolism and indicate proper TE re-application intervals that maintain season-long yield suppression. Development of a functional GDD model would result in accurate prediction of the magnitude of growth regulation and indicate when TE re-applications are required to maintain the growth suppression phase. These objectives were accomplished via two experiments over three growing seasons. The GDD model was first developed through calibration during Experiment #1 and then validated during the following two seasons on a different creeping bentgrass putting green in Experiment #2.

METHODS

Experiment 1: Model Calibration

Site Description

A field experiment was conducted on a creeping bentgrass (‘L93’) putting green at the O.J. Noer Turfgrass Research and Education Facility in Madison, WI during 2008. The green was constructed in 2002 to USGA specifications (USGA Green Section Staff, 1993) with 20% peat-amended sand (by volume). Overhead irrigation supplemented precipitation to 80% of estimated potential evapotranspiration. The plot was fertilized weekly with 5 kg N ha^{-1} as liquid urea (46-0-0). Approximately 10 mm of irrigation was applied to the plot immediately after fertilizer application. Mehlich-3 soil testing indicated supplemental phosphorus and potassium were not required (Mehlich, 1984). Chlorothalonil was applied weekly to control disease; demethylation inhibiting pesticides were avoided because of their growth regulatory properties. The plots were topdressed with sand monthly with approximately 1000 kg ha^{-1}.

Experimental Design

Plots measured 1.8 x 0.9 m. Treatments consisted of six TE re-application intervals arranged in a randomized complete block design with four replicates. Re-application intervals were 100, 200, 400, 800 GDD, four week, and non-treated control. The GDD model was based on a summation of the mean daily air temperature (°C) with a base of 0°C. Mean daily average air temperature was measured with an on-site weather station. After the accumulated GDD units had been surpassed, TE was re-applied to that treatment. Trinexapac-ethyl was applied at 0.05 kg a.i. ha^{-1} with a CO_2 powered backpack sprayer with TeeJet XR 11004 nozzles calibrated to deliver 810 L ha^{-1} at 276 kPa. TE applications began on 22 June and continued to 19 August 2008 (Table 2.2).

Table 2.2. Trinexapac-ethyl (TE) application dates in 2008 for all treatments in Experiment #1. Application rate was 0.05 kg a.i. ha^{-1} for all treatments.

TE Re-application Interval	TE Re-application Date	Total TE Applied
		kg a.i. ha^{-1}
100 GDD†	6/22, 6/27, 7/2, 7/13, 7/19, 7/24, 7/29, 7/31, 8/6, 8/12, 8/17	0.55
200 GDD	6/22, 7/2, 7/13, 7/22, 8/1, 8/12	0.30
400 GDD	6/22, 7/12, 7/30, 8/19	0.20
800 GDD	6/22, 7/30	0.10
Four Week	6/22, 7/17, 8/14	0.15
Non-Treated Control	none	0.00

† Growing degree day (GDD) was the summation of mean daily air temperature (base °C) after TE application. After the GDD threshold was surpassed, TE was re-applied and the model was reset to zero.

Data Collection

Clippings were collected five times per week from 25 June until 21 Aug 2008, weather permitting, by mowing one 1.3 m pass down the center of each plot 24 h (1200 ± 2 h) after the previous mowing. Prior to clipping collection, 27 cm buffer alleys were mowed at the top and bottom of each plot to reduce variation caused by starting and stopping the mower. Clippings were then brushed from the mower collection bucket into a paper bags. Sand debris was removed from clipping samples by agitating and decanting the clippings with water from a 600 mL glass beaker into a towel of known mass. The sample and towel were then placed into a drying oven set to 60°C for 24 hours. Samples were weighed, the towel mass subtracted, and the sample mass from treatments receiving TE were divided by the mass of the non-treated plot within the appropriate block to obtain relative clipping yield.

Turfgrass visual quality and color index were measured weekly. Turfgrass visual quality was rated on a 1 to 9 scale with 1 representing completely dead, 6 minimally acceptable, and 9 perfect putting green quality. Turfgrass color was quantified with the mean of 10 color index measurements from a CM-1000 reflectometer (Spectrum Technologies, Inc, Plainfield, IL) on a 0-999 scale, where higher numbers indicate less 700 nm light reflectance from the turf canopy and therefore greater color. The CM-1000 was held one meter from the turfgrass surface with the incidental light meter pointed in the direction of the sun. Measurements occurred on sunny days between 1100 and 1500 h. Relative CI and visual quality were calculated the same way as relative yield.

Statistical Analysis

The daily mean relative clipping yield from all five TE treatments were plotted as a function of cumulative GDD after TE re-application to estimate stages of growth regulation and identify the GDD threshold that maintained consistent growth suppression. Fourth order sine regression was used to estimate the GDD point where the growth suppression phase transitioned to the rebound phase with SAS statistical software using the regression procedure from treatments that exhibited both the suppression and rebound growth phases (SAS Institute, Cary, NC). Base temperature was determined by fitting fourth order sine regression to relative yield with GDD re-calculated with base temperatures ranging from 0-12°C by 2°C. The most appropriate base temperature, as determined by R^2 value, was selected for model development. The area under the growth curve (AUGC) was calculated from the actual clipping yield for each treatment. Treatment AUGCs were compared with Fisher's protected LSD at α=0.05 separation to estimate season long yield in SAS. Visual turfgrass quality and CI differences were quantified using repeated measures analysis and Fisher's protected LSD at α=0.05 means separation with JMP (version 8.0.2, SAS Institute, Cary, NC).

Experiment 2: Model Validation

Site Description

A field experiment was conducted on a creeping bentgrass ('Penncross') at the O.J. Noer Turfgrass Research and Education Facility in Madison, WI during 2009 and 2010. The green was constructed to USGA specifications in 2005 with sand amended with 20% peat by volume (USGA Green Section Staff, 1993). All cultural practices, including mowing, irrigation, and fertilization were identical to those described in Experiment #1.

Experimental Design

Plots measured 3.6 x 0.9 m. Treatments consisted of a 3 x 2 factorial of TE application rate (0.00, 0.05, 0.10 kg a.i. ha^{-1}) and TE re-applications interval (200 GDD and four week). Plots were arranged in a completely randomized design with four replicates. GDD was calculated as stated in Experiment #1 where TE was re-applied every 200 GDD for those treatments. TE was applied with a CO_2 powered backpack sprayer equipped with TeeJet AI 11004 nozzles calibrated to deliver 810 L ha^{-1} at 276 kPa. Applications began on 29 April through 14 October 2009 and resumed on 1 April 2010 until 11 July 2010 (Table 2.3).

Data Collection

Clippings were collected three times per week from 7 May until 19 Oct 2009 and 22 April 2010 until 9 July 2010; weather permitting following the same methods described in Experiment #1 at within one hour of 12 h. Visual quality and color index were measured bi-weekly with the methods described in Experiment #1.

Statistical Analysis

Relative clipping yields from the four week re-application interval treatments were fitted to the predicted values calculated by the model from the 400, 800, and four week interval treatments in Experiment #1 using the nonlinear procedure in SAS (SAS Institute, Cary, NC) to calculate sum of squares. The sum of squares was then divided by the least squares that resulted from fourth order sine regression of the four week treatments in Experiment #2 to calculate an F statistic and p-value. Relative clipping yields from treatments only exhibiting growth suppression (200 GDD interval) were compared to values calculated in the 100 and 200 GDD treatment of Experiment #1 using the GLM procedure with Fisher's protected LSD in SAS (SAS Institute, Cary, NC). Quality and CI differences were found using repeated measures analysis and Fisher's protected LSD at $\alpha=0.05$ means separation with JMP 8 (version 8.0.2, SAS Institute, Cary, NC).

Table 2.3. Trinexapac-ethyl (TE) application dates in 2009-2010 for all treatments in Experiment #2. Application rates were 0.00, 0.05, and 0.10 kg a.i. ha^{-1} for each re-application interval.

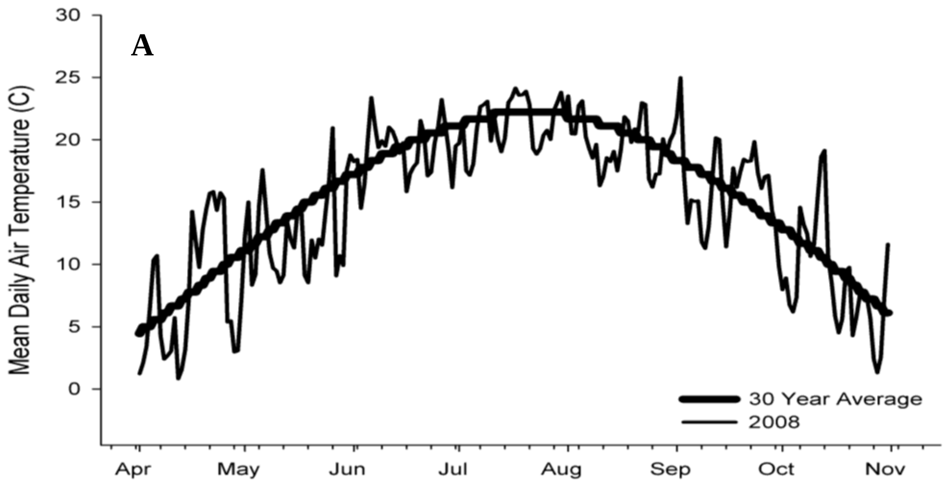
Re-application Interval	Year	TE Re-application Date
200 GDD†	2009	5/5, 5/21, 6/4, 6/16, 6/24, 7/6, 7/15, 7/27, 8/10, 8/22, 9/8, 9/22, 10/14
	2010	4/19, 5/5, 5/22, 6/2, 6/12, 6/23, 7/3, 7/12
Four Week	2009	5/5, 6/20, 7/15, 8/14, 9/11
	2010	4/19, 5/18, 5/22, 6/25

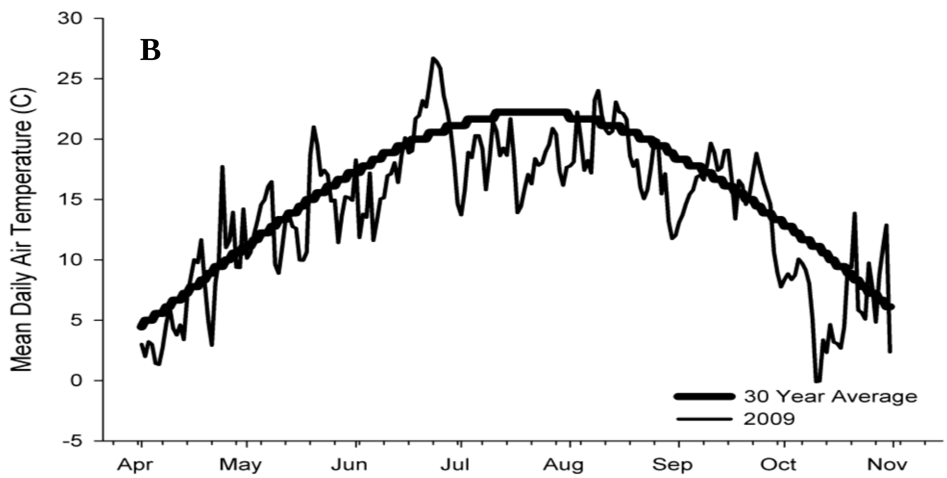
† Growing degree day (GDD) was the summation of mean daily air temperature (base °C) after TE application. After the GDD threshold was surpassed, TE was re-applied and the model was reset to zero.

RESULTS AND DISCUSSION

Weather Data

Mean daily air temperatures ranged from -0.1 to 26.7°C during the times of data collection from 2008 until 2010 (Figure 2.1). The 2008 and 2009 growing seasons had below average temperatures, 1.0 and 2.7°C day^{-1} below normal, respectively. The summer of 2009, with exception of late June, was particularly below average. In 2010, mean daily air temperatures averaged 2.4°C day^{-1} above normal. The above average temperatures in spring 2010 allowed for earlier TE application than in 2008 and 2009.





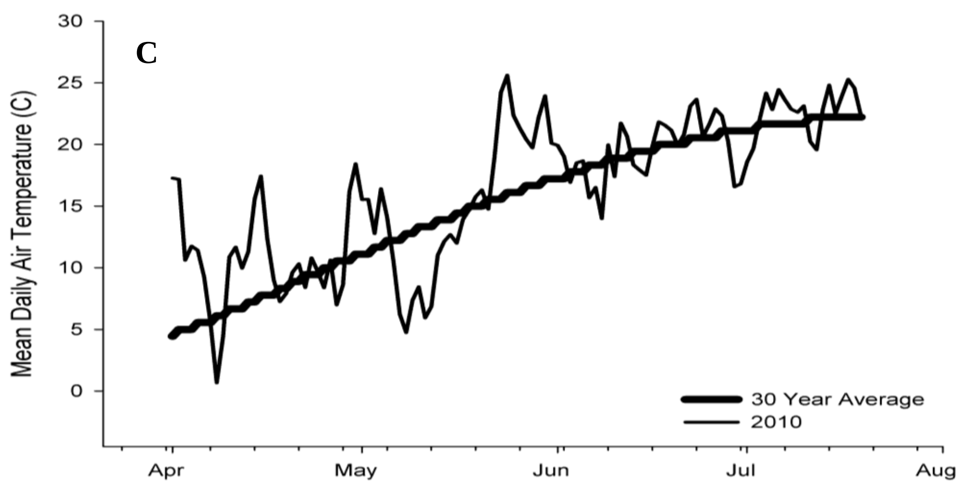


Figure 2.1. Mean daily air temperature in Madison, WI during the 2008 (A), 2009 (B), and 2010 (C) with respect to 30 year average temperature (°C). 2008 was close to the daily average air temperature while 2009 and 2010 were below and above average, respectively.

Experiment 1: Model Calibration

Clipping Yield

The treatments resulted in a wide range of relative clipping yield responses. Clipping yield suppression and rebound occurred when TE re-applications occurred every 400 GDD, 800 GDD, and every four weeks. Therefore, the relative clipping yields from those treatments were pooled together and subjected to regression analysis with relative yield a function of cumulative GDD following the most recent TE application (Figure 2.2). This analysis produced a model that predicted the affect of TE on the relative clipping yield of creeping bentgrass putting green turfgrass following TE application. Four parameter sine regression proved to be the most appropriate model; $y = 1.0090 + 0.1597 * \sin(2\pi * GDD / 805.1719 – 2.5712)$, domain of 0 to 740 GDD, $R^2 = 0.519$, and p-value <0.001 (Figure 2.1). This model is appropriate because it describes how TE application causes relative yield suppression followed by rebound before the turfgrass returns to a clipping yield similar to the non-treated turfgrass. The domain is limited to 740 GDD because the slope of the function continues to decrease after 740 GDD and has no experimental or theoretical basis. Base temperature analysis indicated that the 0°C had the greatest R^2 value compared to models where GDD was re-calculated with base temperatures ranging from 2 to 12°C (Figure 2.3).

Mean maximum growth suppression occurred 128 GDD units after TE application and reduced clipping yield by 15.1%. The transition from the suppression phase into the rebound phase occurred at 323 GDD before the maximum rebound occurred at 531 GDD with a 16.9% increase in relative yield. The effect of TE on clipping yield appeared to dissipate 700 to 800 GDD following TE applications (Figure 2.2).

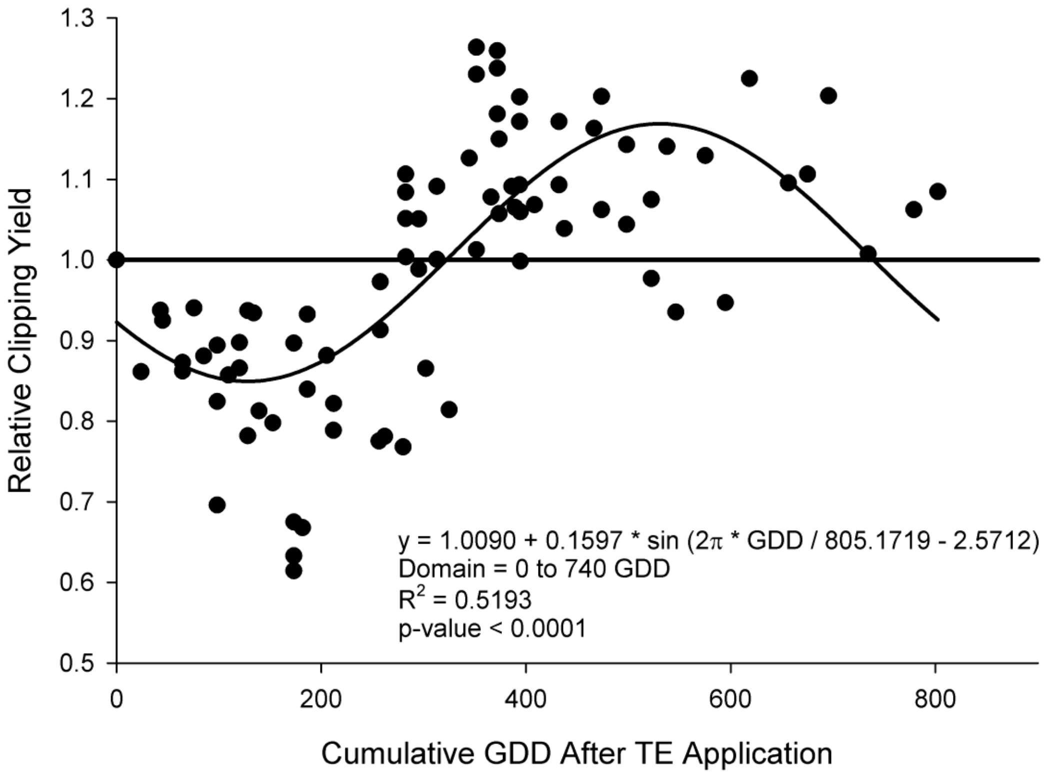


Figure 2.2. The relative clipping yields of the 400 GDD, 800 GDD, and four week trinexapac-ethyl (TE) re-application intervals were pooled together and plotted versus cumulative GDD after previous TE application. Fourth order sine regression analysis was highly significant and was used to develop a TE relative yield response model. Values less than one signifies yield suppression while values greater than one signifies yield enhancement in comparison to the control. Cumulative GDD was calculated as the summation of the mean daily air temperature base 0°C. The GDD was reset to zero following TE application.

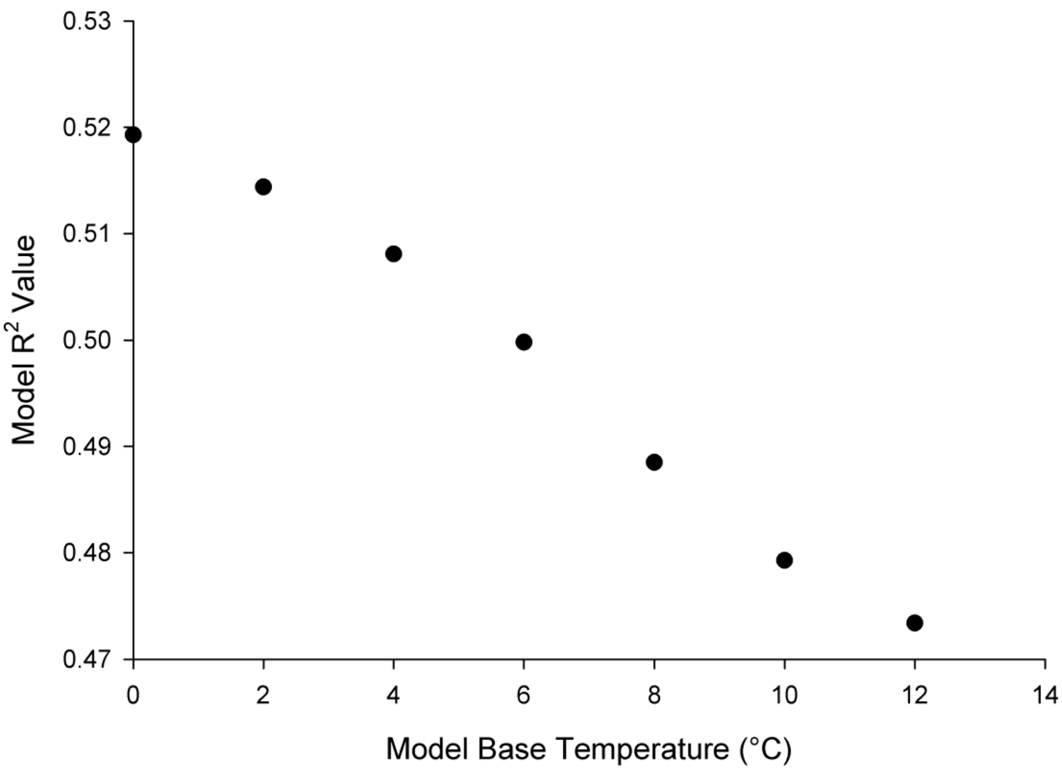


Figure 2.3. The selection of the most appropriate GDD model base temperature was determined by recalculating cumulative GDD in figure 2.1 from 0 to 12°C. The R^2 values for each base temperature were determined following fourth order sine regression. The 0°C had the highest R^2 value and was used as the base temperature.

The 100 and 200 GDD re-application treatments were not included in this model because TE was re-applied prior to 320 GDD transition point and therefore did not exhibit similar yield effects as the four week, 400 GDD, and 800 GDD treatments. Analysis of covariance indicated that the relative clipping yield of the 100 GDD and 200 GDD were not affected by GDD after application (Table 2.4), however, the intercept was highly significant. The lack of a GDD effect indicates that the level of growth suppression was constant during the summer. Re-application interval had a strong effect on relative clipping yield. The relative yield for the 100 GDD treatment was statistically different than the 200 GDD treatment while both were statistically less than the control; 20% and 12% of the non-treated control, respectively

The maximum relative yield of the 200 GDD re-application interval was similar to that of the pooled 400 GDD, 800 GDD, and four week model. This indicates that the mean maximum yield suppression provided by the 0.05 kg a.i. ha^{-1} application rate was 10 to 15% of the non-treated turfgrass. Increased growth suppression observed in the 100 GDD treatment may provide evidence that TE is being re-applied faster than it is being metabolized which resulted in a gradual accumulation of TE in the plant and therefore greater growth suppression. The regression model (Figure 2.2) provides evidence that 200 GDD is the longest possible re-application interval that will maintain yield suppression.

Area under the growth curve calculation indicated that the 100 GDD and 200 GDD TE re-application treatments had statistically lower yield compared to the non-treated control over the study period (Table 2.5). All other TE application intervals, including the labeled four week interval, had similar yields as the control. This is because the rebound phase cancelled out yield suppression for those re-application intervals. More frequent TE applications in the 100 GDD and 200 GDD treatments prevented the development of the rebound phase which resulted in a net yield decrease.

Table 2.4. The ANOVA table for the relative clipping yield of the 100 GDD, 200 GDD, and non-treated control treatments. Slope (GDD) was not significant for both the 100 GDD and 200 GDD treatments. However the intercept (re-application interval) was highly significant and resulted in maintained relative yield suppression regardless of GDD.

Source	df	p-value
Re-application Interval (I)	4	<0.0001
Cumulative GDD (GDD)	1	0.8124
I x GDD	4	0.8661

Table 2.5. The area under the growth curve calculation of net annual clipping yield across all trinexapac-ethyl re-application treatments at the 0.05 kg a.i. ha^{-1}application rate. The 100 GDD and 200 GDD were the only treatments that had reduced net yield compared to the non-treated control.

Re-application Interval	Area Under Growth Curve	
100 GDD	53.54	A
200 GDD	59.41	AB
400 GDD	65.23	BC
800 GDD	65.16	BC
Four Week	68.84	C
Non-treated Control	67.06	C

† GDD is the summation of daily average air temperature in degrees Celsius. TE is re-applied one each GDD threshold is surpassed. GDD is reset to zero after TE is re-applied.

‡ Column means followed by different letters are statistically different according to Fishers LSD (α=0.05).

Color and Quality

TE applications statistically enhanced turfgrass visual quality and color compared to the non-treated control for all re-application intervals except for the 800 GDD interval (Table 2.6). Visual quality and color enhancements are the likely result of the numerous physiological and morphological changes that have been previously well documented (Ervin and Koski, 1998; Stier and Rodgers, 2001; Bunnell et al., 2005; Beasley and Branham, 2007). The 100 GDD re-application interval had the highest visual quality and color index of any re-application treatments (Table 2.7). This may indicate that TE is being re-applied faster than it is being metabolize because Stier et al. (1999) reported that greater color enhancements resulted with increased PGR application rate. The 200 GDD, 400 GDD, and four week intervals all had similar quality and color enhancements. The quality enhancements progressed more rapidly for the 100 GDD treatments compared to the other TE treatments (Figure 2.4).

Date strongly affected both visual turfgrass quality and color index (Figures 2.4 and 2.5). Differences were not statistically different from the control until six to seven weeks after the initial TE applications. This is consistent with findings reported by McCullough et al. (2006b). After this time, enhancements increased in magnitude for remainder of the season for all treatments except for the 800 GDD treatment.

Table 2.6. The ANOVA table for the effect of trinexapac-ethyl re-application interval, date, block, and re-application interval by date interaction on turfgrass visual quality and color.

Source	df	Turfgrass Visual Quality				Color			
		Actual		Relative		Actual		Relative	
Re-application Interval (I)	5	0.0048	**	0.0043	**	0.0043	**	0.0040	**
Date (D)	8	<0.0001	***	<0.0001	*	<0.0001	***	<0.0001	***
I x D	40	0.1768		0.0011	**	0.0004	***	0.6623	
Block	3	0.0921		0.0214	*	0.8095		0.3286	

Table 2.7. The effect of trinexapac-ethyl re-application interval on both actual and relative visual quality and color.

Re-application Interval	Turfgrass Visual Quality				Color Index			
	Actual		Relative		Actual		Relative	
	1 to 9 Scale		% of Control		0-999§		% of Control	
100 GDD	8.2	A	111	A	317	A	111	A
200 GDD	7.8	B	105	B	300	B	106	B
400 GDD	7.8	B	105	B	300	B	106	B
800 GDD	7.6	BC	101	BC	291	BC	103	BC
Four Week	7.8	B	105	B	301	B	106	B
Non-treated Control	7.4	C	100	C	286	C	100	C

† GDD is the summation of daily average air temperature in degrees Celsius. TE is re-applied one each GDD threshold is surpassed. GDD is reset to zero after TE is re-applied.

‡ Column means followed by different letters are statistically different according to Fishers LSD (α=0.05).

§ Measure of 700 nm light absorbed by the turfgrass foliage. Higher color index represents more absorbance and darker green leaf color.

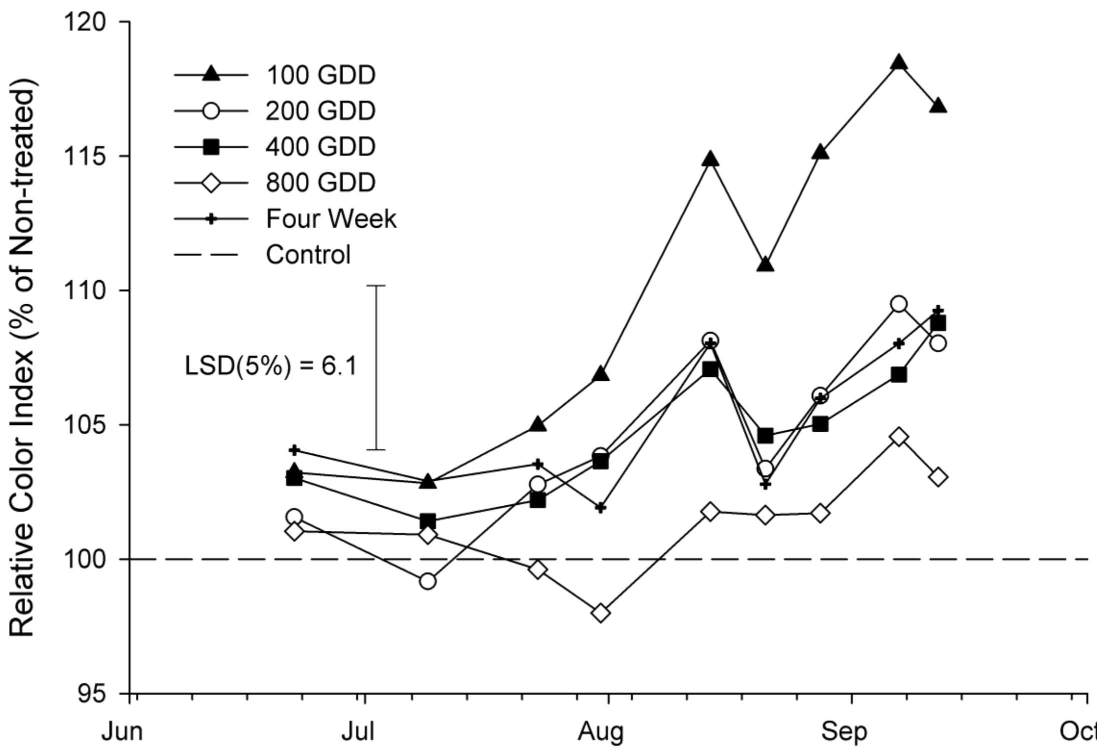


Figure 2.4. The effect of trinexapac-ethyl re-application interval on relative color index in 2008. The dashed line represents the color index non-treated control. Growing degree day (GDD) is the summation of daily average air temperature in degrees Celsius. TE is re-applied once each GDD threshold is surpassed. GDD is reset to zero after TE is re-applied.

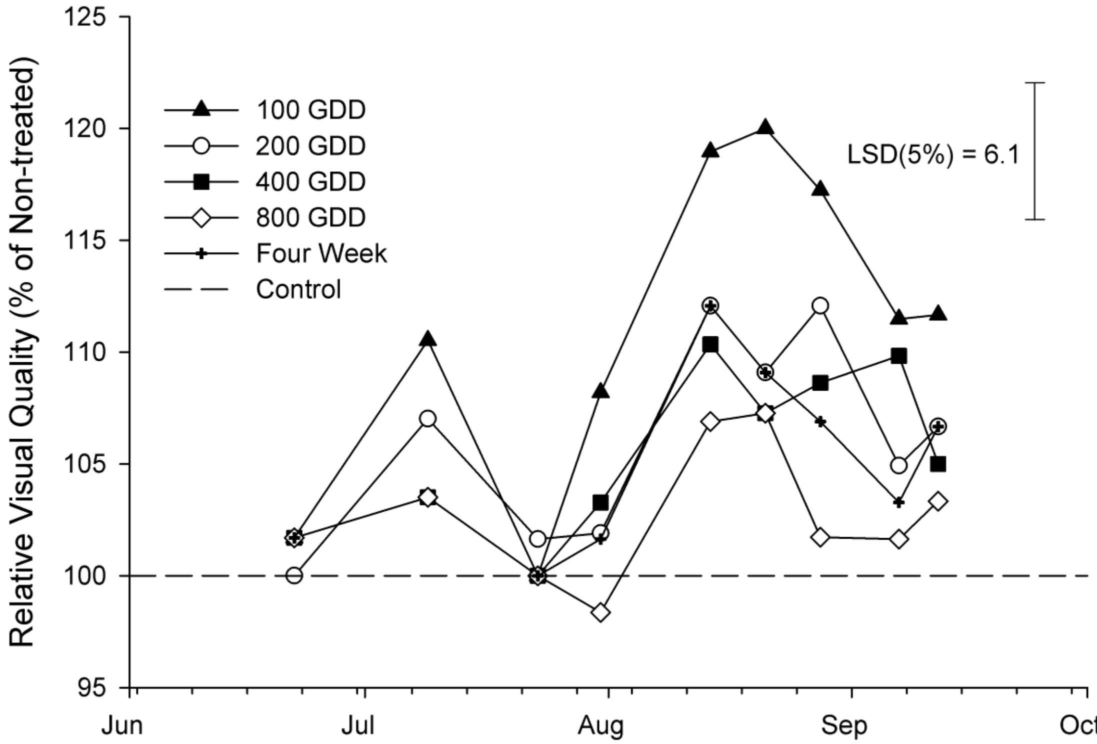


Figure 2.5. The effect of trinexapac re-application interval on relative visual quality in 2008. The dashed line represents the visual quality of non-treated control. Growing degree day (GDD) is the summation of daily average air temperature in degrees Celsius. TE is re-applied once each GDD threshold is surpassed. GDD is reset to zero after TE is re-applied.

Experiment 2: Model Validation

Clipping Yield

In 2009 and 2010, the four-week re-application treatments preformed similarly to the 400 GDD, 800 GDD, and four week re-application intervals from Experiment #1 (Figure 2.1). Relative yield suppression was not held constant at any application when TE was applied every four weeks (Figure 2.5). In both years, relative yield suppression was followed by yield rebound; both the suppression and rebound phases were approximately 20% less and greater than the non-treated control, respectively (Figure 2.6). The cool weather in 2009 drastically limited GDD accumulation in the four week re-application interval and did not allow for sine regression. Above average temperatures in 2010 increased GDD accumulation for the four week re-application treatments compared to 2009.

The 200 GDD re-application frequency sustained growth suppression by 20% regardless of application rate (Figure 2.7). The slope of the GDD by relative yield regression was not significant (p=0.8081) regardless of application rate which is consistent with Experiment #1. There was not a year effect despite below average temperatures in 2009 and above average temperatures in 2010. This indicates that the 200 GDD re-application maintained consistent growth suppression across a wide range of temperatures. The 200 GDD re-application interval decreased relative clipping yield by 21% for both application rates (Table 2.10). The reason for the increased yield suppression at the 0.05 kg a.i. ha^{-1} rate from 11% in Experiment #1 to 21% in Experiment #2 is unclear. There may be morphological differences with between the 'L93' and 'Penncross' cultivars or differences in sprayer nozzles (flat fan vs. air induction) that may have resulted in higher TE absorption in Experiment #2.

The level of growth suppression was approximately 20% in the 100 GDD treatment in Experiment #1 and both application rates during both years in Experiment #2, substantially below the 50% yield suppression stated on the product label. Increasing application rate in Experiment #2 did not increase relative yield suppression or duration of TE efficacy. Such a similarity in yield suppression may indicate the turfgrass' inability to absorb the TE at putting green height of cut. Decreased herbicide efficacy has been observed before at putting green height creeping bentgrass with herbicide bispyribac-sodium. Branham and Calhoun (2005) speculated that the low leaf area index of putting green turf limited herbicide absorption. Data from Experiments #1 and #2 support this notion as doubling re-application frequency approximately doubled yield suppression while doubling application rate had no effect on magnitude of growth suppression.

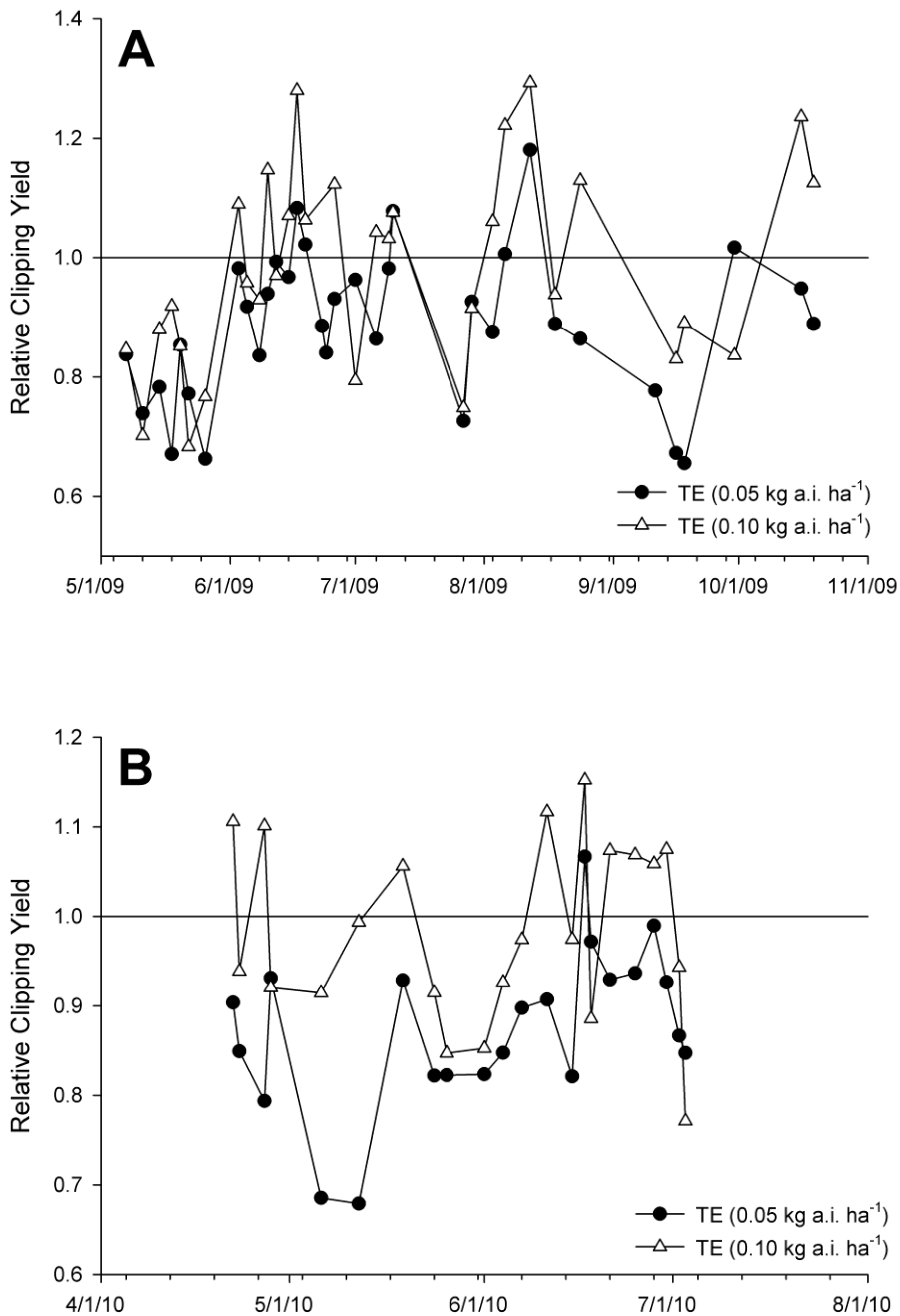


Figure 2.5. The effect of trinexapac-ethyl (TE) application rate on relative clipping yield when TE is re-applied every four weeks in 2009 (A) and 2010 (B).

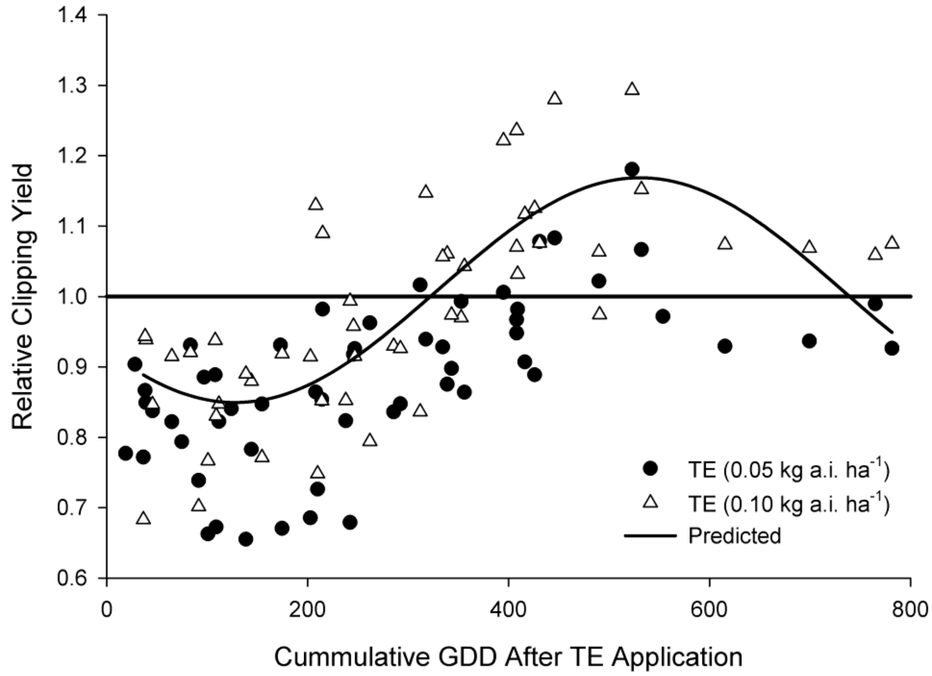


Figure 2.7. The effect of four week trinexapac-ethyl (TE) re-applications at different rates on relative clipping yield from Experiment #2. The predicted relative clipping yield from Experiment #1 is indicated by the solid line. The solid line represents the relative clipping yield of the control. Data from 2009 and 2010 were pooled together at each application rate for greater model resolution.

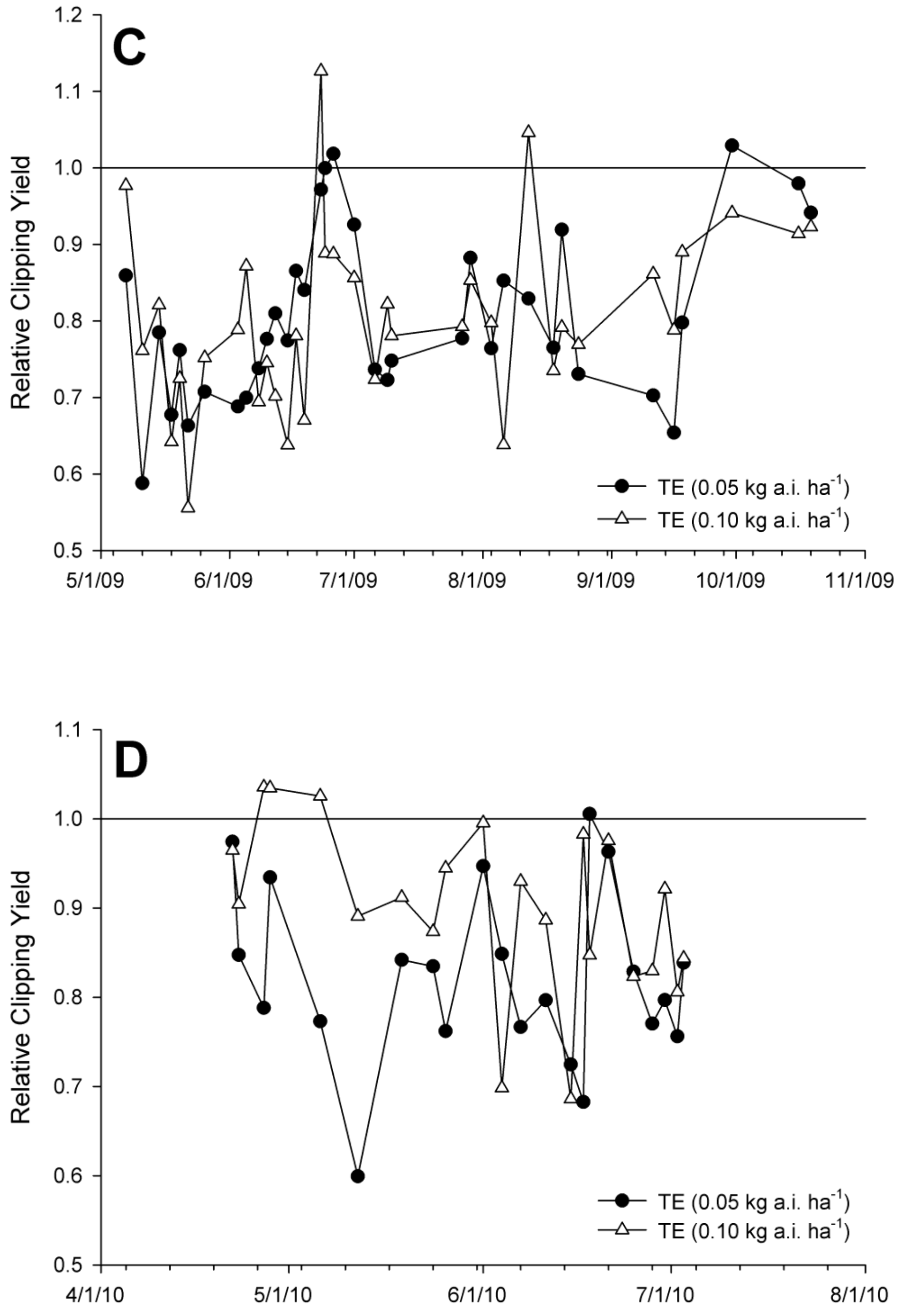


Figure 2.8. The effect of trinexapac-ethyl (TE) application rate on relative clipping yield when TE is re-applied every 200 GDD in 2009 (C) and 2010 (D). TE is re-applied one each GDD threshold is surpassed. GDD is reset to zero after TE is re-applied.

Table 2.8. The effect of trinexapac-ethyl (TE) application rate and re-application interval on relative clipping yield from treatments in Experiment #1 and #2 where season-long clipping suppression was maintained. Data from 2009 and 2010 were pooled together at for each respective application rate. Application rate did not affect the magnitude of clipping suppression in Experiment #2. Both application rates re-applied every 200 growing degree day in Experiment #2 had similar relative yield suppression as the 100 GDD re-application interval in Experiment #1.

Experiment	Application Rate	Re-application Frequency	Relative Clipping Yield
	kg a.i. ha^{-1}		% of Non-treated
# 1	0.05	100 GDD	81 A
# 1	0.05	200 GDD	88 B
# 2	0.05	200 GDD	80 A
# 2	0.10	200 GDD	79 A
# 2	0	n/a	100 C

† Column means followed by different letters are statistically different according to Fishers LSD (α=0.05).

Color and Quality

TE application rate and frequency significantly affected turfgrass quality ratings (Table 2.10). The 200 GDD application rate at the 0.10 kg a.i. TE ha^{-1} application rate consistently provided the highest quality. The 0.05 kg a.i. ha^{-1} application rate had turfgrass quality ratings similar to the control (Table 2.11). The 200 GDD re-application interval increased quality ratings by 0.3 units. Application rate but not frequency had an effect on color index (Table 2.11). The 0.10 kg a.i. ha^{-1} application rate enhanced color index by 11 and 9 units compared to the 0.00 and 0.05 kg a.i. ha^{-1} application rates, respectively. These results are similar to those reported by Stier et al. (1999) and are similar to the 100 GDD treatment in Experiment #1.

There was a strong date and date x application rate effect in both 2009 and 2010. In both years, turfgrass color and visual quality increased after winter during the spring and early summer (Figure 2.8). The slight decline in visual quality and CI in early July 2009 may have resulted from of above average temperatures that occurred in late June 2009 (Figure 2.8). Visual quality and CI enhancements occurred four to six weeks after the initial TE applications and caused the rate x date interactions in both years (Figures 2.8). These color enhancements improved as the growing season progressed.

The color index and visual quality enhancements have been previously reported in the literature (Ervin & Koski, 2001). In both of our experiments, these enhancements became statistically different than the non-treatments four to eight weeks after initial TE applications (Figure 2.9); a result consistent with McCullough et al. (2006a). After such time quality and CI enhancements increased in magnitude with time.

Re-applying TE every 200 GDD caused sustained enhancements in turfgrass quality while the four week re-application interval did not statistically affect quality compared to the non-treated control. Maintaining the suppression phase by re-applying TE every 200 GDD may sustain increases in total nonstructural carbohydrate (TNC) and cytokinin concentrations within the plant which result in the sustained CI and quality enhancements. Too infrequent of TE applications may play a role in variability of TNC concentrations experienced by several researchers (Han et al., 1998, 2004; Richie et al., 2001; Waltz and Whitwell, 2005).

Table 2.9. The ANOVA table for the effects of trinexapac-ethyl application rate, re-application interval, date, and interactions on visual quality and color index.

Source	df	Turfgrass Visual Quality		Color Index	
Application Rate (R)	2	0.0003	***	0.0152	*
Application Interval (I)	1	0.0152	*	0.1088	
Date[Year] (D)	14	<0.0001	***	<0.0001	***
R x I	2	0.1920		0.5819	
R x Year	2	0.6184		0.6324	
I x Year	1	0.2096		0.8347	
R x I x Year	2	0.3959		0.3206	
D x R	28	<0.0001	***	0.0448	*
D x I	14	0.3644		0.0865	
R x I x D	28	0.3530		0.9871	

Table 2.10. The effect of trinexapac-ethyl (TE) application rate on turfgrass visual quality and color index during Experiment #2.

TE Application Rate	Turfgrass Quality	Color Index
kg a.i. ha^{-1}	1 to 9 Scale	0-999
0.00	6.9 B	236 B
0.05	7.1 B	238 B
0.10	7.4 A	247 A

† Column means followed by different letters are statistically different according to Fishers LSD (α=0.05).

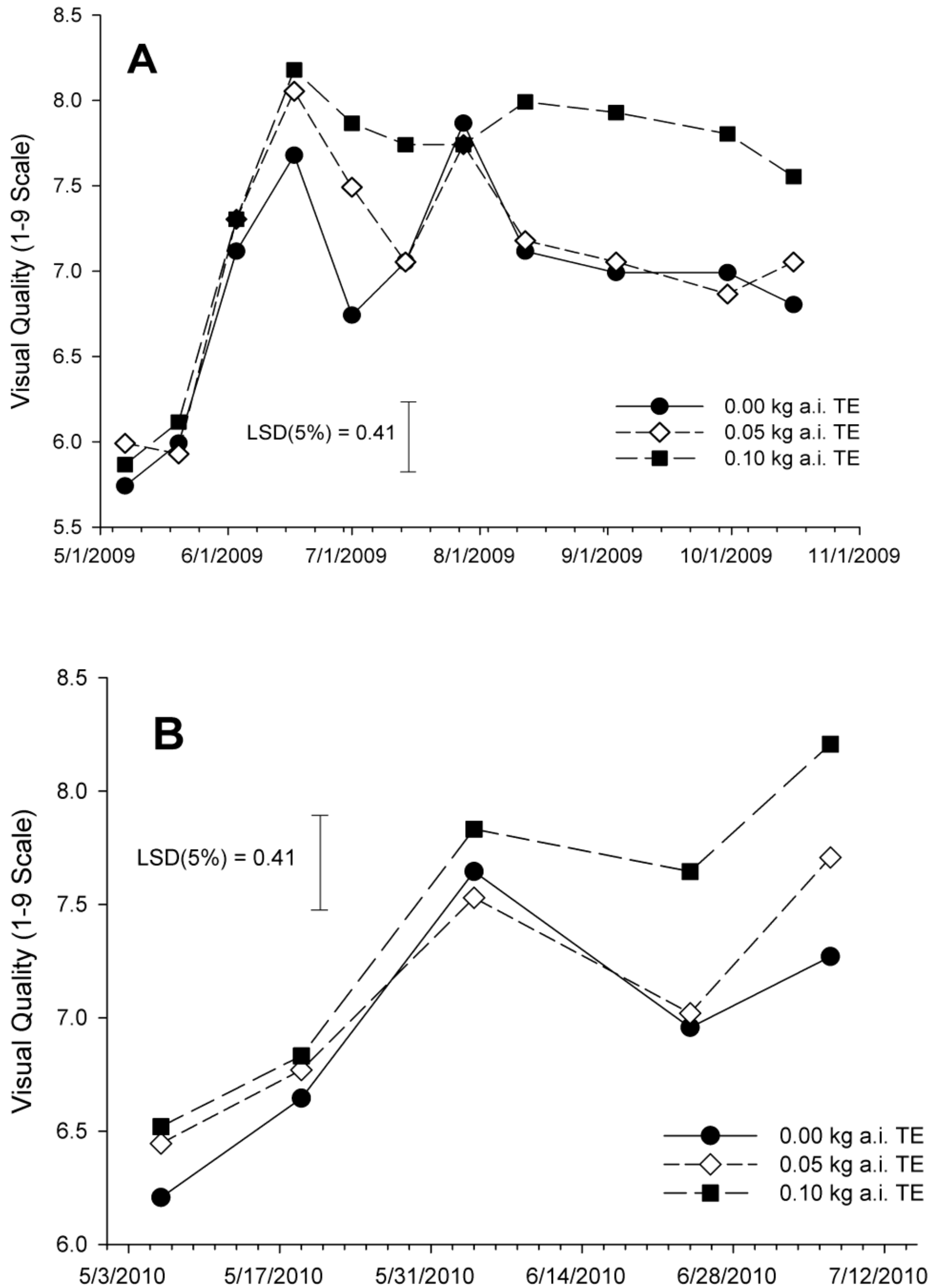


Figure 2.9. The effect of trinexapac-ethyl (TE) application rate on turfgrass visual quality during 2009 (A) and 2010 (B).

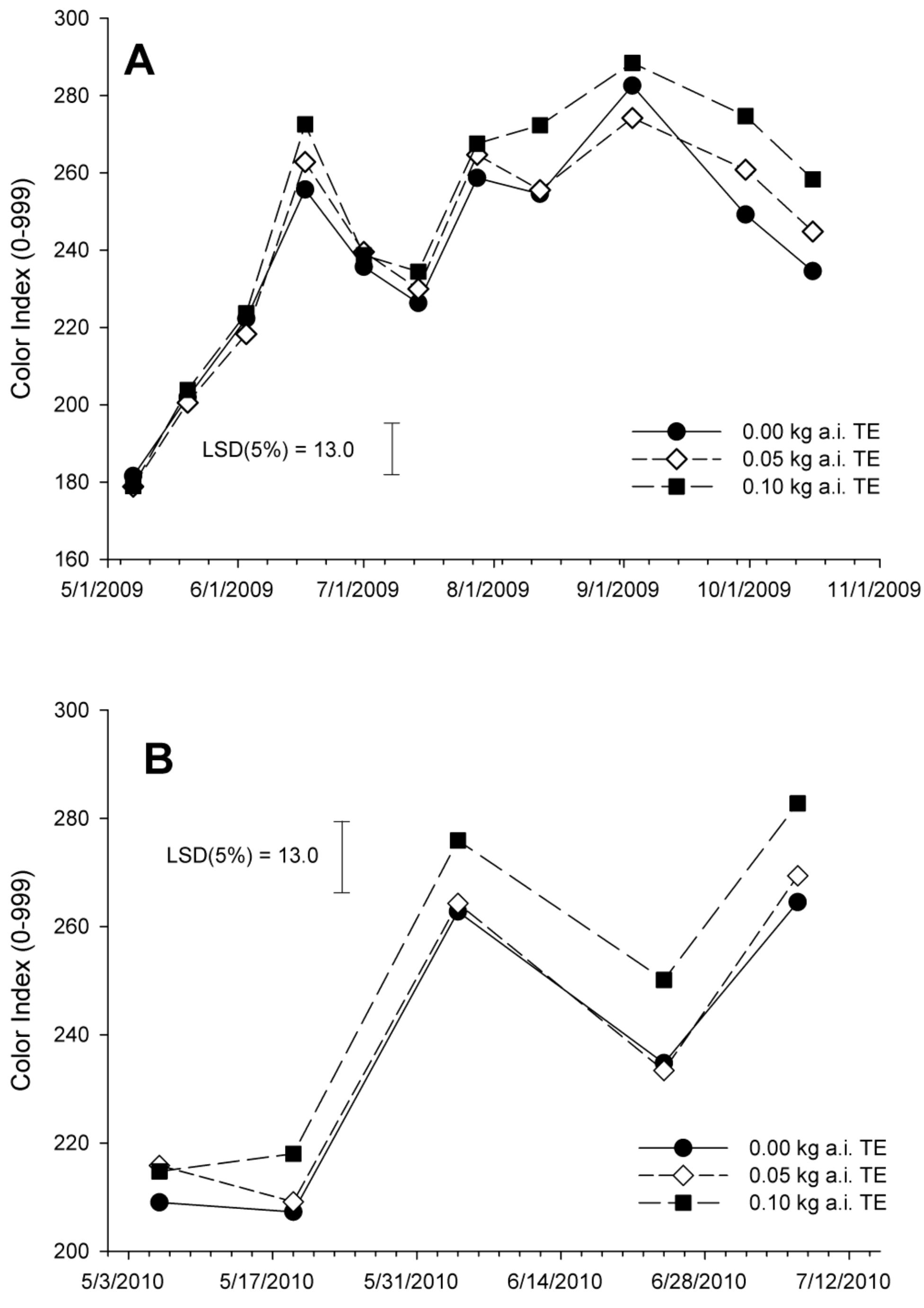


Figure 2.10. The effect of trinexapac-ethyl (TE) application rate on turfgrass color index during 2009 (A) and 2010 (B).

CONCLUSIONS

The results of these experiments indicate that a GDD model can serve as an effective tool for scheduling TE re-applications to maintain season long growth suppression on creeping bentgrass golf putting greens. Clipping yield suppression ranged from an average of 11 to 20% of the non-treated control; much less than the 50% inhibition reported on the product label. Four-week re-application intervals resulted in no net yield reduction over the season due to the rebound phase. Sustained yield suppression through multiple TE applications created enhancements in turfgrass color and quality six to eight weeks after initial TE applications. Consistent yield reductions may also reduce creeping bentgrass nutrient requirements because of reduced clipping and nutrient removal. However, further evaluation is required.

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