Growing Degree-Days Optimize Trinexapac-ethyl Reapplications for Ultradwarf Bermudagrass Putting Greens

by

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Abstract

Applying trinexapac-ethyl (TE) is a standard practice for growth suppression and quality improvement of ultradwarf bermudagrass [*Cynodon dactylon* (L.) Pers. x *C. transvaalensis* Burtt-Davy] putting greens, but research is lacking on proper reapplication frequency and rate. Making properly-timed reapplications is necessary to maintain turfgrass suppression and quality benefits. Ability to predict the maximum suppression point (MSP) that follows a TE application is helpful for making a proper reapplication; however, predicting the MSP is difficult because suppression duration is affected by environmental conditions, especially temperature. Previous research shows that growing degree-days (GDD), a variable unit that accounts for temperature, effectively predicts the suppression of creeping bentgrass (*Agrostis stolonifera* L.) following a TE application. Research was conducted to identify the optimal variable for predicting the MSP after a TE application on a 'MiniVerde' ultradwarf bermudagrass putting green. Tested variable units included: calendar days, GDD (base temperatures of 0 to 12ºC), soil temperature (2.5 cm), global horizontal irradiance, and photosynthetically active radiation. Pseudo-R^2 values from the resulting models suggest that GDD_0 (GDD with a base temperature of 0ºC) predicts the MSP better than the other variables. The GDD_0 model (pseudo-R^2: 0.564, SE: 0.195) indicates that the MSP occurred at 262 GDD_0. From these results, we hypothesized that reapplying TE before the MSP on a 200-GDD_0 interval would maintain suppression and quality benefits throughout the season. The objective of the second experiment was to test a GDD_0 reapplication schedule for an entire growing season. We included 4 GDD$_0$ intervals (100, 200, 400, and 600) and 2 TE rates (0.022 and 0.044 kg ai ha^{-1}). As expected, the 100- and 200-GDD$_0$ intervals resulted in consistent suppression throughout the experiment, and suppression magnitude increased with the higher TE rate. The 400- and 600-GDD$_0$ intervals caused fluctuation in suppression magnitude from day-to-day. Phytotoxicity occurred after initial applications and was more severe for the high rate.

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Table of Contents

List of Tables

List of Figures

List of Abbreviations

DAT	Days after treatment
DAIT	Days after initial treatment
g m^{-2} d^{-1}	grams of dry clippings per square meter per day
GA_1	the first bioactive gibberellic acid
GA_{20}	a non-bioactive gibberellic acid, the precursor to the first bioactive form
GDD	Growing degree-days
GDD_0	Growing degree-days with a base temperature of 0°C
GDD_{10}	Growing degree-days with a base temperature of 10°C
kg ai ha^{-1}	kilograms of active ingredient per hectare
MSP	Maximum suppression point
PGR	Plant growth regulator
TE	trinexapac-ethyl
TA	trinexapac acid
WAIT	weeks after initial treatment

Chapter 1. Literature Review

Introduction

Turfgrass is beautiful. It enriches landscapes, provides sport surfaces, reduces soil erosion, filters runoff, creates jobs, and even produces oxygen (Turgeon, 2002). But turfgrass grows, and this growth necessitates regular mowing to maintain acceptable quality. In addition to being dangerous, mowing is labor intensive and stresses the turfgrass (Christians et al., 2011). In the 1940s researchers developed chemicals that could slow turfgrass growth and reduce the mowing requirement (Watschke et al., 1992). However, most chemicals that slowed growth also caused unacceptable turfgrass injury. According to Watschke (1992), the ideal characteristics for a plant growth regulator (PGR) for high-quality turfgrass include:

1. Inhibits vertical foliar growth.
2. Inhibits seedhead development.
3. Does not inhibit lateral regrowth.
4. Does not inhibit root growth.
5. Does not cause unacceptable turfgrass injury.
6. Does not favor the growth of weeds.

By the 1990s several synthetic chemicals that specifically targeted cell elongation and division fulfilled these requirements (or at least most of them). Today, about 80% of golf courses in the southeast apply PGRs to reduce mowing requirement and enhance turfgrass quality (Auburn, 2016).

Plant Growth Regulators

The term “plant growth regulator” refers to any exogenously applied chemical that alters plant growth and development (Christians et al., 2011). This includes both natural and synthetic compounds, but in turfgrass “PGR” most frequently refers to the latter. While the purpose of this literature review is to examine PGRs in turfgrass systems, note that many of the same active ingredients are important for crops, rights-of-way, and other agricultural systems. For instance, prohexadione-calcium is applied to peanuts for maintaining row definition, which increases harvest efficiency; and it is applied in turfgrass for suppression of vertical growth and turfgrass quality benefits. This is to say, implications of research completed in turfgrass systems may also apply to these agricultural applications, and vice versa.

For high-quality turfgrass, like golf courses and athletic fields, PGRs can significantly reduce mowing requirement and simultaneously generate “secondary” benefits. Reducing mowing is valuable because the average golf course has 40 acres of fairways that are mowed approximately 3 times per week. Anecdotally, as no published research is available, a golf course might be able to reduce this mowing to 2 times a week if a PGR is applied at a proper rate and frequency. This reduction should save about 8 hours of labor and equipment wear per week; however, the monetary savings of applying a PGR on fairways is dependent on the costs of labor and fuel, in addition to the cost of the PGR. Analysis of this cost is not available for fairways, but an analysis for Primo

Maxx, the most commonly applied PGR in turfgrass, applied to golf course rough indicates that an application can save around \$600 per month on maintenance costs by reducing mowing requirement from twice a week to only once (Kowalewski et al., 2014)—savings based on 2014 fuel cost (\$3.67 per gallon) and labor (\$7.25 per hour). However, these savings were eliminated by the \$1900 per month cost of application—Primo Maxx at \$35 per acre. This could be reduced to about \$1000 by using a generic version of Primo Maxx, which currently (2019) retail for about \$18 per acre. If applied solely for reducing mowing, PGRs will only be cost-effective when labor and fuel costs outweigh the cost of the PGR.

Importantly, mowing reduction is not the only benefit of PGR applications. In fact, for golf putting greens, PGRs are not applied to reduce mowing requirement. Most golf courses mow putting greens 6 times per week, with or without a PGR—90% of Southeastern golf courses apply PGRs to putting greens (Auburn, 2016). For putting greens, the secondary benefits that result from reduced vertical growth are the primary reason for making the application: sustained green speed (ball roll), enhanced color, increased tiller density, increased root mass, improved shade tolerance, reduced water and nutrient requirement, and others (Baldwin et al., 2009; Fagerness et al., 2000; King et al., 1997; McCarty et al., 2011; McCullough et al., 2005a). These secondary benefits are generated by the suppression of vertical foliage growth, so to maintain these secondary benefits, it is necessary to maintain suppression with properly-timed sequential applications.

The problem for turfgrass managers is knowing when to make a sequential application. Too soon may lead to turfgrass injury; but too late may not maintain turfgrass suppression and allow turfgrass to enter a period of accelerated growth (“rebound”). When rebound occurs, growth is greater than non-treated turfgrass, and this can cause serious quality reductions and increased mowing requirement. Consequently, a substantial portion of the literature is devoted to determining the duration of suppression following a PGR application so that turfgrass managers can make informed reapplications.

The purpose of this literature review is to examine the research conducted on turfgrass suppression following a PGR application, specifically Primo Maxx (trinexapac-ethyl). I will begin by discussing the available PGR modes-of-action, then focus on the mode-of-action of trinexapac-ethyl and the available suppression research. I will conclude by presenting the most recent research that suggests an optimal reapplication schedule should not be based on calendar days, but it should be based on growing degree-days.

Turfgrass Growth Regulators

Regulating turfgrass growth is a delicate process. Turfgrass managers need to apply enough PGR to effectively reduce growth, but also avoid applying too much, which can cause excessive growth reduction and turfgrass injury. The earliest PGRs were not commonly used on high-quality turfgrass because the risks of injury outweighed the benefits. In fact, the earliest PGRs were applied primarily to reduce seedhead production in low-quality turfgrass. By the 1990s several new chemicals targeting gibberellic acid (GA) biosynthesis emerged that could effectively reduce both vertical turfgrass growth and seedhead production, without causing unacceptable injury. Many of these chemicals are still popular today.

Originally, turfgrass scientists separated PGRs into two types: “Type I” and “Type II.” The Type I PGRs interfered with cell division and differentiation, and the Type II PGRs interfered with GA biosynthesis. The Type I PGRs were considered growth *inhibitors* because they completely stopped growth, whereas the Type II PGRs were known as *suppressors* because growth continued at a reduced rate (Watschke et al., 1992). However, by 1995 the influx of new chemicals rendered it necessary to divide PGRs into five new classes (Turgeon, 2002; Watschke and DiPaola, 1995):

Class A: Late Gibberellic Acid Biosynthesis Inhibitors
Class B: Early Gibberellic Acid Biosynthesis Inhibitors
Class C: Mitosis Inhibitors
Class D: Herbicides
Class E: Hormones

Class C includes PGRs, like mefluidide, that are used primarily for seedhead control (Watschke et al., 1992). Class D includes the herbicides that, when applied at low rates, can slow plant growth without resulting in plant death, but these are not used on high-quality turfgrass. This class predominately includes glyphosate and the acetolactate synthase (ALS) inhibitors. Class E, the natural plant growth regulators, includes ethephon, gibberellic acid, and other natural hormones. Turfgrass managers can apply gibberellic acid to increase cell elongation (and, thus, turfgrass growth), which may be helpful in the case of an overapplication of a class A or B PGR, the inhibitors of gibberellic acid biosynthesis. Today, class A and B are the most important in the turfgrass industry. The remainder of this literature review will focus on these PGRs—starting with a review of GA biosynthesis.

Gibberellic Acid Biosynthesis

Scientists first identified gibberellic acid (GA) in rice infected with *Gibberella fujikuroi*, a pathogen that causes bakanae disease (Hedden and Phillips, 2000). Rice infected by this pathogen has excessive stem elongation, and this was determined to be a result of the pathogen producing GA. “Gibberellic acid” does not refer to a single compound, but rather a collection of tetracyclic diterpenoids that include many non-bioactive forms and a few bioactive forms (Rademacher, 2000). They are named by a number system (GA_1 – GA_n) with the number indicating the order of identification by scientists, which was largely a factor of abundance in the plant.

As described by Rademacher (2000): synthesis of GA begins in the proplastids with the conversion of mevalonic acid (MVA) into isopentenyl diphosphate (IPP). The IPP is transformed into the 20-carbon compound geranylgeranyl diphosphate (GGPP), then the GGPP is converted to *ent*-kaurene. Next, in the endoplasmic reticulum, the *ent*-kaurene is oxidized to *ent*-kaurenoic acid. Importantly, this oxidation requires O_2 and NADPH and involves cytochrome P450-dependent monooxygenases—these P450-dependent monooxygenases are the site of action for the early GA inhibitors (Hedden and Phillips, 2000). *ent*-kaurenoic acid is then hydroxylated to *ent*-7α-hydroxykaurenoic acid, which is transformed to GA_{12}-aldehyde.

The remaining conversions take place in the cytosol—note that no bioactive GA has been formed to this point. In the cytosol, GA_{12}-aldehyde is converted into GA_{12}, which starts a series of conversions to other non-bioactive GAs (this is dependent on species). The conversion leading to the first bioactive form is the 3β-hydroxylation of GA_{20} to GA_1. Importantly, this hydroxylation reaction that forms the bioactive GA_1 requires catalyzation by soluble dioxygenases that use 2-oxoglutaric acid as a co-substrate—this catalyst is the site of action for the late GA inhibitors, which are structural mimics of 2-oxoglutaric acid (Hedden and Phillips, 2000).

Following this reaction, the bioactive GA_1 signals for cell elongation, and, thus, turfgrass growth. The exact mechanism of signaling is not completely understood but involves the degradation of proteins that inhibit the phytochrome-interacting transcription factor (PIF). When GA_1 degrades these proteins that inhibit PIF, transcription of cell elongation genes is possible (Gupta and Chakrabarty, 2013; Santner et al., 2009). Eventually, the same catalyst involving 2-oxoglutaric acid converts GA_1 to GA_8 by a 2β-hydroxylation (Rademacher, 2000). This renders the compound inactive.

Both the early and late GA inhibitors prevent the formation of GA_1, but the early GA inhibitors prevent the production of all GA forms. The following section will detail the mechanism of late gibberellic acid inhibitors.

Late Gibberellic Acid Biosynthesis Inhibitors

Early and late GA inhibitors are the most prevalent PGRs in turfgrass. In most cases, they provide remarkable growth suppression without causing unacceptable turfgrass injury. Early GA inhibitors, as the name suggests, inhibit GA early in the biosynthesis pathway, while late GA inhibitors do so late in the GA pathway. This varied site of action is one source of efficacy differences between the two classes. Another important difference is that early GA inhibitors are both foliar and root absorbed, while late GA inhibitors are only foliar absorbed. Several combination products mix early and late GA inhibitors, but there is little research available to attest to the benefit of the mixture (Cooper, 2003). Today, the most commonly applied PGR is trinexapac-ethyl, a late GA inhibitor (Auburn, 2016).

Trinexapac-ethyl. Late GA inhibitors are members of the acylcyclohexanediones family, which is similar in structure to the family of sethoxydim and clethodim (Fagerness and Penner, 1996). The most common class A inhibitor is trinexapac-ethyl [ethyl 4-[cyclopropyl(hydroxy)methylidene]-3,5-dioxocyclohexane-1-carboxylate]. Trinexapac-ethyl (TE) was first sold under the trade name “Primo Maxx.”

After the foliar application, Fagerness and Penner (1998) suggest that the plant base (leaf sheaths surrounding the crown) absorbs 80% of the applied TE within 1 hr and 96% by 24 hrs, while the leaf blade absorbed only 31% in 1 hr but reached 70% by 24 hrs. In contrast, the roots absorb <5% of applied TE in 24 hrs. Of the TE absorbed at the plant base, <5% moved to the roots and >60% moved acropetally. Of the TE absorbed by the leaf blade, <5% moved to the roots; and approximately 70% of TE applied to the leaf blade remained in the leaf blade, while about 20% moved basipetally accumulating at the growing points. Additionally, <3% of TE applied to the leaf blade or plant base was translocated to the rhizomes (or daughter plants).

Fagerness and Penner hypothesize that this lack of translocation to the roots and rhizomes allows for continued root growth and lateral spreading—an extremely important quality for sports fields and golf courses where turfgrass managers need vertical growth suppression *but not* lateral growth suppression that might hinder recovery from injury (Fagerness and Penner, 1998). These results are not without challenge though. A similar experiment conducted on young wheat seedlings suggests that 60% of applied TE is translocated to the roots before being translocated back to the plant base (Rademacher, 2014). Given that reduced root growth is not normally associated with TE applications, we should expect that the bioactive form of TE is not active in the roots, and the applicability of Rademacher (2014) results from young wheat seedlings is suspect.

Importantly, TE must first be converted into bioactive trinexapac acid (TA) before inhibition can begin, and research suggests that this conversion occurs almost completely by 24 hrs (Syhre et al., 1997). However, this conversion is dependent on temperature and light intensity (Rademacher, 2014). After conversion to this bioactive form, inhibition of GA_1 will begin, and turfgrass growth suppression will result.

This inhibition is possible because TA is structurally similar to 2-oxoglutaric acid. It interrupts GA biosynthesis by competing with 2-oxoglutaric acid to fulfill a role as a co-substrate of the dioxygenases that catalyze the 3β-hydroxylation of GA_{20} into GA_1 in the cytoplasm (Rademacher, 2000). When TA acts as the co-substrate, the hydroxylation does not occur, which halts GA biosynthesis at GA_{20}. Importantly, each step in the process continues up to this point, which results in a backlog of GA_{20} (Tan and Qian, 2003). Interestingly, 2-oxoglutaric acid also acts as a co-substrate in the 2β-hydroxylation of GA_1 to GA_8 (a non-bioactive GA), and Rademacher (2000) suggests that this may result in increased cell elongation rates if the dioxygenases are unable to convert already present GA_1 to GA_8, allowing already produced GA_1 to remain active in the cell. Researchers have not reported this problem in turfgrass.

Inhibition of GA biosynthesis continues until the plant metabolizes TA (Kreuser and Soldat, 2011). This degradation rate is highly correlated with temperature. In creeping bentgrass, the half-life of TA is about 6.4 d at 18ºC but only 3.1 d at 30ºC, and in Kentucky bluegrass the half-life is about 5.3 d at 18ºC and 3.4 d at 30ºC (Beasley and Branham, 2005). As a result, efficacy fluctuates throughout the growing season, and to compound the problem, plant GA production may increase with increasing temperatures and changing light intensity (Tan and Qian, 2003). That is to say, predicting turfgrass suppression magnitude and duration after a TE application is challenging. Specifically, this fluctuation renders the standard calendar-based reapplication schedule ineffective, but recent research indicates that reapplications based on growing degree-days, a unit that accounts for temperature, may solve this problem. The next section will survey the literature on TE with an emphasis on analyzing the Primo Maxx label claim about turfgrass suppression magnitude and duration. The final section will show that growing degree-days can help turfgrass managers make more informed reapplications of TE that will maintain turfgrass suppression and the secondary benefits.

Primo Maxx (trinexapac-ethyl)

Primo Maxx is the most commonly applied PGR in turfgrass (Auburn, 2016). Synthesized by Ciba-Geigy in 1983, it was sold beginning in 1993 under the trade name “Primo” (DiPaola and Shepard, 1996). Syngenta acquired TE in the late 1990s and renamed the product “Primo Maxx” in the early 2000s (USPTO). In late 2005, the patent covering TE expired, which resulted in many generic formulations and a subsequent reduction in application cost.

According to the Primo Maxx label, turfgrass managers should apply Primo Maxx to "well-maintained, quality turfgrass areas" for "managing growth, improving quality and stress tolerance, and edging of warm- and cool-season turfgrasses" (Primo Maxx label). More specifically, the label explicitly makes 13 claims about label rate applications:

1. Reduces vertical growth by approximately 50% over a 4-week period
2. Enhances color
3. Increases density
4. Increases root mass
5. Increases total nonstructural carbohydrates
6. Reduces water requirement
7. Improves drought tolerance
8. Improves shade/reduced light environment tolerance
9. Improves heat and cold tolerance
10. Suppresses seedheads
11. Extends the duration of paint for field stripes
12. Enhances the performance of fungicides
13. Suppresses anthracnose

The legally permitted rate and reapplication frequency of Primo Maxx are vague. The label allows reapplications of Primo Maxx as soon as the "turf resumes growth or more suppression is desired," and for increased suppression duration "a maximum of twice" the label rate may be applied. A turfgrass manager could legally apply double the label rate as often as desired as long as they do not apply more than 2.67 kg ai ha^{-1} per year—although the label does say "applications of Primo Maxx can be made as frequently as weekly," so applying more than once a week falls into the gray area. At "label" rates, these claims are not always supported by peer-reviewed research, especially the first claim of 50% vertical growth suppression over a 4-week period.

The goal of the next section is to determine the accuracy of the first label claim—50% suppression over a 4-week period. This review will focus on data available for creeping bentgrass (*Agrostis stolonifera* L.) putting greens, Kentucky bluegrass (*Poa pratensis* L.) sports fields, ultradwarf bermudagrass [*Cynodon dactylon* (L.) Pers. X. *C. transvaalensis* Burtt Davy] putting greens, and hybrid bermudagrass fairways [*Cynodon dactylon* (L.) Pers. X. *C. transvaalensis* Burtt Davy].

Turfgrass Suppression

Primo Maxx (referred to as TE or TA hereafter) suppresses turfgrass growth. This claim is not contested. The question is: do label rates "provide approximately 50% growth inhibition over a 4-week period…[beginning] about 3-5 days after application?" Many interpret this claim to mean that TE will begin to suppress growth 3-5 days after application, then this suppression will reach 50% (compared to the non-treated) and hold at 50% for a 4-week period. However, even a rudimentary understanding of chemical degradation should make us question this claim—if the common interpretation is correct.

As this claim is examined, careful consideration of word choice is necessary. For this review, "suppression magnitude" refers to turfgrass growth reduction relative to the non-treated (calculated as a percent suppression). The "maximum suppression point" (MSP) is the point of the greatest suppression magnitude following a single TE application. Additionally, the "suppression phase" is the period where growth is less than the non-treated (i.e., suppressed), and "post-inhibition growth enhancement" (rebound) is the period where growth rate is greater than the non-treated that may follow the suppression phase. In the following literature, it is important to differentiate "suppression magnitude at the MSP" from the "average suppression over the suppression phase." The Primo Maxx label refers to the latter, not the magnitude at the MSP.

Sinusoidal Suppression. Before examining the literature on turfgrass suppression, it is helpful to understand the progression of turfgrass suppression after an application. After a TE application, we should expect that suppression may take a few days to begin as TE must be converted to bioactive TA and available GA_1 is deactivated. As explained in the previous section, bioactive GA_1 signals for cell elongation. By preventing the conversion of non-bioactive GA_{20} to GA_1, TA reduces the concentration of GA_1 and increases the concentration of GA_{20}. Two weeks after treatment, TA reduced GA_1 by 47% and increased GA_{20} by 146% in Kentucky bluegrass (Tan and Qian, 2003). In synchrony with this GA_1 reduction, the turfgrass growth should decrease as cell elongation slows. Eventually, TA will reach its maximum concentration and, subsequently, GA_1 will reach its lowest concentration. At this point, turfgrass growth will be at the MSP, the point of slowest growth. After this point, TA concentrations begin to decrease, and GA_1 concentrations will slowly increase, which will also slowly increase turfgrass growth rates back to the non-treated levels. However, in the rebound phase, the GA_1 concentrations may continue to increase beyond the non-treated concentration as the elevated GA_{20} concentration is quickly converted to GA_1 (discussed below). Although, rebound varies with cultivar, temperature, TE rate, and TE frequency (Beasley et al., 2007; Fagerness and Yelverton, 2000; Kreuser and Soldat, 2011). Failure to prevent the rebound phase may lead to scalping and decreased quality. Therefore, the ability to predict suppression phase duration is very important for turfgrass managers.

When represented graphically, suppression magnitude over time is sinusoidal—where the amplitude is the suppression magnitude (below the x-axis is suppression, above is rebound); the local minimum, or trough, is the MSP; and the wave period is the duration of the suppression and rebound phases (Kreuser et al., 2018; Reasor et al., 2018). This graphical conception of turfgrass suppression following a TE application is helpful for understanding the following experiments and evaluating the label claim of a 50% reduction over a 4-week period.

Creeping Bentgrass Putting Greens. On creeping bentgrass putting greens (CBG), several studies indicate that the suppression magnitude, even at doubled rates, does not yield a 50% reduction over 4 weeks. When applied at the label rate (0.05 kg ai ha^{-1}) to CBG putting greens mowed at 3.20 mm, TE reduced growth by almost 40% at the MSP, but subsequent applications resulted in only about 30% suppression at the MSP (McCullough et al., 2007). As for suppression duration, this study suggests that the CBG returns to non-treated growth rates within 3 weeks. We can conclude, considering the sinusoidal nature of TE suppression, the *average suppression magnitude* over the 3-week period is less than 30% (again, not directly reported in this study, but mathematically necessary based on the reported suppression at the MSP).

Similarly, another study with both a label rate and double rate (0.05 and 0.10 kg ai ha^{-1}) indicates that suppression at the MSP may be slightly over 30% for both rates and the average suppression magnitude is less than 20% during the suppression phase (Kreuser and Soldat, 2011). This study noted that suppression duration is highly dependent on temperature, which is expected given the half-life data reported by Beasley and Branham (2005). Kreuser determined that suppression duration was 23 d in early spring, 17 d in late spring, but only 14 d by July. This suggests that a calendar-based reapplication schedule will not be effective for properly timing reapplications to maintain suppression, and Kreuser shows that a growing degree-day schedule is more effective (discussed below). Interestingly, Kreuser also reported that doubling the rate did not increase suppression magnitude or duration. Both studies indicate that TE at the label rate does not suppress CBG growth by 50% at the MSP—let alone over a 4-week period—and suppression duration is highly dependent on temperature.

Kentucky Bluegrass Sports Fields. While generally more favorable to the label claim, research on Kentucky bluegrass (KBG) also shows that both suppression magnitude and duration are highly dependent on temperature. A study with both a label rate and increased rate (0.27 and 0.40 kg ai ha^{-1}) on KBG mowed at 32.0 mm found that TE reduced growth in the spring and fall by close to 50% at the MSP; however, magnitude was closer to 35% at the MSP during the summer (Beasley et al., 2007). As for duration, TE maintained suppression close to 50% for 4 weeks in the spring and fall before returning to non-treated levels at 5 weeks. In the summer, the suppression only lasted 4 weeks and magnitude constantly decreased each week after reaching the MSP at 1 week after treatment. This suggests that the label claim is not supported during periods of elevated temperatures. Another study on KBG (32.0 mm) with TE at 0.29 kg ai ha^{-1} also showed reductions close to 50% at the MSP, and the average suppression magnitude for the entire season-long study was 41% when TE was reapplied every 4 weeks (Lickfeldt et al., 2001). Suppression duration was approximately 4 weeks during the summer but closer to 5 weeks in the spring. Similar to the CBG study, the authors note that suppression magnitude is dependent on temperature, and duration is not extended by increasing rate.

A study on KBG mowed at 55.0 mm (lawn height) with TE at 0.20 kg ai ha^{-1} reported around 50% suppression at the MSP. This single application maintained a 50% reduction for 3 weeks, while growth remained less than the non-treated for almost 6 weeks (King et al., 1997). Again, the authors noted that suppression duration was longer in fall and spring than in summer. In contrast with other studies, they report an increase in both suppression magnitude and duration with an increase in rate (0.20 to 0.60 kg ai ha^{-1}); however, there was not an increase in duration to the MSP, only an increase in average suppression magnitude and suppression phase duration. Also, the authors reported greater turfgrass discoloration (bronzing) at the higher rate. In conclusion, label rates of TE on KBG may provide an average suppression magnitude of 50% over 4-weeks, though duration is dependent on temperature; but TE does not maintain this 50% suppression for the entire suppression duration, rather suppression follows the sinusoidal waveform and averages to 50%.

Ultradwarf Bermudagrass Putting Greens. Compared to CBG, TE suppresses ultradwarf bermudagrass putting greens (UDG) much more, both in magnitude and duration. McCullough (2007) tested TE on a 'TifEagle' UDG maintained at 3.20 mm.

Both the label and increased rate (0.03 and 0.05 kg ai ha^{-1}) provided an average suppression magnitude of 55% over 3 weeks—this corroborates the results from a previous study at this location (McCullough et al., 2006a). Also, two greenhouse studies report similar conclusions: greater than 50% average suppression magnitude over at least 3 weeks (McCullough et al., 2005a; McCullough et al., 2006b).

In contrast, a field study with TE at the label rate on three popular UDG cultivars (‘TifEagle,’ ‘MiniVerde,’ ‘Champion’) found suppression magnitude to be between 49 and 62% at the MSP (Reasor et al., 2018). While the suppression duration in terms of calendar days was not explicitly stated in the paper, based on temperature data, the suppression duration was 3 to 4 weeks. Considering the sinusoidal waveform of suppression, it is unlikely that the average suppression magnitude the suppression phase was 50%. I argue that the previous studies on UDG (and possibly KBG) do not accurately present the average suppression magnitude because they did not collect clippings (to quantify growth suppression) often enough. The Reasor (2018) study is more likely to present an accurate depiction of suppression magnitude because they collected clippings approximately 3 times per week during the suppression phase. From the available literature, we can conclude that, as with CBG and KBG, suppression duration depends on temperature, and it is unlikely that a single application at the label rate will provide an average of 50% suppression over 4 weeks.

Hybrid Bermudagrass Fairways. As with UDG, field research on hybrid bermudagrass fairways (HBG) is limited, but the available research does indicate that HBG, at label rates, responds similarly to UDG. In a study on ‘Tifway’ HBG maintained at 16 mm (fairway height), TE at the label rate (0.10 kg ai ha^{-1}) provided a suppression magnitude of 50% at the MSP and suppression duration of about 4 weeks (Fagerness and Yelverton, 2000). The average suppression magnitude was 40% over the 4 weeks. A reduced rate (0.07 kg ai ha^{-1}) yielded similar results at the MSP, but the suppression phase only lasted 2 weeks. Another field study with TE at 0.11 kg ai ha^{-1} corroborates these results with a reported magnitude of 50% at the MSP and duration of 4 weeks; and, though not reported, the average suppression magnitude would necessarily be less than 50% (Fagerness et al., 2004).

Testing the influence of temperature on suppression, a study conducted in a growth chamber had 2 temperature environments: 35/25ºC and 20/10ºC (Fagerness et al., 2002). In the 35/25ºC environment, suppression magnitude and duration were similar to the previous studies (50% at the MSP, duration of 4 weeks), but in the 20/10ºC environment, magnitude increased to almost 60% at the MSP and duration was over 6 weeks. Once again, it is clear that TE efficacy is highly correlated with temperature, but it is also evident that the Primo Maxx label claim is suspect.

To conclude this section on suppression, only the research on Kentucky bluegrass supports the label claim that TE will "provide approximately 50% growth inhibition over a 4-week period." For the other turfgrasses, it is likely that any rate capable of suppressing growth by an average of 50% over 4 weeks would also cause unacceptable injury. The following section will examine the rebound phase that may follow the suppression phase.

Turfgrass Rebound

Mechanism. While it is clear that TE does reduce vertical growth, an examination of this claim would not be complete without mentioning the period of accelerated growth—rebound—that may follow the suppression phase. While not all studies report rebound, many do, and when the rebound phase does occur, it may eliminate any benefits that the suppression phase provided. The mechanism for this phenomenon is still debated, but two theories are most prevalent. The most widely accepted theory is that this accelerated growth occurs as the backlog of GA_{20} is quickly converted into GA_1. Tan and Qian (2003) determined that at 2 weeks after treatment GA_{20} concentration increased by 146%. If the 3β-hydroxylation of GA_{20} is catalyzed properly again and the backlogged GA_{20} is converted, then the GA_1 concentration would increase dramatically. Now the limiting factor for cell elongation is available energy from photosynthesis and stored reserves in conjunction with other environmental factors, viz., temperature and photoperiod (Fagerness and Yelverton, 2000). The second theory is that this accelerated growth is partly due to the accumulation of total nonstructural carbohydrates (TNC) during the suppression phase (McCarty et al., 2011). This theory is compatible with the first theory since this available energy source combined with the GA_1 signaling for growth means the only limiting factor is other environmental stimuli.

A third proposal, not really a theory, is that the appearance of rebound is, at least partially, the result of flawed experiment design for data collection. Turfgrass suppression is typically measured by collecting clippings from a treated plot and comparing the weight to that of a non-treated plot. However, a secondary benefit of TE is increased tiller density, so by the end of the season the treated plot should have more tillers and, thus, produce more clippings per area than the non-treated—even if both are growing at the same rate (Lickfeldt et al., 2001). This proposal does not explain the anecdotal evidence of significant scalping after TE applications end, but it could mean that the reported magnitude of rebound is exaggerated in experiments, especially if there were multiple TE applications that increased turfgrass density.

Creeping Bentgrass Putting Greens. In CBG, Kreuser and Soldat (2011) reported that rebound occurred beginning around 2 weeks after treatment (depending on temperature) when TE was applied on a 4-week interval at the label rate. This rebound phase was equal in both magnitude and duration to the suppression phase, which resulted in overall clipping yield being equivalent to the non-treated by the end of the 4-week period. In contrast, the McCullough (2007) study with the same CBG cultivar (L-93), height-of-cut (3.20 mm), and TE label rate (0.05 kg ai ha^{-1}) did not report any significant rebound during the 3-week clipping collection period. This result would be expected if the McCullough study took place in a cooler environment, but it was conducted in South Carolina during the summer, whereas the Kreuser study took place in Wisconsin. It is possible that the McCullough study missed the rebound because they only collected data once per week, compared to 5 times per week in the Kreuser study.

Kentucky Bluegrass Sports Fields. As for KBG, all three KBG studies mentioned in the “Primo Maxx and Turfgrass Suppression” section indicated rebound following suppression. Importantly, rebound may be much less of a problem for KBG because, at label rates, the suppression phase lasts at least 4 weeks, even during the warmer weather. Lickfeldt (2001) indicated that the rebound phase consisted of 20 to 60% increased growth for 2 weeks. This report is corroborated by Beasley and Branham (2007), but they noted that rebound was not as pronounced during the cooler year of the study.

Ultradwarf Bermudagrass Putting Greens. No field study has reported rebound following the suppression phase of UDB, which may be a result of the relatively low TE rates (McCullough et al., 2007; Reasor et al., 2018). We suggest that rebound does occur following the end of season-long sequential applications (Chapter 3).

Hybrid Bermudagrass Fairways. Research on 'Tifway' HBG indicates that rebound may occur. Fagerness (2000) reported significant rebound of nearly 50 and 25% increased growth over 4 weeks following the suppression phase when TE was applied at a reduced rate (0.07 kg ai ha^{-1}) or the label rate (0.10 kg ai ha^{-1}), respectively. From the previous research on other turfgrass species, it is not expected that the lower rate would produce more rebound, and this is not reported elsewhere in the literature.

While rebound is normally undesirable, Lickfeldt (2001) points out that turfgrass managers could use this accelerated growth to their advantage by timing the cessation of applications to have rebound occur during periods of heavy turfgrass use, such as the end of football season in the South when turfgrass growth is typically slowing. For golf course putting greens, rebound will rarely be beneficial, so knowing when to reapply TE is very important for turfgrass managers. As the previous section noted, determining suppression duration is difficult because it fluctuates with temperature. The following section will detail a potential solution that will help turfgrass managers make properly-timed reapplications.

Reapplying Trinexapac-ethyl

Growing Degree-Days. The previous sections demonstrated that suppression duration is dependent on temperature (and possibly other environmental conditions). This is problematic for turfgrass managers because, to maintain the benefits of TE and avoid the rebound phase, they need to properly time reapplications. Recently, a solution that accounts for this temperature dependency has emerged.

Growing degree-days (GDDs), a unit that accounts for temperature, more accurately predicts temperature-dependent events such as germination, flowering, and maturity, compared to calendar days (Cross and Zuber, 1972). For plant growth regulators in turfgrass, GDD was first used for predicting proper reapplication timing of mefluidide applications for seedhead control (Danneberger et al., 1987). For turfgrass growth suppression with TE on creeping bentgrass putting greens, Kreuser and Soldat (2011) suggest a 200-GDD_0 (base temperature of 0ºC) interval will maintain suppression all season, irrespective of temperature, but the 200-GDD_0 interval would accomplish this with 5 fewer applications than a calendar-day schedule that would also ensure suppression was maintained (assuming a season average of 18ºC from 15 April to 31 August). By more accurately predicting the MSP, this GDD model provides a practical benefit for turfgrass managers.

For TE applications on ultradwarf bermudagrass putting greens, only one publication on GDD schedules is available. Reasor et al. (2018) demonstrated that the MSP occurs after 166, 166, and 177 GDD_{10} (base temperature of 10ºC) for 'TifEagle' in Mississippi, 'Champion' in North Carolina, and 'MiniVerde' in Tennessee, respectively, following a TE application. They recommend that reapplications occur on a GDD_{10} interval determined by multiplying GDD_{10} accumulation at the MSP by 1.3 (the "1.3x method"). This suggests that TE should be reapplied to UDG approximately every 215 GDD_{10}. While the 1.3x method may be effective for creeping bentgrass, a season-long GDD reapplication schedule has not been tested for ultradwarf bermudagrass (Kreuser et al., 2018; Kreuser and Soldat, 2011).

Currently, GDD models are only available for TE and paclobutrazol (an early GA inhibitor) applications on creeping bentgrass and ultradwarf bermudagrass putting greens, though models are being developed for hybrid bermudagrass fairways. A GDD reapplication schedule will be advantageous for turfgrass managers if it provides a practical benefit, like reducing total applications or minimizing initial phytotoxicity. We expect that the benefits will be greatest in locations with fluctuating season temperatures. We designed our research at Auburn University to (i.) determine the best predictor variable for scheduling TE reapplications on ultradwarf bermudagrass and (ii.) test a reapplication schedule for an entire growing season. We hypothesized that a variable would better predict suppression duration if it accounts for the environmental conditions that may affect TA degradation rate. Tested variables included: calendar days, GDD (base temperature 0 to 12ºC), soil temperature (2.5 cm), global horizontal irradiance, and photosynthetically active radiation. In a separate experiment, we tested 4 GDD_0 intervals and 2 TE rates for an entire season.

Chapter 2. Predicting the Maximum Suppression Point to Optimize Trinexapac-ethyl Reapplications on Ultradwarf Bermudagrass

Introduction

Applying trinexapac-ethyl (TE) to ultradwarf bermudagrass putting greens [*Cynodon dactylon* (L.) Pers. x *C. transvaalensis* Burtt-Davy] is a standard practice. Trinexapac-ethyl is a plant growth regulator that suppresses vertical turfgrass growth, while minimally affecting root and lateral growth (Fagerness and Penner, 1998). Primary benefits for putting greens include sustained green speed, enhanced color, increased root mass, improved shade tolerance, and reduced water and nutrient requirement (Baldwin et al., 2009; Fagerness et al., 2000; King et al., 1997; Kreuser and Soldat, 2012; McCarty et al., 2011; McCullough et al., 2005a). Importantly, these benefits are derived from the vertical growth suppression, so maintaining suppression with properly-timed reapplications is necessary to preserve these benefits. Also, failure to reapply before the suppression phase ends could result in an undesirable period of accelerated turfgrass growth ("rebound"), which will reduce turfgrass quality (Beasley et al., 2007; Fagerness and Yelverton, 2000; Kreuser and Soldat, 2011). Because the duration of the suppression phase varies, the problem for turfgrass managers is knowing when to reapply (Kreuser and Soldat, 2011). An examination of TE on a molecular level illuminates a cause of this variation and a potential solution for turfgrass managers.

After an application, suppression does not begin until TE is absorbed by the foliage and converted to trinexapac acid (TA), the bioactive form (Fagerness and Penner, 1998; Rademacher, 2000). A majority of this conversion to TA takes place between 4 and 24 hrs after application (Syhre et al., 1997), but it may be affected by both temperature and light intensity (Rademacher, 2014). The bioactive TA suppresses turfgrass growth by inhibiting the 3β-hydroxylation of GA_{20} to GA_1, the first bioactive GA that signals for cell elongation (Rademacher, 2000). Eventually, this inhibition of GA_1 will subside as TA is degraded, and this degradation rate is highly correlated with temperature. For instance, the half-life of TA in creeping bentgrass (*Agrostis stolonifera* L.) is 6.4 d at 18ºC but only 3.1 d at 30ºC (Beasley and Branham, 2005). If temperature affects TA degradation rate, it follows that suppression duration will also vary with temperature. Therefore, an optimal reapplication schedule for TE should take temperature into account (Kreuser and Soldat, 2011).

Arguably, the solution to this problem is growing degree-days (GDD), a unit that accounts for temperature. Previous studies have shown that, compared to calendar days, GDD more accurately predicts temperature-dependent events such as germination, flowering, and maturity (Cross and Zuber, 1972). For turfgrass, GDD was first used for predicting proper timing of mefluidide applications for seedhead control (Danneberger et al., 1987), and more recently, Kreuser and Soldat (2011) developed a GDD reapplication schedule for TE reapplications on creeping bentgrass putting greens. They determined that a 200-GDD_0 (base temperature of 0ºC) interval will maintain suppression all season, irrespective of temperature. As for ultradwarf bermudagrass putting greens, Reasor et al. (2018) recommended that TE be reapplied to 'MiniVerde' ultradwarf bermudagrass every

230 GDD_{10} (base temperature of 10ºC). They calculated this interval by multiplying the total GDD_{10} accumulation at the maximum suppression point (MSP) by 1.3, which was shown to be an effective method for creeping bentgrass (Kreuser et al., 2018; Kreuser and Soldat, 2011). Knowing when the MSP will occur is important for making properly-timed reapplications because, after the MSP, the GA_1 concentration begins returning to the non-treated level—and turfgrass growth rate follows in synchrony.

Our objective was to identify the optimal variable unit for predicting the duration to the MSP after a TE application on ultradwarf bermudagrass putting greens. We hypothesized that a variable will better predict the MSP if it accounts for the environmental conditions that affect TA degradation rate. Tested variables included: calendar days, GDD (base temperature 0 to 12ºC), soil temperature (2.5 cm), global horizontal irradiance (GHI), and photosynthetically active radiation (PAR). The results should allow us to recommend an optimal TE reapplication interval that will maintain both suppression and quality benefits. In a separate experiment, we tested four GDD intervals and two TE rates for an entire season (Chapter 3).

Materials and Methods

Research was designed to examine how environmental conditions affect vertical turfgrass suppression following a TE application. A field experiment was conducted on a MiniVerde bermudagrass putting green at the Sports Surface Field Laboratory in Auburn, AL during 2016 and repeated in 2017. The putting green was constructed in 1994 according to United States Golf Association specifications (USGA, 1993) and sprigged with MiniVerde in April 2004. On 1 April 2016, 9 September 2016, and 2 March 2017, the green was hollow-tine aerated and topdressed heavily. During the experiment, plots were not topdressed or cultivated. Following green-up in April, the plots were fertilized with liquid urea at 12.2 kg N ha^{-1} wk^{-1}. Phosphorus and potassium were added based on soil test results. Preventative fungicides to control mini-ring (*Rhizoctonia zeae*) and dollar spot (*Sclerotinia homoeocarpa*) were applied beginning in May. This included azoxystrobin, chlorothalonil, and mancozeb, which are not known to regulate plant growth. Plots were irrigated daily at approximately 80% of the evapotranspiration rate.

Treatments included a single TE application at 0.044 kg ai ha^{-1} (Primo Maxx, Syngenta, Greensboro, NC) to a previously untreated plot on 7 dates spread across the growing season (ideally comprising different environmental conditions): 1 May, 15 May, 1 June, 15 June, 1 July, 15 July, and 1 August. Applications were made with a CO_2 sprayer calibrated to deliver 375 L ha^{-1}. Irrigation was withheld for at least 1 hr following TE applications. Treatments were arranged in a randomized complete block design with four replicates on 1.5 by 1.5 m plots, and a non-treated control was included in each replication.

Environmental conditions. A weather station (WMR300, Oregon Scientific, Tualatin, OR) positioned 1.5 m above the experiment area recorded air temperature and rain, and a soil thermometer (Decagon EM50, METER, Pullman, WA) recorded temperature at 2.5 cm. A separate weather station (Enviromonitor, Davis Instruments, Hayward, CA) in a field located 2 km northeast of the experiment area recorded global horizontal irradiance (Wh m^{-2}) for 2016 and 2017. In 2017, photosynthetically active radiation (μmol m^{-2} s^{-1}) was also recorded above the experiment area. The daily soil temperature, global horizontal irradiance (GHI), and photosynthetically active radiation (PAR) were calculated by taking the average of data points recorded every minute. Total accumulation is the sum of the daily averages beginning on the application date.

The daily high and low air temperatures were recorded in Celsius for each 24 hr period beginning at midnight (Fig. 1). Daily GDD was calculated with the equation:

$$\text{Daily GDD} = \frac{T_{\text{high}} + T_{\text{low}}}{2} - T_{\text{base}}$$

Where if $[(T_{\text{high}}+T_{\text{low}})/2]<T_{\text{base}}$, then the GDD for that day is set to 0, which prevents negative GDD accumulation (McMaster and Wilhelm, 1997). Total GDD accumulation is the sum of daily GDD beginning on the application date. We tested base temperatures between 0 and 12ºC to determine which is most appropriate for a TE model on ultradwarf bermudagrass in the southeastern United States.

Growth Suppression. Vertical growth suppression was approximated by collecting clippings three days per week at 1100 ± 1 hr with a Jacobsen walking greens mower (Greens King 522, Jacobsen, Augusta, GA) set at 3.4 mm. Clipping collections began 2–3 days after the TE application and continued for at least 30 days after application (in 2016 clipping were collected for all treatments dates until September 2).

Before collection, alleys were mowed down the edges of plots (perpendicular to the collection mowing direction) to ensure a total collection area of 0.535 m^2 per plot. Collection mowing direction was altered by 180-degrees each collection date to reduce grain formation. For collection, a single pass was mowed across the center of each plot, then the clippings were removed from the clipping basket using a handheld vacuum (PHV1810, Black & Decker). Clippings were emptied out of the vacuum into a bag, then oven-dried at 60ºC for at least 48 hrs before weighing to the nearest centigram. Following the collection, the entire experiment area was mowed at 3.4 mm with the same Jacobsen greens mower (and plots were mowed only on collection days).

Relative clipping yield (g g^{-1}) was calculated by dividing the weight of the treated by the non-treated control within each replication. For each of the 7 treatment dates of 2016 and 2017, relative clipping yield was plotted as a function of days after treatment (DAT), soil temperature accumulation, GHI accumulation, PAR accumulation, and GDD accumulation—with base temperatures of 0, 2, 4, 6, 8, 10, and 12ºC.

Using SigmaPlot (version 14, Systat Software, Inc., San Jose, CA), models were fit individually for each treatment date of each year and the 11 variable units (Table 1). From the resulting nonlinear regressions, we calculated (Table 3): (i.) duration to the MSP using the MINIMIZE function in Wolfram Mathematica (version 11.2, Wolfram Research, Inc., Champaign, IL); (ii.) suppression magnitude at the MSP with the MINIMIZE function; and (iii.) total suppression phase duration with the FINDROOT function. Addionally, from the DAT regression of each treatment date, we calculated the 28-d average suppression magnitude by integrating the regression from 0 to 28 DAT with the NINTEGRATE function and then divided by 28. Finally, these calculated data points were subjected to ANOVA with SAS (version 9.4, SAS Institute, Inc., Cary, NC). Means were separated with Fisher's protected LSD with α=0.05 when appropriate.

Turfgrass Quality. Visual color ratings based on the NTEP scale were recorded weekly following the initial application (Morris and Shearman, 1998). A visual color rating of 6 was considered minimally acceptable and 9 was considered optimal turfgrass.

Additionally, NDVI was measured weekly, with the exception of May 2016. Research suggests that NDVI has a moderate correlation with turfgrass density and percent live cover (Bell et al., 2002; Sullivan et al., 2017). Turfgrass color ratings and NDVI readings were analyzed using the MIXED procedure in SAS. Means were separated with Fisher's protected LSD with $\alpha=0.05$.

Results and Discussion

Comparison of MSP Predictor Variables

Ability to predict the maximum suppression point (MSP) following a TE application is helpful to properly time reapplications (Kreuser et al., 2018; Reasor et al., 2018). We tested variable units that take environmental conditions into account to see if they predict the duration to the MSP more accurately than calendar days (DAT).

For duration to the MSP, ANOVA indicated that there was not a significant difference by year for all tested variable units, so data were pooled by year. As expected, duration to the MSP by DAT was significantly different across the treatment dates (Table 3). In contrast, duration to the MSP by GDD accumulation (all tested base temperatures), soil temperature accumulation, and GHI accumulation was not significant. Therefore, all data were pooled across treatment date then plotted by these variable units (i.e., all except DAT). Nonlinear regressions were fitted for these pooled data (by the same method as previously described for the individual treatment dates).

Based on pseudo-R^2 values, a five-parameter, amplitude-damped sine regression is the most appropriate model for ultradwarf bermudagrass suppression following a TE application:

$$\text{Relative clipping yield }(\mathrm{g\,g^{-1}}) = \mathrm{Y_{int}} + \text{Amplitude} \times \mathrm{e}^{\frac{-variable}{\text{Decay}}} \times \sin(2\pi\frac{variable}{\text{Period}} + \mathrm{C})$$

The damped amplitude is appropriate because minimal rebound occurred following the suppression phase, which is similar to other research on ultradwarf bermudagrass (Reasor et al., 2018). All parameters were significant ($P<0.05$) for the GDD (all tested base temperatures) and the soil temperature models (Table 2). However, amplitude was not significant for the GHI model ($P=0.185$) and the 2017 PAR model ($P=0.2322$).

The soil temperature accumulation model (pseudo-R^2: 0.573, SE: 0.193) predicts suppression slightly better than the GDD_0 model (pseudo-R^2: 0.564, SE: 0.195), but soil temperature is not practical for turfgrass managers. The GHI (pseudo-R^2: 0.537, SE: 0.201) and 2017 PAR (pseudo-R^2: 0.517, SE: 0.209) models are not as predictive as temperature; but PAR levels influence leaf elongation rates and could affect suppression duration (Stanford et al., 2005). More precise models might take a combination of these variables into account, but a simple GDD model is most practical for turfgrass managers (Kreuser et al., 2018). We conclude that GDD is better than DAT for predicting the MSP and, thus, also better for scheduling reapplications (Fig. 4). The following section will present data that support a GDD_0 reapplication schedule, as opposed to GDD_{10}.

GDD Base Temperature. The difficulty of creating GDD models is determining the base temperature of the turfgrass species, which should be set at the temperature where plant metabolism (and TA degradation) does not progress (McMaster and Wilhelm, 1997). The literature is inconclusive about the base temperature for warm-season turfgrasses. Unruh et al. (1996) indicates that the base temperature for ‘Midiron’ hybrid bermudagrass is not significantly different from 5ºC, with models predicting 0.3 and 3.1ºC. However, this cultivar may not be representative of ultradwarf bermudagrass, and the PAR was low during this experiment (400 µmol m^{-2} s^{-1}). Another experiment conducted in a naturally-lit chamber in Mississippi determined that the base temperature for TifEagle bermudagrass is 12.6ºC (Flournoy, 2017), but considering that both temperature and light intensity affect internode and leaf elongation (Berry and Björkman, 1980; Stanford et al., 2005), we should expect that the optimal base temperature is relative to other environmental conditions. That is, the optimal base temperature for MiniVerde in Auburn, AL (32.6099° N, 85.4808° W) may not apply to other latitudes. While a universal base temperature for ultradwarf bermudagrass may not exist, we can still compare GDD models to determine which base temperature best predicts suppression after a TE application.

Comparison of pseudo-R^2 values (Table 2; Fig. 3, inset) indicated that the GDD_0 model (pseudo-R^2: 0.564, SE: 0.195) provided a better fit than the GDD_{10} model (pseudo-R^2: 0.533, SE: 0.202). This superiority is most evident when comparing the predicted duration to the MSP at a temperature observed during the experiment. For example, at an average temperature of 15 and 19ºC, the GDD_{10} model predicts that the MSP will occur 30 and 16 DAT, respectively, which is similar to the GDD_{10} model of Reasor et al. (2018). In comparison, the GDD_0 model predicts that the MSP will occur at 16 and 13 DAT, respectively.

We can test these predictions at 19ºC—roughly the average temperature during the 1 May treatment. The unpooled DAT regressions for 1 May indicate that the MSP occurred at 12 DAT in 2016 (20.4ºC) and 13 DAT in 2017 (19.2ºC). These data suggest that the GDD_0 model correctly predicts the observed duration (~13 d) to the MSP at 19ºC, while the GDD_{10} does not (predicts 16 d). Additionally, while Beasley and Branham (2005) showed that TA half-life doubled when air temperature dropped from 30 to 18ºC in creeping bentgrass, there is no evidence to corroborate the GDD_{10} model prediction that a decrease from 19 to 15ºC would almost double duration to the MSP from 16 to 30 d.

These results do not indicate that 0ºC is the most appropriate base temperature for ultradwarf bermudagrass, but a GDD_0 model predicts TA degradation and suppression better than the other tested based temperatures in climates like Auburn, AL. Future research should quantify the half-life of TA in ultradwarf bermudagrass with methods similar to Beasley and Branham (2005) and test GDD base temperatures in other climates.

Turfgrass Growth

In the previous section, we determined that GDD_0 was the most predictive and practical variable unit for a TE reapplication schedule—we are also presenting the GDD_{10} data to aid in comparing results with previous and future research. The following sections will detail turfgrass growth following a TE application on ultradwarf bermudagrass, including: (i.) duration to the MSP, (ii.) suppression magnitude at the MSP, (iii.) total suppression phase duration, (iv.) average suppression magnitude over 28 d, and (v.) rebound. Also, combining these results with Reasor et al. (2018) and Brown et al. (Chapter 3) provides insight about TE rate effect. In the final section, these data will be used to support our recommended TE reapplication schedule.

Duration to the MSP. As expected, duration to the MSP was significantly different for DAT across treatment date, ranging from 7.5 to 12.6 d (Fig. 4, Table 3). On the other hand, duration to the MSP was not significantly different for the other tested variables, so data were pooled by treatment date. The pooled regression indicates that the MSP occurred at 262 GDD_0 [157 GDD_{10}] following a 0.044 kg ai ha^{-1} TE application (Fig. 2). In comparison with previous research, this is practically shorter (assuming daily GDD_{10} accumulation of 10 to 15 GDD_{10}) than the 177 GDD_{10} determined on MiniVerde with TE at 0.034 kg ai ha^{-1}, but similar to the 166 GDD_{10} of both 'TifEagle' and 'Champion' ultradwarf bermudagrass (Reasor et al., 2018). Interestingly, a pooled regression with only the 1 June to 1 August treatments (same time period as Reasor et al.) indicates the MSP occurred at 166 GDD_{10} (Fig. 3). In a separate experiment, we tested TE at 0.022 kg ai ha^{-1} and found the MSP occurred at 272 GDD_0 [158 GDD_{10}], practically equivalent to the higher rates (Chapter 3). This suggests that doubling the rate from 0.022 to 0.044 kg ai ha^{-1} is not an effective method for extending the duration to the MSP.

Suppression Magnitude at the MSP. For all tested variables, there was not a significant difference in suppression magnitude at the MSP by year or treatment date (Table 3). The suppression at the MSP averaged over the 1 June to 1 August treatments was 66%, which is similar to the 62% reported by Reasor et al. (2018). In contrast, the 1 May treatment only had 48% suppression at the MSP. This could be due to cooler temperatures that decreased uptake of TE and conversion to bioactive TA (Beasley et al., 2007). This theory is reasonable considering that reducing TE rate to 0.022 kg ai ha^{-1} resulted in only

38% suppression at the MSP, on average from June to August (Chapter 3). These results suggest that increasing rate from 0.022 to 0.044 kg ai ha^{-1} does affect suppression magnitude at the MSP (Fig. 3).

Total Suppression Phase Duration. For all tested variables (including DAT), the duration of the suppression phase was not significantly different by year or treatment date (Table 3). The pooled regression suggests that the suppression phase ended at 997 GDD$_0$ [650 GDD$_{10}$]. In contrast, our experiment with TE at 0.022 kg ai ha^{-1} indicates the suppression phase ended at 622 GDD$_0$ [385 GDD$_{10}$], which suggests that TE rate may affect suppression phase duration (Chapter 3). This rate effect is also noted in experiments on tall fescue (*Festuca arundinacea* Schreb.), perennial ryegrass (*Lolium perenne* L.), and 'Tifway' bermudagrass (Fagerness and Yelverton, 2000; King et al., 1997). On the other hand, this is inconsistent with results from creeping bentgrass and Kentucky bluegrass (*Poa pratensis* L.) that indicate increasing rate does not alter the suppression phase duration (Beasley et al., 2007; Kreuser and Soldat, 2011; Lickfeldt et al., 2001). This discrepancy could be due to species differences or indicate that increasing rate only extends the duration up to a certain point.

The Primo Maxx label claims that a label rate (0.026 kg ai ha^{-1}) application will suppress turfgrass growth by 50% for 28 d. The DAT regressions for TE at 0.044 kg ai ha^{-1} (1.7x rate) indicate that total suppression phase duration ranged from 25 to 45 d. This fluctuation appeared to have little correlation with temperature, as the suppression phase of both the 1 May (19.8 ºC) and 1 August (27.2 ºC) treatments lasted approximately 40 d.

Results with TE at 0.022 kg ai ha^{-1} indicate that the suppression phase lasted about 21 d (Chapter 3).

Average Suppression Magnitude over 28 Days. As for the average suppression magnitude over a 28-d period, it ranged from 10 to 36% with TE at 0.044 kg ai ha^{-1} (Table 3). In comparison, TE at 0.022 kg ai ha^{-1} only reduced growth 18% during the suppression phase (Chapter 3). These data suggest that, at the label rate, ultradwarf bermudagrass putting greens will not be suppressed by an average of 50% over a 28-d period. The TE rate required to suppress growth to this extent would likely produce unacceptable turfgrass phytotoxicity if applied in a single application; but properly-timed reapplications at low rates will hold suppression at greater than 50%, while also increasing turfgrass quality (Chapter 3).

Rebound. If the suppression phase ends, it is possible that a period of accelerated (rebound) growth will occur; however, this varies with cultivar, temperature, and TE rate (Beasley et al., 2007; Fagerness and Yelverton, 2000; Kreuser and Soldat, 2011). As with Reasor et al. (2018), a definite rebound phase did not occur following the suppression phase (Fig. 2), though we did note slightly enhanced growth for a few treatment dates (data not shown). Experiments on Tifway bermudagrass suggest that rebound may occur following a single TE application (Fagerness and Yelverton, 2000), but substantial rebound following a single application has not been reported for ultradwarf bermudagrass putting greens.

Significant rebound following repeated TE applications has been reported on ultradwarf bermudagrass (Chapter 3). Rebound is hypothesized to be the result of accumulated GA_{20} being quickly converted to GA_1 after TA is degraded, since the gibberellic acid biosynthesis pathway up to GA_{20} remains uninhibited (King et al., 1997). Two weeks after a second TE application on Kentucky bluegrass, GA_{20} concentration was increased 146%, in contrast to the 47% decrease of GA_1 (Tan and Qian, 2003). We suspect that dwarf cultivars may respond differently, and future research should examine the GA pathway of ultradwarf bermudagrass.

Properly-Timed Reapplications. In this final section, we will use the aforementioned data on turfgrass suppression to recommend an optimal TE reapplication schedule.

Previous research suggests that the suppression phase can be maintained by a reapplication frequency calculated by multiplying the GDD accumulation at the MSP by 1.3 (Kreuser et al., 2018; Reasor et al., 2018). With the MSP at 262 GDD_0, this method suggests approximately a 350-GDD_0 interval. However, it is possible that this will fail to maintain suppression at lower TE rates because our experiments indicate that TE rate *does* alter total suppression phase duration—but it does *not* alter duration to the MSP. We prefer another reapplication strategy for ultradwarf bermudagrass.

Given the greater efficacy of TE on ultradwarf bermudagrass (compared to creeping bentgrass), it is feasible to recommend a reapplication interval that provides consistent daily growth rates, i.e., minimal fluctuation in suppression magnitude from day-to-day. We hypothesize that consistent growth rates will result from reapplying TE at least 50 GDD_0 before the MSP occurs, which should allow time for TE uptake and conversion to TA before the MSP. The goal is to keep TA concentration constant in the plant and, thus, keep GA_1 concentration constant throughout the entire season. The MSP is an important point because, after the MSP, GA_1 concentration begins to increase as TA concentration decreases. We suspect that this reapplication method will maintain the turfgrass quality benefits of TE without causing unacceptable phytotoxicity.

With the MSP at 262 GDD_0, we recommend a 200-GDD_0 reapplication interval. This should apply to all commonly used TE rates (0.01 to 0.05 kg ai ha^{-1}) for ultradwarf bermudagrass putting greens since TE rate did not affect duration to the MSP. In a separate experiment, we tested a 200-GDD_0 interval with TE at 0.022 and 0.044 kg ai ha^{-1} and noted that this application schedule did yield consistent growth rates and improved turfgrass quality (Chapter 3).

Turfgrass Quality. Color ratings are presented by weeks after initial treatment (WAIT) since it was a significant factor (Table 4). For all treatment dates, turfgrass color decreased following the TE application but was never lower than minimally acceptable after 2 WAIT. By 3 WAIT, color ratings for all treatment dates were not significantly different from the non-treated. Relative NDVI readings followed a similar pattern with lowest readings at 1 WAIT, and by 3 WAIT, relative NDVI for all treatment dates were not significantly different from the control (data not shown). This is consistent with previous experiments that reported initial phytotoxicity until 2 or 3 WAIT followed by quality similar to the control (McCullough et al., 2007; McCullough et al., 2006a).

Other management factors may influence turfgrass quality following TE applications. Repeated applications of TE decreased TifEagle quality for much of the experiment with a N rate below 6 kg ha^{-1} 7 d^{-1} (Unruh et al., 2005). McCullough et al. (2006a) also indicated that N rate (6, 12, 18, and 24 kg ha^{-1} 7 d^{-1}) had a significant interaction with TE (0.05 kg ai ha^{-1}) for turfgrass color.

Conclusion

Previous research indicates that GDD schedules are superior to calendar-day schedules for TE reapplications on cool-season turfgrasses (Kreuser et al., 2018; Kreuser and Soldat, 2011). Taking temperature into account more accurately predicts the degradation of TA, but other factors also influence TA degradation and GA_1 production, such as cultivar, light intensity, moisture, and environmental stresses. Interestingly, previous research does not suggest that there is a N rate and TE interaction for average suppression magnitude (McCullough et al., 2006a); and our results with a N rate of 12 kg ha^{-1} applied weekly compared to the 10 kg ha^{-1} applied biweekly of Reasor et al. (2018) also suggests that N rate has a minimal impact on both duration to the MSP and suppression magnitude at the MSP (relative to the control). Other factors may influence suppression, but a GDD schedule will be advantageous for turfgrass managers if it provides a practical benefit, like reducing total applications or minimizing phytotoxicity.

Based on the MSP at 262 GDD_0, a 200-GDD_0 reapplication interval should provide consistent turfgrass growth rates and maintain quality benefits. In a separate experiment, we tested 4 GDD_0 intervals (100, 200, 400, 600) and 2 rates (0.022 and 0.044 kg ai ha^{-1}) for an entire growing season (Chapter 3). Future research should test a GDD_0 schedule in the transition zone.

Table 1. Parameters for the GDD_0, GDD_{10}, and DAT models presented by treatment. Data were pooled by year. When the following parameters are entered into the five-parameter, amplitude-damped equation, the resulting regression depicts turfgrass clipping yield following a TE application on that treatment date in terms of the predictor variable (GDD_0, GDD_{10}, or DAT).

		Regression Parameters[†]				
Model[‡]	Treatment Date	Y_{int}	a	b	c	d
GDD_0	1 May	0.960*	1.091	1218*	2.822*	350.7*
	15 May	1.226*	211.1	92900	3.139	199.5*
	1 June	1.217*	431.2	245800	3.141	237.4*
	15 June	0.834*	1.307	1349*	2.822*	344.1*
	1 July	1.015*	3.605	1924	2.814*	239.0*
	15 July	0.920*	3.330	1640 *	2.877*	222.6*
	1 August	0.964*	112.3	22020	3.111	116.5
GDD_{10}	1 May	0.977*	1.231*	704.3*	2.932*	170.6*
	15 May	1.222*	249.3	67620	3.140	118.6*
	1 June	1.225*	482.0	178500	3.141	149.7*
	15 June	0.835*	1.335	846.4*	2.831*	210.6*
	1 July	1.009*	3.568	1202	2.796*	152.2*
	15 July	0.915*	3.476	1053*	2.868*	138.4*
	1 August	0.962*	124.6	15490	3.114	73.72
DAT	1 May	0.935*	0.863	51.53*	2.790*	18.89*
	15 May	1.230*	275.7	5348	3.141	8.091*
	1 June	1.195*	382.4	8566	3.141	8.690*
	15 June	0.834*	1.181	50.64	2.936*	13.26*
	1 July	1.024*	3.175	71.03	2.928*	8.743*
	15 July	0.937*	3.125	61.98*	3.001*	7.959*
	1 August	0.970*	81.33	770.7	3.118	4.347*

[†] Parameters with an "*" are statistically significant ($P<0.05$).

[‡] Data were fit to a five-parameter, amplitude-damped sine regression: $y=y_{int}+a*\exp(-x/d)*\sin(2\pi*x/b+c)$. Where "a" is the amplitude, "b" is the period, "d" is the decay, "x" is GDD accumulation, and "y" is relative clipping yield.

Table 2. Parameters for the tested environmental variable models. Data were pooled by year and treatment date. When the following parameters are entered into the five-parameter, amplitude-damped equation, the resulting regression predicts turfgrass clipping yield following a TE application in terms of the specified predictor variable.

Pooled Model[†]	Parameters[‡]					Psuedo-R^2	SE
	y_{int}	a	b	c	d		
Soil Temp[§]	0.994*	2.840*	1791*	2.907*	242.9*	0.573	0.193
GDD$_0$	0.988*	2.624*	1722*	2.915*	244.9*	0.564	0.195
GDD$_2$	0.984*	2.556*	1571*	2.915*	227.4*	0.561	0.196
GDD$_4$	0.980*	2.473*	1421*	2.916*	210.3*	0.557	0.197
GDD$_6$	0.975*	2.368*	1273*	2.920*	193.5*	0.554	0.198
GDD$_8$	0.969*	2.234*	1128*	2.927*	177.4*	0.544	0.200
GDD$_{10}$	0.962*	2.061*	986.7*	2.943*	162.3*	0.533	0.202
GDD$_{12}$	0.953*	1.837*	851.3*	2.972*	148.7*	0.516	0.206
GHI	1.047*	3.241	460400*	3.041*	48590*	0.537	0.201
PAR (2017)	1.018*	2.613	3601*	3.046*	449.0*	0.517	0.209

[†] Data were fit to a five-parameter, amplitude-damped sine regression:
$y = y_{int} + a*\exp(-x/d)*\sin(2\pi*x/b+c)$. Where “a” is the amplitude, “b” is the period, “d” is the decay, “x” is GDD accumulation, and “y” is relative clipping yield.

[‡] Parameters with an “*” are statistically significant ($P<0.05$).

[§] Soil temperature was calculated at 2.5 cm. The subscript number following “GDD” indicates the model base temperature, i.e., “GDD$_0$” is growing degree-days with a base temperature of 0 degree Celsius. “GHI” is global horizontal irradiance, and “PAR” is photosynthetically active radiation.

Table 3. Turfgrass suppression duration and magnitude after a single TE application. Treatment dates were not significantly different by year, so data were pooled by year. All results calculated from regressions of each treatment date.

		Turfgrass Suppression[†]						
Treatment Date	Average Temperature[‡]	Duration to MSP			Duration of Suppression Phase		Magnitude of Suppression	
	ºC	DAT	GDD$_0$	GDD$_{10}$	DAT	GDD$_0$	at MSP	28-d Average
1 May	19.8	12.6 a	271	135	38.8	907	48%	15%
15 May	23.0	9.0 bc	232	133	29.9	759	62%	27%
1 June	25.4	9.1 bc	261	160	44.7	1255	68%	36%
15 June	25.7	10.2 abc	290	178	26.3	954	65%	34%
1 July	27.8	10.3 ab	311	200	30.2	871	63%	20%
15 July	28.1	8.3 bc	267	174	24.8	1379	78%	36%
1 August	27.2	7.5 c	232	147	39.5	1165	58%	10%
LSD		2.74	NS	NS	NS	NS	NS	NS

[†] Column means not sharing any letter are significantly different according to Fisher's protected LSD (α=0.05).

[‡] The average temperature was calculated from daily averages beginning on the application date and ending at the MSP.

Table 4. Visual Turfgrass color rating separated by year, treatment date, and rating date.

Visual Turfgrass Color Rating[†] (2016)						
	WAIT[‡§]					
Treatment Date	1	2	3	4	5	6
1 May	5.75 c	6.00 c	6.75 c	7.00 c	7.00 b	7.00 a
15 May	6.75 a	7.00 a	7.00 bc	7.00 c	7.00 b	7.00 a
1 June	6.75 a	6.50 b	7.00 bc	7.00 c	7.25 b	7.00 a
15 June	6.00 bc	6.00 c	7.25 b	7.00 c	7.00 b	7.00 a
1 July	6.00 bc	7.00 a	7.00 bc	7.00 c	7.00 b	7.00 a
15 July	5.00 d	5.00 d	6.75 c	8.00 a	7.75 a	7.00 a
1 August	6.25 b	6.00 c	8.00 a	7.50 b	7.00 b	7.00 a
Non-treated	7.00 a	7.00 a	7.00 bc	7.00 c	7.00 b	7.00 a

Visual Turfgrass Color Rating[†] (2017)						
	WAIT[‡§]					
Treatment Date	1	2	3	4	5	6
1 May	6.00 c	5.00 d	8.00 a	8.00 a	8.00 a	7.00 b
15 May	5.00 d	6.00 c	7.75 a	8.00 a	7.00 b	7.00 b
1 June	7.25 a	5.00 d	8.00 a	8.00 a	8.00 a	7.75 a
15 June	7.00 b	6.00 c	8.00 a	8.00 a	8.00 a	7.00 b
1 July	6.00 c	6.00 c	7.00 b	7.00 b	7.00 b	7.00 b
15 July	5.00 d	8.00 a	8.00 a	7.00 b	7.00 b	7.00 b
1 August	5.00 d	7.00 b	7.00 b	7.00 b	7.00 b	7.00 b
Non-treated	7.00 b	7.00 b	7.00 b	7.00 b	7.00 b	7.00 b

[†] A color rating of 6 was considered minimally acceptable and 9 was considered optimal turfgrass.

[‡] Weeks after initial treatment (WAIT).

[§] Column means not sharing any letter are significantly different according to Fisher's protected LSD (α=0.05).

Figure 1. Average daily temperature at the Sports Surface Field Laboratory in Auburn, AL during 2016 and 2017. The average temperature during the experiment was 26.4ºC in 2016 and 25.3ºC in 2017. The 25-yr average is from a weather station about 2 km northeast of the experiment location.

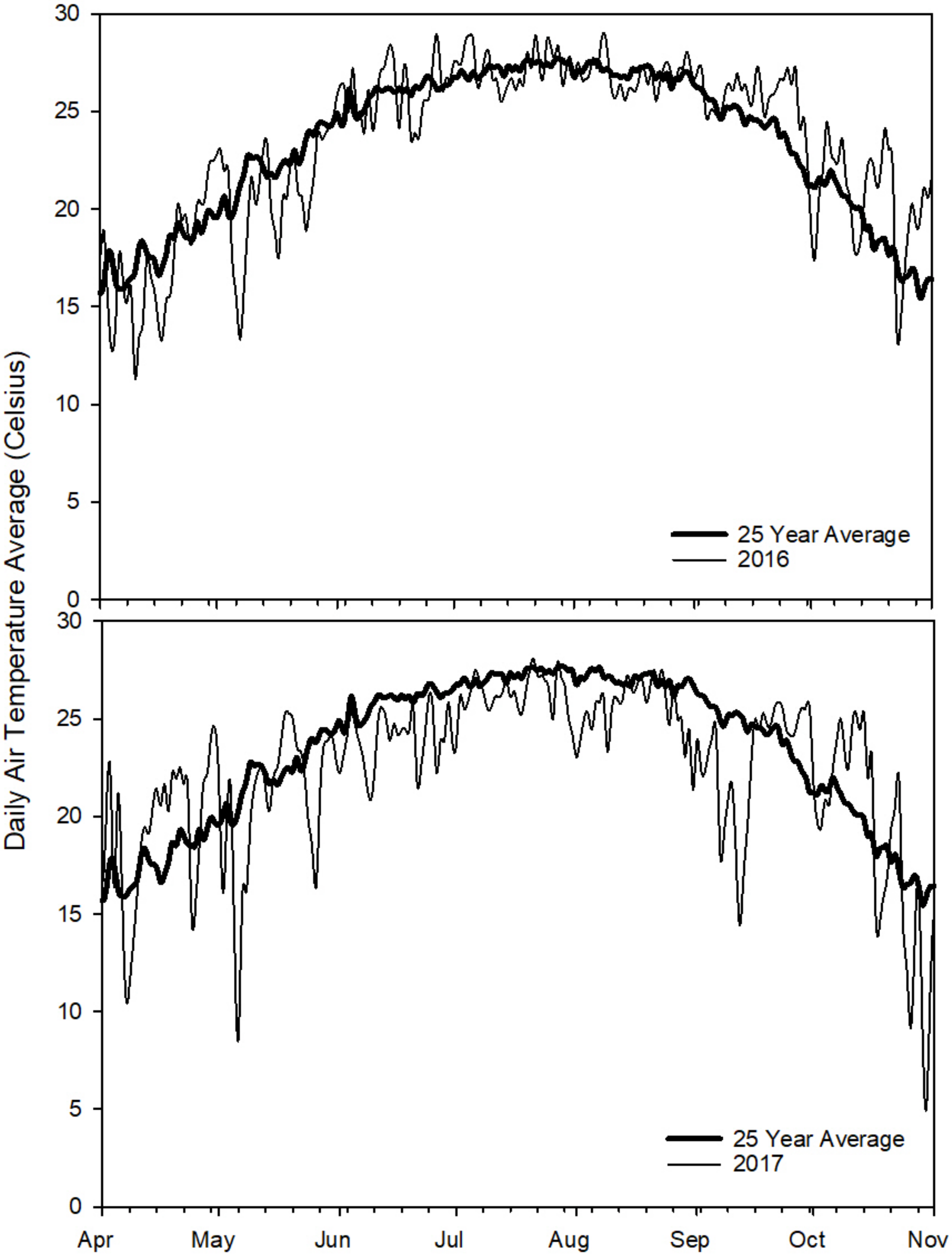


Figure 2. Relative clipping yield plotted by GDD_0 after the TE application. This includes data pooled by year and treatment date. The solid line represents the five-parameter, amplitude-damped sine regression, which was chosen because it resulted in the largest pseudo-R^2 value (0.564, SE: 0.195; see Fig. 3). All parameters are significant ($P<0.05$). The data points are from the individual plots on each collection date during the experiment. A definite rebound phase was not indicated by the model, but we did note accelerated growth for some treatment dates.

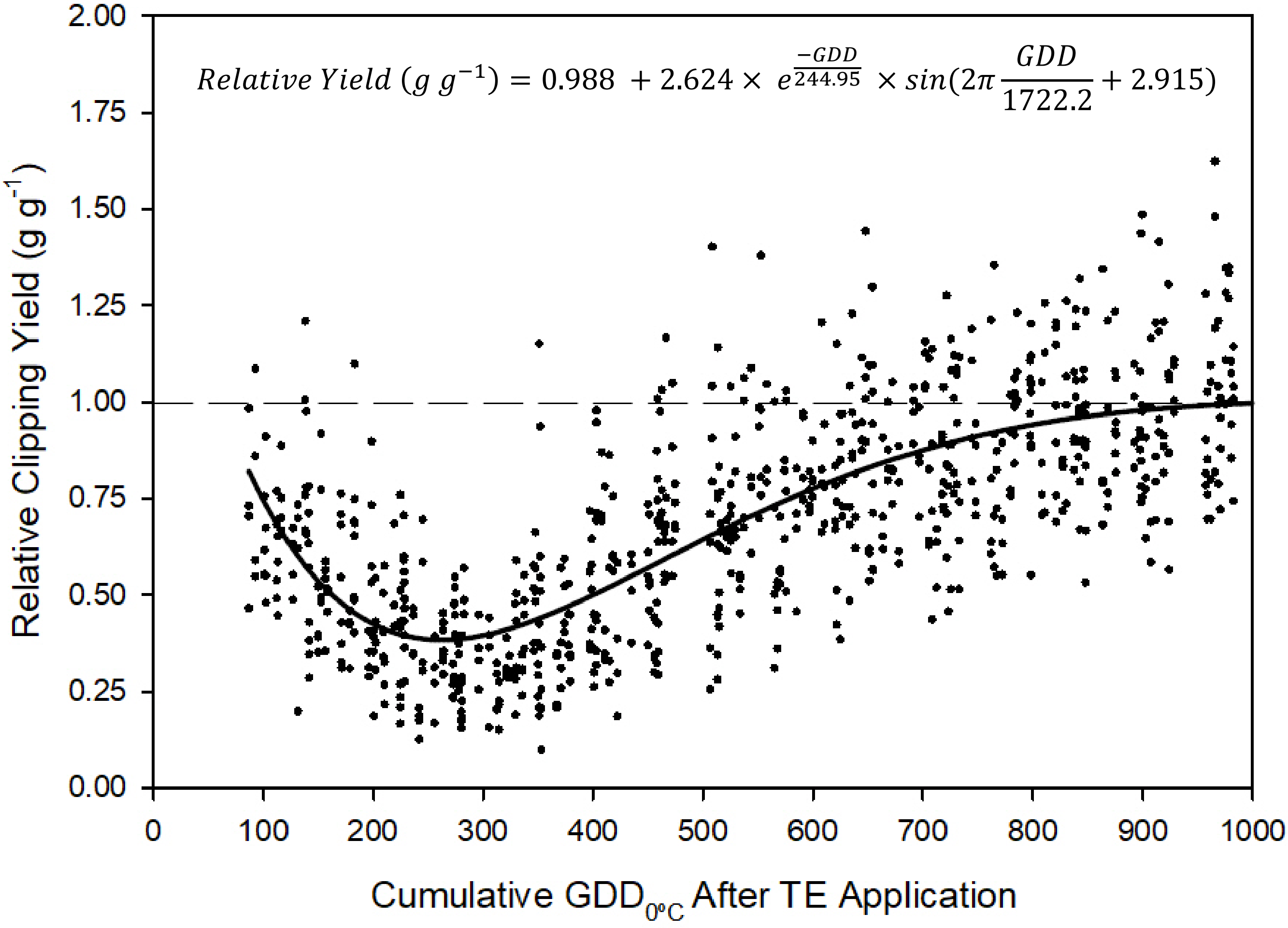


Figure 3. Relative clipping yield plotted by GDD_{10} after the TE application. These models only include the 1 June to 1 August treatments for comparison to Reasor et al. (2018). The 0.044 kg ai ha^{-1} model is from this experiment. The 0.022 kg ai ha^{-1} model is from Chapter 3. The 0.034 kg ai ha^{-1} model is from the equation reported for MiniVerde in Reasor et al. (2018), which took place from June to August. The dot on each regression line represents the MSP. The MSP occurred at 165, 177, and 166 GDD_{10} for 0.022, 0.034, and 0.044 kg ai ha^{-1}, respectively. Reasor et al. (2018) also reported that the MSP occurred at 166 GDD_{10} on TifEagle and Champion at 0.034 kg ai ha^{-1} (not shown). We recommend the GDD_0 model because it resulted in the largest pseudo-R^2 (inset; Table 2), and we only present these GDD_{10} models for comparison to previous and future research.

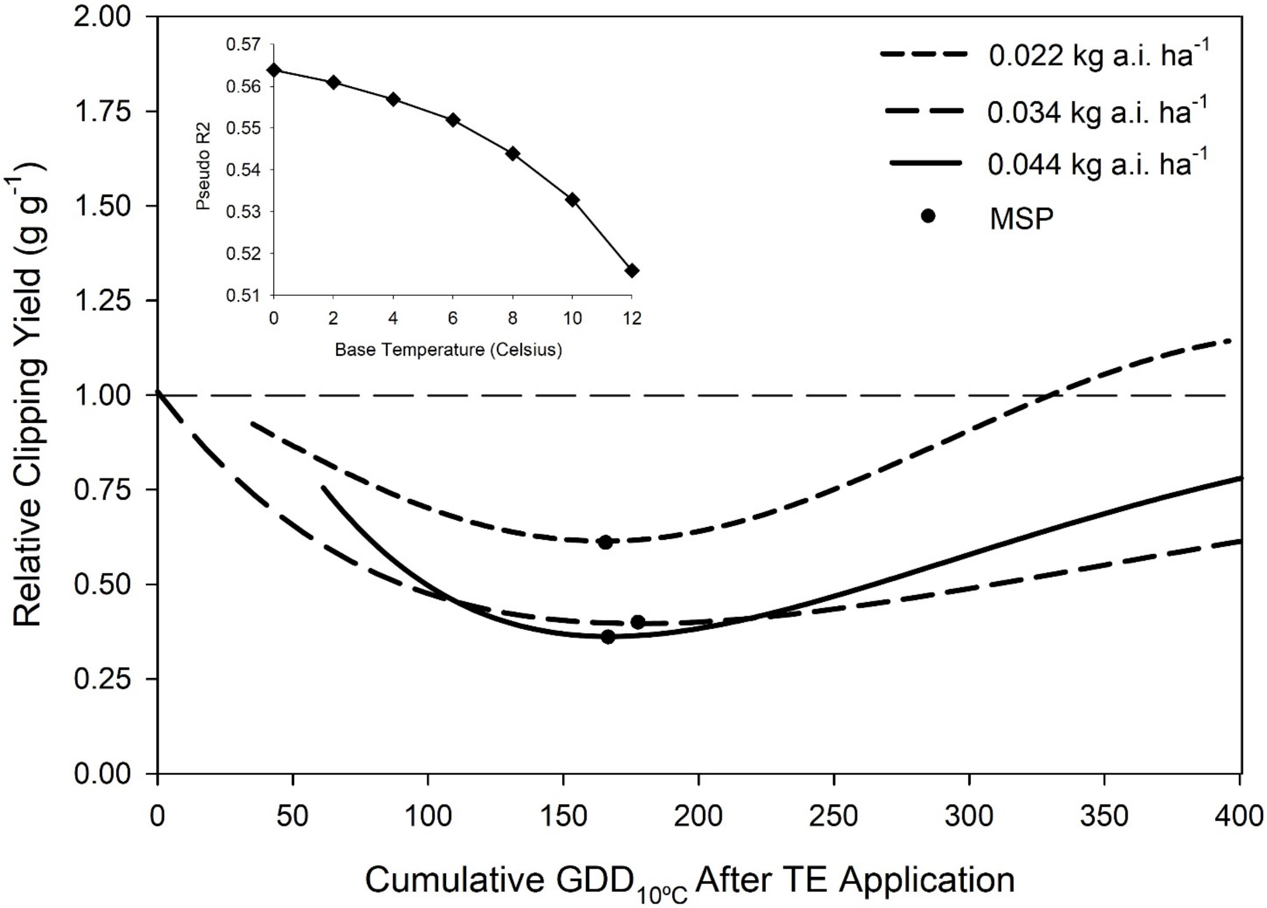
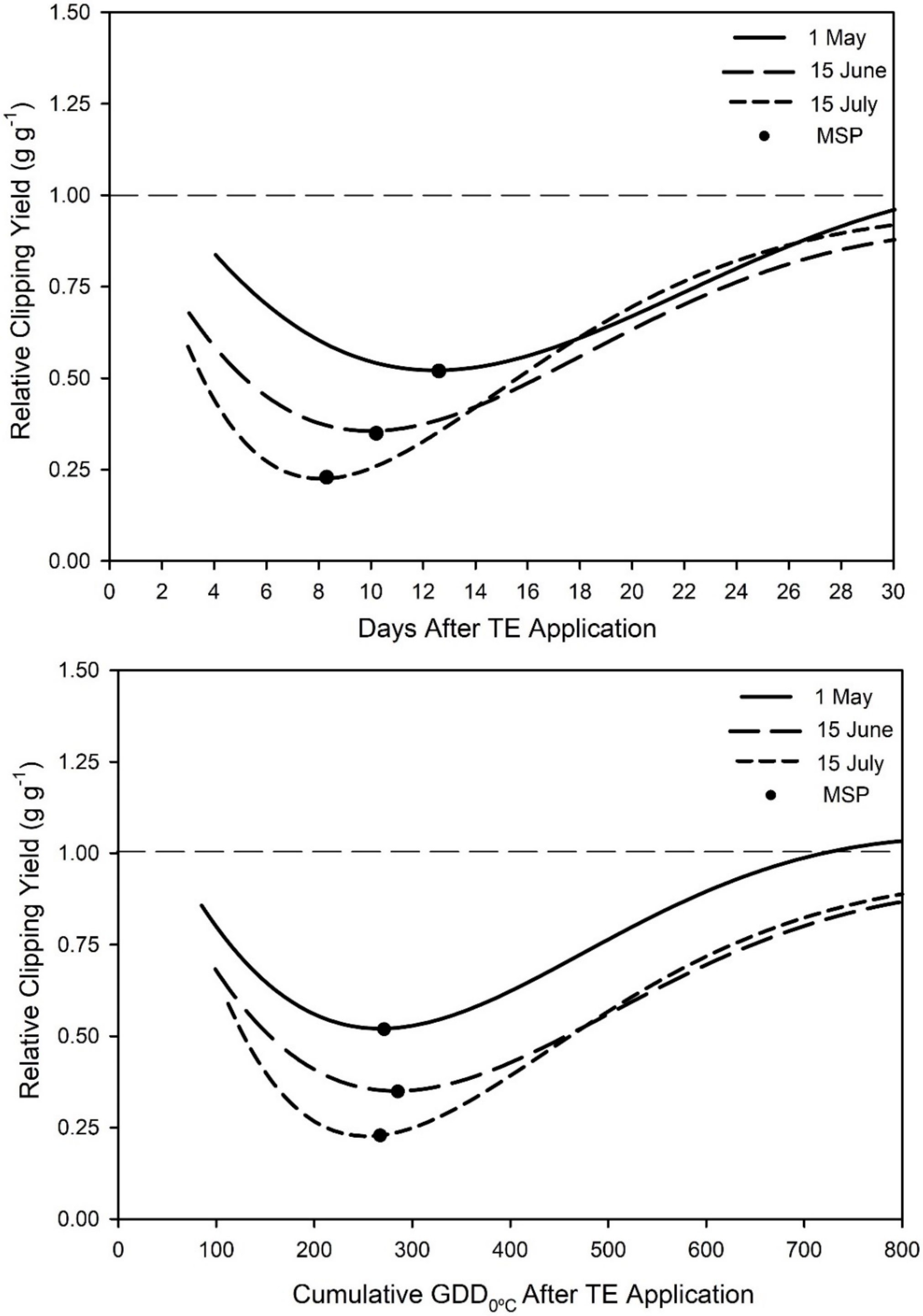


Figure 4. Comparison of relative clipping yield plotted by DAT (top) and GDD_0 (bottom) after a single TE application. The lines are the regressions for the 1 May, 15 June, and 15 July treatments (pooled by year). The dots represent the MSP, which occurred at 12.6, 10.2, and 8.3 DAT and at 271, 290, and 267 GDD_0 for 1 May, 15 June, and 15 July, respectively. The duration to the MSP by GDD_0 is practically similar for these treatment dates (assuming daily accumulation between 20 and 30 GDD_0). Suppression magnitude at the MSP increased as temperatures increased from May to July (Table 3).



Chapter 3. Testing a Growing Degree-Day Reapplication Schedule for Trinexapac-ethyl on Ultradwarf Bermudagrass Putting Greens

Introduction

Turfgrass managers apply trinexapac-ethyl (TE) to ultradwarf bermudagrass [*Cynodon dactylon* (L.) Pers. x *C. transvaalensis* Burtt-Davy] putting greens primarily to sustain ball roll and improve surface quality (Baldwin et al., 2009; Fagerness et al., 2000; King et al., 1997; Kreuser and Soldat, 2012; McCarty et al., 2011; McCullough et al., 2005a). These enhancements are possible because TE suppresses vertical turfgrass growth, so failure to maintain suppression will result in a loss of these benefits. The problem for turfgrass managers is determining when to reapply since suppression duration varies with environmental conditions, like temperature (Kreuser and Soldat, 2011). This renders a calendar-based reapplication schedule ineffective.

Recent research suggests that growing degree-days (GDD), a unit that accounts for temperature, more accurately predicts suppression duration after a TE application. A 200-GDD_0 (GDD with a base temperature of 0°C) will maintain suppression of creeping bentgrass (*Agrostis stolonifera* L.) putting greens all season (Kreuser and Soldat, 2011). As for ultradwarf bermudagrass putting greens, Reasor et al. (2018) recommended that TE be reapplied to 'MiniVerde' ultradwarf bermudagrass every 230 GDD_{10} (GDD with base temperature of 10ºC). They calculated this interval by multiplying the total GDD_{10} accumulation at the maximum suppression point (MSP) by 1.3, which was shown to be an effective method for creeping bentgrass (Kreuser et al., 2018; Kreuser and Soldat, 2011). Knowing when the MSP will occur is important for making properly-timed reapplications. After the MSP, the GA_1 concentration, a hormone the signals for cell elongation, begins returning to the non-treated level—and turfgrass growth rate follows in synchrony. Reapplying before the MSP may prevent this return to the non-treated level.

In an experiment on a MiniVerde putting green, we determined that GDD_0 is better than calendar days and GDD_{10} for predicting the MSP (Chapter 2). According to the GDD_0 model, the MSP occurs at 262 GDD_0 after a TE (0.044 kg ai ha^{-1}) application. We theorized that a reapplication frequency occurring at least 50 GDD_0 before the MSP—allowing time for TE uptake and conversion to trinexapac acid, the bioactive form, before the MSP—would result in a consistent growth rate from day-to-day and enhanced turfgrass quality.

The objective of this experiment was to test a GDD_0 reapplication schedule for an entire growing season on a MiniVerde putting green. We included 4 GDD_0 intervals (100, 200, 400, and 600) and 2 TE rates (0.022 and 0.044 kg ai ha^{-1}). We hypothesized that the 100- and 200-GDD_0 intervals would occur frequently enough to maintain consistent daily growth and enhance turfgrass quality, while the 400- and 600-GDD_0 intervals would result in fluctuating suppression magnitude.

Materials and Methods

A field experiment was conducted on a MiniVerde bermudagrass putting green at the Sports Surface Field Laboratory in Auburn, AL during 2016 and repeated in 2017. The putting green was constructed in 1994 according to United States Golf Association specifications (USGA, 1993) and sprigged with MiniVerde in April 2004. On 1 April 2016, 9 September 2016, and 2 March 2017, the green was hollow-tine aerated and topdressed heavily. During the experiment, plots were not topdressed or cultivated. Following green-up in April, the plots were fertilized with liquid urea at 12.2 kg N ha^{-1} wk^{-1}. Phosphorus and potassium were added based on soil test results. Preventative fungicides to control mini-ring (*Rhizoctonia zeae*) and dollar spot (*Sclerotinia homoeocarpa*) were applied beginning in May. This included azoxystrobin, chlorothalonil, and mancozeb, which are not known to regulate plant growth. Plots were irrigated daily at approximately 80% of the evapotranspiration rate.

Treatments included a low and high TE rate (0.022 and 0.044 kg ai ha^{-1})—both commonly used on ultradwarf putting greens—and 4 GDD_0 reapplication intervals (100, 200, 400, and 600). These application intervals began on the first week of May and ended at the beginning of August (2 May – 8 August 2016, 1 May –7 August 2017). Applications were made with a CO_2 sprayer calibrated to deliver 375 L ha^{-1}. Irrigation was withheld for at least 1 hr following TE applications. Treatments were arranged in a randomized complete block design with four replicates on 1.5 by 1.5 m plots, and a non-treated control was included in each replication.

Weather Data. A weather station (WMR300, Oregon Scientific, Tualatin, OR) positioned 1.5 m above the experiment area recorded air temperature and rain. The daily high and low air temperature were recorded in Celsius for each 24 hr period beginning at midnight (Fig. 5). Daily GDD was calculated with the equation:

$$\text{Daily GDD} = \frac{T_{\text{high}} + T_{\text{low}}}{2} - T_{\text{base}}$$

Where if $[(T_{high}+T_{low})/2]<T_{base}$, then the GDD for that day is set to 0, which prevents negative GDD accumulation (McMaster and Wilhelm, 1997). Daily GDD was calculated with a 0°C base temperature (Chapter 2). Total GDD accumulation is the sum of daily GDD_0 beginning on the application date. Sequential applications for each interval were made on the day after the GDD_0 threshold was crossed.

Turfgrass Growth. Growth was approximated by collecting clippings three days (1100 ± 1 hr) per week with a Jacobsen walking greens mower (Greens King 522, Jacobsen, Augusta, GA) set at 3.4 mm. Clipping collections began 2 d after the first application and ended 25 d after the final application. Clippings were not collected on 21 June 2017 due to a tropical storm.

Before collection, alleys were mowed down the edges of plots (perpendicular to the collection mowing direction) to ensure a total collection area of 0.535 m^2 per plot. Collection mowing direction was altered by 180-degrees each collection date to reduce grain formation. For collection, a single pass was mowed across the center of each plot, then the clippings were removed from the clipping basket using a handheld vacuum (PHV1810, Black & Decker). Clippings were emptied out of the vacuum into a bag, then oven-dried at 60ºC for at least 48 hrs before weighing to the nearest centigram. Following the collection, the entire experiment area was mowed at 3.4 mm with the same Jacobsen greens mower, and the area was mowed only on collection dates.

Relative clipping yield (g g^{-1}) was calculated by dividing the weight of the treated by the non-treated within each replication. Daily growth (g m^{-2} d^{-1}) was calculated by dividing the weight of each treated plot by the number of days since the previous collection date to obtain an approximate daily growth rate. To more meaningfully analyze the effect of rate and interval on vertical growth, clipping collection dates were separated into two sections: "suppression" and "rebound." The data presented in the suppression section includes collection dates beginning at the maximum suppression point (MSP) following the initial application (18 May 2016 and 17 May 2017) through the collection date on which the final TE application was made (8 August 2016 and 7 August 2017). This section is representative of a season-long TE reapplication schedule. The rebound section includes data from the collection dates that followed the final TE application (17 August – 2 September 2016, and 16 August – 1 September 2017), which will provide insight about the potential effects of ending TE applications during the growing season.

Statistical Analysis. After separating collection dates into the suppression section or rebound section, data were subjected to repeated-measures analysis with the MIXED procedure in SAS (version 9.4, SAS Institute, Inc., Cary, NC), and means were separated with Fisher's protected LSD (α=0.05) when appropriate, such as comparing treatments within a collection date.

In a separate experiment, we determined that GDD_0 was the best predictor of the MSP on ultradwarf bermudagrass for TE at 0.044 kg ai ha^{-1} (Chapter 2). To compare these results with a lower TE rate, we pooled data from the 600-GDD_0 interval with TE at 0.022 kg ai ha^{-1}across year and collection date, and we plotted relative clipping yield by total GDD_0 accumulation since the most recent TE application—similar to the method of Kreuser and Soldat (2011). These pooled data were then fit to a five-parameter, amplitude-damped sine regression in SigmaPlot (version 14, Systat Software, Inc., San Jose, CA)—as described in Chapter 2. We followed the same procedure to create a GDD_{10} model for comparison to previous and future research. From the resulting regressions, we calculated using Wolfram Mathematica (version 11.2, Wolfram Research, Inc., Champaign, IL): (i.) duration to the MSP, (ii.) suppression magnitude at the MSP, (iii.) total suppression phase duration, and (iv.) average suppression magnitude over the suppression phase. We only pooled data from the 600-GDD_0 interval because it was the only treatment with the low TE rate that consistently resulted in the suppression phase followed by a return to a non-treated growth rate before the sequential application (Fig. 7.2).

Turfgrass Quality. Visual color ratings based on the NTEP scale were recorded weekly following the initial application (Morris and Shearman, 1998). A visual color rating of 6 was considered minimally acceptable and 9 was considered optimal turfgrass. Additionally, NDVI was measured weekly, except for May 2016. Research suggests that NDVI has a moderate correlation with turfgrass density and percent live cover (Bell et al., 2002; Sullivan et al., 2017). Turfgrass color ratings and NDVI readings were analyzed using the MIXED procedure in SAS. Means were separated with Fisher's protected LSD (α=0.05).

Results and Discussion

Before analyzing the suppression section and the rebound section, we will compare the GDD_0 model for TE at 0.022 kg ai ha^{-1} with the GDD_0 model for TE at 0.044 kg ai ha^{-1} from Chapter 2.

GDD Model Comparison

Ability to predict the maximum suppression point (MSP) following a TE application allows for more precise reapplications that will maintain the suppression phase and the associated benefits (Kreuser et al., 2018; Kreuser and Soldat, 2011; Reasor et al., 2018). For TE at 0.044 kg ai ha^{-1}, we determined that GDD_0 is the best predictor of the MSP for ultradwarf putting greens (Chapter 2). We pooled data from the 600-GDD_0 interval with TE at 0.022 kg ai ha^{-1} across year (not significant) and collection date to create a GDD_0 model for TE at 0.022 kg ai ha^{-1}.

The resulting model indicates that the MSP occurs at 272 GDD_0 [158 GDD_{10}]. Assuming a daily accumulation of 20 to 30 GDD_0 [10 to 20 GDD_{10}], this is very similar to the 262 GDD_0 [157 GDD_{10}] determined with TE at 0.044 kg ai ha^{-1} (Fig. 6). In contrast, suppression magnitude at the MSP was 38% for TE at 0.022 kg ai ha^{-1}, while it was 61% for TE at 0.044 kg ai ha^{-1}. This reduced suppression at the MSP resulted in a shorter total suppression phase duration of only 622 GDD_0 [385 GDD_{10}], compared to the 997 GDD_0 [650 GDD_{10}] of TE at 0.044 kg ai ha^{-1}. Additionally, the average suppression magnitude over the suppression phase was only 18%, compared to 25% for the higher rate. These results suggest that TE rate does not affect duration to the MSP (at least at typical TE rates), but rate does affect suppression magnitude at the MSP and total suppression phase duration.

We conclude that, at the label rate (0.026 kg ai ha^{-1}), a single TE application will not suppress ultradwarf bermudagrass putting greens by an average of 50% over a 28-d period, as claimed on the Primo Maxx label. The TE rate required to suppress growth to this extent would likely produce unacceptable phytotoxicity if applied in a single application. However, the following section will show that properly-timed reapplications can maintain greater than 50% suppression and increase turfgrass quality.

Suppression

Year was significant for both relative clipping yield (g g^{-1}) and daily growth (g m^{-2} d^{-1}), so results are presented by year. For both years, interval, rate, and the interval by rate interaction were significant, so treatments are presented separately. Also, collection date and collection date by interval (but not rate) were significant, so data are presented by collection date (Fig. 7). Turfgrass color ratings were significantly different by year, rate, interval, and rate by interval, so these data are also presented separately (Table 6).

Before examining the suppression data, one flaw in the methodology of turfgrass growth regulator research should be noted. As the season progressed, the clipping production of treated plots gradually increased relative to the non-treated, i.e., the relative clipping yield increased (Fig. 7). This could suggest that suppression magnitude was gradually decreasing because TE was less efficacious as temperature increased; however, before drawing this conclusion, it is important to differentiate “relative clipping yield” from “suppression magnitude.” Suppression magnitude refers to turfgrass growth rate on an *individual tiller basis* (relative to the non-treated growth rate per tiller), while relative clipping yield refers to the *total clipping production* of the plot (relative to the clipping production of the non-treated plot). In most research, relative clipping yield is measured as a proxy for suppression magnitude, but this is flawed in that it assumes an equal tiller density for the treated and non-treated plots. Previous research suggests that repeated TE applications may significantly increase tiller density (Ervin and Koski, 1998; Ervin and Koski, 2001). While density was not quantified in this experiment, NDVI is moderately correlated with turfgrass density (Bell et al., 2002), and it increased as the season progressed (data not shown). Therefore, we suggest that this gradual increase in relative clipping yield during the season is caused, at least partially, by the increased tiller density of the treated plots, compared to the non-treated plots (Lickfeldt et al., 2001). In fact, our TE experiment that did not include repeated applications indicated that relative clipping yield at the MSP decreased as temperature increased, which could suggest that suppression magnitude was greater at higher temperatures (Chapter 2). In accordance with the previous literature, we will refer to “relative clipping yield” as “suppression magnitude,” but we are careful to avoid drawing conclusions about the increased relative clipping yield that occurred as the season progressed.

TE Rate. Before examining suppression by interval and collection date, it is interesting to note that the average suppression magnitude for treatments with a similar total TE rate (sum of all TE applied during the experiment) was almost equal (Table 5). For example, total applied TE was 0.29 and 0.31 kg ai ha^{-1} for the 200 GDD$_0$ at 0.022 kg ai ha^{-1} and 400 GDD$_0$ at 0.044 kg ai ha^{-1}, respectively. For these treatments, the average suppression magnitude was not significantly different (55 vs. 54% in 2016 and 41 vs. 43% in 2017); but when analyzed by collection date, the suppression magnitude was significantly different on 12 collection dates in 2016 and 7 in 2017. Similarly, total applied TE was 0.55 and 0.57 kg ai ha^{-1} for the 100 GDD$_0$ at 0.022 kg ai ha^{-1} and 200 GDD$_0$ at 0.044 kg ai ha^{-1}, respectively. The average suppression was not significantly different in 2016 (80 vs. 76%) and similar, though significantly different, in 2017 (75% vs. 68%). Daily suppression was not significantly different for these treatments on any collection date in 2016, though it was for 2 in 2017.

These data suggest that, while the average suppression may be similar, the daily fluctuation in growth was greater for the less frequent intervals, especially the 400-GDD$_0$ that occurred after the MSP. Fluctuation in daily growth is undesirable for turfgrass managers, and the following sub-sections demonstrate that more frequent TE applications will reduce fluctuation and maintain turfgrass quality.

100 GDD$_0$. In both years, the 100 GDD$_0$ interval at both rates was significantly less than the non-treated on every collection date during the suppression period. The high and low rate were significantly different on 9 collection dates in 2016 and only 1 in 2017 (Fig. 7.1). The average suppression for the low rate was 80% in 2016 and 75% in 2017, compared to 92% in 2016 and 82% in 2017 for the high rate (Table 5). Both rates resulted in very consistent daily growth, but the high rate every 100 GDD$_0$ is unacceptable for high-quality turfgrass because it resulted in color ratings that remained significantly lower than the non-treated until 67 days after the initial treatment (DAIT) in 2016 and 53 DAIT in 2017—though the ratings were considered minimally acceptable by 37 and 30 DAIT in 2016 and 2017, respectively (Table 6). On the other hand, the low rate was equal to or significantly better than the non-treated by 21 DAIT in both years, and color was never unacceptable in 2016. In climates like Auburn, AL, this interval occurs approximately every 4 to 5 d during the growing season, so it would not be practical for most turfgrass managers to reapply this often.

200 GDD_0. For the low rate, growth was significantly different from the non-treated on every collection date in 2016 and on all but 4 in 2017. The average suppression was 55 and 41% for 2016 and 2017, respectively (Table 5). The high rate was significantly less than the non-treated on every collection date in both 2016 and 2017, and the average suppression was 76% in 2016 and 68% in 2017. It was significantly different from the low rate on every collection date, except for 7 collections in 2016 and 2 in 2017 (Fig. 7.1).

We designed this experiment to determine if a TE reapplication frequency that occurs before the MSP will result in consistent daily growth (minimal suppression magnitude fluctuation across collection dates). Based on our other experiment (Chapter 2), the 200-GDD_0 interval should occur before the MSP. Supporting our hypothesis, the high rate resulted in daily growth ranging from 0.20 to 2.3 g m^{-2} d^{-1} in 2016 and from 0.40 to 2.6 g m^{-2} d^{-1} in 2017, compared to the non-treated range of 1.2 to 6.0 and 1.3 to 5.2 g m^{-2} d^{-1} in 2016 and 2017, respectively (Fig. 8). Given that TE rate did not affect duration to the MSP, we also expected that the low rate applied every 200 GDD_0 would maintain a consistent daily growth rate, though suppression magnitude would be less. For the low rate, growth ranged from 0.45 to 3.4 and from 0.50 to 3.4 g m^{-2} d^{-1}in 2016 and 2017, respectively.

For the low rate, color ratings were significantly equal to or greater than the non-treated by 21 DAIT for both years—and were never considered unacceptable (Table 6). The high rate was significantly less than the non-treated until about 28 DAIT in both years, but the color rating was never unacceptable in 2016. After the phytotoxicity subsided, turfgrass color ratings were equal to or significantly higher than the non-treated for the remainder of the TE application period. Previous research also suggests that frequent TE applications at low rates will prevent phytotoxicity and still maintain suppression of ultradwarf bermudagrass and creeping bentgrass putting greens (McCullough et al., 2005a; McCullough et al., 2005b; McCullough et al., 2007).

400 GDD_0. The 400-GDD_0 interval occurs about 150 GDD_0 after the MSP, so we expected that both rates would result in fluctuating daily growth. For the low rate, growth was significantly different from the non-treated on 23 and 9 of the collection dates in 2016 and 2017, respectively (Fig. 7.2). The average suppression for the low rate was 28% in 2016 and 9% in 2017 (Table 5). For the high rate, growth was significantly different from the non-treated on every collection date, except for 3 in 2016 and 3 in 2017. The average suppression was 54% in 2016 and 43% in 2017. The high rate was significantly different from the low rate on every collection date in 2017 and all but 5 in 2016. In terms of daily growth rate, the high rate ranged from 0.47 to 3.0 and from 0.42 to 4.8 $g\ m^{-2}\ d^{-1}$ in 2016 and 2017, respectively (Fig. 8). The high rate of the 400-GDD_0 interval should maintain the suppression phase, but the daily growth rate is not as consistent as the 200-GDD_0 interval.

For both years, the color rating of the low rate was never significantly less than the non-treated, though some phytotoxicity did occur (Table 6). Like the 200-GDD_0 interval, the color rating for the high rate was significantly less than the non-treated until 28 DAIT for both years.

600 GDD_0. For the low rate, the 600-GDD_0 interval did not maintain the suppression phase, and it was not significantly different from the non-treated on 18 and 19 collection dates in 2016 and 2017, respectively (Fig. 7.2). The average suppression was only 24% in 2016 and 20% in 2017 (Table 5). The high rate maintained suppression on more collection dates than the low rate, but it was not significantly different from the non-treated on 8 and 18 collection dates in 2016 and 2017, respectively. The high rate provided greater season-long average suppression of 46% in 2016 and 27% in 2017, and it was significantly different from the low rate on 23 and 9 collection dates in 2016 and 2017, respectively. Color ratings were never significantly lower than the non-treated for either rate (Table 6).

Rebound

The occurrence of accelerated growth (rebound) after the suppression phase varies with cultivar, temperature, and TE rate (Beasley et al., 2007; Fagerness and Yelverton, 2000; Kreuser and Soldat, 2011). Rebound is hypothesized to be the result of accumulated GA_{20} (a non-bioactive GA) being quickly converted to GA_1 (the first bioactive GA) as trinexapac acid is degraded (King et al., 1997). Two weeks after a second TE application on Kentucky bluegrass, GA_{20} concentration was increased 146%, in contrast to the 47% decrease of GA_1 (Tan and Qian, 2003).

We did not report significant rebound on ultradwarf bermudagrass in our experiment without repeated TE applications (Chapter 2), which agrees with similar research (Reasor et al., 2018). To date, no research has reported rebound following TE applications on ultradwarf bermudagrass putting greens. In contrast, the following data suggest that rebound may occur. This rebound section includes collection dates beginning about 260 GDD_0 after the final TE application, which is where the MSP occurred; and the data end on the last collection date, approximately 700 GDD_0 after the final TE application (Fig. 9).

As expected, turfgrass growth rate began returning to the non-treated rate for all treatments within 260 GDD_0 (~10 d) after the final TE application. Relative clipping yield during this period was significantly different for the two years, so results are presented by year. For both years, no treatment was ever significantly higher than the non-treated when separated by collection date, which was due to high variability across replications. In 2016, average suppression magnitude during this period was significantly higher than the non-treated for the 400-GDD_0 (low rate) and 600-GDD_0 (low rate). The low rate of the 400-GDD_0 interval had the greatest increase in growth with an average of 62% more than the non-treated (Table 7), and it peaked at 140% more on the last collection date (Fig. 9). While not significantly different, the growth of the 400-GDD_0 and 600-GDD_0 at the high rate was more than the non-treated by approximately 420 GDD_0 after the final TE application in 2016. This was unexpected since we reported that the suppression duration following a single TE application at the high rate was over 1100

GDD_0 during this same collection period—as the experiments were conducted concurrently (Chapter 2).

Given that environmental conditions were the same, we propose two explanations for this conflict in suppression duration. First, we hypothesize that sequential TE applications may have reduced efficacy as GA_{20} accumulates in the plant. We suspect that as this GA_{20} concentration increases, so does the likelihood of the catalyzation of GA_{20} to GA_1—assuming trinexapac acid concentration remains constant in the plant. This would result in a quicker return to a non-treated growth rate (and rebound may follow). Second, we should also consider if increased tiller density following repeated TE applications exaggerated the apparent shortening of suppression duration. Because we did not measure tiller density, we are hesitant to draw conclusions about turfgrass growth rate during the "rebound period." Based on visual observation in the weeks following the last clipping collection, we suspect that the growth rate of the treated plots did exceed the non-treated plots, especially in mid-October when the non-treated plots entered dormancy before the treated plots.

In 2017, only the 400-GDD_0 interval (low rate) was significantly higher than the non-treated during this period, with only 16% more growth than the non-treated. To better understand the potential rebound period for ultradwarf bermudagrass, future research should examine the GA pathway of ultradwarf bermudagrass, and experiments should be designed to continue clipping collections until turfgrass dormancy and resume collections with spring green-up.

Conclusion

A GDD reapplication schedule will be advantageous for turfgrass managers if it provides a practical benefit over a calendar-based schedule. We expect that the benefits of a GDD schedule will be greatest in locations with fluctuating season temperatures. These results indicate that reapplying before the MSP will provide consistent daily growth and improved turfgrass quality. Applying too often can negatively affect turfgrass quality following initial applications, as noted with the 100-GDD_0 interval at the high rate. On the other hand, not applying frequently enough will not maintain the suppression phase. We are hesitant to draw conclusions from the rebound section, but turfgrass managers should be aware of the potential for accelerated growth after applications end. If timed correctly, rebound growth could shorten recovery time required after cultural practices, like core aeration, or other injury to the putting surface (Lickfeldt et al., 2001). This could be helpful since previous research suggests that TE applications slow turfgrass recovery from aeration while growth is suppressed (McCullough et al., 2007). Future research should be designed to analyze the growth of ultradwarf bermudagrass after the cessation of repeated TE applications.

Most turfgrass managers have limited flexibility in the scheduling of maintenance practices, like spraying TE. A strict GDD reapplication interval may be difficult to adhere to, but we suggest that GDD can be used in combination with a calendar schedule. For example, TE could be reapplied every 7 d only if 200 GDD_0 has accumulated since the previous application (or if forecasts indicate that this threshold will be crossed within the next couple of days). Also, GDD can be used to ensure that reapplications occur frequently enough to prevent potential rebound growth. Future research should test a

GDD_0 reapplication schedule in another climate and find ways to reduce phytotoxicity following initial applications.

Table 5. Average relative clipping yield over the suppression period.

Treatment		Average Relative Clipping Yield[†][‡]	
Interval	Rate	2016	2017
100 GDD_0	0.022	0.20 b	0.25 b
	0.044	0.08 a	0.18 a
200 GDD_0	0.022	0.45 c	0.59 d
	0.044	0.24 b	0.32 c
400 GDD_0	0.022	0.72 e	0.91 g
	0.044	0.46 c	0.57 d
600 GDD_0	0.022	0.76 e	0.80 f
	0.044	0.54 d	0.73 e
Non-treated	0	1.00 f	1.00 h

[†] This includes collection dates beginning at the maximum suppression point (MSP) following the initial application (18 May 2016 and 17 May 2017) through the collection date of the final TE application (8 August 2016 and 7 August 2017).

[‡] Column means not sharing any letter are significantly different according to Fisher's protected LSD (α=0.05).

Table 6. Turfgrass color rating separated by year, treatment, and rating date.

Turfgrass Color Ratings[†] (2016)							
Treatment		DAIT[‡§]					
Interval	Rate	7	21	28	46	67	88
100 GDD$_0$	0.022	6.25 a	6.50 ab	6.75 a	8.00 a	7.25 a	8.00 a
	0.044	5.75 a	5.25 c	5.50 b	6.00 c	7.00 a	8.00 a
200 GDD$_0$	0.022	6.25 a	6.75 ab	7.00 a	8.00 a	7.25 a	8.00 a
	0.044	6.00 a	6.25 b	6.50 a	8.00 a	7.50 a	8.00 a
400 GDD$_0$	0.022	6.00 a	6.75 ab	7.00 a	8.00 a	7.25 a	8.00 a
	0.044	5.75 a	6.25 b	6.75 a	8.00 a	7.25 a	8.00 a
600 GDD$_0$	0.022	6.25 a	6.75 ab	6.75 a	8.00 a	7.50 a	8.00 a
	0.044	6.50 a	6.50 ab	7.00 a	8.00 a	7.50 a	8.00 a
Non-treated	0	6.00 a	7.00 a	7.00 a	7.00 b	7.00 a	7.00 b

Turfgrass Color Ratings[†] (2017)							
Treatment		DAIT[‡§]					
Interval	Rate	9	14	21	30	53	93
100 GDD$_0$	0.022	5.75 bc	5.25 d	6.00 a	7.75 ab	8.00 a	8.00 a
	0.044	5.00 d	4.00 e	5.00 c	6.75 d	8.00 a	8.00 a
200 GDD$_0$	0.022	6.25 ab	6.00 bc	6.75 a	8.00 a	8.00 a	8.00 a
	0.044	5.25 cd	4.50 e	6.00 b	7.5 abc	8.00 a	8.00 a
400 GDD$_0$	0.022	6.75 a	7.00 a	7.00 a	8.00 a	8.00 a	8.00 a
	0.044	5.75 bc	5.75 cd	6.25 b	8.00 a	8.00 a	8.00 a
600 GDD$_0$	0.022	6.25 ab	7.00 a	7.00 a	8.00 a	7.25 b	7.00 b
	0.044	6.25 ab	6.50 ab	7.00 a	7.25bcd	7.75 a	8.00 a
Non-treated	0	6.75 a	7.00 a	7.00 a	7.00 cd	7.00 b	7.00 b

[†] A color rating of 6 was considered minimally acceptable and 9 was considered optimal turfgrass.

[‡] Days after initial treatment (DAIT).

[§] Column means not sharing any letter are significantly different according to Fisher's protected LSD (α=0.05).

Table 7. Average relative clipping yield over the rebound period.

Treatment		Average Relative Clipping Yield[†‡]	
Interval	Rate	2016	2017
100 GDD_0	0.022	1.27 bc	0.87 c
	0.044	0.85 e	0.94 c
200 GDD_0	0.022	1.17 cd	1.09 ab
	0.044	0.97 de	0.96 bc
400 GDD_0	0.022	1.62 a	1.16 a
	0.044	1.27 bc	0.91 c
600 GDD_0	0.022	1.49 ab	0.91 c
	0.044	1.28 bc	0.88 c
Non-treated	0	1.00 cde	1.00 bc

[†] This includes collection dates beginning at the maximum suppression point (MSP) after the final TE application and ending on the last collection date.

[‡] Column means not sharing any letter are significantly different according to Fisher's protected LSD (α=0.05).

Figure 5. The average daily temperature during the experiment was 26.4°C in 2016 and 25.3°C in 2017. Total GDD_0 accumulation over the 98-d experiment was 2580 GDD_0 in 2016 and 2460 GDD_0 in 2017. This resulted in a total of 25, 13, 7, and 5 reapplications for the 100, 200, 400, and 600 GDD_0 treatments, respectively.

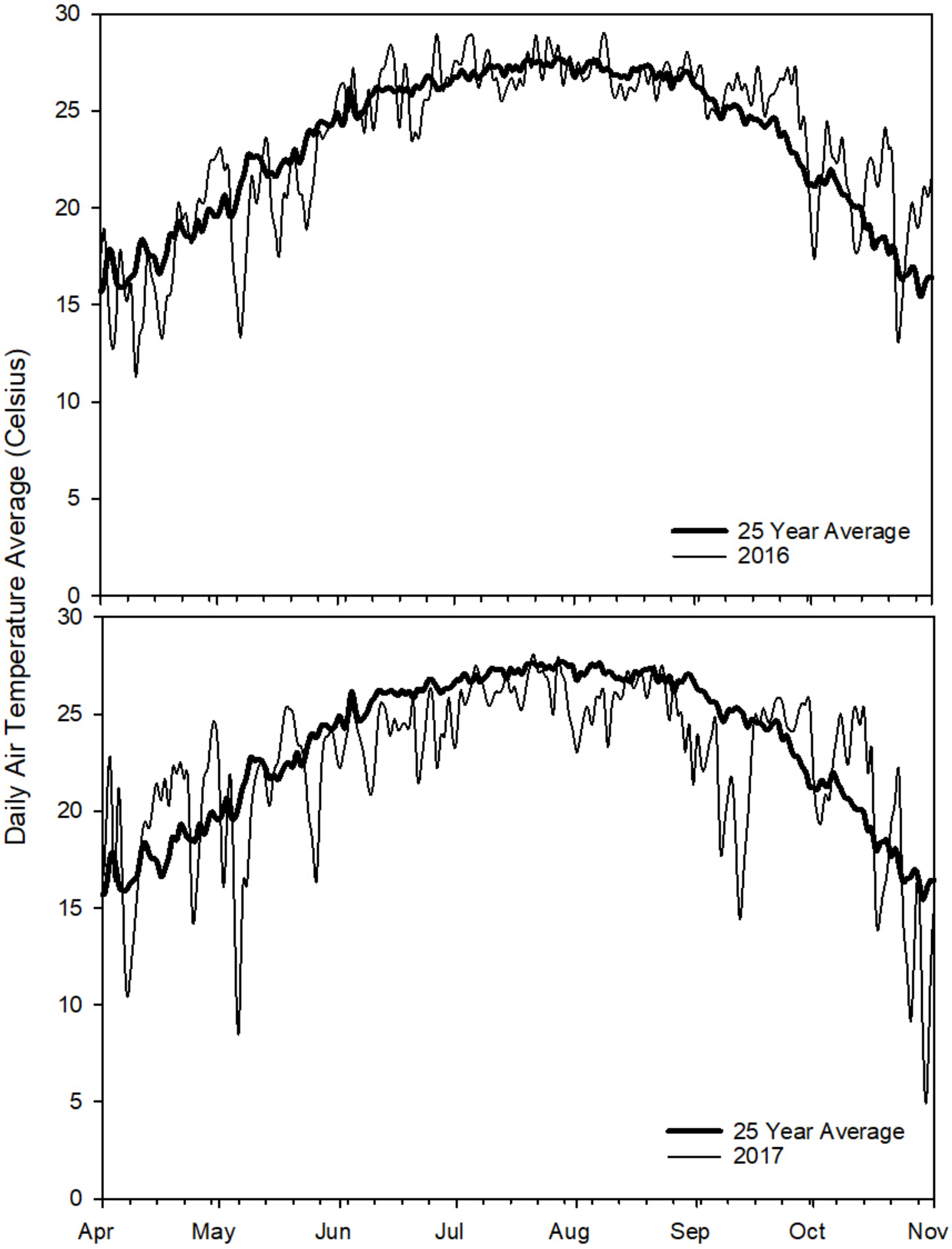


Figure 6. Relative clipping yield plotted by GDD_0 after a TE application. The solid line regression includes data pooled by year and collection date for the 600-GDD_0 interval with TE at 0.022 kg ai ha^{-1}. These data were fit to a five-parameter, amplitude-damped sine regression (equation above x-axis), which was chosen because it resulted in the largest pseudo-R^2 value (0.264, SE: 0.215). The dashed line regression is for TE at 0.044 kg ai ha^{-1} (Chapter 2). The MSP occurred at 272 and 262 GDD_0 with a suppression magnitude of 38 and 61% for the 0.022 and 0.044 kg ai ha^{-1}, respectively.

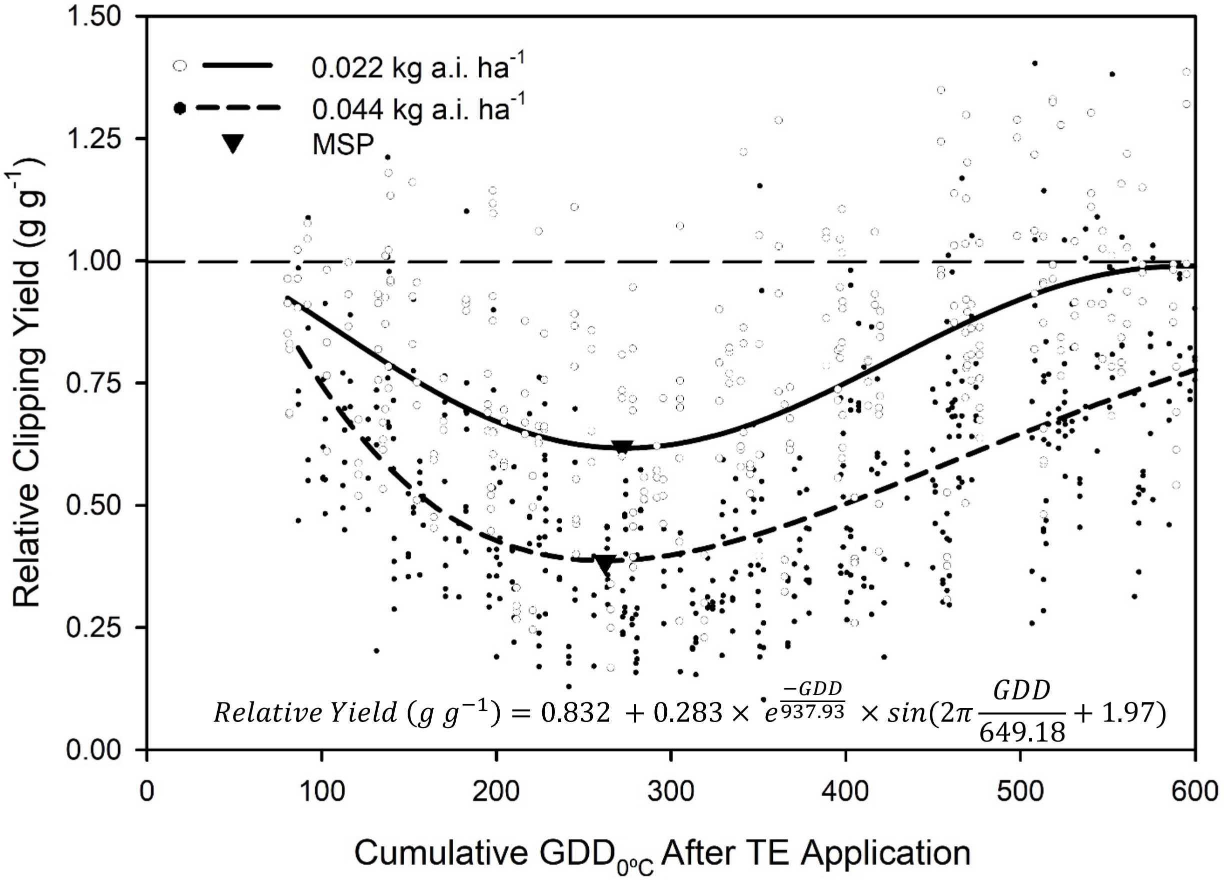


Figure 7.1. Relative clipping yield plotted by collection date for the suppression period (100- and 200-GDD$_0$ intervals). Separated by year, interval, and rate. The graphs begin after the MSP that followed the initial TE application and end on the date of the final TE application. For each graph, the top row of asterisks (L/NT) indicates if the low rate was significantly less than the non-treated on that collection date (according to Fisher's protected LSD with α=0.05), and the middle row of asterisks (H/NT) indicates if the high rate was significantly less than the non-treated. The bottom row (L/H) indicates if the low rate was significantly different from the high rate on that collection date.

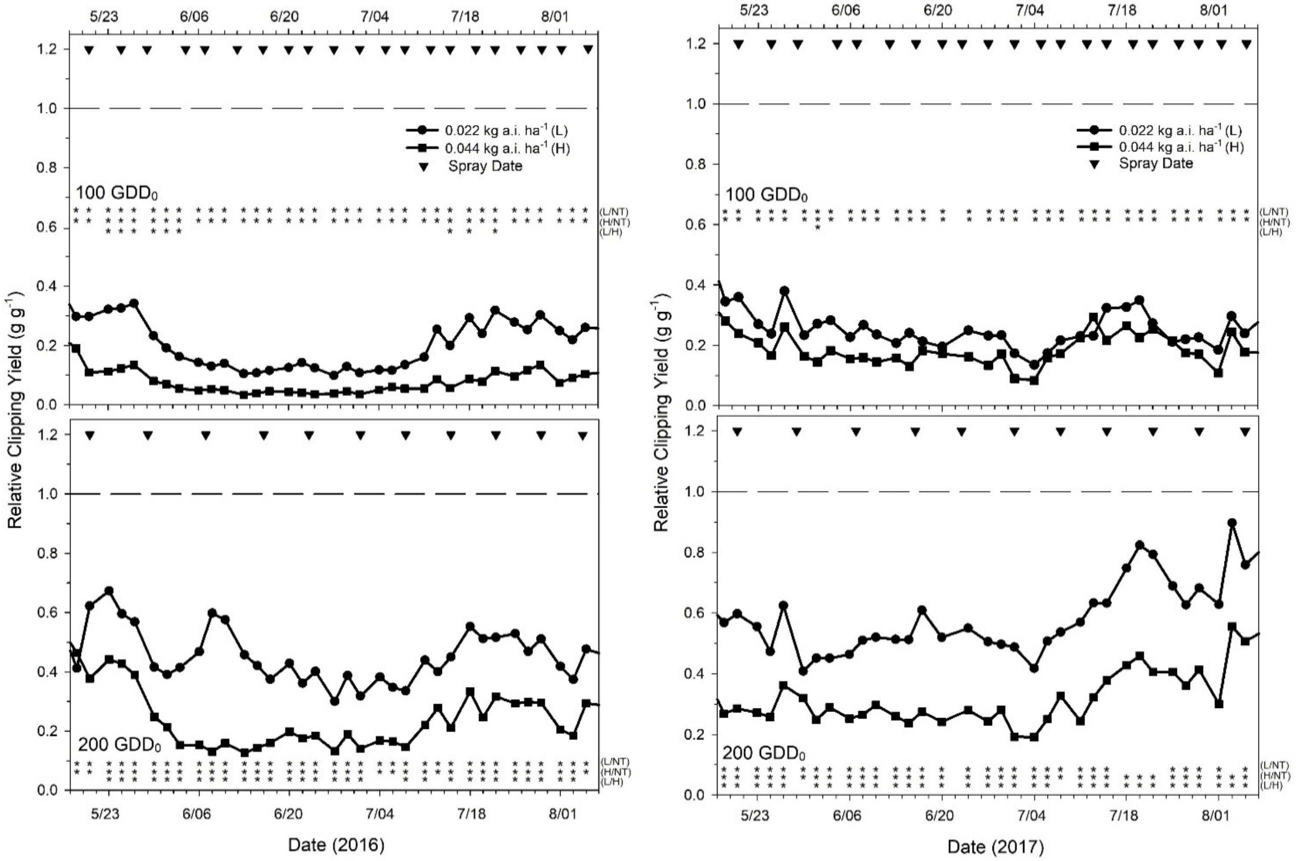


Figure 7.2. Relative clipping yield plotted by collection date for the suppression period (400- and 600-GDD$_0$ intervals). Separated by year, interval, and rate. The graphs begin after the MSP that followed the initial TE application and end on the date of the final TE application. For each graph, the top row of asterisks (L/NT) indicates if the low rate was significantly less than the non-treated on that collection date (according to Fisher’s protected LSD with α=0.05), and the middle row of asterisks (H/NT) indicates if the high rate was significantly less than the non-treated. The bottom row (L/H) indicates if the low rate was significantly different from the high rate on that collection date

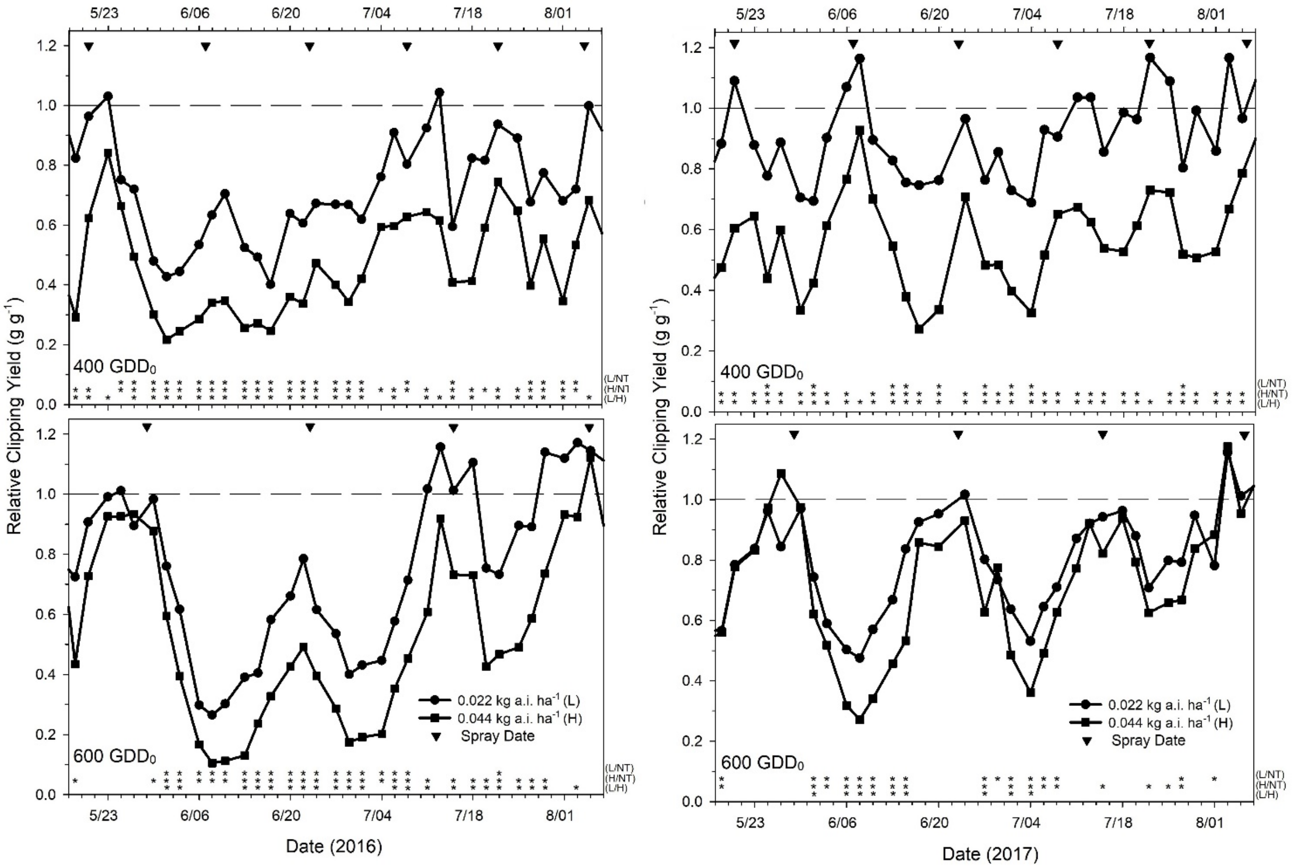


Figure 8. Daily growth plotted by collection date for the suppression period. This graph shows that the more frequent reapplication interval minimized clipping yield fluctuation across the season. The diamonds and triangles at the top of each graph represent spray dates for the 200- and 400-GDD$_0$ interval, respectively. The top row of asterisks (200/NT) indicates if the 200-GDD$_0$ interval was significantly less than the non-treated on that collection date (according to Fisher's protected LSD with α=0.05), and the middle row of asterisks (400/NT) indicates if the 400-GDD$_0$ interval was significantly less than the non-treated. The bottom row (200/400) indicates if the 200-GDD$_0$ interval at the high rate was significantly different from 400-GDD$_0$ interval at the high rate on that collection date.

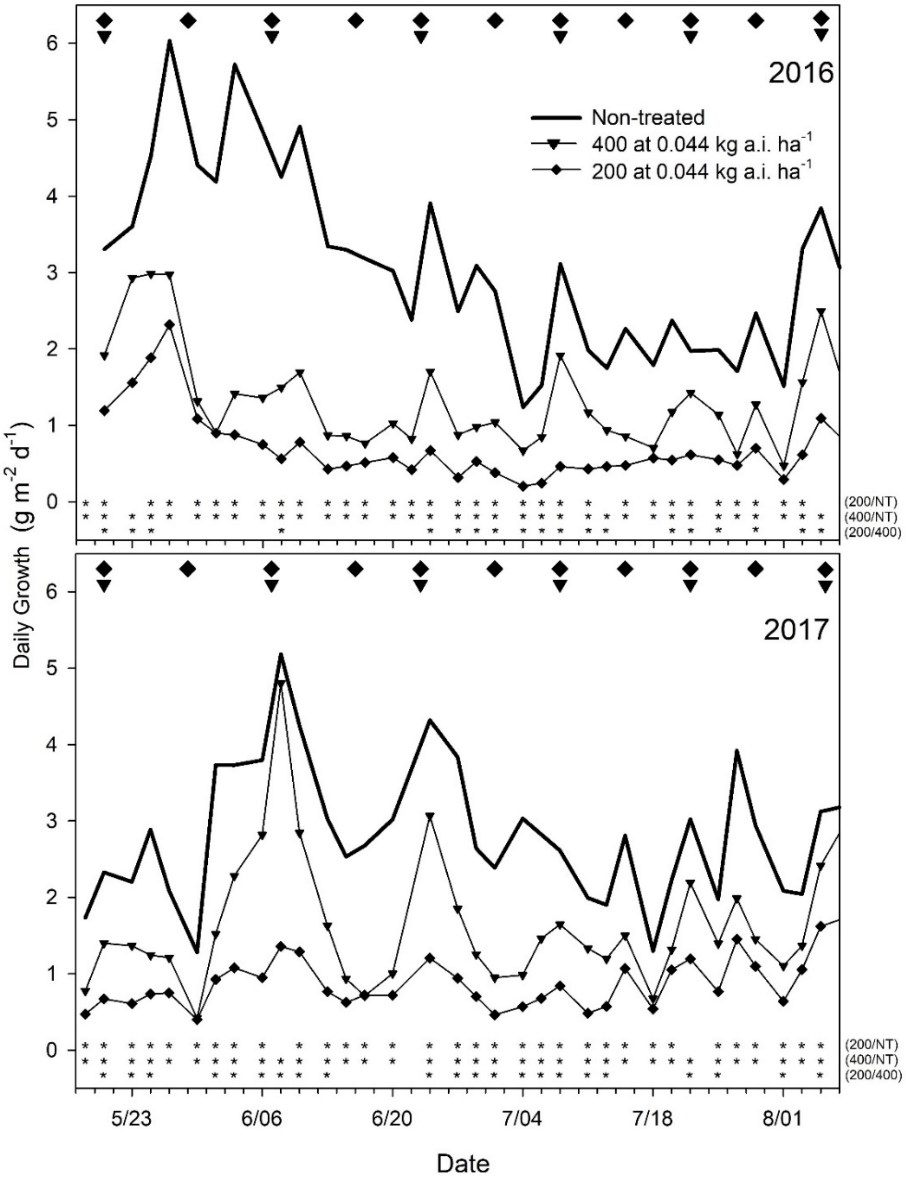
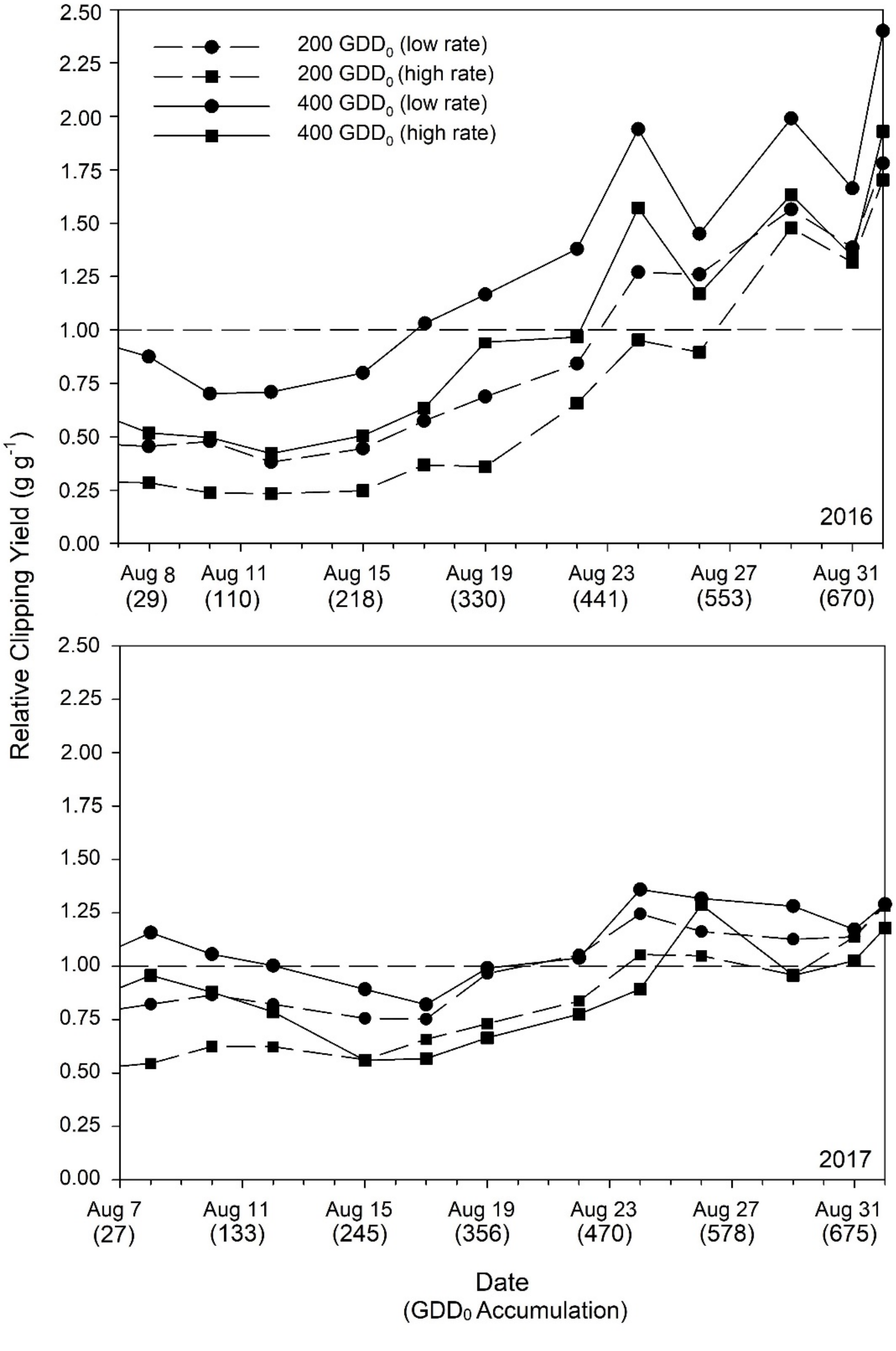


Figure 9. Relative clipping yield plotted by collection date for the rebound period. The number on the x-axis below the date is the total GDD_0 accumulation after the final TE application. This graph begins on the collection date following the final TE application and ends on the last collection date, but analysis presented in the "rebound section" only includes data starting with 17 and 16 August in 2016 and 2017, respectively, and continuing through the last collection date.



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