Turfgrass in an ever-changing world

by

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DEDICATION

This thesis is dedicated to my family and friends that have been so instrumental in who I am today.

TABLE OF CONTENTS

Page

NOMENCLATURE v

ACKNOWLEDGMENTS vi

ABSTRACT vii

CHAPTER 1. GENERAL INTRODUCTION 1
  Origins of Turfgrass Usage 1
  Turfgrass and the Ecosystem 1
  Aesthetics and cool-season turfgrass 2
  Turfgrass Safety and Sport 4
  Turfgrass Assemblages 5
  References 6

CHAPTER 2. SPORTS FIELD RECOVERY FROM UNPLANNED SUSPENSION OF MAINTENANCE ACTIVITIES 10
  Abstract 10
  Introduction 11
  Materials and Methods 14
    Plot construction and Maintenance 14
    Treatments 15
    Height-of-Cut Reduction 15
    Nitrogen Applications 16
    Plant Growth Regulator Treatments 16
    Simulated Traffic 16
    Data Collection 17
    Statistical Analysis 18
  Results and Discussion 18
    Results 18
    Discussion 19
  Conclusion 20
  References 20
  Figures and Tables 24

CHAPTER 3. EVALUATING THE SUSTAINABILITY OF TURFGRASS LAWN ASSEMBLAGES 33
  Abstract 33
  Introduction 33
  Materials and Methods 36
    Experimental Design and Treatments 36
    Establishment 37
    Greenhouse Conditions 37
    Establishment Rate and Maturity Tracking 37

    Drought Tolerance 39
    Statistical Analysis 39
  Results and Discussion 40
    Establishment and Plant Productivity 40
    Simulated Drought 41
    Discussion 41
  Conclusion 43
  References 44
  Tables and Figures 47

CHAPTER 4. GENERAL CONCLUSION 59

NOMENCLATURE

C	Cultivar
CIST	Clegg Impact Surface Tester
Db	Bulk Density
DIA	Digital Image Analysis
FF	Fine Fescue
HOC	Height-of-Cut
KBG	Kentucky bluegrass
LOI	Loss On Ignition
LSD	Least Significant Difference
NTEP	National Turfgrass Evaluation Program
PGC	Percent Green Cover
PGR	Plant Growth Regulator
PR	Perennial Ryegrass
RCBD	Randomized Complete Block Design
S	Species
STE	Simulated Traffic Event
TE	Trinexapac-ethyl
TF	Tall fescue
TP	Total Porosity

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ABSTRACT

Turfgrasses are estimated to cover 2% of the United States land area, it is important to ensure lawns are sustainable while still maintaining ecosystem services (Robbins and Birkenholtz, 2003). Previous research has evaluated the sustainability of mixing species and cultivars. Cultivar performance was not accounted for in prior studies. Because of previous work done with biodiversity and ecosystem function theory, it was hypothesized that more genetic diversity would result in less inorganic nitrate and ammonium leaching (Thompson et al., 2016).

Sixteen cultivars were selected from the National Turfgrass Evaluation Program (NTEP) trials based on turfgrass quality, and commercial availability [4 Kentucky bluegrass (*Poa pratensis* L.; KBG) cultivars, 4 tall fescue (*Schedonorus arundinaceus* (Schreb.); TF) cultivars, 4 fine fescue (*Festuca* spp.; FF) cultivars, and 4 perennial ryegrass (*Lolium perenne* L.; PR) cultivars]. Cultivars were grouped into various assemblage types (monocultures, 3-species mixes, 3-cultivar blends, and 3-species by 3-cultivar mixes). Turfgrass assemblages were all similar in drought tolerance, soil organic carbon and soil microbial biomass. Three cultivar assemblages exhibited higher percent green cover from 42-days after seeding until the end of establishment. Three species and three cultivar assemblages leached similar or lower nitrate than 3-species by 3-cultivar assemblages and monocultures.

Additionally, athletic field managers often face periods of unplanned suspension from maintenance activities. These suspensions can be due to a variety of reasons including natural disasters, budgetary concerns, or pandemics as recently seen with COVID-19. When turfgrass managers return, turf height-of-cut (HOC) can be much greater than desired. Research was conducted on mature ‘Moonlight’ Kentucky bluegrass grown on a native soil. Turfgrass was left unmaintained from the beginning of the growing season through the end of May. Treatments were subjected to various maintenance regimes including combinations of three different mowing treatments (⅓ rule, cut in half and then ⅓ rule, and scalp to final HOC of 5.1cm), two fertility programs (36.6 kg N ha^{-1} and 73.2 kg N ha^{-1}) and were either subjected to Trinexapac-ethyl (TE) applications or not. Twenty-five simulated traffic events (STEs) were applied with a modified Baldree Traffic Simulator. Simulated traffic occurred in the fall of 2020 and 2021. Digital images for percent green cover, surface hardness, and rotational resistance were tested after every five STEs. While there are differences in the slope, for the loss of percent green cover, treatment differences were lacking.

Orthogonal contrasts determined no differences between regime variables in percent green cover. After 25 STEs, small treatment differences were observed for rotational resistance and surface hardness. Although these differences were very minor between treatments. No hardness measurements were greater than 100 GMAX. Soil bulk density at a 10.2 cm depth was not different between any treatments after 25 STEs. The minimum of a 2-month delay after achieving desired HOC before applying STEs may have allowed the turfgrass to acclimate and thus not result in STE stress to the degree that might be expected if simulated traffic occurred sooner after the desired HOC was reached.

There is a need in the turfgrass industry to challenge and update long-standing rules about management practices and decisions. In the case of these two studies, differing societal circumstances and improved varieties of modern day don’t require vastly different changes in management practices.

CHAPTER 1. GENERAL INTRODUCTION

Origins of Turfgrass Usage

Turfgrasses have been an important part of human society for a long time, with references seen as early as 1400 BCE (Roberts et al., 1992; Beard, 1998). They are commonly used in western civilization to create places for recreation and sport (Beard and Green, 1994). A turfgrass stand can also lead to an overall better quality of life due to its aesthetic, recreational and functional qualities (Beard and Green, 1994). Turfgrass usage does have some environmental setbacks; including the time for home lawn carbon sequestration, negating at an average of one hundred and eighty-four years in the United States (Selhorst and Lal, 2013). While these concerns exist, turfgrass usage is quintessential for recreation and sport in the United States (Adams and Gibbs, 1994).

Turfgrass and the Ecosystem

Numerous ecosystem services that are beneficial to the ecosystem and humans are provided by turfgrasses. Including urban heat island dissipation, lessened weed pressure and allergens, CO2 conversion, reduced erosion, groundwater filtration, and a low-cost space for safe recreation (Beard and Green, 1994). Additionally, they mitigate runoff, absorb pollutants, and enhance mental health (Beard and Green, 1994). Concerns about water usage, pesticides, and lack of suitable wildlife are often raised (Stier et al., 2013; Beard and Green, 1994). Turfgrass has evolved from usage as a forage grass and cover crop to now being used by homeowners, golf courses and sport as a crossroads between nature and recreation (Adams and Gibbs, 1994; Beard, 1998). Turfgrasses are estimated to cover 1.9% of the United States land area (Thompson et al., 2017; Milesi et al., 2005), it is important to ensure lawns are sustainable while still maintaining ecosystem services (Robbins et al., 2003). Concerns for turfgrass usage arise regarding irrigation, pesticide usage, and lack of suitable wildlife habitat. Upon comparison to hardscapes, concrete or asphalt in an urban environment, the ecosystem services turfgrass provides become much more pronounced (Christians et al., 2017).

Reliance on our ecosystem services is essential to survival, sustainable practices help promote a healthy ecosystem and those in it (Zhang et al., 2010). Humans are connected to nature emotionally, philosophically, experientially, and materially, and promotion of connecting with nature is becoming increasingly popular (Ives et al., 2018). Proper management practices can help turfgrass become more productive, retain nutrients, limit soil erosion, increase traffic and drought tolerance, and minimize pest pressures (Chrisitans et al., 2017, Beard and Green, 1994).

Aesthetics and cool-season turfgrass

Turfgrass aesthetics are important for property values for homeowners, people aged 45 to 64 prefer landscapes with more turfgrass and fewer trees (Des Rosiers et al., 2002). Home values with quality landscaping have higher home valuations than homes with poor landscaping quality (Henry, 1994; McKenzie, 2005). Looking at natural areas like turfgrass increases sense of tranquility, and mental functioning (Frumkin, 2001). A study by Moore (1981) showed that prisoners that had a window view of greenspace had 24% fewer sick visits than prisoners who didn’t have a view overlooking greenery. Maintained turfgrasses attract recreation and leisure, and physical activity, and provide a softer surface than paved surfaces (Coady and Micheli, 1997).

Cool-season turfgrass lawns in the northern United States often have a mixture of species and cultivars to account for microclimate conditions, disease resistance and differences in sun exposure (Stier et al., 2013). In sports fields it is typical to see multiple cultivars of a species blended to provide diversity in genetics and traits (Vargas and Turgeon, 1980; Brede, 2007).

Ecologically speaking, diversity of species influences ecosystem function, and greater genetic diversity provides ecosystem services more reliably in changing environments (Hooper et al., 2005).

Turfgrass cover can be affected by disease, insect or weed infestation, improper fertilization, or improper watering (Christians et al., 2017). A weed is a plant that is widely adapted to succeed in environments that are not easily inhabitable by turfgrasses (Zimdahl, 2007). Weeds grow in undesirable places and succeed in a wide range of environments (Zimdahl, 2007). Weeds specifically establish most readily in weak turfgrass stands (Engel & Ilnicki, 1969). Many common turfgrass weeds have been shown to have less traffic tolerance than turfgrass, and result in lower plant cover, which increases surface hardness in areas covered in weeds rather than turfgrass (Brosnan et al., 2014). This increase in surface hardness can result in a higher injury incidence. Without proper mowing practices provided by regular maintenance activities, weeds can establish more readily, and are at a competitive advantage over turfgrasses (Engel and Ilnicki, 1969). Reducing turfgrass stress is helpful in reducing common turfgrass pests (Christians et al., 2017).

Higher fertilization rates have generally been shown to resist broadleaf weed growth and development (Voigt et al., 2001). Miltner et al. (2005) found that a 294 kg N ha^{-1} yielded fewer weeds than rates with 66% and 33% less N respectively. A study done by Calhoun et al. (2005) showed a decreased weed incursion with fertilized treatments compared to an untreated control. It has been observed in bermudagrass (*Cynodon dactylon* L.) that shoot density increases with N rate (Guertal and Hicks, 2009). It can be inferred that an increase in shoot density could be the reasoning for increased weed deterrence.

Turfgrass Safety and Sport

Many factors can contribute to the safety of turfgrass surfaces, including soil compaction, turf height-of-cut, and turf cover. Over 20% of concussions suffered by high school athletes are a result of head impacts with the playing surface (Gessel et al., 2007). Rogers and Waddington, (1992) and Griffin et al. (2006) both reported that soil compaction and turf cover are critical to athletic field safety, due to compacted soil increasing the rate of concussions. Steffen et al. (2007) reported the incidence of serious injury was significantly higher on artificial turf than natural grass for young female soccer players. Artificial turf has also shown to cause great temperature differences during sunny conditions than natural grass, which can exacerbate heat related injuries (Jim, 2017). Even the former National Football League (NFL) players are calling for the reduction of artificial turf usage due to an increased rate of knee and ankle injuries seen when compared to natural grass (Hershman et al., 2012). A well-maintained natural grass playing surface is desired as a result.

There are costs associated with reducing HOC, including inadequate carbohydrate supply from the loss of productive biomass, increased susceptibility to environmental stresses and lack of root growth (Voigt et al., 2001). Lower HOC leads to less traffic tolerance but reduces friction seen on an athlete’s foot as they interact with the playing surface (Strunk et al., 2021). Reduction and lower HOC on turfgrass can be stressful on the plant and the use of a nitrogen fertilizer has been shown to be beneficial in improving turfgrass quality (Dernoeden et al., 1993). Plant growth regulators (PGRs) are also used in the turf industry to improve turfgrass quality. Trinexapac-ethyl (TE) is the most widely used PGR in the turf industry, it has been shown to increase chlorophyll concentrations, shoot density and turfgrass quality (Lickfeldt et al., 2001). Trinexapac-ethyl has also been shown to improve photosynthetic efficiency and provide greater carbohydrate and N availability to stem and root tissues (Ervin and Koski, 1998; Zhang and

Schmidt, 2000). The use of TE has also been shown to increases rooting (McCullough et al., 2006) and plant tillering (Beasley et al., 2005).

Athletic field managers will inevitably face periods of unplanned suspension from maintenance activities. The reasoning behind these unplanned suspensions can be due to a variety of reasons including budgetary constraints, natural disasters and pandemics seen recently with COVID-19 across the world (Brosnan et al., 2020). When turf managers return, HOC can be much higher than desired. The process of reducing HOC can be stressful for the plant and turf managers alike, and currently there isn't a set protocol for the most efficient and effective way to reduce turf HOC. The second chapter of this research seeks to determine an effective way to reduce turf HOC and demonstrate what inputs (fertilizer and growth regulator) would be best for returning turf conditions back to normal for a high performing athletic field that fall.

Turfgrass Assemblages

The basis of the third chapter of this thesis is inspired by the works of Thompson and Kao-Kniffin (2016), where they found that with increased turfgrass assemblage diversity, came decreased nitrate leaching, and increased microbial diversity. Basis for cultivar selection was not based on cultivar performance and incorporated some species that would be deemed weeds in manicured turfgrass setting. Proven performance-based cultivars were a priority for this study. Biodiversity and ecosystem function theory also suggests that more genetic diversity would result in less inorganic nitrate and ammonium leaching (Thompson and Kao-Kniffin, 2016). Work has been done to show that species richness and plant productivity associated with it has a correlation to soil respiration (Dias et al., 2010). Because of this we believe the greater the number of species and varieties, the higher the amount of soil organic carbon and overall turfgrass quality. Finally, due to potentially differing root architecture among species and cultivars, potentially greater drought tolerance would be observed with greater diversity.

Differing root architecture among species and cultivars has also been concluded to help improve soil health and performance (Bowman et al., 2002). The increased stability from a more genetically diverse assemblage has also been shown to decrease pest pressure (Naeem, 2002).

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CHAPTER 2. SPORTS FIELD RECOVERY FROM UNPLANNED SUSPENSION OF MAINTENANCE ACTIVITIES

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Abstract

Athletic field managers often face periods of unplanned suspension from maintenance activities. These suspensions can be due to a variety of reasons including budgetary constraints, natural disasters, and pandemics as recently seen with COVID-19. When turfgrass managers return, turf height-of-cut (HOC) can be much greater than desired. Research was conducted on mature 'Moonlight' Kentucky bluegrass grown on a native soil. Turfgrass was left unmaintained from the beginning of the growing season through the end of May in 2020 and 2021. Treatments were subjected to various maintenance regimes including combinations of three different mowing treatments (⅓ rule, cut in half and then ⅓ rule, and scalp to final HOC of 5.1cm), two fertility programs (36.6 kg N ha^{-1} and 73.2 kg N ha^{-1}) and were either subjected to Trinexapac-ethyl (TE) applications or not. Twenty-five simulated traffic events (STEs) were applied with a modified Baldree Traffic Simulator. Simulated traffic occurred in the fall both years to model after a typical American football season. Digital images for percent green cover, surface hardness, and rotational resistance were tested after 10, 20, and 25 STEs. While there are differences in the slope, for the loss of percent green cover, treatment differences were lacking. Orthogonal contrasts determined no differences between regime variables in percent green cover. After 25 STEs, small treatment differences were observed for rotational resistance and surface hardness. Although these differences were very minor between treatments. No hardness measurements were greater than 100 GMAX. Soil bulk density at a 10.2 cm depth was not

different between any treatments. The minimum of a 2-month delay after achieving desired HOC before applying STEs may have allowed the turfgrass to recover fully and thus not result in STE stress to the degree that might be expected if STE occurred immediately following desired HOC reduction.

Introduction

Turfgrasses have been an important part of human society for a long time, with some of the earliest references to turfgrass usage found in the Bible (Roberts et al., 1992; Beard, 1998). They are commonly used in western civilization to create places for recreation and sport (Beard and Green, 1994). They offer some ecosystem services including groundwater recharging, erosion control, decreased weed pressure, and a low-cost cushion against impact injuries (Beard and Green, 1994). A turfgrass stand can also lead to an overall better quality of life due to its aesthetic, recreational and functional qualities (Beard and Green, 1994). There are also concerns regarding water usage, pesticide and fertilizer pollution and lack of suitable wildlife habitat (Stier et al., 2013). While these concerns exist, turfgrass usage is vital for sporting and recreation in the U.S. (Adams and Gibbs, 1994).

Many factors can contribute to the safety of turfgrass surfaces, including soil compaction, turf height-of-cut (HOC), and turf cover. Over 20% of concussions suffered by high school athletes are a result of head impacts with the playing surface (Gessel et al., 2007). Rogers and Waddington, (1992) and Griffin et al. (2006) both reported that soil compaction and turf cover are critical to athletic field safety, due to compacted soil increasing the rate of concussions. Steffen et al. (2007) reported the incidence of serious injury was (4.7%) higher on artificial turf than natural grass for young female soccer players. Artificial turf is also shown to cause great temperature differences during sunny conditions than natural grass, which can exacerbate heat related injuries (Jim, 2017). Non-infilled artificial turf has been observed to be 30 to 60 °C warmer than natural turf (Buskirk et al., 1971). Additionally infilled artificial turf surfaces have been reported to exceed 65.6 °C during the month of August (Lim and Walker, 2009). Even the former Nation Football League (NFL) players are calling for the reduction of artificial turf usage due to higher instances of knee and ankle injuries compared to natural grass (Hershman et al., 2012). Hershman et al. (2012) reported that eversion ankle sprains were 67% more likely on artificial turf compared to natural turf, additionally ACL sprains were 31% higher on artificial turf than natural grass. A well-maintained natural grass playing surface is desired as a result, to keep players safer.

Turfgrass cover can be affected by disease, insect or weed infestation, improper fertilization, or improper watering (Christians et al., 2017). A weed is a plant that is widely adapted to succeed in environments that are not easily inhabitable by turfgrasses (Zimdahl, 2007). Weeds grow in undesirable places and succeed in a wide range of environments (Zimdahl, 2007). Weeds specifically establish most readily in weak turfgrass stands (Engel and Ilnicki, 1969). Foot traffic due to sports causes tearing of leaf tissue and the compaction of soil (Carrow and Petrovic, 1992). Openings in the canopy and compacted soil can result in weeds. Many common turfgrass weeds have been shown to have less traffic tolerance than turfgrass, and result in lower plant cover, which increases surface hardness in areas covered in weeds rather than turfgrass (Brosnan et al., 2014). This increase in surface hardness can result in a higher injury incidence. Without proper mowing practices provided by regular maintenance activities, weeds can establish more readily, and are at a competitive advantage over turfgrasses (Engel and Ilnicki, 1969). Reducing turfgrass stress is helpful in reducing common turfgrass pests.

Unplanned suspension of maintenance activities also allows the turfgrass height to increase, the reduction of turf leaf material can cause significant stress to the turfgrass including scalping and stunting (Beard, 1972). Removing no more than 1/3 of the leaf tissue is a single mowing is a commonly taught practice (Christians et al., 2017), however this is not always practical when HOC is well above desired. Providing an adequate method for turfgrass managers to reduce turfgrass HOC while minimizing stresses and outside pressures would be beneficial for overall turfgrass quality and therefore safety of natural grass athletic fields. If a field is uniform and has a higher amount of turf cover, it is a safer field for athletes to compete on (Rodgers and Waddington, 1992). If fields returning from unplanned suspension of maintenance activities are safer, there is far less liability from turf managers and groups tasked with turf maintenance

There are costs associated with reducing HOC, including inadequate carbohydrate supply from the loss of productive biomass, increased susceptibility to environmental stresses and lack of root growth (Voigt et al., 2001). Lower HOC leads to less traffic tolerance but reduces friction seen on an athlete's foot as they interact with the playing surface (Strunk et al., 2021). Strunk et al. (2021) found that hybrid bermudagrass [*Cynodon dactylon* (L.) Pers. X *C. transvaalensis* Burtt Davy] cut at 13 mm had 50 percent green cover (PGC) 1.8 STE sooner than hybrid bermudagrass mowed at 22 mm on average. Reduction and lower HOC on turfgrass can be stressful on the plant and the use of a nitrogen fertilizer has been shown to be beneficial in improving turfgrass quality (Dernoeden et al., 1993). Plant growth regulators (PGRs) are also used in the turf industry to improve turfgrass quality. Trinexapac-ethyl (TE) is the most widely used PGR in the turf industry, it has been shown to increase chlorophyll concentrations, shoot density and turfgrass quality (Lickfeldt et al., 2001). Trinexapac-ethyl has also been shown to improve photosynthetic efficiency and provide greater carbohydrate and N availability to stem and root tissues (Ervin and Koski, 2001; Zhang and Schmidt, 2000). The use of TE has also been shown to increases rooting (McCullough et al., 2006) and plant tillering (Beasley et al., 2005).

Athletic field managers will inevitably face periods of unplanned suspension from maintenance activities. The reasoning behind these unplanned suspensions can be due to a variety of reasons including budgetary constraints, natural disasters and pandemics seen recently with COVID-19 across the world (Thoms et al., 2020). When turf managers return, HOC can be much higher than desired. The process of reducing HOC can be stressful for the plant and turf managers alike, and currently there isn't a set protocol for the most efficient and effective way to reduce turf HOC for athletic field use. This research seeks to determine an effective way to reduce turf HOC and demonstrate what inputs (fertilizer and growth regulator) would be best for returning turf conditions back to normal for a high performing athletic field that fall.

Materials and Methods

Plot construction and Maintenance

A field trial was conducted at The Iowa State University Horticulture Research Station near Ames, Iowa over two years (2020-2021). The study was conducted on 'Moonlight' Kentucky bluegrass (*Poa pratensis* L.) on a disturbed native Clarion loam soil (Fine-silty, mixed, superactive, mesic Typic Hapludalfs organic matter 5.6%). The same plot areas were used for both years of study. The experimental design was a 2 x 10 randomized block that was replicated 4 times. The entire plot area was left undisturbed from the beginning of the growing season through the end of May (the unplanned suspension of maintenance period) in each year of the study. A preliminary post-emergence herbicide application of quinclorac (Drive XLR8®, BASF; Ludwigshafen, Germany) was used at a rate of at a rate of 4.67 L ha^{-1} to help provide consistent turf surfaces among treatments prior to experimentation applied on 10 August 2020 and 09

August 2021. Following the unplanned suspension of maintenance, a series of various maintenance treatments were carried out as treatments.

Treatments

Three commonly used variables (fertilizer, HOC reduction, and PGR) made up the maintenance regime for each treatment, these regimes were in place to help with turfgrass maintenance recovery program.

Height-of-Cut Reduction

Each maintenance regime had a specified HOC reduction strategy including: ⅓ rule, cut in half followed by ⅓ rule, and cut to final HOC (scalp). All plots were mowed twice a week once maintenance was resumed. The ⅓ rule states that no more than one third of the plant material can be removed at one time. This resulted in each mowing only removing 1/3 of the plant tissue per mowing. The cut in half followed by 1/3 rule HOC reduction strategy removed half of the vertical height with the first mowing, and every subsequent mowing would only remove ⅓ of the remaining leaf tissue until the desired HOC was achieved. Lastly, scalping reduces the HOC down to the target height of 5 cm in one mowing event, and then all subsequent mowing events were at 5 cm HOC. A Honda (Honda HRN216VKA; Motor Company, Tokyo, Japan) 55 cm push mower was used to apply the HOC reduction treatments with clippings removed for the scalp and cut in half treatment applications with the bagging attachment. For the 1/3 rule and once the final HOC was achieved, a John Deere (John Deere Z955; John Deere, Moline, IL) rotary mower maintained the plot at 5 cm HOC. To accurately reduce the HOC described by the treatments, prior to each mowing, the height of the grass was measured, and the correct amount of leaf tissue was determined to be removed. Turf was irrigated as determined by the investigator to achieve 2.54 cm of water a week at the beginning at HOC reduction each year.

Nitrogen Applications

A 28-0-3 nitrogen fertilizer (Lesco Incorporated, Cleveland, Ohio) was applied monthly following the suspension of maintenance at rates of either 36.6 kg N $ha^{-1}$and 73.2 kg N ha^{-1} N per month from June to November in each year of the study.

Plant Growth Regulator Treatments

The final treatment in the treatment regime was either the use of trinexapac-ethyl (Primo Maxx®, Syngenta, Basel, Switzerland), a common PGR, or no application of TE. The PGR was applied every two weeks following the resumption of maintenance until two weeks before simulated athletic traffic 1 Aug. 2020 and 6 Aug. 2021. TE was applied at a rate of 584.6 ml ha^{-1}. Trinexapac-ethyl has been shown to increase tiller density, which in turn can result in a more traffic tolerant plant (Ervin and Koski, 2001). With traffic and PGR application combined, Ervin and Koski (2001) found that heavy traffic and TE suppressed bluegrass resulted in poor turf quality. Because of this, PGR applications ceased two weeks prior to simulated traffic, similar to Haselbauer's et al. (2010) research on hybrid bermudagrass. The PGR was applied with a CO_2-pressurized backpack sprayer with TeeJet 8004XR nozzles calibrated to apply 815 L water carrier ha^{-1}. The full treatment regime combination is listed in **Table 1**.

Simulated Traffic

Athletic field traffic was simulated with a modified Baldree Traffic Simulator (Kowalewski et al., 2013). The modified Baldree Traffic Simulator has been used effectively to simulate athletic field traffic in several previous research projects (Dalsgaard et al., 2020 Pease et al., 2020 Lindsey et al., 2021). Simulated athletic traffic was applied at a rate of three events per week until a total of 25 simulated events (STE) were applied, modified slightly from the rate and total events described by Dalsgaard et al. (2020). Simulated traffic began on 14 Aug. 2020 and

12 Aug. 2021 to align with the prototypical start to the high school football season in Iowa, and ended on 19 Oct. 2020 and 15 Oct. 2021, respectively.

Data Collection

Athletic field performance was quantified by digital image analysis of percent green cover (PGC) and was measured with methods described by Dalsgaard et al. (2020). A light box was used to allow for consistent light quality across images like those described by Thoms et al. (2011). A Canon G9X, (Canon Inc. Ota, Japan) digital camera was used to collect images. Images were taken in the same location on each plot from year-to-year to avoid variance by location within plots. Images were analyzed using Turf Analyzer similar to Karcher et al. (2017). Which allowed green pixels in the images to be divided by the total number of pixels in the image to produce a value between 0 and 100%, measuring green cover in the image. Percent green cover data were taken before, after 10, 20 and 25 STE. Data collection after five STE was missed in both years of the study due to COVID-19 protocols at the University.

Surface hardness was tested with a 2.25 kg Clegg Impact Soil Tester (CIST; Lafayette Instrument Company, Lafayette, IN) similar to methods by Thoms et al. (2016). Rotational resistance (shear) followed methods described by Thoms et al. (2019) and was measured with a shear vane similarly to how the NFL tests for surface stability. These data points were randomly taken from each plot before, after 10, 20 and 25 STE.

Previous research has reported a negative correlation between surface hardness and soil moisture (Iwasaki et al., 2020), as a result, volumetric water content of the soil was tested with a Time Domain Reflectance Probe (TDR 300, Precision Technologies, Inc., Aurora, IL) at 7.5 cm depth at the same location as every CIST reading. Soil physical properties including bulk density and total porosity were evaluated to investigate any changes in the soil after 25 STE each year. Three cores were collected from random locations in each plot, then saturated through an extractor vacuum for 8 h. Saturated cores were weighed and allowed to dry to field capacity and weighed to determine bulk density (Db) and Total Porosity (TP) similar to previously used methods used by Dalsgaard et al. (2020).

Statistical Analysis

Data from this experiment were analyzed using an analysis of variance (ANOVA) in SAS version 9.4. There was a significant date interaction across all variables (**Table 2**), and as a result data will be presented for individual collection dates. Means were separated with Fisher's protected least significant difference at the 0.05 level of probability. For PGC, linear regression analysis was performed to determine slope and intercept for each treatment's loss of green cover (R^2=0.93). Orthogonal contrasts were completed to determine if each HOC reduction strategy, fertilizer rate, or use of PGR was significant to turf cover reduction resulting from simulated traffic events.

Results and Discussion

Results

Percent green cover data were pooled due to a lack of a treatment*year interaction **(Table 2)**. Maintenance regime differences for PGC were only present after 10 STE, when the 1/3 rule with 36.6 kg N ha^{-1} and no PGR lost (5%) less PGC than 1/3 rule with 73.2 kg N ha^{-1} and no PGR (**Table 3**). The only difference between these treatment regimes is the rate of N fertility. All other STE collection dates showed no differences between maintenance regimes. The loss of PGC was a linear relationship between STE (R^2 value 0.93) (**Table 3**). The 1/3 rule with 73.2 kg N ha^{-1} and no PGR (-25 PGC per 5 STE^{-1}) had the greatest PGC lost per 5 STE, while the 1/3 rule with 36.6 kg N ha^{-1} and PGR (-18 PGC per 5 STE^{-1}) had the least cover lost per 5 STE. Contrast statements were used to decipher differences in PGC by regime factor (HOC reduction, fertilizer rate, or PGR use) (**Table 4**). The 36.6 kg N ha^{-1} fertility rate resulted in 7% more PGC (less loss of PGC) than the 73.2 kg N ha^{-1} fertility rate after 10 STE (**Table 5**).

No STE rating dates in either year of the study had any differences in surface hardness values (GMAX) (**Table 6**).

Differences in rotational resistance were seen after 10 and 25 STE (**Table 7**). The 36.6 kg N ha^{-1} rate had 8% greater rotational resistance than the 73.2 kg N ha^{-1} after 10 and 25 STE (**Tables 7 and 8**).

Soil bulk density and total porosity were compared based on management regime factor, but no differences were found based on orthogonal contrast statements (**Table 9**).

Discussion

Decreased traffic tolerance due to a high N fertility rate has been shown previously (Ervin and Koski, 2001). Nitrogen rates past 200-300 kg N ha^{-1} yr^{-1} have been shown to decrease wear tolerance (Canaway, 1984). The total of 73.2 kg N ha^{-1} for 5 months from the return of maintenance activities to the end of the simulated football season results in an annual rate of 366 kg N ha^{-1} yr^{-1} while the 36.6 kg N ha^{-1} mo^{-1} rate had a 5-month total of 183 kg N ha^{-1} yr^{-1}. Plant growth regulator usage with traffic conflicts with findings from Haselbauer et al. (2010). Haselbauer et al. (2010), found PGR usage until 14 days before STE resulted in greater PGC after 20 STE than the no PGR, the difference could be that Haselbauer's work was on hybrid bermudagrass compared to Kentucky bluegrass in his study. Finding no differences between treatments for soil bulk density and total porosity agrees with results from Hasselbauer et al. (2010). Lindsey et al. (2021) also reported no differences in surface hardness due to differing fertility rates under simulated traffic. However, Lindsey's results showed that fertilizer rate didn't have an impact on shear strength, which contradicts the results of this study (Lindsey et al., 2021). Past findings have shown that excessive N results in forced shoot growth and limited root growth, which could explain the lower rotational resistance values on the 73.2 kg N ha^{-1} rate (Beard, 2001).

Conclusion

The minimum of a 2-month delay after achieving desired HOC before applying STEs may have allowed the turfgrass to acclimate and thus not result in STE stress to the degree that might be expected if STE occurred sooner after the desired HOC was reached. This 2-month delay could be potential reasoning for a lack of treatment effects across all metrics. 'Moonlight' Kentucky bluegrass that was treated with a 36.6 kg N ha^{-1} mo^{-1} either was the same or better than 73.2 kg N ha^{-1} mo^{-1} rate across many rating dates in rotational resistance, PGC, and surface hardness. These results suggest that a lower N rate, without PGR use and any HOC reduction method will result in a similar playing surface performance in the fall. This research is limited in scope due to the specific time of unplanned suspension of maintenance and STE. This study is modeled similarly to the high school football schedule commonly seen in the United States. Future research could test unplanned suspension of maintenance in different seasons, and test different timings between HOC reduction and STE.

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Figures and Tables

Table 1. List of management regimes (height-of-cut [HOC], nitrogen fertility, and plant growth regulator [PGR]) implemented on ‘Moonlight’ Kentucky bluegrass (*Poa pratensis* L.) after an unplanned suspension of maintenance at the Horticulture Research Station, Ames, IA in 2020 and 2021.

HOC Reduction Strategy	**Fertilizer Rate**[a]	**PGR**[b]
1/3 rule[c]	36.6 kg N ha^{-1}	No
1/3 rule	36.6 kg N ha^{-1}	Yes
Scalp[d]	36.6 kg N ha^{-1}	No
Scalp	73.2 kg N ha^{-1}	No
Cut in half, then 1/3 rule[e]	36.6 kg N ha^{-1}	Yes
Cut in half, then 1/3 rule	36.6 kg N ha^{-1}	No
Cut in half, then 1/3 rule	73.2 kg N ha^{-1}	Yes
Cut in half, then 1/3 rule	73.2 kg N ha^{-1}	No
1/3 rule	73.2 kg N ha^{-1}	No
1/3 rule	73.2 kg N ha^{-1}	Yes

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[a] Fertility was supplied with a 28-0-3 (N-P-K) fertilizer (Lesco Inc, Cleveland, Ohio) applied monthly following the start of maintenance 1 June – 1 Nov. each year.

[b] PGR was Trinexapac-ethyl (TE) (Primo Maxx®, 584.6 ml ha^{-1} ,Syngenta, Basel, Switzerland), was either applied as part of the maintenance regime every two-weeks following the resumption of maintenance at 584.6 ml ha^{-1} until two weeks before simulated athletic traffic 1 Aug. 2020 and 6 Aug. 2021. TE was applied with a CO2-pressurized backpack sprayer with TeeJet 8004XR nozzles calibrated to apply 815 L water carrier ha^{-1}

[c] ⅓ rule- remove ⅓ of the plant material in a single mowing to get desired HOC with two mowing events a week.

[d] Scalp- removed all leaf tissue to the desired 5 cm HOC in one mowing event.

[e] Cut in half, then ⅓ rule- removed half of the vertical height with the first mowing, and every subsequent mowing would only remove ⅓ of the remaining leaf tissue until the desired HOC with two mowing events per week.

Table 2. Modified analysis of variance (ANOVA) describing the effects of replication, year, treatment regime and date and their interactions for surface hardness with a Clegg Impact Soil Tester (CIST), Percent green cover (PGC) as determined with digital image analysis, rotational resistance (Shear) and soil volumetric water content (TDR). Treatment effects include height-of-cut (HOC) reduction, nitrogen rate, and plant growth regulator regimes tested for turfgrass performance and safety to fall simulated athletic field traffic in Ames, IA in 2020 and 2021.

	df	CIST	PGC	Shear	TDR
Rep (R)	3	***	**	NS	***
Year (Y)	1	***	**	***	***
Y*R	3	***	NS	***	NS
Treatment(T)	9	**	***	**	*
Y*T	9	NS	NS	NS	NS
Y*R*T	54	**	***	NS	***
Date (D)	3	***	***	***	***
Y*D	3	***	***	***	***
T*D	27	NS	NS	NS	NS
Y*T*D	27	NS	NS	NS	NS

[a] Surface hardness was measured with a Clegg Impact Soil Tester equipped with a 2.25 kg missile and accelerometer in units of GMAX with the average of three random locations per plot.

[b] PGC is evaluated using digital image analysis (DIA) from a picture taken before, and after 10, 20 and 25 simulated traffic events. Digital image analysis was performed using TurfAnalyzer.

[c] Rotational resistance (shear) followed methods described by Thoms et al. (2021) and was measured with a shear vane similarly to how the National Football League tests for surface stability. One data point was randomly taken from each plot on the same schedule as PGC.

[d] Volumetric water content of the soil was tested with a Time Domain Reflectance Probe (TDR 300, Spectrum Technologies, Inc., Aurora, IL) at 7.5 cm depth at the same location as every CIST reading.

[e] *** denotes a significant difference with a p-value ≤ 0.001.

[f] ** denotes a significant difference with a p-value ≤ 0.01.

[g] * denotes a significant difference with a p-value ≤ 0.05.

[h] NS denotes no significance.

[I] HOC reduction included three treatments: never removing more than 1/3 of leaf tissue at a time, scalp to 5 cm in a single mowing and reduce HOC in half and then use 1/3 rule with every mowing following to reduce HOC to final 5 cm HOC. Fertility was supplied with a 28-0-3 (N-P-K) fertilizer (Lesco Inc, Cleveland, Ohio) applied monthly following the start of maintenance at rates of either 36.6 kg N ha^{-1} mo^{-1} or 73.2 kg N ha^{-1} mo^{-1} during the growing season (1 June- 1 Nov.). PGR = plant growth regulator was Trinexapac-ethyl (TE) applied at 584.6 ml ha^{-1} applied with TeeJet 8004XR nozzles calibrated to apply 815 L water carrier ha^{-1}. TE was applied until two weeks before simulated athletic traffic 1 Aug. 2020 and 6 Aug. 2021.

Table 3. Percent green cover (PGC) of 'Moonlight' Kentucky bluegrass (*Poa pratensis* L.) grown on a native soil. A linear regression is given to demonstrate the PGC lost for every five simulated traffic events. Percent green cover was pooled for years 2020 and 2021 in Ames, IA.

Regime	**% Green Cover lost every 5 STE** [a]	**Intercept**
1/3 rule[b] for height-of-cut (HOC) reduction with 36.6 kg N [c] ha^{-1} without plant growth regulator (PGR) [d]	-20	121.2
1/3 rule for HOC reduction with 36.6 kg N ha^{-1} with PGR [f]	-18	118.1
Scalp[e] HOC reduction with 36.6 kg N ha^{-1} without PGR	-20	119.5
Scalp HOC reduction with 73.2 kg N ha^{-1} without PGR	-22	121.1
Cut-in-half then 1/3 rule HOC reduction with 36.6 kg N ha^{-1} with PGR	-24	122.8
Cut-in-half then 1/3 rule HOC reduction with 36.6 kg N ha^{-1} without PGR	-19	118.3
Cut-in-half then 1/3 rule HOC reduction with 73.2 kg N ha^{-1} with PGR	-21	117.7
Cut-in-half then 1/3 rule HOC reduction with 73.2 kg N ha^{-1} without PGR	-19	117.5
1/3 rule HOC reduction with 73.2 kg N ha^{-1} without PGR	-25	119.7
1/3 rule HOC reduction with 73.2 kg N ha^{-1} with PGR	-19	119.8

[a] R^2 value for linear regression for percent green cover was 0.93. Linear regression was chosen because it had the highest correlation of any other line of fit. Simulated traffic events (STE) simulate football game traffic using a Modified Baldree Traffic Simulator. Three STE were performed per week for 25 events, data was taken before and after 10, 20 and 25 STE.

[b] ⅓ rule- never removed more than ⅓ of the plant material in a single mowing to get to desired HOC.

[c] Fertility was supplied with a 28-0-3 (N-P-K) fertilizer (Lesco Inc, Cleveland, Ohio) applied monthly following the start of maintenance during the growing season (1 June -1 Nov.).

[d] Trinexapac-ethyl (Primo Maxx®, 584.6 ml ha^{-1},Syngenta, Basel, Switzerland), was either applied or not as part of the maintenance regime every two-weeks following the resumption of maintenance at 584.6 ml ha^{-1}. Trinexapac-

ethyl was applied with a CO_2 sprayer using an H_2O carrier.

[e] Scalp- removed all leaf tissue to the desired HOC (5.08cm) in one mowing event.

[f] ½, then ⅓ rule- removed half of the vertical height with the first mowing, and every subsequent mowing would only remove ⅓ of the remaining leaf tissue until the desired HOC.

Table 4. Orthogonal Contrast Statements used to differentiate based on maintenance regime factors to determine differences in percent green cover (PGC[a]) from 2020 and 2021 in Ames, IA.

Maintenance Regime Factor	**STE[b]**			
	0	**10**	**20**	**25**
1/3 rule[c] vs. ½ then 1/3[d]	NS[e]	NS	NS	NS
1/3 rule vs. scalp[f]	NS	NS	NS	NS
½ then 1/3 vs. scalp	NS	NS	NS	NS
PGR[g] vs. None	NS	NS	NS	NS
Fertilizer rate[h]	NS	**[i]	NS	NS

[a] PGC is evaluated using digital image analysis (DIA) from a picture taken before, and after 10, 20 and 25 STE. Digital image analysis was performed using Images were analyzed using TurfAnalyzer similar to Karcher et al. (2017).

[b] Simulated traffic events (STE) simulate football event traffic using a Modified Baldree Traffic Simulator. Three STE were performed per week for 25 events, data was taken before, after 10, 20 and 25 STE from 14 Aug. to 19 Oct. 2020 and 12 Aug. to 15 Oct. 2021.

[c] ⅓ rule- never removed more than ⅓ of the turfgrass plant material in a single mowing to get to desired 5 cm HOC with two mowing's per week

[d] ½, then ⅓ rule- removed half of the vertical turfgrass height with the first mowing, and every subsequent mowing would
only remove ⅓ of the remaining leaf tissue until the desired 5 cm HOC with two mowing's per week.

[e] NS = not significantly different.

[f] Scalp- removed all leaf tissue to the desired HOC (5cm) in one mowing event and then maintain at 5 cm.

[g] PGR = plant growth regulator was Trinexapac-ethyl (TE) applied at 584.6 ml ha^{-1} applied with TeeJet 8004XR nozzles calibrated to apply 815 L water carrier ha^{-1}. TE was applied until two weeks before simulated athletic traffic 1 Aug. 2020 and 6 Aug. 2021.

[h] Fertility was supplied with a 28-0-3 (N-P-K) fertilizer (Lesco Inc, Cleveland, Ohio) applied monthly following the start of maintenance at rates of either 36.6 kg N ha^{-1} or 73.2 kg N ha^{-1} during the growing season (1 June -1 Nov.).
h

[i] ** denotes a significant difference with a p-value ≤ 0.01.

Table 5. Percent green cover as determined by digital image analysis for two fertility rates after 10 simulated traffic events for ‘Moonlight’ Kentucky bluegrass (*Poa pratensis* L.) in Ames, IA in 2020-2021.

Fertilizer Rate [a]	**Mean 10 STE** [b]
36.6 kg N ha^{-1}	89.7
73.2 kg N ha^{-1}	84.4
LSD (0.05)	4.8

[a] Fertility was supplied with a 28-0-3 (N-P-K) fertilizer (Lesco Inc, Cleveland, Ohio) applied monthly following the start of maintenance at rates of either 36.6 kg N ha^{-1} mo^{-1} or 73.2 kg N ha^{-1} during the growing season (1 June-1 Nov.).

[b] Simulated traffic events (STE) simulate football game traffic using a Modified Baldree Traffic Simulator. Three STE were performed per week for 25 events.

Table 6. Surface hardness values tested with orthogonal contrast statements for various maintenance regime factors applied to 'Moonlight' Kentucky bluegrass (*Poa pratensis* L.) under simulated athletic field traffic with the 2.25 kg Clegg Impact Soil Tester (CIST)[a] in Ames, IA from fall of 2020 and 2021.

Regime Factor	**STE[b]**			
	0	**10**	**20**	**25**
1/3 rule[c] vs. 1/2 then 1/3[d]	NS[e]	NS	NS	NS
1/3 rule vs. scalp[f]	NS	NS	NS	NS
1/2 then 1/3 vs. scalp	NS	NS	NS	NS
PGR[g] vs. None	NS	NS	NS	NS
Fertilizer rate[h]	NS	NS	NS	NS

[a] The Clegg Impact Surface Tester (CIST) measures surface hardness and is measured in GMAX,.with three measurements per plot and averaged for each rating.

[b] Simulated traffic events (STE) simulate football event traffic using a Modified Baldree Traffic Simulator. Three STE were performed per week for 25 events, data was taken before and after 10, 20 and 25 STE from 14 Aug. to 19 Oct. 2020 and 12 Aug. to 15 Oct. 2021.

[c] ⅓ rule- never removed more than ⅓ of the turfgrass plant material in a single mowing to get to desired 5 cm HOC with two mowing's per week

[d] ½, then ⅓ rule- removed half of the vertical turfgrass height with the first mowing, and every subsequent mowing would

only remove ⅓ of the remaining leaf tissue until the desired 5 cm HOC with two mowing's per week.

[e] NS = not significantly different.

[f] Scalp- removed all leaf tissue to the desired HOC (5cm) in one mowing event and then maintain at 5 cm.

[g] PGR = plant growth regulator was Trinexapac-ethyl (TE) applied at 584.6 ml ha^{-1} applied with TeeJet 8004XR nozzles calibrated to apply 815 L water carrier ha^{-1}. TE was applied until two weeks before simulated athletic traffic 1 Aug. 2020 and 6 Aug. 2021.

[h] Fertility was supplied with a 28-0-3 (N-P-K) fertilizer (Lesco Inc, Cleveland, Ohio) applied monthly following the start of maintenance at rates of either 36.6 kg N ha^{-1} or 73.2 kg N ha^{-1} during the growing season (1 June -1 Nov.).

Table 7. Effects of various maintenance regimes on Kentucky bluegrass (*Poa pratensis* L.) under simulated traffic for rotational resistance[a] tested with orthogonal contrasts in Ames, IA from 2020 and 2021.

Regime Factor	**STE[b]**			
	0	**10**	**20**	**25**
1/3 rule[c] vs. 1/2 then 1/3[d]	NS[e]	NS	NS	NS
1/3 rule vs. scalp[f]	NS	NS	NS	NS
1/2 then 1/3 vs. scalp	NS	NS	NS	NS
PGR[g] vs. None	NS	NS	NS	NS
Fertilizer rate[h]	NS	**[i]	NS	**

[a] Rotational resistance (shear) followed methods described by Thoms et al. (2021) and was measured with a shear vane similarly to how the National Football League tests for surface stability. one data point was randomly taken from each plot before, after 10, 20 and 25 simulated traffic events.

[b] Simulated traffic events (STE) simulate football event traffic using a Modified Baldree Traffic Simulator. Three STE were performed per week for 25 events, data was taken before and after 10, 20 and 25 STE from 14 Aug. to 19 Oct. 2020 and 12 Aug. to 15 Oct. 2021.

[c] ⅓ rule- never removed more than ⅓ of the turfgrass plant material in a single mowing to get to desired 5 cm HOC with two mowing's per week

[d] ½, then ⅓ rule- removed half of the vertical turfgrass height with the first mowing, and every subsequent mowing would

only remove ⅓ of the remaining leaf tissue until the desired 5 cm HOC with two mowing's per week.

[e] Scalp- removed all leaf tissue to the desired HOC (5cm) in one mowing event and then maintain at 5 cm.

[f] NS = not significantly different.

[g] PGR = plant growth regulator was Trinexapac-ethyl (TE) applied at 584.6 ml ha^{-1} applied with TeeJet 8004XR nozzles calibrated to apply 815 L water carrier ha^{-1}. TE was applied until two weeks before simulated athletic traffic 1 Aug. 2020 and 6 Aug. 2021.

[h] Fertility was supplied with a 28-0-3 (N-P-K) fertilizer (Lesco Inc, Cleveland, Ohio) applied monthly following the start of maintenance at rates of either 36.6 kg N ha^{-1} or 73.2 kg N ha^{-1} during the growing season (1 June -1 Nov.).

[i] ** denotes a significant difference with a p-value ≤ 0.01.

Table 8. Rotational resistance as determined by a shear vane on 'Moonlight' Kentucky bluegrass after 10 and 25 simulated traffic events (STE)[a] with two different fertility rates in Ames, IA in 2020-2021.

Fertilizer Rate [b]	**Mean**	
	10 STE	**25 STE**
36.6 kg N ha^{-1}	18.55 N·m[c]	23.54 N·m
73.2 kg N ha^{-1}	17.14 N·m	22.25 N·m
LSD (0.05)	0.34	0.54

[a] Simulated traffic events (STE) simulate football event traffic using a Modified Baldree Traffic Simulator. Three STE were performed per week for 25 events, data was taken before and after 10, 20 and 25 STE from 14 Aug. to 19 Oct. 2020 and 12 Aug. to 15 Oct. 2021.

[b] Fertility was supplied with a 28-0-3 (N-P-K) fertilizer (Lesco Inc, Cleveland, Ohio) applied monthly following the start of maintenance during the growing season (1 June -1 Nov.).

[c] Rotational resistance (shear) followed methods described by Thoms et al. (2021) and was measured with a shear vane similarly to how the National Football League tests for surface stability. One data point was randomly taken from each plot before.

Table 9..Bulk density and Total porosity values tested with orthogonal contrast statements for various maintenance regime factors on 'Moonlight' Kentucky bluegrass (*Poa pratensis* L.) under simulated athletic field traffic in Ames, IA in 2020 and 2021.

Regime Factor	**Measurement**[a]	
	Bulk Density	Porosity
1/3 rule [b] vs. 1/2 then 1/3[c]	NS[d]	NS
1/3 rule vs. scalp[e]	NS	NS
1/2 then 1/3 vs. scalp	NS	NS
PGR[f] vs. None	NS	NS
Fertilizer rate[g]	NS	NS

[a] Bulk density and total porosity were evaluated to investigate any changes in the soil after 25 STE each year. Three cores were collected from random locations in each plot, then saturated through an extractor vacuum for 8 h. Saturated cores were weighed and allowed to dry to field capacity and weighed to determine bulk density (Db) and Total Porosity (TP) similar to previously used methods used by Dalsgaard et al. (2019).

[b] ⅓ rule- never removed more than ⅓ of the turfgrass plant material in a single mowing to get to desired 5 cm HOC with two mowing's per week.

[c] ½, then ⅓ rule- removed half of the vertical turfgrass height with the first mowing, and every subsequent mowing would

only remove ⅓ of the remaining leaf tissue until the desired 5 cm HOC with two mowing's per week.

[d] NS = not significantly different.

[e] Scalp- removed all leaf tissue to the desired HOC (5cm) in one mowing event and then maintained at 5 cm.

[f] PGR = plant growth regulator was Trinexapac-ethyl (TE) applied at 584.6 ml ha^{-1} applied with TeeJet 8004XR nozzles calibrated to apply 815 L water carrier ha^{-1}. The TE was applied until two weeks before simulated athletic traffic 1 Aug. 2020 and 6 Aug. 2021.

[g] Fertility was supplied with a 28-0-3 (N-P-K) fertilizer (Lesco Inc, Cleveland, Ohio) applied monthly following the start of maintenance at rates of either 36.6 kg N ha^{-1} or 73.2 kg N ha^{-1} during the growing season (1 June -1 Nov.).

CHAPTER 3. EVALUATING THE SUSTAINABILITY OF TURFGRASS LAWN ASSEMBLAGES

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Abstract

Turfgrasses are estimated to cover 2% of the United States land area, it is important to ensure lawns are sustainable while still maintaining ecosystem services). Previous research has evaluated the sustainability of mixing species and cultivars. Cultivar performance was not accounted for in prior studies. Sixteen cultivars were selected from the National Turfgrass Evaluation Program (NTEP) trials based on turfgrass quality, and commercial availability [4 Kentucky bluegrass (*Poa pratensis* L.; KBG) cultivars, 4 tall fescue (*Schedonorus arundinaceus* Scherb.; TF) cultivars, 4 fine fescue (*Festuca* spp.; FF) cultivars, 4 perennial ryegrass (*Lolium perenne* L.; PR) cultivars]. Cultivars were grouped into various assemblage types (monocultures, 3-species mixes, 3-cultivar blends and 3-species by 3-cultivar mixes). Turfgrass assemblages were all similar in drought tolerance, soil organic carbon and soil microbial biomass. Three cultivar assemblages exhibited higher percent green cover from 42-days after seeding until the end of establishment. Three species and three cultivar assemblages leached similar or lower nitrate than 3-species by 3-cultivar assemblages and monocultures.

Introduction

Turfgrasses provide many beneficial ecosystem services and benefits to humans. Including urban heat island dissipation, lessened weed pressure and allergens, CO_2 conversion, reduced erosion, groundwater filtration, and a low-cost space for safe recreation (Beard and Green, 1994). Additionally, they absorb pollutants, mitigate runoff, and enhance mental health, though concerns about water usage, pesticides, and lack of suitable wildlife are often raised (Stier et al., 2013). Turfgrass has evolved from usage as a forage grass and cover crop to now being used by homeowners, golf courses and sports as a crossroads between nature and recreation (Adams and Gibbs, 1994; Beard, 1998). Turfgrasses are estimated to cover 2% of the United States land area, it is important to ensure lawns are sustainable while still maintaining ecosystem services (Robbins and Birkenholtz, 2003). Upon comparison to hardscapes, concrete or asphalt in an urban environment, the ecosystem services turfgrass provides become much more pronounced (Christians et al., 2017).

Reliance on our ecosystem services is essential to survival, sustainable practices help promote a healthy ecosystem and those in it (Zhang et al., 2010b). Humans are connected to nature emotionally, philosophically, experientially, materially, and promotion of connecting with nature is becoming increasingly popular (Ives et al., 2018). Proper establishment for turfgrasses are important for the reduction of erosion and inclusion of ecosystem services turfgrass provides (Christians et al., 2017). Species and cultivar selection is important for turfgrass performance, and speed to proper establishment must be considered to help avoid additional erosion and nutrient losses (Christians et al., 2017).

Turfgrass aesthetics are important for property values for homeowners, people aged 45 to 64 prefer landscapes with fewer trees and more turfgrass (Des Rosiers et al., 2002). Home values with quality landscaping have higher home valuations than homes with lackluster landscaping (Henry, 1994; McKenzie, 2005). Observing natural areas like manicured turfgrass increases mental functioning and sense of tranquility (Frumkin, 2001). A study by Moore (1981) showed that prisoners that had a window view of greenspace had 24% fewer sick visits than prisoners who didn’t have a view overlooking greenery. Maintained turfgrasses attract recreation and leisure, physical activity, and provide a softer surface than paved surfaces (Coady and Micheli, 1997).

Cool-season turfgrass lawns in the northern United States often have a mixture of species and cultivars to account for changing shade conditions and disease resistance (Stier et al., 2013). In sports fields it is typical to see multiple cultivars of a species blended to attempt to provide benefits of having diverse genetics and traits (Vargas and Turgeon, 1980; Brede, 2007). Ecologically speaking, diversity of species influences ecosystem function, and that more species provide ecosystem services more reliably in ever-changing environments (Hooper et al., 2005).

The basis of this study is inspired by the work of Thompson and Kao-Kniffin (2016), where they found that with increased turfgrass assemblage diversity, came decreased nitrate leaching, and increased microbial diversity. Basis for cultivar selection was not based on cultivar performance and incorporated some species that would be deemed weeds in a turfgrass setting. Turfgrass research is constantly improving cultivars for disease resistance, lower inputs, and drought tolerance through the National Turfgrass Evaluation Program (NTEP) with testing locations across the United States (National Turfgrass Evaluation Program, 2020). Based on these results, cultivars used commercially are often selected from this testing program. Due to the biodiversity and ecosystem function theory, it was hypothesized that more genetic diversity would result in less inorganic nitrate and ammonium leaching (Thompson and Kao-Kniffin, 2016). Additionally, it was hypothesized the greater the number of species and varieties, the higher the amount of soil organic carbon and overall turfgrass quality. Finally, it was hypothesized that greater drought tolerance would be observed with greater diversity. Differing root architecture among species and cultivars was believed to be the reasoning for improved soil health and performance (Bowman et al., 2002). The increased stability from a more genetically diverse assemblage would decrease pest pressure (Naeem, 2002).

Materials and Methods

Experimental Design and Treatments

A greenhouse trial was conducted at the Iowa State University Horticulture Greenhouses in Ames, Iowa in 2021. Sixteen cultivars [four Kentucky bluegrass (*Poa pratensis* L.; KBG), four tall fescue (*Schedonorus arundinaceus* Scherb.; TF), four fine fescue (*Festuca spp.;* FF) and 4 perennial ryegrass (*Lolium perenne* L.; PR)] were selected from the NTEP trials due to the highest turfgrass quality, and commercially available varieties for their respective species (National Turfgrass Evaluation Program, Beltsville, Maryland). These species were chosen based on the commonly found turfgrasses in American cool-season lawns. Thirty-four turfgrass assemblages were evaluated and organized in a 12 x 34 randomized complete block design (RCBD), all 16 cultivars were grown in monoculture, to operate as a control for mixes and blends observed in other treatments. This experiment also compared randomly selected cultivars from a mix of three different species, as well as a mix of three cultivars*three species. Finally, all species had three treatments with three randomly selected cultivars. These treatments were randomly selected using Microsoft Excel (Microsoft Corporation, Redmond, WA). In total there were 34 treatments, each containing 12 replications (n=407). Six replications were be used to evaluate establishment and density (**Table 1**). Once establishment occurred, those replicates were destructively harvested to analyze soil organic matter with loss on ignition following methods described by Thoms et al. (2011). The other six replicates were put through a simulated drought for 28-days to evaluate drought tolerance, then destructively harvested to test for soil organic matter and microbial biomass.

Establishment

Each treatment was grown in a 10.16 x 10.16 x 10.16 cm plastic pot in a topsoil-like media (Baccto top soil, Houston, Texas) compacted to a bulk density of 1.3 g cm^{-3}. The top 0.5 cm of the pot was left without media to ensure seeds and water remained in each pot. Each treatment was seeded with ≈300 seeds, which is the average seeding density for 103.23 cm^2. Treatments were seeded on 26 Mar. 2021.

Greenhouse Conditions

The Greenhouse conditions were maintained at 22°C with supplemental light used to extend the daylength to 16 hours when necessary. Supplemental light was provided by high pressure sodium (HPS) lamps at a light intensity of 300 to 350 $\mu mol \cdot m^{-2} \cdot s^{-1}$. Light intensity and temperature were autonomously regulated using an Argus Control System (White Rock, British Columbia, Canada). All treatments were irrigated with overhead irrigation 5 times $week^{-1}$ at a rate of 0.5 cm irrigation $event^{-1}$. Irrigation amounts totaled 2.5 cm $week^{-1}$ ensured turfgrass establishment but also limited excess watering and nutrient leaching.

Establishment Rate and Maturity Tracking

Establishment was tracked every three-days using digital image analysis (DIA) to measure percent green cover (PGC), modified slightly from the methods of Pease et al. (2022). Percent Green Cover was tracked for the first 54-days until establishment was completed. A photobox was used similar to methods used by Thoms et al. (2011) each time to capture the digital images and ensure consistent lighting and distance. Digital images were analyzed for percent green cover in each container and compared using TurfAnalyzer (hue 71-176 saturation

10 to 100) with a Cannon G9X (Canon Inc. Ota, Japan) (Karcher et al., 2017). Turfgrass pots were deemed established at 90% green cover, modified from Pease et al. (2022) and in accordance with Iowa state regulations (Iowa State Department of Natural Resources, 2018).

Once turfgrass was established, two clipping yields were taken to track above ground biomass growth. Each pot was trimmed with hand shears and clippings collected. Every five weeks (weeks 5, and 10, after 90% cover was reached across all assemblage types) clippings were collected weighed (wet weight) and then dried (dry weight) at 40ºC for 72 hours, which was modified slightly from Thompson Kao-Kniffin (2016). Treatments were mowed once a week with grass shears at a height of 7.62 +/- 0.5 cm to align with the average homeowner's height-of-cut (HOC). Mowing began as soon as treatments grew above the 7.62 cm threshold. Treatments were left unmown for two weeks prior to clipping collection to allow for growth to be adequately measured, modified from methods described by Thompson and Kao-Kniffin (2016).

Every four weeks after the establishment period (weeks eight, 12, and 16 weeks after seeding), nitrogen leaching was measured using a modified pour through method similar to Whipker et al. (2001). Where distilled water was used to saturate the soil, and subsequent drainage was collected and analyzed similar to methods used on turfgrass before with Pease et al. (2022). Both inorganic nitrate and inorganic ammonium leaching was measured using methods similar to Studt et al. (2021). A synergy HTX™ Multi-Mode Microplate reader (Agilent Technologies, Santa Clara, CA) was used to colorimetrically measure of treatments combined with ammonium salicylate and ammonia cyanurate reagent packets for inorganic ammonium (Hach Company). The Synergy HTX™ Multi-Mode Microplate reader was also used to analyze inorganic nitrate when the solution is combined with a vanadium solution and allowed to incubate in the dark for five hours. At the end of the 16-week establishment and normal growth period, six replicates were destructively harvested to collect total standing biomass. Additionally, soil microbial biomass was measured similar to the methods of Studt et al. (2021) and McDaniel and Grandy (2016). Where 5 grams of field moist soil is fumigated with chloroform for 24-hours compared to a set of samples that isn't fumigated. The difference in the dissolved organic C and N were run on a TOC-TN analyzer (Shimadzu Corporation, Kyoto, Japan) and compared to measured amounts of microbial biomass of a soil (Shimadzu Scientific Instruments Inc. Columbia, MD, USA). Upon destructive harvest, lost on ignition (LOI) was used to measure the amount of organic carbon in the soil after the establishment and eight-weeks of maturity with methods similar to Nelson and Sommers, (1996).

Drought Tolerance

After the first six replications were destructively harvested, another six replications were subjected to a simulated drought. Irrigation was terminated and over four weeks DIA was taken every three-days to track drought stress in the treatments. Percent green cover was tracked to determine when treatments completely undergo drought-induced dormancy (Richardson et al., 2001). Once all treatments reached dormancy, above ground biomass was collected and compared.

Statistical Analysis

Data collected were analyzed using the analysis of variance (ANOVA) in SAS version 9.4. Means were separated using Fishers protected least significant difference at the 0.05 probability level. There was an observed treatment x date interaction, as a result data will be presented by individual collection dates. For DIA data, linear regression was used to determine slope and intercept for each treatment to show an increase and decrease of PGC. Orthogonal contrasts were used to determine if there were significant differences between monocultural mixes, species mixes, cultivar mixes or species and cultivar mixes.

Results and Discussion

Establishment and Plant Productivity

Due to the nature of all 34 treatments falling into a normal distribution, treatments were pooled by assemblage type to best compare performance based on plant diversity. No differences in PGC were present for 9 of the 17 collection dates (**Table 2**). Monocultures had a 5% greater PGC than the three species at 6-days after seeding (DAS), and the 3-species*3-cultivar mix (42%) had greater PGC than the three species mix (32%) at 9 DAS. At 39 DAS, the three cultivar blend assemblages (63%) had increased PGC than the 3 species*3 cultivars mix (53%) and monocultures (52%). The three-cultivar mix (76%, 81%, 90%, 101%, and 105%, respectively) performed better than all other turfgrass assemblages during the last five collection dates (**Table 2**).

Clipping yields for the 3-cultivar blends were the highest for both collection dates (**Table 3**). The clipping yield of the 3-cultivar blends (0.016g) was greater than the 3 species*3 cultivar mixes (0.001g) five WAS, and greater than all other assemblages 10 WAS (**Table 3**).

No differences in nitrate leaching were seen between any turfgrass assemblage types for two of the three collection dates (**Table 4**). At the 12 WAS, the monoculture assemblages (0.09 ppm) had greater nitrate leaching, followed by the 3-species*3-cultivar mixes (0.08 ppm) while the 3-species mixes (0.07 ppm) and the 3-cultivar blends (0.07 ppm) had the least amount of nitrate leaching (**Table 4**).

Ammonium leaching was greatest among the 3-cultivar mixes for 2 of the 3 collection dates, with the 3-species mixes resulting in the greatest ammonium leaching at 12 WAS (**Table 5**). The assemblage type that demonstrated the least ammonium leaching differed between each collection date. There isn't an observed theme of ammonium leaching compared to quantity of species or cultivars across all dates.

There were no differences in microbial biomass or organic carbon in the soil after 16 weeks after seeding. (**Table 6**).

Simulated Drought

No differences in turfgrass assemblage types were reported across all dates of simulated drought conditions for PGC (**Table 7**).

Discussion

Turfgrass assemblage types made no difference regarding drought tolerance. This differs from Kanapeckas et al. (2008) who reported differing turfgrass species makes a difference in drought tolerance, with ornamentality ranging from 41.1% to 16.7% after a 75-day drought. Potential reasoning for similar drought tolerance is nature of the high performing cultivars chosen, and that cultivars were evaluated for turfgrass quality by similar traits in NTEP trials. Cultivars were bred and selected for similar traits like dark green color, drought tolerance and vigor.

Microbial biomass was found to be similar across all treatments, which differs from the results shown from Thompson and Kao-Kniffin (2016) but is similar to results found by Groffman et al. (1996) that shows differences in microbial biomass are determined more by soil type. Since our soil type differed greatly from the native soils of the Northeastern United States, we observed microbial biomass values 4.8 times higher than Groffman et al. (1996).

Inorganic ammonium leaching was inconsistent between dates. Previous literature has shown ammonium leachate levels are negligible, or inconsistent in turfgrass ecosystems (Mancino and Troll, 1990; Bowman et al., 2002)

The three-species and three-cultivar assemblage types performed similarly or better (0.07 ppm for both) when evaluating nitrate leaching than the 3S*3C and monoculture assemblages (0.08 and 0.09 ppm, respectively). The highest diversity assemblage type not performing the best differs from Thompson and Kao-Kniffin (2016) and Tilman et al. (1997). Our findings indicate a relationship between species richness and nutrient retention aren’t necessarily linear unlike previous studies (Hooper et al., 2005; Zhang et al., 2010a; Tilman et al., 1996).

The 3C assemblages had greater PGC than all other assemblage types after 42 DAS. This could be explained by a potential synergistic relationship between multiple cultivars within a blend (Brede, 2007). Brede (2007) found that 9 blend combinations performed better than a monoculture of ‘Award’, showing the potential for between cultivars blends to perform better than monocultures.

It is without mentioning that potential disease pressure associated with limited diversity of turf assemblages coincides with biodiversity and ecosystem functioning theory (Cardinale et al., 2013), the principal findings conclude that 3C blends performed similarly or better than all other assemblage types in most rating dates. Vargas and Turgeon (1980) found that combining KBG cultivars provided similar or mildly more disease resistance. Whitman et al. (2022) reported that cultivar blends perform better than monocultures, but additional, more diverse assemblages like 3S or 3S*3C performing similarly or worse contradicts Thompson and Kao-Kniffin (2016).

Sampling effects may have played a factor in the mix and blend treatments, meaning with more varieties there is an increased likelihood that a high performing selection is included (Wardle, 1999). With this study, we believe these effects are minimized due to the selection of high performing cultivars, and that differences in PGC during establishment between species weren't significant. Additionally for the metrics of organic carbon, and microbial biomass, high performing varieties aren't necessarily beneficial to all facets of biological function (van der Heijden et al., 1999). Since no significant differences were found for microbial biomass and soil organic carbon, further proving sampling effects were minimized (**Table 6**).

The limitations for this study should also be noted. Greenhouse conditions were idealized for this experiment prior to simulated drought, and the 1048cm^3 pot size only allows to evaluate a brief moment of varietal expression during establishment and maturation phases of development, in order to truly evaluate assemblage differences in elite turfgrasses in all phases of maturity, a long-term field trial would be necessary.

Conclusion

Findings from this study contradict many of the findings for previous work for diversity and species, however improved cultivars used in this study could have resulted in the differences from previous work. It can be noted that improved cultivars result in few differences for drought tolerance, microbial biomass, organic carbon and ammonium leaching, while offering improved PGC during this short greenhouse study. Species and cultivar diversity can offer lower ammonia leaching during the early stages of turfgrass growth. Future research should investigate cultivar and species diversity studies on established stands of turfgrass.

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Tables and Figures

Table 1. Turfgrass monocultures, blends and mixes used to evaluate turfgrass diversity contributions to sustainability or performance of cool-season turfgrasses in the Horticulture Hall Greenhouses in Ames, IA in 2021.

Treatment #	Turfgrass Cultivar and Species Treatments[a]
1	‘STARR’ Kentucky Bluegrass (*Poa pratensis* L.) [KBG]
2	‘AFTER MIDNIGHT’ KBG
3	‘SKYE’ KBG
4	‘BASERATTI’ KBG
5	‘DRAGSTER’ (*Schedonorus arundinaceus* Schreb.) [TF]
6	‘BULLSEYE’ TF
7	‘RAPTOR III’ TF
8	‘HEMI’ TF
9	‘COMPASS II’ (Festuca spp.) [FF]
10	‘RADAR’ FF
11	‘SEAMIST’ FF
12	‘JETTY’ FF
13	‘SLUGGER 3GL’ (Lolium perenne L.) [PR]
14	‘FIESTA CINCO’ PR
15	‘FASTBALL 3GL’ PR
16	‘APPLE 3GL’ PR
17	‘SKYE’ KBG*’RAPTOR’ III TF* ‘COMPASS II’ FF
18	‘RAPTOR III’ TF*’COMPASS II’*’FASTBALL 3GL’ PR
19	‘SKYE’ KBG*’COMPASS II’ FF*’FASTBALL 3GL’ PR
20	KBG (‘STARR’* ‘SKYE’* ‘BASERATTI’)*TF (‘DRAGSTER’* ‘BULLSEYE’* ‘HEMI’) *FF (‘RADAR’* ‘SEAMIST’* ‘JETTY’)
21	TF (‘DRAGSTER’* ‘BULLSEYE’* ‘HEMI’) *FF (‘RADAR’* ‘SEAMIST’* ‘JETTY’)*PR (‘SLUGGER 3GL’* ‘FIESTA CINCO’* ‘FASTBALL 3GL’)
22	KBG (‘STARR’* ‘SKYE’* ‘BASERATTI’)* FF (‘RADAR’* ‘SEAMIST’* ‘JETTY’)* PR (‘SLUGGER 3GL’* ‘FIESTA CINCO’* ‘FASTBALL 3GL’)
23	KBG ‘STARR’* ‘AFTER MIDNIGHT’* ‘SKYE
24	KBG ‘STARR’* ‘AFTER MIDNIGHT’* ‘BASERATTI’
25	KBG ‘STARR’* ‘SKYE’* ‘BASERATTI’
26	TF ‘DRAGSTER’* ‘BULLSEYE’* ‘RAPTOR III’
27	TF ‘BULLSEYE’* ‘RAPTOR III’* ‘HEMI’
28	TF ‘DRAGSTER’* ‘BULLSEYE’* ‘HEMI’
29	FF ‘COMPASS II’* ‘RADAR’* ‘SEAMIST’
30	FF ‘RADAR’* ‘SEAMIST’* ‘JETTY’
31	FF ‘COMPASS II’* ‘SEAMIST’* JETTY
32	PR ‘SLUGGER 3GL’* ‘FIESTA CINCO’* 'APPLE 3GL’

Table 1. Continued.

33	PR ‘FIESTA CINCO’* ‘FASTBALL 3GL’* ‘APPLE 3GL’
34	PR ‘SLUGGER 3GL’* ‘FASTBALL 3GL’* ‘APPLE 3GL’

[a] Each treatment was grown in a 10.16 x 10.16 x 10.16 cm plastic pot in a topsoil-like media (Baccto top soil, Houston, Texas) compacted to a bulk density of 1.3 g cm^{-3}. The top 0.5 cm of the pot was left without media to ensure seeds and water remained in each pot. Each treatment was seeded with ≈300 seeds.

Table 2. Percent green cover (PGC) of container as determined by digital image analysis (DIA) of various turfgrass assemblage types. Images were captured every three days as soon as germination was observed until greater than 90 PGC of container was achieved with every assemblage type. Assemblages were grown in the horticulture greenhouses in Ames, IA in 2021.

Assemblage Type	**Days After Seeding**																
	6	**9**	**12**	**15**	**18**	**21**	**24**	**27**	**30**	**33**	**36**	**39**	**42**	**45**	**48**	**51**	**54**
Monocultures [a]	19 [b]	37	37	59	66	67	70	62	56	50	43	52	64	68	75	89	96
3 Species (S) Mix [c]	14	32	37	56	64	66	70	65	60	54	51	57	67	71	78	89	95
3 Cultivar (C) Blend [d]	18	37	35	57	64	65	70	66	59	57	50	63	76	81	90	101	105
3 S * 3 C Mix [e]	18	42	38	64	71	71	76	70	62	55	49	53	55	67	73	87	97
LSD (0.05) [f]	**4**	**9**	**NS [g]**	**NS**	**NS**	**NS**	**NS**	**NS**	**NS**	**NS**	**NS**	**8**	**8**	**8**	**8**	**8**	**7**

[a] Monocultures dictate a singular cultivar was grown by itself as an assemblage.

[b] PGC is represented in a percentage of green cover of a container based on Karcher et al. (2017). Some averages may be above 100 due to assemblages growing over the edges of their containers

[c] 3-species mix indicates an assemblage of three species each containing one individual cultivar.

[d] 3-cultivar blend contains three cultivars from the same species.

[e] 3-species*3-cultivars mix contains three species each containing three cultivars within their species, resulting in a total of nine cultivars in these assemblages.

[f] Means were separated using Fishers protected least significant difference at the 0.05 probability level. There was an observed treatment x date interaction, as a result data will be presented by individual collection dates.

[g] NS = not significantly different.

Table 3. Clipping yield for various turfgrass assemblages during and after establishment. Clippings were collected at 5 cm height-of-cut and dried at 40º C for 72 hours at the horticulture hall greenhouses in Ames, IA in 2021.

Assemblage Type	**Weeks after Seeding**	
	5	**10**
Monocultures [a]	0.007 [b]	0.100
3 Species (S) Mix [c]	0.005	0.110
3 Cultivar (C) Blend [d]	0.016	0.226
3 S * 3 C Mix [e]	0.001	0.128
LSD (0.05) [f]	**0.012**	**0.078**

[a] Monocultures dictate a singular cultivar was grown by itself as an assemblage.

[b] Clippings were collected and then dried at 40'C for 72 hours, which was modified slightly from Thompson and Kao-Kniffin (2016).

[c] 3-species mix indicates an assemblage of three species each containing one individual cultivar.

[d] 3-cultivar blend contains three cultivars from the same species.

[e] 3-species*3-cultivars mix contains three species each containing three cultivars within their species, resulting in a total of nine cultivars in these assemblages.

[f] Means were separated using Fishers protected least significant difference at the 0.05 probability level. There was an observed treatment x date interaction, as a result data will be presented by individual collection dates.

Table 4. Nitrate leaching for turfgrass assemblage types of cool-season grasses grown in the horticulture hall greenhouses in Ames, IA in 2021. Nitrate leaching was measured using a modified pour through method similar to Whipker et al. (2001). Where distilled water was used to saturate the soil, and subsequent drainage was collected and analyzed similar to methods used on turfgrass before with Pease et al. (2022).

Assemblage Type	**Weeks After Seeding**		
	8	**12**	**16**
Monocultures [a]	0.08 [b]	0.09	0.11
3 Species (S) Mix [c]	0.08	0.07	0.09
3 Cultivar (C) Blend [d]	0.08	0.07	0.10
3 S * 3 C Mix [e]	0.08	0.08	0.11
LSD (0.05) [f]	**NS [g]**	**0.01**	**NS**

[a] Monocultures dictate a singular cultivar was grown by itself as an assemblage.

[b] Nitrate leaching is measured in parts per million (ppm).

[c] 3-species mix indicates an assemblage of three species each containing one individual cultivar.

[d] 3-cultivar blend contains three cultivars from the same species.

[e] 3-species*3-cultivars mix contains three species each containing three cultivars within their species, resulting in a total of nine cultivars in these assemblages.

[f] Means were separated using Fishers protected least significant difference at the 0.05 probability level. There was an observed treatment x date interaction, as a result data is presented by individual collection dates.

[g] NS = not significantly different.

Table 5. Ammonium leachate for different turfgrass assemblage types of cool-season grasses grown in the horticulture greenhouse in Ames, IA in 2021. Ammonium leaching was measured using a modified pour through method similar to Whipker et al. (2001). Where distilled water was used to saturate the soil, and subsequent drainage was collected and analyzed similar to methods used on turfgrass before with Pease et al. (2022).

Assemblage Type	**Weeks After Seeding**		
	8	**12**	**16**
Monocultures [a]	0.08 [b]	0.08	0.12
3 Species (S) Mix [c]	0.08	0.10	0.10
3 Cultivar (C) Blend [d]	0.12	0.09	0.13
3 S * 3 C Mix [e]	0.07	0.09	0.12
LSD (0.05) [f]	**0.01**	**0.01**	**0.01**

[a] Monocultures dictate a singular cultivar was grown by itself as an assemblage.
[b] Ammonium leaching is measured in parts per million (ppm).
[c] 3-species mix indicates an assemblage of three species each containing one individual cultivar.
[d] 3-cultivar blend contains three cultivars from the same species.
[e] 3-species*3-cultivars mix contains three species each containing three cultivars within their species, resulting in a total of nine cultivars in these assemblages.
[f] Means were separated using Fishers protected least significant difference at the 0.05 probability level. There was an observed treatment x date interaction, as a result data is presented by individual collection dates.

Table 6. Microbial biomass and Organic Carbon (Loss on ignition) for different cool-season turfgrass assemblages grown in the horticulture hall greenhouse in Ames, IA in 2021. The difference in the dissolved organic C and N were determined with a TOC-TN analyzer and compared to measure the amount of microbial biomass of a soil.

Regime Factor	**Microbial Biomass (mg C kg^{-1})**	**Organic Carbon (g)**
Monocultures[a]	530[b]	6.01
3 Species Mix[c]	408	6.17
3 Species*3 Cultivars Mix[d]	367	6.69
3 Cultivar Mix [e]	367	6.37
LSD (0.05) [f]	**NS [g]**	**NS**

[a] Monocultures dictate a singular cultivar was grown by itself as an assemblage.

[b] Soil microbial biomass was measured similar to the methods of Studt et al. (2021) and McDaniel and Grandy (2016).

[c] 3-species mix indicates an assemblage of three species each containing one individual cultivar.

[d] 3-cultivar blend contains three cultivars from the same species.

[e] 3-species*3-cultivars mix contains three species each containing three cultivars within their species, resulting in a total of nine cultivars in these assemblages.

[f] Means were separated using Fishers protected least significant difference at the 0.05 probability level.

[g] NS = not significantly different.

Table 7. Percent green cover (PGC) as determined by digital image analysis of variouscool-season turfgrass assemblage types. Pictures were taken every three days of simulated drought until less than 10 PGC was observed for multiple collection dates with every assemblage type. Assemblages were grown in the horticulture greenhouses in Ames, IA in 2021.

Assemblage Type	**Days of Simulated Drought**							
	0	**3**	**6**	**9**	**12**	**15**	**18**	**21**
Monocultures [a]	53.5 [b]	38.3	30.5	18.6	14.2	12.7	8.9	9.1
3 Species (S) Mix [c]	54.1	39.3	29.3	17.7	13.7	11.6	7.2	7.1
3 Cultivar (C) Blend [d]	50.8	38.0	29.2	18.9	14.6	12.2	8.3	8.6
3 S * 3 C Mix [e]	58.7	44.2	33.3	19.2	14.6	12.5	8.6	8.1
LSD (0.05) [f]	**NS** [g]	**NS**	**NS**	**NS**	**NS**	**NS**	**NS**	**NS**

[a] Monocultures dictate a singular cultivar was grown by itself as an assemblage.

[b] PGC is represented in a percentage of green cover based on Karcher et al. (2017).

[c] 3-species mix indicates an assemblage of three species each containing one individual cultivar.

[d] 3-cultivar blend contains three cultivars from the same species.

[e] 3-species*3-cultivars mix contains three species each containing three cultivars within their species, resulting in a total of nine cultivars in these assemblages.

[f] Means were separated using Fishers protected least significant difference at the 0.05 probability level. There was an observed treatment x date interaction, as a result data is presented by individual collection dates.

[g] NS = not significantly different.

CHAPTER 4. GENERAL CONCLUSION

The minimum of a 2-month delay after achieving desired height-of-cut (HOC) before applying simulated traffic events (STE) may have allowed the turfgrass to acclimate and thus not result in STE stress to the degree that might be expected if simulated traffic occurred sooner after the desired HOC was reached. This 2-month delay could be potential reasoning for a lack of treatment effects across all metrics. ‘Moonlight’ Kentucky bluegrass (*Poa pratensis* L.) that was treated with a nitrogen rate of 36.6 kg ha^{-1} mo^{-1} either was the same or better than the 73.2 kg ha^{-1} mo^{-1} rate across most rating dates for rotational resistance, percent green cover (PGC), and surface hardness. This research is limited in scope due to the specific time of unplanned suspension of maintenance and STE. This study is modeled similarly to the high school football schedule commonly seen in the United States. Future research could test unplanned suspension of maintenance in different seasons, and test different timings between HOC reduction and STE.

The 3 cultivar C blend exhibited the highest PGC 42 days after seeding (DAS) until the end of establishment. Inclusion of more species can reduce damage from disease and other pests as seen in previous research. The 3 Species (S) and 3 C treatments leached similar or lower nitrate than the 3 S* 3 C mixes and the monocultures. Turfgrass assemblages made no difference in drought tolerance. Microbial biomass and organic carbon were similar across turfgrass assemblage type. The least amounts of ammonium leaching varied between assemblages. These results indicate that more diversity may not be beneficial after establishment.