

Zoysiagrass sod performance, rooting characteristics, and thatch accumulation
among genotypes

by

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Abstract

Zoysiagrass (*Zoysia spp.*) is valued for its adaptability, drought tolerance, and low input requirements, making it ideal for diverse landscapes, including golf courses and home lawns in the U.S. transition zone and southern regions. This thesis evaluates the performance, and resilience, of zoysiagrass under varying conditions, focusing on sod establishment, drought avoidance mechanisms, and thatch accumulation. Three studies were conducted to assess turfgrass adaptation and quality (i) the influence of aerification and grow-in treatments on sod performance, (ii) root traits for drought avoidance mechanisms, and (iii) thatch accumulation and genotype traits. The first study (Chapter 2) investigated three cultivars that were grown from sprigs under two nitrogen rates (1.6 lbs. or 3.6 lbs./1000 ft²/yr) and two mowing heights (0.75 in. or 1.5 in.). After harvest sod was installed at two Kansas locations (Olathe and Manhattan), on an aerified and nonaerified soil. After comparing the performance of the three cultivars, we found that aerification prior to sodding declined the NDVI of DALZ 1808 in both locations and the overall quality in Olathe. Meyer and Innovation NDVI and overall quality were not affected by aerification. In Olathe, sod grown under low mowing heights prior to harvest had 8.8% more root numbers than the higher mowing height. In Manhattan, Meyer zoysiagrass had a slightly longer root length than Innovation or DALZ 1808 within 9 days after sod was installed. Drought symptoms (visually on a 1 to 9 scale, 9 = no wilting, no stress) during the three weeks of dry-down showed that ‘Meyer’ had more significant drought symptoms (6.2) compared to Innovation (7.1) and DALZ 1808 (7.2) in Olathe, suggesting that cultivars respond differently to drought stress. The second study (Chapter 3) analyzed the rooting characteristics including rooting depth and shoot-to-root ratio (SRR) of 12 zoysiagrass genotypes in a greenhouse. The genotypes, grown in PVC tubes filled with calcined clay, were

subjected to maximum root length evaluation at the end of the experiment and weekly ratings of shoot quality. Several additional rooting architecture traits in 10 cm depth zones were assessed using WinRHIZO software. The maximum root lengths were 53.5 cm (2023, Chisholm) and 61.5 cm (2024, DALZ 1311); all others were not significantly different in maximum root length except Zeon (2024) and KSUZ 1201 (2023, 2024). The highest root length among all genotypes was between 0 to 10 and 10 to 20 cm. As we evaluated deeper into the rootzone, cultivars that consistently showed lower root lengths, such as 20 to 30 cm, were DALZ 1702, KSUZ 1201 (2023), and Zeon (2024). Zeon consistently had high shoot quality, but KSUZ 1201 and DALZ 1702 had lower shoot quality (< 6 on a 1 to 9 scale), likely resulting in lower root development. The third study (Chapter 4) evaluated thatch accumulation and its possible influence on zoysiagrass performance across multiple environments as part of the National Turfgrass Evaluation Program (NTEP) and United States Golf Association (USGA) research trials in Kansas, Oklahoma, Indiana, Texas, and Florida. Only a few sites had differences among genotypes in thatch depth and mass. ‘Emerald’ accumulated the greatest thatch depth in Texas (NTEP trial), while DALZ 1311 had the lowest in Oklahoma (NTEP). Correlation of thatch depth was evaluated for ratings of genotypes’ traits; Thicker thatch depth was correlated with reduced turfgrass firmness in Texas (NTEP) and Citra (USGA) sites. Thatch depth was positively correlated with increased NDVI levels in Texas and Citra (NTEP) while it was negatively correlated with NDVI levels in Indiana (USGA). Increased thatch depth was positively correlated with increased tiller numbers in Oklahoma, Texas and Ft. Lauderdale. Overall, this thesis provides an understanding of the influence of different grow-in strategies on the performance of zoysiagrass genotypes, the adaptation of genotypes to specific locations

based on their rooting abilities, and the effect of thicker thatch depth and higher thatch mass on turfgrass performance.

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Dedication

To God, the source of all wisdom and strength, whose guidance and grace have carried me through every step of this journey. I dedicate this work to His glory and thank Him for the endless blessings, strength, and perseverance that have sustained me.

“Commit to the Lord whatever you do, and He will establish your plans.” – Proverbs 16:3

Chapter 1 - Introduction

Research conducted in this thesis is focused on three areas: zoysiagrass sod performance after harvest; rooting comparison of zoysiagrass cultivars and genotypes; and thatch accumulation of cultivars and genotypes over several years.

The term "sod" refers to strips of turfgrass with the soil that clings to their roots, used in vegetative planting for rapid establishment. Sodding history goes back to the 1600s when it was sourced from pastures and meadows with low-growing perennial grass species in the United Kingdom (Beard and Rieke, 1969). Historical sodding recommendations called for moving up to 4 inches of soil along with the sod while cutting the sod (Beard and Rieke, 1969). Several commercial sod production operations were also initiated in Michigan in the 1920s. The United States turfgrass sod industry had grown significantly from its infancy in the 1960s when an estimated \$100 million was generated at the farm level in the U.S. from ~1,000 sod farms covering ~105,000 acres (Adrian et al., 2003) to 2017 when 1,500 sod farms generated US \$1.2 billion in annual sales from harvesting approximately 340,000 acres (USDA-NASS, 2019).

Zoysiagrass

Zoysiagrass (*Zoysia* spp. Willd.) is a warm-season turfgrass species originating from East Asia and the Pacific Islands (Patton et al., 2017). According to Lamson-Scribner (1895) and governmental recordkeeping, it is possible to say *Z. matrella* was introduced into the U.S. from Japan in 1892 and *Z. japonica* was introduced from Korea into the U.S in 1894 (Barron, 1906; Corbett, 1906). It is well adapted for use as turfgrass in transitional and warm climatic U.S. regions. Three species of zoysiagrass [*Z. japonica* Steud., *Z. matrella* (L.) Merr., and *Z. pacifica* (Goudswaard) M. Hotta and S. Kuroki] have been widely used by turfgrass breeders in the development of advanced lines (Patton et al., 2017). Still, demand for low-input turfgrasses

(e.g., less irrigation, fertilization, mowing, pesticide inputs) is also high in the southwestern United States. Therefore, there is a need for breeding advancements in zoysiagrass with less of a focus on leaf texture and more focus on reducing inputs to decrease long-term management costs in turfgrass systems (Braun et al., 2021) and a focus on good sod-forming properties, such as shorter sod production cycles and greater transplanting success.

In 2006, approximately 6,594 hectares of zoysiagrass could be found on golf courses across the U.S. with 81% being used on golf courses in the transition zone and 18% used on golf courses in the southeastern U.S. (Lyman et al., 2007). Zoysiagrass use in the southern U.S. has since been gaining popularity as the availability of cultivars increases, especially those that perform well in warmer regions (Patton, 2009). Thirty-eight zoysiagrass cultivars are currently used in the U.S. or previously used, and 32 were commercially available in 2009. Fifteen were released in 2000, 24 in 1990, and 29 in 1980 (Patton, 2009).

Comparison between Zoysiagrass Sodding, Sprigging, and Plugging

One of the challenges of zoysiagrass is its limited availability of seeded cultivars and this dates back to the 1940s, when zoysiagrass had started increasing (Halsey, 1956). In the 1940s, *Z. matrella* was marketed as a seed named Flawn (Childers and White, 1947). However, because of the limited seed availability, zoysiagrass seed was widely regarded as a hoax by many. Significant progress in the development of *Z. japonica* cultivars from seed did not occur until the 1980s, and it wasn't until 1993 that two cultivars developed in the United States became commercially available, Zenith and Compadre (Nesbitt, 1993; Samudio, 1996; Patton et al., 2017). Before this, most *Z. japonica* seeds entering the U.S. were imported from China (Samudio, 1996). While zoysiagrass seed is now produced domestically, mainly in Georgia, its

production remains minimal compared to other grass species (Patton et al., 2017). Zoysiagrass has since been primarily established vegetatively through sod, plugs, and sprigs.

Turfgrass established by sod has numerous benefits, such as providing immediate green space, preventing soil erosion, reducing runoff, and filtering pollutants from rainwater and air. Sodding instead of seeding allows revenue production for cool-season turfgrasses to begin a year earlier due to the elimination of the requirement for a typical grow-in period. With at least a four to six-month advantage over seeding, differences in establishment time for zoysiagrass are evident (Scott, 1996).

An alternative to completely sodding an area is strip-sodding, which involves laying 15-31 cm wide sod strips in rows with 30-61 cm empty spaces between rows that will allow the strips to grow together. This method is less expensive than sodding the entire area; however, proper management practices are crucial to achieve full coverage and minimal weed encroachment in a reasonable amount of time. (Fresenburg, 1997).

Plugging is the vegetative method of inserting ~5 by 5 cm small piece of sod material into the soil at an even spacing. To facilitate planting and improve plug-to-soil contact, cores of soil the same size as the sod plugs are removed using a special instrument called a plugger, dibbler, or long-handled bulb planter (Beasley et al., 2019; Samples and Sorochan, 2007; Fresenburg, 1997). The home lawn market is the main target audience for plugs and smaller damaged areas in the turfgrass system.

Sprigging involves planting a portion of plant material with roots, leaves, and segments of the stolon or rhizome-containing nodes (Landry 2013; Fresenburg, 1997). Broadcast planting also known as “stolonizing” involves planting only stolons on the soil surface. The stolons and rhizomes are either broadcast on the soil surface or incorporated into closely spaced furrows to

promote sprig-to-soil contact (McCarty, 2011). Sprigging is generally less expensive and can provide a faster establishment time than plugs (Fresenburg, 1997). Sprigs need more care while planting than plugs and are less likely to survive unfavorable conditions (Fresenburg, 1997). Similar to strip-sodding or plugging, it is important to monitor soil moisture until the plant material is rooted to prevent desiccation and death after planting. (Christians et al., 2017). Plugs are frequently used to establish these warm-season turfgrasses, especially zoysiagrass, on research plots or smaller home lawn areas (Christians et al., 2017).

Maintenance Inputs of Zoysiagrass Sod

The main objective of sod production is to encourage plant growth and development by promoting root and rhizome or stolon growth and preventing weed infestation. These structures hold the sod intact and allow harvesting and handling (Beard and Rieke, 2015).

Mowing

A recommended mowing height of 0.5 to 6.4 cm is optimal for zoysiagrass sod production (Patton et al., 2017; Beard and Rieke, 1969). Mowing guidelines for sod establishment differ depending on the species, location, and intended use (Wherley et al., 2011; Beard, 2002; Christians, 1998). A higher height of cut will increase the amount of leaf area available for photosynthesis, improving carbohydrate availability and storage for root growth and vertical and lateral shoot growth. Zoysiagrass leaves contain high concentrations of cellulose, hemicellulose, and lignin (Trappe, 2015; Lulli et al., 2012; Washburn and Seamans, 2012) compared to other turfgrass species, which makes it difficult to cut with mowers and may require more equipment maintenance than other turfgrass species to maintain a high-quality cut (Beard and Beard, 2005). It has been reported that *Z. matrella* has a lower quality cut than *Z. japonica* types (Patton et al., 2010). Mowing can also help to reduce sod heating on pallets after harvest.

Sod heating comes from respiration in the living plant leaf (Ruser and White, 1980) while the sod rolls or slabs are being transported or stored for a short duration before transplanting. Generally, sod producers keep the mowing height high through the production phase to maximize root and rhizome growth and then slowly lower the mowing height in the weeks before harvest (Christians et al., 2017). The height of cut and the rate of vertical shoot growth have an impact on mowing frequency. Warm-season turfgrass species usually require mowing every 5 to 10 days during the growing season; however, there are vertical growth differences among cultivars and turfgrass species. Mowing frequency should not be conducted at a fixed schedule, but instead, it should be dependent on the growth rate of the turfgrass, which will change during a growing season due to various factors (Christians et al., 2017). A typical guideline is to never remove more than one-third of the leaf area at once to reduce the amount of stress caused by mowing (Law et al., 2016), this guideline will help determine when mowing is required throughout the growing season.

Fertilization

Adequate plant nutrition will facilitate a faster grow-in period for the sod producer, which a faster grow-in may correlate to a shorter production cycle. Nitrogen is one of the main macronutrients that turfgrass plants require most often to sustain growth and development and improve stress tolerance. In warm, humid climates, such as Texas, Louisiana, and Georgia, 342 kg N ha⁻¹ yr⁻¹ of fertilizer was advised for zoysiagrass in the 1960s (Holt, 1969). Since then, turfgrass science research has revealed that N fertility recommendations for zoysiagrass are significantly lower than recommendations made 50 years ago (Christians et al., 2017). Dunn et al. (1995) reported 98 kg N ha⁻¹ yr⁻¹ was sufficient for ‘Meyer’ zoysiagrass (*Z. japonica*) in Missouri and this N level promoted the fastest root growth. Similar findings were made by

Patton et al. (2010b), who discovered that while three zoysiagrass cultivars' turfgrass quality was always satisfactory (turfgrass quality 6 on a 1–9 scale) for the nonfertilized plots, it was at its highest when 98 kg N ha⁻¹ yr⁻¹ was implemented in Arkansas. Trappe and Patton (2013) noted that when fertilized at a rate of 98 kg N ha⁻¹ yr⁻¹, Meyer and 'Cavalier' zoysiagrass (*Z. matrella*) produced satisfactory turfgrass quality regardless of the N source. Braun and Bremer (2018) reported similar findings with nonfertilized and fertilized (98 kg N ha⁻¹ yr⁻¹) zoysiagrass in Kansas. Kenworthy and Engelke (1999) reported the highest-quality zoysiagrass in Texas was produced at 98 to 195 kg N ha⁻¹ yr⁻¹, which the higher N fertilization amount was likely because of a longer growing season in Texas.

Excessive N fertilization rates may lead to problems, for example, *Z. matrella* cultivars receiving annual N rates of 146 kg N ha⁻¹ yr⁻¹ led to a higher likelihood of scalping after excessive thatch formation (Hale, 2006). Higher N applications may also lead to more disease problems. For example, Schwartz (2008) reported excessive N applications led to more disease development, such as Bipolaris leaf spot [*Bipolaris spicifera* (Bainier) Subramanian)]. Overall, zoysiagrass will produce high-quality turfgrass with fewer fertilization inputs than other warm-season grasses such as bermudagrass (*Cynodon* spp).

Zoysiagrass Rooting

The relationship between root architecture and drought avoidance mechanisms has been well documented with larger root systems contributing to better drought resilience (Koevoets et al., 2016; May and Milthorpe, 1962). Turfgrass species exhibit drought avoidance by minimizing water loss and maintaining tissue hydration. Key traits include deeper root systems, which allow access to water from deeper soil layers, and stomatal regulation to reduce transpiration (Wang et al 2024; Fan et al., 2020). Drought resistance involves a combination of mechanisms that help

the plant withstand prolonged water deficit conditions. Drought-resistant grasses like bermudagrass (*Cynodon spp.*) and buffalograss (*Bouteloua dactyloides*) manage drought stress through enhanced water use efficiency and maintaining green cover for extended periods under drought conditions (Huang 2008; Kim and Beard, 2018). Drought tolerance on the other hand allows turfgrass to survive extreme water stress by altering metabolic processes to endure dehydration. Grasses like buffalograss and zoysiagrass exhibit these traits, enabling them to survive through water stress and quickly recover once water becomes available (Fan et al., 2020; Kim and Beard, 2018).

Zoysiagrass, though often less drought-tolerant than bermudagrass or other warm-season grasses, exhibits specific drought avoidance traits that are increasingly recognized in newer cultivars. Early studies on Meyer zoysiagrass showed reduced shoot growth and increased root elongation under deep, infrequent irrigation regimes (Qian et al., 1996). More recent studies, especially on new genotypes, highlight improved performance during drought (Wherley et al., 2014), with certain cultivars like *Z. japonica* showing better drought tolerance than their predecessors (Patton et al., 2017). Although zoysiagrass generally has a lower evapotranspiration rate compared to species like bermudagrass (*Cynodon spp.*) and St. Augustinegrass (*Stenotaphrum secundatum*), it typically exhibits shallower roots, leading to earlier wilting during drought conditions (Huang, 2008). Research has shown that under severe drought, zoysiagrass can enter dormancy and fully recover after substantial rainfall, even following extended periods of water deficit (Beard and Sifers, 1997; Steinke et al., 2009). This trait positions zoysiagrass as a viable option in regions prone to chronic drought, where water conservation is a priority (Patton et al., 2017).

Recent technologies such as the use of root scanning and image analysis software like WinRHIZO (Christensen et al., 2017; Regent Instruments, 2024), allow for precise characterization of root morphology, including traits such as root length, surface area, and root volume (Bouma et al., 2000; Himmelbauer et al., 2004). Such characterization is particularly valuable for zoysiagrass, as deeper rooting has been identified as a key drought avoidance mechanism in zoysiagrass (Jespersen and Schwartz, 2018).

Zoysiagrass Thatch Accumulation

Thatch formation is a prominent issue in zoysiagrass. Zoysiagrass has dense, slow-decomposing leaves and stems that contribute to the rapid accumulation of organic matter in the form of thatch. Thatch is a layer of intermingled living and dead plant material, that forms between the soil surface and the green vegetation (Beard, 1973; McCarty, 2018). In zoysiagrass, this accumulation often leads to management challenges due to the species' slower decomposition rate compared to other species (Dunn et al., 1981). Zoysiagrass species such as *Z. japonica* and *Z. matrella* are known for producing substantial thatch layers over time, especially under intensive management practices (Patton et al., 2017). Excessive thatch can affect turfgrass quality by reducing water infiltration, restricting root growth, and providing an environment conducive to pests and diseases (McCarty et al., 2016). In addition, thatch accumulation in zoysiagrass can complicate mowing, leading to scalping and impeding turfgrass uniformity (Dunn et al., 1981).

Managing thatch in zoysiagrass is crucial to maintaining turfgrass health, with mechanical practices such as vertical mowing and core aeration being commonly recommended to mitigate excessive accumulation. Regular maintenance of thatch levels can

enhance water movement, improve root development, and maintain overall turfgrass quality (McCarty et al., 2016).

Conclusion

A recent release ‘Innovation’ zoysiagrass from K-State and Texas A&M, had accumulated thatch at some sod producers resulting in difficult management practices for the cultivar after it was harvested and installed at multiple golf courses. Chapter 2 in this thesis examines how grow-in management practices influence the performance of sod after it was harvested and installed in two different sites.

Studies have shown that zoysiagrass offers a promising solution for low input turfgrass systems, particularly in regions where water conservation and reduced management are priorities. However, the specific rooting behaviors of newer zoysiagrass genotypes, especially in comparison to older, established cultivars, have not been fully explored. Deeper and more extensive root systems are known to provide an advantage in drought-prone regions, yet the variability in rooting across different genotypes remains a knowledge gap. A thorough evaluation of rooting is presented in Chapter 3.

Thatch accumulation among cultivars and genotypes across multiple different states has not been thoroughly evaluated. In Chapter 4 greater details on these sites and genotypes are presented. The objective of this research is to bridge these gaps by comparing post-harvest sod performance, evaluating rooting characteristics of zoysiagrass cultivars and thatch accumulation of various genotypes across multiple locations for extended periods. By addressing these aspects, this work aims to improve the management of zoysiagrass in sod production and long-term maintenance.

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Chapter 2 - Evaluating the Sod Performance of Three Zoysiagrass Genotypes on Aerified vs. Non-Aerified Soil After Variations in Nitrogen Fertilization and Mowing Height During Grow-in

This chapter has been prepared using style guidelines for submission to *Crop, Forage, and Turfgrass Management*

Abstract

Cultural practices at sod producers' farms can influence the performance of zoysiagrass sod after harvesting and installation. The objectives of this study were to: (i) evaluate the influence of aerification and grow-in practices on sod performance after installation; (ii) evaluate the root development of sod within two weeks of sodding; and (iii) evaluate sod response based upon treatments prior to sodding on weed infestation, leaf yellowing, and drought tolerance. Cultivars evaluated in this experiment were 'Meyer', 'Innovation', and DALZ 1808 grown from sprigs during a previous experimental study (high N vs. low N and high mowing vs. low mowing). This experiment was established in the summer of 2023 after the sod was harvested at the Olathe Horticulture Research and Extension Center, it was installed on 24 May 2023 (Olathe) and 15 June 2023 (Rocky Ford Turfgrass Research Center in Manhattan, KS). Aerification before sodding reduced the NDVI of DALZ 1808 on multiple dates at both locations compared to Meyer and Innovation on aerified and non-aerified soil. Cultivars that had a low mowing height during grow-in had 8.8% more root numbers in Olathe compared to high mowing height and no differences occurred in Manhattan. The root length of Meyer (0.24 in.) nine days after sodding in Manhattan was longer compared to 'Innovation' (0.20 in.) and DALZ 1808 (0.19 in.). After plots were subjected to three weeks of dry down Meyer, both aerified and non-aerified, showed more significant drought stress in Olathe (6.2 on a 1 to 9 scale; 9 = no wilting, no leaf firing) compared to Innovation (7.1) and DALZ 1808 (7.3). In conclusion, aerifying the soil of the planting site before sodding may have a significant influence on some zoysiagrass genotypes, e.g., DALZ 1808. Initial rooting can be influenced by grow-in mowing height and cultivar, and Meyer zoysiagrass showed more drought stress compared to Innovation and DALZ 1808.

Introduction

Zoysiagrass (*Zoysia* spp. Willd.), is recognized for its low maintenance requirements when compared to cool-season turfgrasses in terms of less water, less fertilizer, and less pesticide outputs are used across the world where warm-season turfgrasses are grown on golf courses, home lawns, and landscape areas (Fry et al., 2008; Hanna et al., 2013). Three species [*Zoysia japonica* Steud; *Zoysia pacifica* (Goudswaard) M. Hotta and S. Kuroki; and *Zoysia matrella* (L.) Merr] introduced into the U.S. around 1900 have been widely used by turfgrass breeders to develop advanced lines (Patton et al., 2017). Interspecific hybridization of *Z. matrella* and *Z. japonica* has successfully increased the adaptation of *Z. matrella* in the transition zone (Chandra et al., 2017). Zoysiagrass grows well on well-drained sandy loams and even clay soils with a pH of 5 or less to pH 8 (strongly acidic to neutral or mildly alkaline). This would make them tolerate temporary waterlogging but not permanent waterlogging or wet clay soils (Loch, 2013; Brosnan and Deputy, 2008).

Public and private sector breeding programs are now focusing on understanding and improving the zoysiagrass germplasm for improved turfgrass performance, winter hardiness, and fine-textured genotypes (Braun et al., 2022; Meeks et al., 2022; Patton et al., 2017). In 2021, approximately 23,670 acres of zoysiagrass were on golf facilities across the U.S., with 97% concentrated in the southeast and transition zone regions (Shaddox et al., 2023).

While most sod producers' goal is to generate maximum production in the shortest possible time, variety selection, establishment timing, irrigation, fertilization, mowing, and pest management may influence the outcome of the sod during and after harvest (Braun et al., 2021). However, despite the release of several new zoysiagrass cultivars, there is limited agronomic information on the impact of cultural practices during the grow-in phase and before sodding.

Several studies have been performed to evaluate the establishment rate of several zoysiagrass genotypes from sprigs (Richardson and Boyd, 2001; Stiglbauer et al., 2009; Zhang et al., 2021). Further, the effects of nitrogen rate and mowing height on zoysiagrass have also been well-documented (Dunn et al., 1981; Patton et al., 2010; Schwartz et al., 2018). However, many of these previous studies have focused on the visual turfgrass quality, with minimal reports on how nitrogen rates and mowing height influence thatch accumulation and post-harvest root development in zoysiagrass sods. Studies have also shown that nitrogen fertility sources don't necessarily influence the growth rate of zoysiagrass cultivars when plugged and sprigged (Fry and Dernoeden, 1987; Carroll et al., 1996; Richardson and Boyd, 2001).

Aerification is the process of perforating the soil with holes that allow for root penetration (Heinrichs, 2020). On established turfgrass, aerifying the soil helps loosen hard, compacted soil and allows the circulation of air and moisture in the root zone. The exchange of air increases microbial activity while breaking down thatch (Heinrichs, 2020). Skipping aerification can increase thatch buildup and soil organic matter accumulation (Bevard, 2011). Excessive thatch and soil organic matter buildup can result in decreased water infiltration, decreased airflow, increased pest problems, and reduced pesticide effectiveness (Gaussoin et al., 2013; Landreth et al., 2008). Core aerification can be done before installing sod to alleviate layering problems that occur when the soil on the sod is different from the soil at installation (Anonymous, 1996). According to TPI Turf News (Anonymous, 1996), aerification helps new roots grow and enhances smoothening and leveling when the soil is loosened. It also aerates the soil when compacted and bulk density is high. However, research has not been reported on how aerification before laying sod can influence zoysiagrass.

The recent release ‘Innovation’ zoysiagrass from K-State and Texas A&M is a fine textured, cold-hardy zoysiagrass (Chandra et al, 2017). Grow in of Innovation at sod producers went well, but there were post-planting problems with sod after it was installed at some golf courses. A review of Innovation at some of those sites showed some accumulated thatch. In addition, soil compaction of areas on golf courses may also have affected sod performance. The impact on Innovation at some of these sites resulted in the grow-in experiment of zoysiagrass (McFadden, 2024). After grow-in of this aforementioned experiment, the sod was harvested in this study to evaluate zoysiagrass cultivar performance after it was installed. Therefore, this research was initiated to: (i) evaluate the influence of core aerification on the planting site for its impact on sod performance after laying; (ii) evaluate the root development of sod within two weeks of sodding; and (iii) evaluate sod response based upon treatments prior to sodding on weed infestation, leaf yellowing, and drought tolerance.

Materials and Methods

Field Management

Meyer (*Zoysia japonica* Steud.), Innovation (*Z. japonica* x *Z. matrella*), and DALZ 1808 were sprigged at the Olathe Horticulture Research and Extension Center in June 2021 in a separate experiment evaluating management practices influencing grow-in. Treatments included low (1.6 lbs./1000 ft²/yr) or high (3.6 lbs./1000 ft²/yr) nitrogen level and low (0.75 in.) or high (1.5 in.) mowing height. Nitrogen was applied shortly after planting, and mowing heights began in spring 2022 and continued into spring 2023. During the summer of 2022, the trial was rearranged to a strip-split plot design to incorporate two mowing treatments (1.9 cm and 3.8 cm). Genotype was the vertical strip, nitrogen treatment was the horizontal strip, and mowing

treatments were the sub-plots nested within each intersection for twelve total treatments replicated four times. The influence of cultural practices on grow-in has been reported by the graduate student who oversaw this experiment (McFadden, 2024).

After sod harvest, experiments were conducted at the Olathe Horticulture Research and Extension Center in Olathe, KS (long. 39.48° N, lat. 95.66° W) and the Rocky Ford Turfgrass Research Center in Manhattan, KS (long. 39.13° N, lat. 96.36° W). Planting site soil was conducted through a soil textural analysis and routine soil chemical test for each location after soil was randomly collected at the planting site (Kansas State Soil Test Laboratory). The soil at Olathe was an Oska-Martin silty clay loam with a pH of 6.0 and the Manhattan soil was characterized as a Chase silty clay loam (fine, smectitic, mesic Aquertic Argiudolls) with a pH of 7.1.

The experiment on the performance of grow-in strategies was arranged as a randomized complete block design with a split-plot treatment structure and three replications in the two locations. Aerification was the whole (main) plots (aerified and non-aerified) measuring 4 × 18 ft (Fig. 2.1) and grow-in cultivars (Table 2.1) that received high or low mowing heights, and high or low nitrogen rates in the previous strip split plot study were the subplots which totaled twelve. Each subplot measured 4 × 1.5 ft., and the alleyway in-between replications measured 2 × 18 ft. (Fig. 2.1). The alleyway had Meyer present which had previously been mowed at 0.75 in.

Olathe Horticultural Research and Extension Center, KS

To prepare the site, the pre-existing Meyer zoysiagrass growing at 0.75 in. height was treated with 0.82 fl. oz./gallon of glyphosate (N- (phosphonomethyl) glycine) to prevent its growth from competing with the experimental cultivars. The application was made twice, two weeks before Meyer was cut and removed using a sod cutter (Ryan Turf, Johnson Creek, WI) at

1 inch deep. This exercise was done to mimic common practices in golf course renovation.

Aerification was done on 24 May 2023 (Fig. 2.2) on one of the whole plots (aerified plot) in each replication. An aerator (Ryan GreenSaire 24 aerator) was used to aerify with core (hollow) tines set at 0.5-inch diameter to penetrate at 2 inches deep and a total of 30 holes ft⁻² was created. A starter fertilizer 24-11-3 (N-P-K) [Scotts Turf Builder Starter Fertilizer (Scotts Miracle-Gro Company, Marysville, OH.)] was applied at 1.5 lb. N/1000 ft² to individual plots of the experimental area using a shaker glass bottle.

Sod was cut from the initial grow-in experiment site and was firmly installed in both the aerified and non-aerified plots in a brick-like pattern with staggered ends to reduce the number of the long seam and ensure each end was in contact. Irrigation was applied afterward through an aluminum pipe with 3 sprinklers 20 ft. from each other at 1 inch for 45 mins to allow the sod and soil to gather enough moisture for the rooting process. A roller measuring 2 ft. x 18 inches in diameter was used on the sod to firmly press it into the soil after irrigation. Irrigation was applied every day for the first 10 days after sodding to allow establishment.

Mowing began on 30 May 2023 [the second week after sodding (WAS)] with a walk-behind rotary mower at 1.5 inches (3.8 cm) height of cut with clippings returned for one week. Mowing was done at this height to avoid scalping the newly installed sod. On 7 June 2023 (3 WAS), mowing was done slowly over the sod surface at 1-inch with a triplex reel mower [Toro Greensmaster 1000, The Toro Company, Bloomington, MN.] in two directions, N-S and W-E. Mowing was continued at 1 inch in two directions through 4 WAS and in this week, mowing was done twice. At 5WAS, height-of-cut was reduced to and remained at 0.75 inches (1.9 cm) thrice a week with a reel mower [Toro Reelmaster 3100D, The Toro Company, Bloomington, MN.]. Scalping started occurring when mowing height was reduced to 0.75 inches and was noticeable

on plots mowed at a higher height of 1.5 inches during grow-in. Plots were fertilized once on 16 June 2023 with urea (24-0-8) with NS-54 sulfur-coated slow-release N at 1 lb. N/1000 ft² to provide uniform, extended growth with extended nitrogen fertilization [Anderson Professional Turf Fertilizer, The Andersons Inc., Maumee, OH.]. Acclaim (Fenoxaprop-p-ethyl [(+)-ethyl 2-[4-[(6-chloro-2-benzoxazolyl)oxy]phenoxy] propanoate, Bayer Environmental Science]), a postemergence herbicide, was applied to control annual and perennial grass weeds (crabgrass, goosegrass, and bermudagrass) at 30 fl. oz/acre on 28 June 2023. On 5 July 2023, a combination postemergence broadleaves herbicide, quinclorac 8.43%, sulfentrazone 0.69%, 2,4-D 11.81%, and dicamba 1.49% [Q4 Plus, PBI/Gordon corporation] was applied at 2.5 fl. oz/1000 ft². Plots were irrigated every other day for 25 minutes and frequently when wilt (leaf folding) was significant.

Between 20 August to 31 August 2023, irrigation was withheld, and plots were subjected to dry down, after which irrigation was resumed at one inch three times a week. A total of 1 to 1.5 inches of water was applied weekly throughout the study unless significant rainfall occurred.

Data collection and details are provided below.

Rocky Ford Turfgrass Research Center, Manhattan KS

To prepare the site, Meyer zoysiagrass at fairway height was also treated with 3 oz/gallon glyphosate on 30 May 2023 to prevent it from competing with the experimental cultivars and was stripped removed with a sod cutter (Ryan turf professional, Johnson Creek, WI) at 1-inch deep on 13 June 2023. On the same day, the aerified whole plots were aerified with an aerator (Plugr PL800, IMSCORP, Lincoln, NE.) with 0.6-inch diameter core (hollow) tines at 1.5 inches deep. On 14 June 2023, a 24-11-3 (N-P-K) starter fertilizer [Scotts Turf Builder, (Scotts Miracle-Gro Company, Marysville, OH.)] was applied on the individual plot area using a shaker bottle at

0.5 lb. N/1000 ft². Sod was harvested from the grow-in experiment at Olathe on 13 June and installed at RFTRC on 14 June 2023. Sod treatments were installed in a brick-like pattern with both ends in contact to reduce the number of long seams. To ensure there was enough moisture for the sod and soil to start the rooting process, plots were irrigated at 1-inch for 45 mins, after which a 24-gallon tow behind lawn roller (Brinyl hardy Co., Jefferson, IN.) was rolled over to ensure a firm press of the sod to the soil. In the first week, irrigation was continued at 1 – 1.5 inch, and that weekly amount continued throughout the study unless rainfall occurred.

Mowing began on 20 June (2 WAS) with a walk-behind rotary mower [Honda HRX 217, American Honda Motor Company, Inc., Torrance, CA.] at a 1.5-inch height of cut with clippings returned. The turfgrass was mowed at a higher height to prevent scalping the newly installed sod. On 28 June 2023 (3WAS), mowing was reduced to 1 inch in two directions, twice a week for 2 weeks with a reel mower [Hustler FJ 50 greens. Hustler, Hesston, KS.], and on 12 July (6WAS), mowing height-of-cut was reduced and kept at 0.75 inches (1.9cm) thrice a week (MWF). A postemergence herbicide, (fenoxaprop-p-ethyl [(+)-ethyl 2- [4- [(6-chloro-2 benzoxazolyl) oxy] phenoxy] propanoate - Acclaim Herbicide, Bayer Environmental Science]) was applied to the plots on 18 July and 8 August 2023 at 0.64 oz 1000 ft² to control annual and perennial weeds. Plots were fertilized again on 27 July 2023 with urea (24-0-8) with NS-54 sulfur coated slow-release nitrogen at 1 lb. N/000 ft². [Anderson Professional Turf Fertilizer, The Andersons Inc., Maumee, OH.]. Irrigation was done on plots every other day for 25 minutes. Plots in Manhattan were also subjected to 3 weeks of dry down from 22 August to 8 September 2023, although there was 0.1 inch of rainfall on 26 August. After that, irrigation continued to apply a total of one inch weekly until zoysiagrass went into winter dormancy.

Data Collection and Measurement

Both the Olathe and Manhattan study areas were rated weekly for visual turfgrass quality (TQ) and Normalized Differences Vegetative Index (NDVI). TQ was rated based on turfgrass color, leaf texture, density, and uniformity on a 1 to 9 scale where 1 = poorest quality; 6= minimum acceptable quality; and 9= optimum color, uniformity, texture, and density (Morris and Shearman, 1998). Measurement of NDVI was done on a scale of 0 to 1 [where 1= best quality] by using a handheld NDVI meter (FieldScout CM 1000, Spectrum Technologies, Aurora, IL.) at wavelengths 660 nm and 840 nm by holding the sensor 50 inches above turfgrass level. NDVI measurements are usually used to indicate plant health status for many crops (Atshan et al., 2018; Straw et al., 2016; Chapman and Barreto, 1997). Nine days after sodding, data were collected to evaluate the maximum root length and maximum root numbers of each genotype. Data was collected on 1 June in Olathe and 23 June in Manhattan by cutting and flipping over 1 ft. of sod in each plot area and randomly throwing two 2 in.- diam. rings on each flipped plot (Fig. 2.5). In each ring, the maximum root length was measured using an electronic digital caliper, and the maximum root number that had emerged within 2 WAS was counted. Data from each ring were averaged across all subplots.

Weed infestation was rated by visually rating the percentage of the plot covered with crabgrass (*Digitaria sanguinalis* (L.) Scop), goosegrass (*Eleusine indica* (L.) Gaertn.), and bermudagrass (*Cynodon spp.*) on 23 June 2023 at Olathe and on 27 June in Manhattan.

There were instances of leaf yellowing in Olathe and Manhattan. Leaf discoloration was visually rated on a scale of 1 to 9 [9= no yellowing; 1= discolored leaves, not acceptable] on 7 August and 9 August, respectively.

Dry down levels and tolerance of the 3 weeks of drought stress were evaluated by visually rating the plots on a 1 to 9 scale [9 = no wilting, no leaf firing, 1 = complete wilting, completely stressed] (NTEP, 2000) This marked the end of data collection in 2023 for both locations.

The frequency of NDVI and (TQ) ratings was reduced to every other week until 3 November 2023. Data collection ceased after this date because the sod had gone dormant (Fig. 2.3B). Winter recovery started in April 2024 (Fig. 2.4A and B) and the study area in Olathe was rated on 17 April and 14 May 2024 for visual turfgrass quality, NDVI, greenup, and weed infestation. The experiment in Manhattan was rated on 15 and 24 April 2024, respectively, for visual turfgrass quality, NDVI, winter greenup, and weed infestation (particularly tall fescue). Mowing resumed after all plots greened up in May 2024 (Fig. 2.3 C) at a height of 0.625 inches every other day. On 18 June 2024, data was collected on NDVI and TQ.

Spring greenup measures the transition from winter dormancy to active spring growth. It is based on plot color and not genetic color; the visual rating is based on a 1 to 9 scale (1 = straw brown and 9 = completely green) (NTEP, 2000). During this period, no irrigation or other management practices were applied except that plots were mowed. The study area in Olathe began experiencing drought stress compared to Manhattan and on 8 July, NDVI and turfgrass quality ratings were taken again in this site.

Data Analysis

Data at each site were analyzed separately due to differences in sodding date and data collection frequency. All data for each of the ratings were subjected to Analysis of Variance (ANOVA) in a Statistical Analysis System (SAS Institute Inc., Cary, NC). A mixed model analysis was conducted using the proc mixed procedure to evaluate the effects of various factors

on the response variables NDVI, turfgrass quality, weed Infestation, leaf yellowing, and drought responses. The primary factors of interest, aerification, cultivars, mowing height, nitrogen fertilization, and date were specified as fixed effects. These fixed effects were selected because they represent specific, controlled treatments in this study and are of primary interest in assessing their influence on the response variables. Replications were incorporated as random effects to account for variability. Post hoc comparisons were conducted using least squares means (LSMEANS) and were separated according to Fisher's protected Least Significant Differences ($P \leq 0.05$).

Results and Discussion

Zoysiagrass Sod Performance in Olathe and Manhattan, KS

The NDVI and TQ of Innovation and Meyer zoysiagrass were not affected by aerification at either location. However, the analysis revealed a significant three-way interaction ($P < .0001$) among cultivar x aerification x date for NDVI (Table 2.2). Further analysis indicated that this interaction was driven specifically by DALZ 1808. A significant decline in NDVI was observed for DALZ1808 on aerified soil compared to Meyer and Innovation on aerified or nonaerified on June 1, June 9, June 23, and June 28 (Fig. 2.6A), indicating that this genotype's NDVI response was sensitive to aerification effects on these dates.

Similar to the NDVI effect, turfgrass quality showed a significant effect on the three-way interaction (cultivar x aerification x date) exclusively in DALZ 1808 on the aerified plot in Olathe. DALZ 1808 had lower TQ in Olathe than all other cultivars on aerified or nonaerified soil on June 9, June 28, July 7, July 14, and July 21 (Fig 2.6B), indicating the specific periods where the combination of cultivar and aerification influenced TQ of DALZ 1808 under certain seasonal conditions.

In Manhattan, the three-way interaction (cultivar x aerification x date) had a significant impact on the NDVI values of DALZ 1808 when compared to Meyer and Innovation on aerified and nonaerified soil, this is similar to the result in Olathe. DALZ 1808 on aerified soil had lower NDVI levels for 3 out of the 20 rating dates: on June 23, June 30, and July 14 (Fig. 2.7). However, aerification did not affect the TQ (Table 2.2) of either DALZ 1808 or Meyer and Innovation. There are limited studies related to sod performance after installation on aerified soil. Aerification with hollow and solid tines on compacted soil prior to installing Kentucky bluegrass (*Poa pratensis*) enhanced sod performance (Lee and Rieke, 1993). Specifically, rooting was enhanced weeks after its installation, and the force required to remove sod from the surface increased. In contrast, our study showed that aerification prior to installing sod on non-compacted soil in Olathe and Manhattan had a negative impact on DALZ 1808 NDVI values but not on Meyer or Innovation (Fig. 2.6 & Fig. 2.7). Also, there was no rooting effect on the three zoysiagrasses compared by aerification, nine days after installation (Table 2.3).

In Olathe, mowing height had a small but significant ($P = 0.04$) effect on NDVI (Table 2.2). High mowing height had an NDVI of 0.69 and low mowing height had an NDVI of 0.68. Visual TQ was not affected, so mowing height had no impact on zoysiagrass' visual appearance.

The interaction between mowing height and date ($P = 0.023$) was also significant, indicating that the slight effects of mowing height on NDVI varied over time (Table 2.2).

Effects of grow-in conditions on rooting in Olathe, KS, and Manhattan, KS

There were no significant effects of aerification, nitrogen, or cultivar on rooting. Furthermore, there were no significant interactions among these factors. However, the analysis of variance (ANOVA) for root length in Olathe, KS (Table 2.3) shows a significant effect of mowing height (MH) on root number ($P < 0.012$). Low mowing height (0.75 in.) had higher root

numbers (554 ft⁻²) compared to high mowing height (1.5 in.) which had a low count of root numbers (473 ft⁻²). This suggests that the grow-in mowing height influences the number of roots formed in the early stages of sod establishment in Olathe. Interestingly, studies have shown that lowering the mowing heights can reduce root mass and root density in warm-season grasses like zoysiagrass, although it may also increase tiller numbers (Wherley and Sinclair, 2009; Busey 2003; Trenholm and Unruh 2002).

In Manhattan, the ANOVA showed significant differences among cultivars (Table 2.3; $P = 0.038$) with Meyer having the longest root length (0.243 in.), followed by Innovation (0.196 in) and DALZ 1808 (0.190 in.) nine days after sod installation. However, similar to Olathe, no significant effects were observed for other factors such as aerification, mowing height, or nitrogen fertilization, nor were there significant interactions among these factors. Early root growth doesn't reflect total rooting. More information on genotype rooting is presented in chapter 3 of the thesis.

Zoysiagrass Weed infestation, Leaf Yellowing, and Drought tolerance in Olathe and Manhattan, KS

Analysis of variance (ANOVA) on weed infestation ratings in Olathe indicated some interactions between mowing height x nitrogen levels, and cultivar x mowing height x nitrogen levels (Table 2.4). However, weed levels were extremely low (< 4%), so detailed results are not presented here.

In Manhattan, differences in leaf yellowing across cultivars were statistically significant (Table 2.4; $P = 0.004$) based on ANOVA, with observed values of 8.4 (Meyer), 8.6 (Innovation) and 8.9 (DALZ 1808), indicating variations across cultivars.

In Olathe, cultivars showed different responses to drought stress (Table 2.4; $P = 0.004$), with Meyer exhibiting the most stress (rating 6.2 on a 1-9 scale) compared to Innovation (7.1) and DALZ 1808 (7.3). Other compared zoysiagrass genotypes demonstrated better drought response than Meyer in a study conducted in Georgia (Jespersen and Schwartz, 2018). Specifically, an experimental genotype '09-TZ-54-9' had a TQ rating of 7.8 after 20 days of drought compared to 5.3 in Zeon, 5.2 in Meyer, and 5.0 in Palisades. This supports our result that showed that other genotypes performed better in drought compared to Meyer and some other commercial cultivars.

The ratings of NDVI, TQ, green up, and weed infestation that were done at both locations in 2024 showed no significant differences (ANOVA not provided) among cultivars that had been sodded, regardless of the earlier mowing heights and nitrogen fertilization applied during the grow-in period.

Conclusion

Aerification before sodding reduced the NDVI and TQ for DALZ 1808 in Olathe and NDVI in Manhattan, but did not influence Meyer or Innovation. The NDVI and TQ for DALZ 1808 improved over time, so aerification only had a negative influence for a couple of weeks after the installation of DALZ 1808 sod. While aggressive aerification can disrupt soil structure, further research is needed to understand its effects on various zoysiagrass genotypes. In Olathe, the sod root number was higher for cultivars at low mowing heights, while Meyer had slightly greater root length in Manhattan. Weed infestation was most notable for DALZ 1808 under low mowing and nitrogen conditions and for Innovation under high mowing and low nitrogen, although the differences were minimal under 4%. Drought stress was more significant in Meyer than in Innovation and DALZ 1808 in Olathe. In Manhattan, Meyer also exhibited significant

leaf discoloration (yellowing). No significant differences were observed in spring green-up across cultivars in 2024 at either location.

8	11		4	9		5	9
3	6		5	8		3	1
10	4		2	10		1	2
1	7		11	6		4	6
7	1		9	11		10	4
9	5		3	5		12	12
12	8		10	7		6	5
11	2		12	12		11	8
2	12		1	2		7	7
6	10		7	1		9	11
4	9		6	3		2	3
5	3		8	4		8	10

Figure 2.1 Whole plot layout with three replicates of each. The yellow color indicates an aerified plot, the white color is non-aerified and the blue color is the alleyway. Numbers indicate sod installed in each based upon the grow-in strategy as indicated in Table 2.1.



Figure 2.2 Preparation of experimental plots with aerified whole plots marked as ‘A’.



Figure 2.3 All pictures were taken in Manhattan. (A) The picture was taken on 22 September 2023 before zoysiagrass dormancy in the fall; (B) Picture was taken on 1 December 2023 during zoysiagrass dormancy; (C) The picture was taken 18 June 2024 showing full recovery from dormancy.



Figure 2.4 (A) Spring green up of experimental plot on April 17, 2024. (B) Winter recovery of experimental plot in Manhattan on April 24, 2024.



Figure 2.5 Nine days after the sod was installed, one-foot length of each plot was flipped, and two 2-inch diam. rings were randomly thrown to count new roots after sod establishment.

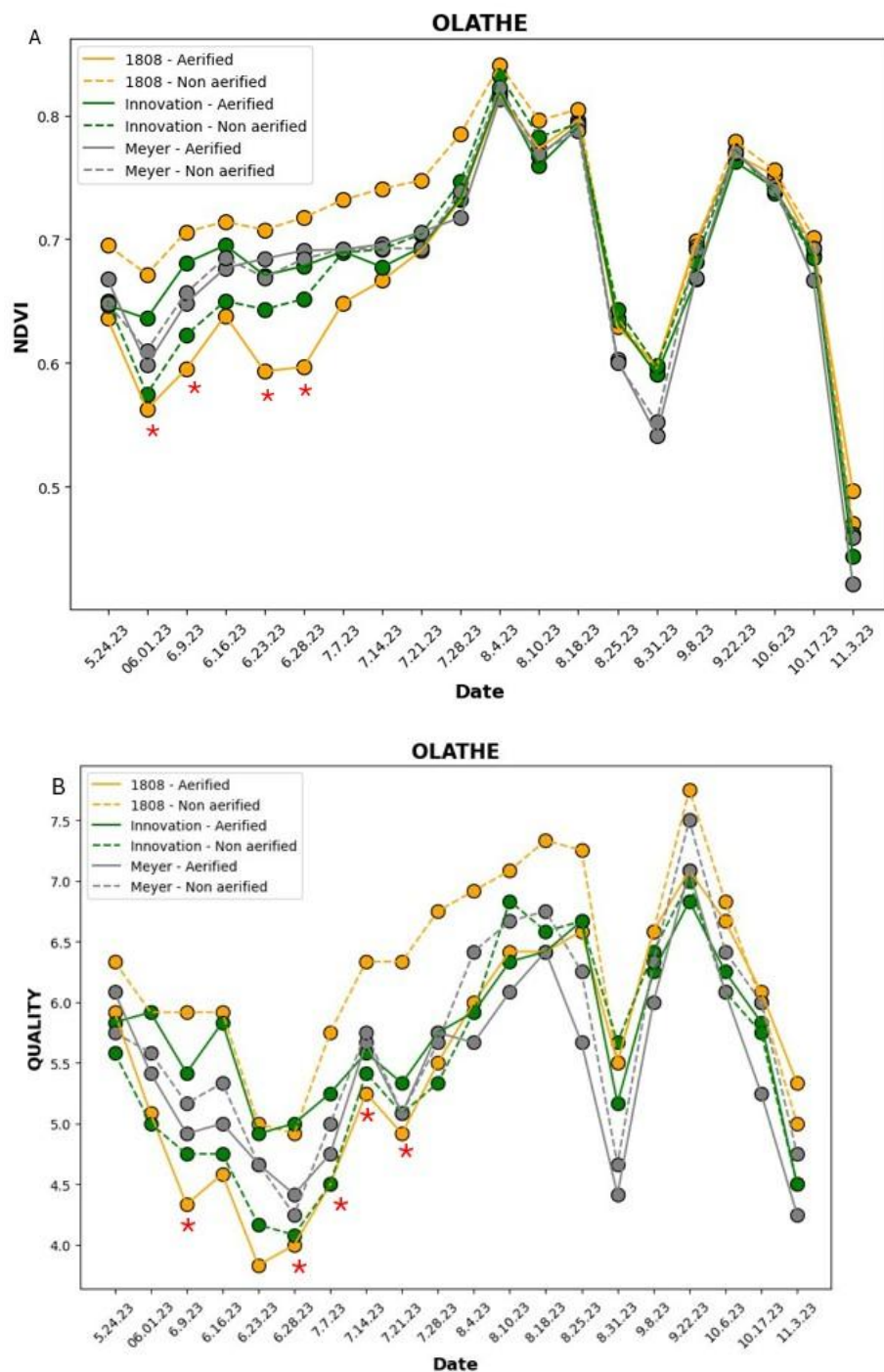


Figure 2.6 Cultivar $\times$ aerification $\times$ date interaction effects on the NDVI (A) and quality ratings (B) of Meyer, DALZ 1808 and Innovation on aerified and nonaerified soil in Olathe. Data points reflect the mean NDVI (A) and quality (B) values comparing the performance of each cultivar across the study period. Asterisks (*) on specific dates indicate significant declines in NDVI or quality values of DALZ 1808 sodded in aerified soil compared to Meyer and Innovation that had no significant differences on aerified or nonaerified soil.

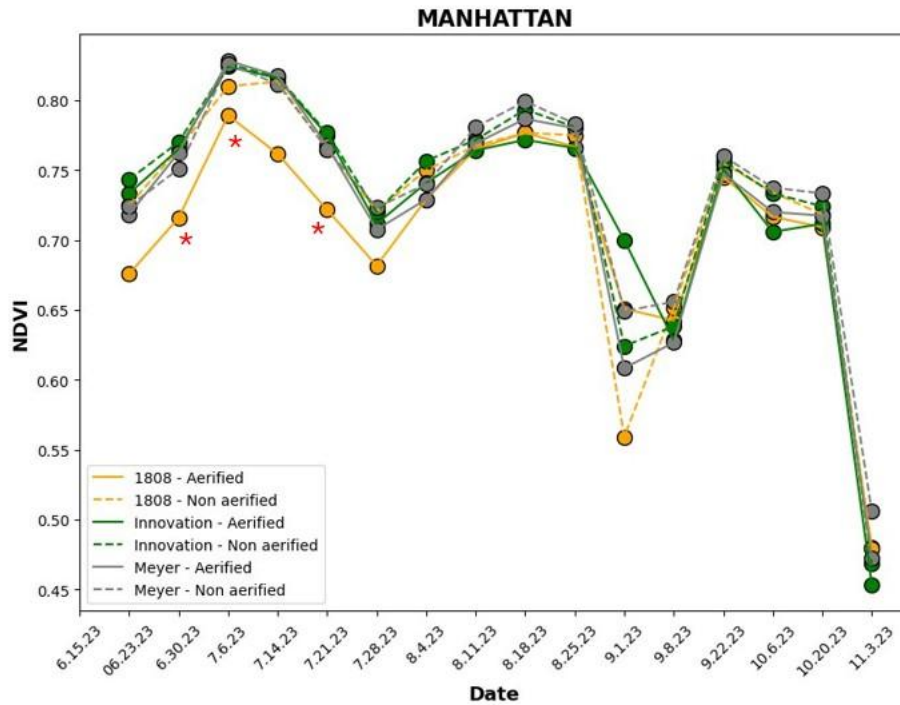


Figure 2.7 Effects of cultivar $\times$ aerification $\times$ date interaction on the NDVI of Meyer, DALZ 1808 and Innovation zoysiagrass cultivars on aerified and nonaerified soil in Manhattan over the study period. Data points illustrate the mean NDVI values, reflecting seasonal fluctuations and cultivar responses to aerification practices across various dates. Asterisks (*) on specific dates indicate significant declines in NDVI values for DALZ 1808 as compared to Meyer and Innovation, marking periods of notable response in Manhattan.

Table 2.1 Twelve total subplots from the grow-in experiment in June 2021. Each cultivar received either 1.6 lbs./1000 ft²/yr (low) or 3.6 lbs./1000 ft²/yr (high) and 0.75 in. (low) or 1.5 in. (high) mowing heights.

Plot number	Cultivar	Mowing Height	Nitrogen Fertilization
1	Innovation	Low	Low
2	Innovation	Low	High
3	Innovation	High	Low
4	Innovation	High	High
5	DALZ 1808	Low	Low
6	DALZ 1808	Low	High
7	DALZ 1808	High	Low
8	DALZ 1808	High	High
9	Meyer	Low	Low
10	Meyer	Low	High
11	Meyer	High	Low
12	Meyer	High	High

Table 2.2 Analysis of variance on the effects of aerification, cultivars, nitrogen levels, Mowing height Date, and their interaction on NDVI and turfgrass quality (TQ) in Manhattan and Olathe, KS.

Source	Manhattan				Olathe			
	df [†]	NDVI	df	TQ	df	NDVI	df	TQ
Block	2		2		2		2	
Aerification (A)	1	ns	1	ns	1	ns	1	ns
Cultivar (C)	2	ns	2	ns	2	ns	2	ns
Nitrogen (N)	1	ns	1	ns	1	ns	1	ns
Mowing Height (MH)	1	ns	1	ns	1	*	1	ns
Date (D)	10	***	11	***	14	***	14	***
C×A	2	ns	2	ns	2	***	2	**
MH ×A	1	ns	1	ns	1	ns	1	ns
C×MH	2	ns	2	ns	2	ns	2	ns
C×MH ×A	2	ns	2	ns	2	ns	2	ns
N ×A	1	ns	1	ns	1	ns	1	ns
C ×N	2	ns	2	ns	2	ns	2	ns
C×N×A	2	ns	2	ns	2	ns	2	ns
MH ×N	1	ns	1	*	1	ns	1	ns
MH ×N×A	1	ns	1	ns	1	ns	1	ns
C×MH×N	2	ns	2	ns	2	ns	2	ns
C×MH×N×A	2	ns	2	ns	2	ns	2	ns
A×D	10	*	11	ns	14	ns	14	ns
C×D	20	ns	22	ns	28	**	28	*
C×A×D	20	*	22	ns	28	***	28	*
MH × D	10	ns	11	ns	14	*	14	ns
MH×A×D	10	ns	11	ns	14	ns	14	ns
C×MH×D	20	ns	22	ns	28	ns	28	ns
C×MH×A×D	20	ns	22	ns	28	ns	28	ns
N× D	10	ns	11	ns	14	ns	14	ns
N×A×D	10	ns	11	ns	14	*	14	ns
C×N×D	20	ns	22	ns	28	ns	28	ns
C×N×A×D	20	ns	22	ns	28	ns	28	ns
MH×N×D	10	*	11	ns	14	ns	14	ns
MH×N×A× D	10	ns	11	ns	14	ns	14	ns
C×MH×N×D	20	ns	22	ns	28	*	28	ns
C×MH×N×A×D	20	ns	22	ns	28	ns	28	ns

[†] df is the degree of freedom associated with each factor.

[‡] *, **, *** significant at $P < 0.05$, 0.01 and 0.001, respectively.

ns means not significant

Table 2.3 Analysis of Variance (ANOVA) on the effects of aerification (A), cultivar (C), mowing height (MH), and nitrogen level (N) on root length and root number in Manhattan and Olathe

Manhattan					Olathe				
Source	df	Root length	df	Root number	Source	df	Root length	df	Root number
A	1	ns	1	ns	A	1	ns	1	ns
C	2	*	2	ns	C	2	ns	2	ns
MH	1	ns	1	ns	MH	1	ns	1	*
N	1	ns	1	ns	N	1	ns	1	ns
C*A	2	ns	2	ns	C*A	2	ns	2	ns
MH*A	1	ns	1	ns	MH*A	1	ns	1	ns
C*MH	2	ns	2	ns	C*MH	2	ns	2	ns
MH*N	1	ns	1	ns	MH*N	1	ns	1	ns
N*A	1	ns	1	ns	N*A	1	ns	1	ns
C*MH*A	2	ns	2	ns	C*MH*A	2	ns	2	ns
C*N*A	2	ns	2	ns	C*N*A	2	ns	2	ns
MH*N*A	1	ns	1	ns	MH*N*A	1	ns	1	ns
C*MH*N	2	ns	2	ns	C*MH*N	2	ns	2	ns
C*MH*N*A	2	ns	2	ns	C*MH*N*A	2	ns	2	ns

[†] df is the degree of freedom associated with each factor.

[‡] *, **, *** significant at $P < 0.05$, 01 and 0.001, respectively.

ns means not significant

Table 2.4 Analysis of Variance (ANOVA) on the effects of aerification (A), cultivar (C), mowing height (MH), and nitrogen level (N) on weed infestation, leaf yellowing, drought, and their interactions in Manhattan and Olathe, KS.

Manhattan							Olathe						
Source	df	Weed Infestation	df	Leaf yellowing	df	Drought	Source	df	Weed Infestation	df	Leaf yellowing	df	Drought
A	1	ns	1	ns	1	ns	A	1	ns	1	ns	1	ns
C	2	ns	2	*	2	ns	C	2	ns	2	ns	2	**
MH	1	ns	1	ns	1	ns	MH	1	ns	1	ns	1	ns
N	1	ns	1	ns	1	ns	N	1	ns	1	ns	1	ns
C*A	2	ns	2	ns	2	ns	C*A	2	ns	2	ns	2	ns
MH*A	1	ns	1	ns	1	ns	MH*A	1	**	1	ns	1	ns
C*MH	2	ns	2	ns	2	ns	C*MH	2	ns	2	ns	2	ns
MH*N	1	ns	1	ns	1	ns	MH*N	1	ns	1	ns	1	ns
N*A	1	ns	1	ns	1	ns	N*A	1	ns	1	ns	1	ns
C*MH*A	2	ns	2	ns	2	ns	C*MH*A	2	ns	2	ns	2	ns
C*N*A	2	ns	2	ns	2	ns	C*N*A	2	ns	2	ns	2	ns
MH*N*A	1	ns	1	ns	1	ns	MH*N*A	1	ns	1	ns	1	ns
C*MH*N	2	ns	2	ns	2	ns	C*MH*N	2	*	2	ns	2	ns
C*MH*N*A	2	ns	2	ns	2	ns	C*MH*N*A	2	ns	2	ns	2	ns

† df is the degree of freedom associated with each factor.

‡ *, **, *** significant at $P < 0.05$, 01 and 0.001, respectively.

ns means not significant

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Chapter 3 - Rooting Characteristics of Selected Zoysiagrass Genotypes in a Controlled-environment

This chapter has been prepared using style guidelines for submission to the *HortScience* Journal

Abstract

Zoysiagrass (*Zoysia spp.*) exhibits drought avoidance potential, due to its deep rooting ability, allowing water uptake from deeper soil. However, specific root architecture traits of different genetic lines have not been examined in detail. The objectives of this study were to (i) quantify the differences in root characteristics of twelve genotypes and experimental genotypes of zoysiagrass under greenhouse conditions; and (ii) determine potential correlations between rooting characteristics and shoot quality that can contribute to drought avoidance mechanisms. The study was conducted for 12 weeks in 2023 (Run 1) and 2024 (Run 2) at the Throckmorton Plant Science Center, Manhattan, Kansas. Each genotype was grown in calcined clay-filled polyvinyl chloride (PVC) tubes with weekly measurements taken on shoot quality visually on a 1-9 scale and maximum root length after 12 weeks. At the end of the experiment, root traits (length, surface area, and volume) at 0-10, 10-20, 20-30, 30-60, and 60-90 cm were analyzed using WinRHIZO Pro. Shoot-to-root ratio (SRR) was calculated as total shoot mass by total root mass. Differences in root length, surface area, and volume were observed among genotypes ($P < 0.0001$), with Chisholm and FAES 1319 performing better than a few others including Zeon, DALZ 1702 and KSUZ 1201, which had lower root growth and shoot quality. Root traits were greatest in the top 10 cm, with substantial declines at deeper depths (60-90 cm). The SRR in Run 1 had no significant differences; however, in run 2, KSUZ 1201 was significantly greater than Innovation, DALZ 1311, Meyer and Gateway while Zeon and Emerald were statistically greater than Gateway. Conversely, Zeon had good shoot quality in each experiment but exhibited low root growth. Numerous genotypes with relatively good shoot quality generally had comparable rooting results in both experimental runs. Field-based validation of these findings may be required to further inform genotype recommendations.

Introduction

Zoysiagrass (*Zoysia spp.*), a warm-season turfgrass species, is recognized for its drought tolerance and efficient water use. Studies have reported that its daily evapotranspiration rate may vary between 2.5 and 6.0 mm per day under typical conditions, though this can vary based on climate, soil type, and irrigation practices (Colmer and Barton, 2017). Zoysiagrass may avoid drought by developing an extensive root system that enables water extraction deep in the soil (Huang et al., 2014; Hurd, 1975; Qian et al., 1997). Drought avoidance is a major strategy of drought resistance (Huang et al., 2014). Key traits for drought avoidance include deeper root systems, which allow access to water from deeper soil layers, and stomatal regulation to reduce transpiration (Wang et al 2022; Fan et al., 2020). Drought-avoiding genotypes may possess different characteristics in response to drought, including enhanced root plasticity, deeper root extension, decreased leaf growth, and modification in the number and aperture of stomatal complex (Duncan and Carrow, 1999; Huang et al., 2014; Nilsen and Orcutt, 1996; Simpson, 1981). Management strategies can also influence drought avoidance characteristics. Meyer zoysiagrass (*Zoysia japonica* Steud) had reduced shoot growth rates, increased root elongation, and reduced drought symptoms during dry-down when deep infrequent irrigation was applied (Qian et al., 1996). Turfgrasses, including zoysiagrass, are typically maintained at a height of < 8 cm, which limits most roots to the upper 10 cm of the soil profile (Madison, 1971). Therefore, periodic precipitation or irrigation is required to prevent drought stress. Evaluation and selection of turfgrass species, including zoysiagrass genotypes and cultivars, with deep root systems that tap greater volumes of soil water is critical (Boeker, 1974). Newer cultivars, especially genotypes developed from *Z. japonica*, perform more favorably in response to drought (Wherley

et al., 2014). Much of the initial research on zoysiagrass drought response was conducted in Georgia or Texas with Meyer or Emerald zoysiagrass (Patton et al., 2017).

Deep rooting has been considered a drought-avoidance mechanism in several plants (Rosa et al., 2019; Jespersen and Schwartz, 2018). Characterizing root morphological properties such as total root length, root system surface area, average root diameter, and branching patterns can be assessed accurately using new root analysis software based on image analysis (Rosa et al., 2019). According to Bouma et al. (2000) and Himmelbauer et al. (2004), root length and root diameter distribution measurements provided by the WinRHIZO software (Regent Instruments, 1991) are accurate when the correct scanning protocol is followed. Therefore, computer-assisted root imaging is an opportunity to facilitate the analysis process and improve accuracy (Rosa et al., 2019).

Drought tolerance, on the other hand, allows turfgrass to survive extreme water stress by altering metabolic processes to endure dehydration. Grasses like buffalograss and zoysiagrass exhibit these traits, enabling them to survive through water stress and quickly recover once water becomes available (Fan et al., 2020; Kim and Beard, 2018). Zoysiagrass tolerates severe drought by entering a dormant state and subsequently fully recovers after significant rainfall even after long dormancy periods (Beard and Sifers, 1997; Steinke et al., 2009; Fry and McFadden, 2021).

The objectives of this greenhouse study were to (i) examine the differences in root characteristics of twelve experimental genotypes and commercialized cultivars; and (ii) determine potential correlations between rooting characteristics and shoot quality that can contribute to drought avoidance mechanisms. Experimental genotypes and commercialized cultivars will now be referred to as genotypes in this study.

Materials and Methods

Material Preparation

An experiment was conducted under a temperature-controlled greenhouse facility at the Throckmorton Plant Science Center at Kansas State University, Manhattan, KS from 25 May to 24 August 2023 (Run 1) and 16 May to 14 August 2024 (Run 2) to determine the differences in the root growth of zoysiagrass breeding genotypes and commercial cultivars (all referred to genotypes herein). Small polyvinyl chloride (PVC) pipes were used to hold media to grow 12 genotypes of zoysiagrass that were replicated five times in a completely randomized design. The pipes were 113 cm long and 6.4 cm in diameter.

In the PVC pipe, a clear polyethylene plastic measuring 121 cm long and 5 cm in diameter served as the root tube to minimize restrictions in root growth and allow the harvest of the entire root system with minimal damage. Each pipe was one experimental unit with 60 columns used for each experimental Run. Each root tube was filled with 1020 g of calcined clay (Turface Athletics, PROFILE Products LLC, Grove, IL.) as a growing media and vibrated by hand until a uniform column was formed. Tubes were perforated at the base to allow for drainage. A 24-4-13 N-P-K soluble fertilizer (Scotts Miracle-Gro, Marysville, OH.) at 1.4 g was mixed with 1400 ml of water and 20 ml of the solution was applied to each clay-filled PVC tube. The soil mix was saturated with water, after which zoysiagrass genotypes were plugged. Each tube was set in the PVC columns in a completely randomized design and was supported at a 20° angle from vertical in a tube rack to observe root growth along the side of root tubes (Fig. 3.1).

The 12 zoysiagrass genotypes evaluated were Meyer, Emerald, Zeon, Innovation, Chisholm, Gateway, KSUZ 1201, DALZ 1701, DALZ 1702, DALZ 1808, DALZ 1311, and FAES 1319. These plugs were collected from a pre-existing turfgrass plot at the Olathe

Horticulture Research Center, Olathe KS on 23 May 2023 for Run 1, and on 14 May 2024 for Run 2 with a 3.2 cm diameter soil auger. Soil was hand washed, and roots were trimmed to the crown to accelerate the formation and growth of new roots from the crown and nodes. Each plug was planted in a root tube as described above.

The greenhouse temperature for (Argus Control Systems, Manitoba, CN) day and night was set at 29°F and 21°C, respectively with no supplemental light. The actual temperature was not monitored. For the first three weeks in Run 1, 20 ml of the fertilizer solution was applied to each tube weekly, and subsequently, the application was reduced to 10 ml of the fertilizer solution twice weekly to give 0.731g N/m². for each application. Irrigation mist was applied manually using an irrigation hose three times a day to establish root growth. The actual amount of water applied as mist was not measured. Irrigation was reduced to twice a day in the third week after the roots had become visible in clear tubes for all genotypes.

Seven weeks after planting, irrigation was reduced to once a day after root growth had fully developed. In the ninth week, irrigation was applied every other day until the end of the study. Mowing began in the third week of establishment and clipped at 2.54 cm once weekly for the first five weeks, subsequently, clippings were collected and bagged twice weekly because of the aggressive growth of the shoot.

Data collection

Maximum root length at the end of the experiment was calculated by summing the individual weekly root lengths measured (Fig. 3.2). Turfgrass shoot quality was visually estimated weekly on a 1-9 scale, (1 = poor quality; 6 = minimally acceptable quality; 9 = optimal density, texture, color, and uniformity). At the end of the 12-week experiment, root tubes were placed horizontally, and the shoots were removed using scissors and bagged while root tubes

were sectioned into 0 to 10, 10 to 20, 20 to 30, 30 to 60, and 60 to 90 cm. Soil media was hand-washed from each root with tap water, and packed in resealable zipper storage plastic bags, which were stored in a 4°C storage room.

Roots from each section were placed in a transparent acrylic tray 20 cm wide and 30 cm long in a thin layer of water (6-8 mm height) and scanned using a root scanner (Epson Perfection; Epson model V850 Pro). Images were processed using the WinRHIZO Pro image analysis system (Regent Instrument) to estimate the root length by depth, surface area by depth, and root volume by depth for each 10-cm depth zone. Maximum root length was defined as the cumulative length of all root segments within each depth, indicating the potential reach for water for each genotype. A greater surface area generally indicates a higher efficiency in resource acquisition. Additionally, the larger root surface area created by root hairs and branching patterns enhances the plant's capacity to absorb water enabling roots to anchor water plants effectively (Cai and Ahmed, 2022). The total root volume estimates the entire three-dimensional space occupied by the roots. This measurement can indicate the plant's overall root biomass and storage capacity, playing a critical role in water retention (Regents Instruments 2024). After imaging, the roots and shoots collected were dried at 100°C and weighed when constant dry weight was achieved.

The shoot/root ratio (SRR) was calculated by dividing the cumulative total clipping and shoot mass by the total root mass of each genotype per replication; it is the ratio of dry root weight to dry shoot weight.

Data Analysis

Data were analyzed using PROC GLIMMIX and PROC MIXED procedures in SAS to assess maximum root extension, shoot quality, SRR, and WinRHIZO-derived root traits across

different depths, genotypes, and years. Genotype, year, and depth were treated as fixed effects to evaluate specific influences on trait performance. Random effects included replications and years to account for variability. The slice option was used for interactions, such as genotype by year to examine interactions within specific years or depths. Root traits were tested for treatment and year effect. The least square means statement was used to calculate and compare among genotypes with Tukey's adjustment applied to control for multiple comparisons.

Results and Discussion

Genotypes evaluated in this study exhibited variation for all measured traits which aligns with other studies (Christensen et al., 2017; Jespersen and Schwartz, 2018; Marcum et al., 1995).

Maximum Root Length (MRL)

The effect of genotypes, year, and their interaction on MRL showed that there were differences among genotypes ($P < 0.0001$) and across years ($P = 0.0094$) (Fig. 3.3). Maximum root length varied significantly among genotypes in both years (Fig. 3.3), with some genotypes like Chisholm (53.5 cm in 2023 and 56.7 cm in 2024) and DALZ 1311 (40.5 cm in 2023 and 61.5 cm in 2024) showing consistently greater MRL across 2023 and 2024. These findings align with research suggesting that zoysiagrass genotypes with deeper or more extensive root systems tend to have enhanced drought avoidance capabilities, as deeper roots allow access to water reserves in the soil (Colmer and Barton, 2017). Specifically, Chisholm's consistent MRL across both years suggests a genetic predisposition for root development that supports greater drought avoidance, a finding supported by Hurd (1975) and Qian et al. (1997), who documented similar root traits in drought-avoidance turfgrasses.

On the other hand, Zeon (28.7 cm in 2023 and 28 cm in 2024) consistently had shorter MRL across both years (Fig. 3.3), which may indicate limited drought avoidance capability.

Shorter root systems could result from genetic limitations in root growth or higher allocation of resources to shoot growth rather than root development. This aligns with research suggesting a trade-off between shoot and root growth in turfgrasses, where genotypes with denser shoots often have restricted root development (Wherley and Sinclair, 2009). This genotype might perform better under well-irrigated conditions but could struggle in environments with limited water availability due to their limited MRL.

Root Length by Depth (RL)

There were significant effects for interaction between genotype and depth ($P < 0.0001$), indicating that both genotype and depth influenced root length, and the effect of depth varied by genotype. The root length (RL) of zoysiagrass genotypes across five soil depths (0-10 cm, 10-20 cm, 20-30 cm and 30-60 cm) for Run 1 and Run 2 is shown in Fig. 3.4. The data shows differences in rooting patterns among genotypes across years, with the highest root lengths generally observed in the upper soil layers (0-30 cm), while root development was limited in the deeper layers (30-60 cm).

The 0-10 cm depth exhibited the greatest RL across most genotypes in both years, particularly for Chisholm, DALZ 1701, DALZ 1808, and FAES 1319. This pattern aligns with the findings of Christensen et al., 2017, who classified zoysiagrass genotypes with dense rooting systems at the upper portion of the soil, which is effective in initial water uptake, supporting early-stage drought avoidance.

At the 10-20 cm depth, DALZ 1701 had the highest root length in 2023, and DALZ 1808 had the highest in 2024. In 2023 several genotypes were equivalent to DALZ 1701; Zeon, DALZ 1702, and KSUZ 1201 had lower lengths compared to DALZ 1701. In 2024, there were

few differences among genotypes; DALZ 1808 was only greater in length at this depth than Zeon.

Root length at 20-30 cm depth showed that Zeon numerically had the lowest root length in each year but was statistically similar to several other genotypes. At depths 30-60 cm and 60-90 cm, RL was minimal across all genotypes in both years, with no significant differences. At depths 30-60 cm and 60-90 cm, RL was minimal across all genotypes in both years, with no significant differences. This finding aligns with the work of Qian et al. (1997), who observed that Meyer zoysiagrass primarily allocates root biomass to upper soil layers, especially when moisture is not limited. While deep rooting capacity is present in some genotypes, it may be constrained by the experimental conditions or limited water availability at these depths, note that environmental conditions were not considered in this study. However, the findings of Wang et al. (2022) support this, suggesting that environmental factors and soil properties play significant roles in limiting root length at deeper levels.

Genotype Surface Area by Depth

The surface area showed significant variation among genotypes, depths, and their interactions ($P < 0.05$). Fig 3.5 shows the surface area of all genotypes by depths for both experimental runs. Surface area is a key indicator of a plant's potential for water absorption, with larger surface areas at shallower depths typically indicating greater access to surface moisture.

In Run 1 and Run 2, the 0-10 cm depth displayed the highest surface area across most genotypes, indicating significant rooting activity near the soil surface. The majority of the genotypes had surface areas greater than or close to 100 cm² except for KSUZ 1201 and DALZ 1702 with surface areas lower than 60 cm². These observations align with studies by Christensen et al. (2017) and Colmer and Barton (2017), which found that zoysiagrass genotypes adapted to

dry environments tend to concentrate roots near the soil surface to maximize water capture during early-stage drought.

At the 10-20 cm depth, the surface area was quite lower than 0-10 cm with all the genotypes falling below 100 cm² (Run 1) and 80 cm² (Run 2). This could reflect a genetic predisposition toward maintaining mid-depth rooting structures, as noted by Jespersen and Schwartz (2018). The stability of these genotypes suggests resilience to moisture fluctuations at this depth. However, genotypes like KSUZ 1201, DALZ 1702 (Run 1), and Zeon (Run 2) still displayed lower root surface area, potentially limiting their ability to extract water effectively from mid-level soil layers.

At deeper depths of 20-30 cm, 30-60 cm and 60-90 cm, root surface area decreased significantly for all genotypes in both years, with only minor differences among them. Particularly, at depths 30-60 cm and 60-90 cm, most genotypes showed minimal surface area (< 40 cm²).

Genotype Root Volume by Depth

The root volume is another important indicator of a plant's ability to absorb water, with greater root volume typically providing improved access to soil resources. Fig. 3.6 shows the root volume of the genotypes across the same depths over two years, highlighting the broader distribution and statistical significance. Similar to the surface area, 0-10 cm was highest among the shallow depths in Run 1 and Run 2, with the majority of the genotypes exhibiting larger root volumes at these depths.

At depth 10-20 cm, most of the genotypes had similar root volumes which maintained high root volumes relative to each other. This consistent rooting pattern across shallow to mid-level depths shows the adaptability of these genotypes to draw from multiple soil layers,

supporting improved drought avoidance. Jespersen and Schwartz (2018) found similar patterns, showing that zoysiagrass genotypes with significant root presence at upper 20 cm are better equipped to withstand drought conditions due to their ability to access water at intermediate depths.

At depths, 20-30 cm, 30-60 cm, and 60-90 cm, root volume was minimal with all genotypes corresponding to each other. Potentially, deeper rooting in genotypes like Chisholm, DALZ 1707, FAES 1319 and some others could suggest a strategic distribution of root biomass to access more stable moisture levels during extended dry periods, a trait often seen in drought-tolerant turfgrass genotypes.

These observations associated with maximum root depth and root length, surface area, and root volume by depth align with studies by Christensen et al. (2017) and Colmer and Barton (2017), who found that zoysiagrass genotypes adapted to dry environments tend to concentrate roots near the soil surface to maximize water capture during early-stage drought. Results also align with Qian et al. (1997), who observed that Meyer zoysiagrass primarily allocates root biomass to upper soil layers, especially when moisture is not limited.

Zoysiagrass genotypes that exhibited maximum rooting and extensive root characteristics at various depths have the potential for greater drought avoidance. Beard (1973) and Qian et al. (1997) emphasized that deep-rooted genotypes in warm-season grasses tend to be more resilient to drought, as their roots can access subsurface water reserves during dry periods. Conversely, genotypes that exhibited shallow maximum rooting and reduced rooting across depths, such as Zeon, suggest that shallow-rooted cultivars are best suited to environments with frequent rainfall or frequent irrigation (Christensen et al., 2017; Wang et al. (2022) suggest the same.

Shoot-to-Root Ratio (SRR)

The shoot-to-root ratio is an important metric for evaluating the allocation of resources between above-ground (shoot) and below-ground (root) biomass. Data analysis showed that the genotype effect was significant (Fig. 3.7 $P = 0.0010$) indicating that SRR varied significantly among the 12 genotypes studied. The effect of the year was not significant ($P = 0.317$), and neither was the interaction between genotypes and years ($P = 0.222$). When comparing Run 1 and Run 2, SRR among genotypes is similar, with only minor fluctuations noticeable in Run 2. The overall trends suggest that genotypes with higher SRR such as KSUZ 1201, Zeon, Emerald, and other genotypes (Fig. 3.7) prioritize above-ground growth, which could enhance shoot density and aesthetic quality in well-watered settings. However, this comes at the potential cost of reduced drought avoidance due to less developed root systems.

Higher SRR ratios typically indicate greater allocation of biomass above-ground, potentially favoring shoot quality and visual appeal but possibly at the expense of root development (Beard, 1973; Qian et al. 1997), which is critical for drought resilience. Conversely, lower SRR suggests more balanced or even root-dominant allocation, which can be beneficial for drought avoidance as it enhances water absorption. This pattern is consistent with findings from Huang et al. (2014), who documented that warm-season grasses with higher SRR may have increased water demands, as they rely more heavily on surface water uptake to sustain shoot biomass. Similarly, a trend was observed in previous studies where tall fescue with lower SRR exhibited significantly deeper and more extensive rooting than warm-season grasses like Meyer zoysiagrass (Qian et al., 1997). In contrast, genotypes like Gateway, Meyer, DALZ 1311, and Innovation, with lower SRR, display a growth strategy favoring root development, which may

support drought avoidance through increased water absorption capacity which could make them better suited for regions with limited water availability.

Conclusion

This study provides an understanding of the root and shoots characteristics of various zoysiagrass genotypes, highlighting their adaptability to different water conditions. Genotypes like Chisholm, DALZ 1701, and FAES 1319 demonstrated extensive root length and surface area, particularly at shallow and mid-level depths, supporting robust water uptake and drought resilience. These genotypes also maintained high shoot quality with moderate SRR, making them well-suited for environments with fluctuating water availability. Conversely, genotypes like KSUZ 1201, Zeon, and Emerald, with higher SRR and shallower root systems, prioritized shoot growth, which could enhance turfgrass quality in irrigated landscapes but may reduce drought avoidance. Gateway's consistently low SRR reflects a strong root-focused allocation, making it more suitable for water-limited regions. The variation in root volume and distribution among genotypes emphasizes the importance of selecting zoysiagrass types based on site-specific needs, whether for aesthetic quality under frequent irrigation or for drought avoidance. Future research may focus on field-based validation of these findings to further inform genotype recommendations for regions experiencing very predominant drought stress. These results are also essential for breeding programs to emphasize root architecture traits in developing more drought-avoiding zoysiagrass genotypes.



Figure 3.1 PVC pipes with Polyethylene tubes rested at 20 degrees in PVC columns in the greenhouse



Figure 3.2 Weekly measurements of root length with lines drawn to mark the end of each root for the week.



Figure 3.3 Relationship between maximum root length and shoot quality of zoysiagrass genotypes for 2023 (A) and 2024 (B). Maximum root length was defined as the average root lengths across replications after 12 weeks of experiment. The letters above bars represent statistical results ($P < 0.05$); those which have the same letter are not significantly different.

Root Length by Cultivar and Depth (2023 and 2024)

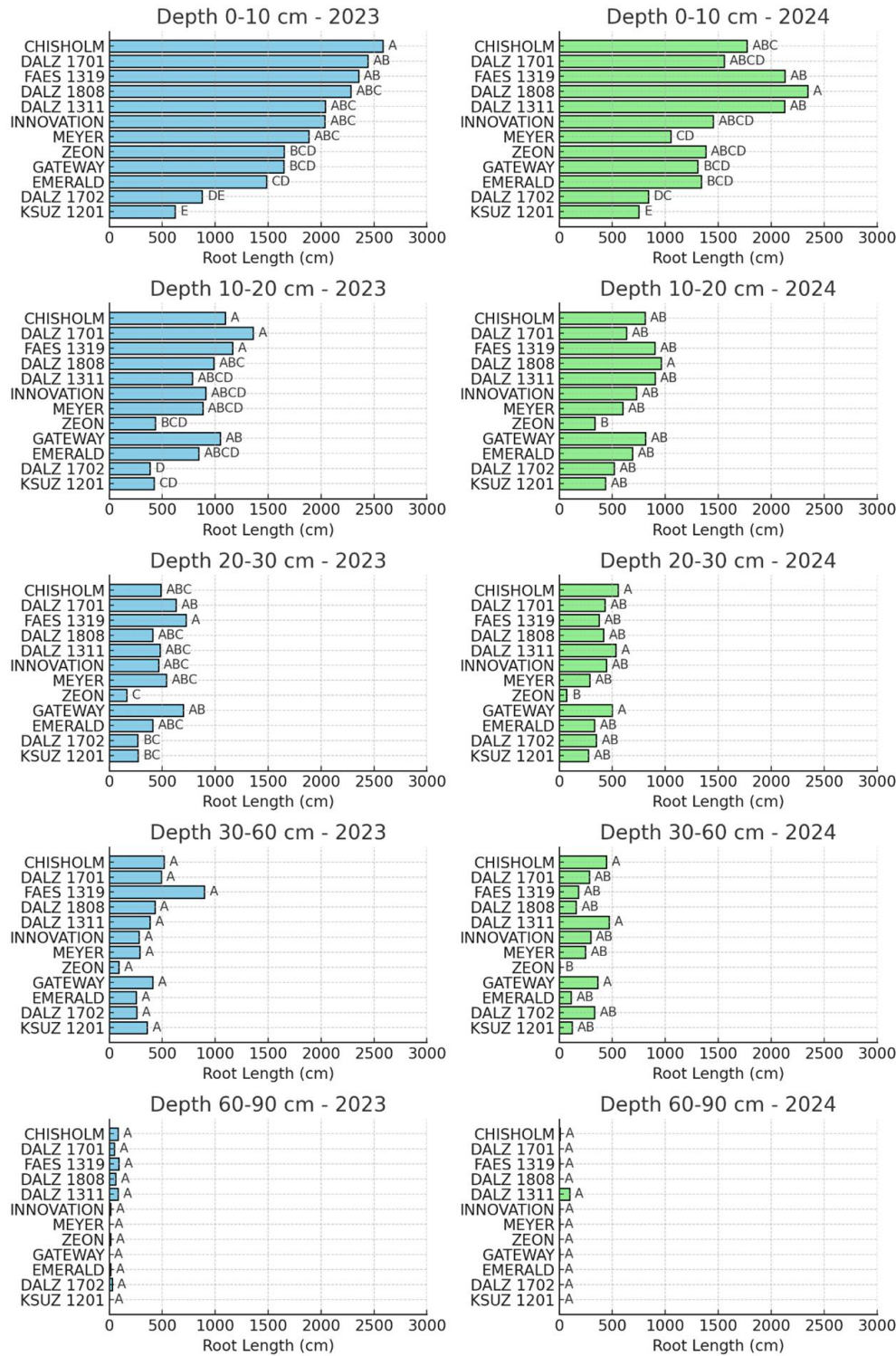


Figure 3.4 Root length of zoysiagrass genotypes at different depths across years. Letters above bars represent statistical results ($P \leq 0.05$); those with the same letter are not significantly different.

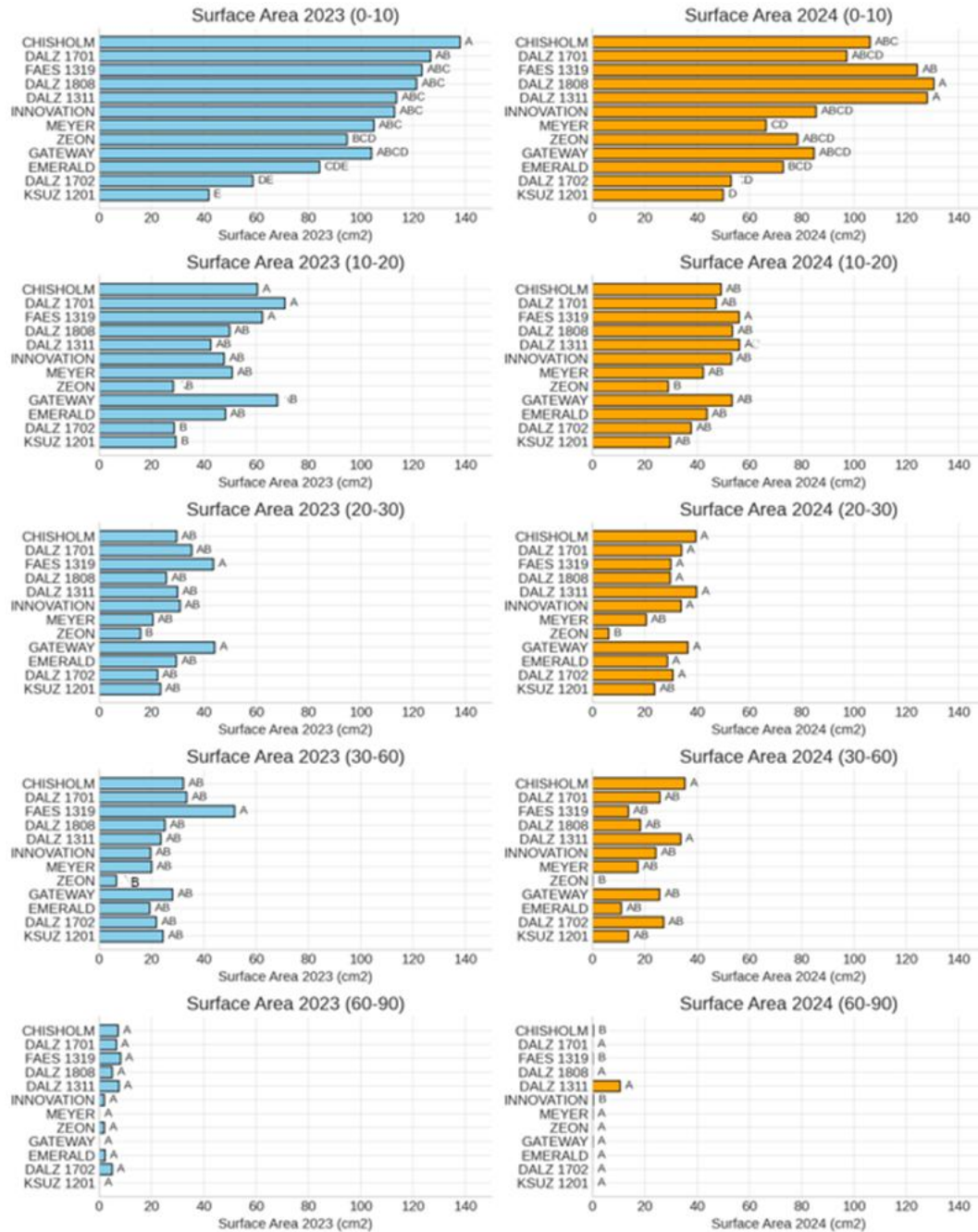


Figure 3.5 Genotype comparison of surface area across depths in 2023 and 2024. Letters above bars represent statistical results ($P \leq 0.05$); those with the same letter are not significantly different.

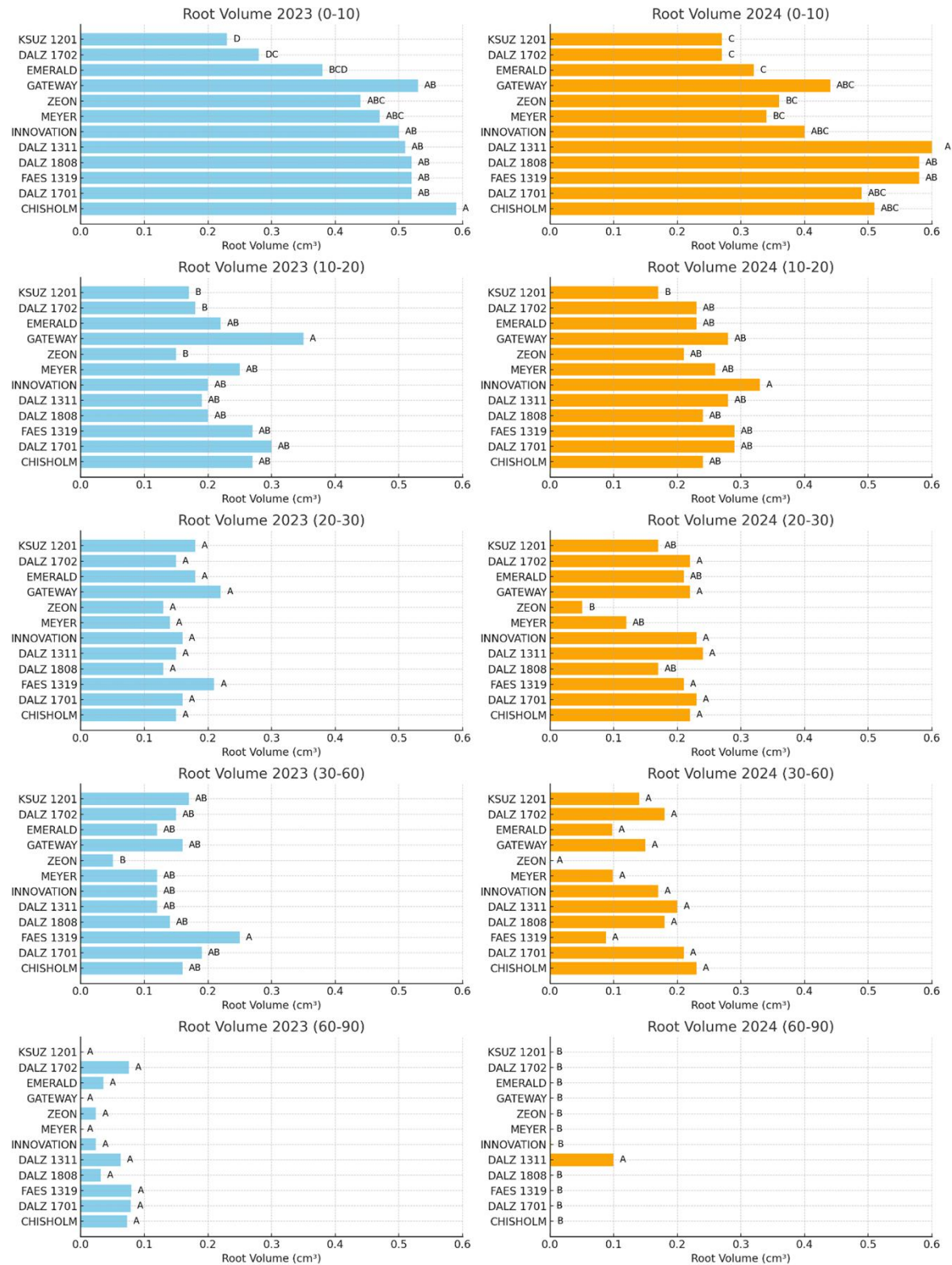


Figure 3.6 Genotype comparison of root volume in 2023 and 2024 across depth. Letters above bars represent statistical results ($P \leq 0.05$); those with the same letter are not significantly different.

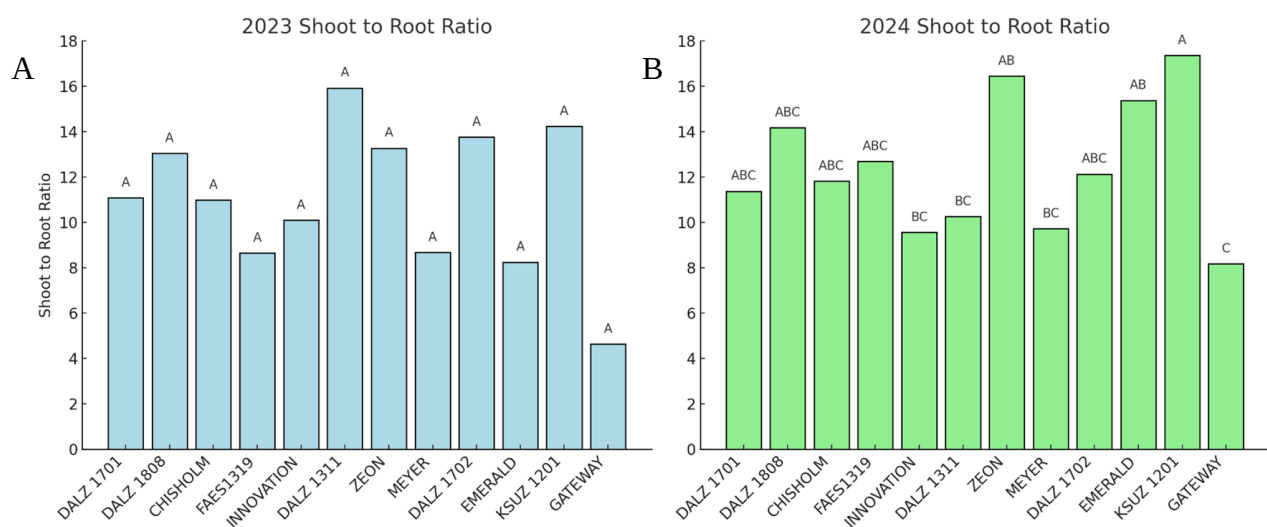


Figure 3.7 Shoot-to-root ratio. This was determined by dividing the combination of total clipping and shoot mass throughout the study period by total root mass across depths. Letters above bars represent statistical results ($P = 0.0004$); those with the same letter are not significantly different.

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Chapter 4 - Evaluation of Thatch Accumulation among Zoysiagrass Genotypes in the Transition Zone and Southern States in the U.S.

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Abstract

Thatch is an intertwined layer of dead and living stems and roots, that accumulates in turfgrass when organic material production outpaces decomposition, affecting turfgrass quality and playability. This study evaluated thatch accumulation and performance of various zoysiagrass genotypes across six locations (Kansas, Oklahoma, Indiana, Texas, and Ft. Lauderdale & Citra, Florida) for National Turfgrass Evaluation Program (NTEP) and United States Golf Association (USGA) trials. Thatch accumulation in the NTEP trial was measured five years after planting, while the USGA trial was assessed three years after planting. The objectives were to: (i) evaluate thatch accumulation across cultivars and breeding genotypes and (ii) evaluate the correlations between zoysiagrass thatch depth or mass vs. other genotype morphological traits. Thatch depth, mass, turfgrass quality, and other traits were measured across locations using a randomized complete block design. In the NTEP trial, cultivar differences in thatch depth were observed only in Dallas, TX, where ‘Emerald’ had the greater thatch depth, while ‘Meyer’, ‘Empire’, and DALZ 1808 had the lowest. The USGA trial showed no significant thatch depth differences in Kansas and Indiana, but minor variations were noted in Citra, FL. Correlations between thatch depth and other traits showed that thatch depth was negatively correlated with turfgrass firmness at Texas and Citra (NTEP trial) and with NDVI at Indiana (USGA trial). Positive correlations were found with tiller numbers at Oklahoma, Texas, and Ft. Lauderdale (NTEP trial). Thatch depth was positively correlated with the thatch mass in Kansas (USGA trial). Environmental conditions could impact the performance of zoysiagrass genotypes, although these conditions were not considered in this study. The correlations noted at each location can give guidance on what may impact thatch development or its influence over time.

Introduction

Thatch, a closely entwined layer of dead and living stems and roots that forms between the soil surface and the green vegetation zone (McCarty, 2018; Beard, 1973) occurs naturally and is caused by organic material from turfgrass that is actively growing at a faster rate than it is breaking down (Couillard and Turgeon, 1997). Excessive thatch can reduce turfgrass quality, playability, pesticide efficacy, and water infiltration (McCarty et al., 2016). The wear resistance of the turfgrass may be improved by a thin layer of thatch, which also acts as a buffer against temperature and moisture extremes. However, excessive thatch accumulation may negatively impact the growth of turfgrasses as well as the properties of the underlying soil, increasing the risk of disease and insect problems, making the soil more susceptible to heat, cold, and drought stress, and increasing soil compaction (Couillard and Turgeon, 1997).

A previous study (Ledeboer and Skogley, 1967) reported that thatch composition is mainly made up of cellulose, hemicellulose, and lignin. Thatch is made up of intact fibrous roots, sclarified vascular strands of leaf sheaths and stem nodes, and crown tissue (Couillard and Turgeon, 1997). Thatch development is favored in turfgrass species that produce lateral stems and other plant structures high in lignin (Ledeboer and Skogley, 1967). Higher maintenance input levels lead to a rate of plant organic matter buildup that exceeds decomposition, which directly produces thatch formation (Hurto et al., 1980; McWhirter and Ward, 1976). Plant tissues often are produced faster than they decompose especially in high-maintenance turfgrass settings such as the putting green, thereby resulting in thatch accumulation (Weaver et al., 2020).

Studies have shown that the efficacy of herbicides can be significantly affected by thatch layers, which act as a barrier that limits herbicide movement into the soil (McCullough et al., 2007; Hurto and Turgeon, 1979). Zoysiagrass scalping can occur when mowers are driven

unevenly on thatched turfgrass (Dunn et al., 1981). Many people find walking on deep thatch turfgrass uncomfortable, which can lead to poor footing or unstable balance on golf course fairways and tees (Dunn et al., 1981). Thatch management is an important practice to ensure acceptable turfgrass quality and performance (Shaddox and Unruh, 2019). Various methods of measuring thatch include weight-loss-on-ignition (WLOI) for organic matter, rulers for depth, balances for weight, and thatchmeter for compression. Studies have reported that the thatch meter increases the speed of thatch measurements (Shaddox and Unruh, 2019).

Zoysiagrass (*Zoysia* spp. Willd) is a perennial sod-forming species in the United States that is well adapted for use as a turfgrass in the transitional and southern regions (Patton et al., 2017). Zoysiagrass acreage is expanding to golf courses, home lawns, and commercial sites throughout the transition zone and southern U.S. Three main species of zoysiagrass [*Zoysia japonica* Steud., *Zoysia matrella* (L.) Merr., and *Zoysia pacifica* (Goudswaard) M. Hotta and S. Kuroki] were introduced into the United States and have since been used directly as turfgrass or by turfgrass breeders in the development of advanced lines (Patton et al., 2017). These species are adapted to various regions and have variable adaptation, growth, and development that was once considered “unimportant” (Hitchcock, 1935). Since its first adoption over 125 years ago, many scientists and breeders have contributed to the advancement of zoysiagrass in the United States (Patton et al., 2017). It is difficult to maintain a sustainable turfgrass culture when plant roots are developed in thatch because of its ability to dry and the difficulty in rewetting dry thatch (Dunn et al., 1981).

Zoysiagrass breeding can be dated back to 1940 in Beltsville, MD where new germplasms were created using irradiation, directed crosses, and open pollination (Hanson, 1965). Most of the older cultivars are not well adapted to the southern climates, with poor

adaptability to the U.S. transition zone except for Chisholm. During the development of new cultivars, breeders often face challenges with genotype $\times$ environment (GE) interaction, defined as a differential response of genotypes from one environment to another (Kang and Gorman, 1989).

Multi-environment trials are conducted to assess genotype performances across environments (years, locations, or seasons). The aim is to select the best and superior genotypes across different environmental conditions and for specific environments (Comstock, 1977; Annicchiarico, 2002; Smith et al., 2005). Zoysiagrass breeding efforts have been focused on improving traits such as turfgrass quality, salt, and drought tolerance, tolerance to extreme temperature, shade tolerance, and biotic stress resistance. All breeding programs aim to develop regionally adapted cultivars to broaden the use of zoysiagrass. In 2019, 74 genotypes sponsored by the United States Golf Association (USGA) were established in Kansas, Florida, and Indiana and 39 genotypes sponsored by the National Turfgrass Evaluation Program (NTEP) were established in Kansas, Texas, Oklahoma, Florida, and Indiana. The objectives of this study were to: (i) evaluate the variability of thatch accumulation among several zoysiagrass genotypes and cultivars from these diverse geographical locations; and (ii) evaluate the correlations between zoysiagrass thatch depth or mass vs. other genotype morphological traits.

Materials and Methods

Research Site Information

Experimental studies to evaluate thatch accumulation and turfgrass morphological characteristics in 2024 were done on breeding genotypes and commercialized cultivars in National Turfgrass Evaluation Program (NTEP) and United States Golf Association (USGA)

trials. Before planting plugs, the soil was prepared by tilling and leveling. At all locations, plugs were planted in randomized complete block experiments and individual plots measured 5 x 5 or 6 x 6 ft. Approximately 24 plugs measuring approximately 2 inches in diameter. were planted in individual plots for both studies. All locations had a 1 or 2-foot-wide alleyway between all plots. The NTEP trial was initiated in 2019 in Olathe, KS; Stillwater, OK; West Lafayette, IN; Dallas, TX; Fort Lauderdale, FL; and Citra, FL. Thirty-nine zoysiagrass genotypes and cultivars were planted across all locations, comprising 4 commercial cultivars and 35 experimental breeding genotypes. However, only four breeding genotypes and four commercial cultivars were selected for the thatch study due to their overall excellent performance ‘Meyer’, ‘Emerald’, ‘Zeon’, ‘Empire’, DALZ 1808, DALZ 1311, DALZ 1701, and FAES 1319 in the NTEP trial. The performance of these cultivars and experimental genotypes were evaluated in an NTEP trial for comparison to Meyer zoysiagrass in Olathe, KS (Fry and McFadden, 2022). The performance of these genotypes at multiple sites can also be seen at ntep.org.

The USGA experiment was initiated in June 2021 in Olathe, KS; July 2021 in West Lafayette, IN; and October 2021 in Citra, FL. (Fig. 4.1) Seventy-four genotypes and cultivars were established across the three locations, comprising 5 commercial and 69 experimental breeding genotypes in the trial. Only those that had overall excellent performance were selected for the thatch study, in Olathe, which included 14 genotypes including DALZ 1701, DALZ 1808, 6782-75, 6782-79, 6782-104, 6829-36, 6844-36, 6844-74, 6844-104, 6844-150, 6844-202 and three commercial cultivars included Meyer, Innovation, and Emerald. In Citra, Florida, twelve genotypes were selected for this study including breeding genotypes; 6829-69, 6941-36, 6782-75, 6942-22, 6782-104, 6785-19, 6789-23, 6792-44, 6789-40, DALZ 1701, and cultivars; ‘Zeon’, and ‘Palisades’. In West Lafayette, Indiana, 12 genotypes were evaluated for this study

including, 6844-74, DALZ 1701, 6844-150, 6829-36, 6844-36, 6844-104, 6844-202, DALZ 1808, Palisades, Meyer, Emerald, and Innovation. Evaluation of the USGA zoysiagrass genotypes was done across multiple states and those included in this study had good performance according to USGA standards in the southern U.S. and the transition zone (Chandra et al. 2023). These high performers were provided back to the USGA who sponsored the project, and details can be found in McFadden (2024).

The following provides details for each experimental site, including planting date, soil type, fertilization, mowing practices, and irrigation. These factors, along with the environmental conditions of each location, can influence thatch accumulation over time. Preemergence herbicides were applied at the onset of planting and routinely every year across all sites, with postemergence herbicides commonly used routinely throughout the trial period. In addition, some locations utilized fungicides to control large patch disease. However, details on these were not included as they may have minimal impact on thatch accumulation.

Olathe, Kansas

An NTEP experimental study was initiated at the K-State Olathe Horticulture Research and Extension Center in Olathe, KS (long. 39.48° N, lat. 95.66° W) on 11 July 2019. The soil type was a silty clay loam with a pH of 6.5. Fertilizer of 1 lb. N/1000 ft² from a 16-0-4 (The Andersons Inc., Maumee, OH.) was applied on 1 May 2020, 26 May 2021, and 9 June 2023. In 2022, 1.25 lbs. N/1000 ft² was applied on 1 June 2022 from the same fertilizer. Before the zoysiagrass plugs reached full coverage, the areas were mowed at approximately 1.5 inches with a rotary mower. Full coverage was attained in mid-summer of 2020 and plots were continued to be mowed at 0.75 inches three times weekly with a reel mower. Irrigation was applied at 0.75

inches in weeks when there was less or no rainfall. Plugs (4-inch diameter) to measure thatch from the NTEP trial were harvested on 14 May 2024.

The USGA experimental trial was initiated on 17 June 2021. On 6 July, after the plugs were established, 0.75 lb. N/1000 ft² from a 16-0-4 fertilizer (The Andersons Inc., Maumee, OH.) was applied. Some genotypes gained their full coverage in late August 2021. In 2022, the same source, date, and amount of N was applied as described above in the NTEP trial. Similar to the NTEP trial, 0.75 inches of irrigation was applied on weeks when there was little or no rainfall. Plugs from the USGA trial were also harvested on 14 May 2024.

Stillwater, Oklahoma

The NTEP zoysiagrass field experiment was initiated at the Turfgrass Research Center in Stillwater, OK, (long. 36.123° N, lat. 97.103° W) on 1 July 2019. A soil textural analysis was conducted and showed that the soil is a sandy clay loam with a 7.2 pH and 1.95% OM. A 25-0-10 fertilizer (Lebanon Seaboard Corporation, Lebanon, PA) was applied at 1 lb. N/1000 ft² after plugging and again in August 2019. In 2020, the same fertilizer was applied at 0.5 lb. N/1000 ft² in April, May, Aug, and early September, a 46-0-0 urea fertilizer was applied at the same rate. In 2021, 2022, and 2023, the plots were treated with a total of 4 lbs. N/1000 ft² from the 25-0-10 fertilizer applied in June, July, early August, and early September (a total of 2 lbs. N/1000 ft² applied annually). Throughout the study, mowing was maintained at 1.5 inches height using a reel mower and irrigation was applied accordingly based on the ET and soil moisture to prevent stress and wilting. Plugs were harvested on 13 June 2024 for the thatch study.

West Lafayette, Indiana

Experiments for the NTEP and USGA studies were conducted at the William H. Daniel Turfgrass Research and Diagnostic Center, West Lafayette, IN (40°26'31"N; 86°55'47"W). The soil type was silt loam.

For the NTEP trial, plugs were planted on 11 July 2019. At planting, a 6-24-24 starter fertilizer (The Andersons Inc.) was applied at 0.44 lb. P/1000 ft² and throughout the first experimental year in 2019, urea fertilizer was applied at the rate of 1 lb. N/1000 ft² on 1 August and 1 September. In 2020, at 1 lb. N/1000 ft² from urea fertilizer was applied to plots on 9 June and was reduced to 0.5 lb. N/1000 ft² on 8 and 31 July. In 2021, Shaw's 24-0-22 fertilizer (Knox Fertilizer Company, Knox, IN) was applied at 0.75 lb. N/1000 ft² on 21 May and the same fertilizer was applied at 1 lb. N/1000 ft² on 17 August. In 2021, the same Shaw 24-0-22 fertilizer was applied at 0.75 lb. N/1000 ft² on 24 May. A 28-0-10 surfcote Shaw fertilizer (Knox Fertilizer Company, Knox, IN.) was applied at 0.75 lb. N/1000 ft² on 30 May and 1 September 2022, and on 30 May and 1 August 2023. Throughout the experimental study, mowing was kept at a 3-inch bench height and irrigation was applied as needed to prevent wilting.

For the USGA experiment, genotypes were plugged on 21 July 2021. A. On the same day, a 6-24-24 starter fertilizer (The Andersons Inc., Maumee, OH.) was applied at 0.44 lb. P/1000 ft², and on 17 August, a 24-0-22 Shaw fertilizer (Knox Fertilizer Company) was applied at 1 lb. N/1000 ft². In 2022, Shaw fertilizer 24-0-22 was applied once at 0.75 lb. N/1000 ft² on 24 May. In 2023, 28-0-18 Surfcote Shaw fertilizer was applied at 0.75 lb. N/1000 ft² on 30 May and 1 September. In 2024, prior to thatch plug harvest, a single application of 0.75 lb. N/1000 ft² from the same N source last mentioned was done. Mowing was maintained at a 1.75-inch bench height during grow-in and was later reduced to 0.75 inches after full coverage in the summer of

2022. Irrigation was applied as needed to prevent wilting. Plugs were removed to evaluate thatch on 18 June for NTEP and 5 July for the USGA.

Dallas, Texas

The NTEP zoysiagrass experiment was planted in the Dallas Station of Texas A&M University on 19 July 2019. A 25-5-10 fertilizer (Harells, LLC, Lakeland, FL.) was applied at 1.5 lb. N/1000 ft² the same day. Throughout the field trial in 2019, mowing height was maintained at 1.5 inches starting from 16 August; mowing frequency was once a week. In 2020, fertilizer was applied at 0.5 lb. N/1000 ft² on 18 April and 4 June. A 42-0-0 (Harells, LLC) fertilizer was applied at 1.0 lb. N/1000 ft² on 16 September 2020. In 2021, fertilizer was applied at 1 lb. N/1000 ft² on 27 April and was later reduced to 0.5 lb. N/1000 ft² on 16 June, fertilizer was however raised back to 1 lb. N/1000 ft² on 21 September. In 2022, urea fertilizer at 1 lb. N/1000 ft² was applied on 19 May and 22 September. In 2023, fertilizer at 0.75 lb. N/1000 ft² was applied on 18 April. In 2020, mowing began on 25 March with a John Deere reel mower. Mowing height was maintained at 1.5 inches until June 2020 after which mowing height was reduced to 1.25 inches and 1 inch by July until the end of 2020. In 2020, the plots were subjected to drought twice (June and July) followed by a 2-inch heavy rain. In 2021, mowing was continued at a 1-inch height from March to December with a reel mower. Dry down was initiated on 9 July and recovery was initiated by scheduling the irrigation system for 20 minutes every day for a couple of days, manual irrigation was done for an hour on the fifth day. In 2022, irrigation was done in January and February for 20 minutes each and for 30 minutes on 18 May and 7 June after which dry down was initiated on 13 June for a recovery on 12 July. On 29 July, another dry down was initiated and interrupted by a 1.2-inch rainfall on 10 August and a 4-inch

rainfall from 21 August to 23 August. During this period, mowing height was maintained at 1 inch until May 2024. Plugs were harvested to evaluate thatch on 15 May.

Fort Lauderdale, Florida

An NTEP zoysiagrass experiment was initiated on 4 November 2019 in Ft. Lauderdale. Unlike other locations, zoysiagrass plugs were planted on a Hallandale fine sand (siliceous, isohyperthermic Lithic Psammaquents) soil type with a pH of 7.5, soil fertility measures 199 lb. P acre⁻¹ and 35 lb. K acre⁻¹. From the beginning of the study in 2019 to the time of harvest in Spring 2024, a total of 4 lb. N/1000 ft² was applied each year. To prevent drought, 75% of ET through irrigation was applied. Irrigation was withheld when precipitation occurred throughout the study period. Mowing was maintained at 0.58 inches three times a week during the summer period and once or twice a week during the winter period with a reel mower (Toro Company). Plugs were harvested on 16 January 2024 to evaluate thatch.

Citra, Florida

An NTEP experiment was initiated in Citra Florida in late June 2019. A 13-4-13 granular fertilizer was applied in March and May 2020 and 2021 at 1.2 lb. N/1000 ft² and 0.9 lb. N/1000 ft², respectively. Following those treatments, another fertilizer application was continued each year with a 15-0-15 granular fertilizer at 0.9 lb. N/1000 ft² in July. In 2022, 1 lb. N/1000 ft² of a 15-0-15 granular fertilizer was applied in late March and was later reduced to 0.9 lb. N/1000 ft² in mid-August and early October. The same application procedure was followed in 2023, starting with 1 lb. N/1000 ft² of a 15-0-15 granular fertilizer in late April and was continued with a 0.9 lb. N/1000 ft² 15-0-15 granular fertilizer in late May, mid-July, and late September.

Mowing was done once weekly at 2.5 inches mowing height (Fig. 4.1). Irrigation was only applied when drought stress was beginning. However, the harvest of plugs for the thatch study was conducted on 20 February 2024.

The USGA zoysiagrass experiment was initiated in 2021. Fertilizer was first applied in late October using a 15-0-15 granular fertilizer at 1 lb. N/ 1000 ft² and in 2022, the same amount was applied in late March. The same fertilizer was applied at 0.9 lb. N/1000 ft² in mid-August and early October. In 2023, a similar procedure to 2022 was followed with 1 lb. N/ 1000 ft² of 15-0-15 applied in late April and 0.9 lb. N/1000 ft² was applied in late May, mid-July, and late September. Mowing was done with a reel mower at 0.75 in. two or three times weekly. Irrigation was applied only to prevent drought stress. However, the harvest of plugs for the thatch study was done on 2 April 2024.

Data Collection and Measurement

For the thatch accumulation measurements, the soil and roots directly beneath the thatch layer were removed and discarded. Thatch depth was measured using a digital vernier caliper. This method is widely accepted for accurate thatch measurement (Shaddox and Unruh, 2019; Smith 1979; Menchyk et al., 2014; Mu and Carroll, 2013). Thatch depth was measured from the base of tillers (crown) to the base of organic matter that was visible (Fig. 4.2c). Thatch depth measurements were taken at three random angles around the core of the 4-inch diameter plug, and the results were averaged. Tillers were counted on each 4-inch diameter plug before all green leaf tissues were cut using handheld scissors (Shaddox and Unruh, 2019). The soil and roots below the thatch layer were also cut off and washed leaving a bare core thatch which was further oven-dried at 70°C for 72h. After 72h, thatch mass was weighed and recorded using a weighing balance (Ohaus Explorer Analytical EX224 balance, Parsippany, NJ.)

All measurements below were collected once on the day of thatch evaluation, just prior to plugs being collected at each site. Turfgrass quality was visually rated on a 1 to 9 (1 = poor quality, 6 = minimum acceptable quality, and 9 = overall best quality) scale. Data was also collected on the Normalized Difference Vegetation Index (NDVI) of each plot in each location on a 0 to 1 scale to measure the plant health, and vegetative cover (Fig. 4a). The leaf width was measured using a digital vernier caliper, three strands of leaves were measured by laying the leaf flat on a surface and measuring the west to east at the same hour plugs were removed to prevent wilting and inaccurate measurement. Three measurements of leaf width were made on each core and the results were averaged. Other data that were collected on site were turfgrass color, which was visually rated on a 1 to 9 scale (1 = completely brown; 6 = minimum acceptable green color, and 9 = optimum dark green color), turfgrass density also visually rated on a 1 to 9 scale (1 = very thin turfgrass, 6 = minimum acceptable density, and 9 = very dense turfgrass), turfgrass texture on a scale of 1 to 9 (1 = very coarse textured, 6 = minimally coarse-textured, and 9 = very fine-textured). Data was also collected on turfgrass firmness using a Clegg hammer which measures the deceleration impact of a free-falling hammer, providing a quantified assessment of surface hardness or firmness, which directly influences playability (O'Brien, 2019) (Fig. 4.2b). Clegg Hammer measurement in surface units is “gravities”; higher levels of gravities indicate harder turfgrass surface. Finally, data was collected on the number of tillers of each core by counting the number of lateral shoots that grow from the base of the main stem in each 4-inch diameter plug.

Data Analysis

All data for each of the ratings were subjected to ANOVA in SAS v. 9.4 (SAS Institute Inc., Cary, NC). A mixed model was used to analyze the effects of cultivars, locations, and

replicates on average thatch depth and thatch mass. The MIXED procedure was used for thatch depth and thatch mass with cultivars and locations as fixed effects, allowing for the estimation of their main effects and their interaction. Replicates nested within the location were specified as a random effect to account for variability due to differences among replicates at each location. GLIMMIX procedure was used to examine genotype performance within each location, enhancing our understanding of location-specific effects on thatch traits. Correlation procedure was conducted by location to explore relationships among key turfgrass traits such as thatch depth, turfgrass color, turfgrass density, and NDVI. Least square means were calculated for cultivars, locations and their interactions, with Tukey's adjustment applied for pairwise comparisons among cultivars, locations, and their interactions at $P \leq 0.05$.

Results and Discussion

Thatch Depth across Genotypes and Locations

NTEP

In the NTEP trial, there was a significant cultivation x location interaction, indicating that cultivars performed differently across locations (Fig. 4.3). Only one site, Dallas, showed statistical differences in thatch depth among genotypes. Emerald had the largest thatch depth. Meyer, Empire, and DALZ 1808 had the smallest.

Although there were no statistical differences in thatch depth across cultivars at the other locations, the range of thatch depth at other locations for the NTEP study was as follows: Olathe, KS, Empire (0.59 in.) to DALZ 1311 (0.81 in.); Stillwater, Oklahoma, DALZ 1311 (0.13 in) to DALZ 1808 (0.36 in); West Lafayette, Indiana, DALZ 1311 (0.71 in) to DALZ 1701 and Emerald (1.08 in.); Fort Lauderdale, FL, DALZ 1311 (0.29 in) to Emerald and DALZ 1701 (0.52 in); and Citra, Florida, Emerald (0.54 in) to DALZ 1808 (1.21 in). A higher range of thatch

depth is visible in West Lafayette, IN, and Citra, FL where mowing heights are higher than other sites. This is similar to what (McFadden, 2024) observed when Innovation mowed at a higher depth of 1.5 inches had a significantly higher thatch depth of 0.70 inches compared to innovation mowed at 0.75 inches, indicating that mowing height and genotype x mowing height had more influence on thatch depth. Although (McFadden, 2024) had reported a smaller thatch depth for DALZ 1808 when mowed at 0.75 and 1.5 inches

A study conducted by Kauffman et al. (2013) on thatch-mat depth showed that ‘TifEagle’ and ‘Champion’ bermudagrass accumulated thatch-mat deeper in the soil profile (1.06 in and 1.09 in, respectively) than ‘Diamond’ zoysiagrass and ‘SeaDwarf’ seashore paspalum due to their dense, aggressive growth habits. Kentucky bluegrass was also shown to have differences among genotypes in thatch accumulation in Nebraska as mowing height increased from 0.9 inches to 1.9 inches (Shearman et al., 1980). For example, S-21 Kentucky bluegrass had less thatch (0.19 in.) than Cheri and Glade (0.54 and 0.58 in., respectively). The differences in thatch accumulation across cultivars can also be traced to their genetic makeup as studies have shown that denser, aggressive growing genotypes can accumulate more thatch. Studies have also shown that *Z. matrella* and its hybrids have a higher tendency of thatch accumulation than *Z. japonica* (Loch, 2013).

USGA

In the USGA zoysiagrass genotype trial, there were no differences among genotypes in Kansas ($P = 0.08$) and Indiana ($P = 0.40$). However, there were differences among genotypes in Citra, Florida ($P = 0.002$). DALZ 1701 (0.52 in.) had the greatest thatch depth and genotypes like 6782-104 and 6785-19 had the smallest thatch depth (0.15 in.) (Fig. 4.4). Ranges in thatch depth across locations included: Olathe, KS, DALZ 1808 (0.48 in.) to DALZ 1701 (0.73 in.);

West Lafayette, IN, 6844-150 (0.64 in.) to DALZ 1701 (0.82 in.). The thatch depth levels among cultivars in Kansas and Indiana suggest that environmental factors may play a stronger role in thatch accumulation in these regions compared to cultivar genetics. Factors such as soil type, climate, and management practices might overshadow cultivar-specific performance in these locations. In a study by Kauffman et al. (2013), thatch-mat depth could be linked to a reduction in water infiltration and oxygen content and the impact on plant traits is not well established, but could be a result of many environmental, soil characteristics and/or management practices (Carrow, 2000; Horst et al., 1996; Taylor and Blake, 1982; Harris, 1978; Murray and Juska, 1977; Thompson, 1967).

Thatch Mass across Genotypes and Locations

NTEP

Differences in thatch mass were only observed in Olathe, KS ($P = 0.015$) (Fig. 4.5). In Olathe, DALZ 1311 (0.0072 oz.) had the highest mass compared to DALZ 1701 (0.0045 oz.) and DALZ 1808 (0.0049 oz.). More likely, variability between reps might have caused the differences among locations. However environmental factors were not measured.

USGA

Differences among genotypes in thatch mass were observed in Kansas ($P = 0.037$) and Indiana ($P = 0.010$), but not in Florida (Fig. 4.6). In Kansas, Emerald had the highest thatch mass (0.0061 oz.), which was significantly greater than two other cultivars, (6782-75 and 6782-75). In Indiana, Palisades had the highest thatch mass (0.0059 oz.) and was significantly higher than 6844-202 (0.0034 oz.) and DALZ 1701. In Florida, there were no differences, but the means ranged from 6942-22 (0.0023 oz.) to Palisades (0.0055 oz.).

While Emerald and Palisades showed larger thatch mass in Kansas and Indiana, respectively, other cultivars like 6942-22 in Florida and 6844-202 in Indiana consistently showed lower thatch mass production. The overlap of significant groups for many cultivars indicates that while genetic differences exist, they may not practically be significant for turfgrass management unless combined with specific environmental conditions.

Correlation Between Thatch depth and Morphological Traits

For the NTEP and USGA trials correlations were done at specific sites. This was done because genotypes evaluated on the USGA trials were not the same across all sites.

NTEP

Correlations among thatch depth, thatch mass, and turfgrass morphological traits are indicated in Table 4.1. Most correlations were not significant (Table 4.1). Significant correlations are presented in figures and described below.

In Dallas, thatch depth and firmness were negatively correlated (Fig. 4.7). A negative correlation between thatch depth and firmness implies that as thatch accumulates, turfgrass firmness decreases. This accumulation can restrict water movement, leading to moisture retention near the surface, thereby reducing soil compaction and increasing turf softness. This softer surface affects turfgrass firmness and playability, especially in high-traffic areas like golf courses. This is similar to what Beard (1973) and Carrow (2001) observed, that thick thatch layers can act as a buffer, decreasing surface hardness by trapping moisture and organic material at the soil-turf interface. However, Stier et al. (2000) found that light thatch (around 0.197 in.) could support firmness in certain sandy soil conditions. Interestingly, this correlation between thatch depth and firmness was not significant in the other locations.

In Dallas and Citra, turfgrass NDVI was positively correlated with thatch depth (Fig. 4.8). NDVI is a spectral reflectance measure that indicates vegetation vigor, with higher values often reflecting denser, healthier plant coverage. Some genotypes with high density and good quality can have high NDVI values and also accumulate a higher level of thatch depth. Bremer et al. (2011) have shown NDVI to be responsive to biomass accumulation in turfgrass, indicating that more vigorous or dense vegetation tends to yield higher NDVI values. Beard and Rieke (1976) demonstrated that turfgrass with higher shoot density often had higher NDVI values, as denser foliage can elevate NDVI by enhancing the reflectance of healthy vegetation. This could also imply that thicker thatch depth, which might be associated with better moisture retention, especially at the surface area may indirectly boost NDVI. This positive correlation may have resulted from cultivars with high shoot quality, such as Zeon and Emerald, that were included in this study. As time progresses, these high-performing cultivars could accumulate excessive thatch that would require management practices to reduce the amount of thatch.

Tiller numbers may also be associated with thatch accumulation. In Stillwater (Fig. 4.9a), Dallas (Fig. 4.9b), and Fort Lauderdale (Fig. 4.9c), thatch depth increased with an increase in tiller numbers among genotypes. Some genotypes, such as cultivars developed from *Z. matrella*, have high density and tiller numbers. When turfgrass is actively tillering, more biomass is produced, contributing to thatch accumulation, especially if management practices do not frequently remove or break down the organic matter. During this NTEP trial, no cultivation or thatch removal was done. Trenholm et al. (1999) found that dense, tiller-rich turfgrass stands often correlate with increased thatch, as the turfgrass maintains a self-sustaining layer of organic matter, which is quite common in areas that are well-fertilized and irrigated. Huang and Liu (2009) also found that tillering is positively associated with shoot density and biomass

production, which directly contributes to thatch in species like Kentucky bluegrass (*Poa pratensis* L.) and creeping bentgrass (*Agrostis stolonifera* L.).

USGA

Correlations for thatch depth, thatch mass, and turfgrass traits are listed in Table 4.2. Most relationships had no significant correlation. Relationships with significant correlations are presented in figures and described below.

In Kansas, there was a positive correlation between thatch depth and thatch mass (Fig. 4.10). This positive correlation between thatch depth and thatch mass may indicate that as the thickness of the thatch layer increases, so does the total mass of organic material within it; however, this was not consistently observed across the three sites. Beard (1973) reported that as the depth of thatch increases, it correlates with increased organic mass due to ongoing deposition of plant material, but thatch mass was not measured. Turgeon (2002) further explains thatch accumulation in perennial turfgrass systems, noting that mass increases with depth due to the combined effects of organic matter deposition. However, since the environmental conditions in these locations were not considered, we cannot verify if environmental factors could have influenced this result in Kansas.

In Indiana, thicker thatch depth correlated negatively to NDVI (Fig. 4.11), which was the opposite of what occurred between thatch depth and NDVI in Dallas and Citra in the NTEP trial. Studies have shown that excessive thatch buildup limits light, water, and nutrient movement to the root zone (McCarty, 2005). This restriction of light penetration can lead to reduced photosynthetic activity in turfgrass which is often reflected as lower NDVI values. Beard (1973) reported that heavy thatch can result in a thinner, weaker stand of turfgrass due to reduced access to essential resources, which supports the observed negative correlation between thatch depth

and NDVI. However, zoysiagrass in West Lafayette can be significantly impacted by freezing temperatures. Studies have shown that Emerald and Empire exhibit more winter injury in Indiana and are not considered suitable for transition zones compared to other cultivars (Patton and Reicher, 2007; Dunn and Diesburgk 2004; Morris, 2001), and cultivars that are fine in texture may have low NDVI levels. Although we did not take winter temperature or any environmental conditions into consideration, we are not certain if winter injury influenced the negative correlation.

In Florida, thatch depth was negatively correlated with turfgrass firmness (Fig. 4.12). This was comparable to the results shown in Dallas with the NTEP trial. In addition, in Florida, an increase in leaf width correlated with higher levels of thatch depth (Fig. 4.13). The same was shown in Olathe (Table 4.1), but the opposite occurred in Dallas. This may indicate that as the thatch gets deeper, the leaf becomes broader which may have been influenced by the retention of nutrients and moisture near the soil surface, which is then accessible to the turfgrass.

Conclusion

In the NTEP trial, thatch depth differences were observed among genotypes only in Texas, with Emerald showing the greatest thatch depth compared to Meyer and DALZ 1808. Higher thatch depths across all genotypes were observed in West Lafayette and Citra, possibly due to higher mowing heights. In the USGA trial, genotype differences in thatch depth were noted in Citra, Florida, but not in Kansas and Indiana. Notably, correlations between thatch depth and morphological traits were site-specific, such as the negative correlation between thatch depth and firmness in Texas and Citra, Florida. In addition, there were varying correlations between thatch depth and NDVI, which was positive in Dallas and Citra in the NTEP trial, but negative in Indiana in the USGA trial, probably due to genotype-specific factors at locations. In summary,

zoysiagrass thatch accumulation was only significant among NTEP and USGA genotypes at a few sites, but significant correlations between thatch depth vs. multiple morphological traits ratings and measurements across numerous sites occurred, which provides more data and information on understanding thatch production among zoysiagrass genotypes. Future research should investigate the causation between thatch depth and turfgrass morphological traits.

Table 4.1 Statistical results of correlation coefficients between NTEP thatch depth (TD) and thatch mass (TM) vs. morphological traits measured on the day thatch was collected.

Variable	Olathe, Kansas	Stillwater, Oklahoma	West Lafayette, Indiana	Dallas, Texas	Ft. Lauderdale, Florida	Citra, Florida
TD vs Turfgrass Quality	NS	NS	NS	* (0.427) ¹	NS	NS
TD vs Turfgrass Color	NS	NS	NS	NS	NS	NS
TD vs Turfgrass Density	NS	NS	NS	** (0.55)	NS	NS
TD vs Firmness	NS	NS	NS	*** (-0.906)	NS	NS
TD vs NDVI	NS	NS	NS	* (0.458)	NS	* (0.542)
TD vs Leaf Width	* (0.435)	NS	NS	** (-0.635)	NS	NS
TD vs Tillers	NS	* (0.425)	NS	* (0.504)	** (0.536)	NS
TD vs TM	NS	* (-0.406)	NS	NS	NS	NS
TD vs Texture	NS	NS	NS	* (0.5)	NS	* (-0.509)
TM vs Firmness	NS	NS	NS	NS	NS	* (-0.446)
TM vs NDVI	** (-0.601)	NS	NS	NS	NS	NS

*, **, *** significant at $P \leq 0.05$, 0.01 and 0.001, respectively.

NS, not significant at $P \leq 0.05$

¹Correlation coefficient values when significant.

Table 4.2 Statistical results of correlation coefficients between USGA thatch depth (TD) and thatch mass (TM) vs. morphological traits measured on the day thatch was collected.

Variable	Olathe, Kansas	West Lafayette, Indiana	Citra, Florida
TD vs Turfgrass Density	NS	NS	NS
TD vs Firmness	NS	NS	*** (-0.599) ¹
TD vs NDVI	NS	** (-0.451)	NS
TD vs Leaf Width	NS	NS	*** (0.676)
TD vs Texture	NS	NS	*** (-0.644)
TD vs Tillers	NS	NS	* (-0.402)
TD vs TM	*** (0.566)	NS	NS
TD vs Turfgrass Color	* (0.353)	NS	NS
TD vs Turfgrass Quality	* (0.338)	NS	NS
TM vs Turfgrass Color	NS	NS	** (0.477)
TM vs Firmness	NS	* (0.387)	NS
TM vs Leaf Width	NS	** (0.470)	NS
TM vs Texture	NS	* (-0.370)	NS
TM vs Tillers	NS	* (-0.394)	NS
TM vs Turfgrass Quality	NS	** (-0.454)	NS
TM vs NDVI	NS	NS	* (0.362)
TM vs Turfgrass Color	NS	NS	* (0.477)

*, **, *** significant at $P \leq 0.05$, 0.01 and 0.001 respectively.

NS, not significant at $P \leq 0.05$

¹Correlation coefficient values when significant.



Figure 4.1 Zoysia genotypes in Citra, FL mowed at 2.5 inches.



Figure 4.2 (A) NDVI data collection using handheld NDVI device; (B) data collection on firmness using Clegg Hammer on zoysiagrass genotypes in Fort Lauderdale, FL; and (C) measurement of thatch depth with vernier caliper.

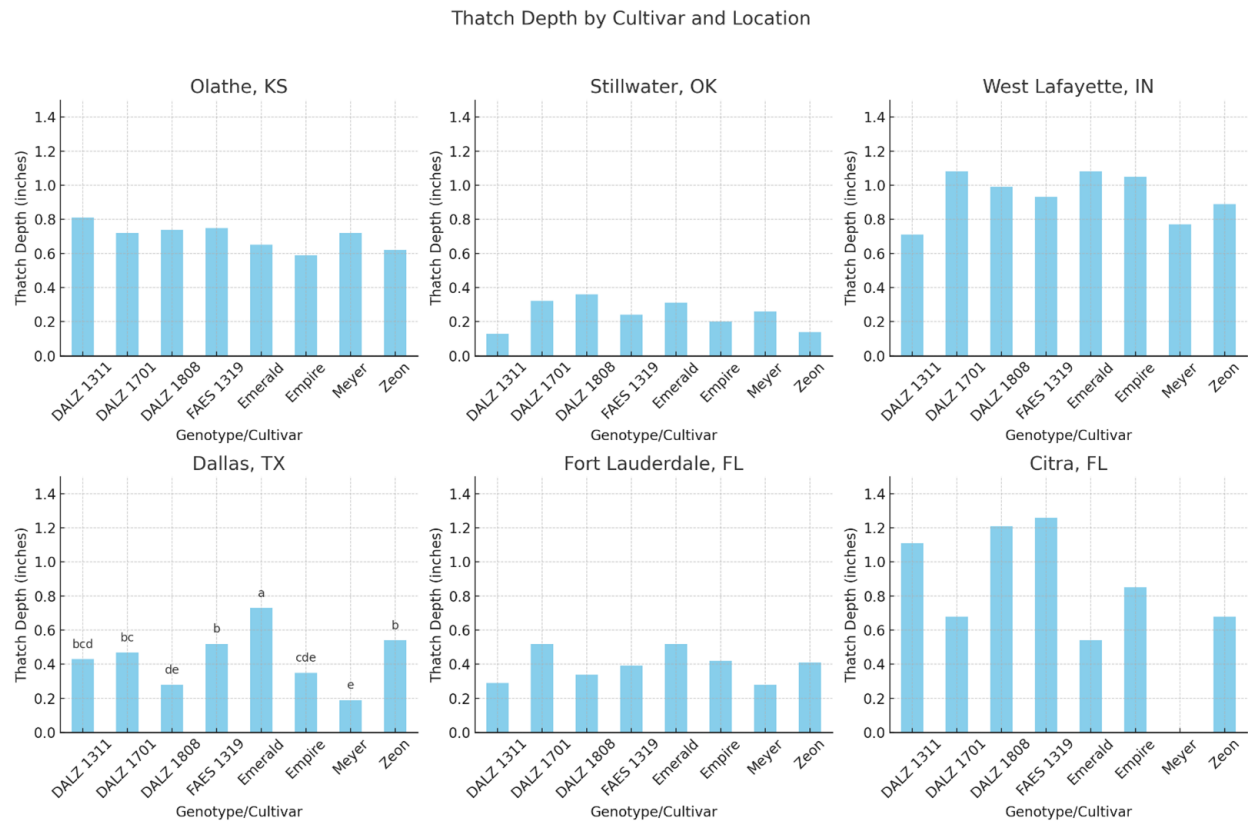


Figure 4.3 Thatch depths among zoysiagrass genotypes in NTEP trial in Olathe, KS, Stillwater, OK, West Lafayette, IN, Dallas, TX, Fort Lauderdale, FL, and Citra, FL. Statistical differences among genotypes in Dallas, TX ($P < 0.05$).

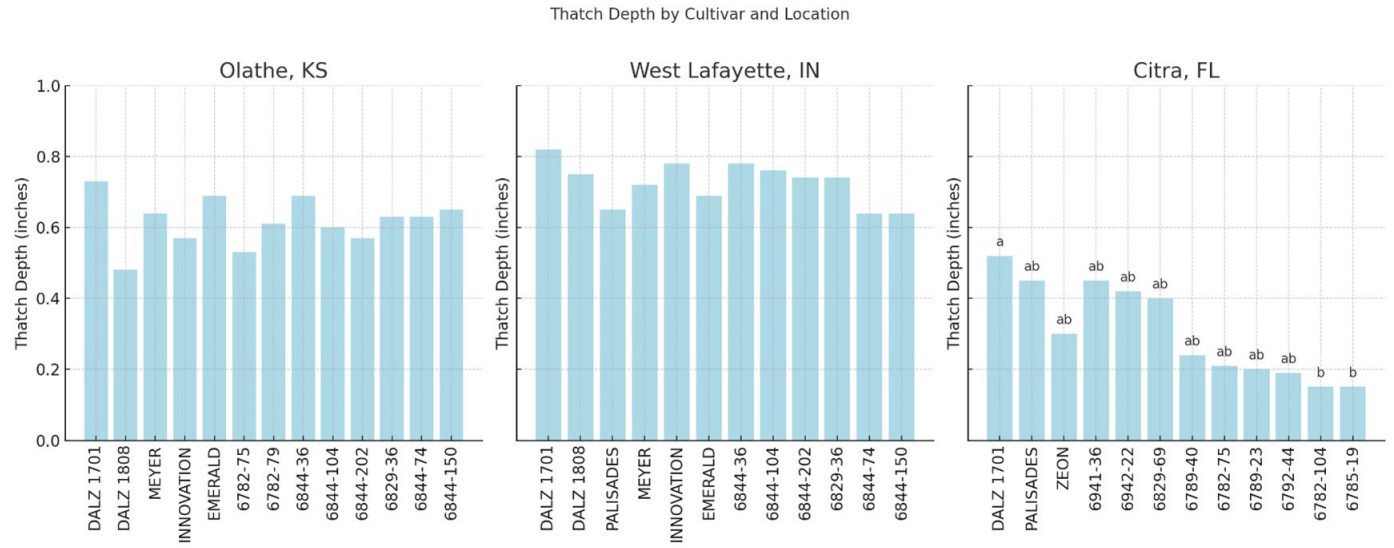


Figure 4.4 Thatch depths among zoysiagrass genotypes in USGA trials in Olathe, KS, West Lafayette, IN, and Citra, FL. Statistical differences among genotypes occurred in Citra, FL only ($P < 0.05$).

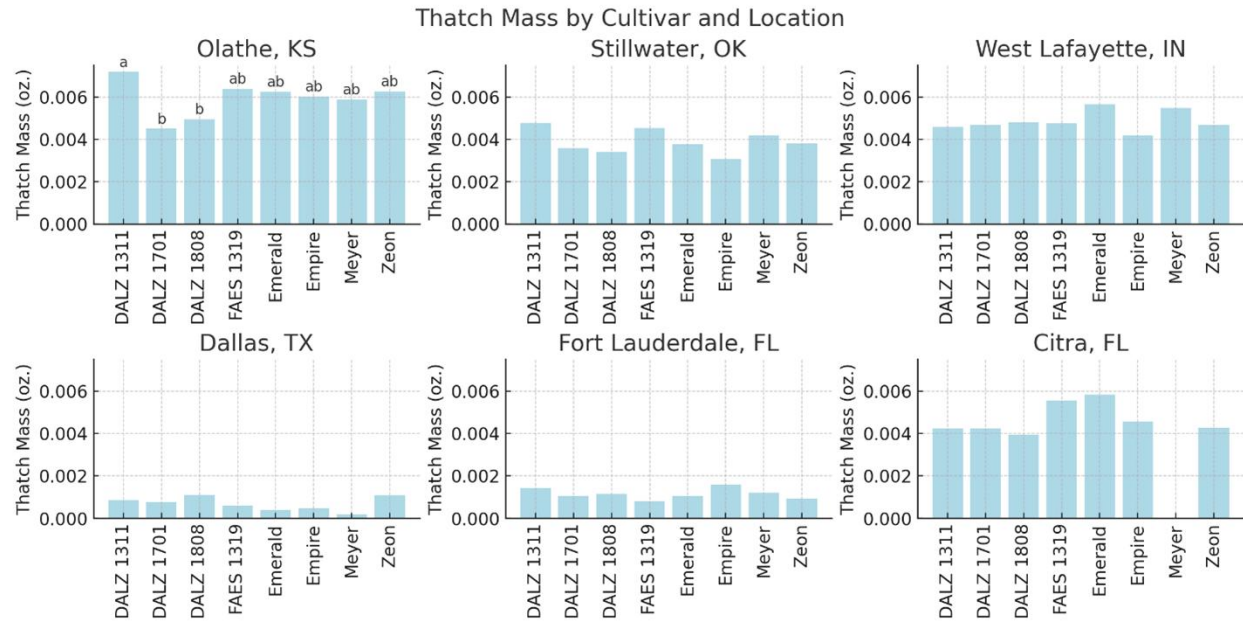


Figure 4.5 Thatch mass among zoysiagrass genotypes in the NTEP trial in Olathe, KS., Stillwater, OK., West Lafayette, IN., Dallas, TX., Fort Lauderdale, FL., and Citra, FL. Statistical differences among genotypes occurred only in Olathe, KS. ($P < 0.05$).

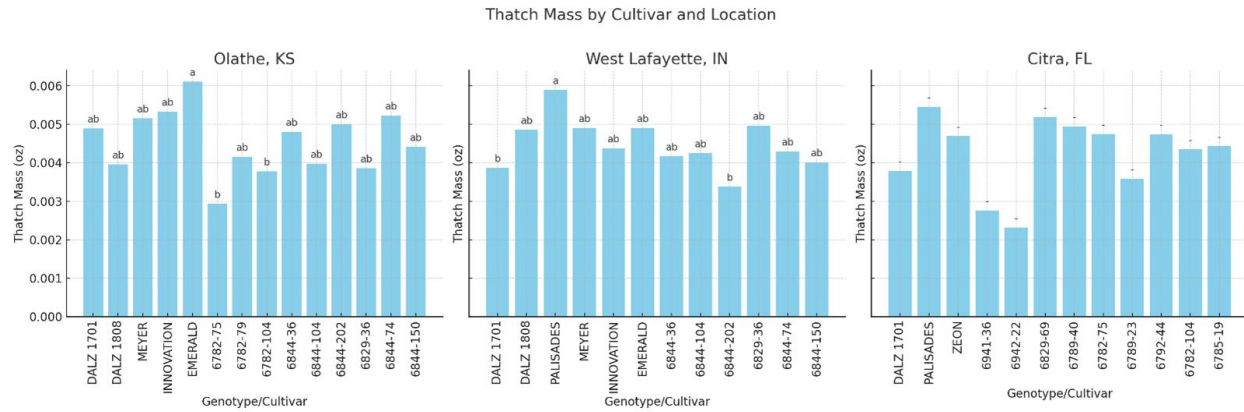


Figure 4.6 Thatch mass among zoysiagrass genotypes in the USGA trial in Olathe, KS., West Lafayette, IN., and Citra, FL. Statistical differences among genotypes only in Olathe, KS, and West Lafayette, IN ($P < 0.05$).

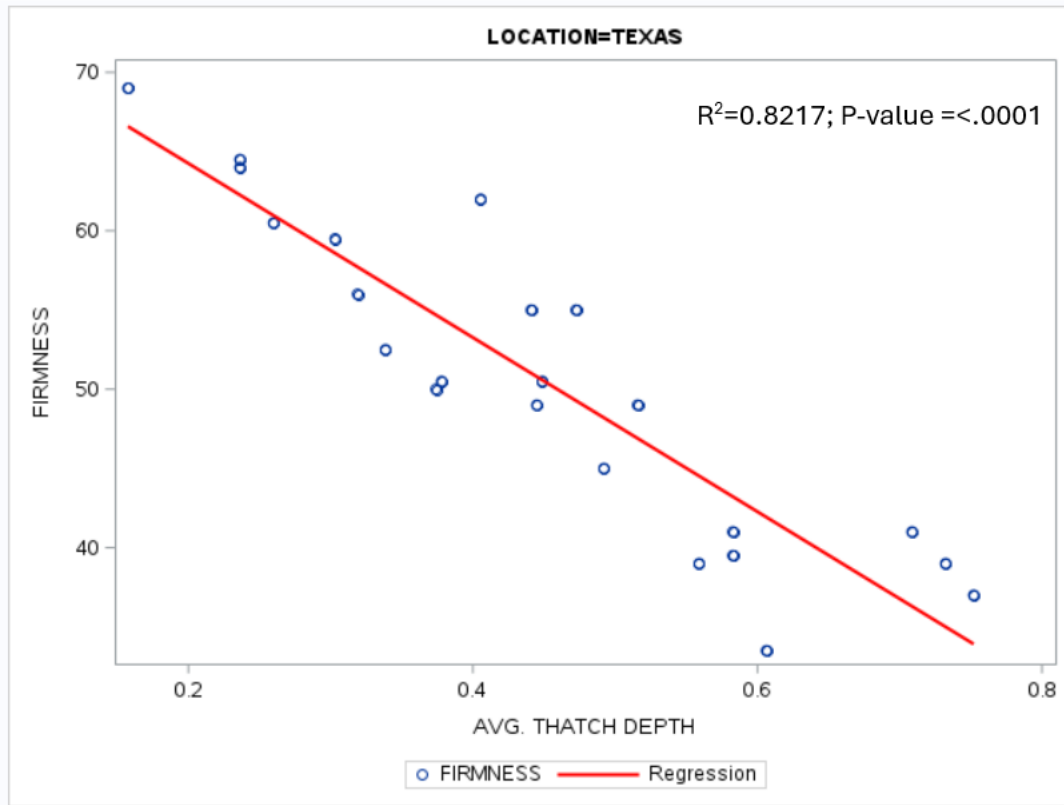


Figure 4.7 Correlation between thatch depth (inches) and firmness (Clegg Hammer measurement in surface units “gravities”), which measures the surface hardness (lower values softer firmness; higher values harder firmness) in the NTEP trial in Dallas, TX ($P < 0.001$). Firmness was measured once in May 2024 across 24 plots.

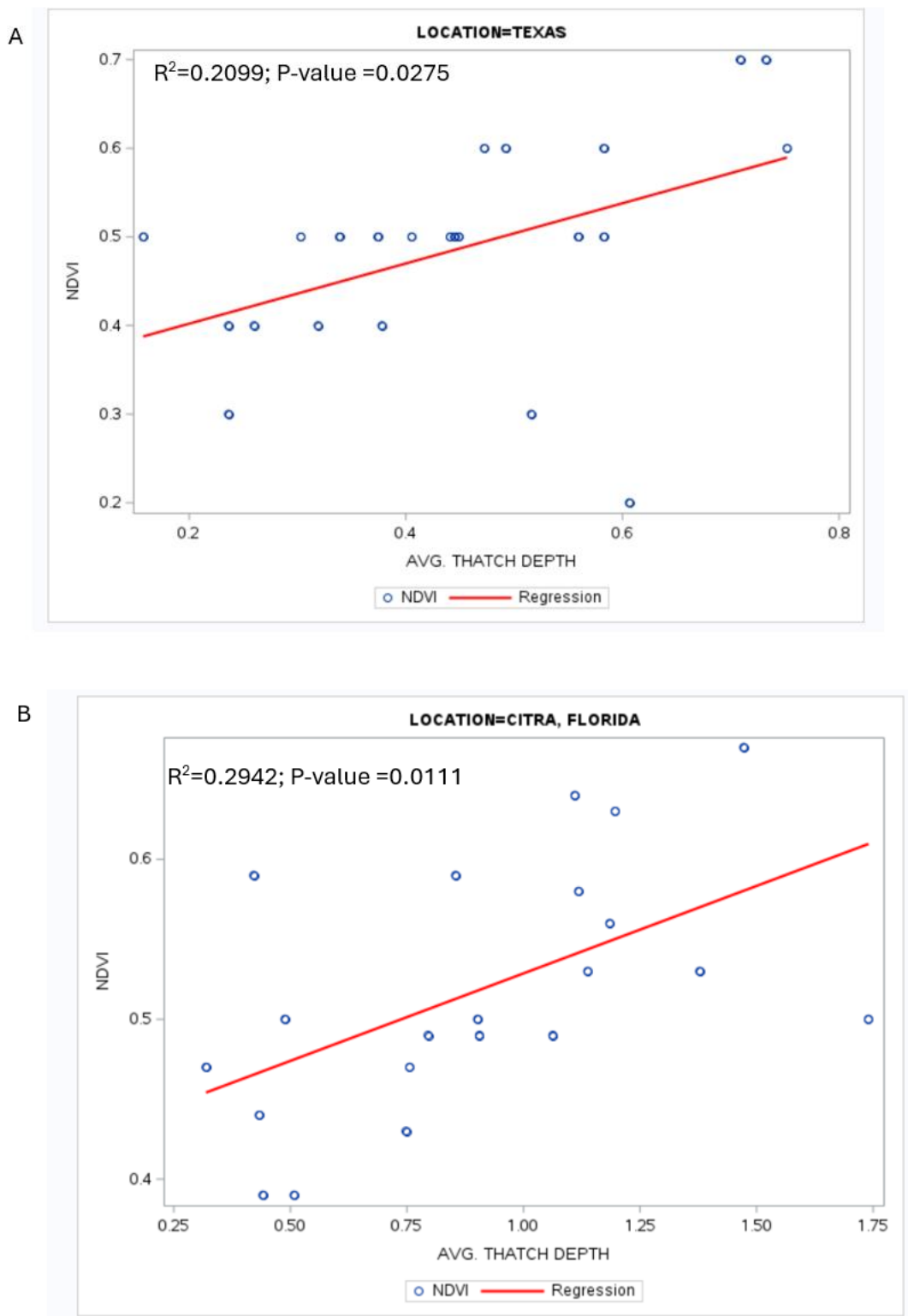
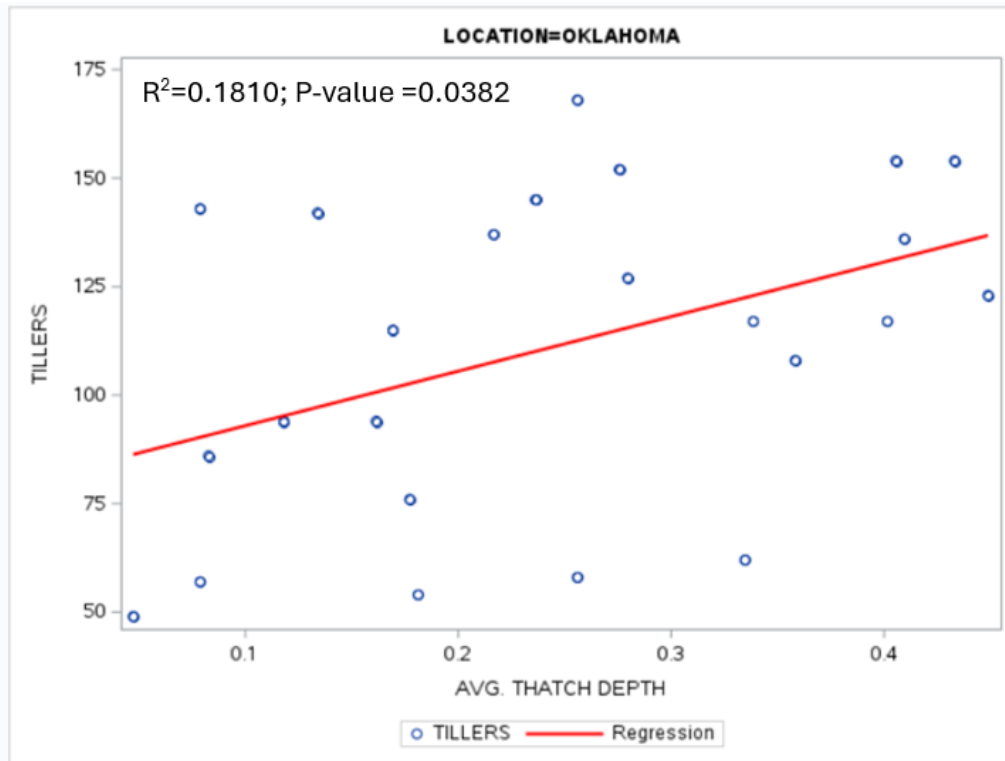
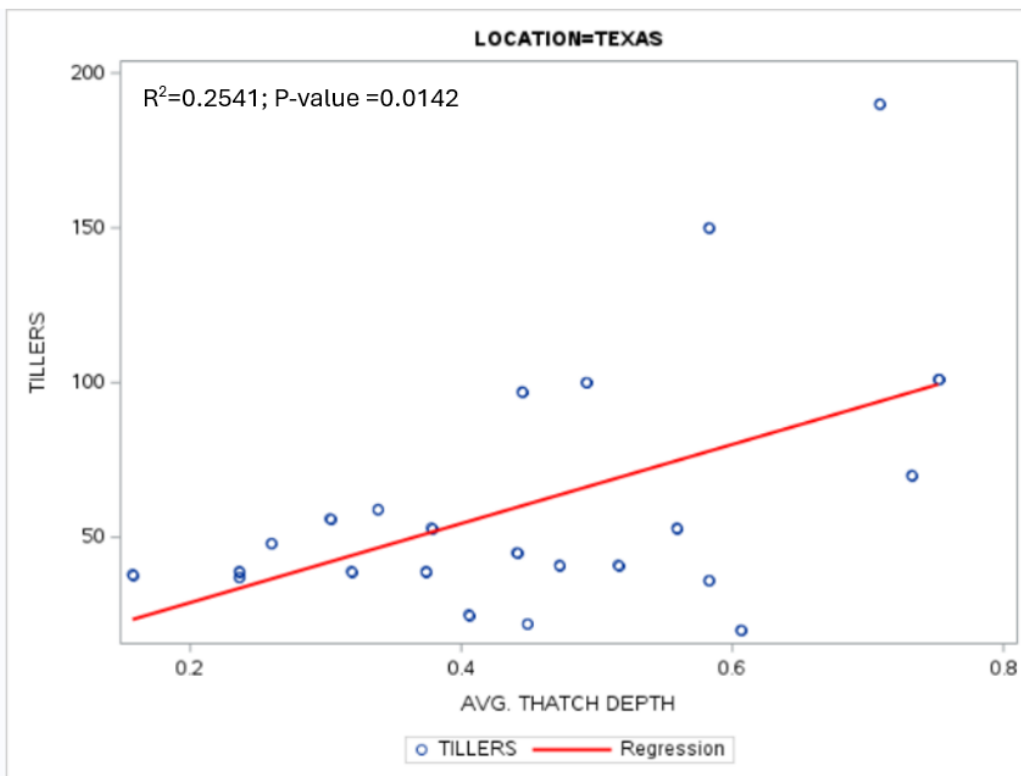


Figure 4.8 Correlation between thatch depth (inches) and NDVI in the NTEP trial in (A) Dallas, TX; and (B) Citra, FL ($P < 0.05$). Thatch depth was measured in May 2024 in Dallas and February 2024 in Citra. Twenty-four plots were measured at each location.

A



B



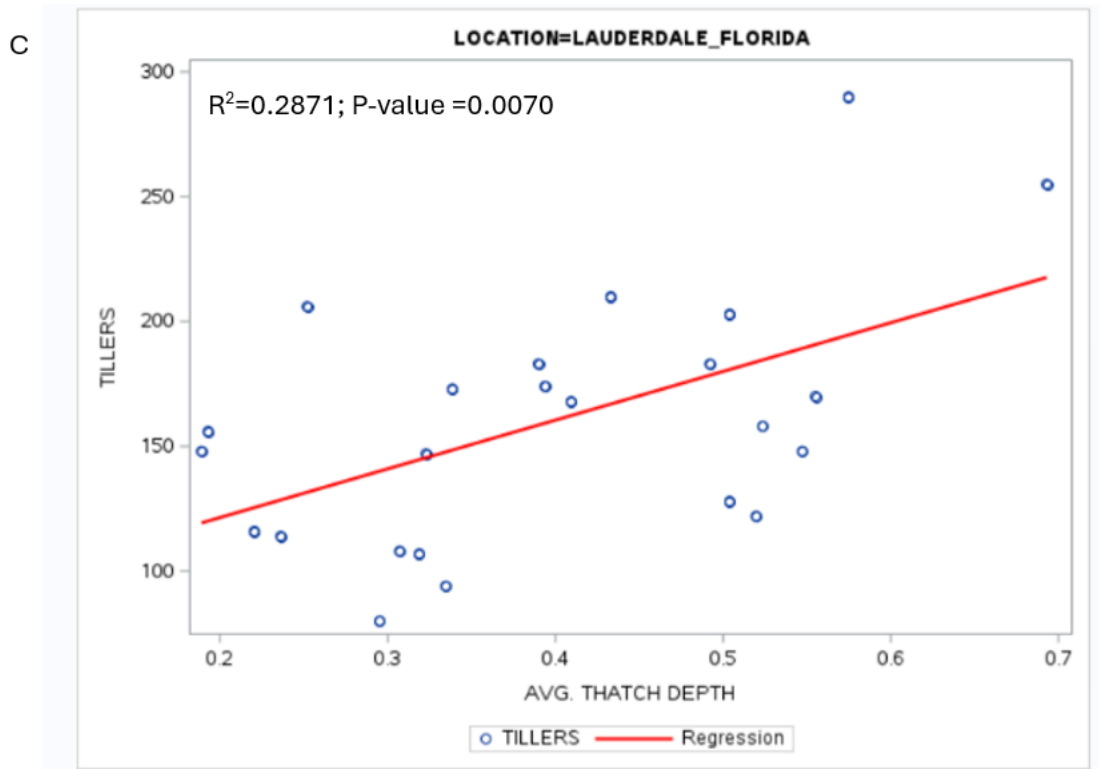


Figure 4.9 Correlation between thatch depth (inches) and tiller number in the NTEP trial in (A) Stillwater, OK ($P < 0.05$).; (B) Dallas, TX ($P < 0.05$); and (C) Ft. Lauderdale, FL ($P < 0.01$). Tillers were measured by counting the number of shoots that emerged from the base of the turfgrass on each 4-inch diameter plug. Tiller numbers were counted from plugs sampled in 24 plots in June (OK), May (TX), and February (FL), 2024.

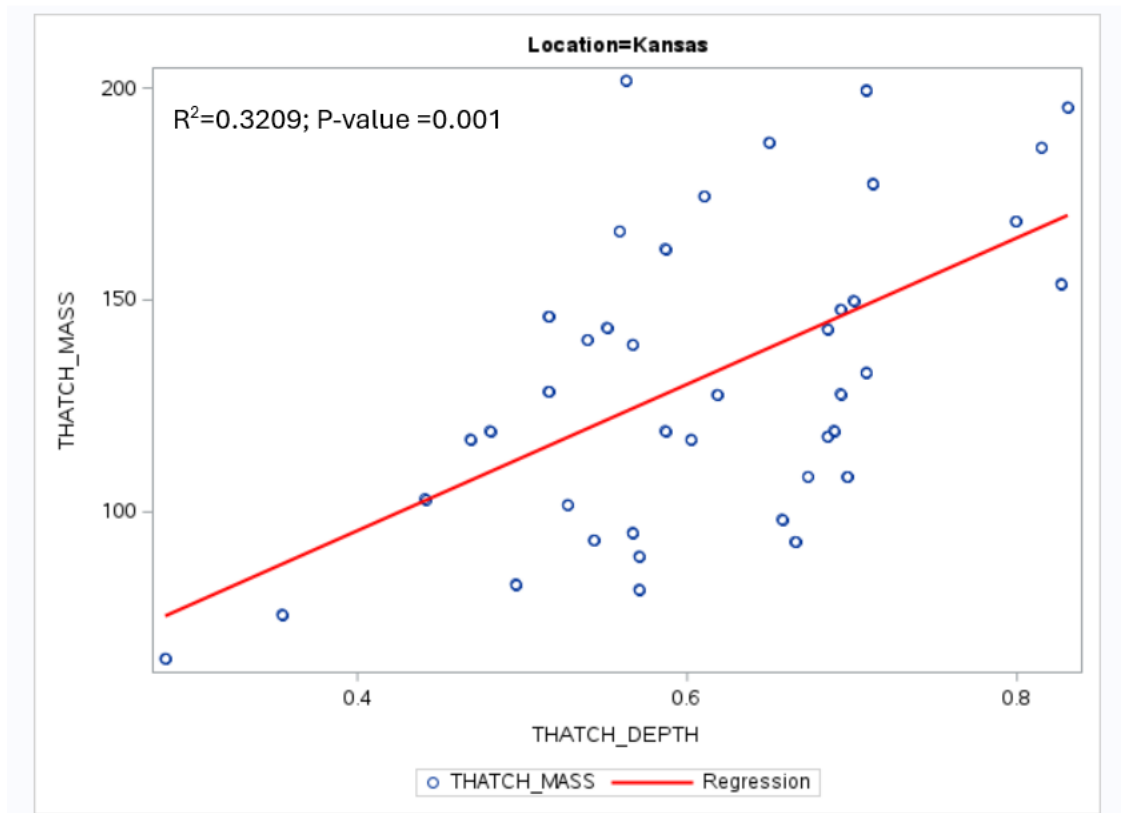


Figure 4.10 Correlation between thatch depth (in.) and thatch mass (mg) in the USGA trial in Olathe, KS ($P < 0.001$). Thatch depth and mass data were collected from plugs sampled in 42 plots in May 2024.

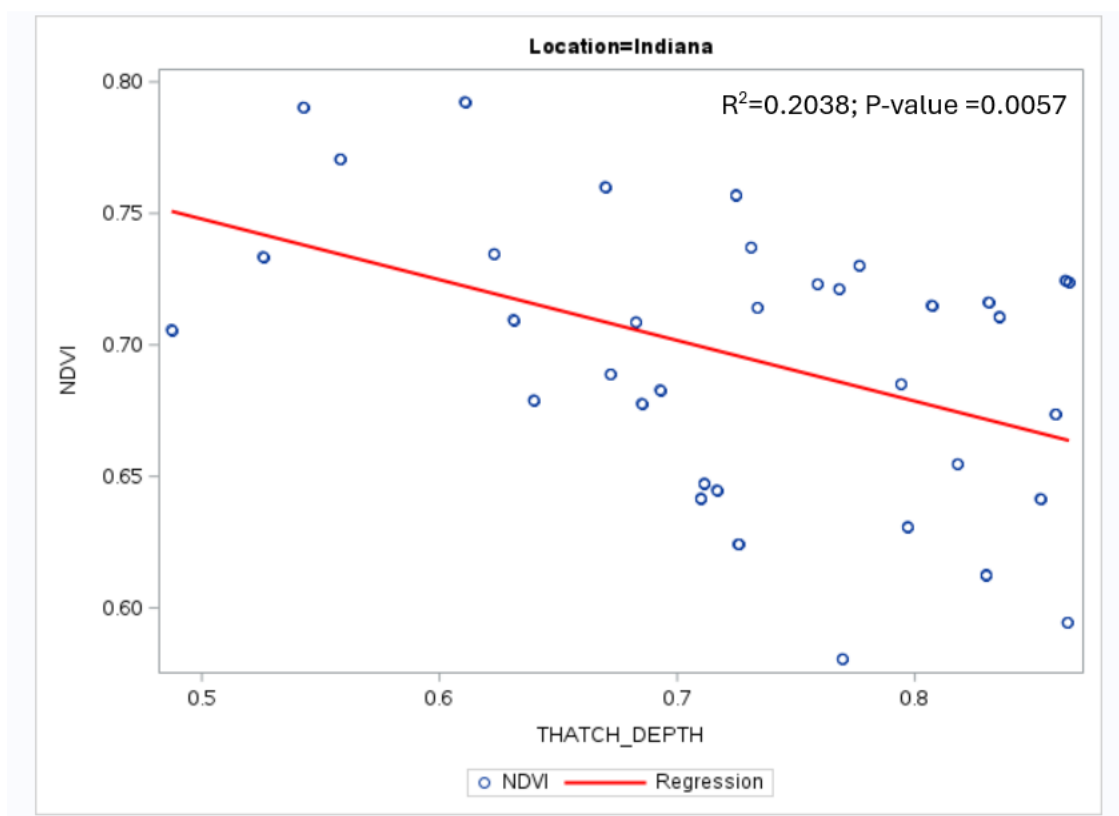


Figure 4.11 Correlation between thatch depth (in.) and NDVI in the USGA trial in West Lafayette, IN ($P < 0.01$). NDVI measurements occurred across 36 plots in July 2024.

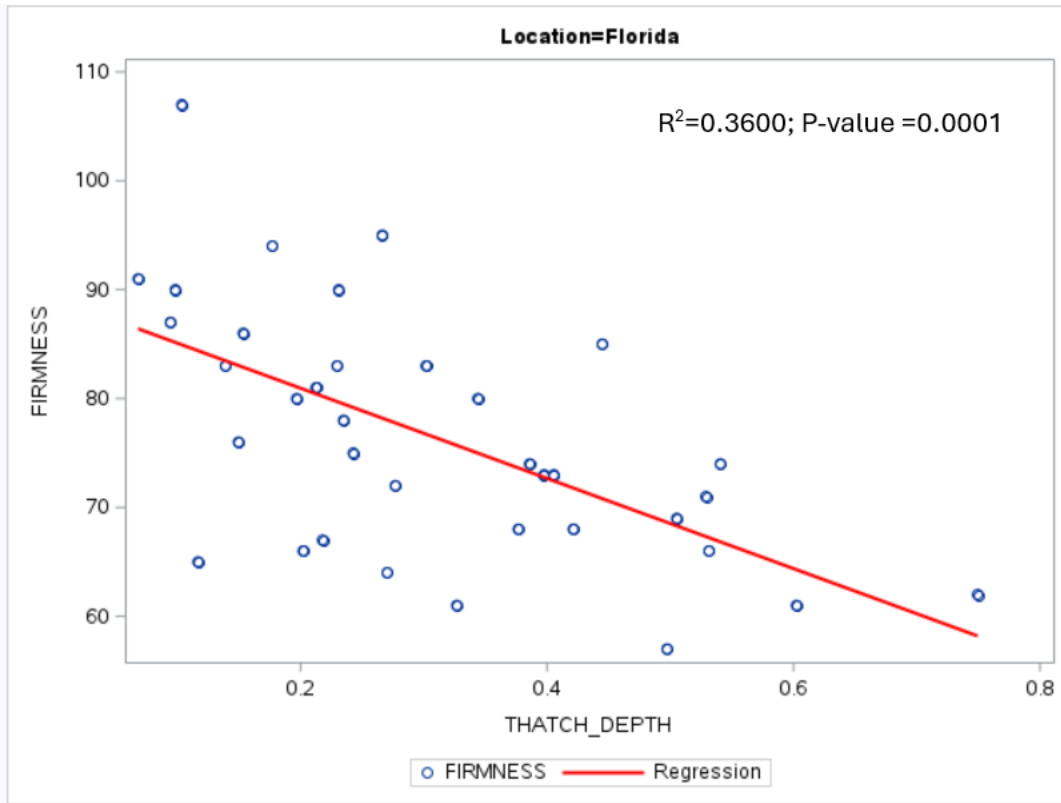


Figure 4.12 Correlation between thatch depth (in.) and turfgrass firmness (Clegg Hammer measurement in surface unit “gravities”) which measures the surface hardness (lower values softer firmness; higher values harder firmness) in the USGA trial in Citra, FL ($P < 0.001$). Firmness was measured once in April 2024 across 36 plots.

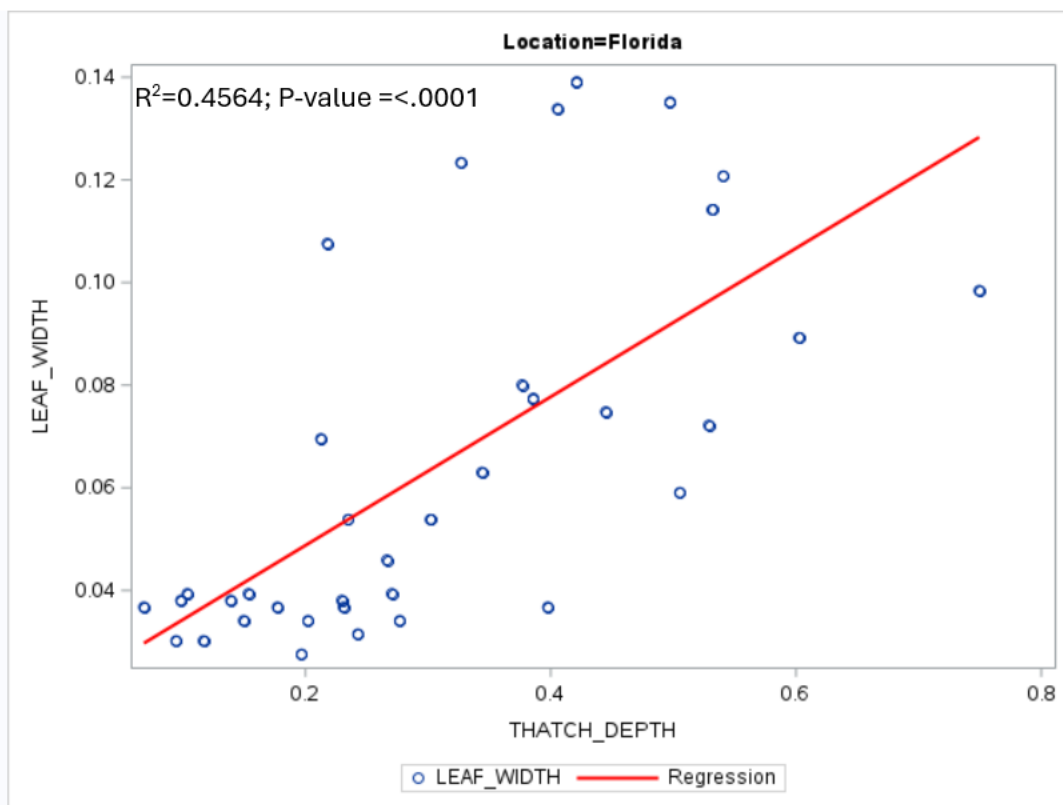


Figure 4.13 Correlation between thatch depth (in.) and leaf width (in.) in the USGA trial in Citra, FL ($P < 0.001$). Data was collected on plugs removed from 36 plots in April 2024.

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