SPRING ESTABLISHMENT OF CREEPING BENTGRASS AND ANNUAL BLUEGRASS ON PUTTING GREENS FOLLOWING SIMULATED WINTERKILL

By

Payton Clare Perkinson

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ABSTRACT

In the springs of 2023 and 2024, two trials were conducted to evaluate spring establishment of creeping bentgrass (*Agrostis stolonifera* L.) and, annual bluegrass (*Poa annua* var. Reptans (Hauskn.)) on putting greens following simulated winterkill. Trial 1 in 2023 evaluated spring establishment on bare soil following sod removal from a putting green at 4 locations; Hancock Turfgrass Research Center (HTRC), East Lansing, MI; O.J. Noer Research Station, Verona, Wisconsin; Horticulture Research Station, Ames Iowa; and Turfgrass Research, Outreach, and Education Center (TROE), St. Paul, Minnesota. Trial 2 in 2023 and 2024 evaluated putting green establishment on an annual bluegrass putting green that had been sprayed with non-selective herbicide and interseeded at Hancock Turfgrass Research Center (HTRC), East Lansing, MI. Both trials consisted of evaluating timing of spring seeding: early, middle, or late spring seeding, and seed entry: four creeping bentgrass cultivars, ‘Penncross’, ‘Pure Distinction’, ‘Declaration’, ‘Penn A-4', and one annual bluegrass, ‘Two-Putt’, and a mixture of Pure Distinction and Two-Putt. All entries including the Pure Distinction and Two-Putt mix were seeded at 49 kg ha^{-1}. In both Trial 1 and Trial 2, timing of spring seeding was significant for the first one or two measurement dates at the beginning of the study but would not drastically change when golf courses opened in the spring following winterkill. In both studies seed entry was significant but numeric differences in percent turfgrass cover were minimal.

Dedicated to my parents, Erin and Will, and my sister Reagan. Thank you for your unconditional love and support.

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TABLE OF CONTENTS

CHAPTER 1: LITERATURE REVIEW

INTRODUCTION

Creeping bentgrass (*Agrostis stolonifera* L.; CBG) is a cool-season turfgrass used on putting greens throughout the world because of its fine texture, high shoot density, uniformity, and quality (Beard, 1973). Due to the intense management of putting greens, perennial biotypes of annual bluegrass (*Poa annua* L.; ABG) have been found to invade and persist on these playing surfaces (Huff, 1999). Annual bluegrass removal is challenging and costly, so it often becomes the dominant species under putting green management. Despite annual bluegrass being a cool season turfgrass with a C3 photosynthetic pathway, a major weakness is its susceptibility to winterkill compared to other turfgrass species such as creeping bentgrass (Beard and Olien, 1963).

The winter of 2013-2014 caused devastating loss of turfgrass throughout the Great Lakes region, with most damage being on annual bluegrass putting greens (Frank, 2014). When winterkill events occur golf course superintendents are left to re-establish playing surfaces as quickly as possible to reduce revenue loss. Interseeding creeping bentgrass is the most popular and cost-effective method of re-establishment, but persistent cold and cloudy conditions in the spring reduce the speed of establishment. Recent growth chamber studies have shown a difference in germination rates between CBG cultivars in low temperatures (Heineck et al., 2019; Carroll et al., 2020). However, spring re-establishment trials out in the field have had varying results and there is little to no research conducted on when golf course superintendents should seed in the spring.

WINTERKILL

Turfgrass winterkill is defined as the death of turfgrass in the winter months (Beard and Beard, 2005). There are various causes of winterkill, both abiotic and biotic, which include direct low-temperature kill, crown hydration, anoxia, desiccation, and low-temperature fungi.

The ability of turfgrass to successfully survive winter is positively correlated to freezing tolerance (Humphreys and Eagles, 1988). Typically, in the autumn, during shorter photoperiods and lower temperatures, overwintering plants go through a cold acclimation process to increase freezing tolerance. During cold acclimation, turfgrass plants develop freezing tolerance through physiological and biochemical changes (Levitt, 1980) such as decreased crown water concentration (Beard, 1969) and an increased storage of carbohydrates such as fructans and sucrose in the turfgrass crown (Dionne et al., 2001; Dionne et al., 2010; Hoffman et al., 2014).

Beard (1966) found that cool-season turfgrasses have differing levels of cold hardiness. Rough bluegrass (*Poa trivialis* L) and creeping bentgrass have excellent low-temperature hardiness, annual bluegrass has intermediate hardiness and perennial ryegrass (*Lolium perenne* L.), and annual ryegrass (*Lolium multiflorium* Lam.) have poor and very poor cold hardiness (Beard, 1969). Gusta et al. (1980) evaluated the crown hardiness of six field cold acclimated turfgrass species by measuring the minimum temperature that causes 50 percent of the sample to die (LT_{50}). Creeping bentgrass had the highest freezing tolerance (LT_{50} –35°C) and perennial ryegrass had the lowest freezing tolerance (LT_{50} –5 to –15°C) (Gusta et al., 1980). Cold acclimated annual bluegrass has been found to have a LT_{50} range of –19 to –31°C and can vary depending on ecotype (Dionne et al., 2010). However, the genetic differences in cold hardiness found in these turfgrass species will not be fully expressed without proper cold acclimation

(Vavrek, 2016). Non-acclimated CBG and ABG were found to have LT_{50} values of –8 and –9°C, respectively (Hoffman et al., 2014).

Cold acclimated turfgrass is most tolerant to freezing injury from December to February with a sharp decline in hardiness in late winter and early spring (Beard, 1969; Dionne et al., 2001). Fowler and Gusta (1977) discovered when winter wheat *(Triticum aestivum* L.) and rye (*Secale cereale* L.) are exposed to increased temperatures, plants began to de-harden (de-acclimate), resume growth, and decrease cold hardiness potential. Studies conducted in cool-season turfgrass have found similar trends existing between warmer soil temperatures, reduced cold hardiness, and increased crown hydration (Tompkins et al., 2000; Hoffman et al., 2014; Hoffman et al., 2014; DaCosta et al., 2020). Annual bluegrass is more susceptible to premature cold de-acclimation and loses freezing tolerance when exposed to lower temperatures for a shorter duration of time compared to creeping bentgrass (Hoffman et al., 2014; Hoffman et al., 2014). CBG's ability to maintain greater freezing tolerance when exposed to warmer temperatures has been associated with maintaining higher fructans and total soluble sugars throughout de-acclimation (Hoffman et al., 2014).

Crown hydration is one of the most destructive and least preventable types of winterkill. When exposed to warmer spring temperatures during freeze/thaw cycles, turfgrass crowns begin rehydrating. In wheat and barley (*Hordeum vulgare* L.), small increases in percent crown moistures at a given temperature have drastic differences in plant survival (Metcalf et al., 1970). When compared, annual bluegrass has a higher percent crown moisture than creeping bentgrass (Tompkins et al., 2000; Hoffman et al., 2014). When plants with high crown hydration are exposed to freezing temperatures, they become more susceptible to lethal ice crystals forming both intracellularly and extracellularly (Beard, 1969). Intracellular freezing is extremely deadly and occurs when hydrated plants are exposed to rapid cooling and ice forms within the cell (Guy, 1990). When exposed to a gradual temperature decrease, temperate plants can move water outside the cell and have ice form extracellularly (Burke, 1976). Extracellular freezing is rather harmless but can have a desiccation effect when extracellular ice acts as a nucleation site for water to be drawn out of the cell and can be lethal (Andrews, 1996).

Layers of ice can form on putting greens from ice storms, freezing rain, poor drainage or under snow cover during freeze/thaw cycles. Annual bluegrass has been documented to be killed between 75 to 90 days of ice encasement (Beard, 1964) and a loss of cold hardiness has been reported after 45 days of ice cover (Tompkins et al., 2004). Creeping bentgrass has been found to survive over 120 days under ice (Beard, 1965). The lethal cause of ice cover is the depletion of oxygen and the buildup of carbon dioxide, ethyl butyrate, and other toxic gasses (Aamlid et al., 2009). Castonguay et al. (2009) found that a lack of oxygen was the main cause of damage independent from carbon dioxide level with ABG being more sensitive to low oxygen levels than CBG. Rochette et al. (2006) found that native soil putting greens and greens that had recurring winter damage had greater O_2 consumption under impermeable covers due to higher levels of organic carbon and increased rates of respiration.

Desiccation is the lethal dehydration of a plant when water loss exceeds water uptake (Charbonneau, 2010). Winter desiccation occurs when there is low atmospheric moisture or when soil is frozen. Desiccation injury is common in elevated areas that are exposed to excessive wind movement and can be exacerbated in areas with high thatch levels (Beard, 1973; Beard and Beard, 2005; Happ, 2004). In Nebraska, Kreuser (2014) observed that creeping bentgrass areas that were exposed to wind and had a thatch layer greater than 1.27 centimeters had more severe damage.

Turfgrasses are also susceptible to winterkill from low-temperature diseases. The two most common low-temperature diseases are *Microdochium* patch or pink snow mold (*Microdochium nivale* Fr. Samuels & I.C. Hallett) and gray snow mold or *Typhula* Blight (*Typhula incarnata* Lash ex Fr.) (Vargas, 2005). Gray snow mold requires snow cover and temperatures between –1 and 12.7°C for infection and produces gray or tan patches that average about 15 to 30 cm in diameter (Vargas, 2005). *Microdochium* patch does not require snow cover to form and is most active at temperatures of 8 to 17°C with greater than or equal to 90% humidity for more than 20 hours (Dwyer, 2004). *Microdochium* patch produces reddish brown or copper patches between 2.5 to 20 cm in diameter (Vargas, 2005).

PREVENTION STRATEGIES

Implementing winterkill prevention strategies is challenging due to the variability of winter weather from year to year. Common practices include reducing shade, increasing fall mowing heights, proper thatch management, and improving drainage (Beard, 1969). Thick and stable snow cover is an excellent insulator and can reduce desiccation and freezing stress by maintaining soil temperatures, turfgrass dormancy, and cold hardiness (Dionne et al., 1999; Tompkins et al., 2000; Tompkins et al., 2004).

If snow cover is limited, permeable or impermeable covers can be used to protect greens from winter stresses such as ice cover. However, a golf course's ability to place covers on greens might be limited due to high cost and labor requirements. Permeable covers allow for gas and water exchange which is important if covers need to be left on for an extended period (Bauer et al., 2014). Impermeable covers do not allow for gas exchange (Rochette et al., 2006) so venting covers periodically through the winter is advised (Frank, 2016). Dionne et al. (1999) found that placing impermeable covers over insulating materials such as straw mats created airspace under the cover and allowed management of crown moisture and temperature. Protective covers provide a similar environment to snow cover so proper application of fungicides to prevent snow mold formation in the fall is required (Frank, 2016; Koch, 2017).

In areas where desiccation injury is a major concern, the use of heavy topdressing and both permeable and impermeable covers has been found to maintain crown moisture content (Michael and Kreuser, 2020). Other management for desiccation includes installing windbreaks, snow fences, and irrigating when temperatures are above freezing using portable water tanks or sprayers (Charbonneau, 2010; Vavrek, 2016).

Removing ice from putting greens is a highly debated topic and can require superintendents to make difficult decisions about ice removal. Anecdotal evidence from Detroit area golf courses suggests the best strategy for preventing ice damage is to break up the ice and remove it from the playing surface when daytime temperatures are below freezing (Vargas and Turgeon, 2004). Similarly, Tompkins et al. (2004) discovered that the best time to remove ice off annual bluegrass putting greens is as soon as possible and that waiting up to 45 days to remove ice will have no benefit. Alternatively, Hollman et al. (2017) found that applying chemical or solar de-icing products can be successful in melting ice on putting greens. However, some of these products were found to cause phytotoxicity when applied directly on turfgrass so solar de-icing products such as black sand or single applications of nitrogen-free chemical de-icing products are recommended.

The effect of autumn nitrogen applications on winter hardiness has had varying results through the years. Beard (1969) states that excessive nitrogen fertilization, irrigation, or other cultural practices that stimulate growth will reduce cold hardiness. In Kentucky bluegrass (*Poa pratensis* L.), Caroll and Welton (1939) found that high rates of nitrogen (1.22 kg N ha^{-1}) applied in September and October reduced Kentucky bluegrass resistance to sub-zero temperatures. More recently, Webster and Ebdon (2005) reported late fall nitrogen applications on perennial ryegrass did not increase winter injury potential. Research on the effects of nitrogen applications on cold hardiness in harsher climates is quite limited, and more work on the topic is needed (Bauer et al., 2012).

Little research has been conducted on the relationship between potassium and cold hardiness on cool-season turfgrass. Webster and Ebdon (2005) found that maximum cold hardiness occurred in perennial ryegrass with higher rates of potassium (235 to 441 kg K ha yr^{-1}). In the spring of 2015, Schmid et al. (2016) observed that ABG plots that had not been fertilized with potassium for the last three years had 58% winter damage while plots that were fertilized with potassium biweekly throughout the growing season had less than 4% winter damage. They found there was no difference between potassium sources or rates greater than 52 kg K ha^{-1}(Schmid et al., 2016).

PUTTING GREEN RE-ESTABLISHMENT

Re-establishing turfgrass following winterkill is challenging due to suboptimal soil temperatures and cloudy conditions. Golf course superintendents can either sod or seed areas depending on the scale and pattern of damage. Areas with distinct kill margins are feasible to sod and seeding should be considered in areas with sporadic kill (Frank, 2020). Superintendents can use winterkill events as an opportunity to repopulate creeping bentgrass in killed annual bluegrass voids and reduce the potential for winter injury in the future (Stier, 2005). On golf courses that have annual bluegrass greens but do not want to introduce creeping bentgrass, the only seed currently available on the market is ‘Two-Putt’ (*Poa annua* L.f. *var.* Reptans Hauskins T. Koyama). ‘PA-33’, a new annual bluegrass seed variety is set to be released soon for putting greens and fairways as part of Dr. David Huff's breeding program at Penn State University (Reitman, 2024).

The optimal germination temperatures for creeping bentgrass seed are 15 to 30°C (Toole and Koch, 1977) and daily average temperatures below 7°C are unacceptable for CBG germination (Heineck et al., 2019). Carroll et al. (2020) found that a significant proportion of creeping bentgrass seed can germinate within 10 to 17 days at 10°C and 8:16 hour light-dark cycles.

Multiple growth chamber studies have found differences in cold germination between CBG cultivars. Heineck et al. (2019) studied the germinability of 21 creeping bentgrass cultivars at four different spring simulated temperatures. The day/night temperatures were based off historical averages from the upper Midwest and were 7/1.6, 12.7/1.5, 15.5/4.4, 21.1/10°C. Their results showed that 'Proclamation', 'Declaration' and 'Pure Select' had good low-temperature germination, and 'Independence' and 'Memorial' had poor low-temperature germination (Heineck et al., 2019). At 10°C, 'Penn A-4', 'Pure Select', 'CrystalBlueLinks', and '007' had some of the highest germination percentages and took the fewest days to germinate (Carroll et al., 2020). Inhibition of photosynthesis and growth when exposed to below-freezing temperatures can vary between cultivars. Seedlings of 12 creeping bentgrass cultivars were exposed to –5°C for eight hours. Three days after cold exposure, 'Piranha', 'Declaration', 'T1', and 'Penn A-4' had the highest visual quality and photosynthetic efficiency, and 'Barracuda', 'Memorial', 'Independence', and 'Luminary' had visual injury (DaCosta and Watkins, 2024).

Winterkill re-establishment has had varied success in the field. Frank et al. (2017) studied spring establishment rates of three creeping bentgrass cultivars ('Alpha', 'Penn A-4', 'Providence') and annual bluegrass florets on preexisting putting greens that had simulated winterkill. In the first year of the study, Providence and Penn A-4 had significantly higher percent cover for two weeks in the first year of the study. In the second year of the study, there was no statistical significance between CBG cultivars, but all three cultivars had a higher percent turfgrass cover compared to ABG florets. Ebdon and DaCosta (2021) evaluated creeping bentgrass, colonial bentgrass, and velvet bentgrass (*Agrostis canina* L.) cultivars on spring establishment. They found that creeping and colonial bentgrass had faster establishment than velvet bentgrass and out of the seven CBG cultivars, 'L-93', 'T-1', and 'Declaration' had the slowest establishment rates.

When studying creeping bentgrass seeding rates from 49 to 390 kg seed ha^{-1}, Madison (1966) found that the effective seeding rate for establishment of creeping bentgrass is 49 to 98 kg seed ha^{-1}. Green et al. (2018) evaluated six creeping bentgrass seeding rates, 6, 12, 24, 37, 49, and 98 kg seed ha^{-1}, on a bare soil site and discovered that seeding rates as low as 37 kg seed ha^{-1} are acceptable when establishing CBG. When seeding into existing putting greens, seeding rates of 73 kg seed ha^{-1} were more effective compared to 36.6 kg seed ha^{-1} (Carroll et al., 2021).

Previous research suggests that slicing is the most effective cultivation method for seeding into pre-existing putting greens. Miltner et al. (2005) observed that verticutting in two directions at a depth of 3.4 mm was the most effective method of establishing 'True-Putt' compared to solid and hollow tine cultivation methods. Carroll et al. (2021) found similar results that slicing into preexisting putting greens had the highest turfgrass cover. The Jobsaver, an aerator attachment, creates small cone-shaped depressions in the putting surface has also had success when establishing creeping bentgrass (Frank et al., 2017), especially at lower seeding rates (Carroll et al., 2021).

Protective covers have been found to increase soil temperatures (Patton et al., 2010) but spring creeping bentgrass re-establishment with protective covers has had mixed results. In Michigan, the use of a clear polyethylene cover when nighttime temperatures were lower than 10°C showed only a small (3-6%) increase in turfgrass cover compared to treatments without the polyethylene cover (Frank et al., 2017). In Nebraska, the use of green pigment, white permeable covers, black landscape fabric, and clear plastic sheeting with small holes were studied on spring establishment of 'L-93' creeping bentgrass. The protective covers never increased turfgrass cover compared to uncovered control (Michael, 2016). Recently, DaCosta and Watkins (2024) found that creeping bentgrass plots that used a permeable cover during spring establishment reached 50% cover 7 to 12 days faster than the uncovered plots but by early June there was no difference in turfgrass cover between treatments.

Application of nitrogen during the first few weeks of establishment increases seedling vigor and turfgrass growth (Watson et al., 2012). Chestnut (2018) found that when establishing creeping bentgrass during late summer and early fall the optimal range for nitrogen is 9.78 to 12.23 kg N ha^{-1} a week. Carroll et al. (2021) evaluated 3 annual nitrogen levels (24, 49, 73 kg N ha yr^{-1}) on establishment of creeping bentgrass on preexisting putting green and reported that 73 kg N ha yr^{-1} had the highest percent cover. Michael (2016) tested 3 nitrogen rates, 0, 4.9, 24.4 kg N ha^{-1}, weekly throughout the spring establishment of 'L-93' CBG and did not find any differences among fertilizer treatments. Similarly, Frank et al. (2017) evaluated spring establishment rates between a nitrogen-only treatment at a rate of 0.5 kg N ha^{-1} weekly and a starter fertilizer treatment (19N-25P-5K) was applied at 1.5 kg N ha^{-1} every 21 days. The starter fertilizer treatment was only significant on re-establishment on three measurement dates out of a two-year study.

Information on winterkill re-establishment is quite limited due to the lack of peer-reviewed research available. The overarching objectives of this thesis are to i) evaluate three spring seeding dates on establishment rate and, ii) evaluate seven seeding entries on establishment in the spring. In chapter two, the objectives were studied on a bare soil putting green in 2023 at four locations: Michigan State University, Iowa State University, University of Wisconsin, and University of Minnesota. In chapter three, the primary objectives were studied at Michigan State University during the spring of 2023 and 2024 on an annual bluegrass putting green that was interseeded following non-selective herbicide application to simulate winterkill.

CHAPTER 2: SPRING ESTABLISHMENT OF CREEPING BENTGRASS AND ANNUAL BLUEGRASS

ABSTRACT

Re-establishing golf course putting greens following winterkill is challenging due to the prevalence of cloudy and cold temperatures during the spring in northern climates. Very little information is available on timing of spring seeding and what cultivar should be selected in the field. The objective of the study was to evaluate spring establishment rate of creeping bentgrass (*Agrostis stolonifera* L.), annual bluegrass (*Poa annua* var. Reptans (Hauskn.)), and a 50/50 by weight mixture of creeping bentgrass and annual bluegrass, seeded three consecutive weeks in the spring. The experiment was conducted in East Lansing, MI; St. Paul, MN; Ames, IA; and Verona, WI. The experimental design was a randomized complete block split-plot design with four replications. The whole plot was the timing of spring seeding with three levels: early, middle, and late spring seeding, and the subplot was seed entry. Seed entry treatments were four creeping bentgrass cultivars, ‘Penncross’, ‘Pure Distinction’, ‘Declaration’, ‘Penn A-4', one annual bluegrass, ‘Two-Putt’, and a mixture of Pure Distinction and Two-Putt, and an unseeded control. Seed was applied with hand shaker bottles at a rate of 49 kg ha^{-1} for three consecutive weeks once soil temperature at each respective location reached 7°C at 5 cm soil depth. Data was collected for five weeks during the establishment period with digital image analysis to assess percent turfgrass cover. Statistical analysis was conducted separately for each location. Timing of spring seeding was significant for the first measurement date in Michigan and Iowa and the first two measurement dates in Wisconsin but numerical differences in turfgrass cover between treatments was small. Seed entry was significant at three locations with Pure Distinction, Two-Putt, and the Pure Distinction and Two-Putt mix treatment having the highest turfgrass cover

across locations. Although there were statistical differences among entries, turfgrass cover differences were small and relatively insignificant at the end of the study.

MATERIALS AND METHODS

The study was conducted in 2023 at four locations: Michigan State University, Hancock Turfgrass Research Center (HTRC), East Lansing, MI; University of Wisconsin, O.J. Noer Research Station, Verona, Wisconsin; Iowa State University, Horticulture Research Station, Ames, Iowa; and University of Minnesota, Turfgrass Research, Outreach, and Education Center (TROE), St. Paul, Minnesota (Table 1). The total experimental area at each location was 128 m^2. The putting greens in Michigan, Wisconsin, and Minnesota were constructed under USGA recommendations and the Iowa location was a native soil putting green grown on Clarion Loam (fine-loamy, mixed, superactive, mesic Typic Hapludolls).

The experimental design was a randomized complete block split-plot design with four replications. The whole plot (timing of spring seeding) included three levels: early, middle, and late. The subplot (seed entry) included seven levels: ‘Penn A-4’, ‘Declaration’, ‘Penncross’, ‘Pure Distinction’, ‘Two-Putt’, ‘Pure Distinction’ and ‘Two-Putt’ 50/50 by weight mix, and an unseeded control. The Two-Putt and Pure Distinction 50/50 by weight mix treatment was added to simulate creeping bentgrass seed competing with annual bluegrass seed from the soil seed bank. The same seed lot for each seed entry was used across all four sites and seed was from the years of 2019 and 2020. Each putting green was divided into twelve 20 m^2 whole plots and each whole plot was split into eight 0.8 m^2 subplots.

In the autumn of 2022, the sod from pre-existing creeping bentgrass stands maintained as putting greens were removed at each site using a sod cutter. In spring 2023, the first seeding date occurred when daily average soil temperatures reached 7°C at a 5 cm soil depth at each location. Seed treatments were weighed out individually and applied by hand shaker bottles at a rate of 49 kg ha^{-1}. Plots were lightly raked in two directions with a leaf rake placed upside down to facilitate soil-to-seed contact. Starter fertilizer (ContecDG, 12N-48P-8K) (The Andersons, Maumee, OH) was applied at 21.5 kg P ha^{-1}. The seeding process was repeated every seven days for three weeks (Table 2).

Plots were mowed three times a week at 0.31 cm with a walking greens reel-mower. After the first seedling emergence, granular urea (46N-0P-0K) was applied bi-weekly with hand shaker bottles for eight weeks at a rate of 9.7 kg N ha^{-1} and was immediately watered in after application with 0.64 cm of irrigation. Irrigation was applied when necessary to provide adequate germination and establishment. Precipitation, soil temperature, and air temperature data was collected daily at each site using on-site weather stations. Due to weather station malfunctions, only air temperature and daily precipitation were recorded at the Iowa location.

Data collection

Single images were captured weekly for five weeks (Table 3) during the establishment period from the center of the plot. Images were then analyzed to assess turfgrass cover by quantifying the number of green pixels in the image (Karcher and Richardson, 2013).

Statistical analysis

Data from each location was analyzed separately through PROC GLIMMIX using SAS software (version 9.4; SAS Institute Inc., Cary, NC). Block was treated as a random effect and timing of spring seeding was nested within each block. Analysis of Variance (ANOVA) was conducted on the fixed effects of timing of spring seeding and seeding entry to determine significant differences among treatment means ($P \leq 0.05$). Fisher's Least Significance Difference (LSD) were used when significant differences between treatments occurred.

RESULTS AND DISCUSSION

Timing of spring seeding x seed entry interaction

The interaction between timing of spring seeding x seed entry was significant on 12 May and 18 May at the Wisconsin location (Table 4). Within the early, middle, and late spring seeding dates, Pure Distinction and Two-Putt mix had the highest percent turfgrass cover but was not statistically different from Pure Distinction at the early spring seeding date (Table 7). The timing of spring seeding within the Pure Distinction seed entry was significant. On 12 May, the early spring seeded Pure Distinction had significantly higher turfgrass cover with 17.6% compared to the middle and late seeding which had 6.2% and 8.7%. On 18 May, Pure Distinction again had significance within timing of spring seeding treatments. The early spring seeding date had 25.7% and the middle and late seeding dates had 13.6% and 17.2% (Table 8). However, by 26 May, there was not a significant interaction among timing of spring seeding x seed entry at the Wisconsin location.

In Michigan, the interaction between timing of spring seeding x seed entry was significant on 10 May and 17 May (Table 5). On 10 May, within the early spring seeding treatments, Two-Putt had the highest percent turfgrass cover with 11.3% and was statistically different from the middle and late spring seeding date of Two-Putt with 5.8% and 3.3% (Table 9). All entries seeded on the late seeding date averaged 3% including the unseeded control. These results suggest that minimal emergence had occurred by 10 May. On 17 May, within the early spring seeding date, Two-Putt had the highest percent turfgrass cover with 19.0% but was not statistically different from Pure Distinction and Two-Putt mix (Table 10). Within the middle and late spring seeding dates, entries were not significantly different from each other except for the ‘Declaration’ and control treatments which had the lowest percent turfgrass cover.

In Minnesota, timing of spring seeding x seed entry interaction was significant on 26 May and 2 June (Table 6). On 26 May, the early spring seeding of the Pure Distinction and Two-Putt mix, had the highest percent turfgrass coverage with 85.5% but was not statistically different from the Penn A-4, Pure Distinction, and Two-Putt treatments (Table 11). The timing of spring seeding within the Two-Putt seed entry was significant. The early seeding of Two-Putt had 70.3% while the middle and late had 49.4% and 44.5%. On 2 June, seed entries within the early and middle spring seed dates (excluding the control) were not statistically different from each other (Table 12). Within the late seeding date, Pure Distinction and Two-Putt mix, Pure Distinction, Penncross, and Penn A-4 were not statistically different from each other. Declaration and the control had the lowest percent turfgrass cover out of entries in the late spring seeding date.

Timing of spring seeding main effect

In Wisconsin, the date of spring seeding was significant on the 12 May and 18 May measurement dates (Table 4). On 12 May, all three spring seeding dates were statistically different from one another (Table 13). The early spring seeding treatment had the highest percent turfgrass cover with 9.3%, the late spring seeding date had 6.6%, and the middle spring seeding date had 4.8%. On 18 May, the early spring seeding date had the highest percent turfgrass cover with 15.6% and was different from the middle and late spring seeding dates which had 10.7% and 12.5%, respectively. Although statistically different, there was only a 4.6% cover difference between the highest and lowest treatments on 12 May and a 4.9% difference on 18 May.

In Michigan, the timing of spring seeding was significant only on the 10 May measurement date (Table 5). The early spring seeding treatment had 6.4% turfgrass cover and was not different from the middle spring seeding date which had 5.0% turfgrass cover (Table 14).

The late spring seeding date had the lowest percent turfgrass cover with 3.3%. Despite there being differences, the percentage of turfgrass cover on 10 May averaged less than 10% across all treatments.

The main effect of timing of spring seeding was also significant in Iowa on the 15 May measurement date (Table 15). The middle spring seeding date had the highest percent turfgrass cover with 25.9% but was not statistically different from the late spring seeding date which had 23.3% (Table 16). The late spring seeding date was not statistically different from the early seeding date which had 16.4% turfgrass cover.

In Pennsylvania, research evaluated spring establishment of creeping bentgrass at three different spring seeding dates: 200, 400, and 600 Growing Degree Days (GDD) base 0°C with GDD accumulation beginning on 1 March (Carroll, 2019). On the first rating date, the GDD 200 treatment had a higher visual percent turfgrass cover compared to GDD 400 and 600 treatments. On the second rating date, the GDD 200 and 400 treatments had greater visual seedling vigor and turfgrass cover compared to the 600 GDD treatment. Although Carroll's study was conducted for only one year these results are similar to the results observed at the Wisconsin and Michigan locations.

Seed entry main effect

Seed entry main effect was significant in Michigan on every measurement date (Table 5). On 10 May and 17 May, Two-Putt had the highest turfgrass cover with 6.8% and 13.9% (Table 17). On 24 May and 31 May, Two-Putt had the highest turfgrass cover but was not statistically different from the Pure Distinction and Two-Putt mix, Penn A-4, and Penncross entries. Throughout the study in Michigan, Declaration had the lowest percent turfgrass cover out of the seed entries, excluding the unseeded control. Although there were differences among treatments, by 7 June no entry had reached above 40% turfgrass cover.

In Wisconsin, seed entry was significant on every measurement date (Table 4). On 12 May, 18 May, and 26 May measurement dates, the Pure Distinction and Two-Putt mix treatment had the highest percent turfgrass cover out of all the seed entries (Table 18). On 2 June and 9 June, the Pure Distinction and Two-Putt mix, Pure Distinction, and Penncross treatments had the highest percent turfgrass cover. Declaration had the lowest percent turfgrass cover across seeded entries and on 12 May and 18 May, was not statistically different from the control.

In Minnesota, seed entry was significant on four of five measurement dates (Table 6). On the first measurement date, 26 May, the Pure Distinction and Two-Putt mix had the highest percent turfgrass cover with 78.5% (Table 19). On 2 June, the Pure Distinction and Two-Putt mix again had the highest percent turfgrass cover but was not statistically different from Penn A-4 and Pure Distinction. By the 16 June measurement date, the only differences among treatments were the seeded entries compared to the control treatment and on 24 June, there were no differences between any of the treatments. On the next measurement date, 30 June, the Pure Distinction and Two-Putt mix had the highest percent turfgrass cover but was not different from Pure Distinction, Penncross, Penn A-4, and Declaration.

The lack of significant differences in turfgrass cover among all the treatments and the unseeded control on 24 June in Minnesota and in Iowa (Table 15) can be attributed to high levels of annual bluegrass contamination. Despite the control treatments not having any seed applied to it, these treatments averaged 93% and 94% by the fifth measurement date in Iowa (Table 29) and Minnesota (Table 19). In preparation for the study, every site removed the pre-existing turfgrass from the putting green which exposed bare soil and likely promoted germination of annual bluegrass. In Maryland, Kaminski and Dernoeden (2007) observed invasion of annual bluegrass in bare soil areas between the months of March and May and recorded counts as high as 69 annual bluegrass seedlings per 930 cm^2. Research has also found that annual bluegrass seedbanks can vary between locations. Green et al. (2019) sampled putting green root zones from four different golf courses in Michigan. They found that annual bluegrass seedling emergence varied between golf courses and hypothesized these results were due to differences in sand topdressing practices. This could explain why, in the current study, only two out of four locations had heavy annual bluegrass contamination.

The Pure Distinction and Two-Putt mix treatment was the top performing seed entry in Wisconsin for the first three measurement dates and in Minnesota on the first measurement date. In a growth chamber experiment, Heineck et al. (2019) tested 21 CBG cultivars for germinability under cool temperatures. They found that Pure Distinction accumulated the fourth highest area under the germination curve ($AUGC_{98}$) in growth chambers set to day/night temperatures of 12.7/1.5°C. In addition, annual bluegrass has been found to have a minimum germination temperature of 5°C (Sherner et al., 2017). In Scandinavia, researchers evaluated annual bluegrass along with rough bluegrass (*Poa trivialis* L.) and perennial ryegrass (*Lolium perenne* L.) as nurse species with 'Independence' creeping bentgrass to aid in faster spring establishment due to these respective species having lower germination temperature requirements. They found Two-Putt and Independence creeping bentgrass mix had 10% higher turfgrass cover 3 weeks after seeding compared to the Independence only treatment. Despite the faster establishment, Two-Putt was more prone to winter damage and required to be reseeded the following year (Heltoft et al., 2022). In the present study, although the Pure Distinction and Two-Putt mix was the highest performing seed entry in the beginning of the study in Wisconsin and Minnesota, reintroducing annual bluegrass perpetuates the issue of future winterkill due to its susceptibility to winter stress compared to creeping bentgrass.

Declaration had the lowest percent turfgrass cover on most measurement dates in Michigan, Wisconsin, and Minnesota. When evaluating seven CBG cultivars in Massachusetts, Ebdon and DaCosta (2021) found similar results with Declaration being one of the poorest establishing cultivars. However, these results contradict growth chamber studies that found that ‘Declaration’ was one of the highest performing cultivars in sub-optimal temperatures (Heineck et al., 2019). Possible explanations for the differences in performance between Declaration is the variability in germination between seed lots as well as germination rate discrepancies between growth chamber and field studies. When evaluating rough bluegrass for overseeding bermudagrass (*Cynodon dactylon* (L.) Pers. X *C. transvaalensis* Burtt-Davy) putting greens, Liu et al. (2001) discovered that germination rate varied by seed lot especially in low temperatures. Carroll et al. (2020) found similar results with creeping bentgrass having variable germination within cultivars and seed samples at 10°C. Liu et al. (2001) also found that germination rate was significantly lower in the field compared to the growth chambers set at lower temperatures. In the present study, a basic germination test revealed that ‘Declaration’ had the lowest germination percentage out of all seeded entries tested with 70.5% which could have been a factor for slower establishment (Table 20).

CONCLUSIONS

The interaction between timing of spring seeding x seed entry was only significant for two measurement dates at the beginning of establishment at the Wisconsin, Michigan, and Minnesota locations. Pure Distinction, Two-Putt, and the Pure Distinction and Two-Putt mix treatments were the top performing seed entries across locations within each spring seeding timing. The early spring seed timing had greater percent turfgrass cover compared to the middle and late timings for Pure Distinction, Two-Putt, and the Pure Distinction and Two-Putt mix treatments. By the third measurement date, at each location, timing of spring seeding x seed entry was no longer statistically significant. The main effect of timing of spring seeding was significant at Wisconsin for the first two measurement dates and in Iowa and Michigan for the first measurement date. However, numerical differences in percent turfgrass cover were small, and no treatment averaged greater than 25% turfgrass cover when significance occurred but there was no evidence to suggest that early spring (early April) seeding had any negative effects on re-establishment.

The main effect of seed entry was significant at every measurement date at the Wisconsin and Michigan locations and at every measurement date except for 24 June in Minnesota. On the first measurement date, the Pure Distinction and Two-Putt mix treatment had the highest turfgrass cover among seed entry in Wisconsin and Minnesota and Two-Putt had the highest turfgrass cover in Michigan. While initially there was statistical differences occurred among seed entries, by the fifth measurement date, differences between creeping bentgrass entries, excluding Declaration, were minimal or statistically insignificant. However, Declaration should not be excluded as a poor-performing cultivar in spring seedings as the results from this study were due to low seed viability from the seed lot. Two-Putt demonstrated the ability to germinate in early spring, but superintendents should be cautious of re-introducing annual bluegrass into putting greens as this might potentially impact future winterkill events.

Turfgrass cover at the end of the study in Wisconsin and Michigan, regardless of treatment, would be deemed unacceptable for effective and efficient putting green re-establishment. Future research should investigate various seeding rates for faster establishment. In addition, future field work should evaluate spring establishment of creeping bentgrass from various seed lots.

TABLES AND FIGURES

Table 1. Research locations for 2023 experiment evaluating spring establishment of creeping bentgrass and annual bluegrass.

Research location	Coordinates	USDA Plant Hardiness Zone[a]
East Lansing, MI	42.74 N, 84.48 W	6a
Verona, WI	42.99 N, 89.53 W	5b
Ames, IA	42.03 N, 93.63 W	5b
St. Paul, MN	44.95 N, 93.09 W	5a

[a] U.S. Department of Agriculture Plant Hardiness Zone (2023).

Table 2. Calendar dates for evaluating spring establishment of creeping bentgrass and annual bluegrass at each location, 2023.

Timing of spring seeding (T)	Calendar seeding dates			
	East Lansing, MI	Verona, WI	Ames, IA	St. Paul, MN
Early	7 April	13 April	3 April	11 April
Middle	14 April	20 April	10 April	18 April
Late	21 April	27 April	17 April	25 April

Table 3. Calendar measurement dates for assessing spring establishment of creeping bentgrass and annual bluegrass at each location, 2023.

	Date of measurement			
	East Lansing, MI	Verona, WI	Ames, IA	St. Paul, MN
1	10 May	12 May	15 May	26 May
2	17 May	18 May	22 May	2 June
3	24 May	26 May	30 May	8 June
4	31 May	2 June	5 June	16 June
5	7 June	9 June	12 June	23 June

Table 4. Analysis of variance (ANOVA) for main treatment factors, timing of spring seeding (T), seed entry (S), and their interaction on turfgrass cover at O.J. Noer Research Center, WI, 2023.

Treatment	Turfgrass cover (%)[†]				
	12 May	18 May	26 May	2 June	9 June
Timing of spring seeding (T)	*[‡]	**	NS	NS	NS
Seed entry (S)	***	***	***	***	***
TxS	**	*	NS	NS	NS

[†] Turfgrass cover using digital image analysis for 5 weeks during the establishment period

[‡] NS = not significant ($P > 0.05$); * = significant ($P \leq 0.05$); ** = significant ($P \leq 0.01$); *** = significant ($P \leq 0.001$)

Table 5. Analysis of variance (ANOVA) for main treatment factors, timing of spring seeding (T), seed entry (S), and their interaction on turfgrass cover at Hancock Turfgrass Research Center, East Lansing, MI, 2023.

Treatment	Turfgrass cover (%)[†]				
	10 May	17 May	24 May	31 May	7 June
Timing of spring seeding (T)	**[‡]	NS	NS	NS	NS
Seed entry (S)	***	***	***	***	***
TxS	***	*	NS	NS	NS

[†] Turfgrass cover using digital image analysis for 5 weeks during the establishment period

[‡] NS = not significant ($P > 0.05$); * = significant ($P \leq 0.05$); ** = significant ($P \leq 0.01$); *** = significant ($P \leq 0.001$)

Table 6. Analysis of variance (ANOVA) for main treatment factors, timing of spring seeding (T), seed entry (S), and their interaction on turfgrass cover at Turfgrass Research, Outreach, and Education Center (TROE), St. Paul, MN, 2023.

Treatment	Turfgrass cover (%)[†]				
	26 May	2 June	16 June	24 June	30 June
Timing of spring seeding (T)	NS[‡]	NS	NS	NS	NS
Seed entry (S)	***	***	***	NS	**
TxS	*	*	NS	NS	NS

[†] Turfgrass cover using digital image analysis for 5 weeks during the establishment period

[‡] NS = not significant ($P > 0.05$); * = significant ($P \leq 0.05$); ** = significant ($P \leq 0.01$); *** = significant ($P \leq 0.001$)

Table 7. The interaction of timing of spring seeding and seed entry on percent turfgrass cover on 12 May at O.J. Noer Research Center, Verona, WI, 2023.

Seed entry (S)	Turfgrass cover (%)[†]		
	Timing of spring seeding (T)		
	Early	Middle	Late
Pure Distinction + Two-Putt	18.1 a[‡]A[§]	9.3 aB	15.7 aAB
Pure Distinction	17.6 aA	6.2 bB	8.7 bB
Penncross	12.5 bA	6.4 abB	9.2 bAB
Penn A-4	8.6 bcA	5.9 bcA	6.5 bcA
Two-Putt	5.9 cdA	2.9 cdB	4.0 cdAB
Declaration	1.8 deA	1.8 dA	0.8 dA
Control	0.8 eA	0.9 dA	1.5 dA

[†] Turfgrass cover using digital image analysis for 5 weeks during the establishment period

[‡] Means followed by the same lowercase letter within the columns are not significantly different according to Fisher's Protected LSD (*P*=0.05)

[§] Means followed by the same uppercase letter within the rows are not significantly different according to Fisher's Protected LSD (*P*=0.05)

Table 8. The interaction of timing of spring seeding and seed entry on percent turfgrass cover on 18 May at O.J. Noer Research Center, Verona, WI, 2023.

Seed entry (S)	Turfgrass cover (%)[†]		
	Timing of spring seeding (T)		
	Early	Middle	Late
Pure Distinction + Two-Putt	29.1 a[‡]A[§]	19.7 aB	25.6 aAB
Pure Distinction	25.7 abA	13.6 bB	17.2 bAB
Penncross	21.2 bA	14.8 abB	17.9 bAB
Penn A-4	15.2 cA	13.5 bA	12.9 bcA
Two-Putt	12.3 cA	7.4 cB	9.2 cAB
Declaration	4.0 dA	3.2 cA	1.9 dA
Control	1.5 dA	2.6 cA	3.0 dA

[†] Turfgrass cover using digital image analysis for 5 weeks during the establishment period

[‡] Means followed by the same lowercase letter within the columns are not significantly different according to Fisher's Protected LSD (*P*=0.05)

[§] Means followed by the same uppercase letter within the rows are not significantly different according to Fisher's Protected LSD (*P*=0.05)

Table 9. The interaction of timing of spring seeding and seed entry on percent turfgrass cover on 10 May at Hancock Turfgrass Research Center, 2023.

Seed entry (S)	Turfgrass cover (%)[†]		
	Timing of spring seeding (T)		
	Early	Middle	Late
Two-Putt	11.3 a[‡]A[§]	5.8 abB	3.3 aB
Pure Distinction + Two-Putt	7.4 bA	6.1 aA	3.4 aB
Penncross	6.3 bcA	4.8 abAB	3.1 aB
Pure Distinction	5.2 cA	5.5 abA	3.2 aB
Declaration	5.1 cA	4.2 abAB	3.0 aB
Penn A-4	5.1 cA	5.1 abA	3.7 aA
Control	4.5 cA	3.8 bAB	3.2 aB

[†] Turfgrass cover using digital image analysis for 5 weeks during the establishment period

[‡] Means followed by the same lowercase letter within the columns are not significantly different according to Fisher's Protected LSD (*P*=0.05)

[§] Means followed by the same uppercase letter within the rows are not significantly different according to Fisher's Protected LSD (*P*=0.05)

Table 10. The interaction of timing of spring seeding and seed entry on percent turfgrass cover on 17 May at Hancock Turfgrass Research Center, 2023.

Seed entry (S)	Turfgrass cover (%)[†]		
	Timing of spring seeding (T)		
	Early	Middle	Late
Two-Putt	19.0 a[‡]A[§]	11.6 aB	11.2 aB
Pure Distinction + Two-Putt	13.9 abA	11.1 aA	10.2 abA
Penncross	10.8 bcA	10.9 aA	8.8 abcA
Pure Distinction	8.8 bcAB	10.3 abA	6.2 bcB
Declaration	7.7 cA	7.2 bcA	5.1 cA
Penn A-4	11.0 bcA	11.5 aA	10.9 aA
Control	6.8 cA	5.3 cB	4.8 cB

[†] Turfgrass cover using digital image analysis for 5 weeks during the establishment period

[‡] Means followed by the same lowercase letter within the columns are not significantly different according to Fisher's Protected LSD (*P*=0.05)

[§] Means followed by the same uppercase letter within the rows are not significantly different according to Fisher's Protected LSD (*P*=0.05)

Table 11. The interaction of timing of spring seeding and seed entry on percent turfgrass cover on 26 May at Turfgrass Research, Outreach, and Education Center (TROE), St. Paul, MN, 2023.

Seed entry (S)	Turfgrass cover (%)[†]		
	Timing of spring seeding (T)		
	Early	Middle	Late
Pure Distinction + Two-Putt	85.5 a[‡]A[§]	72.8 aA	77.1 aA
Pure Distinction	74.8 abA	60.1 abA	71.5 abA
Penncross	67.5 bA	59.4 abA	64.8 abA
Penn A-4	74.7 abA	64.0 abA	61.4 bA
Two-Putt	70.3 abA	49.4 bcB	44.5 cB
Declaration	44.7 cA	39.4 cdAB	25.6 dB
Control	28.7 dA	24.0 dA	16.7 dA

[†] Turfgrass cover using digital image analysis for 5 weeks during the establishment period

[‡] Means followed by the same lowercase letter within the columns are not significantly different according to Fisher's Protected LSD (*P*=0.05)

[§] Means followed by the same uppercase letter within the rows are not significantly different according to Fisher's Protected LSD (*P*=0.05)

Table 12. The interaction of timing of spring seeding and seed entry on percent turfgrass cover on 2 June at Turfgrass Research, Outreach, and Education Center (TROE), St. Paul, MN, 2023.

Seed entry (S)	Turfgrass cover (%)[†]		
	Timing of spring seeding (T)		
	Early	Middle	Late
Pure Distinction + Two-Putt	97.3 a[‡]A[§]	91.3 aA	92.1 aA
Pure Distinction	93.4 aA	84.3 aA	90.7 aA
Penncross	85.5 aA	77.0 abA	83.6 abA
Penn A-4	96.1 aA	91.7 aA	84.3 abA
Two-Putt	92.2 aA	78.8 abAB	70.3 bB
Declaration	87.1 aA	78.7 abA	48.9 cB
Control	64.9 bA	59.3 bAB	32.6 cB

[†] Turfgrass cover using digital image analysis for 5 weeks during the establishment period

[‡] Means followed by the same lowercase letter within the columns are not significantly different according to Fisher's Protected LSD (*P*=0.05)

[§] Means followed by the same uppercase letter within the rows are not significantly different according to Fisher's Protected LSD (*P*=0.05)

Table 13. The effect of timing of spring seeding on turfgrass cover measured 5 consecutive weeks during establishment at O.J. Noer Research Center, Verona, WI, 2023.

Timing of spring seeding (T)	Turfgrass cover (%)[†]				
	12 May	18 May	26 May	02 June	9 June
Early	9.3 a[‡]	15.6 a	30.8	45.3	59.7
Middle	4.8 c	10.7 b	29.2	44.3	62.4
Late	6.6 b	12.5 b	27.8	42.8	60.3

[†] Turfgrass cover using digital image analysis for 5 weeks during the establishment period

[‡] Means followed by the same letter within the same column are not significantly different according to Fisher's Protected LSD (*P*=0.05)

Table 14. The effect of spring seeding timing on percent cover measured 5 consecutive weeks during establishment at Hancock Turfgrass Research Center, East Lansing, MI, 2023.

Timing of spring seeding (T)	Turfgrass cover (%)[†]				
	10 May	17 May	24 May	31 May	7 June
Early	6.4 a[‡]	11.1	16.4	18.4	31.4
Middle	5.0 a	8.2	16.3	15.8	28.3
Late	3.3 b	9.7	14.0	18.1	26.7

[†] Turfgrass cover using digital image analysis for 5 weeks during the establishment period

[‡] Means followed by the same letter within the same column are not significantly different according to Fisher's Protected LSD (*P*=0.05)

Table 15. Analysis of variance (ANOVA) for main treatment factors, timing of spring seeding (T), seed entry (S), and their interaction on turfgrass cover at Horticulture Research Station, IA, 2023.

Treatment	Turfgrass cover (%)[†]				
	15 May	22 May	30 May	5 June	12 June
Timing of spring seeding (T)	*[‡]	NS	NS	NS	NS
Seed entry (S)	NS	NS	NS	NS	NS
TxS	NS	NS	NS	NS	NS

[†] Turfgrass cover using digital image analysis for 5 weeks during the establishment period

[‡] NS = not significant ($P > 0.05$); * = significant ($P \leq 0.05$); ** = significant ($P \leq 0.01$); *** = significant ($P \leq 0.001$)

Table 16. The effect of spring seeding timing on percent cover measured 5 consecutive weeks during establishment at Horticulture Research Station, IA, 2023.

Timing of spring seeding (T)	Turfgrass cover (%)[†]				
	15 May	22 May	30 May	5 June	12 June
Early	16.4 b[‡]	30.6	59.5	80.8	91.6
Middle	25.9 a	33.6	64.1	84.8	94.8
Late	23.3 ab	37.6	67.6	90.2	97.3

[†] Turfgrass cover using digital image analysis for 5 weeks during the establishment period

[‡] Means followed by the same letter within the same column are not significantly different according to Fisher's Protected LSD (*P*=0.05)

Table 17. The effect of seed entry on percent cover measured 5 consecutive weeks during establishment at Hancock Turfgrass Research Center, East Lansing, MI, 2023.

Seed entry (S)	Turfgrass cover (%)[†]				
	10 May	17 May	24 May	31 May	07 June
Two-Putt	6.8 a[‡]	13.9 a	21.8 a	25.2 a	35.9 a
Pure Distinction + Two-Putt	5.6 b	11.7 b	20.2 a	23.1 a	37.4 a
Penn A-4	4.6 cd	11.1 b	18.8 a	21.9 a	35.9 a
Penncross	4.7 c	10.2 bc	18.3 ab	21.0 ab	35.8 a
Pure Distinction	4.7 c	8.5 cd	15.0 b	16.5 b	32.9 a
Declaration	4.1 cd	6.7 de	9.0 c	9.6 c	17.1 b
Control	3.8 d	5.6 e	5.8 c	4.9 c	6.6 c

[†] Turfgrass cover using digital image analysis for 5 weeks during the establishment period

[‡] Means followed by the same letter are not significantly different according to Fisher's Protected LSD (*P*=0.05)

Table 18. The effect of seed entry on percent cover measured 5 consecutive weeks during establishment at O.J. Noer Research Center, Verona, WI, 2023.

Seed entry (S)	Turfgrass cover (%)[†]				
	12 May	18 May	26 May	02 June	09 June
Pure Distinction + Two-Putt	14.4 a[‡]	24.8 a	42.3 a	53.8 a	65.9 a
Pure Distinction	10.8 b	18.8 b	37.2 b	49.4 a	66.3 a
Penncross	9.4 b	18.0 b	37.1 b	51.8 a	70.2 a
Penn A-4	7.0 c	13.8 c	32.3 c	43.7 b	60.4 b
Two-Putt	4.3 d	9.6 d	27.9 d	40.9 bc	54.2 cd
Declaration	1.5 e	3.0 e	16.0 e	37.7 c	57.4 bc
Control	1.0 e	2.4 e	12.0 f	31.7 d	51.2 d

[†] Turfgrass cover using digital image analysis for 5 weeks during the establishment period

[‡] Means followed by the same letter are not significantly different according to Fisher's Protected LSD (*P*=0.05)

Table 19. The effect of seed entry on percent turfgrass cover measured 5 consecutive weeks during establishment at Turfgrass Research, Outreach, and Education Center (TROE), St. Paul, MN, 2023.

Seed entry (S)	Turfgrass cover (%)[†]				
	26 May	02 June	16 June	24 June	30 June
Pure Distinction + Two-Putt	78.5 a[‡]	93.5 a	99.1 a	98.6	99.3 a
Pure Distinction	68.8 b	89.5 abc	98.1 a	92.6	99.1 a
Penncross	63.9 b	82.0 bc	98.9 a	96.8	97.5 ab
Penn A-4	66.7 b	90.7 a	99.5 a	99.3	99.5 a
Two-Putt	54.8 c	80.4 cd	96.6 a	94.5	95.1 b
Declaration	36.6 d	71.9 d	96.9 a	92.9	98.0 a
Control	23.2 e	52.3 e	87.0 b	91.3	94.1 b

[†] Turfgrass cover using digital image analysis for 5 weeks during the establishment period

[‡] Means followed by the same letter within the same column are not significantly different according to Fisher's Protected LSD (*P*=0.05)

Table 20. Germination percentage of each seed entry, 2023.

Seed Entry	Germination (%)[†]
Penn A-4	89.9a[‡]
Pure Distinction	85.9a
Penncross	84.0a
Two-Putt	82.4ab
Declaration	70.5b

[†] Germination percentage following AOSA Rules for Testing Seeds (2016)

[‡] Means followed by the same letter within the same column are not significantly different according to Fisher's Protected LSD (*P*=0.05)

Figure 1. Daily average soil temperature, average air temperature, and total daily rainfall at the Hancock Turfgrass Research Center in East Lansing, MI from 7 April to 15 June 2023. Black filled-in triangles indicated the three spring seeding dates.

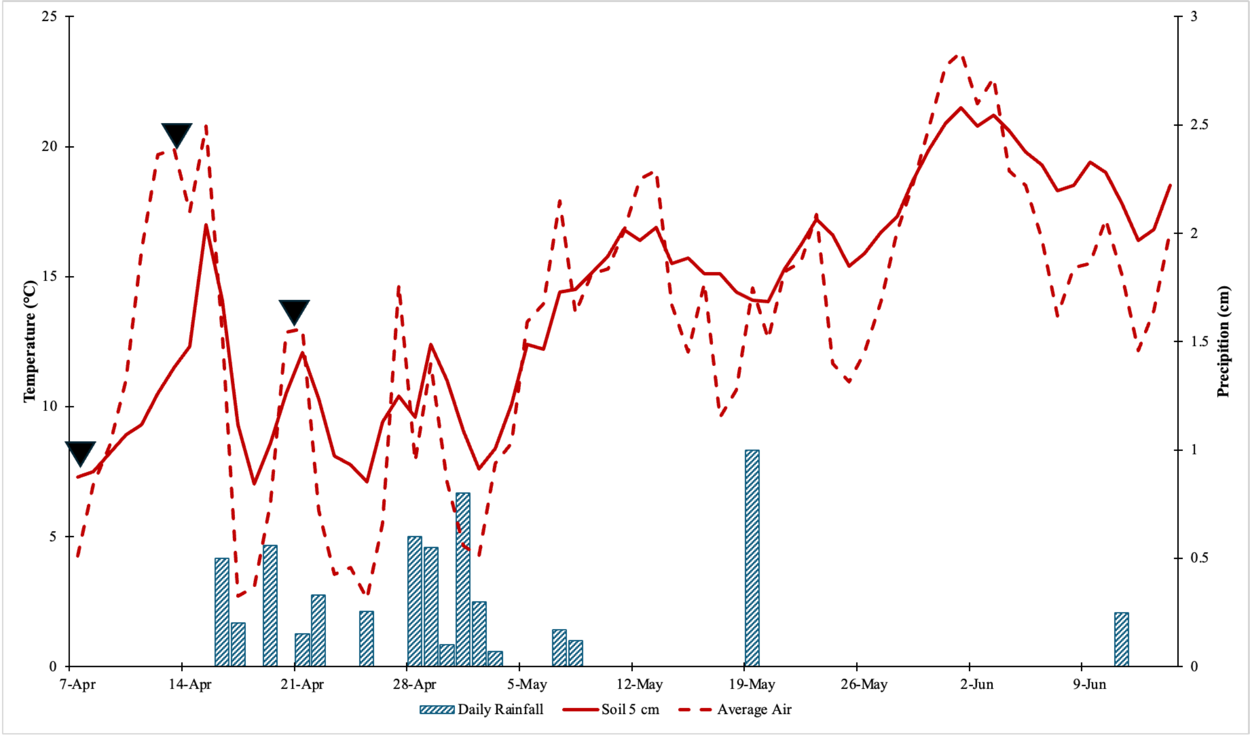


Figure 2. Daily average soil temperature, air temperature, and total daily rainfall at the O.J. Noer Research Station, Verona, WI from 13 April to 9 June 2023. Black filled-in triangles indicate when spring seeding occurred.

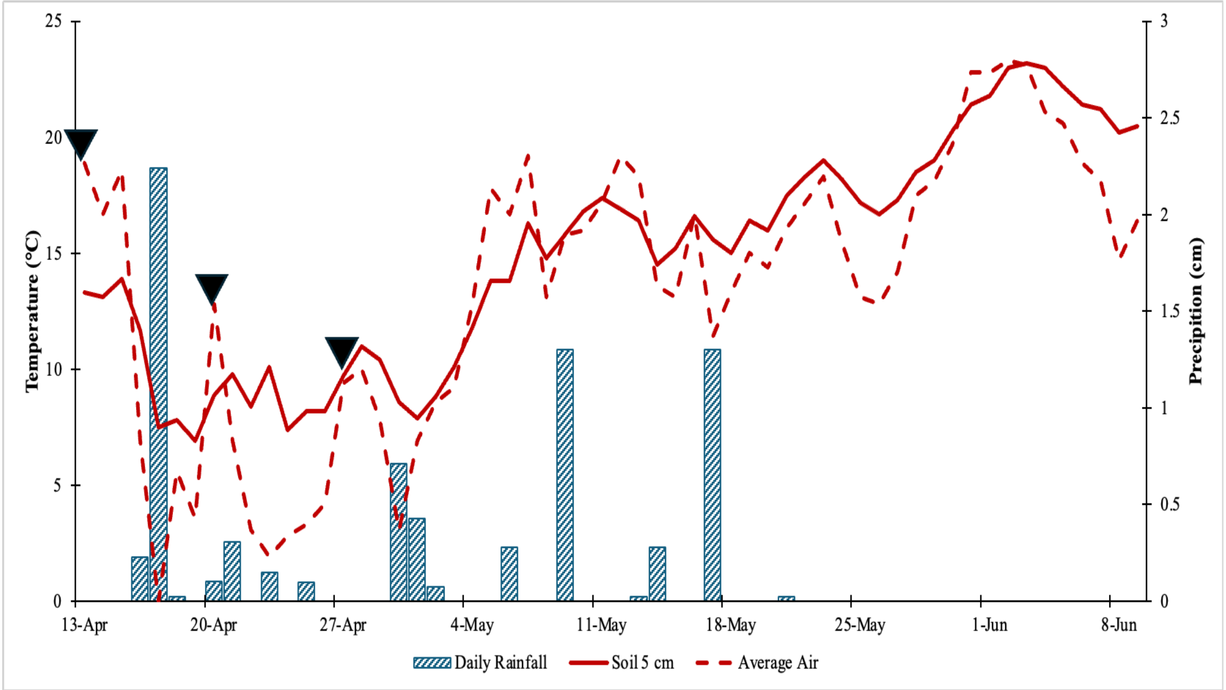


Figure 3. Daily average soil temperature, air temperature, and total daily rainfall at the Turfgrass Research, Outreach, and Education Center, St. Paul, MN from 11 April to 30 June 2023. Black filled-in triangles indicate when spring seeding occurred.

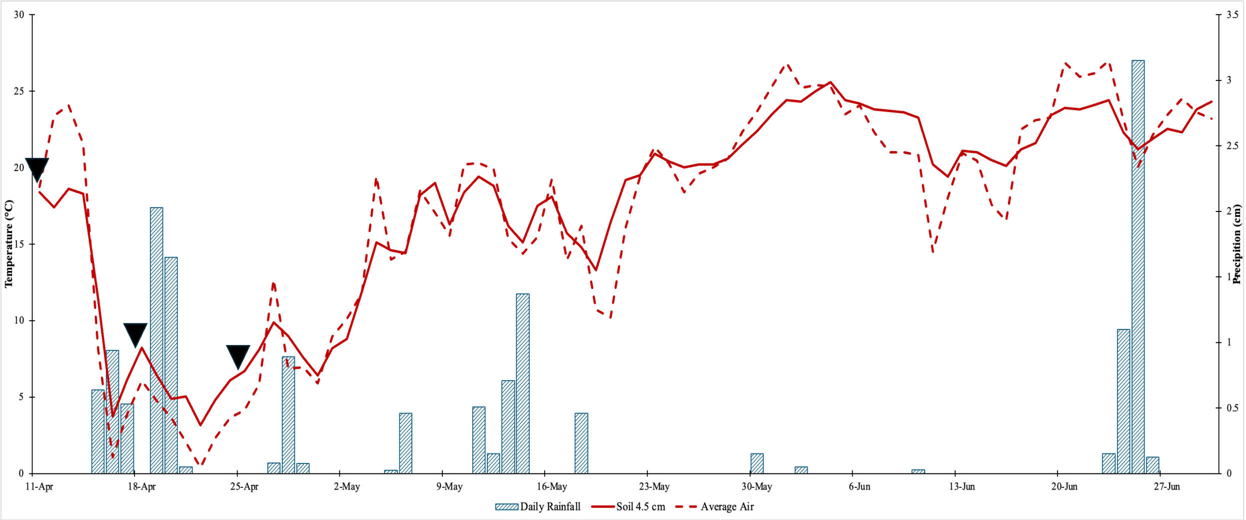
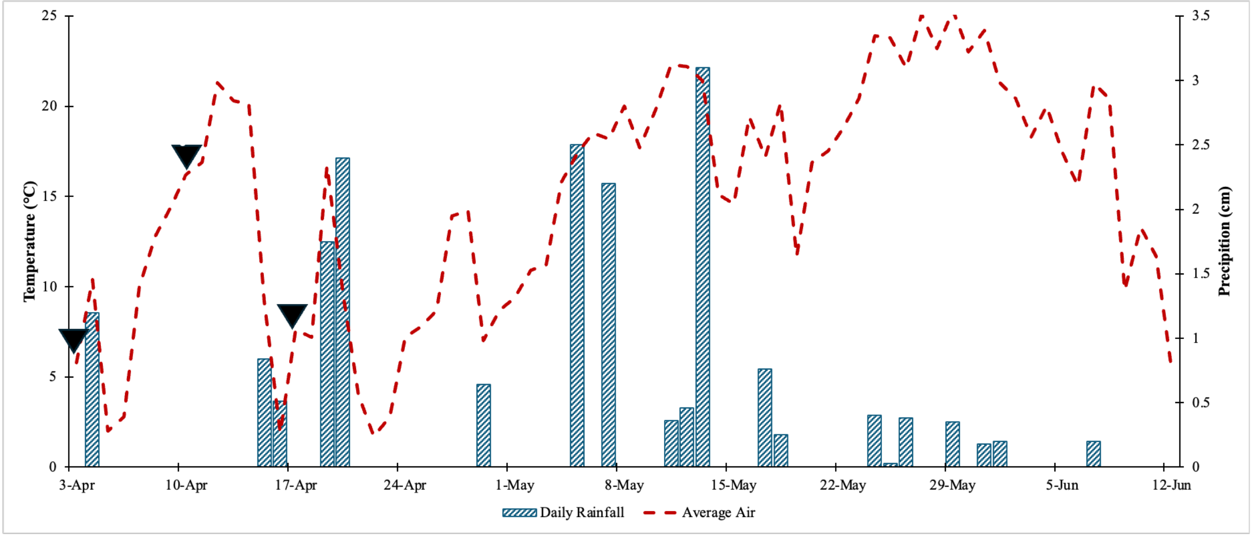


Figure 4. Daily average air temperature and total daily rainfall at the Horticulture Research Station, Ames, Iowa from 3 April to 12 June 2023. Black filled-in triangles indicate when spring seeding occurred.



CHAPTER 3: SPRING ESTABLISHMENT OF INTERSEEDED CREEPING BENTGRASS AND ANNUAL BLUEGRASS FOLLOWING SIMULATED WINTERKILL ON AN ANNUAL BLUEGRASS PUTTING GREEN

ABSTRACT

One common method golf course superintendents use when re-establishing putting greens following winterkill is to verti-cut and apply seed. Very little information is available on when spring seeding should occur and what type of seed should be selected in the field. The objective of this two-year study was to evaluate spring establishment rate of creeping bentgrass (*Agrostis stolonifera* L.), annual bluegrass (*Poa annua* var. Reptans (Hauskn.)), and a 50/50 by weight mixture of creeping bentgrass and annual bluegrass, seeded three consecutive weeks in the spring. Seeding treatments included four creeping bentgrass cultivars, 'Penncross', 'Pure Distinction', 'Declaration', 'Penn A-4', and one annual bluegrass, 'Two-Putt', and a mixture of Pure Distinction and Two-Putt, and an unseeded control. The experimental design was a randomized complete block split-plot design with 4 replications. The whole plot was spring seeding timing, and the subplot was seed entry. Plots were verti-cut in two directions and seeding occurred at rate of 49 kg seed ha^{-1} three consecutive weeks once soil temperature reached 7°C at 5 cm. Starter fertilizer and sand topdressing was applied immediately following seeding. Data was collected for five weeks during the establishment period with digital image analysis to assess percent turfgrass cover. Statistical analysis was conducted separately for each year. The timing of spring seeding was significant at the beginning of the study on two measurement dates in 2023 and one measurement date in 2024. By the end of the study, timing of seeding was not significant, and all treatments had above 90% turfgrass cover. Seed entry was significant at all but one measurement date through the two-year study. Penn A-4 and Penncross had some of the

highest turfgrass cover out of all the seed entries, but differences were fairly small and numerically insignificant.

MATERIALS AND METHODS

Research was conducted in 2023 and 2024 on an annual bluegrass [*Poa annua* (L.)] native soil putting green at Michigan State University, Hancock Turfgrass Research Center (HTRC), East Lansing, MI. The putting green was grown on Colwood-Brookston loams (Fine-loamy, mixed, active and superactive, mesic Typic Endoaquolls and Argiaquolls) that had been sand capped with years of light frequent topdressing to a depth of 4 cm. The particle size analysis of the native soil putting green below the sand topdressing layer was 75% sand, 16% silt, 9% clay with 2.7% organic matter and 7.1 pH. Mowing was conducted three times per week at 0.31 cm with a triplex reel mower (Greensmaster 3150-Q, The Toro Company, Bloomington, MN). The total area of the site was 256 m^2. In 2023, 128 m^2 of the area was used for the experiment and in 2024 the other 128 m^2 of the putting green was used to reduce treatment effects from creeping bentgrass that had been established the prior year.

The experimental design was a randomized complete block split-plot design with four replications. The whole plot (timing of spring seeding) included three levels: early, middle, late spring seeding date. The subplot (seed entry) included seven treatments: ‘Penn A-4’, ‘Declaration’, ‘Penncross’, ‘Pure Distinction’, ‘Two-Putt’, ‘Pure Distinction’ and ‘Two-Putt’ 50/50 by weight mix, and an unseeded control. The same seed lot was used for each seed entry in the 2023 and 2024 trial years and seed was harvested in 2019 and 2020. Each putting green was divided into twelve 20 m^2 whole plots and each whole plot was split into eight 0.8 m^2 subplots.

To simulate winterkill, glyphosate and diquat dibromide [Roundup QuikPRO; Bayer Environmental Science, Cary, NC; (N-(phosphonomethyl) glycine, in the form of its ammonium salt), (6,7-dihydrodipyrido (1,2-a:2’,1’-c pyrazinediium dibromide)) was applied with a CO_2 pressurized sprayer calibrated to deliver 6.7 kg a.i. ha^{-1} and 2.7 kg a.i. ha^{-1}, respectively. Before the 2023 trial, herbicide was applied on 30 March 2023 and 12 April 2023 and before the 2024 trial, herbicide was applied 18 October 2023 and 15 March 2024. After the herbicide applications there was no visible green turfgrass tissue on the putting green. Before seeding, plots were verti-cut in two directions with a walk mower (GREENSMASTER FLEX 2100, The Toro Company, Bloomington, MN) with a verticutter attachment (TRUE-SURFACE, Moscow Mills, MO) at a depth of 0.4 cm. The organic material removed from verticutting was blown off the putting surface using a backpack leaf blower (STIHL BR 600 238 mph 677 CFM Gas Backpack Leaf Blower, STIHL, Virginia Beach, VA). When spring soil temperatures reached a daily average of 7°C at a 5 cm depth, seeding was initiated. Seed treatments were weighed out individually and applied to the putting green with hand shaker containers at 49 kg ha^{-1}. Starter fertilizer (10 N-8P-8K) was applied with hand shaker containers to deliver 21.5 kg P ha^{-1}. Sand topdressing was applied over the plots to maximize soil to seed contact at a rate of 5,612 kg sand ha^{-1}. The seeding process was repeated every seven days for three weeks in 2023 and in 2024. In 2023 seeding occurred on 14 April, 21 April, and 28 April and in 2024 on 8 April, 15 April, and 22 April.

Starting on 22 May 2023, and 17 May 2024, plots were mowed with a reel-mower three times a week starting at 0.635 cm and gradually dropped to 0.31 cm (GREENSMASTER FLEX 2100, The Toro Company, Bloomington, MN). Overhead irrigation was applied when necessary to provide adequate germination and establishment, generally this averaged about 0.64 cm per day (Rain Bird 750 series, Rain Bird, Azusa, CA). Granular urea (46 N-0P-0K) was applied bi-weekly with hand shaker bottles for eight weeks after the first seedling emergence at a rate of 9.7 kg N ha^{-1} and was immediately watered in with 0.64 cm of irrigation. Two applications of tebuconazole (Torque; Nufarm; Alsip, IL, α-[2-(4-chlorophenyl) ethyl]-α-1,1-dimethylethyl)-1H-

1,2,4-triazole-1-ethanol) was applied in 2024 with a CO_2 pressurized sprayer calibrated to deliver 1.91 L ha^{-1} to prevent summer patch (*Magnaporthe poae*). Precipitation, soil temperature, and air temperature data was collected daily by an on-site weather station.

Data collection

Turfgrass cover was assessed with digital image analysis (DIA) weekly beginning on 23 May 2023 and 2 May 2024. Using a light box, single images captured from the center of the plot with a SONY RX100 VI (Sony Corporation of America, New York, NY) with fixed camera settings (manual, focal length= 26 mm, F-stop=4.5, exposure= 1/200, ISO: 800). Images were then analyzed to assess turfgrass cover using ImageJ (National Institute of Health, USA) (Karcher and Richardson, 2013) and was configured to quantify green turfgrass cover based on the number of green pixels in the image (HUE: 32-135 SAT: 0-255 and BRIGHTNESS: 8-255).

Statistical analysis

Data from each year was analyzed separately through PROC GLIMMIX using SAS software (version 9.4; SAS Institute Inc., Cary, NC). Block was treated as a random effect and timing of spring seeding was nested within each block. Analysis of Variance (ANOVA) was conducted on the fixed effects of timing of spring seeding and seed entry to determine significant differences among treatment means ($P \leq 0.05$). Fisher's Least Significance Difference (LSD) were used when significant differences between treatments occurred.

RESULTS AND DISCUSSION

Timing of spring seeding x seed entry interaction

The timing of spring seeding x seed entry interaction for turfgrass cover was only significant on one date in two years of the research, 28 May 2024 (Table 22). On this date, Pure Distinction and Two-Putt mix, Pure Distinction, Penncross, Penn A-4, and Declaration treatments were not statistically different from each other within each timing of spring seeding (Table 23). Two-Putt and the control treatments had the lowest turfgrass cover of seed entries at all three timing of seeding treatments but averaged 89% or greater turfgrass cover. The timing of spring seeding of Declaration was significantly different. The early spring seeding averaged the lowest turfgrass cover with 94.1% and was statistically different from the late spring seeding of 98.1%. Although there was statistical significance in the interaction between timing of spring seeding and seed entry, differences in percent turfgrass cover among treatments were small and lack practical significance.

Timing of spring seeding

The main effect of timing of spring seeding was significant on 30 May and 6 June in 2023 (Table 21). On 30 May, the early spring seeding date had the highest percent turfgrass cover with 59.7% compared to the middle and late spring seeding dates which both had 44% (Table 24). On 6 June, the early spring seeding date had 85.3% turfgrass cover and was statistically different from the middle and late seeding dates which had 78.9% and 76.9%, respectively. By 13 June, there were no differences, and all three treatments had 92% or 93% turfgrass cover.

In 2024, timing of spring seeding was significant on 2 May (Table 22). The early and middle spring seeding dates had a statistically higher percent turfgrass cover with 36.6% and

34.7% compared to the late spring seeding date that had 24.0% turfgrass cover (Table 25). The 2 May measurement date was the only date in 2024 that was statistically significant and by 28 May, all three treatments had greater than 94% turfgrass cover.

In 2023 and 2024 timing of spring seeding was significant at the beginning of the establishment period but by the end there were no differences among treatments. The differences between the timing of spring seeding treatments in the first few measurement dates in the study would likely not re-open a golf course earlier. However, there was no evidence to suggest seeding earlier in the spring had a negative impact on re-establishment.

Seed entry main effect

Seed entry main effect was significant on all five measurement dates in 2023 (Table 21). On 23 May, 30 May, and 6 June, Penn A-4, and Penncross had the highest percent turfgrass cover among all seed entry treatments (Table 26). On 13 June, Penn A-4 had the highest percent turfgrass cover but was not statistically different from Penncross and Pure Distinction. On 20 June, Penn A-4 had the highest percent turfgrass cover with 96.9% but was not statistically different from Penncross, Pure Distinction, or Pure Distinction and Two-Putt mix. Although there were statistical differences among treatments, by 20 June every seed entry had greater than 93% turfgrass cover.

In 2024, the main effect of seed entry was significant on four of the five measurement dates (Table 22). On the first measurement date, 2 May, there was no statistical difference between seed entry treatments (Table 27). On the next four measurement dates, Penn A-4 had the highest percent turfgrass cover but was not statistically different from any of the other creeping bentgrass entries. Two-Putt consistently had the lowest percent turfgrass cover out all the seed entries. These results are similar to Frank et al. (2017) who evaluated spring establishment of three CBG cultivars: Penn A-4, ‘Alpha’, and ‘Providence’ and annual bluegrass florets on putting greens follow simulated winterkill. In this study, researchers found that all creeping bentgrass cultivars had a higher percent turfgrass cover compared to the annual bluegrass florets and that the differences between cultivars was low or insignificant, depending on measurement date (Frank et al., 2017).

The lack of significance between CBG seed entries in 2024 could be attributed to warmer soil temperatures. In 2023, daily average soil temperatures at a 5 cm depth fell below 10°C multiple times during the first month of the trial (Figure 5) whereas in 2024, daily average soil temperatures were consistently at or above 10°C following study initiation (Figure 6). Both growth chamber and field studies have concluded that most CBG cultivars can germinate and emerge at approximately 10°C and even small increases of 1.0°C can have a significant effect on turfgrass cover (Carroll et al., 2020; Ebdon and DaCosta 2021). Heineck et al. (2019) found greater contrasts in germination between CBG cultivars at average temperatures of 7°C compared to 10°C. Warmer soil temperatures could have been the reason for the lack of statistical differences between CBG seed entries in 2024.

By the end of the study in 2023 and 2024, the control plots averaged 93% turfgrass cover despite not having any seed applied to it. It was observed that annual bluegrass had emerged and established in these plots each year. This could be due to annual bluegrass seed germinating from the soil seed bank and filling in these bare areas. It is also possible that some of the pre-existing turfgrass was not completely killed by the non-selective herbicide applications and started to recover during the study. Similar results were observed by Michael (2016) in a three-year study in Nebraska that was evaluating re-establishment of ‘L-93’ creeping bentgrass following winterkill. At 10 weeks after the first seedling emergence, the non-seeded controls recovered

84.7% turfgrass where ‘L-93’ seeded treatment had 92.4% turfgrass cover. The researchers hypothesized that the large portion of turfgrass in the non-seeded controls was because the non-selective herbicides did not get completely kill the pre-existing turfgrass and that natural regeneration of bentgrass occurred (Michael, 2016). Although both Michael (2016) and the present study found that some turfgrass regenerates following non-selective herbicide applications, this is representative of natural winterkill with some of the turfgrass being able to recover in the spring.

CONCLUSIONS

Timing of spring seeding was significant for two measurement dates in the 2023 trial and for one measurement date in the 2024 trial. When there was significance in the timing of spring seeding, the early spring seeding date had the highest percent turfgrass cover. In 2023, the highest percent turfgrass cover was the early spring seeding date and was significantly different from both the middle and late spring seeding dates and in 2024 the early spring seeding date was significantly different from the late spring seeding date. The differences between the timing of spring seeding treatments in the first few measurement dates in the study would likely not re-open a golf course earlier. However, there was no evidence to suggest seeding earlier in the spring had a negative impact on re-establishment.

Seed entry was significant at all five measurement dates in 2023 and four out of five measurement dates in 2024. In 2023, Penn A-4 and Penncross had the highest percent turfgrass cover at the beginning part of the study but by the end of the trial, differences among treatments were small. In 2024, there were no significant differences in turfgrass cover among creeping bentgrass cultivars which may have been due to warmer soil temperatures. In both years, all treatments, including the unseeded control, had 93% or greater turfgrass cover. These results suggest that either annual bluegrass germinated from the soil seed bank or that some of the pre-existing turfgrass stand was able to regenerate following non-selective herbicide applications. However, this is representative of natural winterkill where some turfgrass eventually recovers and superintendents should still seed following winterkill to increase rate of establishment. Future research should consider evaluating different strategies for elevating soil temperatures early in the spring during re-establishment. More work needs to be done to evaluate various covers and dark colored materials such as black sand on winterkill re-establishment.

TABLES AND FIGURES

Table 21. Analysis of variance (ANOVA) for main treatment factors, timing of spring seeding (T), seed entry (S), and their interaction on turfgrass cover at Hancock Turfgrass Research Center, East Lansing, MI, 2023.

Treatment	Turfgrass cover (%)†				
	23 May	30 May	6 June	13 June	20 June
Timing of spring seeding (T)	NS‡	**	*	NS	NS
Seed entry (S)	**	***	***	***	**
TxS	NS	NS	NS	NS	NS

†Turfgrass cover using digital image analysis for 5 weeks during the establishment period

‡NS = not significant (P > 0.05); * = significant (*P* ≤ 0.05); ** = significant (*P* ≤ 0.01); *** = significant (*P* ≤ 0.001)

Table 22. Analysis of variance (ANOVA) for main treatment factors, timing of spring seeding (T), seed entry (S), and their interaction on turfgrass cover at Hancock Turfgrass Research Center, East Lansing, MI, 2024.

Treatment	Turfgrass cover (%)†				
	2 May	7 May	14 May	21 May	28 May
Timing of spring seeding (T)	**‡	NS	NS	NS	NS
Seed entry (S)	NS	*	**	**	***
TxS	NS	NS	NS	NS	*

†Turfgrass cover using digital image analysis for 5 weeks during the establishment period

‡NS = not significant (P > 0.05); * = significant (*P* ≤ 0.05); ** = significant (*P* ≤ 0.01); *** = significant (*P* ≤ 0.001)

Table 23. The interaction of timing of spring seeding and seed entry on percent turfgrass cover on 28 May at Hancock Turfgrass Research Center, East Lansing, MI, 2024.

Seed Entry (S)	Turfgrass cover (%)†		
	Timing of spring seeding (T)		
	Early	Middle	Late
Pure Distinction + Two-Putt	95.1 ab‡A^c	97.3 aA	97.4 abA
Pure Distinction	95.7 abA	97.6 aA	98.0 aA
Penncross	95.2 abA	94.0 aA	97.9 aA
Penn A-4	98.0 aA	97.4 aA	97.2 abA
Two-Putt	90.8 bA	89.1 bA	94.4 cA
Declaration	94.1 abB	95.2 aAB	98.1 aA
Control	89.3 bB	96.6 aA	94.8 bcA

†Turfgrass cover using digital image analysis for 5 weeks during the establishment period

‡Means followed by the same lowercase letter within columns are not significantly different according to Fisher’s Protected LSD (*P*=0.05)

[c] Means followed by the same uppercase letter within rows are not significantly different according to Fisher’s Protected LSD (*P*=0.05)

Table 24. The effect of timing of spring seeding on turfgrass cover measured 5 consecutive weeks during establishment at Hancock Turfgrass Research Center, East Lansing, MI, 2023.

Timing of Spring Seeding (T)	Turfgrass cover (%)†				
	23 May	30 May	6 June	13 June	20 June
Early	33.0	59.7 a‡	85.3 a	92.5	95.6
Middle	24.9	44.3 b	78.9 b	93.1	95.8
Late	25.5	44.0 b	76.9 b	92.1	95.4

†Turfgrass cover using digital image analysis for 5 weeks during the establishment period

‡Means followed by the same letter within the same column are not significantly different according to Fisher’s Protected LSD (*P*=0.05)

Table 25. The effect of timing of spring seeding on turfgrass cover measured 5 consecutive weeks during establishment at Hancock Turfgrass Research Center, East Lansing, MI, 2024.

Timing of Spring Seeding (T)	Turfgrass cover (%)†				
	2 May	7 May	14 May	21 May	28 May
Early	36.6 a‡	56.8	73.0	90.5	94.0
Middle	34.7 a	57.7	77.6	92.0	95.3
Late	24.1 b	47.6	75.7	92.5	96.8

†Turfgrass cover using digital image analysis for 5 weeks during the establishment period

‡Means followed by the same letter within the same column are not significantly different according to Fisher’s Protected LSD (*P*=0.05)

Table 26. The effect of seed entry on percent cover measured 5 consecutive weeks during establishment at Hancock Turfgrass Research Center, East Lansing, MI, 2023.

Seed Entry (S)	Turfgrass cover (%)[†]				
	23 May	30 May	6 June	13 June	20 June
Penn A-4	32.8 a[‡]	55.5 ab	86.1 a	95.2 a	96.9 a
Penncross	31.8 a	55.8 a	85.5 ab	94.6 ab	96.9 a
Pure Distinction	27.3 b	50.0 c	81.2 bc	93.3 abc	96.0 ab
Pure Distinction + Two-Putt	27.1 b	48.5 c	78.8 c	92.5 bc	95.5 ab
Two-Putt	27.1 b	50.0 bc	80.1 c	92.0 c	94.7 bc
Declaration	26.6 b	45.5 cd	79.0 c	92.3 bc	95.9 ab
Control	21.9 c	40.1 d	71.7 d	88.1 d	93.2 c

[†] Turfgrass cover using digital image analysis for 5 weeks during the establishment period

[‡] Means followed by the same letter are not significantly different according to Fisher's Protected LSD (*P*=0.05)

Table 27. The effect of seed entry on percent cover measured 5 consecutive weeks during establishment at Hancock Turfgrass Research Center, East Lansing, MI, 2024.

Seed Entry (S)	Turfgrass cover (%)[†]				
	2 May	7 May	14 May	21 May	28 May
Penn A-4	33.7	56.2 a[‡]	78.9 a	93.8 a	97.6 a
Pure Distinction + Two-Putt	33.7	55.8 ab	78.1 a	92.6 a	96.6 a
Pure Distinction	32.0	57.1 a	77.7 a	93.5 a	97.1 a
Declaration	31.7	54.3 abc	77.9 a	92.6 a	95.8 a
Penncross	31.4	55.4 ab	74.9 ab	92.1 a	95.7 a
Two-Putt	29.7	49.2 c	69.6 c	88.1 b	91.4 c
Control	30.21	50.5 bc	71.1 bc	88.9 b	93.6 b

[†] Turfgrass cover using digital image analysis for 5 weeks during the establishment period

[‡] Means followed by the same letter within the same column are not significantly different according to Fisher's Protected LSD (*P*=0.05)

Figure 5. Daily average soil temperature, air temperature, and total daily rainfall represented by at the Hancock Turfgrass Research Center in East Lansing, MI from 14 April to 20 June 2023. Black filled-in triangles indicate the three seeding dates.

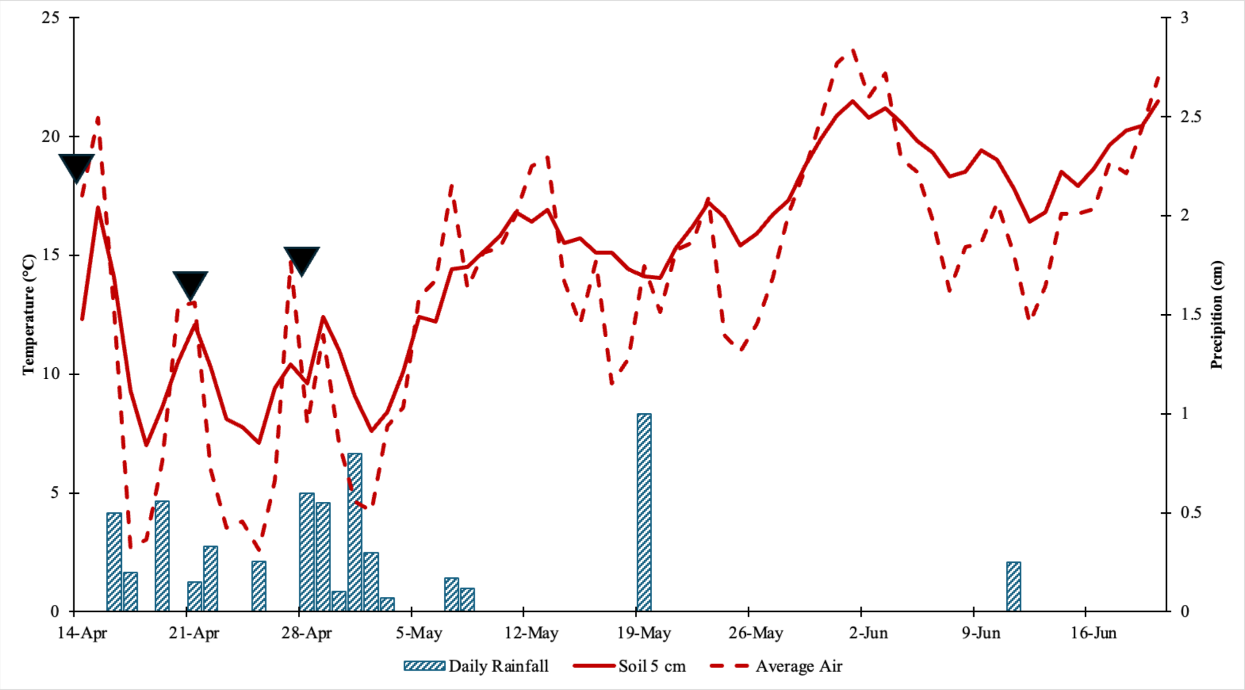
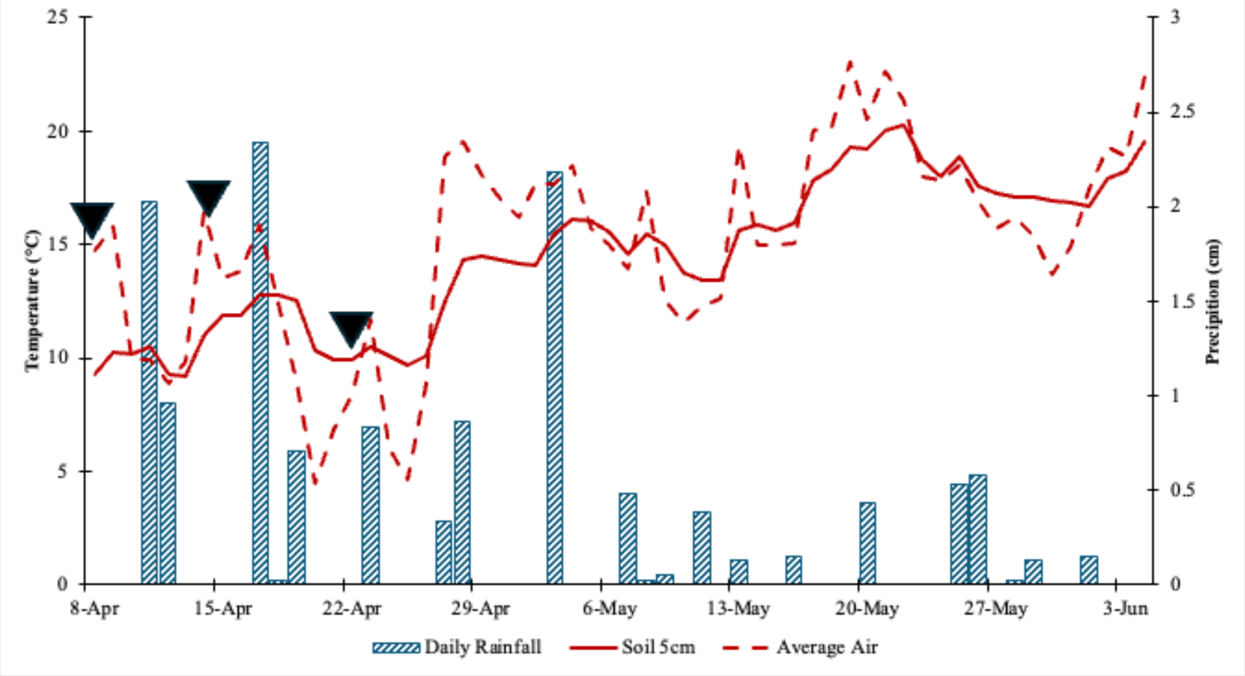


Figure 6. Daily average soil temperature, average air temperature, and total daily rainfall at the Hancock Turfgrass Research Center in East Lansing, MI from 8 April to 4 June 2024. Black filled-in triangles indicate the three seeding dates.



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APPENDIX

Table 20. Growing degree day accumulation at each timing of spring seeding treatment at each location, 2023.

Timing of Spring Seeding (T)	Growing degree days (GDD)†			
	East Lansing, MI	Verona, WI	Ames, IA	St. Paul, MN
Early	236‡	361	190	241
Middle	423	487	323	318
Late	555	558	505	359

†Growing degree day accumulation beginning on 1 March at 0°C base temperature

‡Data retrieved from GDD*Tracker* 4.0 (2024)

Table 21. The effect of seed entry on percent turfgrass cover measured 5 consecutive weeks during establishment at Horticulture Research Station, IA, 2023.

Seed Entry (S)	Turfgrass cover (%)†				
	15 May	22 May	30 May	5 June	12 June
Two-Putt	20.0	30.5	58.3	83.4	95.5
Pure Distinction + Two-Putt	25.3	33.2	62.5	86.2	96.2
Penn A-4	24.0	39.8	66.9	85.7	97.1
Penncross	24.8	35.7	66.4	86.7	96.0
Pure Distinction	18.8	30.9	62.7	80.6	89.1
Declaration	19.6	32.4	63.1	86.9	94.2
Control	20.6	35.1	66.5	87.2	93.8

†Turfgrass cover using digital image analysis for 5 weeks during the establishment period

Table 22. The effect of timing of spring seeding on turfgrass cover measured 5 consecutive weeks during establishment at Turfgrass Research, Outreach, and Education Center (TROE), St. Paul, MN, 2023.

Timing of Spring Seeding (T)	Turfgrass cover (%)†				
	26 May	2 June	16 June	24 June	30 June
Early	63.8	88.1	97.3	88.1	97.7
Middle	52.7	80.2	96.1	80.2	97.1
Late	51.7	71.9	96.4	71.9	98.0

†Turfgrass cover using digital image analysis for 5 weeks during the establishment period

Table 23. Growing degree day accumulation at each timing of spring seeding treatment at Hancock Turfgrass Research Center in 2023 and 2024.

Timing of Spring Seeding (T)	Growing degree days (GDD)[†]	
	2023	2024
Early	423[‡]	390
Middle	555	543
Late	626	666

[†] Growing degree day accumulation beginning on 1 March at 0°C base temperature

[‡] Data retrieved from GDD*Tracker* 4.0 (2024)