SCREENING AGROSTIS SPECIES AND CULTIVARS FOR GERMINATION AT LOW TEMPERATURES

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SUMMARY

Bentgrass (Agrostis spp.) is typically planted in late summer through early fall under warm soils to promote rapid germination and establishment, which coincides with germination and ingress of annual bluegrass (Poa annua L.) into turf stands. Soil temperature is often one of the major limiting factors affecting seed germination rate. Cold soils during spring months can significantly delay turfgrass establishment and cover and postpone play on damaged areas under repair. The objectives of this study were (i) conduct a growth chamber study to evaluate germination characteristics of cultivars of creeping, colonial, and velvet bentgrass in response to temperatures ranging from 50 to 77°F, and (ii) conduct a field study to evaluate germination characteristics and turfgrass cover during early spring establishment. Twelve cultivars from all three Agrostis species were evaluated in early spring by assessing grassy cover following an 8 April 2013 planting date. Soil temperatures and weekly grassy cover on 3 by 3 ft plots were measured in the field. Seeding rates for Agrostis species was applied at 0.75 pound per 1,000ft² adjusted using pure live seed. In the growth chamber, nine cultivars were assessed for their germination rate to achieve 50% germination at four temperatures (50, 59, 68, and 77°F). Curve fitting using a sigmoid model was used to calculate days to achieve 50% germination at the four controlled temperatures. In the field study, curve fitting was also used to calculate the number of days and soil temperature needed to achieve 10, 50 and 90% grassy cover. In the field study, soil temperatures ranged from 50 to 55°F during the establishment period. Agrostis species and cultivars that were able to reach 10, 50, and 90% grassy cover in the fewest number of days or under the lowest soil temperatures were associated with significantly less ingress of broadleaf weeds. Colonial bentgrass and creeping bentgrass species and cultivars were able to achieve 90% grassy cover by as much as 8 to 9 days sooner and at soil temperatures 2.5°F colder than velvet bentgrass. After emergence, some Agrostis cultivars such as Capri colonial bentgrass and 13-M, Memorial, and Penncross creeping bentgrass cultivars were statistically equal to perennial ryegrass in achieving 90% grassy cover in 20 days or less. Many of these same species and cultivars with superior germination and establishment vigor were associated with less ingress of broadleaf weeds into seeded plots. Results among species were similar between field and the growth chamber studies except when species were evaluated for germination characteristics in the growth chamber at 77°F. At this higher growth chamber temperature of 77°F, velvet bentgrass was observed to exhibit superior germination vigor than either colonial or creeping bentgrass species. This study will be repeated in 2014.

INTRODUCTION AND OBJECTIVES

Winter damage to cool-season turfgrasses used for putting green, fairway, and other fine turf is a significant concern in New England. In particular, greens and fairways comprised of considerable populations of annual bluegrass may experience as much as 70-90% turf loss from winter damage. Stand loss due to freezing injury can lead to costly re-establishment, enhanced weed pressure, and significant reduction in the function and aesthetic quality of highly managed turfgrass areas. Managers utilize a number of cultural practices to either prepare for winter injuries or to promote recovery from winter damage. As such, overseeding into damaged turf areas to rapidly reestablish turf cover is an important step in the recovery process.

Bentgrass (Agrostis spp.) is typically planted in late summer through early fall. which coincides with germination and ingress of annual bluegrass (*Poa annua* L.) into turf stands. Soil temperature is often one of the major limiting factors affecting seed germination rate, which can significantly delay turfgrass establishment and cover during spring months and postpone play on damaged areas under repair. To aid in warming soil temperatures under less favorable periods such as late winter-early spring, turfgrass managers can use geotextile covers or light topdressing of black sand to improve germination rate and renovation success (Golob et al., 2010). In addition, selecting cultivars with rapid germination rates at lower soil temperatures can also serve as an additional option to enhance overseeding success during late winter and early spring periods that are less favorable to the ingress of annual bluegrass. Germination temperatures for cool-season grasses range anywhere from 60 to 85°F (Beard, 1973). This wide range in temperature can be attributed to different temperature requirements among species and cultivars. Research specific to identifying species and cultivars responses to varying temperatures including favorable and unfavorable and their correlation to actual field establishment success have not been conducted. Accordingly, this research is the basis of our study. The specific objectives were as follows:

- (i) Conduct a growth chamber study to evaluate germination characteristics of cultivars of creeping bentgrass (*Agrostis stolonifera* L.), colonial bentgrass (*Agrostis capillaris* Huds.), and velvet bentgrass (*Agrostis canina* L.) in response to temperatures ranging from 50 to 77°F.
- (ii) Conduct a field study to evaluate germination characteristics and turfgrass cover during early spring establishment.

METHODS

Growth Chamber Study

Nine *Agrostis* cultivars including four cultivars of creeping bentgrass (007, A-4, Penncross, and T-1), three colonial bentgrass (Capri, Greentime, and Tiger II), and two velvet bentgrass cultivars (Legendary and Villa) were selected based on commercial availability and use in the turfgrass industry.

Seed was surface sterilized using 95% ethanol and 2% sodium hypochlorite solution, and then stored at 41°F prior to germination tests. Sterilized seed was placed

into petri dishes containing moistened filter paper. Seeds (50 total) per petri dish for each cultivar, and a total of 2 petri dishes per cultivar as sub-samples were used. Petri dishes were placed into a growth chamber under an 8 h photoperiod and low light levels (approximately 60 PAR). Distilled water was applied as necessary to prevent filter paper and seed from drying out, and dishes were rotated to minimize any chamber effects. The selected germination temperatures included 50, 59, 68, and 77°F. Germination was recorded daily for up to 21 days or until maximum germination percentage was reached and no additional germination was observed. Seed germination was defined as a 1 mm emerged shoot, as visible under a microscope (McCarty and Dudeck, 1993).

Three replicates for each cultivar was used to calculate different germination parameters including days to 50% germination and seed weight. Seed weight was determined based on the weight of 50 seeds. Days to 50% germination was determined by curve fitting using a 4-parameter sigmoid model as described by Webster and Ebdon (2004) using days as the independent variable and daily germination percentage as the response variable with the day of initial germination defined as day 1.

Field Study

A field study was prepared and planted at the Troll facility on 8 April, 2013 to assess germination vigor and percent grass cover during establishment. A standard seeding rate of 0.75 pound per 1,000ft² adjusted for pure live seed was used to establish 3 by 3 ft plots. Plots were seeded by hand immediately after ground thaw and when the ground could be properly prepared for planting. Twelve bentgrass cultivars including seven cultivars of creeping bentgrass (007, 13-M, Declaration, L-93, Memorial, Penncross, and T-1), two colonial bentgrass cultivars (Capri and Tiger II), and three velvet bentgrass cultivars (Legendary, SR-7200, and Villa) were selected. Plots were planted as a randomized complete block design using three replicates. *Agrostis* plots were not mowed during the establishment period until total plant height of 0.5 inch was achieved. Thereafter, all *Agrostis* plots were mowed 3 times per week at 0.375 inch height of cut with light weight (walk-behind) reel mowers with clippings collected. Perennial ryegrass (*Lolium perenne* L.) check plots were mowed weekly at 2.0 inch height of cut using rotary mowers with clipping collected.

Weekly grass cover during establishment was recorded until all plots had achieved at least 90% visual grassy cover. Grassy cover was recorded as a visual estimate. Temperature probes were installed into adjacent non-seeded check plots to record soil surface temperatures at a 0.5 inch soil depth. Soil temperatures were recorded on a weekly basis and at the time (day) when plots were initially planted and initial emergence was observed. No covers were used at the time of planting so that initial emergence and germination date as well as visual estimates of weekly grassy cover could be effectively assessed visually. Barbeta perennial ryegrass was also included as part of the field test as a reference species because of its rapid germination and establishment rate. Irrigation was applied as needed to maintain adequate soil moisture for germination. After all plots had shown initial emergence, 18-24-5 fertilizer was applied using a drop spreader at 1.1 pounds P_2O_5 per 1,000ft² as recommended by soil test results.

Days to achieve 10%, 50%, and 90% cover following initial seedling emergence was calculated by curve fitting using a 4-parameter sigmoid model as described above for

the growth chamber study using days as the independent variable and daily percent cover as the response variable with the day of initial emergence defined as day 1. Similarly, soil temperature to achieve 10%, 50%, and 90% cover following initial seedling emergence was also calculated by curve fitting using soil temperature as the independent variable and daily percent cover as the response variable. The soil temperature on the day of initial emergence was defined as day 1 soil temperature. No postemergence herbicides were applied for several weeks before or after planting. Tupersan (Siduron) was applied on 14 May, 2013 at 12 pounds of total material per acre for preventative control of annual grassy weeds. Weed cover as the number of total weeds per plot from the ingress of broadleaf weeds was also recorded on 13 May, 2013. All data was analyzed as a randomized complete block design. Mean separation among cultivar treatments were based on Fisher's Protected Least Significant Difference at α =0.05 level.

RESULTS AND DISCUSSION

Field Study

Perennial ryegrass was the first species to emerge following planting on 8 April, 2013. Barbeta perennial ryegrass emerged at day 9 when soil temperatures were 49.6°F followed by creeping bentgrass, colonial bentgrass, and velvet bentgrass, which emerged at 10.3, 11.0, and 13.0 days after planting, respectively (Table 1). Significant differences were observed among cultivars of two of the three Agrostis species. No difference was observed among cultivars of velvet bentgrass. The rate of increase in percent cover per day following emergence (expressed as cover d⁻¹, Table 1) is an indication of the speed of establishment from 10 to 90% grassy cover and factors out any advantages or disadvantages among cultivars due to their early or late emergence during the establishment period. Higher "cover d⁻¹" values indicate a greater percent grassy cover per day during establishment in the development from an immature stand (10% cover) to a fully established stand (90% cover). The greatest increases in cover was observed with creeping bentgrass cultivars (6.5% cover d⁻¹) with velvet bentgrass cultivars exhibiting significantly lower vigor and grassy cover in early spring (5.4% cover d⁻¹). No statistical difference was observed between colonial bentgrass (6.1% cover d⁻¹) and creeping and velvet bentgrass species. Memorial and 13-M creeping bentgrass cultivars were statically equal to Barbeta perennial ryegrass, which exhibited 9.6, 8.7, and 7.8% increases in grassy cover per day, respectively, during establishment (Table 1).

Generally, *Agrostis* species and cultivars exhibiting greater increases in grassy cover per day during establishment were able to achieve greater increases in grassy cover with soil warming; cover per day and cover per °F were highly correlated (r=0.92, P≤0.001, n=42). Perennial ryegrass showed a 30% increase in grassy cover per °F increase in soil temperature while *Agrostis* species and cultivars averaged only 20% increase per °F with soil warming in early spring (Table 1). Capri colonial bentgrass and 13-M and Memorial creeping bentgrass cultivars were statically equal to Barbeta perennial ryegrass averaging approximately 25% increase in grassy cover per °F increase in soil temperature during establishment (Table 1).

Soil temperatures (°F) required to achieve 10, 50, and 90% cover are reported in Table 1 along with days required to achieve 10, 50, and 90% cover. Colonial bentgrass cultivars averaged 19.4 days to achieve 90% cover followed by creeping bentgrass and velvet bentgrass, which required significantly longer periods of 21.5 and 25.1 days, respectively, until fully established (90% cover, Table 1). Barbeta perennial ryegrass required only 16.4 days until 90% cover was reached. Compared to perennial ryegrass, colonial bentgrass, creeping bentgrass, and velvet bentgrass required approximately 3, 5, and 9 additional days until 90% cover was achieved. Significantly lower soil temperatures were also associated with perennial ryegrass during establishment. Perennial ryegrass averaged 53.2°F soil temperature to achieve 90% grassy cover compared to 55.0, 55.2, and 56.5°F soil temperature before 90% grassy cover was established for colonial bentgrass, creeping bentgrass, and velvet bentgrass, respectively (Table 1). Velvet bentgrass cultivars required significantly higher soil temperatures and more days after emergence compared to other *Agrostis* species to establish 10, 50, and 90% cover (Table 1).

Generally, species and cultivars that were able to establish quickly during early spring were able to compete more effectively with broadleaf weeds and diminish the ingress of weeds into stands during establishment. The total number of broadleaf weeds decreased as days to achieve 10, 50, and 90% grassy cover decreased (r=0.57, 0.65, and 0.47, respectively, $P \le 0.001$, n=42). As such, colonial bentgrass cultivars on average were associated with fewer weeds (25 total per plot) while velvet bentgrass as a species allowed the greatest number (94 total per plot). Barbeta perennial ryegrass was observed to average only seven broadleaf weeds, which was statistically equal to the total weed count observed with Capri and Tiger II colonial bentgrass cultivars and Penncross and 007 creeping bentgrass cultivars (Table 1). Similar correlations between weed encroachment and establishment vigor were observed among Agrostis species and cultivars with soil temperature (°F) indicating better vigor (grassy cover) under lower soil temperature was associated with fewer weeds. The number of weeds at planting also decreased with seed count (r=0.48, $P\le0.001$, n=42) indicating larger seeds (lower seed count) were associated with fewer weeds (Table 1). In addition, lower seed count (larger seeds) was positively correlated with greater establishment vigor and fewer days required for achieving 10, 50, and 90% grassy cover; r values ranged from 0.34 (P=0.035, n=42, days to 90% cover) to 0.45 (*P*=0.004, n=42, days to 10% cover).

Growth Chamber Study

Days to achieve 50% germination under controlled conditions varied with temperature; germination time in days decreased significantly with increasing temperature from 50 to 77°F (Table 2). Germination time to achieve 50% germination decreased from 10.0 to 3.8 days at 50 and 77°F, respectively (Table 2). Soil temperatures in the field (Table 1) for *Agrostis* species and cultivars range from approximately 50 to 55°F during early spring establishment. Moreover, soil temperatures to achieve 50% grassy cover in *Agrostis* species in the field (Table 1) averaged approximately 53°F, which correlate well with the growth chamber temperature of 50°F to achieve 50% germination. Initial emergence in the field study was generally observed at soil temperatures of 50°F. Treatment temperatures corresponding to 59, 68, and 77°F from

the growth chamber study do not necessarily correlate to actual soil temperatures that were observed in the field study at the time of early spring planting, emergence, and establishment.

In the field study, colonial and creeping bentgrass species were observed to require significantly fewer days and lower soil temperature compared to velvet bentgrass to achieve 10, 50 and 90% grassy cover (Table 1). Similarly in the growth chamber study, velvet bentgrass species exhibited significantly more days and reduced germination vigor to achieve 50% germination compared to colonial and creeping bentgrass at 50, 59, and 68 °F (Table 2). At higher temperatures of 77°F, however, velvet bentgrass exhibited significantly fewer days and greater germination vigor to achieve 50% germination than either colonial or creeping bentgrass species.

Generally, the number of days to 50% germination decreased significantly with each incremental increase in temperature from 50 to 77°F (Table 2). However, increases in germination vigor with temperature occurred at a greater rate in velvet bentgrass compared to creeping and colonial bentgrass (Table 2). When compared to all other *Agrostis* species, velvet bentgrass exhibited significantly more days (10.3 days) to achieve 50% germination at 50°F but required significantly fewer days (3.4 days) at 77°F to achieve 50% germination (Table 2). Therefore, velvet bentgrass shows a potential for greater gains in germination vigor with soil warming. Velvet bentgrass would most likely show greater germination vigor under higher soil temperature and with greater soil warming typical of late summer to early fall planting periods. Compared to other *Agrostis* species, however, velvet bentgrass shows a diminished capacity to establish quickly and to prevent the ingress of broadleaf weeds under the cooler soil temperatures of 50 to 55°F typical of early spring plantings.

SUMMARY

First year results from 2013 field and growth chamber studies indicate that genetic differences exists in *Agrostis* species and cultivars in their ability to establish quickly on soils with temperatures ranging from 50 to 55°F. Compared to velvet bentgrass, colonial bentgrass and creeping bentgrass species and cultivars were able to achieve 90% grassy cover by as much as 8 to 9 days sooner and at soil temperatures 2.5 °F colder during early spring plantings. Perennial ryegrass emerged by as much as 1 to 4 days sooner on cold soils in spring than *Agrostis* species and cultivars. After emergence, some *Agrostis* cultivars such as Capri colonial bentgrass and 13-M, Memorial, and Penncross creeping bentgrass cultivars were statistically equal to perennial ryegrass in achieving 90% grassy in less than 20 days. Many of these same species and cultivars with superior germination and establishment vigor were associated with less ingress of broadleaf weeds into seeded plots. This study will be repeated in 2014.

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Table 1. Days and soil temperature (°F) to achieve 10, 50 and 90% grassy cover among 12 Agrostis species and cultivars following planting in the field on 8 April, 2013.

					rease over 10 ssy cover per		Days		S	oil temperat	ure	Broadleaf Weed count	
Species-cultivar	Emergence†			day (d) or soil temp (°F)		following initial emergence			following initial emergence			(35 DAP)	Seed count
					_			Co	over				
	Date	DAP	Soil Temp	Cover d ⁻¹	Cover °F-1	10%	50%	90%	10%	50%	90%	No. plot ⁻¹	No. lb ⁻¹
Creeping bentgrass	2013	Days	°F				Days			°F			
007	20 Apr	12b‡	50.7a	5.0de	17.1cd	6.4ef	13.9bcd	22.7a-d	51.4bc	53.4bc	56.1ab	41c-f	6,003,795cd
13-M	18 Apr	10c	49.6b	8.7ab	24.8ab	9.3abc	14.0bcd	18.4bcd	51.3cd	52.9cd	54.5cd	77abc	6,144,686cd
Declaration	18 Apr	10c	49.6b	4.5e	15.9d	7.6cde	15.9a	25.5abc	51.1cd	53.4bc	56.1ab	59cde	5,033,394e
L-93	18 Apr	10c	49.6b	5.6cde	20.2bcd	8.2bcd	15.4ab	23.2abc	51.1cd	53.2bcd	55.6abc	64cd	5,797,206d
Memorial	18 Apr	10c	49.6b	9.6a	25.1ab	9.4abc	14.0bcd	19.6bcd	51.3cd	52.9cd	54.9bc	67bcd	6,248,612cd
Penncross	18 Apr	10c	49.6b	7.3bc	22.2bc	6.8def	12.5cd	18.4bcd	50.7cd	52.5cde	54.5cd	10ef	5,803,477d
T-1	18 Apr	10c	49.6b	4.8de	16.7d	8.6bc	15.3abc	22.7a-d	51.3cd	53.2bcd	55.4abc	116ab	6,512,670c
Colonial bentgrass													
Capri	18 Apr	10c	49.6b	6.7bcd	24.7ab	7.6cde	12.7cd	17.8cd	50.9cd	52.5de	54.3bcd	24def	5,773,852d
Tiger II	20 Apr	12b	50.7a	5.5cde	17.6cd	5.3f	12.4cd	21.1a-d	51.1cd	52.9cde	55.8abc	27def	6,316,327cd
Velvet bentgrass													
Greenwich	21 Apr	13a	50.7a	5.4cde	20.2bcd	10.3ab	17.5a	25.5ab	52.5a	54.7a	56.7a	90abc	7,966,547b
SR-7200	21 Apr	13a	50.7a	5.1de	18.8cd	10.8a	17.5a	26.4a	52.2ab	54.7a	56.8a	65cd	6,405,173c
Villa	21 Apr	13a	50.7a	5.6cde	19.9bcd	9.1abc	15.8ab	23.3abc	52.2a	54.1ab	56.3ab	128a	8,865,503a
Check	_												
No seed	-	-	-	-	-	-	-	-	-	-	-	87ab	-
Barbeta PRG	17 Apr	9d	49.6b	7.8ab	29.7a	6.2ef	11.3d	16.4d	50.5d	51.8e	53.2d	7f	244,507f
Species average	_												
Creeping	-	10.3C	-	6.5A§	20.3A	8.1B	14.4B	21.5B	51.1B	53.1B	55.2B	62AB	5,858,065B
Colonial	-	11.0B	-	6.1AB	21.1A	6.4C	12.5B	19.4C	51.1B	52.7B	55.0B	25B	6,045,090B
Velvet	-	13.0A	-	5.4B	19.7A	10.1A	17.0A	25.1A	52.3A	54.5A	56.5A	94A	7,745,843A

[†]Emergence date and days after emergence (DAP)=number of days to emergence following planting and soil temperature at initial emergence. ‡Numbers followed by the same lower case letter(s) are not statistically different (α =0.05 level) for comparison among cultivars. §Numbers followed by the same upper case letter(s) are not statistically different (α =0.05 level) for comparison among species.

Table 2. Days to achieve 50 % germination among nine *Agrostis* species and cultivars at four controlled temperatures in the growth chamber.

Species-cultivar		Difference				
	50°F	59°F	68°F	77°F	Average	(50-77°F)
Creeping bentgrass				Days		
007	11.2b‡	6.4ab	4.8a	4.2b	6.7b	7.0b
A-4	8.8e	5.1d	3.8b	3.3cd	5.3e	5.5e
Penncross	10.2cd	5.9bc	4.9a	3.9bc	6.2c	6.2cd
T-1	9.0e	5.3с-е	3.8b	3.2d	5.3e	5.7de
Colonial bentgrass						
Capri	9.6d	5.7cd	4.7a	5.5a	6.4bc	4.1f
Greentime	8.7e	4.9e	3.6b	3.0d	5.0e	5.7de
Tiger II	11.9a	7.0a	5.0a	4.0b	7.0a	7.9a
Velvet bentgrass						
Legendary	10.0c	6.0b	4.4a	3.0d	5.9d	7.0b
Villa	10.5c	5.9b	4.7a	3.7bc	6.2c	6.8bc
Species average						
Creeping	9.8C§	5.7B	4.3B	3.7B	5.9B	6.1B
Colonial	10.1B	5.9A	4.4B	4.2A	6.1A	5.9B
Velvet	10.3A	5.9A	4.6A	3.4C	6.0AB	6.9A
Average	10.0	5.8	4.4	3.8	_	-

[†]Days after initial germination to achieve 50% germination by curve fitting using a 4 parameter sigmoid model.

[‡]Numbers followed by the same lower case letter(s) are not statistically different (α =0.05 level) for comparison among cultivars within temperature.

Numbers followed by the same upper case letter are not statistically different (α =0.05 level) for comparison among species within temperature.



