



UNIVERSITA' DEGLI STUDI DI PADOVA

DOCTORAL SCHOOL OF
CROP SCIENCES

CURRICULUM OF ENVIRONMENTAL AGRONOMY - CYCLE XXIV

Department of Environmental Agronomy and Crop Production

***PERFORMANCE OF WARM SEASON TURFGRASSES
AS AFFECTED BY VARIOUS MANAGEMENT PRACTICES
IN A TRANSITION ZONE ENVIRONMENT***

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THESIS SUBMISSION DATE

31 January 2012

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Riassunto

La gestione dei tappeti erbosi è sempre più spesso soggetta a critiche a causa dell'elevato impiego di risorse naturali, tra le quali soprattutto l'acqua per l'irrigazione. In tal senso, le specie *poaceae* macroterme sono preferibili rispetto alle microterme. Infatti, in Italia, durante i mesi estivi i fabbisogni irrigui delle specie macroterme possono ridursi dal 20 al 45% rispetto a quelli delle microterme. Le specie macroterme sono entrate nel mercato italiano solo recentemente ed il loro effettivo impiego è ancora molto limitato, a causa del lungo periodo di dormienza invernale (fino a cinque mesi) durante il quale ingialliscono. La scelta della specie/cultivar è determinante, giacché in commercio vi sono materiali con caratteristiche molto eterogenee. Sebbene la scelta della cultivar sia importante, anche le pratiche colturali possono condizionare la risposta varietale. Con l'intento di approfondire quest'ultimo aspetto, sono stati studiati gli effetti di alcune pratiche colturali sulla qualità del tappeto erboso, la tolleranza al gelo e la durata del periodo di dormienza di alcune *poaceae* macroterme: *Cynodon dactylon*, *Paspalum vaginatum* e *Zoysia* spp. In particolare, sono stati presi in esame la concimazione azotata autunnale, lo *scalping* primaverile e l'applicazione invernale di glifosate. Nel periodo 2009-11, sono state condotte tre prove parcellari presso l'azienda agraria sperimentale dell'Università di Padova.

Relativamente alla concimazione autunnale, la distribuzione dell'azoto in cinque apporti fino ad ottobre, rispetto alla stessa quantità annua distribuita in tre apporti fino ad agosto, ha consentito un modesto anticipo del rinverdimento di *C. dactylon* e *P. vaginatum*. Inoltre, l'apporto di azoto in autunno ha ritardato il disseccamento di *C. dactylon*. Le diverse modalità di somministrazione non hanno influenzato le caratteristiche morfologiche di stoloni e rizomi ed hanno avuto un effetto limitato sull'accumulo delle sostanze di riserva. L'esecuzione dello *scalping* a metà marzo su cultivar di *C. dactylon* e *Z. japonica* ha consentito un anticipo del rinverdimento fino a 15 giorni. Tuttavia, tale pratica è risultata efficace principalmente per le varietà caratterizzate da un rinverdimento tardivo. Infine, l'applicazione di glifosate a febbraio ha consentito un controllo efficiente delle infestanti autunno-vernine, senza conseguenze negative sul rinverdimento di *C. dactylon* e *Z. matrella* o provocando un lieve ritardo in *Z. japonica*. Da tale studio si evince che la scelta della specie/cultivar condiziona le prestazioni dei tappeti erbosi di macroterme in misura maggiore rispetto alle pratiche gestionali studiate.

Summary

The maintenance of turfgrasses has been increasingly criticized due to its high demand of natural resources, especially for the requirements of potable water for irrigation. To address this issue, warm season grasses should be preferred over the cool season in northern areas of Italy, where warm season grasses require 20–45% less water than the cool season in summer. Warm season turfgrasses have been recently introduced in the Italian industry but their widespread use is gained due to a lack of green color during winter dormancy, which can persist up to five months. Since wide variability occurs among commercial turfgrasses, the choice of species and/or cultivar should be taken in high consideration. Despite varietal selection plays an important role, cultural practices are also critical for turf performance. To learn more about this topic, the effects of various management practices have been experimented on turf quality, freeze avoidance, and winter dormancy of some warm season grasses: bermudagrass, seashore paspalum, manilagrass, and zoysiagrass. Field studies on late-season nitrogen fertilization, spring scalping, and winter-application of glyphosate have been carried out from 2009 to 2011 at the experimental agricultural farm of Padova University (northeastern Italy).

Under the same annual amount of nitrogen, five applications of nitrogen until October allowed bermudagrass and seashore paspalum to recover from dormancy slightly earlier compared to three applications until August. In addition, late-season application of nitrogen delayed fall color retention of bermudagrass cultivars. Different nitrogen fertilization schedules had no effects on morphological traits of stolons and rhizomes; and their influence on reserve accumulation was of limited biological importance. Bermudagrass and zoysiagrass cultivars subjected to scalping in mid March reached 80% green cover up to 15 days earlier compared to controls (i.e., unscalped plots). However, the effectiveness of this practice was of practical importance only for bermudagrass cultivars that were slow to green-up in spring. Application of glyphosate in February satisfactorily controlled winter weeds, resulting in no injury for bermudagrass and manilagrass; and little delay of spring green-up for zoysiagrass. Results of this project pointed out that the choice of species/cultivar should be considered of primary importance relative to management practices for improving the performances of warm season turfgrasses.

Introduction

Turfgrasses are involved in several human activities and they have increasingly become more important components of European city architectural landscapes. As a consequence, the turfgrass industry has grown rapidly contributing to the national economy with new employment opportunities. Turfgrasses are important for ornamental purposes and also offer safe recreational surfaces. In addition, turfgrasses provide many environmental benefits, such as preventing soil erosion, reducing air pollution, dust stabilization, and prevention of pesticides leaching (Beard, 1973). Based on their main function, turfgrasses can be referred as ornamental, functional-recreational, or athletic fields; all of each is made by millions of individual plants that together form an unique plant community. Turfgrasses completely differ from any other agricultural crop and require a considerable effort in terms of labor, energy and costs, sometimes exceeding public or private budgets. However, irrigation and the use of herbicides and pesticides are necessary to maintain an adequate quality.

Quality criteria may vary based on the type of turfgrass, the purposes for which it is to be used, and the individual performing the evaluation. However, some traits of turfgrass are widely recognized as the main components of quality: color, density, uniformity, leaf texture, growth habit, and smoothness (Beard, 1973; Patton *et al.*, 2008). These parameters are typically rated by visual estimations, which can be supported by instrumental measurements, such as chlorophyll index (Madison & Andersen, 1963), digital image analyses (DIA) (Richardson *et al.*, 2001; Karcher & Richardson, 2003), and normalized difference vegetation indices (NDVI) (Bell *et al.*, 2009). Depending on user requests and season of the year, the desired quality levels can also widely vary and maintaining high quality turf may result often in a high demand of inputs. Water consumption represents the most important cost to society, since large amount of potable water are generally destined to turfgrass irrigation, rather than domestic use. For example, 30% of potable water is used for turfgrass irrigation in the East coast area of USA, rising to 60% in the arid West (Brede, 2000). According to the requirement of low input managements and based on the warming meteorological trend, the future of turfgrasses is strictly related to the ability to increase water use efficiency, which mainly depends on species selection (Beard & Beard, 2004).

Turf species are typically divided into cool season and warm season grasses. The two groups have different photosynthetic systems, such that cool season grasses begin the production of carbohydrate with a three-carbon (C₃) compound, while warm season species with a four-carbon (C₄) compound (Jones, 1985). Cool season species, such as perennial ryegrass (*Lolium perenne* L.), kentucky bluegrass (*Poa pratensis* L.), tall fescue (*Festuca arundinacea* Schreb.), and creeping bentgrass (*Agrostis stolonifera* L.), are best adapted to the cooler regions of the world and have an optimum temperature range of 15 – 25 °C. Instead, the warm season species, such as bermudagrass [*Cynodon dactylon* (L.) Pers.] and zoysiagrass (*Zoysia japonica* Steud.), are distributed in warmer zones and have an optimum temperature range of 27 – 35 °C. Warm season species lose chlorophyll as they go dormant, while cool season species generally remain green through the winter (Fig. 1). The cool season grasses emerge from winter dormancy in early spring, are intolerant to summer stress period, and have a slow growth increase in the fall (Fig. 2). The warm season break winter dormancy in the spring later than cool season grasses and reach the maximum growth rate in the summer. Cool season grasses have greater winter hardiness than warm season grasses but their tolerance to heat and drought is generally poor. They can be used in arid regions only if irrigation is supplied and they also require water supply in temperate regions during the warmer periods to maintain plant vigour and turf quality. Warm season grasses are also more efficient in rooting, which led to a root mass considerably deeper compared to cool season species (Christians, 1998).

Some areas of the world are known as transition zone, in which the winter is cold enough to make difficult the growth of warm season, while summer is warm enough to limit that of cool season grasses (Christians, 1998). Therefore, the management of grasses is particularly challenging for green keepers and turf maintainers operating in these areas. In the transitional zones of the Mediterranean Europe, which include most of Italy, turfgrasses are mostly composed of cool season species. However, maintaining cool season grasses in these areas has been questioned because of their high irrigation requirements relative to warm season species. Cool season turfgrasses use more water than warm season grasses due to their higher evapotranspiration rates during the summer months, and longer growing period. Evapotranspiration rates for warm season grasses range from 2 to 5 mm d⁻¹ compared to 3 to 8 mm d⁻¹ for cool season grasses (Casnoff *et al.*, 1989; Huang, 2008).

Although warm season turfgrasses are best adapted to warm climates, they can be successfully grown in transitional environments (Harland & de Wet, 1969; de Bruijn, 2010; Volterrani *et al.*, 2004). Moreover, warm season species have relative low nutrient requirement, high fungi resistance, and offer tough competition to summer weeds compared to cool season grasses (Brede, 2000). Warm season grasses spread quickly by stolons and rhizomes, and they are able to respond to variation of sources availability by plastic adjustment in branching intensity, and in length and weight of spacer organs (Dong & Pierdominici, 1995). This morphological plasticity improves the adaptability to heterogeneous environments and competitive ability. In order to reduce water consumption for landscape irrigation, the use of warm season grasses should be encouraged in Mediterranean countries of Europe.



Figure 1 – General view of cool season (upper side of the picture) and warm season (lower side) turfgrass experimental plots at the New Mexico State University saline irrigation turfgrass research station (Las Cruces, NM) as they appeared in Nov. 2009.

Winter dormancy and poor tolerance to low temperatures represent the main impediments to warm season grasses gaining greater acceptance in the Mediterranean countries. In transitional zones, warm season grasses lose color in autumn and are susceptible to low temperature injury during the cooler months (Beard, 1973; Richardson, 2002; Munshaw *et al.*, 2006). Turf leaf tissues turn yellow when temperature drops below 0 °C, and successively survive over winter thanks to the reserves stored in stolons and

rhizomes, which allow to the spring green-up (Macolino *et al.*, 2010). In the northern regions of Italy, the warm season turfgrasses remain dormant for up to five months which strongly inhibits their widespread use in that area. The winter dormancy is not easy to accept in temperate regions, since turf managers and users are used to see green turf over the four seasons. Furthermore, the environmental functions typically associated to the turfgrass are more reduced in warm season than in cool season species during the winter (Beard & Green, 1994).

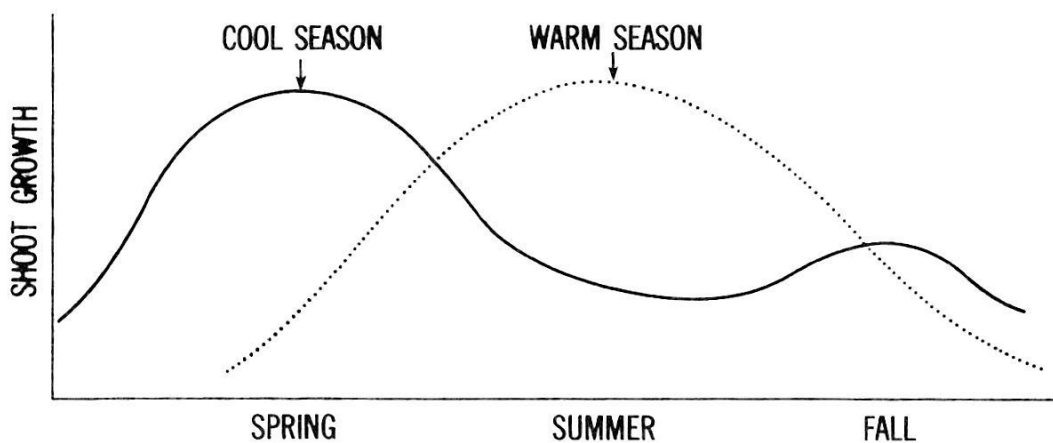


Figure 2 – Shoot growth seasonal distribution of cool season and warm season grasses (Christians, 1998).

Bermudagrass is currently the warm season turfgrass most widely used in the European countries because of its recuperative potential, wear tolerance, and pest resistance (Beard, 1973; Croce *et al.*, 2003). Most of the turf-type bermudagrasses have their center of origin in eastern Africa (Hanson *et al.*, 1969) and are used on athletic fields, lawns, parks, and golf courses. In transitional zones, bermudagrass may be used in pure stands or overseeded in the fall with cool season grasses, such as perennial ryegrass or annual ryegrass (*Lolium multiflorum* Lam.) to provide year-round green color (Goddard *et al.*, 2008; Schmidt & Shoulders, 1980).

In the past few years, other warm season species have increasingly become more important in the Italian turf industry, such as zoysiagrass and seashore paspalum (*Paspalum vaginatum* Swartz). Zoysiagrasses (*Zoysia* spp.) originated in the eastern Asia (Duble, 1989) and are primarily grown in warm humid and transitional regions, where represent a

valuable alternative to bermudagrass due to a very high-quality turf and good cold tolerance. However these species grow very slowly and take longer than bermudagrass to establish and to recover from damages (Christians, 1998). For these reasons zoysiagrass turfs are usually established vegetatively by sod rather than by seeding, leading to high establishment costs, which limits their practical use. Seashore paspalum is native of the seaside dunes of southern Africa (Handreck & Black, 2010) and has been recently introduced in temperate countries as a consequence of progress in turfgrass industry. Turf traits and performances are analogous to those of bermudagrass and, in temperate climate conditions, it could be seriously damaged by cold winter (Casler & Duncan, 2003). Seashore paspalum is characterized by fine leaf texture and very high tolerance of sodium salt. It grows where salt accumulation in soil is so high that limits the survival of other grasses, perhaps its use for turfgrass purposes may represent a solution for specific conditions.

The speed to recovery after winter dormancy and low temperature tolerance have been considered main factors in selecting bermudagrass varieties for the transition zone (Anderson *et al.*, 2007; Patton *et al.*, 2008). Results from controlled regional studies indicated that warm season species and/or cultivars may crucially differ in spring green-up (Macolino *et al.*, 2010; Severmutlu *et al.*, 2011). Several researchers also reported differences between cultivars in term of fall color retention (Munshaw *et al.*, 2006; Schiavon *et al.*, 2011). These findings indicate the possibility to reduce the period of winter dormancy by the proper choice of warm season species and cultivars within species. Therefore, an object of this work was to evaluate the adaptability of warm season species and cultivars to the environmental conditions of the Venetian Valley. Performance of warm season species have been studied with emphasis on fertilization (Goatley *et al.*, 1994; Miller & Dickens, 1996a, 1996b; Trenholm *et al.*, 1998; Munshaw *et al.*, 2006) and other management practices such as winter turf covers (Goatley *et al.*, 2005) and plant growth regulators (White & Schmidt, 1989; Richardson, 2002). Collectively, these studies demonstrated that great potential exists for accelerating spring regrowth and delaying fall color retention through the application of appropriate cultural practices. Perhaps, the main objective was to test the effects of various management practices on spring green-up and fall color retention of warm season grasses in a transition zone environment.

During the course of this doctoral project, three management practices have been investigated: i) late-season fertilization, ii) spring scalping, iii) winter weeds control. These practices have been tested on bermudagrass and seashore paspalum, on bermudagrass and zoysiagrass, and on manilagrass [*Zoysia matrella* (L.) Merr.], zoysiagrass and bermudagrass, respectively. The experimental works are described in the following chapters.

Chapter I

**Effects of three nitrogen fertilization
schedules on four bermudagrass cultivars
and 'Sea Spray' seashore paspalum**

Introduction

Mineral nutrition is one of the most important components of a turfgrass management program as fertilization strongly influences physiological parameters and performance of turfgrass. Among the essential elements, nitrogen (N) is typically used in the largest amount by turfgrass (Hall & Moses, 1931). Nitrogen is needed in highest concentrations in the actively growing tissues, such as young leaves and flowers, and its content in turfgrasses may vary from 3 to 6% on a dry matter basis (Welton & Carrol, 1940; Roberts & Bredakis, 1960; Hodges, 1965). Monthly requirements of N fertilization range between 0 and 10 g m⁻², depending on species/cultivar and on environmental conditions (Beard, 1973). Within limits, plant growth is associated to N supplies, with shoots having priority over roots in responding to N growth stimulation (Lovvorn, 1945; Hanson & Juska, 1961; Zaroni *et al.* 1969). Moreover, the level of N fertilization affects turf performance in a number of ways including density (Jagschitz & Skogley, 1965; Goss & Law, 1967), color (Kopec *et al.*, 2007; Xiong *et al.*, 2007), recuperative potential (Lunt, 1954), and resistance to environmental stresses (Schmidt & Blaser, 1969; Beard, 1973).

In transitional zones, the standard recommendation for N fertility program of bermudagrass and seashore paspalum (Fig. 3) is 5 g m⁻² of N per growing month (Christians, 1998). However, proper adjustments of this general guideline need to be applied according to soil characteristics and local climate conditions in order to increase winter hardiness and to extend growing season. Exposure to low temperatures as winter approaches induces plant acclimation, which is optimized under non-limiting light conditions (Gray *et al.*, 1997). The significant roles of carbohydrate reserves and protein accumulation in turfgrass stress tolerance and cold hardiness have been widely recognized (Di Paola & Beard, 1992; Fry *et al.*, 1993; Gatschet *et al.*, 1994). These molecules are accumulated in storage organs during the autumn when plant growth is slow, which in return improves cold hardiness by reducing the freezing point and delaying cell freezing (Stier & Fei, 2008; Macolino *et al.*, 2010). Rhizomes and stolons have been indicated as the principal locations of overwintering reserves in warm season grasses, with stolons being typically the most important between the two organs (Dunn & Nelson, 1974).

Plant carbohydrates have been classified by White (1973) in two major groups, the nonstructural and the structural carbohydrates. The major carbohydrate reserve constituents of plants, such as glucose, fructose, sucrose, fructosans, and starches, are traditionally referred as total nonstructural carbohydrates (TNC), while hemicelluloses and cellulose are considered structural carbohydrates (White, 1973; Goatley *et al.*, 1994; Bunnell *et al.*, 2005). Among the TNCs, cool season grasses typically store water-soluble carbohydrates (WSC), while warm season species accumulate greater amounts of starch, which need to be hydrolyzed to provide energy (Okajima & Smith, 1964; Bender & Smith, 1973; Borland & Farmer, 1985). The TNC are used as a resource to support the shoot renewing in spring, leading to a positive correlation between their content and early green-up in warm season grasses (Rogers *et al.* 1975; Di Paola & Beard, 1992; Macolino *et al.*, 2010). In addition, several studies indicated that turfgrasses may increase their capacity for protein synthesis during winter acclimation (Cloutier, 1983; Huges & Dunn, 1996; Zhang *et al.*, 2006). In this period, plants synthesize many low-temperature induced proteins, which are able to protect cells from freezing by stabilizing their proteins and membranes (Guy *et al.*, 1985; Taiz & Zeiger, 1998). Several researchers recognized the role of a unique group of proteins, referred as antifreeze proteins, which enable turfgrasses to survive freezing conditions (Sidebottom *et al.*, 2000; Kuiper *et al.*, 2001). Gatschet *et al.* (1994) reported that cold acclimation in bermudagrass promoted the synthesis of proteins with unknown function having low to intermediate molecular weight. It has been also reported that cold acclimation induced an increase of proline, glutamine, and glutamic acid in annual bluegrass (*Poa annua* L.) (Dionne *et al.*, 2001). Anderson *et al.* (2002) suggested that physiological differences in acclimation between bermudagrass cultivars could be based on differences on stress protein induction. Moreover, an increase of protein synthesis has been correlated with greater freezing tolerance in bermudagrass cultivars (Gatschet *et al.*, 1996). Although the functions of many cold-induced proteins are still unclear, collectively these reports denoted that protein accumulation plays a key-role in winter survival of turfgrasses.

In the transition zone, a primary concern in managing bermudagrass and seashore paspalum is the turf survival under low temperatures during dormancy. Conventional management practices that enhance freeze tolerance of bermudagrass have focused on reducing the application of N while increasing those of K in late summer and early fall

(Reeves *et al.*, 1970; Ervin *et al.*, 2004). These practices agree with the recommendations of several textbooks, reporting that fall N fertilization may reduce winter hardiness by promoting grass growth while sustained levels of phosphorus (P) and potassium (K) are important for winter hardiness (Beard, 1973; McCarty, 2001; Turgeon, 2002). However, the results of several studies on late-season N applications during the last 15 years indicated no loss of cold hardiness, contrary to what is commonly believed (Smith & Chalmers, 1993; Richardson, 2002; Munshaw *et al.*, 2006). Goatley *et al.* (1994) found that the application of late-season N on ‘Tifgreen’ bermudagrass delayed fall color retention and provided faster spring green-up with limited effects on TNC levels in rhizomes. Richardson (2002) also found that late-season N applications improved both fall color and spring regrowth of ‘Tifway’ bermudagrass, without any negative effects on rhizome cold tolerance. Munshaw *et al.* (2006) corroborated these findings, reporting that N applications in the fall promoted color retention and spring green-up of several bermudagrass cultivars, with no effects on postfreeze regrowth.



Figure 3 – Turf density and leaf texture of turf-type bermudagrass (left side of the figure) and seashore paspalum (right side).

Previous studies have investigated the effects of late-season N applications on bermudagrass afterwards equal distribution of N throughout the spring and summer (Smith & Chalmers, 1993; Goatley *et al.*, 1998; Richardson, 2002). However, the physiological and agronomical responses of bermudagrass to N fertilization schedules shifted in the late-season, under the same annual amount of N, have not been tested. Moreover, little is known on the effects of late-season N applications on seashore paspalum. Therefore, the objectives

of this study were to determine the effects of three N fertilization schedules on visual quality, spring green-up, fall color retention, stolon morphological features, levels of starch, WSC, and crude protein (CP) of four seeded bermudagrass cultivars and 'Sea Spray' seashore paspalum.

Materials and methods

Site description

This study was conducted from Sept. 2009 to Dec. 2011 at the experimental agricultural farm of Padova University in Legnaro, northeastern Italy (lat. 45°20'N, long. 11°57'E, elevation 8 m). The area has a sub-humid climate, with an annual rainfall of 820 mm mostly distributed during the growing season from April to November. The annual mean temperature is of 12.3 °C and average minimum and maximum temperatures of –5.5 and 32.8 °C, and the area is similar to plant hardiness zone 8 (U.S. Department of Agriculture, 1990). Mean monthly meteorological parameters collected during the study period are shown in Table 1.

Table 1 – Monthly mean air temperatures and monthly precipitations from 2009 to 2011, and long-term averages (1963-2007) at the agricultural experimental farm of Padova University, Legnaro, northeastern Italy.

Month	Mean temperature				Total precipitation			
	2009	2010	2011	45-yr avg.	2009	2010	2011	45-yr avg.
	°C				mm			
Jan.	2.9	2.5	3.1	2	57	59	18	53
Feb.	5.2	4.9	5.0	4.1	57	127	45	49
Mar.	9.0	8.1	9.3	7.8	105	38	98	56
Apr.	14.3	13.8	15.5	11.6	126	45	4	69
May	20.2	17.7	19.5	16.6	26	110	25	74
June	21.6	21.9	22.0	20.2	87	111	59	82
July	24.2	24.7	22.9	22.3	79	96	88	74
Aug.	25.3	22.4	24.6	21.9	21	91	10	76
Sept.	20.8	17.9	22.0	18.2	190	112	60	72
Oct.	14.3	12.7	13.2	13.1	44	87	90	80
Nov.	10.7	9.8	7.8	7.2	92	153	80	76
Dec.	3.8	2.6	4.6	2.9	113	112	23	60
Annual	14.4	13.3	14.1	12.3	996	1141	601	820

Daily growing degree days (GDD) were calculated during Sept. 2009–Apr. 2010 (Year 1), Sept. 2010–Apr. 2011 (Year 2), and the spring (1 Mar.–30 June) of 2010 and 2011 (Fig. 4), using the following formula:

$$\text{GDD} = [(T_{\text{MAX}} + T_{\text{MIN}}) / 2] - T_{\text{BASE}}$$

using 5°C as T_{BASE} (Unruh *et al.*, 1996; Frank *et al.*, 1998; Patton *et al.*, 2004); where if $T_{MAX} < T_{BASE}$, then $T_{MAX} = T_{BASE}$, and if $T_{MIN} < T_{BASE}$, then $T_{MIN} = T_{BASE}$ (Baker *et al.*, 1986; Swanson & Wilhelm, 1996; McMaster & Wilhelm, 1997). The soil at the site was a coarse-silty, mixed, mesic, Oxyaquic Eutrudept (Morari, 2006) containing 14% clay, 69% silt, and 17% sand, with a pH of 8.1, 2.4% organic matter, a carbon (C) to N ratio of 12.2, an Olsen P content of 29 mg kg⁻¹, and an exchangeable K content of 140 mg kg⁻¹.

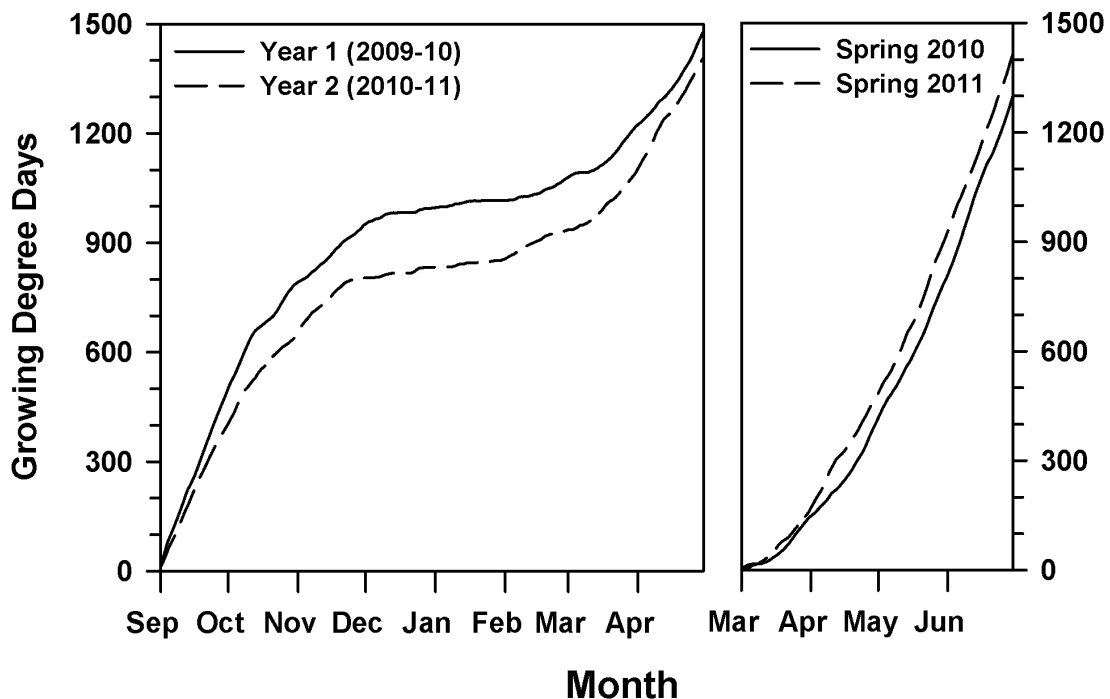


Figure 4 – Cumulative Growing Degree Days from 1 Sept. to 30 Apr. in 2009–10 (Year 1) and 2010–11 (Year 2), and during the spring of 2010 and 2011 at the agricultural experimental farm of Padova University, Legnaro, northeastern Italy. Growing Degree Days were calculated using a base temperature of 5 °C.

Plant material and management practices

The experiment was carried out on turf plots that were seeded on 24 June 2008 using four bermudagrass cultivars and ‘Sea Spray’ seashore paspalum (The Scotts Company, Marysville, OH) at a seeding rate of 48.8 kg pure live seed ha⁻¹. Bermudagrass cultivars used were Princess-77 (SeedsWest, Inc., Maricapa, AZ), Riviera (Johnston Seed Co., Enid, OK), SWI 1014 (SeedsWest, Inc., Maricapa, AZ), and Yukon (Seed Research of Oregon,

Corvallis, OR). Except the first month of the establishment period, no additional irrigation to natural precipitation was provided during the study. During the establishment year prior the onset of the study (2008), all plots (2×3 m) received 20 g m^{-2} of N with applications made as 4 g m^{-2} of N in the ammonium nitrate form (26N-0P-0K) on a biweekly schedule from July to September. Nitrogen fertilization schedules were imposed from 2009 and included: A) 6.7 g N m^{-2} on 15 May, 15 June, and 15 Aug.; B) 5 g N m^{-2} on 15 May, 15 June, 15 Aug., and 15 Oct.; and C) 4 g N m^{-2} on 15 May, 15 June, 15 Aug., 15 Sept., and 15 Oct.

The N fertilizer used was ammonium nitrate (26N-0P-0K) and all plots also received P as triple superphosphate (0N-19P-0K) in July and Aug. 2011 at a rate of 40 kg ha^{-1} of P_2O_5 , to maintain soil P at sufficiency level. Potassium fertilizer and other nutrients were not provided throughout the study according to soil tests recommendations. In addition, plots were mowed weekly until late September at an effective cutting height of 45 mm using a rotary mower (HRD 536; Honda Europe Power Equipment, Ormes, France) and clippings were removed. In October and November, plant vertical growth was monitored by weekly measurements of actual turf canopy height at four randomly selected locations within each plot. In March and November, weeds were removed by hand; and no herbicides were used during the trial as plots presented low levels of weed infestations throughout the rest of the year.

Spring green-up, summer quality, and fall color retention

In 2010 and 2011, visual ratings of spring green-up were assigned weekly from 15 Mar. to 15 June based on a linear percentage (0–100) scale of green cover. A sigmoidal model (GraphPad Prism 5.0 for Windows; GraphPad Software, La Jolla, CA) was used to calculate days of year, beginning 1 Jan., required to reach 80% of green color for each plot (D80) (Macolino *et al.*, 2010). Turf recovery from winter dormancy was also monitored on a biweekly schedule using DIA (Richardson *et al.*, 2001; Karcher & Richardson, 2005) via SigmaScan Pro software (version 5; SPSS Inc., Chicago, IL). Photographic images were obtained using a handheld digital camera (Canon PC1225; Canon Europe Ltd, London,

UK) housed in a closed box with four halogen light bulbs designed to provide uniform covering of light (Shaver *et al.*, 2006).

Plots were visually rated for turf quality from June to September using a 1–9 rating scale, where 1 = dead, 6 = acceptable, and 9 = most desirable. Turfgrass quality ratings were taken according to the guidelines of National Turfgrass Evaluation Program (NTEP), based on combination of color, density, uniformity, texture, weed infestation, and susceptibility to environmental stresses (Krans & Morris, 2007). Bermudagrass and seashore paspalum had their own standard (NTEP, 2011a). Fall color retention was visually rated from September to November using a 1–9 scale, where 1 = completely dormant and 9 = lush green. Color was the primary parameter of interest for fall color ratings (Munshaw *et al.*, 2006). Turf summer quality and fall color ratings were collected on a weekly schedule in 2010 and 2011, and the ratings were averaged every month. In addition, NDVI readings were collected by means of a Greenseeker handheld unit (NTech Industries, Ukiah, CA) from March to December within 24 h of the visual ratings being taken (Bell *et al.*, 2002).

Sampling and measurements

Areas measuring 0.5×3 m were defined within each plot for turf sampling in Year 1 and Year 2, respectively. Turf samples measuring $20 \times 20 \times 4$ (depth) cm were randomly collected on a monthly schedule (Table 2). Soil was added back to the quadrat sample areas immediately after sampling to support turf re-colonization. Turf samples were washed to remove soil from the vegetative parts; and leaves, shoots, and roots were discarded with scissors. Stolons and rhizomes were identified according to their growth habit and morphological traits, such as coloration, internodes length and diameter (Richardson, 2002), and immediately frozen at -22 °C. Stolons and rhizomes were later freeze-dried and weighed to determine their dry weight. Stolon internodes diameter, length, and dry weight were also measured as an average of 12 random three-internodes stolon fragments. Stolon internodes dry weight was determined after drying in forced air oven for 36 h at 105 °C and was added back for calculating the stolon dry weight.

Table 2 – Sampling dates for stolon and rhizome collection.

Year†	Sampling dates							
1	19 Sept.	17 Oct.	14 Nov.	5 Dec.	16 Jan.	12 Feb.	21 Mar.	17 Apr.
2	21 Sept.	16 Oct.	12 Nov.	11 Dec.	15 Jan.	11 Feb.	9 Mar.	15 Apr.

† Year 1 = 2009–2010; Year 2 = 2010–2011.

Non-structural carbohydrates and crude protein determination

After freeze-drying, stolons were ground to pass a 0.5-mm screen. Total WSC were extracted from 100 mg of stolon tissue with 800 mL L⁻¹ ethanol, microfuged, and the sugar concentration of the supernatant determined spectrophotometrically (Ultrospec 2000 Spectrophotometer; Pharmacia Biotech, Cambridge, UK) with anthrone (9,10-dihydro-9-oxoanthracene), using glucose as a standard (Van Handel, 1968). Additional 100 mg of stolon ground tissue were used for starch analysis. The samples were pre-extracted two times with 800 mL L⁻¹ ethanol at ≈80 °C and the sediment was used for analysis. The ethanol-extracted residue was starch digested by adding 3.0 mL thermo stable α -amylase (A3306 from *Bacillus licheniformis*, Sigma-Aldrich, St. Louis, MO) and 0.1 mL amyloglucosidase solution (A7255 from *Rhizopus mold*; Sigma-Aldrich, St. Louis, MO). Tubes were centrifuged and glucose in the supernatant was determined using glucose oxidase. Starch concentration was estimated as $0.9 \times$ glucose concentration (McCleary *et al.*, 1997). Stolon content of TNC was determined as the sum of starch and WSC, and the ground area amount was calculated as stolon dry weight \times stolon content of TNC. Total N was extracted from 700 mg of stolon ground tissue using the method described by Ling (1963) and CP was determined as Kjeldahal N \times 6.25.

Statistical analysis

The experimental design was a completely randomized split plot with three replicates, having cultivars as main plots and N fertilization schedules as subplots. Variance homogeneity of the data was evaluated by panels of studentized residuals, and log-transformation was used for rhizome dry weight. Visual ratings, D80, turf height, tissue measurements, starch, WSC, and CP were statistically analyzed using a repeated measures

analysis of variance (Schabenberger & Pierce, 2002) with SAS Proc Mixed (version 9.2; SAS Institute, Cary, NC). Months were used as variable for repeated measures of visual ratings; years (2010, 2011) as variable for D80; and sampling dates as variable for turf height, tissue measurements, starch, WSC, and CP. Three types of covariance structures were compared for the repeated measures: compound symmetry, autoregressive order one, and spatial power, using the Akaike information criterion (AIC) and the REPEATED statement in Proc Mixed (Littell *et al.*, 1996; Witowska *et al.*, 2008). A compound symmetry covariance structure generally resulted in the best fit for the data (lowest AIC values); therefore repeated measures were analyzed using the compound symmetry structure. Tukey's honestly significant difference test was used at the 0.05 probability level to identify significant differences among means.

Proc Corr and Proc Reg (version 9.2; SAS Institute, Cary, NC) were used to correlate visual ratings with NDVI readings and DIA estimates. Proc Reg was also used to regress D80 against stolon dry weight and content of starch, WSC, and CP. GraphPad Prism software package (version 5.0; GraphPad Software, La Jolla, CA) was used to investigate the relationship between GDD and D80, and between GDD and ground area amount of TNC in stolons. Contrasting regression models were discriminated using the *F* test of differences between sum-of-squares at $P < 0.05$, followed by AIC values comparison (Bozdogan, 1987; Motulsky & Christopoulos, 2003). All regression diagnostics were performed using the Shapiro-Wilk test (Shapiro & Wilk, 1965) to ensure that residuals were normally distributed and if needed the data were log-transformed.

Results and Discussion

The interactions between cultivars and years and between N fertilization schedules and years were significant for D80, which was also affected by the main effects of cultivars and N fertilization schedules (Table 3). Cultivars interacted with months in determining summer quality and autumn color. The interaction terms N fertilization schedule \times cultivar and N fertilization schedule \times month were also significant for fall color retention (Table 3).

All tissue measurements (dry weight of rhizome and stolon; diameter, mass, and length of stolon internodes) were affected by the interactions between cultivars and sampling dates, and their single effects (Table 4). Stolon content of WSC was influenced by the two-way interactions cultivar \times sampling date, and all treatment effects (Table 5). The interaction terms cultivar \times sampling date and N fertilization schedule \times sampling date, and all the main effects were significant for stolon content of starch and CP (Table 5).

Table 3 – Probability values of ANOVA for treatment effects and their interactions on calendar days to reach 80% green cover (D80), summer visual quality, fall color retention, and turf canopy height of five warm season turfgrasses.

Source	D80	Summer quality	Fall	
			Color retention	Canopy height
<i>Pr > F</i>				
Cultivar (Cv)	<.0001	0.0064	<.0001	0.2660
Nitrogen fertilization (N)	<.0001	0.0987	<.0001	0.0979
Cv \times N	0.3189	0.3952	<.0001	0.2323
Time†	0.4685	<.0001	<.0001	<.0001
Cv \times Time	<.0001	<.0001	<.0001	0.0889
N \times Time	0.0241	0.6436	<.0001	0.4730
Cv \times N \times Time	0.3284	0.8763	0.0735	0.9582

† Years were variable for repeated measures of D80; months for summer quality and fall color; sampling dates for canopy height.

Spring green-up

In the spring of 2010, ‘Yukon’, ‘Riviera’, and ‘SWI 1014’ exhibited earliest spring green-up, while ‘Princess-77’ and ‘Sea Spray’ reached D80 15–16 d later than ‘Yukon’ (Fig. 5). In the subsequent spring, ‘Yukon’ and ‘Riviera’ showed again earliest green-up, reaching

D80 by mid April, whereas ‘Princess-77’ and ‘Sea Spray’ reached D80 17 d and 27 d later than ‘Yukon’, respectively. When data were averaged over the two years, ‘Yukon’ and ‘Riviera’ were earliest to achieve green-up, and ‘Sea Spray’ showed slowest green-up (Table 6).

Table 4 – Probability values of ANOVA for treatment effects and their interactions on tissue measurements of five warm season turfgrasses.

Source	Rhizome		Stolon		
	Mass	Mass	Diameter	Internodes length	Internodes mass
<i>Pr > F</i>					
Cultivar (Cv)	0.0001	<.0001	<.0001	<.0001	<.0001
Nitrogen fertilization (N)	0.7443	0.9719	0.2160	0.7460	0.2070
Cv × N	0.8829	0.8589	0.4756	0.7640	0.1998
Sampling date (Time)	<.0001	<.0001	<.0001	<.0001	<.0001
Cv × Time	0.0017	<.0001	<.0001	<.0001	<.0001
N × Time	0.9924	0.9461	0.5297	0.0769	0.0664
Cv × N × Time	0.9730	0.9842	0.7859	0.7132	0.4387

Table 5 – Probability values of ANOVA for treatment effects and their interactions on stolon reserves of five warm season turfgrasses.

Source	Starch	Water soluble carbohydrates	Crude protein
	<i>Pr > F</i>		
Cultivar (Cv)	<.0001	<.0001	0.0054
Nitrogen fertilization (N)	0.0114	0.0004	<.0001
Cv × N	0.8158	0.9934	0.1831
Sampling date (Time)	<.0001	<.0001	<.0001
Cv × Time	<.0001	<.0001	<.0001
N × Time	0.0091	0.5054	<.0001
Cv × N × Time	0.1561	0.6595	0.4011

These results agreed with a previous study of Macolino *et al.* (2010) conducted in northeastern Italy, where ‘Yukon’ exhibited the earliest spring green-up in a bermudagrass varietal comparison. Moreover, similar results have been published by the NTEP’s 2007 bermudagrass trial (NTEP, 2011b), reporting that ‘Riviera’ and ‘Yukon’ showed a significantly faster green-up than ‘Princess-77’ in four locations across the United States. However, the slow spring green-up of ‘Sea Spray’ seashore paspalum relative to

bermudagrass cultivars have not been reported in previous studies (Severmutlu *et al.*, 2011), in which turf was irrigated to prevent drought stress. Moreover, the lower precipitations and higher temperatures occurred in spring 2010 than spring 2011 (Table 1), together with the significant cultivar \times year interaction (Fig. 5), could indicate a high water requirement of ‘Sea Spray’ seashore paspalum relative to bermudagrasses.

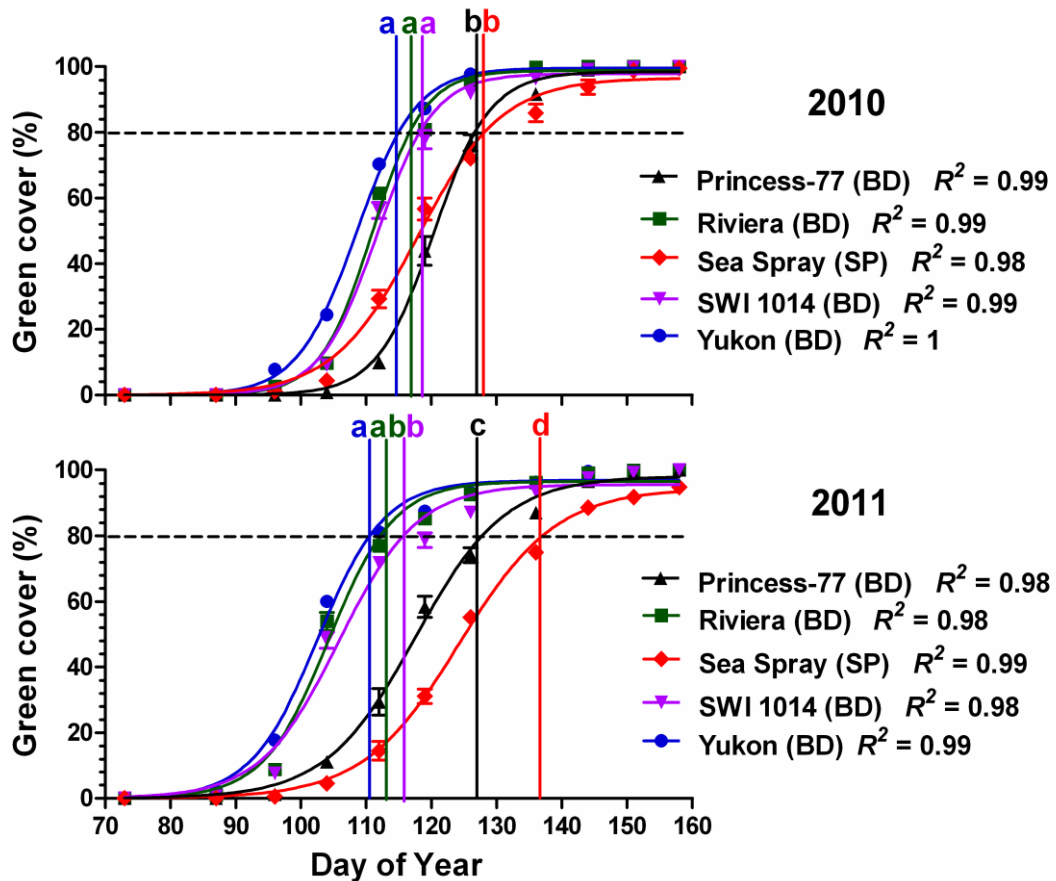


Figure 5 – Percent green cover as a function of day of the year for four bermudagrass (BD) cultivars and ‘Sea Spray’ seashore paspalum (SP) in 2010 and 2011. Data points represent an average of three N fertilization schedules and three replicates; bars indicate \pm standard error. Within each spring, line means sharing a letter are not statistically different according to Tukey’s honestly significant difference test ($\alpha = 0.05$).

In the spring 2010, turf plots N fertilized five times per year (Schedule C) showed earlier green-up than those receiving N in three applications (Schedule A), however the differences were of little practical importance (Table 7). In the following spring, differences in spring green-up occurred among all three N fertilization treatments, with Schedule C

reaching D80 5 d earlier than Schedule A. When data were averaged over the two springs, plots receiving N until August delayed spring green-up by 4 d compared to those N fertilized also in September and October (Table 7). These results concur to previous studies that reported an advantage in spring green-up for bermudagrass after late-season N applications (Goatley *et al.*, 1994; Richardson, 2002; Munshaw *et al.*, 2006).

Table 6 – Days of year to reach 80% green-up (based on sigmoidal model estimates) of four bermudagrass (BD) cultivars and ‘Sea Spray’ seashore paspalum (SP). Data were averaged over three N fertilization schedules and two years, and were collected in Legnaro, northeastern Italy.

Princess-77 (BD)	Riviera (BD)	Sea Spray (SP)	SWI 1014 (BD)	Yukon (BD)
Time to reach 80% green-up (d)				
127 c†	114 ab	133 d	117 b	113 a

† Means sharing a letter are not statistically different according to Tukey’s honestly significant difference test ($\alpha = 0.05$).

Table 7 – Days of year to reach 80% green-up (based on sigmoidal model estimates) of warm season turfgrasses under three N fertilization schedules in spring 2010 and spring 2011. Data were averaged over four bermudagrass cultivars and ‘Sea Spray’ seashore paspalum, and were collected in Legnaro, northeastern Italy.

Nitrogen applications†	Spring 2010	Spring 2011	Overall
Time to reach 80% green-up (d)			
May, June, Aug.	122 b‡	123 c	123 c
May, June, Aug., Oct.	121 ab	120 b	120 b
May, June, Aug., Sept., Oct.	120 a	118 a	119 a

† The three N fertilization schedules provided 20 g m⁻² yr⁻¹ of N.

‡ Within each column, means sharing a letter are not statistically different according to Tukey’s honestly significant difference test ($\alpha = 0.05$).

Across years, the overall green-up was achieved on similar calendar dates, as indicated by the non significant spring effect for D80 (Table 3). When green cover percentages were regressed against spring-accumulated GDD, the GDDs required to reach 80% green cover responded to cultivar, N fertilization, and cultivar × year consistently to D80 (data not presented). Whereas, different sigmoidal associations were found for the overall averages of spring 2010 and 2011, with 92 GDDs difference between years to reach 80% green cover (Table 8; Fig. 6). These results indicate that spring-accumulated GDD was not robust as a solely predictor of green-up. Different responses of turf spring regrowth to

GDD could be possibly due to other environmental differences between years, such as winter hardening conditions, spring evapotranspiration, and incident radiation (Rogers *et al.*, 1977; Rimi *et al.*, 2011; Schiavon *et al.*, 2011).

Table 8 – Equation parameters for sigmoidal regression models relating percent green cover of warm season turfgrasses and cumulative growing degree days (GDD; base = 5 °C) during spring (1 Mar.–30 June) of 2010 and 2011 in Legnaro, northeastern Italy.

Source	Spring 2010	Spring 2011	P value
Bottom	0.0	0.0	—
Top	97.8	96.0	0.2171
Halfway between bottom and top	340.7	385.5	<.0001
Slope	59.4	89.9	<.0001
GDD to reach 80% green-up	430	522	<.0001

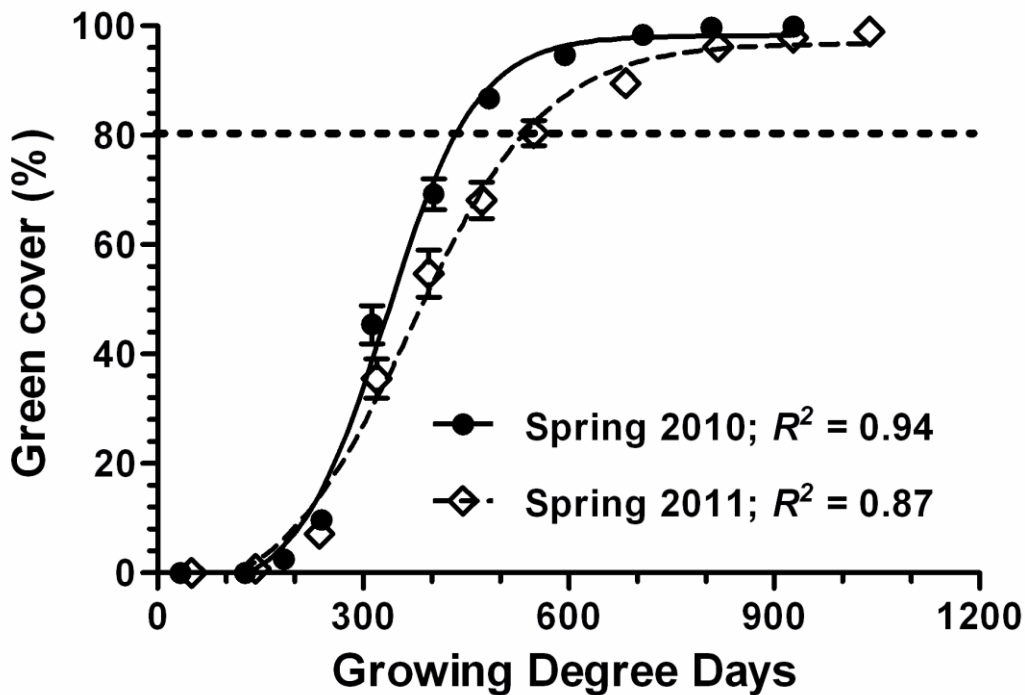


Figure 6 – Percent green cover as a function of cumulative growing degree days (base = 5 °C) during spring (1 Mar.–30 June) of 2010 and 2011 for warm season turfgrasses. Data points represent an average of five cultivars, three N fertilization schedules, and three replicates; bars indicate \pm standard error.

Green cover visual ratings, DIA estimates, and NDVI readings of the test grasses were strongly and positively correlated during the spring green-up (Table 9). The close

association occurred between visual ratings and DIA estimates of green cover agreed with the results of Richardson *et al.* (2001), reporting high correlation between DIA and subjective analyses. These results confirmed that photographic techniques can be effectively used as unbiased substitutes of visual assessments for turf green cover (Richardson *et al.*, 2001). In addition, the strong correlation detected between visual ratings and NDVIs concur to several previous findings that reported a positive correlation between turf color and reflectance values (Bell *et al.*, 2004; Bell & Xiong, 2007). High effectiveness of NDVI optical sensor in detecting green cover can be ascribed to the positive correlation between leaf chlorophyll and near infra-red reflectance (Adcock *et al.*, 1990; Blackmer *et al.*, 1994).

Table 9 – Pearson correlation coefficients relating green cover visual ratings, digital image analysis (DIA) estimates, and normalized difference vegetation index (NDVI) readings of four bermudagrass (BD) cultivars and ‘Sea Spray’ seashore paspalum (SP). Data were collected biweekly from 15 Mar. to 15 June of 2010 and 2011 on plots subjected to three N fertilization schedules in Legnaro, northeastern Italy; and were averaged over three replicates ($n = 36$). Correlations were significant at $P < 0.0001$.

Cultivar	Parameter	r	
		NDVI	DIA
Princess-77 (BD)	Visual ratings	0.992	0.990
	NDVI	—	0.992
Riviera (BD)	Visual ratings	0.927	0.874
	NDVI	—	0.981
SWI 1014 (BD)	Visual ratings	0.938	0.956
	NDVI	—	0.985
Sea Spray (SP)	Visual ratings	0.986	0.992
	NDVI	—	0.981
Yukon (BD)	Visual ratings	0.981	0.913
	NDVI	—	0.970

Digital image analysis estimates and NDVIs were also strongly correlated (Table 9), however different relationships between DIA estimates and visual ratings or NDVI readings occurred for each cultivar (Fig. 7). Whereas, visual ratings and NDVIs fitted on the same line (Fig. 7), suggesting that optical sensor and human evaluator responded similarly to varietal differences. Canopy structure and leaf color intensity, which can widely differ between grasses, could have affected the DIA estimates in a contrasting

manner compared to visual ratings and NDVIs. According to this, reflectance measurements seemed more appropriate than photographic techniques as instrumental substitutes for human evaluation of spring green-up in cultivar-by-cultivar studies.

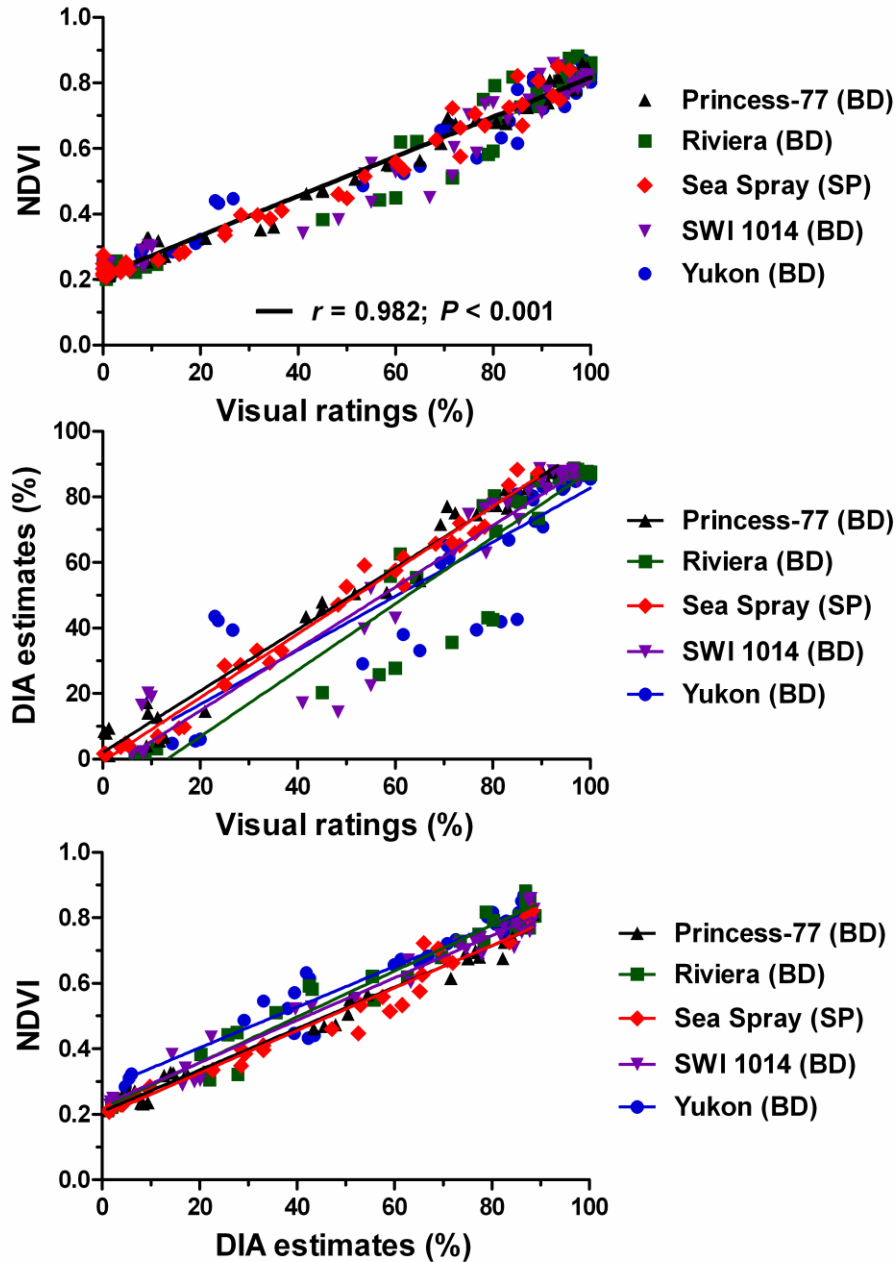


Figure 7 – Linear relationships between green cover visual ratings, Normalized Difference Vegetative Indices (NDVI), and Digital Image Analysis (DIA) estimates of warm season turfgrasses in Legnaro, northeastern Italy. Data were collected biweekly from 15 Mar. to 15 June of 2010 and 2011 on four bermudagrass (BD) cultivars and ‘Sea Spray’ seashore paspalum (SP) subjected to three N fertilization schedules, and were averaged over three replicates ($n = 36$).

Summer quality

Cultivars, months, and their interactions affected summer quality, while N fertilization schedules and all interactions including N fertilization schedule were not significant (Table 3), therefore the data were combined over N fertilization schedules (Table 10). In June 2010, ‘Yukon’ displayed highest quality, whereas visual quality of ‘Sea Spray’ was lowest, and quality of ‘Princess-77’, ‘Riviera’, and ‘SWI 1014’ fell between the other two grasses (Table 10). In June of the following year, ‘Yukon’ provided the highest visual quality, followed by ‘Princess-77’ and ‘SWI 1014’, while ‘Riviera’ and ‘Sea Spray’ had lowest quality (Table 10). In July and August of both years, ‘Sea Spray’ seashore paspalum generally showed lower quality than bermudagrass cultivars, which did not differ between each other (Table 10). Acceptable turfgrass quality (i.e., visual rating of 6.0) was not achieved by ‘Sea Spray’ seashore paspalum in July and August possibly due to a high water requirement relative to bermudagrasses under non irrigated conditions. No differences among the test grasses were observed for turf visual quality in both Sept. 2010 and Sept. 2011 (Table 10).

Table 10 – Monthly visual quality of four bermudagrass (BD) cultivars and ‘Sea Spray’ seashore paspalum (SP) from June to September of 2010 and 2011. Data were averaged over three N fertilization schedules and were collected in Legnaro, northeastern Italy.

Month	Princess-77 (BD)	Riviera (BD)	Sea Spray (SP)	SWI 1014 (BD)	Yukon (BD)	Average
Visual rating†						
2010						
June	7.1 ab‡	6.7 ab	6.2 b	6.9 ab	7.4 a	6.8 A‡
July	7.4 a	6.6 a	5.4 b	6.7 a	7.5 a	6.7 A
August	7.2 a	7.0 a	5.8 b	7.0 a	6.8 ab	6.8 A
September	6.6	6.5	6.5	6.4	6.2	6.5 BC
2011						
June	7.1 ab	6.1 b	6.1 b	6.8 ab	7.4 a	6.7 AB
July	7.4 a	6.6 a	5.4 b	6.7 a	7.5 a	6.7 A
August	6.9 a	6.8 a	5.2 b	6.7 a	6.5 a	6.4 C
September	6.6	6.5	6.3	6.4	6.2	6.4 C
Average	7.0 a	6.6 ab	5.9 b	6.7 a	6.9 a	—

† Visual quality rating on a 1–9 scale, where 1 = dead, 6 = acceptable, 9 = most desirable.

‡ Means sharing a letter are not statistically different according to Tukey’s honestly significant difference test ($\alpha = 0.05$). Lowercase letters denote differences between cultivars (in rows); uppercase letters denote differences of the mean values between months.

Table 11 – Pearson correlation coefficients resulting from linear correlation of normalized difference vegetation index and Log_{10} [visual quality ratings] of warm season turfgrasses from June to September of 2010 and 2011 in Legnaro, northeastern Italy. Data were collected weekly on four bermudagrass cultivars and ‘Sea Spray’ seashore paspalum subjected to three N fertilization schedules with three replicates ($n=180$).

Statistic	2010				2011			
	June	July	Aug.	Sept.	June	July	Aug.	Sept.
<i>r</i>	0.288	0.365	0.322	0.659	0.313	0.211	0.655	0.521
<i>P</i>	0.0018	0.0058	0.0061	<.0001	<.0001	0.0045	<.0001	<.0001

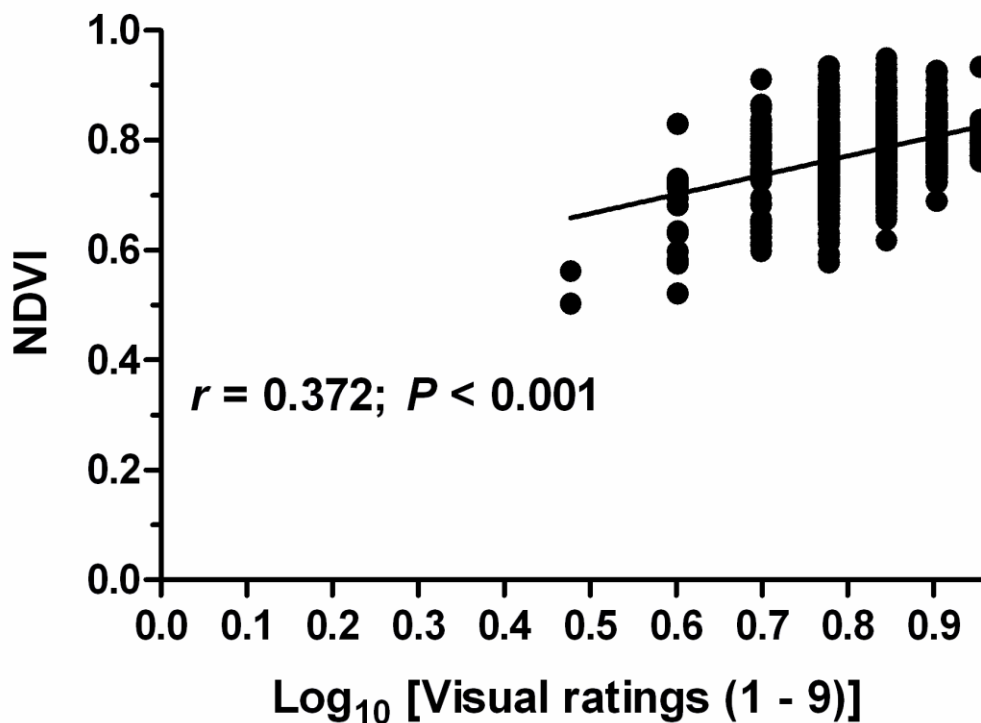


Figure 8 – Correlation between Log_{10} [Visual quality ratings] and Normalized Difference Vegetation Indices (NDVI) of warm season turfgrasses in Legnaro, northeastern Italy. Data were collected weekly from June to September of 2010 and 2011 on four bermudagrass cultivars and ‘Sea Spray’ seashore paspalum subjected to three N fertilization schedules, with three replicates ($n = 1440$).

When the data were averaged over months, ‘Princess-77’, ‘SWI 1014’, and ‘Yukon’ had the highest visual quality, while ‘Sea Spray’ displayed poorest quality (Table 10). On average, all bermudagrass cultivars tested provided a summer visual quality of 6.6 to 7.0 under study conditions. Correlations of NDVI readings and visual ratings resulted in correlation coefficients that ranged from 0.21 to 0.66, depending on month (Table 11).

Among months, sensor measurements were better correlated to human evaluations in Sept. 2010, Aug. 2011, and Sept. 2011 (Table 11), when average grass performances were generally lowest (Table 10). These results suggest that potentials of NDVI in predicting turfgrass visual quality may vary across time probably due to its sensitivity to environmental stresses (Xiong *et al.*, 2007). The overall correlation between visual ratings and NDVI yielded a significant but weak correlation (Fig. 8), due to a wide spread of reflectance values for each individual rating. These results are consistent to previous findings that reported a weak correlation between the two variables for both warm season and cool season grasses (Haendel & Wissemeier, 2008; Bunderson *et al.*, 2009; Sevostianova *et al.*, 2011).

Fall color retention

All possible two-way interactions were significant for fall color retention, however there were no interactions among cultivars, N fertilization schedules, and months (Table 3), consequently the data were pooled over N fertilization schedules (Table 12), months (Table 13), and cultivars (Table 14), respectively. With exception of Sept. 2011, ‘Sea Spray’ seashore paspalum displayed better fall color compared to other grasses (Table 12), indicating high ability to prolong color retention relative to bermudagrasses. Among bermudagrass cultivars, differences were noticed in November of both years, with ‘Princess-77’ maintained color longer than ‘Riviera’ and ‘Yukon’ in 2010; and longer than ‘Yukon’ in 2011 (Table 12). These findings together with cultivar differences observed for spring regrowth (Table 6; Fig. 5) suggest that bermudagrasses characterized by early fall discoloration may green-up faster compared to those prolonging fall color retention. These results are similar to the reports of Munshaw *et al.* (2006), who found early fall discoloration in association with cold-tolerance for bermudagrass cultivars. As expected, there was an average loss of fall color from September to November of both years (Table 12). However, monthly differences of 2011 were of higher magnitude compared to those of 2010, which can be explained by severe drops in air temperature between months of 2011 (Table 1).

Table 12 – Fall color ratings of four bermudagrass (BD) cultivars and ‘Sea Spray’ seashore paspalum (SP) from September to November of 2010 and 2011. Data were averaged over three N fertilization schedules and were collected in Legnaro, northeastern Italy.

Month	Princess-77	Riviera	Sea Spray	SWI 1014	Yukon	Average
	(BD)	(BD)	(SP)	(BD)	(BD)	
Visual rating†						
2010						
September	8.2 b‡	8.3 b	9.0 a	8.1 b	8.1 b	8.4 B‡
October	7.2 b	7.1 b	9.0 a	6.9 b	6.9 b	7.4 C
November	5.7 b	4.6 c	6.8 a	5.2 b	4.4 c	5.3 D
2011						
September	8.7	8.6	9.0	8.5	8.5	8.7 A
October	7.1 b	7.1 b	8.5 a	6.8 b	7.3 b	7.3 C
November	4.8 b	4.2 bc	6.3 a	4.5 bc	4.1 c	4.8 E
Average	7.0 b	6.7 bc	8.1 a	6.7 bc	6.6 c	—

† Color rating on a 1–9 scale, where 1 = dormant and 9 = lush green.

‡ Means sharing a letter are not statistically different according to Tukey’s honestly significant difference test ($\alpha = 0.05$). Lowercase letters denote differences between cultivars (in rows); uppercase letters denote differences of the mean values between months.

Table 13 – Fall color ratings of four bermudagrass (BD) cultivars and ‘Sea Spray’ seashore paspalum (SP) subjected to three N fertilization schedules in Legnaro, northeastern Italy. Data were averaged over six months of evaluation (Sept.–Nov. 2010 and 2011).

Cultivar	Nitrogen Applications†		
	May, June, Aug.	May, June, Aug., Oct.	May, June, Aug., Sept., Oct.
	Visual rating‡		
Princess (BD)	6.5 b§	7.0 a	7.3 a
Riviera (BD)	6.3 c	6.7 b	7.0 a
Sea Spray (SP)	8.1	8.0	8.1
SWI 1014 (BD)	6.3 b	6.6 b	7.2 a
Yukon (BD)	6.1 b	6.6 a	6.9 a
Average	6.7 c	7.0 b	7.3 a

† The three N fertilization schedules provided $20 \text{ g m}^{-2} \text{ yr}^{-1}$ of N.

‡ Color rating on a 1–9 scale, where 1 = dormant and 9 = lush green.

§ Within each row, means sharing a letter are not statistically different according to Tukey’s honestly significant difference test ($\alpha = 0.05$).

Averaged over the six evaluation months, the effects of N fertilization schedule on fall color retention varied depending on grasses, with ‘Sea Spray’ showed no benefit from late-season N applications (Table 13). For all bermudagrass cultivars, a delay in fall

discoloration was observed in plots that received N up to October (Schedule B & C) compared to those N fertilized until August (Schedule A) (Table 13). These results are in agreement with previous findings that reported on the possibility to prolong fall color retention of bermudagrass by late-season N applications (Reeves *et al.*, 1970; White & Schmidt, 1990; Richardson, 2002; Munshaw *et al.*, 2006). This study also pointed out the possibility to prolong the practicability of non-overseeded bermudagrass turf by delaying fall color discoloration under the same annual amount of N fertilizer. Pooled over five cultivars, in October of both years the plots N fertilized five times per year (Schedule C) also displayed better color than those N fertilized four times (Schedule B) (Table 14).

Table 14 – Fall color ratings from September to November of 2010 and 2011 for warm season turfgrasses subjected to three N fertilization schedules in Legnaro, northeastern Italy. Data were averaged over four bermudagrass cultivars and ‘Sea Spray’ seashore paspalum.

Month	Nitrogen Applications†		
	May, June, Aug.	May, June, Aug., Oct.	May, June, Aug., Sept., Oct.
Visual rating‡			
2010			
September	8.3	8.5	8.3
October	7.3 b§	7.3 b	7.8 a
November	4.7 c	5.4 b	5.9 a
2011			
September	8.5 b	8.6 ab	8.9 a
October	7.1 b	7.2 b	7.7 a
November	4.2 b	4.9 a	5.3 a

† The three N fertilization schedules provided 20 g m⁻² yr⁻¹ of N.

‡ Color rating on a 1–9 scale, where 1 = dormant and 9 = lush green.

§ Within each row, means sharing a letter are not statistically different according to Tukey’s honestly significant difference test ($\alpha = 0.05$).

There was a strong positive correlation between color ratings and NDVI readings collected from September to November (Fig. 9), as observed between green cover visual ratings and NDVIs in spring (Table 9; Fig. 7). These results indicated that reflectance measurements have high potentials as substitutes for subjective turf fall color ratings, thus its use could avoid human bias in color evaluations (Horst *et al.*, 1984; Karcher & Richardson, 2003). Moreover, color visual ratings tend to be discrete rather than

continuous, as suggested by Karcher & Richardson (2003); therefore collecting NDVIs instead may avoid issues for applicability of standard analysis of variance.

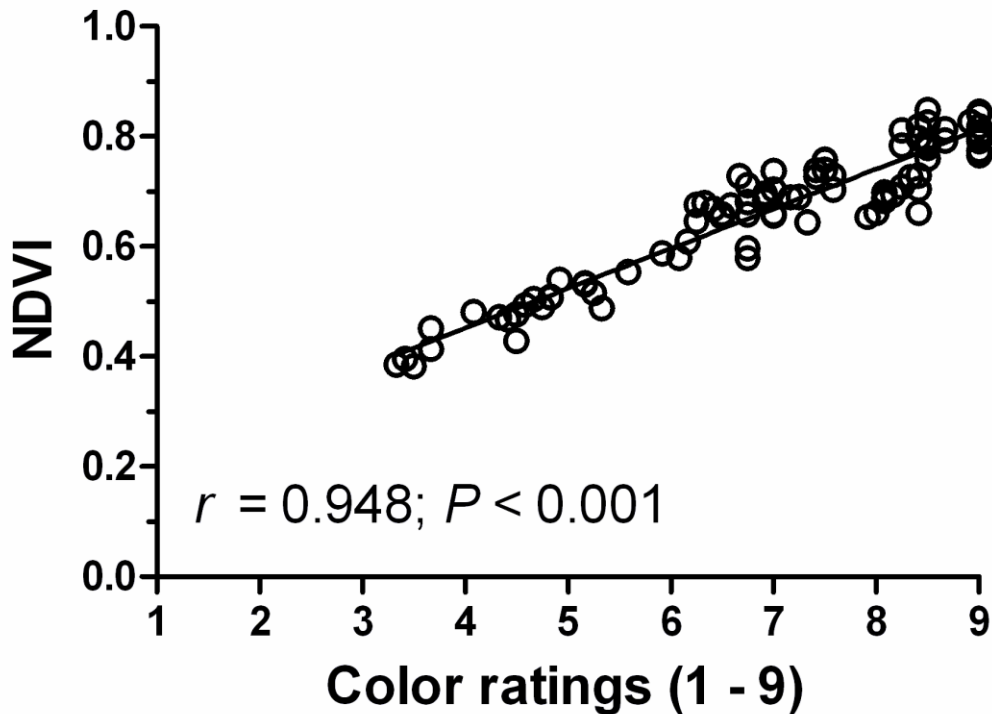


Figure 9 – Correlation between fall color ratings and Normalized Difference Vegetation Indices (NDVI) of four bermudagrass cultivars and ‘Sea Spray’ seashore paspalum subjected to three N fertilization schedules. Data were collected monthly (average of four weeks) from September to November of 2010 and 2011 in Legnaro, northeastern Italy; and were averaged over three replicates ($n = 90$).

Canopy turf effective eight was measured with advance of fall discoloration (October – November) and results revealed that cultivar and N fertilization schedule had no effects on vertical growth (Table 3). The fact N fertilization schedules did not affect vertical growth during the acclimation disagreed with common knowledge, as it has been longer reported that late-season N applications stimulate shoot growth (Harrison, 1931; Carroll & Welton, 1939; Beard, 1973). When the data were pooled over cultivars and N fertilization schedules, increases in turf height were observed only in early Oct. 2010 and mid Oct. 2011 (Fig. 10). The lack of a significant interaction between cultivars and dates (Table 3) suggests that the test grasses can be managed in a similar manner with regard to cessation of mowing.

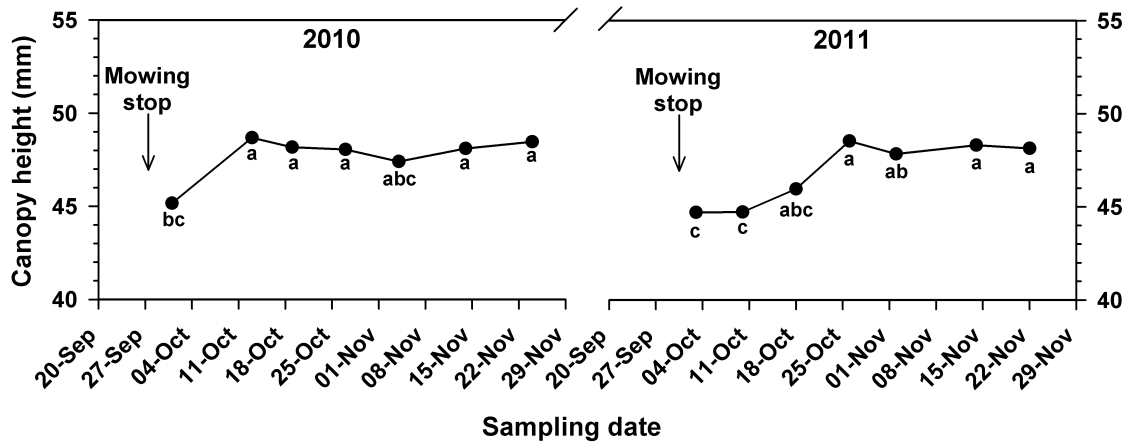


Figure 10 – Turf canopy height of warm season turfgrasses as affected by sampling date from October to November of 2010 and 2011 in Legnaro, northeastern Italy. Data points represent the average of five cultivars, three N fertilization schedules, and three replicates. Means sharing a letter are not statistically different according to Tukey’s honestly significant difference test ($\alpha = 0.05$).

Stolon and rhizome measurements

The analysis of variance revealed significant interactions between cultivars and sampling dates for all measurements (stolon and rhizome dry weight, stolon internodes diameter, length and mass), whereas N fertilization schedules and all their interactions were not significant (Table 4), consequently the data were pooled over N fertilization schedule.

Differences in stolon dry weight among grasses were consistent throughout Year 1 and Year 2, although there was a difference in magnitude that resulted in a cultivar \times sampling date interaction (Fig. 11). Compared with other grasses, ‘Riviera’ had highest mass of stolon on all sampling dates, with dry weight declined from 1643 to 1336 g m⁻² in Year 1 and from 2028 to 1515 g m⁻² in Year 2 (Fig. 11). With the exception of Sept. 2010, differences in stolon dry weight were not detected among the other cultivars (Fig. 11). Morphological development of stolons has been indicated a a primary factor involved in freeze avoidance, and a high stolon density may be of crucial importance for winter survival under traffic conditions (Hensler *et al.*, 1999; Trenholm *et al.*, 2000; Anderson *et al.*, 2007). The results of this study revealed a large difference in stolon mass between

grasses, pointing out that the choice of species/cultivar should be highly considered for managing athletic fields.

When the data were averaged over sampling dates, stolon dry weight of ‘Princess-77’ was significantly lower than other bermudagrass cultivars and not different than ‘Sea Spray’ seashore paspalum (Fig. 11). These results, together with late spring green-up observed for ‘Princess-77’ and ‘Sea Spray’ (Fig. 5), may suggest poor adaptability to cold winter temperatures for these two grasses. In the average of five cultivars, dry weight of stolon was higher in Year 2 than Year 1 on each sampling date with the exception of April (Table 15). With the nearness of spring green-up, the magnitude of decreases in stolon dry weight differed between Year 1 and Year 2 (Table 15). Stolon dry weight increased between Sept. 2009 and Sept. 2010, pointing out that stolons actively expand over years in warm season turf canopies. These results agreed with several previous studies and recommendations of textbooks, which suggest to periodically reduce the stolon mass of warm season grasses by vertical mowing (Beard, 1973; Christians, 1998; Rowland *et al.*, 2009).

For each cultivar tested, patterns for dry weight of rhizome were widely different between Year 1 and Year 2, leading to an interaction between cultivar and sampling date (Table 4, Fig. 12). During Year 1, ‘Riviera’ generally had higher rhizome mass than ‘Princess-77’ and did not differ from the other cultivars. In the second year, differences among cultivars for rhizome dry weight were detected only in Sept. 2010 and Oct. 2010. Rhizome dry weight of ‘Riviera’ decreased from 114 to 61 g m⁻² in Year 1 and from 110 to 8 g m⁻² in Year 2, while that of ‘Princess-77’ declined from 20 to 7 g m⁻² in Year 1 and from 20 to 0.5 g m⁻² in Year 2 (Fig. 12). When the data were pooled over sampling dates, ‘Riviera’ had higher mass of rhizome than ‘SWI 1014’, while ‘Princess-77’ had lowest rhizome dry weight compared to other cultivars (Fig. 12). Limitedly to bermudagrass cultivars, overall differences in rhizome dry weight were generally consistent to those detected for the mass of stolon (Figs. 11 & 12). These results indicate that bermudagrasses widely differed in producing stolons and rhizomes as storage organs, as suggested by de Kroon *et al.* (1994) who reported a marked non-plastic variation in spacer organs of clonal plants.

Averaged over cultivars, rhizome dry weight was smaller in Year 2 than Year 1 on each sampling date with the exception of September (Table 15). These results are similar to those observed for stolon dry weight, but opposite in nature, which suggests that plant tendency in producing stolons or rhizomes may diverge as canopy age advances. In Year 1, the mass of rhizome decreased between Sept. 2009 and Nov. 2009, and between Feb. 2010 and Apr. 2010 (Table 15). In Year 2, significant decreases of rhizome dry weight were observed between Sept. 2010 and Oct. 2010, and between Mar. 2011 and Apr. 2011 (Table 15). These findings indicated a seasonality of rhizome development, with renewed growth during the active growing season, a neat drop in autumn (October – November), and a subsequent decrease in spring with green-up.

Table 15 – Stolon and rhizome dry weight of warm season turfgrasses as affected by sampling date in Legnaro, northeastern Italy, during Sept. 2009–Apr. 2010 (Year 1) and Sept. 2010–Apr. 2011 (Year 2). Rhizome dry weight was log-transformed for data analysis, then means were back transformed for presentation. Data were averaged over five cultivars and three N fertilization schedules.

Sampling date	Stolon dry weight	Rhizome dry weight
	g m^{-2}	
Year 1		
19 Sept. 2009	1062 cd†	77.7 a
17 Oct. 2009	1003 cdef	54.5 abc
14 Nov. 2009	1059 cd	43.8 bcd
5 Dec. 2009	1024 cde	57.3 abc
16 Jan. 2010	927 efgh	56.4 ab
12 Feb. 2010	946 defg	49.2 bc
21 Mar. 2010	897 fgh	41.0 cde
17 Apr. 2010	808 h	31.3 def
Year 2		
21 Sept. 2010	1294 a	56.0 abc
16 Oct. 2010	1278 a	25.0 efg
12 Nov. 2010	1235 ab	17.7 fg
11 Dec. 2010	1230 ab	24.3 efg
15 Jan. 2011	1213 ab	12.7 gh
11 Feb. 2011	1121 bc	15.5 gh
9 Mar. 2011	1040 cde	19.2 fg
15 Apr. 2011	855 gh	5.8 h

† Within each column, means sharing a letter are not statistically different according to Tukey's honestly significant difference test ($\alpha = 0.05$).

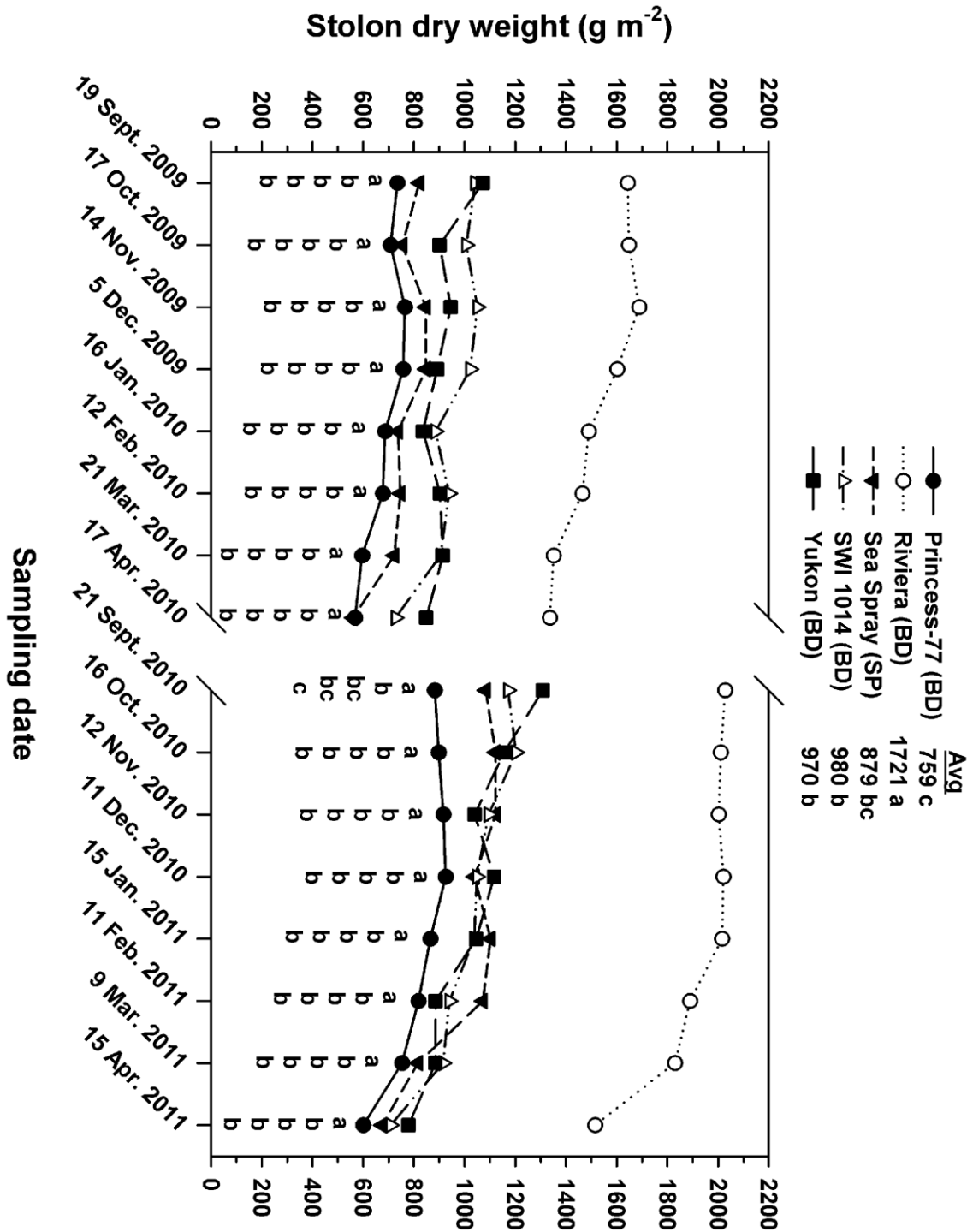


Figure 11 – Stolon dry weight of four bermudagrass (BD) cultivars and ‘Sea Spray’ seashore paspalum (SP) as affected by sampling date in Legnaro, northeastern Italy, during Sept. 2009–Apr. 2010 and Sept. 2010–Apr. 2011. Data points represent the average of three N fertilization schedules and three replicates. Within each date, means sharing a letter are not statistically different according to Tukey’s honestly significant difference test ($\alpha = 0.05$).

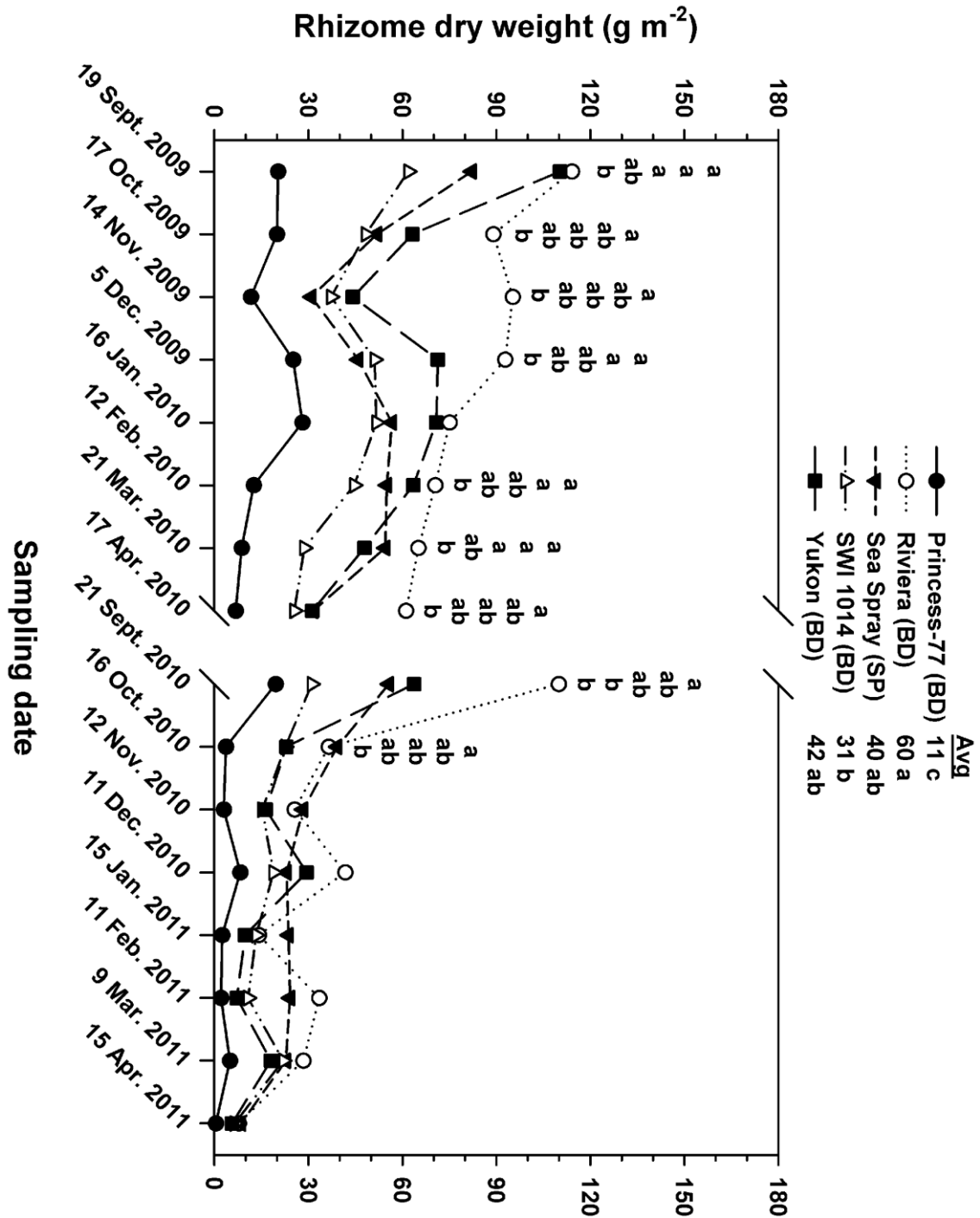


Figure 12 – Rhizome dry weight of four bermudagrass (BD) cultivars and ‘Sea Spray’ seashore paspalum (SP) as affected by sampling date in Legnaro, northeastern Italy, during Sept. 2009–Apr. 2010 and Sept. 2010–Apr. 2011. Data were log-transformed for data analysis, then means were back transformed for presentation. Data points represent the average of three N fertilization schedules and three replicates. Within each date, means sharing a letter are not statistically different according to Tukey’s honestly significant difference test ($\alpha = 0.05$). Mean separation statistics were calculated for transformed data.

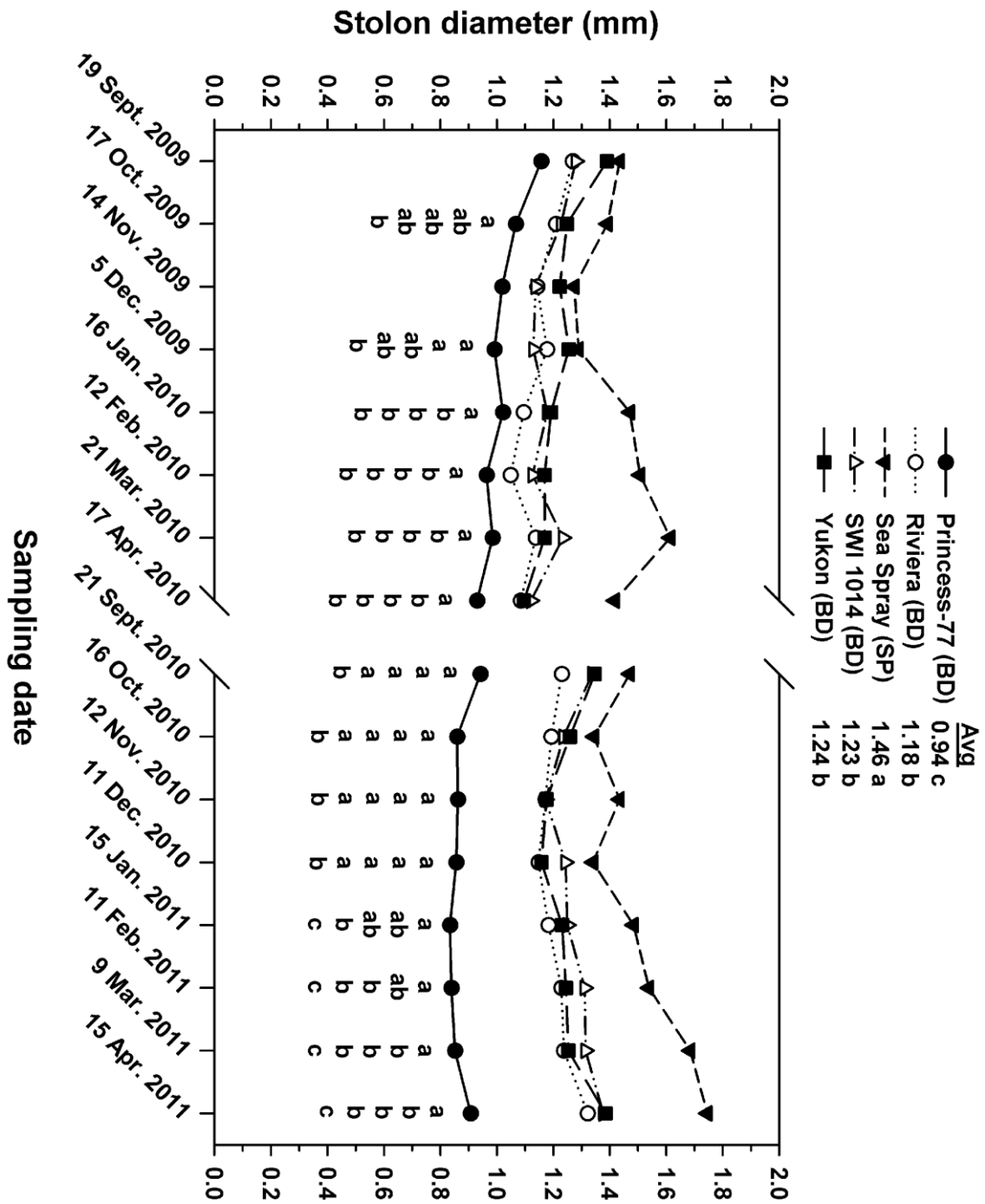


Figure 13 – Stolon diameter of four bermudagrass (BD) cultivars and ‘Sea Spray’ seashore paspalum (SP) as affected by sampling date in Legnaro, northeastern Italy, during Sept. 2009–Apr. 2010 and Sept. 2010–Apr. 2011. Data points represent the average of three N fertilization schedules and three replicates. Within each date, means sharing a letter are not statistically different according to Tukey’s honestly significant difference test ($\alpha = 0.05$).

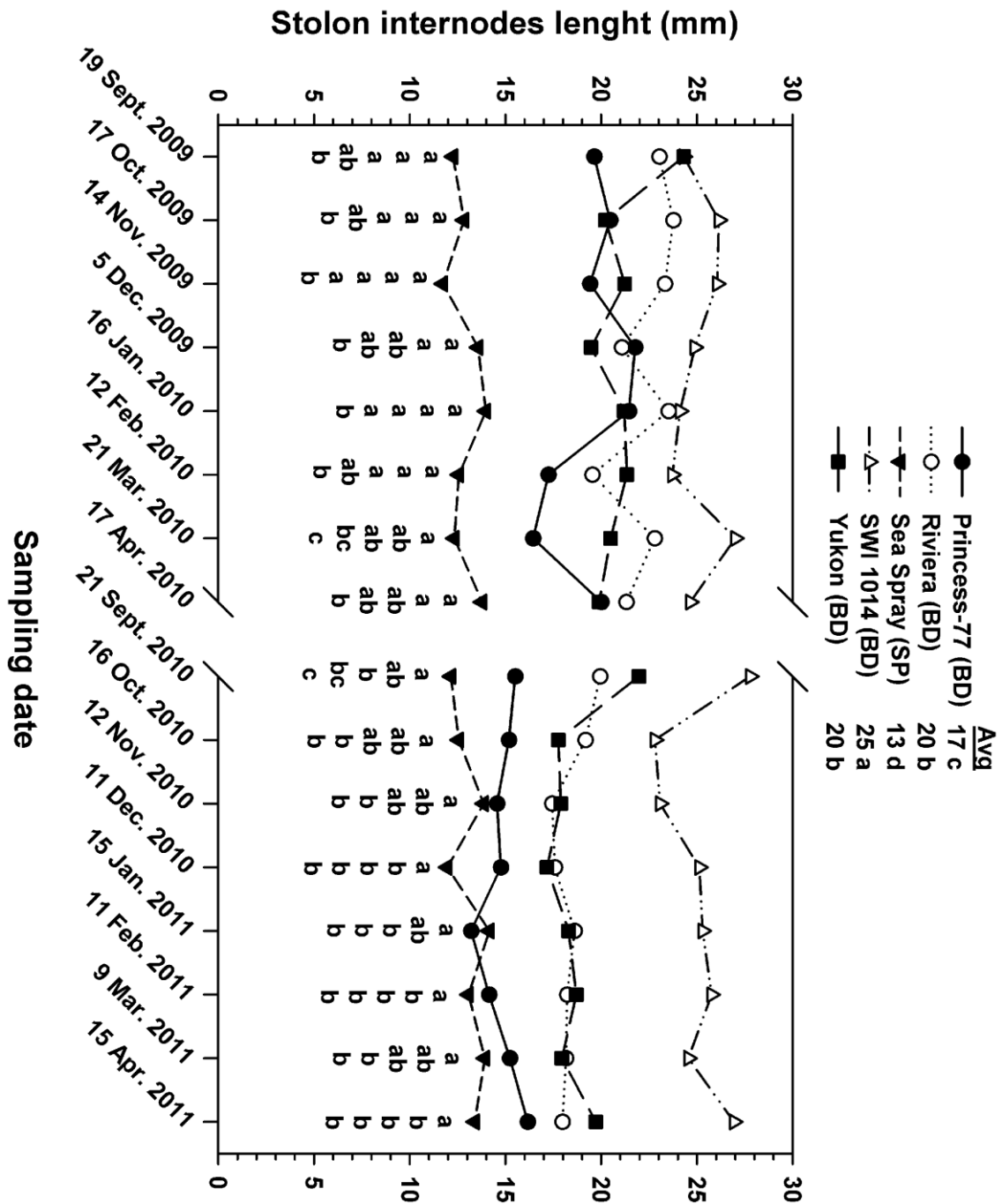


Figure 14 – Stolon internodes length of four bermudagrass (BD) cultivars and ‘Sea Spray’ seashore paspalum (SP) as affected by sampling date in Legnaro, northeastern Italy, during Sept. 2009–Apr. 2010 and Sept. 2010–Apr. 2011. Data points represent the average of three N fertilization schedules and three replicates. Within each date, means sharing a letter are not statistically different according to Tukey’s honestly significant difference test ($\alpha = 0.05$).

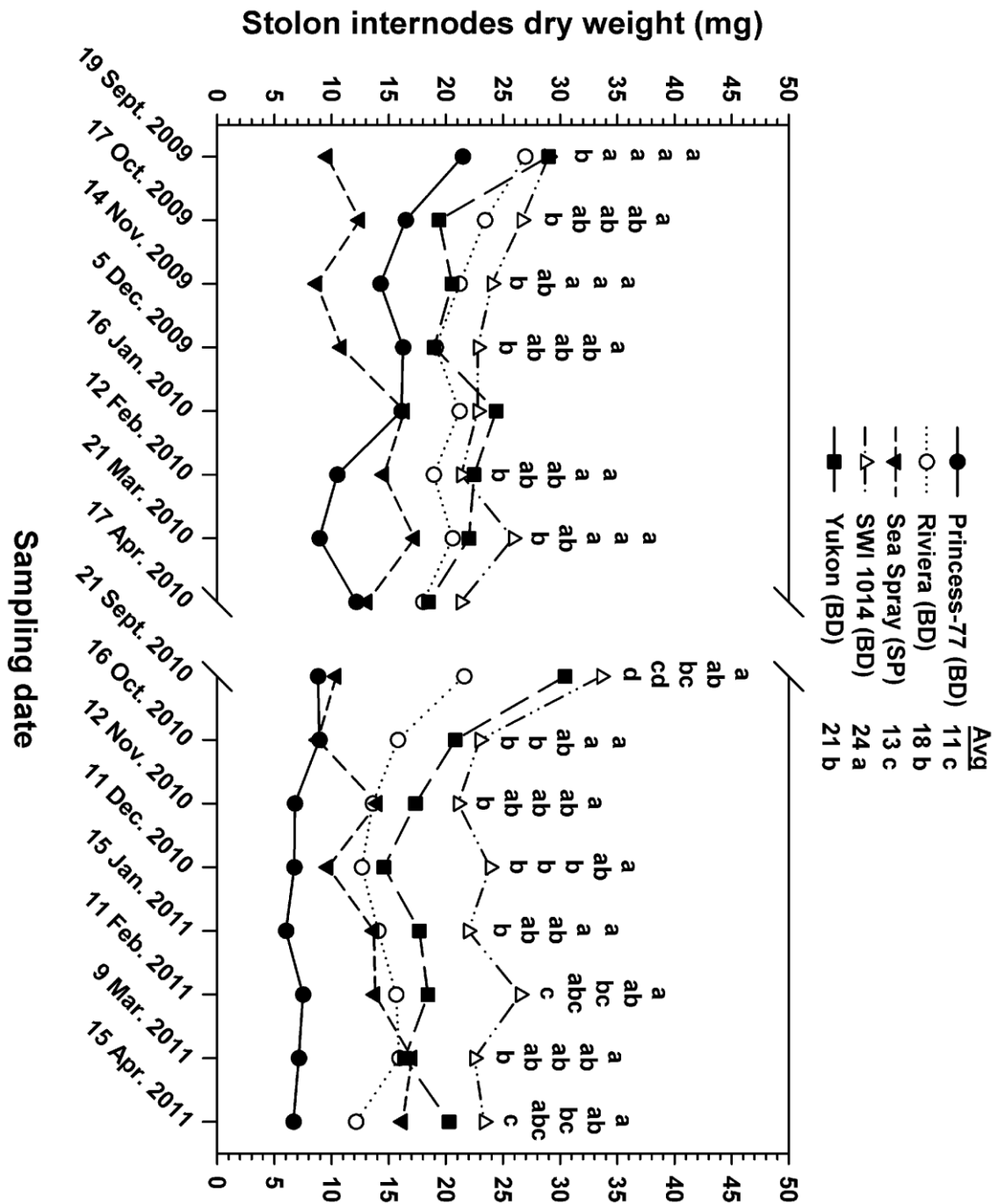


Figure 15 – Stolon internodes mass of four bermudagrass (BD) cultivars and ‘Sea Spray’ seashore paspalum (SP) as affected by sampling date in Legnaro, northeastern Italy, during Sept. 2009–Apr 2010 and Sept. 2010–Apr 2011. Data points represent the average of three N fertilization schedules and three replicates. Within each date, means sharing a letter are not statistically different according to Tukey’s honestly significant difference test ($\alpha = 0.05$).

Cultivars interacted with sampling dates in determining diameter, length, and mass of stolon internodes (Table 4), although the interactions were differently related to environmental differences within and between years (Table 1; Figs. 13, 14, & 15). With exception of Sept. 2009 and Nov. 2009, stolon diameter of ‘Sea Spray’ seashore paspalum was higher than of ‘Princess-77’ on all sampling dates (Fig. 13). The magnitude of differences in stolon diameter between cultivars varied markedly between sampling dates, with ‘Princess-77’ displayed the finest diameter throughout Year 2 (Fig. 13). When data were pooled over sampling dates, stolon diameter of ‘Sea Spray’ was 0.52 mm higher than of ‘Princess-77’, and diameter of ‘Riviera’, ‘SWI 1014’, and ‘Yukon’ differed from the other two cultivars (Fig. 13). Stolon internodes length of ‘SWI 1014’ was greater than of ‘Sea Spray’ seashore paspalum on all sampling dates (Fig. 14). ‘Sea Spray’ generally displayed the shortest stolon internodes in Year 1, while its internodes length was not different than ‘Princess-77’, ‘Riviera’, and ‘Yukon’ since Oct. 2010 (Fig. 14). Averaged over sampling dates, ‘SWI 1014’ had the longest internodes of stolon, followed by ‘Yukon’, ‘Riviera’, ‘Princess-77’, and ‘Sea Spray’ seashore paspalum, respectively (Fig. 14).

Patterns for internodes mass of each grass widely differed between Year 1 and Year 2 (Fig. 15), as observed for the other morphological features of stolon (Figs. 11, 13, & 14). For example, in Year 1 ‘Princess-77’ had internodes mass smaller than ‘SWI 1014’ and ‘Yukon’ only on Feb. 2010 and Mar. 2010, while its mass was the smallest throughout Year 2 (Fig. 15). When the data were averaged over sampling dates, ‘SWI 1014’ had the greatest mass of stolon internodes, followed by ‘Yukon’ and ‘Riviera’, then by ‘Sea Spray’ and ‘Princess-77’, respectively (Fig. 15). Collectively, results of internodes traits indicated that stolon internodes of ‘Sea Spray’ seashore paspalum were shorter and coarser compared to those of bermudagrass cultivars. The stolon internodes of ‘SWI 1014’ were longest and heaviest compared to other grasses, those of ‘Princess-77’ were finest and lightest, while those of ‘Riviera’ and ‘Yukon’ were intermediate. It has been reported that stolon morphological traits of bermudagrass are genetically controlled (de Kroon *et al.*, 1994), and they have been studied with regard to turf quality (Dudeck & Murdock, 1998; Roche & Loch, 2005) and cold tolerance (Hensler *et al.*, 1999; Anderson *et al.*, 2007). While differences in summer quality among bermudagrass cultivars were of limited importance under study conditions (Table 10), there was disparity in spring green-up (Fig. 5) and fall

color retention (Table 12). Stolon morphological features varied widely among test bermudagrasses, suggesting that internodes traits could have more influence on early- and late-season turf performance than on summer quality, under low-maintenance conditions (e.g., non irrigated areas, weekly mowed lawns).

Table 16 – Stolon internodes diameter, length, and mass of warm season turfgrasses as affected by sampling date in Legnaro, northeastern Italy, during Sept. 2009–Apr. 2010 (Year 1) and Sept. 2010–Apr. 2011 (Year 2). Data were averaged over five cultivars and three N fertilization schedules.

Sampling date	Stolon diameter	Stolon internodes length	Stolon internodes mass
	mm		mg
Year 1			
19 Sept. 2009	13.1 ab†	20.7 a	23.2 a
17 Oct. 2009	12.3 bcd	20.7 a	19.7 abc
14 Nov. 2009	11.6 de	20.3 ab	17.7 bcde
5 Dec. 2009	11.7 de	20.2 abc	17.6 bcde
16 Jan. 2010	11.9 cde	20.8 a	20.2 abc
12 Feb. 2010	11.6 de	18.8 abcde	17.6 bcde
21 Mar. 2010	12.2 bcd	19.8 abcd	18.9 bcd
17 Apr. 2010	11.3 e	19.9 abc	16.6 cdef
Year 2			
21 Sept. 2010	12.7 abc	19.4 abcde	21.0 ab
16 Oct. 2010	11.8 cde	17.5 de	15.5 def
12 Nov. 2010	11.6 de	17.4 e	14.6 ef
11 Dec. 2010	11.5 de	17.3 e	13.5 f
15 Jan. 2011	12.0 cde	17.9 cde	14.7 ef
11 Feb. 2011	12.3 bcd	18.0 bcde	16.4 cdef
9 Mar. 2011	12.7 abc	18.0 bcde	15.8 def
15 Apr. 2011	13.5 a	18.8 abcde	15.7 def

† Within each column, means sharing a letter are not statistically different according to Tukey's honestly significant difference test ($\alpha = 0.05$).

When the data were averaged over cultivars, patterns for stolon internodes traits were widely different between Year 1 and Year 2 (Table 16), according to the concept of phenotypic plasticity (Dong & de Kroon, 1994; de Kroon & Hutchings, 1995; Dong & Pierdominici, 1995). Stolon diameter, internodes length, and dry weight in Sept. 2009 were not different than in Sept. 2010 (Table 16). These findings, together with the increment of stolon mass occurred between Year 1 and Year 2 (Table 15) suggest that no. internodes m^{-2}

increased from an active growing season (i.e., 2010) to another (i.e., 2011). Internodes of stolon collected in Sept. 2009 were coarser and heavier compared to those collected in April 2010 (Table 16). During Year 2, mass of internodes in Sept. 2010 was highest compared to other dates; and diameter of stolon in Apr. 2011 was coarser than of those collected from Oct. 2010 to Feb. 2011 (Table 16). Instead, when sampling dates of Year 1 and Year 2 were observed separately, there were no differences among dates for stolon internodes length (Table 16). These results suggest that diameter and mass of stolon internodes may be of greater interest than internodes length for further research on cold tolerance of warm season grasses.

Carbohydrates and protein in stolons

Cultivar and N fertilization schedule significantly affected starch content of stolons, and there were significant interactions between cultivars and sampling dates, and among N fertilization schedules and sampling dates (Table 5). However, the interaction terms cultivar \times N fertilization schedule and cultivar \times N fertilization schedule \times sampling date were not significant, therefore the responses of cultivar and N fertilization schedule were combined over N fertilization schedules and cultivars, respectively. Compared with other grasses, 'Riviera' generally had the highest content of starch in stolons on all sampling dates, whereas starch content was lowest for 'Sea Spray' seashore paspalum (Fig. 16). Starch content in 'Princess-77', 'SWI-1014', and 'Yukon' were intermediate than the other two grasses, and the range of difference occurred between each other varied from 20 (non significant) to 82 g m⁻², depending on sampling date (Fig. 16). When the data were averaged over sampling dates, 'Riviera' had higher content of starch in stolons than other cultivars, followed by 'SWI 1014' and 'Yukon', then by 'Princess-77', and 'Sea Spray', respectively (Fig. 16). These results suggest that seashore paspalum and bermudagrass differ in starch accumulation, although the study was unbalanced in that regard.

Differences among N fertilization schedules in stolon content of starch were observed only in Sept. 2009, Apr. 2010, and Dec. 2010 (Fig. 17). However, when the data were averaged over sampling dates, starch content in stolons of grasses N fertilized three

times per year (Schedule A) was 6% higher than of those N fertilized five times (Schedule C). These results concur to those of Goatley *et al.* (1994), who found that late-season N had limited effect on carbohydrates content of bermudagrass rhizomes. In the average of cultivars and N fertilization schedules, starch concentration declined from 215 to 120 g kg⁻¹ in Year 1, and from 182 to 86 g kg⁻¹ in Year 2 (Table 17). In the area of study, increases in starch were not observed between the sampling dates of September and October of both years. These findings agreed with previous studies reporting that warm season grasses accumulate starch in the storage organs before winter and subsequently use it to support spring regrowth (Okajima & Smith, 1964; Hofstra & Nelson, 1969; Bender & Smith, 1973). The lower content of starch observed in Year 2 compared to Year 1 may be explained by higher dry weight of stolon (Table 15), which could have increased intra-specific competition (Beard, 1973; Firbank & Watkinson, 1985).

Differences in stolon content of WSC among cultivars were consistent throughout the study, although there was a difference in magnitude leading to an interaction between cultivar and sampling date (Fig. 18). Content of WSC in stolons of both ‘Riviera’ and ‘Yukon’ was higher than of ‘Princess-77’ and ‘Sea Spray’ seashore paspalum on ten of the 16 sampling dates (Fig. 18). ‘Yukon’ had a stolon content of WSC significantly higher than ‘Riviera’ in Dec. 2010 and values of ‘SWI 1014’ generally overlapped with other grasses throughout the study (Fig. 18). Averaged over sampling dates, ‘Yukon’ had the highest content of WSC compared to other cultivars, followed by ‘Riviera’, then by ‘SWI 1014’, whereas ‘Princess-77’ and ‘Sea Spray’ had lowest concentration (Fig. 18). These findings, together with differences in starch detected among cultivars (Fig.16), indicated that cultivars characterized by high WSC accumulation (e.g., ‘Yukon’) can differ from those high-starch accumulating (e.g., ‘Riviera’). Moreover, these results corroborated those of Macolino *et al.* (2010), who reported that WSC in stolons during the winter were higher for ‘Yukon’ compared to ‘Princess-77’ and ‘Riviera’.

Stolon WSC was greater in grasses N fertilized until August (Schedule A) than in those that received N until October (Schedule B & C) (Table 18), regardless of cultivar or sampling date as dictated by non significant interaction terms including N fertilization schedule (Table 5). These results are consistent to the response of starch (Fig. 17), as the difference in carbohydrates appeared of limited biological importance for both molecular

groups with regard to N fertilization schedules. When the data were pooled over cultivars and N fertilization schedules, the trends for WSC appeared widely different between Year 1 and Year 2 (Table 17). Stolon WSC markedly increased from Sept. 2009 to Nov. 2009, then gradually decreased between Jan. 2010 and Mar. 2010, and subsequently dropped in Apr. 2010 with spring green-up (Table 17). Instead, stolon content of WSC was generally constant from Sept. 2010 to Jan. 2011, then increased markedly between Jan. 2011 and Feb. 2011, and subsequently declined in Apr. 2011 (Table 17). When comparing the two experimental years, seasonal trends of WSC appeared less regular than those of starch, suggesting that the two molecular groups were contrastingly affected by environmental factors.

Table 17 – Stolon content of starch, water soluble carbohydrates (WSC), and crude protein of warm season turfgrasses as affected by sampling date in Legnaro, northeastern Italy, during Sept. 2009–Apr. 2010 (Year 1) and Sept. 2010–Apr. 2011 (Year 2). Data were averaged over five cultivars and three N fertilization schedules.

Sampling date	Starch	WSC	Crude protein
	g kg ⁻¹ dry wt		
Year 1			
19 Sept. 2009	215 a†	87 e	50 gh
17 Oct. 2009	204 ab	107 c	52 g
14 Nov. 2009	185 cd	121 a	61 f
5 Dec. 2009	176 de	122 a	76 cd
16 Jan. 2010	164 ef	123 a	75 cd
12 Feb. 2010	159 fg	111 bc	83 ab
21 Mar. 2010	142 hi	108 bc	83 ab
17 Apr. 2010	120 j	79 f	85 a
Year 2			
21 Sept. 2010	182 cd	95 de	44 hi
16 Oct. 2010	195 bc	93 de	43 i
12 Nov. 2010	176 de	89 de	64 ef
11 Dec. 2010	174 de	88 e	65 ef
15 Jan. 2011	148 gh	96 d	66 ef
11 Feb. 2011	142 hi	117 ab	68 e
9 Mar. 2011	131 ij	122 a	77 bc
15 Apr. 2011	86 k	88 e	70 de

† Within each column, means sharing a letter are not statistically different according to Tukey's honestly significant difference test ($\alpha = 0.05$).

Table 18 – Water soluble carbohydrates content in stolons of warm season turfgrasses subjected to three N fertilization schedules in Legnaro, northeastern Italy. Data were averaged over five cultivars and 16 sampling dates.

Nitrogen Applications†		
May, June, Aug.	May, June, Aug., Oct.	May, June, Aug., Sept., Oct.
g kg ⁻¹ dry wt		
105.9 a‡	101.7 b	101.3 b

† The three N fertilization schedules provided 20 g m⁻² yr⁻¹ of N.

‡ Means sharing a letter are not statistically different according to Tukey's honestly significant difference test ($\alpha = 0.05$).

Table 19 – Statistics of models relating growing degree days (base temperature = 5 °C) and total non-structural carbohydrates content in stolons per ground sq m of four bermudagrass (BD) cultivars and 'Sea Spray' seashore paspalum (SP) during Sept. 2009–Apr. 2010.

Cultivar	Model	Extra sum-of-square <i>F</i> test	AIC† differences	<i>R</i> ²
Princess-77 (BD)				
	Linear regression	————	0.0	0.18
	Segmental linear regression	<i>P</i> < 0.001	27.7	0.51
	Second order polynomial	<i>P</i> < 0.001	29.0	0.50
	Third order polynomial	<i>P</i> < 0.001	16.6	0.50
Riviera (BD)				
	Linear regression	————	0.0	0.38
	Segmental linear regression	<i>P</i> < 0.001	22.1	0.58
	Second order polynomial	<i>P</i> < 0.001	22.7	0.57
	Third order polynomial	<i>P</i> < 0.001	15.9	0.57
Sea Spray (SP)				
	Linear regression	————	0.0	0.26
	Segmental linear regression	<i>P</i> = 0.010	5.3	0.38
	Second order polynomial	<i>P</i> = 0.051	6.0	0.36
	Third order polynomial	<i>P</i> < 0.022	5.4	0.45
SWI 1014 (BD)				
	Linear regression	————	0.0	0.31
	Segmental linear regression	<i>P</i> < 0.001	21.9	0.52
	Second order polynomial	<i>P</i> < 0.001	23.2	0.52
	Third order polynomial	<i>P</i> < 0.012	19.5	0.52
Yukon (BD)				
	Linear regression	————	0.0	0.26
	Segmental linear regression	<i>P</i> = 0.078	0.8	0.32
	Second order polynomial	<i>P</i> = 0.039	2.2	0.31
	Third order polynomial	<i>P</i> < 0.071	0.7	0.31

† AIC = Akaike Information Criteria values

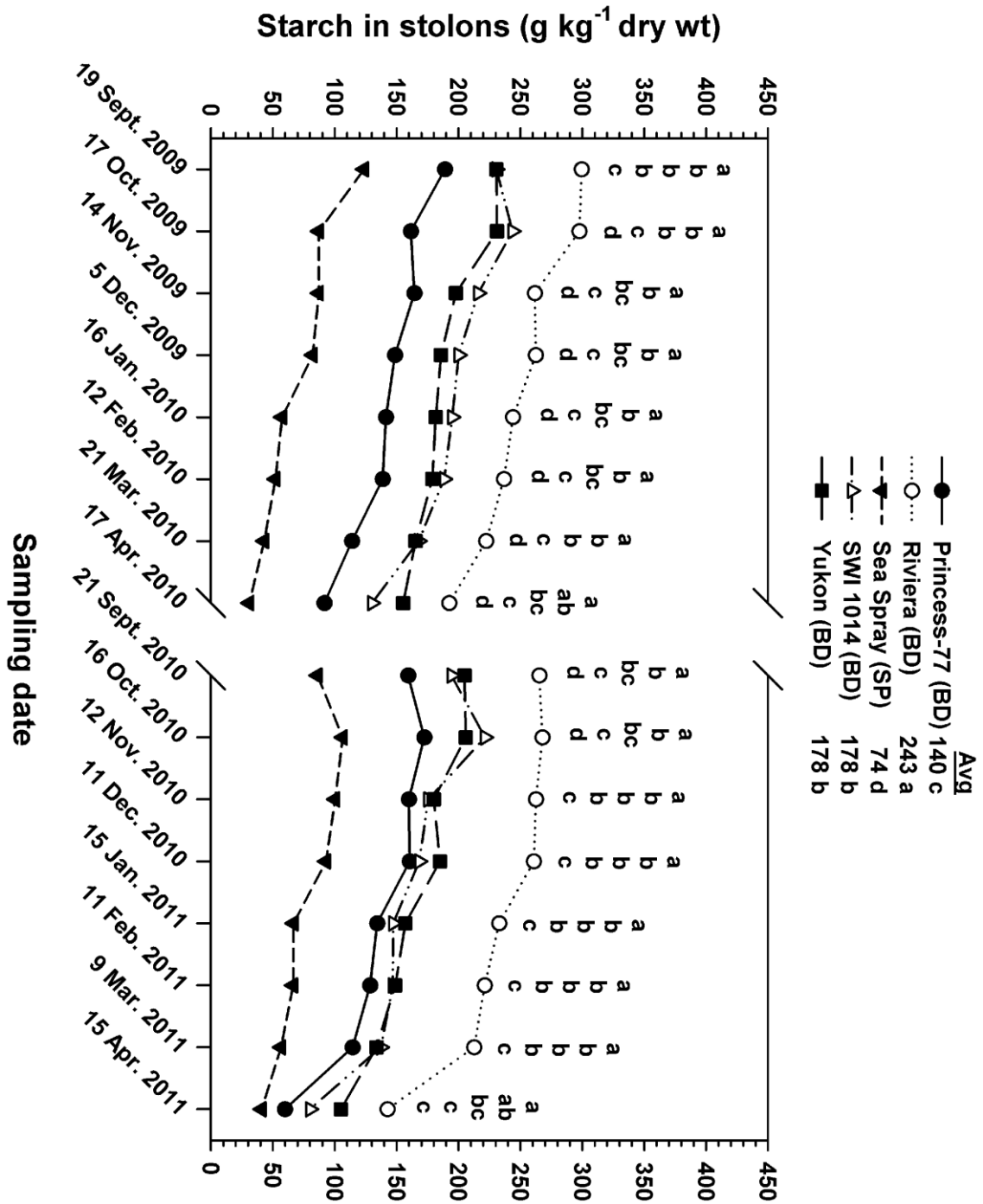


Figure 16 – Starch content in stolon of four bermudagrass (BD) cultivars and ‘Sea Spray’ seashore paspalum (SP) as affected by sampling date in Legnaro, northeastern Italy, during Sept. 2009–Apr. 2010 and Sept. 2010–Apr. 2011. Data points represent the average of three N fertilization schedules and three replicates. Within each date, means sharing a letter are not statistically different according to Tukey’s honestly significant difference test ($\alpha = 0.05$).

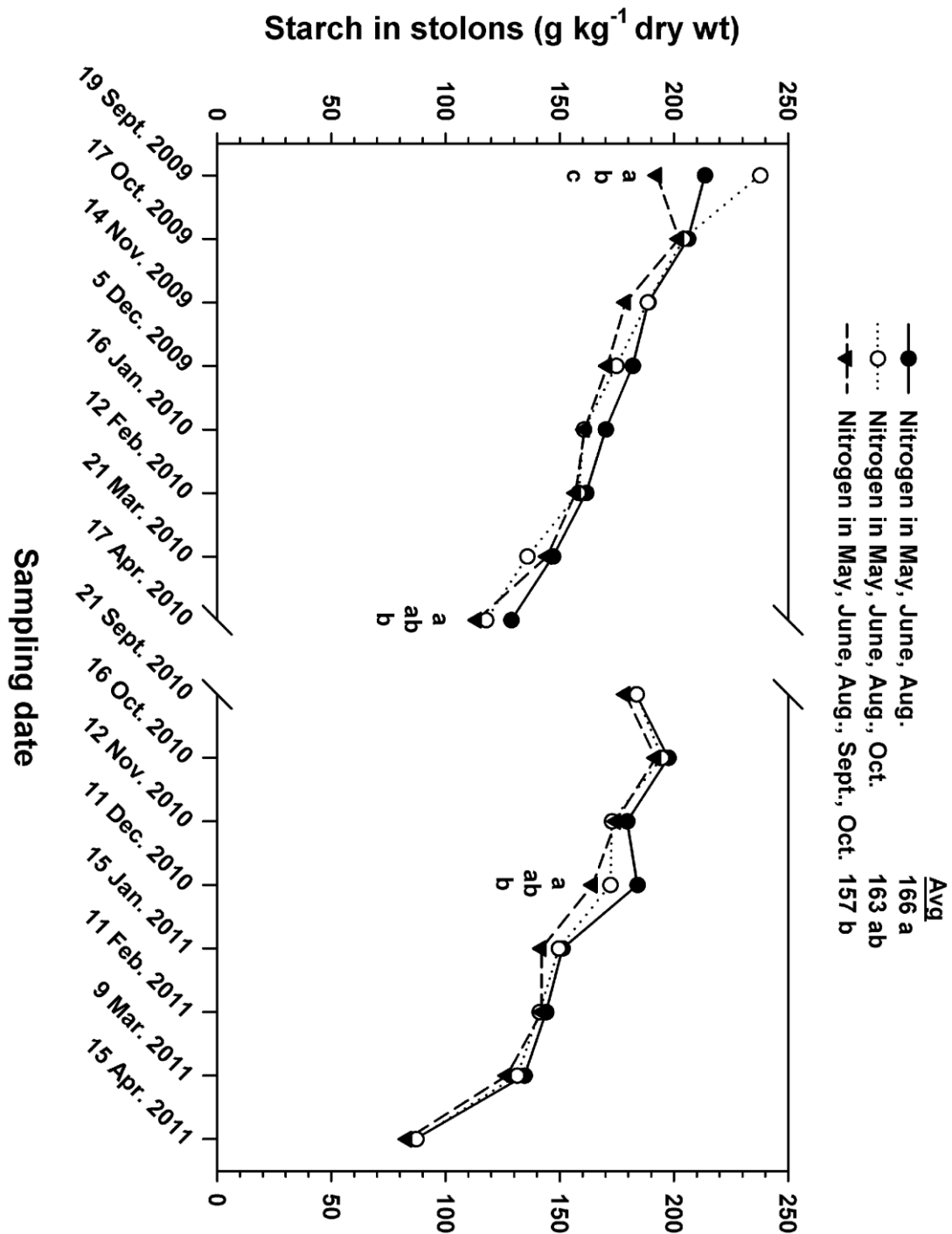


Figure 17 – Starch content in stolon of warm season turfgrasses subjected to three N fertilization schedules as affected by sampling date in Legnaro, northeastern Italy, during Sept. 2009–Apr. 2010 and Sept. 2010–Apr. 2011. Each N fertilization schedule received 20 g m⁻² year⁻¹ of N. Data points represent the average of five cultivars and three replicates. Within each date, means sharing a letter are not statistically different according to Tukey’s honestly significant difference test ($\alpha = 0.05$).

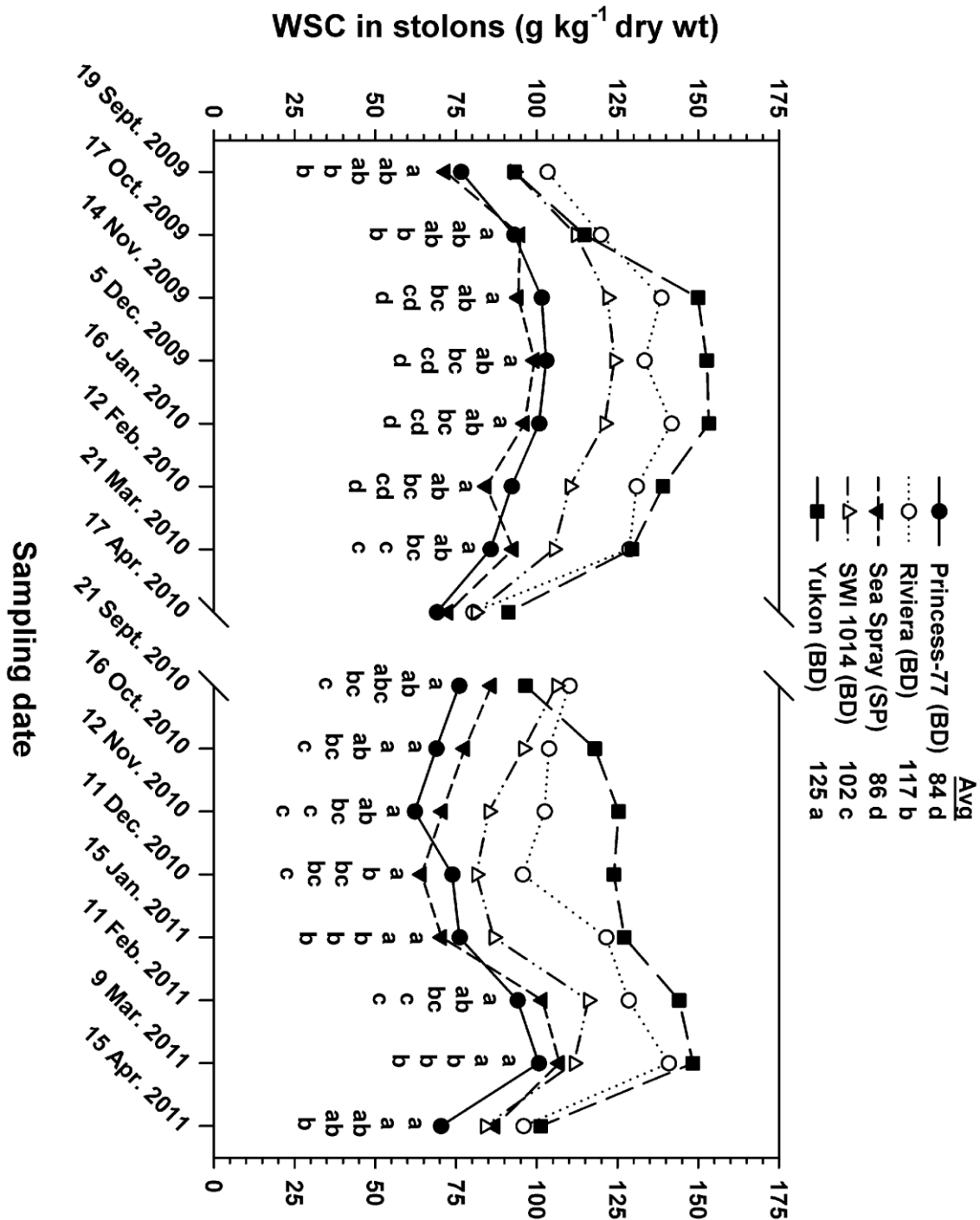


Figure 18 – Content of water soluble carbohydrates (WSC) in stolons of four bermudagrass (BD) cultivars and ‘Sea Spray’ seashore paspalum (SP) as affected by sampling date in Legnaro, northeastern Italy, during Sept. 2009–Apr. 2010 and Sept. 2010–Apr. 2011. Data points represent the average of three N fertilization schedules and three replicates. Within each date, means sharing a letter are not statistically different according to Tukey’s honestly significant difference test ($\alpha = 0.05$).

Table 20 – Statistics of models relating growing degree days (base temperature = 5 °C) and total non-structural carbohydrates content in stolons per ground sq m of four bermudagrass (BD) cultivars and ‘Sea Spray’ seashore paspalum (SP) during Sept. 2010–Apr. 2011.

Cultivar	Model	Extra sum-of-square <i>F</i> test	AIC† differences	<i>R</i>²
Princess-77 (BD)				
	Linear regression	————	0.0	0.35
	Segmental linear regression	<i>P</i> < 0.001	18.2	0.53
	Second order polynomial	<i>P</i> < 0.001	19.5	0.52
	Third order polynomial	<i>P</i> < 0.001	17.2	0.52
Riviera (BD)				
	Linear regression	————	0.0	0.43
	Segmental linear regression	<i>P</i> < 0.001	27.2	0.64
	Second order polynomial	<i>P</i> < 0.001	27.5	0.63
	Third order polynomial	<i>P</i> < 0.001	26.7	0.64
Sea Spray (SP)				
	Linear regression	————	0.0	0.47
	Segmental linear regression	<i>P</i> = 0.003	7.6	0.57
	Second order polynomial	<i>P</i> = 0.002	8.0	0.55
	Third order polynomial	<i>P</i> = 0.084	4.6	0.57
SWI 1014 (BD)				
	Linear regression	————	0.0	0.42
	Segmental linear regression	<i>P</i> = 0.126	-0.1	0.44
	Second order polynomial	<i>P</i> = 0.049	1.2	0.45
	Third order polynomial	<i>P</i> = 0.062	1.1	0.47
Yukon (BD)				
	Linear regression	————	0.0	0.52
	Segmental linear regression	<i>P</i> = 0.076	0.9	0.56
	Second order polynomial	<i>P</i> = 0.030	2.7	0.55
	Third order polynomial	<i>P</i> = 0.052	1.7	0.56

† AIC = Akaike Information Criteria values

For each cultivar, stolon content of both starch (Fig. 16) and WSC (Fig. 18) were differently affected as the result of time progress during Year 1 and Year 2. In addition, the interactions between cultivars and sampling dates for stolon dry weight were contrastingly related to the environmental differences occurred within Year 1 and Year 2 (Table 1, Fig. 11). Despite of these differences, quadratic relationship between ground area amount of TNC and cumulative GDD were identified for all cultivars in both Year 1 (Table 19) and Year 2 (Table 20). Beginning on 1 Sept., the ground area amount of TNC in stolons changed slightly from 300 to ≈800 GDD, then declined towards ≈1300 GDD, regardless of

the experimental Year (Table 21; Fig. 19). These findings suggest that TNC area amounts of various warm season grasses are similarly related to GDD, irrespectively of their stolon mass or concentrations of starch and WSC over time. From a general standpoint, these results concur to previous studies, documenting that GDD is a main environmental factor controlling the use and storage of carbohydrates in several grasses (Brown, 1939; Sullivan & Sprague, 1949; Miller, 1960).

Table 21 – Equation parameters resulting from the non-linear regression of total non-structural carbohydrates content in stolons per ground sq m against cumulative growing degree days (base temperature = 5 °C) for five warm season turfgrasses, during Sept. 2009–Apr 2010 (Year 1) and Sept. 2010–Apr. 2011 (Year 2). The data were collected over three N fertilization schedules, three replicates, and eight sampling dates per year ($n = 72$) in Legnaro, northeastern Italy.

Cultivar†	$a‡$	SEa	$b‡$	SEb	$c‡$	SEc
Year 1						
Princess-77 (BD)	67.62	29.38	0.3623	0.0718	-0.000263	0.000042
Riviera (BD)	520.10	69.46	0.6457	0.1784	-0.000580	0.000108
Sea Spray (SP)	112.40	36.13	0.1642	0.0892	-0.000154	0.000053
SWI 1014 (BD)	228.50	47.30	0.4696	0.1211	-0.000397	0.000073
Yukon (BD)	323.10	44.50	0.1119	0.1139	-0.000146	0.000069
Year 2						
Princess-77 (BD)	156.80	30.09	0.2550	0.0801	-0.000253	0.000051
Riviera (BD)	603.70	70.91	0.7170	0.1891	-0.000721	0.000121
Sea Spray (SP)	185.80	29.97	0.1048	0.0747	-0.000149	0.000046
SWI 1014 (BD)	382.60	61.08	0.0237	0.1627	-0.000193	0.000104
Yukon (BD)	409.30	45.91	0.0149	0.1223	-0.000173	0.000078

† BD = bermudagrass; SP = seashore paspalum.

‡ a , b , & c are coefficients of a quadratic equation, where $y = a + b x + c x^2$

Stolon content of CP was significantly affected by the interaction terms cultivar × sampling date and N fertilization schedule × sampling date, and all main effects were also significant (Table 5). However, the interactions between cultivars and N fertilization schedules, and among cultivars, N fertilization schedules, and sampling dates were not significant, therefore the data were pooled over N fertilization schedules and cultivars, respectively. ‘Princess-77’ had a content of CP in stolons higher than ‘Riviera’ and ‘Yukon’ on four of the 16 sampling dates (Jan. 2010, Mar. 2010, Apr. 2010, Apr. 2011) (Fig. 20). In addition, CP content in stolons of ‘Princess-77’ was greater than of ‘SWI 1014’ and ‘Yukon’ in Feb. 2010 (Fig. 20). Differences among ‘Riviera’, ‘Sea Spray’, ‘SWI 1014’,

and ‘Yukon’ were not detected in these dates, and there were no significant differences between cultivars in other sampling dates (Fig. 20). However, in the average of sampling dates, CP content in stolons of ‘Princess-77’ was 13–19 g kg⁻¹ higher compared to other grasses, which did not differ between each other (Fig. 20). These findings appeared of opposite nature compared to those of Zhang *et al.* (2006), who reported that ‘Riviera’ had greater leaf protein than ‘Princess-77’ after three weeks of cold acclimation in a growth chamber.

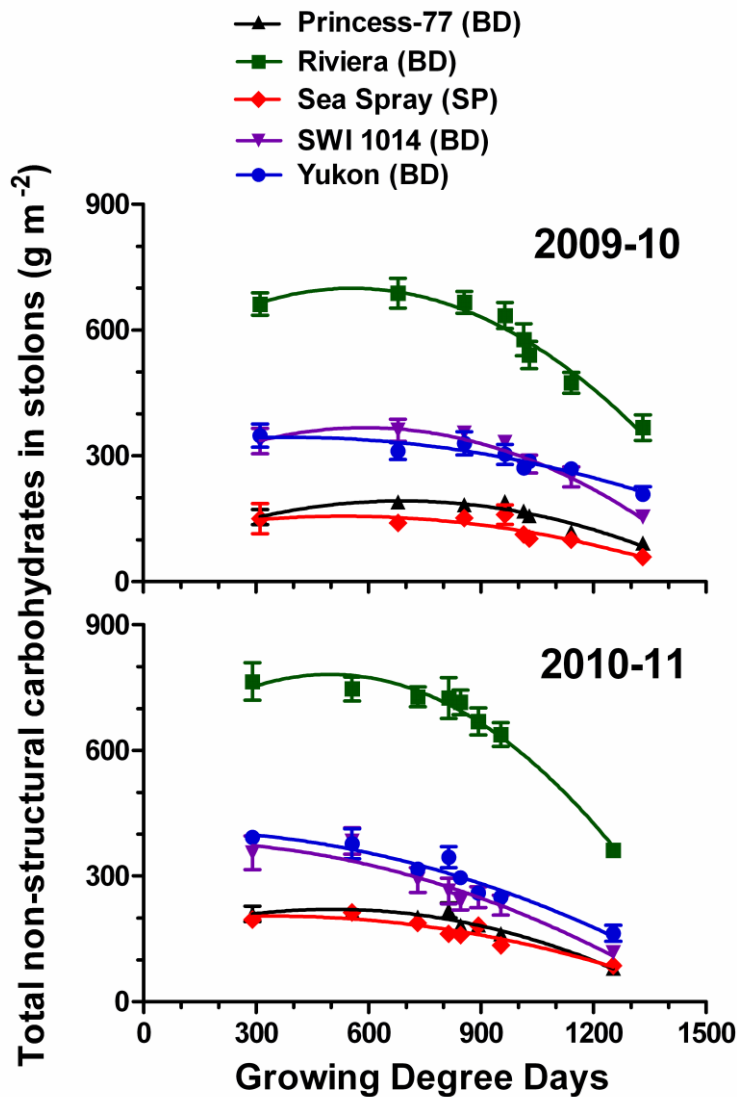


Figure 19 – Total non-structural carbohydrates content in stolons per ground sq m of four bermudagrass (BD) cultivars and ‘Sea Spray’ seashore paspalum (SP) as affected by cumulative growing degree days (base temperature = 5 °C) in Legnaro, northeastern Italy, during Sept. 2009–Apr. 2010 and Sept. 2010–Apr. 2011. Data points represent the average of three N fertilization schedules and three replicates; error bars indicate \pm standard error.

Among N fertilization schedules, grasses that received N in five seasonal applications (Schedule C) had a stolon content of CP higher than those N fertilized three times (Schedule A) on nine of the 16 sampling dates (Fig. 21). In addition, plots receiving N four times per year (Schedule B) had higher CP in stolon compared to plots N fertilized three times (Schedule A) in Dec. 2009, Nov. 2010, Dec. 2010, Jan. 2011, and Apr. 2011 (Fig. 21). In the average of sampling dates, CP content of plants subjected to Schedule C was 6% higher compared to Schedule B, and 11% higher than Schedule A (Fig. 21). The role of soluble protein as osmolyte in bermudagrass has been empathized by Richardson (2002), who hypothesized no deleterious effects of late-season N fertilization as a result of increased protein. In this study, responses of nonstructural carbohydrates to late-season N fertilization were opposite in nature compared to those of protein (Table 18; Figs. 17 & 21). However, differences between N fertilization schedules for starch and WSC appeared of little impact on spring green-up (Table 7) relative to those for CP. According to this, the increase of CP promoted by late-season applications of N could have over-covered for a reduction in carbohydrates, explaining the advantage in spring green-up.

When data were pooled over cultivars and N fertilization schedules, differences in stolon content of CP among dates within Year 1 were consistent to those occurred in Year 2 (Table 17). During the first Year of study, stolon CP increased gradually from Oct. 2009 to Dec. 2009, and subsequently between Jan. 2010 and Feb. 2010. In Year 2, increases in CP were observed between Oct. 2010 and Nov. 2010; and again between Feb. 2011 and Mar. 2011 (Table 17). Moreover, stolon CP content decreased by 93% during the growth season 2010 (Apr. 2010–Sept. 2010), indicating that plants had an accumulation peak during cold acclimation, and concentrated in late winter. Results for CP accumulation during cold acclimation agreed with several previous studies, reporting that various amino acids and soluble proteins serve as osmolytes to prevent cell freezing (Hsiao, 1973; Gatschet *et al.*, 1996; Munshaw *et al.*, 2006). In addition, the increase in stolon content of CP that occurred in late winter (February – March) can be explained by concurrent decrease in the mass of stolon. These findings together with the average trends for starch and WSC (Table 17) pointed out that grasses consumed carbohydrate reserves during dormancy, while there was a concentration of CP levels.

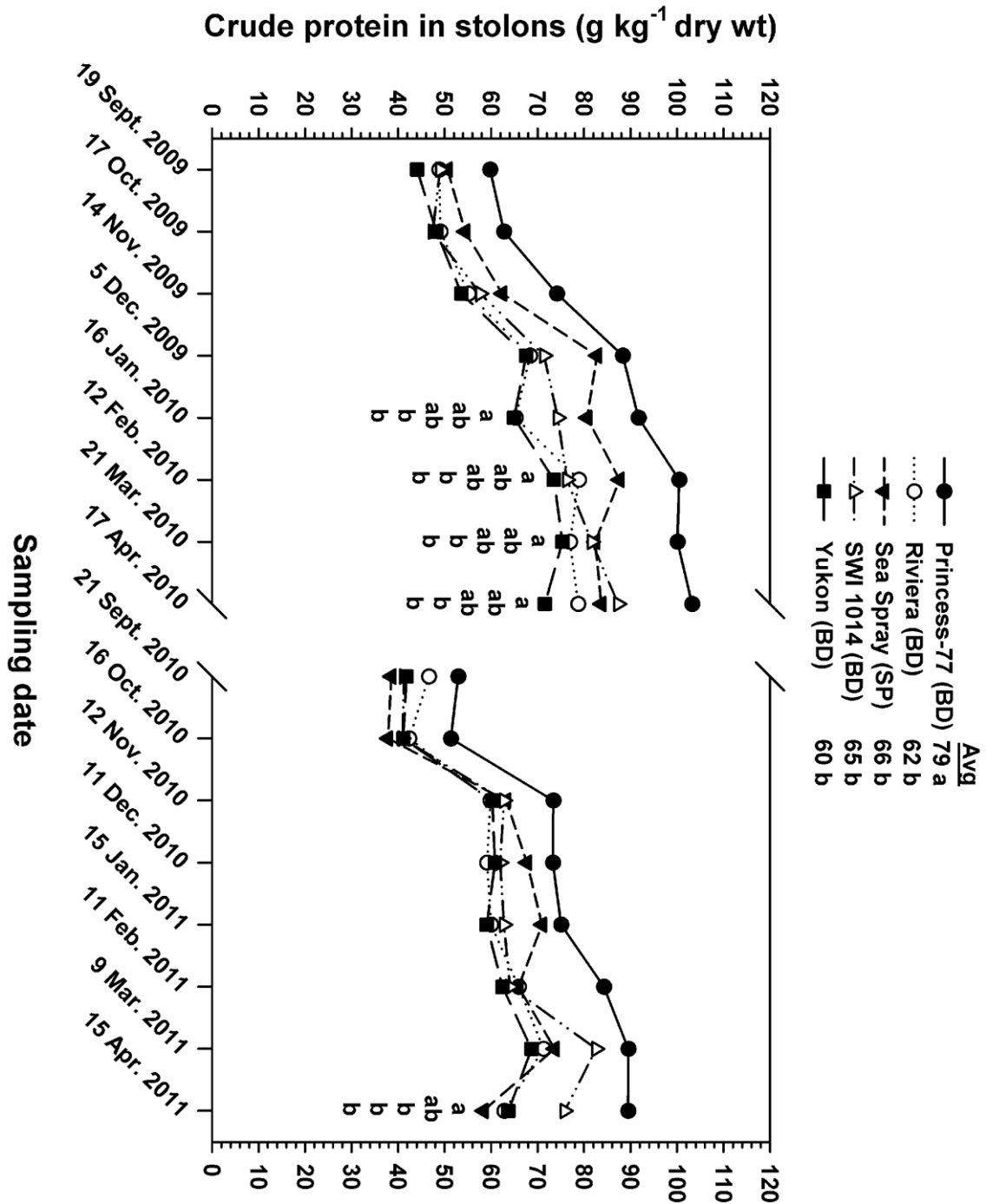


Figure 20 – Content of crude protein in stolons of four bermudagrass (BD) cultivars and ‘Sea Spray’ seashore paspalum (SP) as affected by sampling date in Legnaro, northeastern Italy, during Sept. 2009–Apr 2010 and Sept. 2010–Apr 2011. Data points represent the average of three N fertilization schedules and three replicates. Within each date, means sharing a letter are not statistically different according to Tukey’s honestly significant difference test ($\alpha = 0.05$).

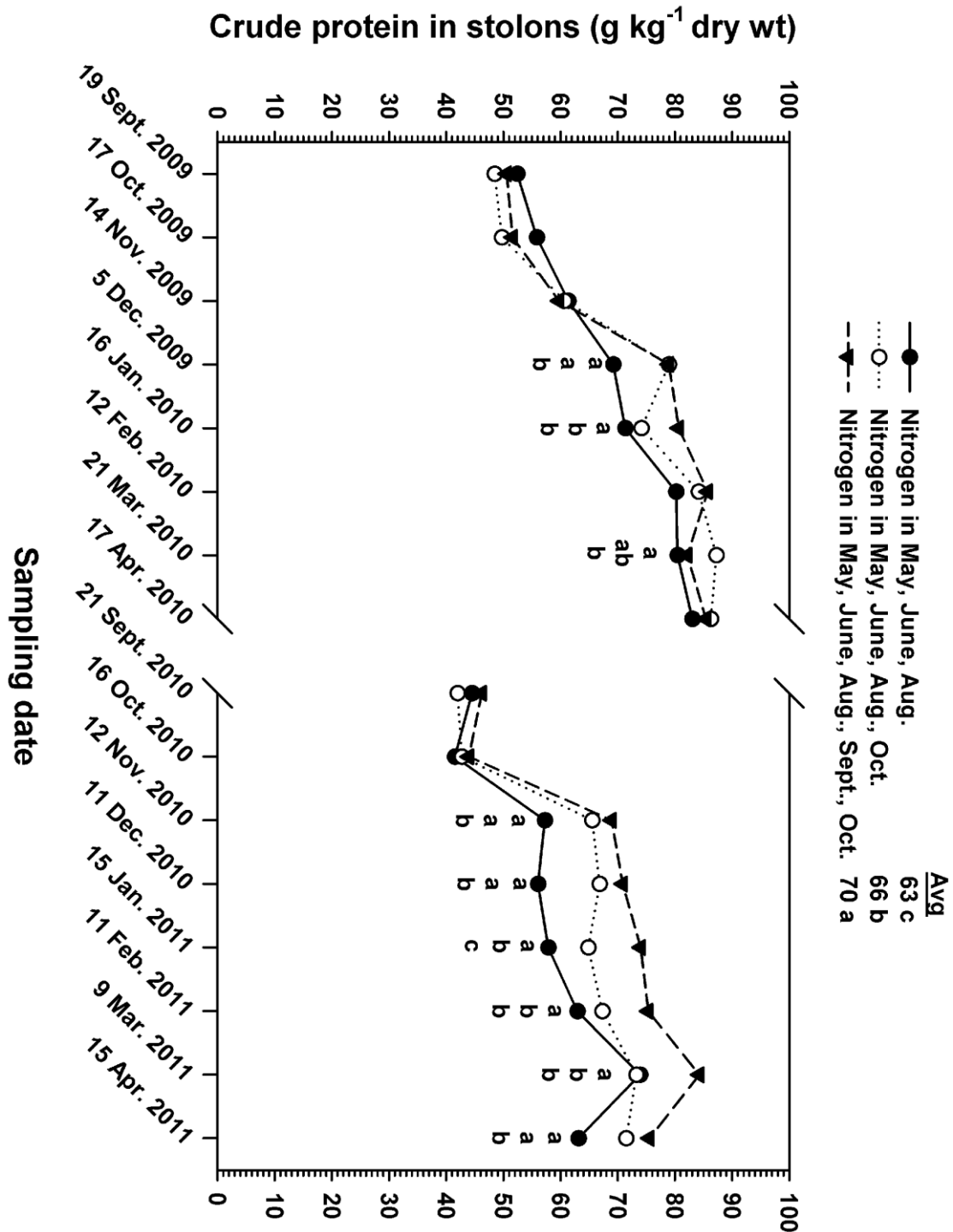


Figure 21 – Content of crude protein in stolons of warm season turfgrasses subjected to three N fertilization schedules as affected by sampling date in Legnaro, northeastern Italy, during Sept. 2009–Apr 2010 and Sept. 2010–Apr 2011. Each N fertilization schedule received 20 g m⁻² year⁻¹ of N. Data points represent the average of five cultivars and three replicates. Within each date, means sharing a letter are not statistically different according to Tukey’s honestly significant difference test ($\alpha = 0.05$).

Predicting spring green-up of bermudagrass cultivars

The regression analysis revealed that values of dry weight, starch and WSC content of stolon measured from September to February of each experimental years were negatively related to bermudagrass D80 (Table 22). These results indicate that varietal differences in stolon dry weight, starch, or WSC concentrations in fall and winter can determine disparity between cultivars in speed to spring green-up. However, the regression between stolon dry weight and D80 yielded coefficients ranging between 0.14 and 0.30 (Table 22), suggesting that relations were significant but of limited practical importance. In Year 1 stolon content of starch and WSC appeared of greater importance for early spring green-up than in Year 2, since highest regression coefficients were yielded during the first year (Table 22). This difference may be due to drought stress occurred in Apr. 2011 (Table 1), which could have interfered with carbohydrate mobilization during spring regrowth. In general, content of WSC was related to D80 more closely than starch, with better prediction of early spring green-up on nine of the 12 dates tested (Table 22). These results concur to those of Macolino *et al.* (2010), reporting that cultivars with highest WSC concentrations during dormancy greened-up earlier than those with low concentrations.

Interestingly, linear regression analysis revealed that stolon content of CP was positively related to D80 (Table 22). These results appeared in contradiction with the responses of N fertilization schedule, as late-season N applications promoted CP accumulation in stolons and also improved spring green-up (Table 7, Fig. 21). However, in the average of N fertilization treatments, a negative correlation was found between starch and CP in stolon collected from September to February of each experimental year (Fig. 22). Therefore, the early green-up resulted by late-season N application may be explained by increase in CP without deleterious effects on carbohydrates storage, regardless of cultivar differences in starch. These findings also suggest a preferential use of starch rather than protein to support spring green-up of bermudagrass cultivars. According to this, breeding selection programs should be tailored to improve starch accumulation in storage organs of bermudagrass, for meeting requirements of early spring green-up.

Table 22 – Linear regression parameters ($y = a + b x$) and regression coefficients for predicting day of year to reach 80% green cover from stolon dry weight (g m^{-2}), content (g kg^{-1} dry wt) of starch, water soluble carbohydrates (WSC), or crude protein (CP) of four bermudagrass cultivars subjected to three N fertilization schedules. Samples were collected from three replicates ($n = 36$) in Legnaro, northeastern Italy.

Sampling date	Parameter	Dry weight	Starch	WSC	CP
19 Sept. 2009	<i>a</i>	126.38	133.91	136.61	94.95
	<i>b</i>	-0.00663	-0.0615	-0.190	0.467
	r^2	0.27 **	0.34 ***	0.33 ***	0.62 ***
17 Oct. 2009	<i>a</i>	125.48	136.61	145.82	101.65
	<i>b</i>	-0.00601	-0.0715	-0.243	0.336
	r^2	0.24 **	0.64 ***	0.51 ***	0.35 ***
14 Nov. 2009	<i>a</i>	124.70	138.09	141.73	102.19
	<i>b</i>	-0.00506	-0.0891	-0.177	0.276
	r^2	0.17 *	0.49 ***	0.59 ***	0.53 ***
5 Dec. 2009	<i>a</i>	125.33	131.25	135.07	106.06
	<i>b</i>	-0.00586	-0.0611	-0.0127	0.166
	r^2	0.18 **	0.32 ***	0.47 ***	0.33 ***
16 Jan. 2010	<i>a</i>	126.89	133.55	143.18	101.84
	<i>b</i>	-0.00751	-0.0775	-0.187	0.233
	r^2	0.29 **	0.48 ***	0.70 ***	0.46 ***
12 Feb. 2010	<i>a</i>	126.00	134.24	143.29	99.85
	<i>b</i>	-0.00695	-0.0817	-0.205	0.234
	r^2	0.23 **	0.42 ***	0.68 ***	0.39 ***
21 Sept. 2010	<i>a</i>	127.62	135.63	136.08	92.26
	<i>b</i>	-0.00828	-0.0954	-0.196	0.511
	r^2	0.30 ***	0.39 ***	0.35 ***	0.28 **
16 Oct. 2010	<i>a</i>	126.90	138.47	138.59	88.06
	<i>b</i>	-0.00792	-0.1010	-0.0231	0.629
	r^2	0.25 **	0.30 ***	0.49 ***	0.33 ***
12 Nov. 2010	<i>a</i>	125.34	130.89	132.65	93.50
	<i>b</i>	-0.00702	-0.0742	-0.173	0.344
	r^2	0.21 **	0.20 **	0.43 ***	0.24 **
11 Dec. 2010	<i>a</i>	124.17	128.34	131.04	88.94
	<i>b</i>	-0.00603	-0.0613	-0.156	0.418
	r^2	0.17 *	0.15 *	0.25 **	0.27 **
15 Jan. 2011	<i>a</i>	125.27	129.24	136.97	—
	<i>b</i>	-0.00709	-0.0745	0.199	—
	r^2	0.22 **	0.18 *	0.43 ***	ns
11 Feb. 2011	<i>a</i>	123.39	130.21	144.22	102.56
	<i>b</i>	-0.00603	-0.0852	-0.230	0.208
	r^2	0.14 *	0.21 **	0.46 ***	0.13 *

ns, not significant; *, **, ***, significant at $P < 0.05, 0.01, 0.001$, respectively.

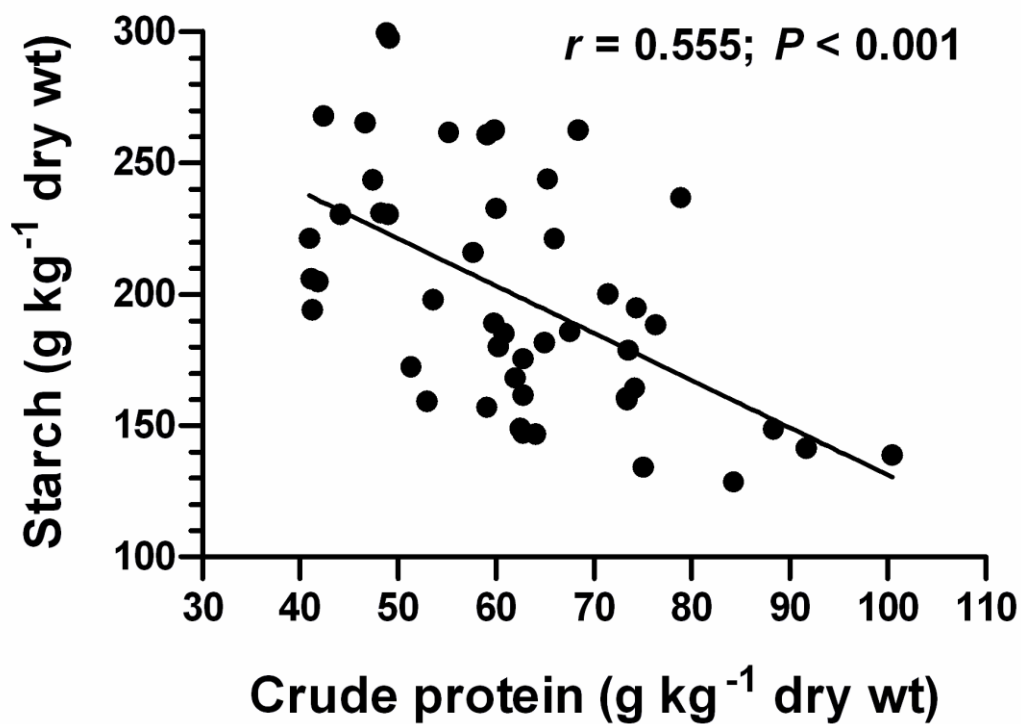


Figure 22 – Correlation between crude protein and starch content in stolons of four bermudagrass cultivars. Data were collected monthly from September to February of 2010 and 2011 in Legnaro, northeastern Italy; and were averaged over three N fertilization schedules and three replicates ($n = 48$).

Conclusions

This study pointed out important differences among seeded bermudagrass cultivars concerning spring green-up, fall color retention, and dry weight of overwintering stolons. Under study conditions, ‘Sea Spray’ seashore paspalum displayed later spring green-up and lower summer quality than bermudagrasses, while prolonged fall color retention. Among bermudagrass cultivars, ‘Riviera’ exhibited high dry weight of stolons and was characterized by early spring green-up along with ‘Yukon’, whereas ‘Princess-77’ had low stolon dry weight, late green-up, and prolonged fall color retention. Carbohydrate levels in stolons during fall and winter months were significantly related to spring green-up, such that bermudagrass cultivars with higher content of starch and WSC had high regrowth capacity. Moreover, levels of CP were negatively correlated with those of starch, indicating that differences in starch were of greater interest than those in CP for early green-up of bermudagrass.

Using the same seasonal amount of N, shifts of fertilizer applications until late-season anticipated spring green-up regardless of cultivars and also delayed fall color retention of bermudagrasses. Late-season N applications had limited influence on stolon storage of carbohydrates, while promoted CP accumulation during acclimation. Moreover, delays of N applications until October had no influence on stolon morphological traits and rhizome production, indicating no contraindications for freeze-avoidance of reserve organs. In summary, the present study revealed that the selection of N fertilization schedules may play a limited practical importance with regard to the duration of the growing season. Therefore, the choice of N fertilization program should be considered of secondary importance than that of species/cultivar to reduce the winter dormancy of warm season turfgrasses.

Additional results of this study confirmed that visual estimation of green cover during spring green up can effectively be replaced by photographic technique and by NDVI readings collected with a handheld optical sensor. Normalized difference vegetation indices also provided rapid and unbiased estimates of fall color retention, supplying quantitative substitutes for discrete visual color ratings. Nevertheless, NDVI readings were weakly

correlated with visual ratings of summer turf quality, discouraging the use of this instrumental method over turf quality assessments.

Chapter II

Green-up of eight bermudagrass cultivars and ‘Companion’ zoysiagrass as influenced by spring scalping¹

¹ During the course of this doctoral project, a public presentation has been made based on part of the work presented in this chapter (Rimi *et al.*, 2011).

Introduction

Winter dormancy and poor tolerance to low temperatures represent the main impediments to warm season grasses gaining greater acceptance in the Mediterranean countries of Europe. Spring recovery of warm season grasses usually begins when the soil temperature reaches a specific threshold level and the active growing period continues until the soil temperature decreases below this level in the fall (Youngner, 1959). However, freeze-avoidance and spring green-up can be improved through appropriate cultivar selection (Richardson *et al.*, 2004; Anderson *et al.*, 2007; Patton *et al.*, 2008) and the application of proper management practices (White & Schmidt, 1989; Miller & Dickens, 1996a, 1996b; Goatley *et al.*, 2005). Mowing, a fundamental practice of turfgrass culture (Beard, 1973), could be considered as a potential tool for extending the growing season of warm season turfgrasses.

Mowing improves turfgrass appearance and uniforms its surface by periodic removal of a portion of leaves or tillers. This continuous defoliation affects turf quality (Beard *et al.*, 1991; Kopek *et al.*, 2007), evapotranspiration rate (Pohjakallio & Antila, 1955), weed control (Dexter, 1936; Beard, 1973), and tolerance to other environmental stresses (Wilkins, 1935; Juska & Hanson, 1963). According to this, proper cutting height and mowing frequency must be defined in order to maintain an attractive and vigorous turf. Cutting height may extremely vary (from ≈ 0.3 up to 10 cm) on the basis of the intended use of turfgrass, species/cultivar, and the period of the year (Beard, 1973). In addition, mowing frequency should be tailored to the shoot growth rate of the turf and environmental conditions, by avoiding single removal of more than 30 – 40% leaf tissue (Fig. 23) (Madison, 1960; Beard, 1973; Christians, 1998).

During the growing season, removing more than 40% of the warm season turf height can cause scalping, resulting in unattractive patches and exposure of the lower part of the plants (Beard, 1973). However, the deliberate application of scalping in early spring removes dead leaf tissue and allows sunlight to reach the new growth next to the ground and can allow for earlier spring green-up (Brede, 2000; Christians, 1998). Despite the suggested use of this cultural practice on bermudagrass (Fig. 3) and zoysiagrass (Fig. 24),

no studies have been published on the effectiveness of spring scalping in promoting green recovery. In order to address this knowledge gap, the influence of spring scalping on spring green-up of eight bermudagrass cultivars and a zoysiagrass was studied for two years in a transition zone environment.



Figure 23 – Standard recommendation for setting mowing frequency of turfgrass areas, commonly referred to as the ‘one-third’ rule. No more than 30 – 40% leaf tissues should be usually removed at a single mowing (Christians, 1998).



Figure 24 – Turf density and leaf texture of manilagrass (left side of the figure) and zoysiagrass (right side).

Materials and methods

A field trial was conducted from March 2009 to June 2010 at the experimental agricultural farm of Padova University in Legnaro, northeastern Italy (lat. 45°20'N, long. 11°57'E, elevation 8 m) to investigate the effect of spring scalping on eight seeded bermudagrass cultivars and a zoysiagrass. The area has a humid subtropical climate and is similar to plant hardiness zone 8 (U.S. Department of Agriculture, 1990) with a rainfall of 820 mm distributed throughout the year (Table 1). The soil at the site was a coarse-silty, mixed, mesic, Oxyaquic Eutrudept, (Morari, 2006), containing 20% clay, 61% silt, and 19% sand, with a pH of 8.3, 2.1% organic matter, a C to N ratio of 11.4, a P content of 30 mg kg⁻¹, and a K content of 147 mg kg⁻¹.

The experiment was carried out on mature turf plots that were established in 2005. Grasses included 'Companion' zoysiagrass and eight bermudagrass cultivars: Barbados, Contessa, La Paloma, Mohawk, NuMex Sahara, Princess-77, SR 9554, and Yukon. During the growing periods of the three years prior to the onset of the study (2005 – 2008) a slow-release fertilizer (20N-2.2P-6.6K) was applied in May, June, July, August, and September at a rate of 40 kg ha⁻¹ of N. In addition, plots were mowed weekly at an effective height of cut of 52 mm using a rotary mower (HRD 536; Honda Europe Power Equipment, Ormes, France) with an effective mowing width of 55 mm and clippings were removed. The mowing height was chosen because it is the height at which warm season grasses in low maintenance (zero irrigation) areas in Italy is typically maintained.

In 2009 and 2010 spring scalping was applied on 13 Mar. by means of a rotary mower set at a height of 28 mm (effective height of cut). Regular weekly mowing started 28 d after spring scalping (DASS). No additional irrigation to natural precipitation was provided during the study and plots were fertilized in May, June, and August with ammonium nitrate at 66 kg ha⁻¹. The experimental design was a completely randomized split-block with cultivars as the main plot and scalping as the subplot treatment. Main plots and sub plots were replicated four times and measured 1.6 × 4.5 m and 1.6 × 2.3 m, respectively.

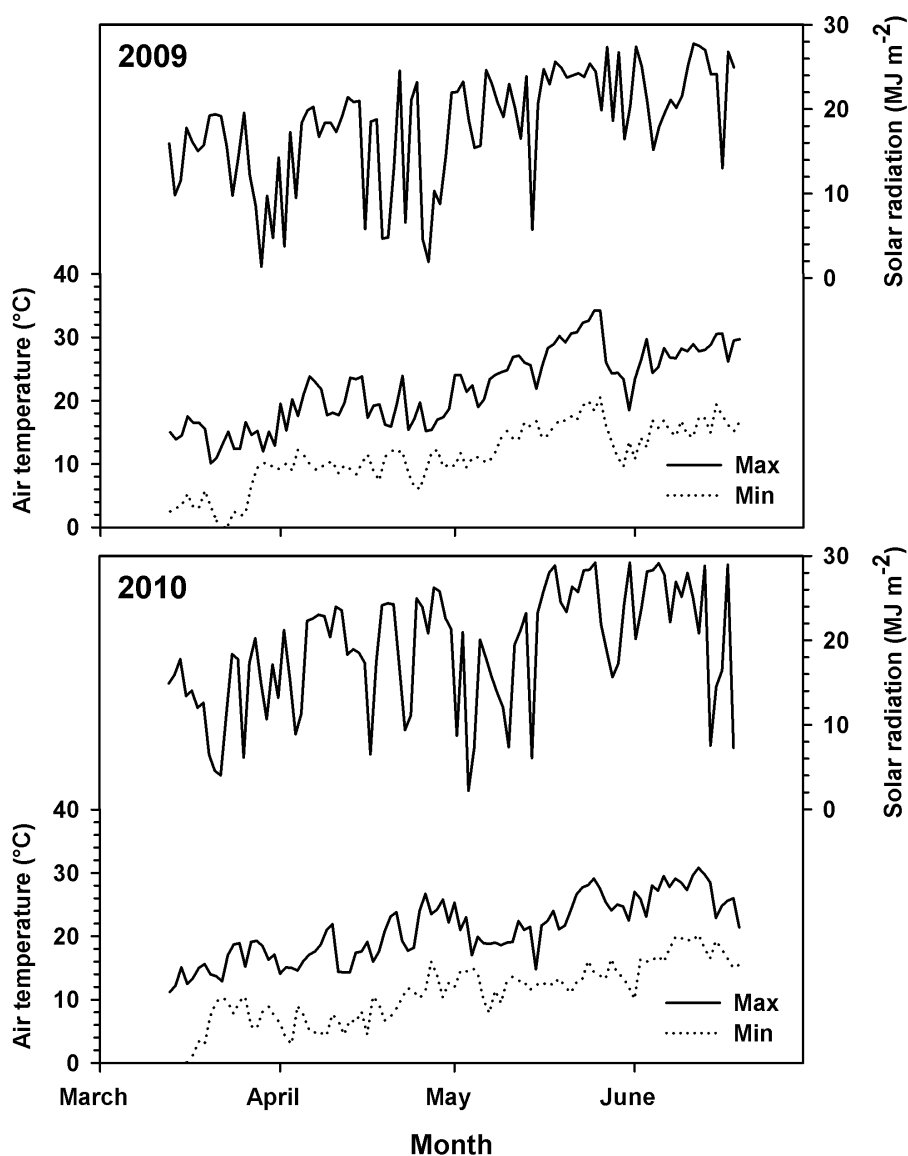


Figure 25 – Maximum (Max) and minimum (Min) air temperature and incident solar radiation from 13 Mar. [0 d after spring scalping (DASS)] to 19 June (98 DASS) during 2009 and 2010 at the experimental farm of Padova University, Legnaro, Italy.

Green cover for each subplot was visually estimated weekly beginning immediately after the scalping treatment until 98 DASS. A sigmoidal model (GraphPad Prism 5.0 for Windows; GraphPad Software, La Jolla, CA) was used to calculate DASS required to reach 80% of green color for each plot (Macolino *et al.*, 2010). Eight temperature sensors (thermocouples) were installed at a soil depth of 2.5 cm in four randomly selected scalped bermudagrass subplots and in the corresponding unscalped subplots. Thermocouples were

connected to a data logger (CR10X; Campbell Scientific, Logan, UT) and soil temperature was recorded hourly throughout the research period. Daily maximum soil temperature difference (DMTD) was calculated by subtracting the recorded minimum temperature from the maximum temperature for each day during the investigative period. Air temperature and incident solar radiation were measured by means of a weather station (WTS 7000; MTX Italia, Modena, Italy) located in close proximity to the research plots (Fig. 25). The DMTDs were then regressed against daily incident solar radiation and daily air temperature. The linear regression was calculated for the time period of 13 Mar. (day when scalping treatments were applied) to 3 May (day when bermudagrass cultivars reached an average of 80% green cover).

The effects of scalping, cultivar and year on DASS to reach 80% of green cover were statistically analysed using a repeated measures analysis of variance with SAS Proc Mixed (version 9.2; SAS Institute, Cary, NC). A compound symmetry covariance structure resulted in the best fit for the data (lowest AIC value). Tukey's honestly significant difference test was used at the 0.05 probability level to identify significant differences among means.

Results and Discussion

The analysis of variance revealed significant three-way interactions among cultivars, scalping, and years ($P = 0.002$). The two-way interactions between cultivars and scalping ($P = 0.002$) and between cultivars and years ($P = 0.005$), and all the main effects ($P < 0.001$) were also significant. The two-way interactions between scalping and year was not significant ($P = 0.381$).

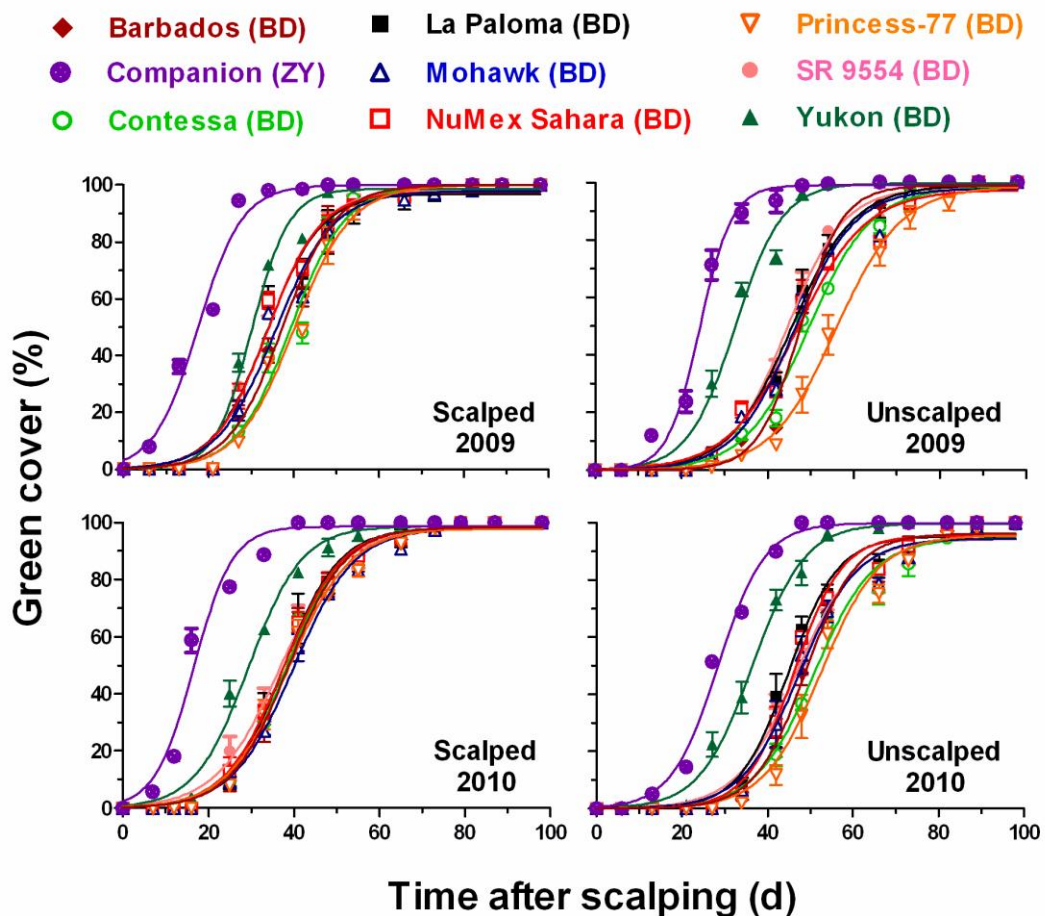


Figure 26 – Percent green cover as a function of number of days after spring scalping for scalped and unscalped bermudagrass (BD) cultivars and ‘Companion’ zoysiagrass (ZY) in 2009 and 2010. Data points represent an average of four replicates and bars indicate \pm standard error.

With the exception of ‘Yukon’, spring green-up was enhanced by scalping for all the test bermudagrass cultivars in both years of the study (Table 23; Fig. 26). For ‘Barbados’, ‘Contessa’, ‘La Paloma’, ‘Mohawk’, ‘NuMex Sahara’, ‘Princess-77’, and ‘SR

9554', scalping reduced the time required to reach 80% green cover by 8 to 18 d compared to unscalped plots in 2009, and by 8 to 16 d in 2010 (Table 23). Scalping application reduced time to reach 80% green recovery for 'Companion' zoysiagrass only in 2010. When subjected to spring scalping, 'La Paloma' did not differ from 'Yukon' in both 2009 and 2010 (Table 23). 'Companion' zoysiagrass showed earlier spring green-up than bermudagrasses and, among those, 'Yukon' exhibited the shortest time to reach 80% green cover in unscalped plots in both years (Table 23; Fig. 26). Macolino *et al.* (2010) studied the concentration of WSC in stolons of the same bermudagrass varieties involved in this research and reported that 'Yukon' exhibited the highest concentration in March. Based on these previous findings and the results of this study, it appears that spring scalping could be less effective for bermudagrass cultivars that are rich in WSC in late winter.

Table 23 – Mean number of days required to reach 80% of green cover (estimates based on sigmoidal model) for scalped and unscalped bermudagrass (BD) cultivars and 'Companion' zoysiagrass (ZY) in 2009 and 2010. Days were counted on both scalped and unscalped plots beginning on the day scalping treatments were applied. Data were collected in Legnaro, northeastern Italy.

Cultivar	2009		2010		Overall
	Scalped	Unscalped	Scalped	Unscalped	
	— Time after scalping treatment to reach 80% green cover (d)† —				
Barbados (BD)	46 c A‡	54 c A	48 c A	60 cd B	52 cd
Companion (ZY)	25 a A	30 a A	22 a A	35 a B	28 a
Contessa (BD)	48 c A	60 cd B	48 c A	64 d B	55 cd
La Paloma (BD)	44 bc A	56 c B	46 bc A	55 c B	51 c
Mohawk (BD)	46 c A	57 c B	50 c A	59 cd B	53 cd
NuMex Sahara (BD)	44 bc A	59 cd B	48 c A	56 cd B	52 cd
Princess-77 (BD)	50 c A	67 d B	49 c A	64 cd B	57 d
SR 9554 (BD)	43 bc A	54 c B	47 c A	60 cd B	51 c
Yukon (BD)	36 b A	40 b A	38 b A	44 b A	40 b

† Spring scalping applied on 13 Mar. 2009 and 13 Mar. 2010.

‡ Values followed by the same letter are not significantly different from one another at the 5% level of significance according to the Tukey's honestly significant difference test. Lower case letters denote differences between cultivars (in columns); upper case letters denote differences between treatments for each cultivar separately.

In 2009, the green-up period (March to May) was warmer than in 2010, while the solar radiation was greater in 2010 than in 2009 (Fig. 25). With respect to scalping,

differences in air temperature and solar radiation between the two experimental years may have subjected the test grasses to different environmental conditions, which may have influenced the effects of scalping, and may explain the significant cultivar \times scalping \times year interaction. Averaged over the two years, ‘Yukon’ was the only bermudagrass cultivar that differed from the others in scalped plots, showing the earliest spring green-up, while differences occurred among the other bermudagrasses in unscalped plots (Table 24). When data were averaged over the nine cultivars and the two years, spring scalping enhanced time to reach 80% green cover by an average of 11 d (Table 24), confirming the hypothesis that scalping in early spring is helpful to accelerate green-up.

Table 24 – Mean number of days required to reach 80% of green cover (estimates based on sigmoidal model) for scalped and unscalped bermudagrass (BD) cultivars and ‘Companion’ zoysiagrass (ZY) in 2009 and 2010. Days were counted on both scalped and unscalped plots beginning on the day scalping treatments were applied. Data were collected in Legnaro, northeastern Italy, and were averaged over two years and two scalping treatments, respectively.

Cultivars	Avg. over two years		Avg. over two scalping treatments	
	Scalped	Unscalped	2009	2010
— Time after scalping treatment to reach 80% green cover (d) † —				
Barbados (BD)	47 c A‡	57 c B	50 c A	54 c A
Companion (ZY)	24 a A	32 a B	27 a A	29 a A
Contessa (BD)	48 c A	62 cd B	54 cd A	56 c A
La Paloma (BD)	45 c A	56 c B	50 c A	51 c A
Mohawk (BD)	48 c A	58 cd B	51 cd A	55 c A
NuMex Sahara (BD)	46 c A	58 cd B	51 cd A	52 c A
Princess-77 (BD)	49 c A	65 d B	58 d A	57 c A
SR 9554 (BD)	45 c A	57 c B	48 c A	53 c B
Yukon (BD)	37 b A	42 b A	38 b A	41 b A
Average	43 A	54 B	48 A	50 B

† Spring scalping applied on 13 Mar. 2009 and 13 Mar. 2010.

‡ Values followed by the same letter are not significantly different from one another at the 5% level of significance according to the Tukey’s honestly significant difference test. Lower case letters denote differences between cultivars (in columns); upper case letters denote differences between treatments for each cultivar separately.

Differences in spring green-up among cultivars were consistent across the two years of study although there was a difference in magnitude that resulted in a cultivar \times year interaction (Table 24). ‘Companion’ zoysiagrass greened-up earlier than any of the test

bermudagrass cultivars, reaching 80% green cover by mid April in both years of study (Table 24). Among bermudagrasses, 'Yukon' showed earliest spring green-up and reached 80% green cover by the end of April in both 2009 and 2010 (38 DASS and 41 DASS, respectively) (Table 24). In 2009, 'Barbados', 'Contessa', 'La Paloma', 'Mohawk', 'NuMex Sahara', and 'SR 9554' reached 80% green cover 11–15 d later than 'Yukon', while 'Princess-77' was 20 d later than 'Yukon' (Table 24). In the subsequent year, 'Barbados', 'Contessa', 'La Paloma', 'Mohawk', 'NuMex Sahara', 'Princess-77', and 'SR 9554' needed 11–15 d longer than 'Yukon' to reach 80% green cover, without significant differences among the cultivars (Table 24). Time necessary to reach 80% green cover was on average 2 d longer in 2010, under cooler early spring conditions, than in 2009 (Table 24, Fig. 25). This result, together with the nonsignificant scalping \times year interaction suggests that the effectiveness of scalping may not be greatly influenced by air temperature conditions in early spring.

Scalping applications influenced soil temperatures of bermudagrass plots in both experimental years. From immediately after scalping application until the end of April, maximum soil temperatures were higher in scalped plots than in unscalped in both years, while minimum temperatures were not affected (Fig. 27). A strong positive relationship was found between DMTD and solar radiation during the period of spring green-up for scalped and unscalped plots (Fig. 28). The two treatments had significantly different slopes, showing a higher increment of DMTD as solar radiation increases for scalped than unscalped plots. Maximum air temperatures showed a weaker relationship with DMTD in both scalped ($r^2 = 0.24$; $P < 0.001$) and unscalped plots ($r^2 = 0.33$; $P < 0.001$) and no differences between the two fitted regressions (data not shown). These results suggest that scalping increases light penetration through the turf canopy, resulting in increased maximum soil temperatures during the spring green-up period. The strong influence of light penetration on scalping effectiveness could also explain the lack of a significant scalping \times year interaction. The greater solar radiations in 2010 may have increased the effectiveness of the scalping treatment, which would have compensated for the lower air temperatures (Fig. 25). Difference in minimum soil temperatures were not observed in this study, suggesting that scalping in early spring had no contraindications for freeze exposures.

However, further research is needed to address the knowledge gap on effects of scalping applied in late fall or during the winter.

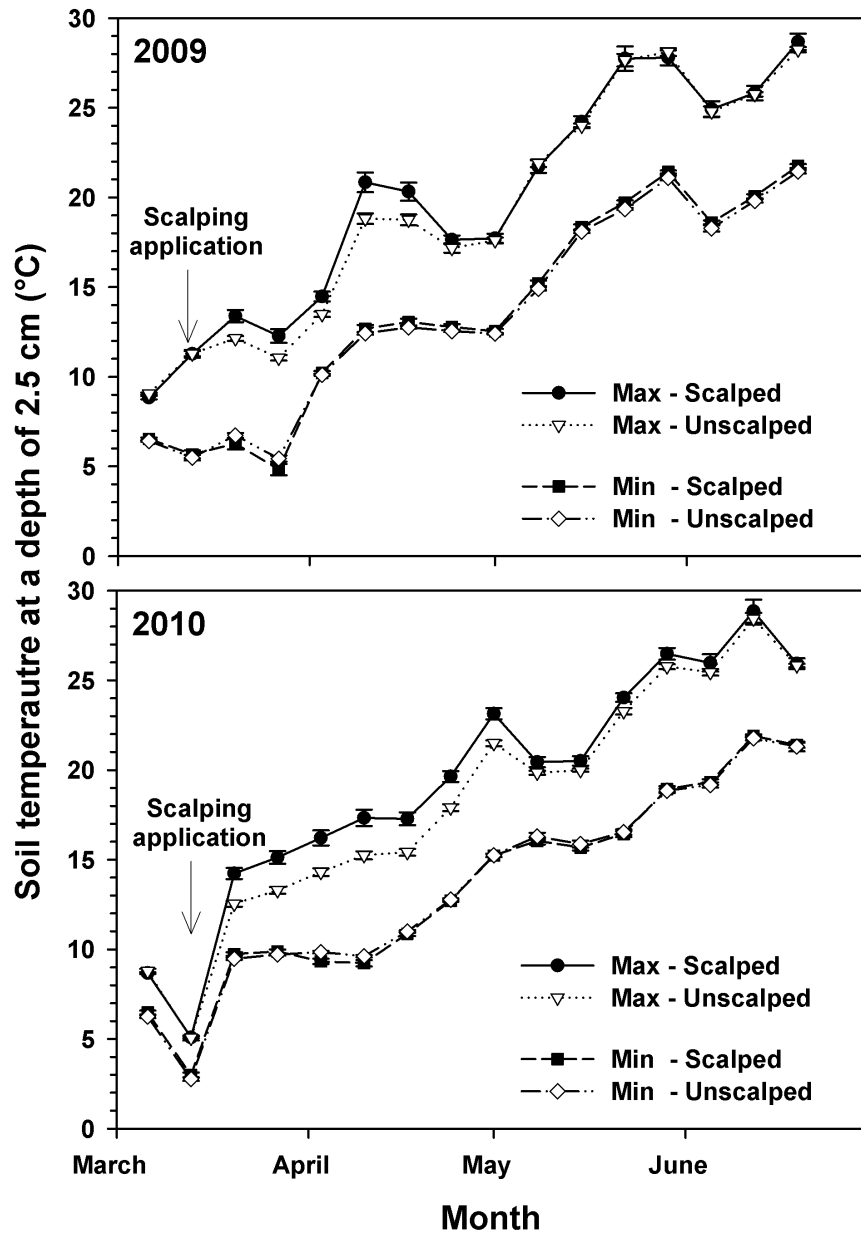


Figure 27 – Weekly average maximum (Max) and minimum (Min) soil temperatures at a depth of 2.5 cm in scalped and unscalped bermudagrass plots in 2009 and 2010. Data points represent an average of four replicates and bars indicate \pm standard error.

When data were averaged over the scalping treatments and the two years, ‘Yukon’ was the earliest bermudagrass cultivar to achieve green-up, reaching 80% green cover by the end of April (40 DASS), while ‘Princess-77’ showed the slowest green-up (Table 23).

These results corroborated the findings of Macolino *et al.* (2010), in which ‘Yukon’ had the earliest spring green-up after establishment, while ‘Princess-77’ was in the slowest group. Similar results have been published by the NTEP’s 2007 bermudagrass trial (NTEP, 2011b), reporting that ‘Yukon’ showed a significantly faster green-up than ‘Princess-77’ in five locations across the U.S. In addition, ‘Companion’ zoysiagrass greened-up earlier than any of the test bermudagrass cultivars, reaching 80% green cover 12 d earlier than ‘Yukon’ (Table 23). These results are in agreement with previous reports from Turkey (Severmutlu *et al.*, 2011), documenting better visual quality for zoysiagrasses in comparison with bermudagrass cultivars in early spring.

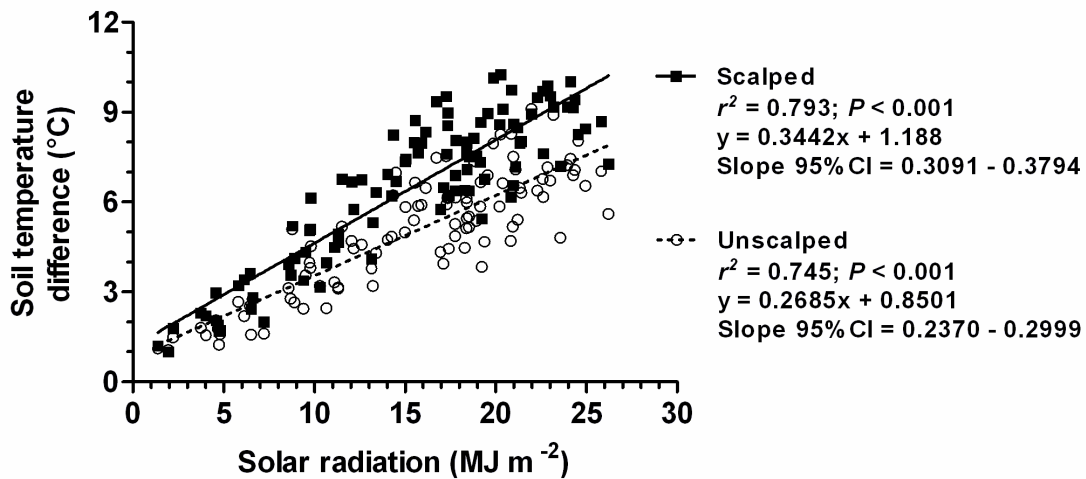


Figure 28 – Relationship between daily soil temperature differences (maximum – minimum temperature) at 2.5 cm depths and incident solar radiation for scalped and unscalped bermudagrass plots. Data are the average of four replicates and were collected over the period beginning on the day scalping was applied until plots reached 80% of green cover across two years (13 Mar. 2009 – 1 May 2009; 13 Mar. 2010 – 3 May 2010).

Synopsis and recommendations

The results of this study demonstrated that the application of spring scalping enhances green-up of bermudagrass by increasing the soil temperatures during the day. However, the effectiveness of this practice was influenced by varietal response. Among the bermudagrass cultivars tested, ‘Yukon’ showed the earliest spring green-up, regardless of the scalping treatment applied. Therefore, spring scalping is highly recommended for bermudagrasses that are slow to green-up in the spring.

‘Companion’ zoysiagrass was faster than any of the bermudagrass cultivars in transitioning out of winter dormancy and the influence of spring scalping on its green-up was affected by environmental conditions. However, early spring green-up of zoysiagrasses relative to bermudagrasses can be counterbalanced by lower quality in summer. In fact, roots of zoysiagrass provide lower soil exploration compared to bermudagrass, as recently noticed by Macolino *et al.* (2012). Consequently, the choice between a zoysiagrass or bermudagrass cultivar in a specific area should be pondered on the requirements for both early spring color and drought tolerance. Further research is needed concerning the effects of scalping on green-up of zoysiagrasses to investigate the influence of genetic diversity on its effectiveness.

Additional results of this study indicate that turf canopy scalped in early spring had no contraindications for freeze exposures. Despite this practice is anecdotically applied also in late fall or during the winter, these timings have not been tested. Thus, more studies are needed to determine the effects of scalping in other periods of the year.

Chapter III

Winter-applied glyphosate effects on spring green-up of zoysiagrasses and 'Yukon' bermudagrass in a transition zone ²

² During the course of this doctoral project, a public presentation has been made based on the work presented in this chapter (Rimi *et al.*, 2012).

Introduction

Over the last few years the use of warm season grasses, such as bermudagrass and zoysiagrass, has rapidly increased in the Mediterranean countries of Europe (Croce *et al.*, 2001; Volterrani *et al.*, 1997). These species are also becoming very popular in the sod production industry due to their good sod-forming characteristics. The primary concern of sod producers is to obtain an adequate quality for harvest in the shortest possible period with minimal inputs. However, the time necessary to obtain a sod ready for harvest depends on grass species, cultivar, environmental conditions, and cultural practices (Beard, 1973; McCalla *et al.*, 2008). High quality sod is expected to have full green color, no weeds, in addition to high density and uniformity, and other technical traits, such as strength for transplanting (McCalla *et al.*, 2008).

Cool season grasses are generally seed propagated and a harvestable sod can be produced after about six months by adding rhizomatous species, such as kentucky bluegrass, to the seed mixture or by using a synthetic netting (Carrow & Sills, 1980). In contrast, several warm season grasses are commonly established by sprigging or plugging, and can be easily re-established by regrowth from rhizomes (Christians, 1998). Under restrictive climate conditions, the time required to produce a sod of warm season species may exceed the period of time in which temperatures are in the optimum range for growth (Beard, 1973). In transitional environments, zoysiagrass often needs longer than one growing season to establish (Severmutlu *et al.*, 2011); and such slow growth rates are the main reason for choosing sodding over seeding as the method of establishment. Studies conducted on seeded cultivars and vegetative hybrids of bermudagrass suggest that a minimum of 16 weeks are needed to produce a marketable sod (McCalla *et al.*, 2008; Mitchell & Dickens, 1979), leading to transplanting in late summer or autumn, under suboptimal conditions for rooting. To avoid these issues, warm season turfgrass sod are often harvested at the onset of the second or third growing season, after reaching full spring green-up.

One of the major problems for producers of warm season sod are winter annual weeds, which germinate in early fall and reduce quality of turfgrasses emerging in from

dormancy in spring (Johnson, 1980). Postemergence herbicides are used extensively to kill winter annual weeds in warm season turfgrasses and application timing may optimize weed control. However, application timing of non-selective herbicides is critical as spring green-up can be inhibited (Johnson, 1977; Johnson & Burns, 1985; Johnson & Ware, 1978). Postemergence non-selective herbicides such as glyphosate [N-(phosphonomethyl)glycine] and paraquat (1,1'-dimethyl-4,4'-bipyridinium ion) effectively control annual weeds and cause no injury to bermudagrass when applied on dormant turf during the winter (Johnson 1980, 1984). However, these herbicides may delay spring green-up of bermudagrass if they are applied when the grass has physiologically transitioned out of winter dormancy (Fagerness *et al.*, 2002; Johnson, 1984).

A delay in spring green-up may shift the date of harvest, narrowing the time frame available for a re-establishment of sod. Based on the requirements of sod producers, weed control should be tailored according to local environmental conditions (Breuninger & Schmidt, 1981; Johnson, 1976). The use of glyphosate during the cooler months is commonly recommended for zoysiagrass and manilagrass turf (Tae, 2005; Velsor *et al.*, 1989) (Fig. 24). Despite the suggested winter-application of glyphosate on *Zoysia* species, research is limited that document the effects of this cultural practice on spring green-up. Experiments were conducted to evaluate effects of glyphosate application timing on three turfgrass species on spring transition.

Materials and methods

Experiment 1

This study was conducted from January to June 2011 at two commercial sod farms in northern Italy: Somma Lombardo (lat. 45°41'N, long. 8°41'E, elevation 256 m), and Sommacampagna (lat. 45°18'N; long. 11°01'E; elevation 30 m). The soil at Somma Lombardo site was a sandy loam (5% clay, 29% silt, and 66% sand) with a 5.1% organic matter content, a pH of 5.1, 6 mg kg⁻¹ of P, and 36 mg kg⁻¹ of K. The soil at the Sommacampagna site was a loamy sand (9% clay, 19% silt, and 72% sand) with a 1.8% organic matter content, a pH of 8.1, 18 mg kg⁻¹ of P and 218 mg kg⁻¹ of K. Both locations have a humid subtropical climate with a bimodal precipitation pattern (Table 25) and are similar to plant hardiness zone 8 (U.S. Department of Agriculture, 1990).

The manilagrass cultivar Zeon was established at both locations in July 2009 by planting 25 mm diameter plugs at a rate of 18 plugs/m². During the establishment phase irrigation was provided at a rate of 5 mm d⁻¹. Following establishment, plots were mowed two times per week with a reel mower set at a height of 27 mm, with clippings returned. At the Somma Lombardo site, a slow-release fertilizer (16N-0P-12.5K) was applied monthly from May to August at the rate of 5 g m⁻² N. A slow-release fertilizer (20N-2P-8.3K) was applied in May, June, and August at the rate of 6.6 g m⁻² N to the Sommacampagna location.

In December 2010, nine plots (2 × 3 m) were established at each of the two research sites. Treatments were randomly assigned to the plots arranged in a randomized complete block design at each location. Treatments consisted of: A) glyphosate at 1.1 kg ha⁻¹ on 8 Feb. 2011, B) glyphosate at 1.1 kg ha⁻¹ on 21 Feb 2011, and C) untreated control. Each treatment was replicated three times. The rate and dates of application were chosen according to typical management practices used in these areas with regard to winter control of annual bluegrass. Daily growing degree days were calculated beginning on 1 Jan. 2011 for the applications of glyphosate for both locations, using 5 °C as a base temperature (Patton *et al.*, 2004; Schiavon *et al.*, 2011; Severmutlu *et al.*, 2011). The accumulated GDD were as follows: Somma Lombardo, 1 Jan.–8 Feb. = 52 GDD and 1 Jan.–21 Feb. = 87

GDD; Sommacampagna, 1 Jan.–8 Feb. = 36 GDD and 1 Jan.–21 Feb. = 73 GDD. The glyphosate formulation (Glifogold; Monsanto Europe, Anversa, Belgium) contained a proprietary surfactant with 360 g L⁻¹ of glyphosate in its isopropylamine salt and was applied in water at a rate of 200 L ha⁻¹.

Table 25 – Long-term monthly average air temperature and precipitation from weather stations located in close proximity to the field sites in Somma Lombardo (2000–2010) and Sommacampagna (1993–2008), northern Italy.

Month	Mean temperature		Total precipitation	
	Somma Lombardo	Sommacampagna	Somma Lombardo	Sommacampagna
	°C		mm	
Jan.	1.2	2.8	31	39
Feb.	3.0	4.4	59	30
Mar.	8.0	8.9	60	40
Apr.	11.7	12.7	134	73
May	17.0	18.1	88	65
June	21.5	21.8	63	64
July	23.8	23.6	49	64
Aug.	22.7	23.1	49	85
Sept.	17.9	18.1	93	95
Oct.	12.2	13.5	110	79
Nov.	6.5	7.8	213	80
Dec.	2.0	3.7	104	58
Annual	12.3	13.2	1053	772

Glyphosate was applied using a backpack sprayer (F200 Electra; Fox Motors, Poviglio, Italy) calibrated to operate at 100 kPa. Weed species were determined and the number of individual plants of each weed species within plots was counted before the glyphosate application. At Somma Lombardo percent ground cover of weeds was ≈10% and the weed population consisted of 80% (6 plants/m²) annual bluegrass and 20% (2 plants/m²) common chickweed [*Stellaria media* (L.) Vill.]. At Sommacampagna percent ground cover of weeds was ≈5% and the main weeds were 95% (2 plants/m²) annual bluegrass and 5% (0.5 plants/m²) tall fescue. A visual estimation of weed control efficiency was conducted 49 d after treatment (DAT) for the 8 Feb. application and 35 DAT for the 21 Feb. glyphosate application. Weed control was based on a scale of 0 to 100, with 0 = no control, and 100 = total control. Weed control ratings of different species were combined to

form unique values based on plant size, discoloration or necrosis, and general plant vigor (Main *et al.*, 2004). Immediately after weed control estimations, plots were hand-weeded in order to avoid weed interference on assessments of turf regrowth.

Plots were visually assessed for spring green-up on 10 Mar., 28 Mar., 9 Apr., 15 Apr., 30 Apr., and 30 May of 2011. Spring green-up ratings were visually assessed on a linear 0 to 100 scale of the green ground cover (Munshaw *et al.*, 2006; Patton *et al.*, 2004). Normalized difference vegetation indices readings were also collected by means of a handheld optical sensor (Greenseeker; NTech Industries, Ukiah, CA) within 24 h of the visual ratings being taken (Bell *et al.*, 2009).

Experiment 2

An additional field trial was conducted from January to June 2011 at the experimental agricultural farm of Padova University in Legnaro, northeastern Italy (lat. 45°20'N, long. 11°57'E, elevation 8 m). The soil at the site was a silty loam (20% clay, 61% silt, and 19% sand) with a 2.1% organic matter content, pH of 8.3, P level of 28 mg kg⁻¹, and K level of 142 mg kg⁻¹. The area has a humid subtropical climate and is similar to plant hardiness zone 8 (U.S. Department of Agriculture, 1990), with the annual rainfall mostly distributed from April to November (Table 1). Grasses used in this study were 'Yukon' bermudagrass and 'Companion' zoysiagrass. The experiment was carried out on mature turf plots established in July 2005. Slow-release fertilizer (20N–2.2P–6.6K) was applied each month from May to September at a rate of 4 g m⁻² of N. Plots were mowed weekly with a rotary mower at a height of 32 mm with clippings removed.

On 8 Feb. 2011 (47 GDD since 1 Jan. 2011), half of the plots for each grass were treated with glyphosate at 1.1 kg ha⁻¹, using the method of application described for experiment 1. Number of weeds and species were counted before glyphosate treatment. Percent ground cover of weeds was ≈12% and the main weeds were 60% (5 plants/m²) annual bluegrass, 30% (2 plants/m²) orchardgrass (*Dactylis glomerata* L.), and 10% (0.8 plants/m²) meadow fescue (*Festuca pratensis* Huds.). Weed control ratings were conducted 49 DAT, following the evaluation criteria used in experiment 1. Spring green-up ratings

and NDVI readings were taken on 10 Mar., 28 Mar., 9 Apr., 15 Apr., 22 Apr., 30 Apr., 17 and 30 May of 2011, as described in experiment 1. Treatments were arranged in a split plot design with cultivar as main plots and glyphosate treatments (treated vs. untreated) as subplots. Main plots and subplots had four replicates and measured 1.6×4.5 m and 1.6×2.3 m, respectively.

Statistical analysis

Spring green-up visual ratings were arcsine-transformed to improve the homogeneity of the error variance, based on visual checks of residual plots; and then presented as back transformed means. Green-cover visual ratings, NDVI readings, and weed control ratings were subjected to analysis of variance ($P = 0.05$) using SAS Proc Mixed (version 9.2; SAS Institute, Cary, NC). A repeated measures procedure was used to analyze green-cover visual ratings and NDVIs. A compound symmetry covariance structure resulted in the best fit for the data (lowest AIC value) for each experiment. Tukey's honestly significant difference test was used at the P level of 0.05 to identify significant differences among means. Pearson's correlation coefficients for visual ratings and NDVI readings were determined using SAS Proc Corr (version 9.2; SAS Institute, Cary, NC).

Results and discussion

Experiment 1

A single application of glyphosate satisfactorily controlled the weeds present, providing 98% control on average, with no differences between the two applications (8 and 21 Feb.) nor between locations. The analysis of variance of spring green-up revealed significant two-way interactions between glyphosate treatment and evaluation date, and between location and evaluation date. Spring green-up was also significantly affected by glyphosate treatment, location, and evaluation date. However, the interaction terms glyphosate treatment \times location and glyphosate treatment \times location \times evaluation date were not significant; therefore the data were pooled over glyphosate treatment or location.

When data were averaged over the locations, untreated plots had 8 to 13% more green cover than other plots on 9 and 15 Apr., with no differences between the two glyphosate applications (Fig. 29). However, all plots reached \approx 90% green cover by the end of April and full green cover by the end of May. These results for 'Zeon' manilagrass are similar to those reported by several studies for bermudagrass, whose spring green-up was not delayed by winter applications of glyphosate (Johnson, 1976, 1977, 1980). Averaged over the three glyphosate treatments, Sommacampagna plots showed earliest spring green-up, with higher green cover than at the Somma Lombardo location from 9 Apr. until the end of the month (Fig. 30). These differences in speed of green-up observed between the two locations could be due to the different meteorological conditions which occurred at the two locations in March and April (Fig. 31). In Somma Lombardo there were lower minimum daily temperatures and higher monthly precipitation than in Sommacampagna, together with relatively low incident solar radiation. These differences may have led to lower temperature in the soil for Somma Lombardo compared to Sommacampagna (Hillel, 1998), which could explain the observed variation in spring green-up between the two sites (Youngner, 1959; Rimi *et al.* 2011). Despite the large disparity in spring green-up between locations, there was no significant interaction between glyphosate treatment and location, which suggests that the responses of glyphosate application are consistent across contrasting environmental conditions.

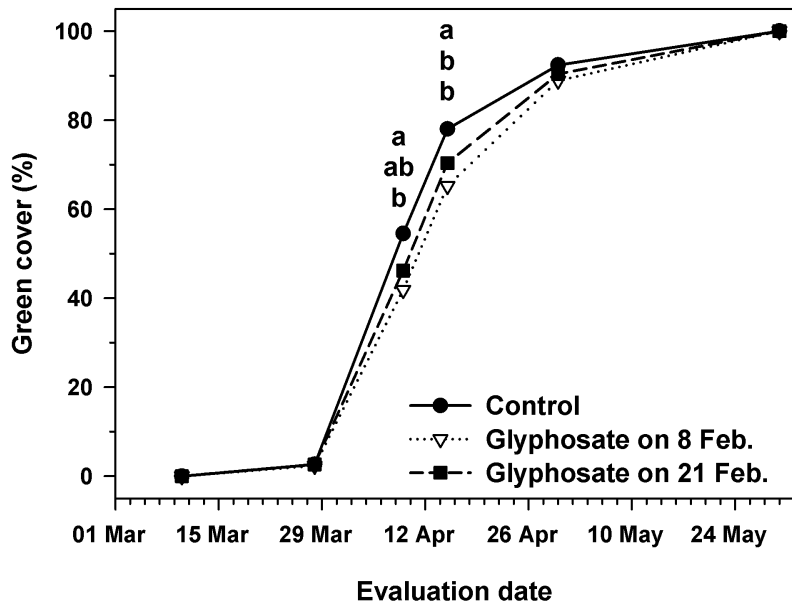


Figure 29 – Percent green cover of ‘Zeon’ manilagrass as affected by three glyphosate treatments from Mar. 2011 to May 2011. Data are averaged over two locations. Within each date, different letters denote statistical differences according to Tukey’s honestly significant difference test ($\alpha = 0.05$).

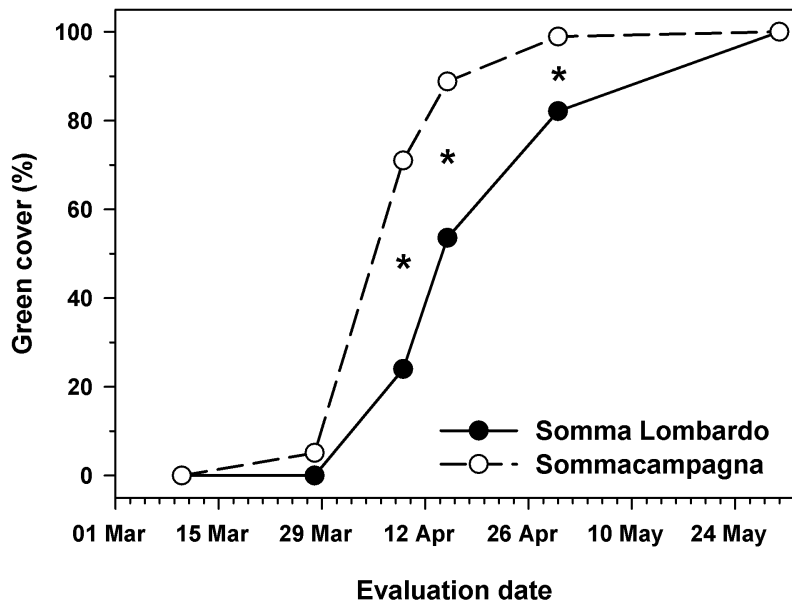


Figure 30 – Percent green cover of ‘Zeon’ manilagrass as affected by location (Somma Lombardo vs. Sommacampagna) from Mar. 2011 to May 2011. Data are averaged over three glyphosate treatments. Differences between locations according to Tukey’s honestly significant difference test ($\alpha = 0.05$) are indicated by *.

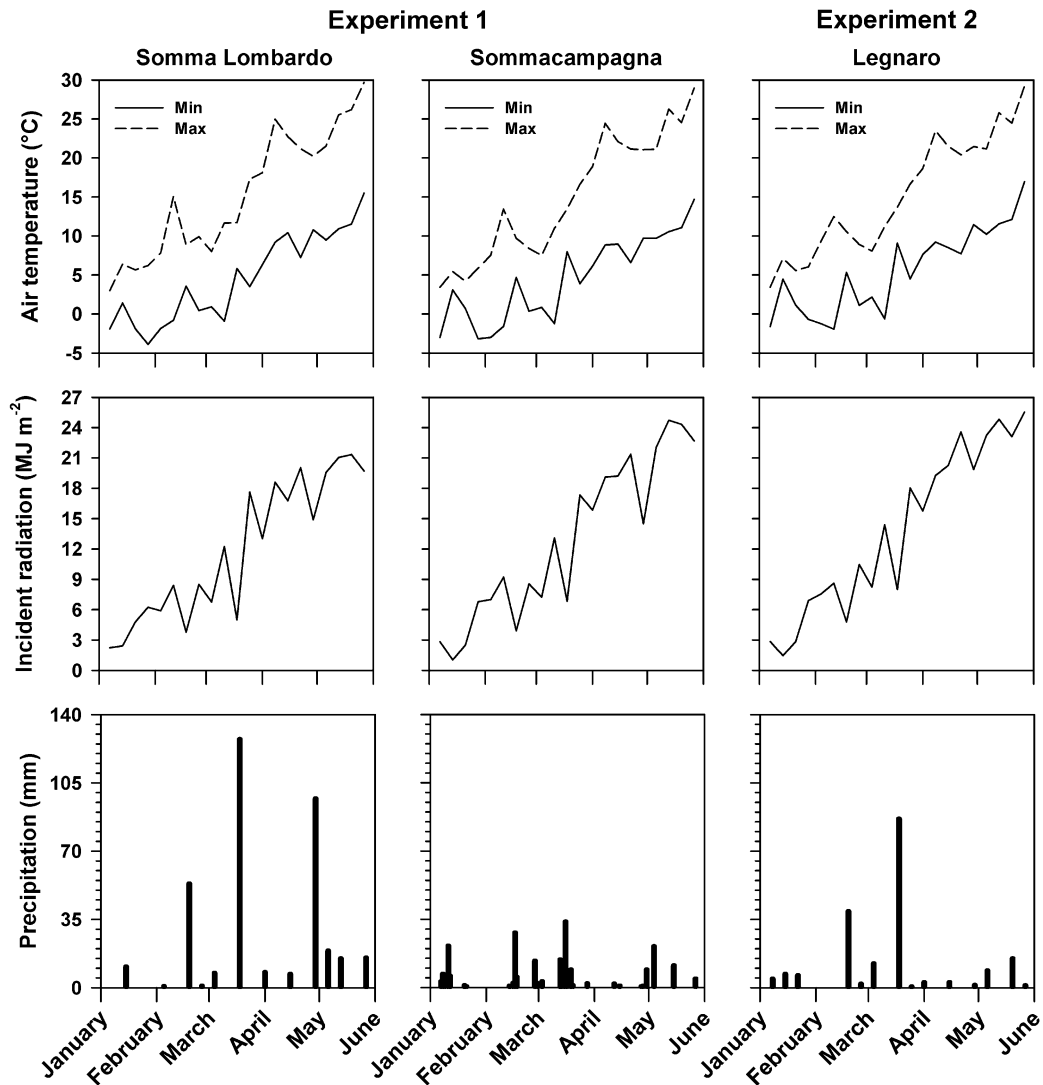


Figure 31 – Weekly maximum (Max) and minimum (Min) air temperature, incident solar radiation, and precipitation from Jan. 2011 to May 2011 collected at weather stations located in close proximity to the field sites in Sommacampagna, Somma Lombardo, and Legnaro, northern Italy.

Responses of NDVIs to the significant interaction terms glyphosate treatment \times evaluation date and location \times evaluation date were consistent with those of the visual ratings (Figs. 32 & 33). The visual ratings of green cover were closely and positively correlated with NDVI readings collected during the green-up period in both locations (Fig. 34). These results agreed with previous findings indicating that NDVI was strongly related to turf color (Bell *et al.*, 2000, 2002). This significant correlation corroborates that NDVI could provide an unbiased estimate of turf color and potentially replace the more time consuming visual ratings (Bell *et al.*, 2009; Schiavon *et al.* 2011).

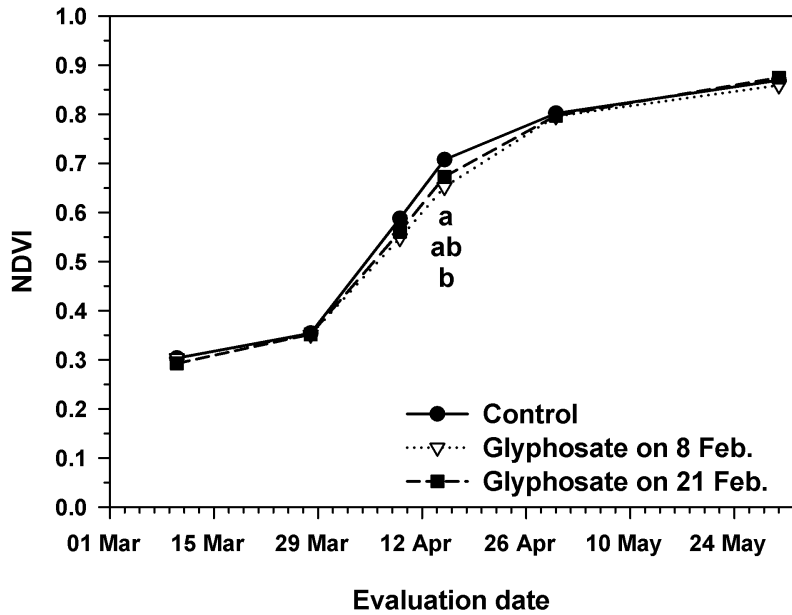


Figure 32 – Normalized difference vegetation index (NDVI) of ‘Zeon’ manilagrass as affected by three glyphosate treatments from Mar. 2011 to May 2011. Data are averaged over two locations. Within each date, different letters denote statistical differences according to Tukey’s honestly significant difference test ($\alpha = 0.05$).

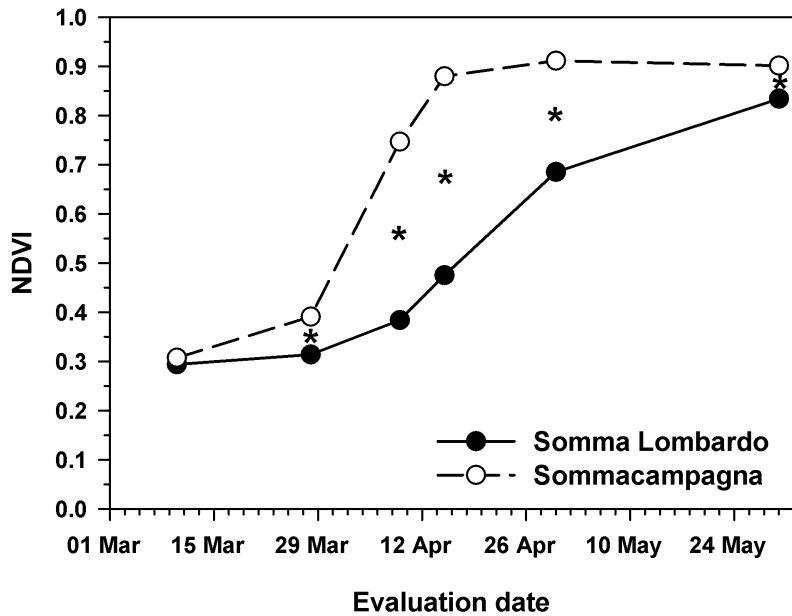


Figure 33 – Normalized difference vegetation index (NDVI) of ‘Zeon’ manilagrass as affected by location (Somma Lombardo vs. Sommacampagna) from Mar. 2011 to May 2011. Data are averaged over three glyphosate treatments. Differences between locations according to Tukey’s honestly significant difference test ($\alpha = 0.05$) are indicated by *.

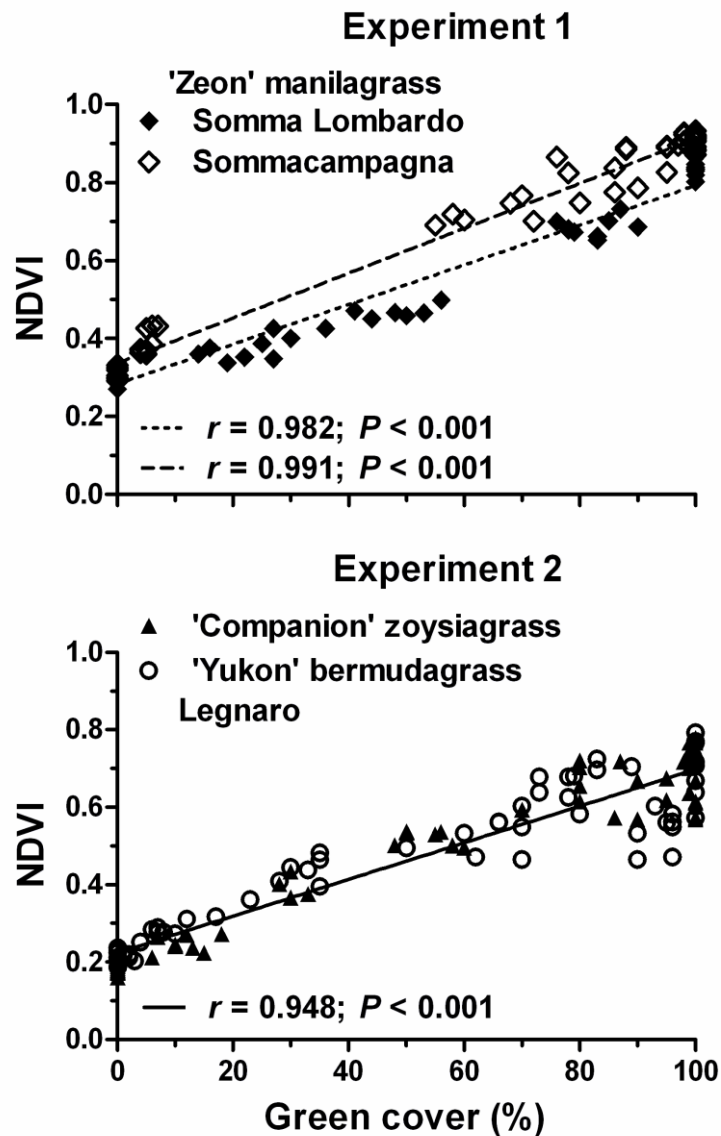


Figure 34 – Correlation between green cover visual ratings and normalized difference vegetation indices (NDVI) during the spring green-up period (Mar. 2011 to May 2011) for ‘Zeon’ manilagrass in Somma Lombardo and Sommacampagna, northern Italy; and for ‘Companion’ zoysiagrass and ‘Yukon’ bermudagrass in Legnaro, northeastern Italy.

Experiment 2

The weed population present at Legnaro was controlled by winter-applied glyphosate, as was observed in experiment 1, with 97% control in both turf species. Results of the analysis of variance of green cover data showed a significant three-way interaction among cultivar,

glyphosate treatment, and evaluation date. The interaction terms cultivar × glyphosate treatment, cultivar × evaluation date, glyphosate treatment × evaluation date, and all main effects were also significant.

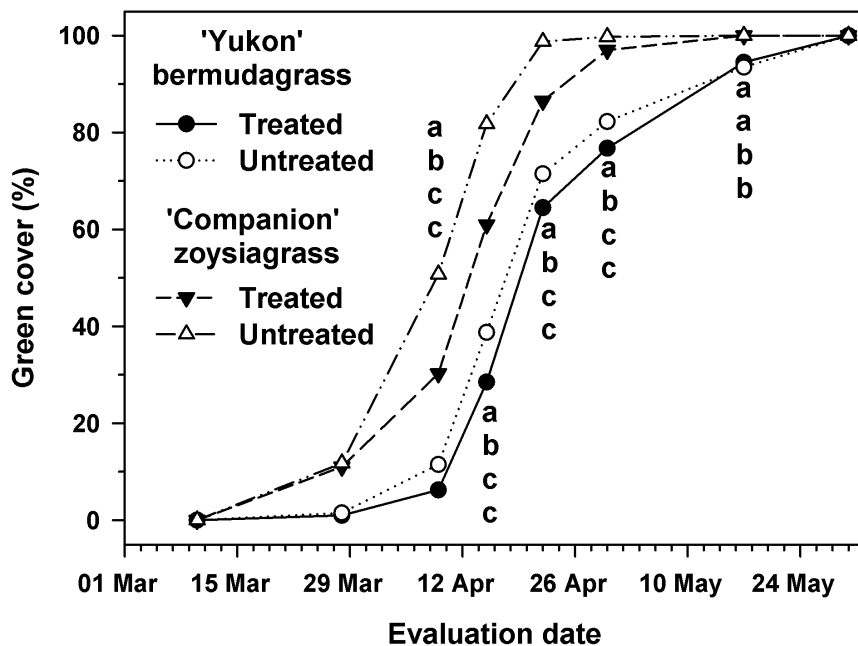


Figure 35 – Percent green cover of ‘Yukon’ bermudagrass and ‘Companion’ zoysiagrass as affected by winter application of glyphosate from Mar. 2011 to May 2011 in Legnaro, northeastern Italy. Within each date, different letters denote statistical differences according to Tukey’s honestly significant difference test ($\alpha = 0.05$).

Green cover of ‘Companion’ zoysiagrass was 21% lower in plots treated with glyphosate compared with controls on 9 and 15 Apr., and 12% lower on 22 Apr. (Fig. 35). Compared to untreated plots, winter-applied glyphosate delayed full spring green-up of ‘Companion’ zoysiagrass from 22 Apr. to the end of April. Meteorological parameters in Legnaro throughout the study period were similar to those recorded in Sommacampagna (Expt. 1) (Fig. 31), where green-up of ‘Zeon’ manilagrass showed no delay as a result of the February applications of glyphosate (Figs. 29 & 32). Therefore, these preliminary findings suggest that the effect of winter-applied glyphosate on spring green-up could differ depending on zoysiagrasses. These results are in agreement with previous research documenting differential tolerance levels of zoysiagrass species/cultivars to other

herbicides. Johnson (1978) reported that the growth of ‘Meyer’ zoysiagrass was less injured than ‘Emerald’ or ‘Matrella’ after applications of benfen [N-butyl-N-ethyl-2,6-dinitro-4-(trifluoromethyl)-benzenamine] or bensulide (O,O-diisopropyl S-2-phenylsulfonylaminoethyl phosphorodithioate). More recently, further tolerance differences among zoysiagrasses have been pointed out with regard to other preemergence (Johnson & Carrow, 1999) and also postemergence herbicides (Patton *et al.*, 2006). Collectively, these findings indicate that weed control in zoysiagrasses should be tailored on the basis of varietal genetic diversity and its influence on herbicide tolerance.

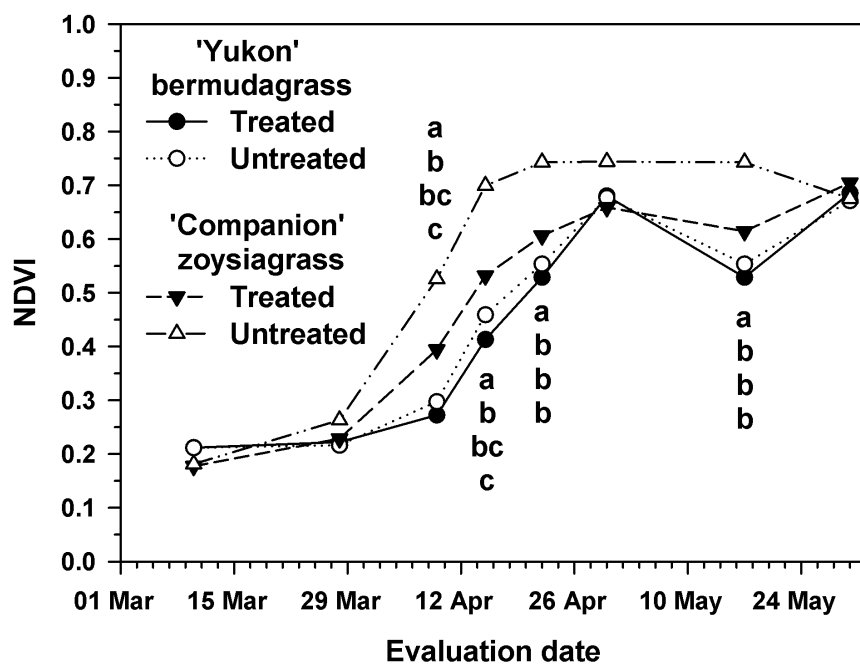


Figure 36 – Normalized difference vegetation index (NDVI) of ‘Yukon’ bermudagrass and ‘Companion’ zoysiagrass as affected by winter application of glyphosate from Mar. 2011 to May 2011 in Legnaro, northeastern Italy. Within each date, different letters denote statistical differences according to Tukey’s honestly significant difference test ($\alpha = 0.05$).

Green cover of ‘Yukon’ bermudagrass was not affected by glyphosate treatments throughout the evaluation dates (Fig. 35). These data support previous studies that reported no effects on spring green-up of bermudagrass treated with glyphosate during the winter (Johnson, 1976, 1977, 1980). Time needed to achieve green-up varied greatly among species, with ‘Companion’ zoysiagrass completing spring green-up by the end of April, while ‘Yukon’ bermudagrass being fully green one month later (Fig. 35). These findings

are similar to those reported from two localities of Turkey, where zoysiagrass cultivars had faster spring green-up than bermudagrass cultivars (Severmutlu *et al.*, 2011). Normalized difference vegetation indices responded to the different treatments similarly to visual ratings, with the significant interaction cultivar \times glyphosate treatment \times evaluation date being likely related to environmental factors (Fig. 36). Green cover ratings were highly correlated with NDVI for both ‘Companion’ zoysiagrass and ‘Yukon’ bermudagrass (Fig. 34), as observed in experiment 1.

Conclusions

Spring green-up of zoysiagrasses and control of winter weeds are both critically important to sod producers to optimize re-establishment planting after harvesting. This research suggests that, under experimental conditions, glyphosate applied as a single treatment at 1.1 kg ha⁻¹ in February can effectively control winter weeds of ‘Zeon’ manilagrass, without injuring turf in the spring. However, additional preliminary findings indicated that winter-applied glyphosate delayed spring green-up of ‘Companion’ zoysiagrass in a similar environment. This suggests that extending this practice to other *Zoysia* species/cultivars should be evaluated on a cultivar-by-cultivar basis. In addition, this study has corroborated that winter-applied glyphosate can efficiently control winter weeds without delaying spring green-up of bermudagrass turf. The results of this study also confirmed a strong positive correlation between visual ratings of turf green cover and NDVI measured with a handheld optical sensor.

Main conclusions and perspectives

In the transition zone, the selection of species and cultivars, along with the appropriate management practices, are critical for reducing the dormancy period of warm season turfgrasses. In the area of study, characterized by a subtropical climate with hot, humid summers and cool winters, the studied management practices had limited influences on the duration of winter dormancy. Applying N fertilizer until late-season moderately anticipated spring green-up of seashore paspalum and bermudagrass cultivars; and also slightly delayed fall color retention of bermudagrasses. The application of spring scalping enhanced green-up of bermudagrass, however its effectiveness was of practical importance only for cultivars that were slow to green-up in the spring. This research also indicated that winter-applied glyphosate effectively controlled winter weeds in bermudagrass, manilagrass, and zoysiagrass resulting in little or no delay of the spring green-up. Therefore, management decisions appeared not critical for improving agricultural performances of warm season turfgrasses in the area of study.

In the Venetian Valley, the choice of species/cultivar should be considered of primary importance relative to management practices for extending the period of practicability of warm season turfgrasses. None of the species and cultivars tested displayed a lack of adaptability to the environmental conditions of the study area, surviving over several winters. However, large difference in spring green-up occurred among species, with zoysiagrass and manilagrass transitioning out of dormancy earlier than bermudagrasses, while seashore paspalum being slowest. Further differences were observed among seeded bermudagrass cultivars concerning spring green-up, fall color retention, and dry weight of overwintering stolons. Additional findings indicated that the cultivars of bermudagrass, seashore paspalum, and zoysiagrass were also different in summer quality as a result of dissimilar water stress avoidance. Therefore, the choice among these warm season grasses in a specific area should be pondered on the requirements for both early spring color and drought tolerance.

Bermudagrass cultivars did not differ in summer quality under study conditions, such that irrigation was not provided and grass was mowed weekly. It would also be

interesting to study the influence of specific cultural practices for high maintenance turfs, such as irrigation strategies and intensive cuttings, on bermudagrasses in northern Italy. From a general standpoint, while effective tools have been pointed out for reducing the period in which warm season turfgrasses remain dormant, this study highlighted that winter dormancy is not avoidable. Further research is needed to investigate alternative solutions for a year-round green cover under no water supply management, such as mixing warm season and cool season grasses. In such plant community, cool season species should dominate in the mixture during the winter months and be naturally replaced by warm season species in summer. In this case, cultural practices may interact with weather patterns in determining the temporal shifts in species dominance within mixtures. Further studies are warranted for transferring and arranging the knowledge achieved during the course of this project into the mixtures of cool season and warm season grasses.

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Acknowledgments



Dr. Stefano Macolino

Prof. Umberto Ziliotto

Prof. Antonio Berti

Prof. Andrea Battisti

Erica Barolo, Andrea Battistella, Marisa Cossu, Pier Alberto Gobbo, Daniela Guglielmi, Alessandro Menegon, Stefania Migliorini, and Davide Zanin for their technical assistance and help in data collection

The workers of agricultural experimental farm ‘Lucio Toniolo’ for field assistance

Vittorio Ferrari (bromatology laboratory, Dept. of Environmental Agronomy and Crop Production) and the personnel of LAZ – LCQA laboratories (Dept. of Animal Science) for their help in the performance of the experiments

Staff of library ‘Pietro Arduino’ for filling literature requests

Claudia Dal Buono and Antonio Timoni for being nice co-workers



Prof. Bernd Leinauer

Dr. Rossana Sallenave

Prof. Leonard Lauriault

Marco Schiavon, Matteo Serena, and Elena Sevostianova for being friendly colleagues

The sod farms **Fedrigo** and **Torrani** for hosting a field trial



Financial support for this doctoral project was provided by ‘Veneto Agricoltura’ – Veneto Region Agency; D.R. 2778 – 12 August 2008

Meeshell for making me a better person