Introduction

Virginia Tech's Worsham Field is considered a high quality turf, implying it must be grown to a level that is functional and aesthetically acceptable for National Collegiate Associate of America-sanctioned Division I collegiate football. This study evaluated traffic tolerance characteristics of two different hybrid bermudagrasses, Tifsport and Patriot, deemed acceptable for high quality athletic field turf in the in the upper transition zone climate of SW Virginia. Tifsport tended to recover poorly from the combined stresses of traffic, fall overseeding with perennial ryegrass (*Lolium perenne* L.) and low temperatures. Field trials conducted as part of the National Turfgrass Evaluation Program (NTEP) indicated that Patriot had similar cold tolerance to Tifsport, but also a much more aggressive growth rate. Patriot was installed on Worsham Field in May 2006 to improve recovery characteristics.

Increased tiller density due to repeated trinexapac-ethyl applications is thought to increase wear tolerance. Also increased endongenous cytokinin levels have been shown to initiate new tiller formation in turfgrasses, consequently increasing turf density (Murphy and Briske, 1992). Denser turfgrass canopies have been associated with improved resistance to wear (Trenhom et al., 2001). Furthermore, increased endogenous cytokinin levels have been shown to increase carbohydrate partitioning to the crown (Ervin and Zhang, 2003). Increased levels of carbohydrates provide energy for auxillary bud growth, resulting in an increase in tiller density. Increased soluble carbohydrates may result in more energy for wear recovery and cold acclimation—an important factor when managing bermudagrasses in cooler climates. Based upon this evidence our hypothesis is that the use of cytokinin-enhancing PGRs may improve the traffic tolerance and repair of overseeded Patriot and Tifsport bermudagrasses

Literature Review

Turfgrasses

All grasses belong to the family *Poaceae.* The family is divided into approximately 7500 species. Only a few dozen of these species are tolerant of mowing and traffic and, therefore, adapted for use as turfgrasses (Turgeon, 1980). Turfgrasses are divided into two groups: cool season (C3) and warm season grasses (C4). The grasses differ in climatic requirements and the way they carry out photosynthesis (Wiecko, 2006). Warm season grasses grow most vigorously at temperatures of 27ºC-35ºC, whereas cool season grasses do best at temperatures of 15ºC -22ºC (Turgeon, 1991). Often sports field managers in the transition zone will grow both. For example, Worsham Field consists of Patriot bermudagrass (*Cynodon dactylon* var. *dactylon* x *Cynodon transvaalensis*), overseeded in the late summer with perennial ryegrass (*Lolium perenne* L.). This is done to maintain fall into spring aesthetics. Without perennial ryegrass overseeding the field would develop unattractive worn areas and become a straw-brown color as the bermudagrass became dormant. The rapid germination and dark green color of perennial ryegrass makes it a good candidate for the maintenance of field aesthetics. In the fall football season, field managers often will apply as much as 3000 kg $ha^{-1}$of perennial ryegrass seed. Overseeding events typically begin in late August-early September in the transition zone of the United States and continue throughout the football season, typically ending around the beginning of November.

Overseeding, from an agronomic perspective is detrimental to bermudagrass vigor and density (Askew et al., 2006). The main effects are seen in the spring as bermudagrass begins to actively grow. During this period, the growing perennial ryegrass vigorously competes with bermudagrass for nutrients, water and light. In the south, heat and drought often result in uniform perennial ryegrass mortality, allowing the bermudagrass to become a monostand (Askew et al., 2006). However, in the cooler transition zone, perennial ryegrass must be chemically controlled with a selective herbicide. There are several herbicides available including pronamide, metsulfuron, trifloxysulfuron sodium, foramsulfuron and rimsulfuron (Askew et al., 2006). These transition-aid herbicides are generally effective in killing the perennial ryegrass without causing bermudagrass phytotoxicity. Often, however, the product requires two to three applications for 100% control.

Leaf cross sections of bermudagrass (C4 species) and perennial ryegrass (C3 species) reveal two different types of anatomy (Parrish, 2006). The perennial ryegrass leaf typically consists of general mesophyll cells (palisade and spongy). Carbon dioxide (CO_2) enters the plant via stomatal openings that lead to intercellular spaces within the leaf. In this area it is dissolved in the water overlying the mesophyll cells, allowing it to move across the mesophyll membrane where it can react with chloroplasts (Hay and Porter, 2006). Within the chloroplast, light energy raises the energy levels of sensitive pigments, leading to the assimilation of CO_2 and consequent synthesis of plant dry matter. This is labeled as being the light dependent reaction of photosynthesis, where photons split water, releasing electrons, protons and O_2 (Hay and Porter, 2006). Electrons move through the “Z scheme” to produce NADPH, ATP, and O_2. These substrates are used to fuel the Calvin-cycle. In this cycle, the large and ubiquitous enzyme, RubisCO (ribulose bisphosphate carboxylase-oxygenase), captures CO_2 to convert it into sugars. RubisCO enzyme structure has an affinity towards CO_2; however, one out of nine times it captures O_2 (Hay and Porter, 2006). The oxygenase activity of RubisCO causes dry matter to be lost. C3 plants have developed a partial remedy to the flaw via photorespiration. When RubisCO becomes oxygenated it forms one molecule of glycollate and one molecule of phosphoglyceric acid (PGA). The PGA can still be used to make dry matter; however, the molecule of glycollate cannot be used in the Calvin cycle. Photorespiration, through a series of reactions, transforms glycollate into PGA. The PGA is then re-able to be processed in the Calvin cycle. As said earlier, photorespiration is only a partial remedy to the oxygenase activity of RubisCO. Three out of four carbons that leave the chloroplast as glycollate return as PGA (Hay and Porter, 2006). In a sense, a quarter of the carbon is lost and not able to be used for dry matter production.

The cross section of a bermudagrass leaf indicates that it has mesophyll cells and bundle-sheath cells. The bundle sheath cells typically comprise 10% of the leaf cells, the remaining are mostly comprised of mesophyll cells (Hay and Porter, 2006). Within the C4 mesophyll cells, the cytosol assimilates bicarbonate (HCO_3^-). The assimilation is catalyzed by phosphoenylpyruvate carboxylase (PEPcase). When this happens, HCO_3^- is carboxylated to phosphoenylpyruvate (PEP) to form oxaloacetate (OAA). Oxaloacetate is reduced in the chloroplast to malate. Malate moves symplastically into the bundle-sheath cells where it is decarboxylated to form pyruvate and CO_2 (Hay and Porter, 2006). Within the bundle sheath cells the concentration of CO_2 to O_2 is much higher than what occurs within the Calvin cycle of C3 plants. This allows RubisCO almost complete efficiency in carboxylating CO_2 molecules. The pyruvate formed by the decarboxylation of malate, moves back to the mesophyll cell where it is reformed into PEP where it can re-start the cycle. The ability of warm-season grasses to carboxylate CO_2 more efficiently than cool-season grasses makes them more efficient in utilizing light and water.

Bermudagrasses

Bermudagrasses are commonly grown on intensive-use areas such as lawns, golf courses and athletic fields. They have many characteristics that make them an attractive choice for turf use. These traits include a long-lived perennial sod forming nature, aggressive spreading ability, tolerance to low mowing heights, good tolerance of wear and compaction, and adaptation to a wide range of growing conditions (Christians, 2004; Taliaferro et al., 2004). Bermudagrasses have good heat, drought and traffic tolerance, and few disease problems. The most desirable trait is its recuperative ability, since sports areas are consistently subjected to injury from foot traffic and divoting (Karcher et al., 2005).

Two out of the nine bermudagrass species of the genus *Cynodon* are used for turf use. *Cynodon dactylon* is the most common and widespread species used. It is distributed throughout the world between the northern and southern latitudes of 45° (Taliaferro et al., 2004). It has at least 20 different common names, including, Bermudagrass, Bahama grass, Devil's grass, Couch grass, Kweek, Indian doob, Dog-tooth grass and Wire grass (Wiecko, 2006). The names imply that it is often more a pest and is considered to be a noxious weed in many parts of the country. The presence of vigorous rhizomes and stolons that allow it to be an ideal grass for heavily trafficked athletic fields also makes it a difficult to control weed.

Bermudagrass will begin the process of cold acclimation when soil temperatures fall below 10ºC. Plant leaves become a straw-brown color as they lose chlorophyll. Typically, a heavy frost will send it into dormancy. It will remain in this dormant phase until soil temperatures rise and remain above 10ºC. As temperatures and sunlight decrease, bermudagrasses begin to acclimate to the change in climate by way of a number of physiological changes. Most importantly, accumulation of cryoproctectants (sugars and polyamines) in the cytoplasm and changes to the composition of cell membranes occurs (Leyser and Day, 2003). The accumulation of sugars, typically in the stolons, also serve as energy for re-growth in the following spring. Root and rhizome growth greatly increase at soil temperatures around 15.5-20ºC. Optimum growth occurs at soil temperatures of 24-30ºC (Christians, 2004).

Cynodon dactylon originated from Africa (Wiecko, 2006). Sub-species can be found world wide, growing in tropical and sub-tropical climates. *Cynodon dactylon* typically has a coarse leaf texture, low shoot density and forms a loose knit sod, although improved varieties of the 21st century are noted for finer leaf texture and improved density.

The other major bermudagrass species used for sports turf is *Cynodon transvaalensis*, (commonly called "African bermudagrass"). *Cynodon transvaalensis* is native to areas of South Africa. Its distinctive traits include a compact growth habit, fine leaf texture, and yellow-green leaf color (Taliaferro et al., 2004).

Cultivar improvement has been achieved by screening for plants with superior characteristics related to turf use (Taliaferro, et al., 2004). Some of the newer varieties have been selected for their cold tolerance. The adaptation to survive colder temperatures has allowed bermudagrass to successfully grow in the northern part of the transition zone.

Most bermudagrasses being produced for turf purposes are interspecific hybrids (*Cynodon hybrida*) derived from the cross of the two *Cynodon* species: *Cynodon dactylon x Cynodon transvaalensis* (Taliaferro, et al., 2004). The breeding strategy for the development of bermudagrass hybrids involves the selection of good parent plants of a respective species and doing genetic crosses to produce large numbers of progeny that can undergo screening tests to be selected for elevated traits, such as cold hardiness, turf quality, sod strength, disease and drought tolerance (Taliaferro et al., 2004). The cultivar Patriot is a recent product of this effort. Patriot is a F_1 hybrid developed by crossing the bermudagrass hybrid, 'Tifton-10' (*Cynodon hybrida*), with a *Cynodon transvaalensis* parent (Taliaferro et al., 2004).

Patriot is a high quality turf that performs well in the transition zone. Its characteristics include dark green foliage, excellent sod-strength, high density, aggressive vertical and lateral shoot growth and superior cold tolerance. Most interspecific F_1 hybrid bermudagrasses are triploid (27 chromosomes). Patriot differs from other F_1 hybrid varieties in that it is tetraploid, receiving 27 chromosomes from the Tifton 10 parent and nine chromosomes from the *C. transvaalensis* parent (Taliaferro et. al, 2004). These characteristics have made Patriot a popular choice for sports turf. Because of its superior cold tolerance many fields in the northern transition zone and northward have been planted to Patriot. Some recognizable universities that have installed Patriot on game fields are Purdue University, George Mason University and Virginia Tech. In 2006, Worsham Field at Virginia Tech was completely renovated and sodded with Patriot.

Tifsport, released in 1997, is another popular hybrid bermudagrass used in the transition zone. It was developed by inducing a mutation of the interspecific hybrid 'Midiron' with gamma-irradiation (Hale, 2003). 'Midiron' is a hybrid bermudagrass developed by crossing *C. transvaalensis* and *C. dactylon* (Hale, 2003). Tifsport's characteristic dark green foliage, medium texture and high shoot density make it a high quality turf (Hale, 2003).

NTEP transition zone data (2004) indicates Tifsport and Patriot have relatively equal qualities (Table 1) (NTEP, 2004). However, in 2005, Tifsport was rated to have had significantly higher quality than Patriot in the North Carolina trial (NTEP, 2005). Fall (November) color ratings reported Patriot to be consistently lighter green than Tifsport, indicating Patriot's tendency to go into fall dormancy earlier than Tifsport. In this trial, both species' fall density and percent winter kill were rated as equivalent. However, laboratory trials have also reported Tifsport has lower freeze tolerance than Patriot, at -7.9ºC and -9.7ºC, respectively (Anderson et. al, 2003).

Table 1. A comparison of 2004 NTEP ratings for bermudagrass cultivars Patriot and Tifsport.

2004 NTEP Turfgrass Characteristics									
	Quality (Oct)§			Fall Color (November)			Density (Fall)		% Winter Kill
	NTEP Location								
Cultivar	VA1¥	VA2	NC	VA1	VA 2	NC	SC	OK	OK
Patriot	4.3	7.7	6.7	3.0	7.0	7.0	7.0	8.3	1.7
Tifsport	5.7	7.3	6.7	7.3	7.7	7.0	7.0	8.7	4.3
LSD†	2.6	0.8	4.2	1.6	0.9	3.8	0.5	0.7	7.0

†Statistical differences can be determined by subtracting one entry's mean from another entry's mean. Means are statistical different when the value is lager than the LSD value.

§Turf quality, fall color and density are rated on a 1-9 scale; 9 being the maximum; % Winter kill is rated on 1-100%; 100% is complete mortality

¥ VA 1 location is Blacksburg Virginia; VA 2 location is Virginia Beach, Virginia.

Table 2. A comparison of 2005 NTEP ratings for bermudagrass cultivars Patriot and Tifsport.

2005 NTEP Turfgrass Characteristics									
	Quality (Sept)§			Fall Color (November)			Density (Fall)		% Winter Kill
	NTEP Location								
Cultivar	VA1	VA2	NC1	VA2	NC	SC	SC	CA	KS
Patriot	7.0	7.3	6.3	6.0	5.0	5.3	7.0	7.3	73.3
Tifsport	6.7	7.7	8.3	6.7	6.0	7.3	7.0	7.0	80.0
LSD	3.4	0.9	1.1	1.3	1.1	1.7	0.3	0.9	23.7

†Statistical differences can be determined by subtracting one entry's mean from another entry's mean. Means are statistical different when the value is lager than the LSD value.

§Turf quality, fall color and density are rated on a 1-9 scale; 9 being the maximum; % Winter kill is rated on 1-100%; 100% is complete mortality

¥ VA 1 location is Blacksburg Virginia; VA 2 location is Virginia Beach, Virginia.

What is traffic?

In regards to turfgrass management, the term traffic refers to two major factors: physical damage and compaction of the soil (Brosnan et al., 2005). High amounts of traffic cause abrasion and tearing of leaf tissue, characterized by the symptoms of leaf necrosis and chlorosis. Chlorosis is caused by the degradation of chlorophyll. This is turn, results in the plant being less photochemically efficient with less chlorophyll available to initiate the conversion of CO_2 into dry matter (Trenholm et al., 2000).

A turf's traffic tolerance is characterized by its ability to withstand heavy foot and/or mechanical traffic. It is important to the management of high use areas, such as sports fields, golf courses, and high-trafficked landscapes (Karcher et al., 2005). The abrasion and tearing of leaf tissue causes the protective cuticle to be damaged, providing pathogens a mode of entry and lowering plant water efficiency. Turf exposed to high amounts of traffic eventually develops bare spots. The bare spots are the result of compacted soils and chronic plant damage. Bare spots allow more sunlight penetration and less moisture competition to weed seeds, ultimately increasing the susceptibility to weed infestations (Trenholm et al., 2000).

Severe compaction of the soil reduces turf density, causes premature senescence of shoot and root tissue and inhibits the plant's ability to recover (Trenholm et al., 2000). However it should be noted that any form of compaction, which reduces aeration and increases root impedance has been shown to limit plant growth (Foil, 1965). Plants growing in soils with high bulk densities have to expend more energy to grow roots (Glinski and Stepniewski, 1985). High levels of root impedance lead to poor root structure and a plant that is much less efficient at harvesting light and soil resources (Brady and Weil, 2002). More energy is required to "push" roots through the soil. Compacted soils develop higher percentages of micropores, causing water to be held in the soil matrix for longer periods of time, ultimately reducing the oxygen diffusion rate (Black, 1984). When the diffusion rate of oxygen is reduced, turf respiration and nutrient and water uptake are lowered, ultimately causing the plant to be less efficient in making food (Black, 1984; Rengel, 2002). Because oxygen diffuses through water at much slower rates than through the air, plant roots become susceptible to suffocation. It was shown that the concentration of ethyl alcohol increased with a decrease in the rate of oxygen. High amounts of accumulated ethyl alcohol results in a decrease in plant growth (Fulton and Erickson, 1964).

Water infiltration rates are lowered when the soil becomes compacted (Black, 1984). This is caused by the disruption of macropores (Foil, 1965). During periods of heavy watering or rainfall, water is not able to infiltrate at fast enough rates, resulting in high amounts of water run-off and erosion (Kramer and Boyer, 1995).

There are many plant growth mechanisms that impact traffic tolerance. Physiological, anatomical, and morphological characteristics of a turf species/variety can play key roles in ability to resist wear (Shearman and Beard, 1975; Trenholm et al., 2000; Brosnan et al., 2005). Kentucky bluegrass (KBG) leaf and stem total cell wall constituents (cellulose, hemicellulose, lignin, and lignocellulose) showed little association to wear tolerance at the interspecies level; however, the combined effects of the constituents accounted for a significant variation in KBG's ability to tolerate wear (Brosnan et al., 2005). Furthermore, it was shown that more sclerenchyma fibers and lignified cells were correlated with improved wear tolerance. The same study also showed that combined effects of increased leaf tensile strength and increased leaf width enhanced the ability to resist tearing (Shearman and Beard, 1975).

Physiologically, the ability for a turfgrass to resist wear has been attributed to cell turgidity. This is achieved by having maximum leaf and stem moisture contents and sufficient concentrations of nutrients, particularly potassium (K). Trenholm (2001), working with hybrid bermudagrass cultivars, reported that wear tolerance was increased based on tiller density, shoot moisture, and tissue K concentration. High K concentrations have been shown to lower total cell wall content (TCW) in hybrid bermudagrasses. Reduction in TCW content in seashore paspalum (*Paspalum vaginatum* Swarz.), has been shown to increase wear tolerance (Trenholm, 2001).

Potassium is released from soil minerals very slowly by the process of chemical and physical weathering (Wiecko, 2006). It is typically readily available in the soil solution or in an exchangeable form on the soil colloid (Wiecko, 2006). Potassium concentrations vary among soils; it is mainly dependent on the soil's clay content. It has been shown that a soil with 21% clay content had higher concentrations of potassium in the soil solution compared to a soil with 4% clay content (Marschner, 1986). Soils with high clay contents have higher cation exchange capacities (CEC) than sandier soils, resulting in high clay content soils having higher concentrations of exchangeable potassium (Marschner, 1986).

Morphologically, the higher the tiller density, the more likely a turfgrass has better wear tolerance. The greater amount of leaf blades per area lessens the amount of force per blade.

Also, some researchers suggest plants with more tillers have more meristematic growth points in which it can grow and repair when damaged (Trenholm et al., 2000).

Wear and turfgrass species

The differences between cool season grasses (C3) and warm season grasses (C4) have been shown to influence wear tolerance. Typically C3 grasses are less wear tolerant than C4 grasses (Trenholm et al., 2000). This is due to the fact that C4 grasses produce more lignified tissue than C3 grasses. High amounts of lignified tissue contribute to stronger blades and stems. Another factor that contributes to wear tolerance is growth habit. The majority of C3 grasses are clump forming, whereas C4 grasses typically spread by large numbers of stolons and/or rhizomes. A heavy stolon/rhizome network forms a dense mat that better tolerates wear. Utilization of the proper species/varieties is needed to maintain the integrity of a high-use turf.

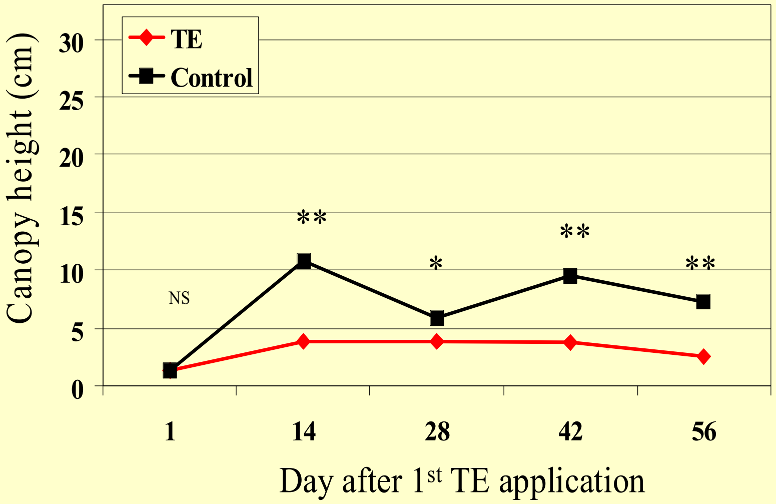
Plant Growth Regulators and Hormones

Plant growth regulators (PGRs) are natural or synthetic chemicals that stimulate or inhibit growth and development. Many PGRs are natural hormones extracted from plant tissues or synthetic compounds that mimic naturally occurring hormones. There are five main natural plant hormones that are widely accepted. They include gibberellins, cytokinins, auxin, ethylene, and abscisic acid**.** Recently it has been discovered that brassinosteroids, jasmonates, salicylic acid and the peptides have hormonal characteristics (Davies, 2005)**.**

There is a wide use of PGRs in turf management. For high maintenance turf they are used to lower mowing frequency and clipping production, suppress annual bluegrass (*Poa annua* L.), promote tiller growth and density, and improve stress tolerances (Ervin and Zhang, 2007). The most commonly used synthetic inhibitory PGRs are flurprimidol, paclobutrazol, mefluidide, ethephon, and trinexapac-ethyl. Out of these, trinexapac-ethyl (TE) is probably the most widely used in fine turf management.

Trinexapac-ethyl [4-(cyclopropyl-[α]-hydroxymethylene)-3,5-dioxo-cyclohexane carboxylic acid ethyl ester] is predominately used to reduce mowing frequency and clipping production (Figure 1) (Ervin and Zhang, 2003; McCullough et al., 2006). TE, when properly applied, reduces leaf elongation at four to seven days after first application and will typically result in a 50% reduction in clipping yield (Ervin and Zhang, 2004). Turf quality (Figure 2) has been shown to be increased with TE applications, namely because of increased density and darkening of leaf color (Ervin and Zhang, 2003).

Figure 1. Canopy height of Bermudagrass as influenced by trinexapac ethyl (Ervin and Zhang, 2003)



‘*’’**’, significant cultivar difference on LSD (0.05), LSD(0.01), respectively; NS, not significant.
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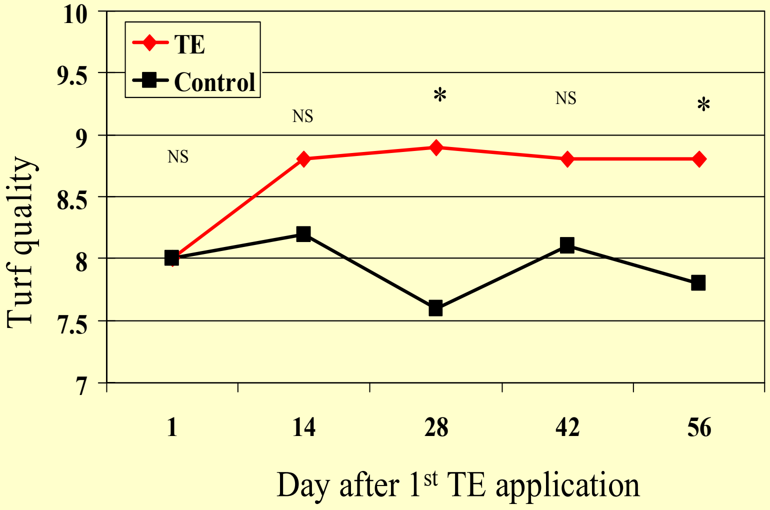
Figure 2. Turf quality of Bermudagrass as influenced by trinexapac ethyl (Ervin and Zhang, 2003)



‘*’’**’, significant cultivar difference on LSD (0.05), LSD(0.01), respectively; NS, not significant.
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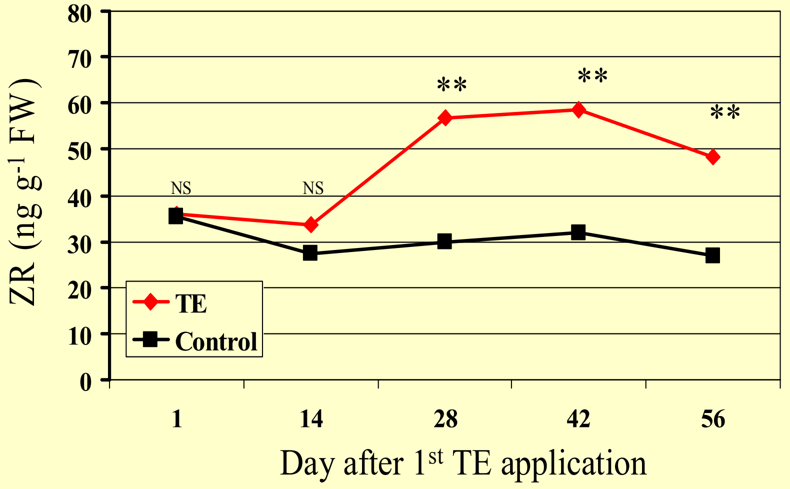
Frequent and long-term use of TE by turf managers has revealed it to have many more beneficial growth effects. It has been shown in some instances that sequential applications of TE reduced evapotranspiration rates, improved shade and heat tolerance, and lessened the severity of dollar spot on creeping bentgrass (*Agrostis stolonifera*) (Ervin and Zhang, 2007).

Trinexapac-ethyl is a Class A PGR that inhibits the conversion of GA_{20} to physiologically- active GA_1 by competitively inhibiting the regulatory enzyme 3-β-hydroxylase (Ervin and Koski, 2001). In short, cell elongation is reduced. Applications of TE result in turf becoming darker green and denser. The darker green appearance is due to an increase in leaf cell and chlorophyll density (Ervin and Koski, 2001). Increased cell density is believed to increase cell wall content. This has been correlated to improve the wear tolerance (Heckman et al., 2005).

Increased turf density is achieved by new tiller formation and growth. In order for tiller formation and growth to occur, energy must be allocated to the crown and apical dominance must be eliminated (Ervin and Koski, 2001)**.** Further measurements of TE effects on turf growth have indicated that cytokinin levels may often be increased. Ervin and Zhang (2003) showed that TE increased canopy photochemical efficiency (Figure 3) and concentration of the plant cytokinin, zeatin riboside (Figure 4).

Figure 3. Leaf zeatin riboside (ZR) content as influenced by trinexapac ethyl in bermudagrass

(Ervin and Zhang, 2003)



‘*’’**’, significant cultivar difference on LSD (0.05), LSD(0.01), respectively; NS, not significant

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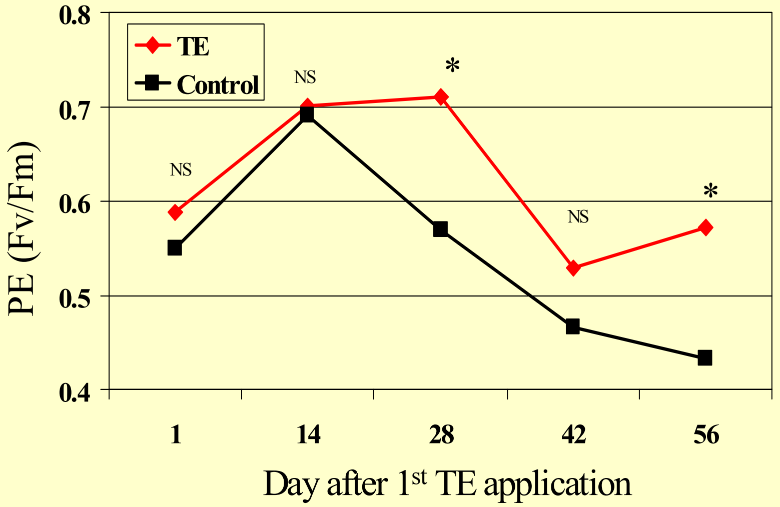
Figure 4. Photochemical efficiency (PE) of Bermudagrass as influenced by trinexapac ethyl (Ervin and Zhang, 2003)



'*''**', significant cultivar difference on LSD (0.05), LSD(0.01), respectively; NS, not significant.
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Cytokinins and bermudagrass traffic tolerance

Cytokinins were first discovered by a group of plant physiologists at the University of Wisconsin who reported that unripe coconut milk (endosperm) contained a substance that promoted cell division (Thimann, 1977). After further investigation for certain extracts that promoted similar growth changes they concluded that effective extracts needed to contain nucleic acids (Thimann, 1977). As a result, they worked with yeast DNA preparations and were able to isolate a substance called "Kinetin" (6-furfurylaminopurine, $C_{10}H_9N_5O$) and observed it to be very active in promoting mitosis and cell division (Moore, 1989). Kinetin was soon coined as the present term "cytokinin", meaning any compound that promotes cell division and regulatory functions in the same manner as kinetin (Thimann, 1977). The term kinetin is still used to describe the natural cytokinin, 6-furfurylaminopurine (Moore, 1989). Since then, many natural cytokinins have been discovered.

Research has shown that cytokinins influence a wide array of important physiological growth processes in plants. Some of the most relevant are their participation in the induction of cell division and shoot/tiller formation, promotion of chloroplast development, increasing the sink activity of cells, preventing leaf senescence, and inducing opening of stomata (Smith et al., 1996).

There is a bit of confusion as to the way in which cytokinin translocates through the plant. Abundant data reveal that exogenous cytokinins, when applied to leaves, stems, and buds, exhibit little if any movement from the site of application. On the other hand, it has been shown that the application of exogenous cytokinins increase endogenous cytokinin levels (Mok and Mok, 1994). Research has shown that cytokinins can induce their own synthesis. It is therefore thought that applications of exogenous cytokinin to cytokinin-requiring tissue could initiate an autoinductive mechanism that induces synthesis of cytokinins (Mok and Mok, 1994).

Cytokinins are a constituent of xylem and phloem sap (Moore, 1989). When cytokinin is present in the vascular tissues, it moves in a process regarded as a source-to-sink relationship. The source or biosynthesis of cytokinin is a bit unclear because cytokinins exist in the plant at extremely low levels (Mok and Mok, 1994). Although cytokinins are primarily synthesized in roots, particularly root apices, they have been found to be biosynthesized in other meristematic tissues and organs (Pharis and Rood, 1990). It was previously thought that cytokinins are synthesized in root tip meristems and translocated throughout the xylem sap. However, it is now understood that cytokinins are at high concentrations in all meristematic regions. This is because cells need cytokinin for the induction of cell division. Cytokinins promote cytokinesis by regulating the event that is necessary for the transition from G2 to mitosis (Fosket et al., 1981).

Axillary buds are formed by cell divisions in the third innermost layer of the apical meristem. Increased tiller density is achieved by the initiation of successive tillers from axillary buds of previous tillers (Murphy and Briske, 1992). In order for axillary buds to grow and form new tillers, apical dominance must be eliminated (Murphy and Briske, 1992). Apical dominance refers to the control exerted on axillary bud formation and growth (Mok and Mok, 1994). Naturally, cytokinins and auxins form a balanced ratio that is responsible for axillary bud formation and growth; changes in the ratio impact the way a plant develops. The plant hormone auxin (indoleacetic acid or IAA), produced in the apical meristem and young leaves, is known to be the controlling factor in apical dominance (Murphy and Briske, 1992). IAA is transported to lateral buds and when at high enough concentrations, a direct inhibition of lateral bud growth occurs. It has been observed that increases in exogenous cytokinin on inhibited axillary buds causes apical dominance to be overcome, allowing for axillary bud growth to occur (Mok and Mok, 1994). One hypothesis is that IAA adheres to vascular traces of axillary buds, causing a blocking and prevention of cytokinin synthesis and utilization (Murphy and Briske, 1992).

Cytokinins are thought to be the main hormone that controls leaf senescence (Thimann, 1977). The role of cytokinin in leaf senescence was first observed with a demonstration that immersed a detached *Xanthium* leaf into water with kinetin. It was observed that the kinetin solution possessed a regulatory function that prevented the *Xanthium* leaf from breaking down, allowing it to retain its green color (Kefeli, 1978; Mok and Mok,1994). Furthermore, in a study using tobacco leaves, nonsenescent leaves were shown to have much higher concentrations of cytokinin than senescent leaves (Mok and Mok, 1994).

When perennial turfgrasses go into dormancy, the aerial portion of the leaf senesces and necrosis continues throughout the length of the leaf until it reaches the basal portion of the sheath (Moore, 1989). The most prominent change is a decline in proteins and nucleic acids, accompanied by chlorosis, due to the loss of chlorophyll (Moore, 1989). Cytokinins have been shown to sustain nucleic acid and protein synthesis (Table 3).

Table 3. Contents of chlorophyll and protein in leaves of wild-type tobacco (WT) and tobacco with autoregulated production of cytokinin (APC) (Moore, 1989).

	Chlorophyll mg/m^{-2}		Protein g/m^{-2}	
	WT	APC	WT	APC
Senescing	70 ±5	177±6	0.50±0.02	1.13±0.05

Leaf senescence is correlated with increases in proteolytic enzyme activity and a decrease of lipase and lipoxygenase. All are involved in membrane breakdown. Maintaining membrane integrity prevents leakage of nutrients and enzymes, resulting in a plant that better resists injury (Mok and Mok, 1994).

The ability of cytokinins to enhance anti-senescence properties is attributed to an increase of in-plant natural production of antioxidants. Antioxidants such as α-tocophenol (vitamin E) and ascorbic acid (vitamin C) are concentrated in the chloroplast. When a plant is subjected to stress, vitamins E and C protect the photosynthetic apparatus by scavenging reactive oxygen species that could potentially damage it (O'Dell, 2003).

A leaf has an elevated respiration rate when it senesces (Wingler et al., 1998). At high levels of respiration, plants use energy at rates that exceed production. Cytokinins, with the ability to prevent leaf senescence, lower the rate of energy used for respiration (Thimann, 1977). Since cytokinins possess the ability to maintain protein and chlorophyll synthesis, it is thought that cytokinins might possibly enhance a plant's photochemical efficiency. If so, the finding that trinexapac-ethyl increased photochemical efficiency (Fig. 4) might be attributed to an increase in ZR content (Fig. 3).

Cytokinins have been found to induce opening of stomata. As a result, transpiration and nutrient flow through the xylem are enhanced. Given sufficient water, it is thought that CO_2 assimilation rates are increased, contributing to more carbohydrates being produced (Mok and Mok, 1994).

Cytokinins are classified into two different groups: synthetic phenylurea derivatives or adenine derivatives. CPPU [*N*-(2-chloro-4-pyridyl)-N'-phenylurea] is classified as a synthetic phenylurea, whereas 6-BA [6-benzylaminopurine] is labeled as an adenine derivative (Kadota and Niimi, 2003). The two different cytokinins have similar structure-relationships and essentially the same activities, i.e., growth promotion of callus and stimulation of shoot formation (Mok and Mok, 1994).

There are a number of synthetic cytokinins on the market that are available for various plant uses. Most are used in fruit production to increase fruit size and delay senescence after harvesting. Synthetic phenylurea derivatives typically have more activities than adenine derivatives.

Grapes (*Vitis labrusca* and *V. labrusca* x *V. vinifera*) sprayed at four CPPU concentrations (0, 5, 10, or 15 $mg \bullet L^{-1}$) showed significant increases in berry mass, cluster mass and compactness. It was also observed that berry firmness was linearly related to CPPU concentration (Zabadal and Bukovac, 2006). Increases in berry volume are associated with increases in sugar after véraison. The berry expands in volume as solutes accumulate. Also, the extent to which cell division occurs at the first stage of berry development has been shown to affect the eventual size of the berry (Kennedy, 2002).

Research was conducted on the effects certain cytokinins (6-BA, CPPU, and thidiazuron (TDZ)) on lateral branching of *Aridisia pusilla*. 6-BA solutions were sprayed at concentrations of 500, 750, 1000, 1,500, or 2,000 $mg \bullet L^{-1}$; CPPU and TDZ solutions were sprayed at concentrations of 10, 20, 30, 40, or 50 mg•L^{-1}. The study showed all the cytokinins induced lateral branching. BA produced the most desirable branching, whereas CPPU and TDZ treated plants developed short and degenerated lateral shoots (Lee, 2005). Cytokinins' ability to induce lateral branching is thought to improve turf traffic tolerance. The more tillers per area may result in the ability to better resist wear.

Rationale and Significance

Traffic stress provides major challenges to sports turf managers and ways to improve traffic tolerance are continually sought. Therefore it is appropriate to investigate the use of cytokinins to improve the quality and traffic tolerance of bermudagrasses. Cytokinins' ability to increase tiller density, delay senescence, and enhance photochemical efficiency are all factors that could improve traffic tolerance. Further investigation is needed to see if this concept is scientifically valid.

Research Methods

Hypothesis:

The use of cytokinin-enhancing PGRs will improve the traffic tolerance and repair of overseeded Patriot and Tifsport bermudagrasses.

Objectives:

Examine if exogenous applications of synthetic cytokinins (CPPU and 6-BA) or TE, increase the traffic tolerance of Patriot and Tifsport bermudagrasses. Cultivar by PGR by traffic interactions in terms of field performance were also investigated.

Blacksburg Climate

The USDA cold hardiness climate (Figure 5 and Figure 6) zone of Blacksburg Virginia is 6b. Temperatures can go as low as -23ºC to -20.5ºC. The lowest recorded temperature during each year of the present study (Figure 5) was -15ºC on February 6, 2007 and -13.9ºC on February 19, 2006. Rainfall (Figure 6) was relatively normal. Some precipitation extremes were a month long drought in September, 2005 and 28 cm of rain in June, 2006.

Figure 5. Monthly High, Low, and Average temperatures for Blacksburg, VA (NOAA, 2007).

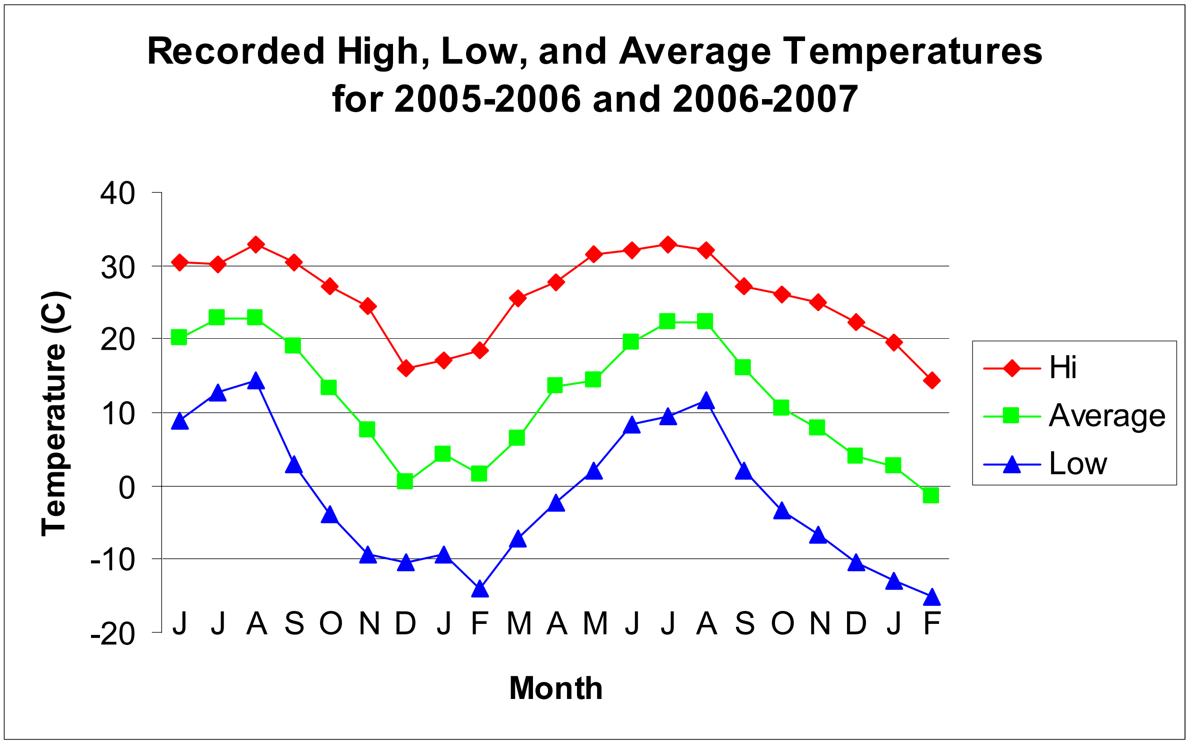
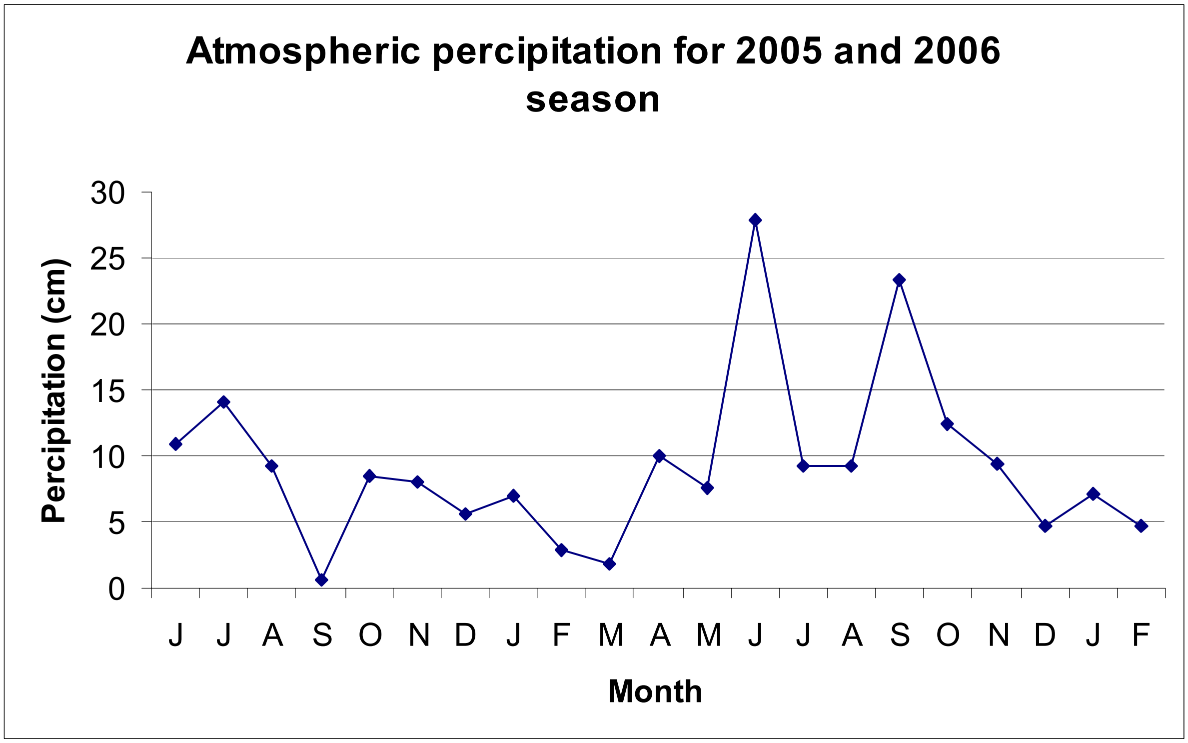


Figure 6. Atmospheric precipitation for 2005 and 2006 for Blacksburg, VA (NOAA, 2007).



Experimental Design

Tifsport and Patriot plots were managed at the Virginia Tech Turfgrass Research Center in Blacksburg, Virginia. Plots were established on a Groseclose silt loam (fine, kaolinitic, mesic typic Hapludult). Each cultivar was previously managed and well established. A split-split plot design was used with three factors and four replications. Main plots were Tifsport and Patriot; sub-plots were traffic and no traffic; sub-subplots were PGRs: TE, CPPU, 6-BA, and control. Experimental units were 1.8 x 1.8 m (36 ft^2).

Traffic and PGR treatments

The two annual trials began on September 8, 2005 and September 7, 2006, respectively. Every two weeks, a traffic treatment was administered using a differential-slip traffic simulator (Picture 1) in an attempt to match the cumulative damage done on Virginia Tech's football facility, Worsham Field, during the football season. The traffic simulator was adapted from a design by Ohio State University by Dr. Robert Grisso of Virginia Tech's Biological and Science Engineering Department.

Picture 1. Differential-slip traffic simulator

In 2005, the amount of roller passes (Table 4) averaged between 10-20 passes per traffic treatment. In 2006, the decision was made to increase the amount of roller passes to better exemplify what was seen on Worsham Field. Roller passes done outside the trial period were done to mimic the traffic exposure Worsham Field experienced outside the football season. Such events were spring games, scrimmages, and camps.

Table 4. Total amount of traffic simulator passes performed on both cultivars to mimic traffic damage observed on Worsham Field during the 2005 and 2006 seasons.

2005		2006	
Number of Simulator Passes			
September 8, 2005	20	January 12, 2006	30
September 23, 2005	10	Febuary 10, 2006	30
October 6, 2005	10	April 19, 2006	20
October 20, 2005	10	June 15, 2006	20
November 3, 2005	20	September 7, 2006	30
Total	70	September 20, 2006	30
		October 4, 2006	12
		October 19, 2006	20
		November 2, 2006	30
		Total	222

PGRs were applied on a two week basis during the simulated football season. Application dates were September 2, September 14, September 29, October 14, October 28, and November 9, of 2005 and August 31, September 15, September 28, October 12, October 31, and November 7, of 2006. The two synthetic cytokinins (6-BA and CPPU) were provided by Valent Biosciences Corporation. The products were applied at the following levels with a CO_2 pressurized walk-behind sprayer:

1. MaxCel (6-Benzyladenine) at 0.185 kg a.i.• ha^{-1}
2. Prestige (2-Chloro-4-pyridyl-phenylurea) at 0.025 kg a.i.• ha^{-1}
3. Primo Maxx (Trinexapac-ethyl) at 0.097 kg a.i. • ha^{-1}

Data Collection

Data were collected on a two week frequency on turfgrass color, quality and percent cover. Pictures were taken along with data collection for later referencing. Data collection dates were September 15, October 17, November 1, and November 15, 2005 in year one and September 18, October 2, October 16, October 31, and November 13, 2006 in year two. Percent turf cover was determined with a 100 section grid platform (Picture 2).

Picture 2.100 section grid platform used to determine percent turf cover.

The platform was positioned at the same orientation for each measurement. Each section of the grid was rated on a 0-5 scale: 0-complete turf coverage; 1-visible damage-chlorotic tissue; 2-broken stolon tissue and visible bare areas forming; 3-significant damage-more than 50% the area is bare; 4-25% or less turf coverage; 5-no turf coverage. The following equation was developed to determine % turf cover: 100-(Total grid ratings/500) * 100 = % turf coverage.

Turf color was rated on a 1-9 scale: 1-brown; 9-dark green. Turf quality was measured in the same manner: 1-poor quality; 9-excellent quality.

Bulk densities for each plot were measured at the end of each season to provide a measure of compaction. Sampling was done in the manner described by Blake (1965). Two samples from each sub-sub plot were taken with a bulk density sampler. The verdure of each sample was removed and placed into plastic bags for later stolon viability and carbohydrate content analyses. The remaining amount of soil was measured for length and trimmed at the bottom so that each sample had a volume of 149.5 cm^3. Soil samples were then completely dried in an oven at 120ºC for 48 hours. Following the drying period, samples were weighed and bulk density was calculated.

Cultural Management

During the active growing season, plots were double-mowed two to three times per week with a reel cutting unit. Mowing height was set at 1.6 cm and clippings were returned. Soil cores were taken on a regular basis to check moisture and thatch status. Irrigation was dependent on soil dryness and plant wilting and was applied to maintain active turfgrass growth. Many times, watering was based on what was received/not-received from rainfall events (Figure 6). Care was taken to relieve compacted soil. Core aeration events occurred on three different dates: September 27, 2005 and May 23, 2006, 0.635 cm cores were removed with a hollow-tine aerator at 5.08 cm x 5.08 cm spacing; July 18, 2006, 1.27 cm cores were taken with a hollow-tine aerator at 5.08 cm x 5.08 cm spacing. Each aeration event consisted of two passes.

For the first year, fertilization was congruent with that done on the sand-based Worsham Field at Virginia Tech's Lane Stadium. In 2006, the decision was made to modify the fertilization program to better suit the native soil plots at the Turfgrass Research Center (Table 5).

Table 5. The fertility program administered to the bermudagrass plots for 2005 and 2006 season.

2005		2006	
Fertility Program			
August 12th	24.2 kg/ha 34-0-0	May 10th	48.4 kg/ha 18-28-24
August 22nd	36.3 kg/ha 10-10-10	June 14th	48.4 kg/ha 46-0-0
August 29th	24.2 kg/ha 10-10-10	July 10th	48.4 kg/ha 46-0-0
September 30th	48.4 kg/ha 12-6-4	August 7th	48.4 kg/ha 46-0-0
October 12th	48.4 kg/ha 24-6-12	September 6th	48.4 kg/ha 10-10-10
Total	181.5 kg/ha	October 11th	48.4 kg/ha 10-10-10
		Total	290.4 kg/ha

Overseeding events (Table 6) started in late August and continued throughout the trial period. Overseeding was done after traffic events. The perennial ryegrass used was "Allsport", supplied by Lesco; this blend had a 90% germination rate and 3.09% inert ingredients. The amount of pure live seed (PLS) was 87 %. When plots were overseeded, seed was applied with a drop spreader in two directions. Attempts to incorporate seed into the turf canopy were made. At first, the strategy involved pulling a cocoa mat over the plots. Later, it was found to be more efficient to use a gasoline-powered leaf blower to force seed down into the canopy.

Table 6. Kilograms (kg) of perennial ryegrass applied per hectare (ha) during overseeding events for 2005 and 2006 season.

2005		2006	
kg of pure live seed/ha			
August 22nd	484.4 kg/ha	August 28th	484.4 kg/ha
August 29th	242.2 kg/ha	September 28th	484.4 kg/ha
September 17th	242.2 kg/ha	October 14th	484.4 kg/ha
September 29th	484.4 kg/ha	November 3rd	387.5 kg/ha
October 12th	484.4 kg/ha	Total	1,840.7 kg/ha
November 7th	581.3 kg/ha		
Total	2,518.9 kg/ha		

Revolver© (foramsulfuron), produced by Bayer, was applied at 1.27 L•ha^{-1}on June 9, 2006 to facilitate summer transitioning. It was later applied on July 26, 2006 to help eliminate goosegrass (*Eleusine indica* L.) populations that had become prevalent, along with crabgrass (*Digitaria ischaemum*) in the highly worn areas. Tifsport particularly showed high weed populations in some of the trafficked plots (Picture 3). Drive© (quinclorac), produced by BASF Corporation, was applied at 0.026 L•ha^{-1} to control crabgrass. A slight discoloration occurred in the Patriot stand as a result of the application (Picture 4).

Picture 3. Images of weed populations becoming an issue in the highly worn areas

Picture 4. Patriot became slightly chlorotic in response to the quinclorac application

Laboratory analyses

A series of laboratory analyses were performed to determine if the application of exogenous cytokinins promoted growth changes. At the end of each season, stolon samples were collected with a bulk density sampler. The sampler had a volume of 149.5 cm^3. Soil samples were separated by removing the top 3.8 cm that contained the vegetative portion of the turf. The vegetative portions were sealed in a bag and stored in a -10ºC freezer until further analysis. Samples were removed from the freezer and allowed to thaw in a refrigerator overnight at 5ºC. Stolons were removed and cleaned.

Electrolyte leakage assay: Small sections (0.5 cm) of stolon tissue were immersed in 20 mL deionized H_2O and placed under the hood for 15 minutes. Samples were then shaken in the deionized H_2O for 24 hours. Electrical conductivity of the solution was measured twice with a conductivity meter. The first one (EC_1) was taken after the shaking and the second (EC_2) was taken after autoclaving at 120ºC for 20 minutes. Electrolyte leakage was determined by the following equation: $EL (\%) = (EC_1/EC_2)*100$.

During the data analysis period it became evident that storing the samples in the -10ºC freezer was the improper protocol. A set of samples were collected to establish a difference between frozen stored samples vs. non-frozen stored samples. The difference between the two was determined and used to develop a correction equation that was then used to adjust readings from the original samples damaged by the freezing.

The remaining stolons not used for the electrolyte leakage readings were dried and used for the nonstructural carbohydrate assay. It was done in the manner described by Hendrix (1993) and Trapley (1993), as modified by Zhang (2004). **Sugar extraction:** The stolons were dried at 50ºC for 48 hours. After drying, stolons were ground with a coffee grinder and further ground by hand with a mortar and pestle. Ground stolons (20 mg) were placed in a tube containing 4 ml 80% ethanol solution. Plant material and ethanol solution were placed into an 80ºC water bath for 30 minutes. Following the water bath, the solution was brought up to 5 ml with ethanol and centrifuged (3000 x g, for 10 minutes). Extraction solution was collected (1.5 ml) and added to a microcentrifuge tube containing 20 mg activated charcoal. Tubes were shaken and centrifuged (2200 x g), for 15 minutes for a second time. The extracted sugars were stored in a -80ºC freezer until further analysis. **Starch extractions:** Extractions were performed with the residual from the sugar extractions. 1.0 ml of 0.1 M KOH was added to the extracted sugars. The solution was placed into a boiling water bath for 60 minutes. Tubes were allowed to cool and 0.2 ml of 1.0 M acetic acid was added. 100 µl of 1.0 Tris buffer (pH 7.2) and 200 µl alpha-amylase preparation was added to the solution. Tubes were placed into an 85°C water bath for 30 minutes. Tubes were shaken several times during this period. Following the water bath treatment, acetic acid was added to the solution to reduce pH to <5.0. The amount of acetic acid added varied between 250-300 µL. Once pH was properly reduced, 1.0 mL of amyloglucosidase preparation was added. The solution was then placed into a 55ºC water bath and heated for 60 minutes. Following this, enzymatic action was stopped by heating samples in a 100ºC water bath for four minutes. Samples were removed from the water bath and brought up to 6.0 ml with deionized H_2O and vortexed. Extraction solution was collected (1.5 ml) and transferred to a microcentrifuge tube and centrifuged. Starch extractions were stored at -80ºC until further analysis.

Sugar assay: Extracted sugars were taken out of the ultracold freezer and allowed to defrost. Samples (20µL) were pipetted into a microtitration plate, and then allowed to evaporate in an oven at 50ºC for 1.5 hours. After evaporation, 20µl deionized H_2O was added to each well. The plate was covered for 1 hour. Following this, 100µl glucose reagent solution (Sigma GAHK-20) was added to each well of the microtitration plate. The plate was covered for 30 minutes and then read with a spectrophotometer at 340 nm. After running the glucose measurement, PGI was added for the determination of fructose. Again, it was covered for 30 minutes and read at 340 nm. Following the fructose measurement, invertase was added to determine sucrose. Plates were covered for 30 minutes and read again at 340 nm.

Starch assay: Extracted starches (20 µl) were pippetted into the microtitration plate. Glucose reagent solution was added and samples were covered for 30 minutes, afterwards to be read at 340 nm.

Two standards were used for the development of standard curves. The first standard was a combination of the three sugars (glucose, fructose and sucrose) at increasing concentrations. The second standard was solely glucose at increasing concentrations. The equations derived from the first standard curve were used for the determination of glucose, glucose + fructose, and glucose + fructose + sucrose. The equation derived from the second standard curve was used for the determination of starch concentrations. Glucose, fructose and sucrose concentrations were multiplied by a dilution factor of 250. Starch concentrations were multiplied by a dilution factor of 300.

Statistical Analysis

The data were analyzed with ANOVA using SAS software (SAS, 2003). Significant main effects and interactions between variables were determined. Mean separations were performed using a LSD at 5% probability level.

Results/Discussion

Bulk Density

In 2005, there was a significant bulk density increase for trafficked plots (Table 7), indicating that soil compaction had occurred. Summer aeration seemed to partially alleviate soil compaction, because in 2006, a significant difference between traffic treatments was not recorded

Table 7. A comparison between traffic and no-traffic soil bulk densities for 2005 and 2006.

2005		2006	
Bulk Density (gcm^{-3})			
Traffic	No-traffic	Traffic	No-traffic
1.51a	1.42b	1.50a	1.46a

Turf Cover

Significant main effects and interactions were determined with ANOVA and means separated by the Least Significant Difference (LSD) test at an α level of 0.05. The ANOVA test revealed a significant difference (<0.0001) between the 2005 and 2006 seasons; therefore, significant main effects and interactions are presented separately on a yearly basis (Tables 8 and 9). Loss of turf cover was greater in 2006 relative to 2005, most likely due to the increased traffic simulator passes made in 2006 in an attempt to more closely match the turf cover loss observed on Worsham Field. The ANOVA showed that the interactions between traffic x PGR and cultivar x traffic were consistently significant across traffic periods for both years (Tables 8 and 9). Given such consistent effects, these interactive effects will be presented below.

In 2005 and 2006, turf cover was not consistently different between Patriot and Tifsport on the no-traffic plots (Table 10). Measurements were taken in the fall, therefore both cultivars had the summer to grow in and fill any damaged areas. If the measurements had included what was seen in early summer they would have shown Tifsport to have less turf cover than Patriot (Marshall, personal observation). Tifsport plots, compared to Patriot, showed more reduction in turf cover, most likely because it was more susceptible to damage from the combined stresses of low winter temperatures and perennial ryegrass competition. It has previously been reported that Patriot is more cold tolerant than Tifsport (Anderson et. al, 2003). Patriot's higher percent turf cover at early summer indicates that it was more cold tolerant than Tifsport. Worsham Field, when it consisted of a Tifsport playing surface, had undesirably high percentages of spring bermudagrass mortality, likely as a result of the combined stresses of traffic, overseeding, and cold temperatures (Marshall, personal observation). The 2006 renovation of Worsham's surface with Patriot has resulted in the fastest fill-in to 100% cover in the last decade or more (Ervin, personal observation).

In 2006, a significant difference in turf cover was seen after the second traffic treatment between no-traffic Patriot and Tifsport plots (Table 10). The loss of turf cover was apparently due to a June beetle infestation that affected the Patriot area but not the Tifsport. After treating, the cultivars were relatively equal in terms of turf cover for the no-traffic plots.

For both years, as the application of traffic progressed, Patriot maintained greater turf cover than Tifsport (Figures 7 and 8). It was observed that Patriot formed a much denser turf canopy and had a more vigorous growth rate than Tifsport. Previous NTEP results suggest that Patriot and Tifsport have relatively equal turf densities (Tables 1 and 2). However, for Blacksburg Virginia, it is quite evident that Patriot formed a denser turf canopy. Patriot's denser turf canopy and more vigorous growth rate contributed to enhanced traffic tolerance. The renovation of Worsham Field from Tifsport to Patriot should result in a field that better withstands heavy foot and mechanical traffic.

PGRs did not show an effect in terms of overall turf cover for no-trafficked plots (Table 11). However, in both years, trafficked plots treated with TE resulted in significantly less turf cover, relative to the other PGRs and the control (Table 12, Figures 9 and 10). Based on these results it appears that TE applied at 0.097 kg a.i.•ha^{-1} every two weeks to trafficked fall acclimating overseeded bermudagrass was detrimental. It was quite evident in 2005, that TE applied at this rate caused a reduction in turf cover. Even when perennial ryegrass became the dominant growing species, significantly lower turf cover was often seen with plots treated with TE (Figures 9 and 10). Previous research done with 'Tifway' bermudagrass showed that autumn applications of TE, applied at a rate of 0.11 kg a.i.•ha^{-1}, led to slower growth and decreased density (Fagerness et al., 2002). The authors suggest that bermudagrass may have difficulties recovering, due to the cooler temperatures, from the initial suppressive effects of TE. Another study done with Kentucky bluegrass (*Poa pratensis* L.), suggest caution while using TE in trafficked areas (Ervin and Koski, 2001).

Table 8. Percent turf cover Pr > F values for 2005.

	2005 Percent turf cover Pr > F values				
	Traffic Treatments				
Source	Traffic 1§	Traffic 2	Traffic 3	Traffic 4	Traffic 5
Cultivar	<0.0001†	NA	<0.0001	<0.0001	<0.0001
Traffic	<0.0001	NA	<0.0001	<0.0001	<0.0001
PGR	0.0260	NA	<0.0001	0.0011	0.0020
Traffic X PGR	0.0108	NA	<0.0001	0.0123	0.0291
Cultivar X PGR	0.5993	NA	0.0014	0.2047	0.1960
Cultivar X Traffic	<0.0001	NA	0.0039	0.0012	0.0003
Cultivar X Traffic X PGR	0.4092	NA	0.8409	0.1190	0.1195

† Significant main effects and interactions were determined with the Anova LSD test at an α level of 0.05

§ Traffic 1 applied from September 8th – September 17th ; Traffic 2 (NA); Traffic 3 applied from September 17th – October 17th; Traffic 4 applied from October 17th – November 1st; Traffic 5 applied from November 1st- November 15th

Table 9. Percent turf cover Pr > F values for 2006.

	2006 Percent turf cover Pr > F values				
	Traffic Treatments				
Source	Traffic 1§	Traffic 2	Traffic 3	Traffic 4	Traffic 5
Cultivar	0.0205†	0.3882	0.0045	<0.0001	0.0046
Traffic	0.0006	<.0001	<0.0001	<0.0001	<0.0001
PGR	0.4483	0.0113	<0.0001	0.0029	<0.0001
Traffic X PGR	0.5810	0.2529	<0.0001	0.0064	<0.0001
Cultivar X PGR	0.5131	0.8060	0.8517	0.1135	0.0755
Cultivar X Traffic	0.0094	0.0053	0.0014	0.0052	0.0003
Cultivar X Traffic X PGR	0.4092	0.1039	0.8409	0.1190	0.1195

† Significant main effects and interactions were determined with the Anova LSD test at an α level of 0.05

§ Traffic 1 applied from September 7th – September 18th ; Traffic 2 applied from September 18th – October 2nd; Traffic 3 applied from October 2nd – October 16th; Traffic 4 applied from October 16th – October 31st; Traffic 5 applied from October 31st - November 13th

Table 10. Percent turf cover in 2005 and 2006 trials expressed as cultivar and traffic interaction averaged over plant growth regulator treatments.

	2005		2006	
Cultivar	Traffic§	No-Traffic	Traffic¥	No-Traffic
		Traffic 1 †		
Patriot	94.0b	99.0a	97.9a	98.8a
Tifsport	90.3c	99.1a	93.5b	99.0a
		Traffic 2		
Patriot	-	-	93.7c	97.3b
Tifsport	-	-	92.5c	99.5a
		Traffic 3		
Patriot	91.1b	98.6a	73.2b	99.0a
Tifsport	89.1c	97.8a	68.8c	99.3a
		Traffic 4		
Patriot	92.4b	99.5a	93.7b	99.7a
Tifsport	89.2c	98.6a	89.3c	98.8a
		Traffic 5		
Patriot	91.6b	99.7a	87.03b	99.16a
Tifsport	88.9c	99.0a	85.0c	99.5a

† Means followed by the same letter are not significantly different at the 0.05 probability level. Mean separations are determined on a per traffic basis.

§ 2005 Traffic 1 applied from September 8th – September 17th ; Traffic 2 (NA); Traffic 3 applied from September 17th – October 17th; Traffic 4 applied from October 17th – November 1st; Traffic 5 applied from November 1st- November 15th

¥ 2006 Traffic 1 applied from September 7th – September 18th ; Traffic 2 applied from September 18th – October 2nd; Traffic 3 applied from October 2nd – October 16th; Traffic 4 applied from October 16th – October 31st; Traffic 5 applied from October 31st - November 13th

Figure 7. Percent turf cover (2005) for overseeded Patriot and Tifsport plots after traffic treatments averaged over plant growth regulators.

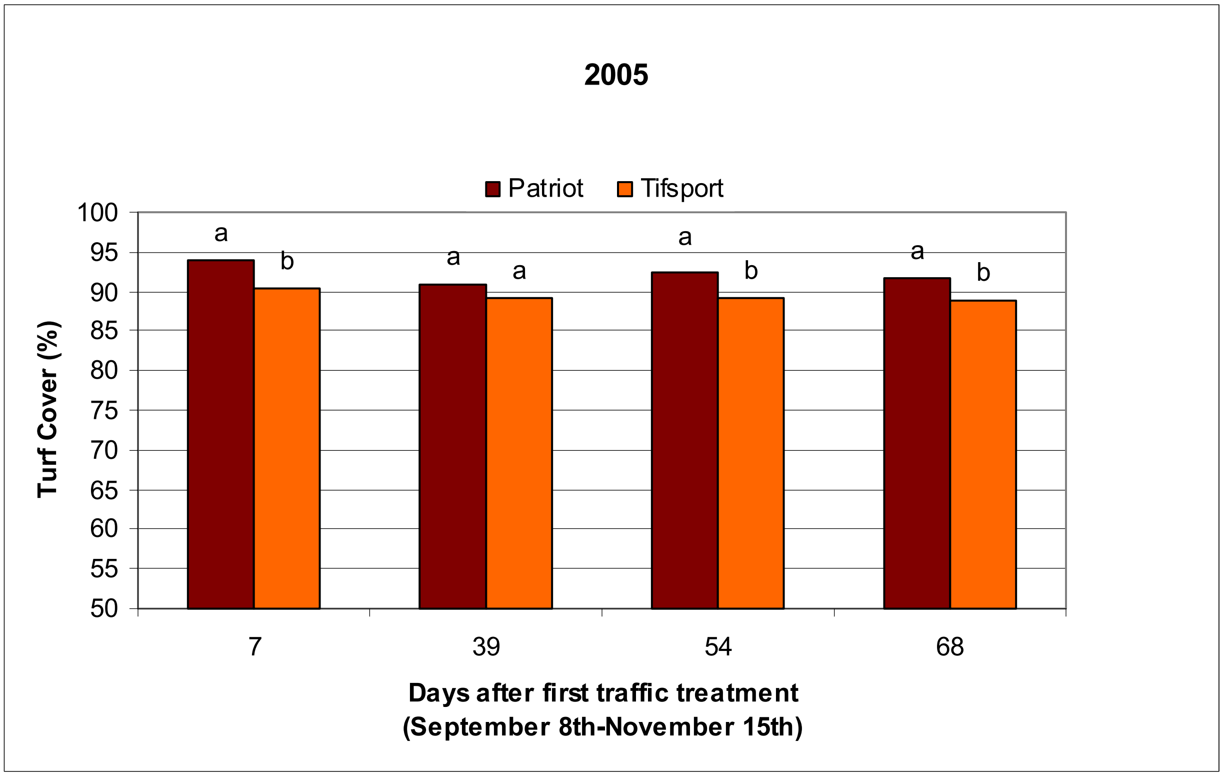


Figure 8. Percent turf cover (2006) for overseeded Patriot and Tifsport plots after traffic treatments averaged over plant growth regulators.

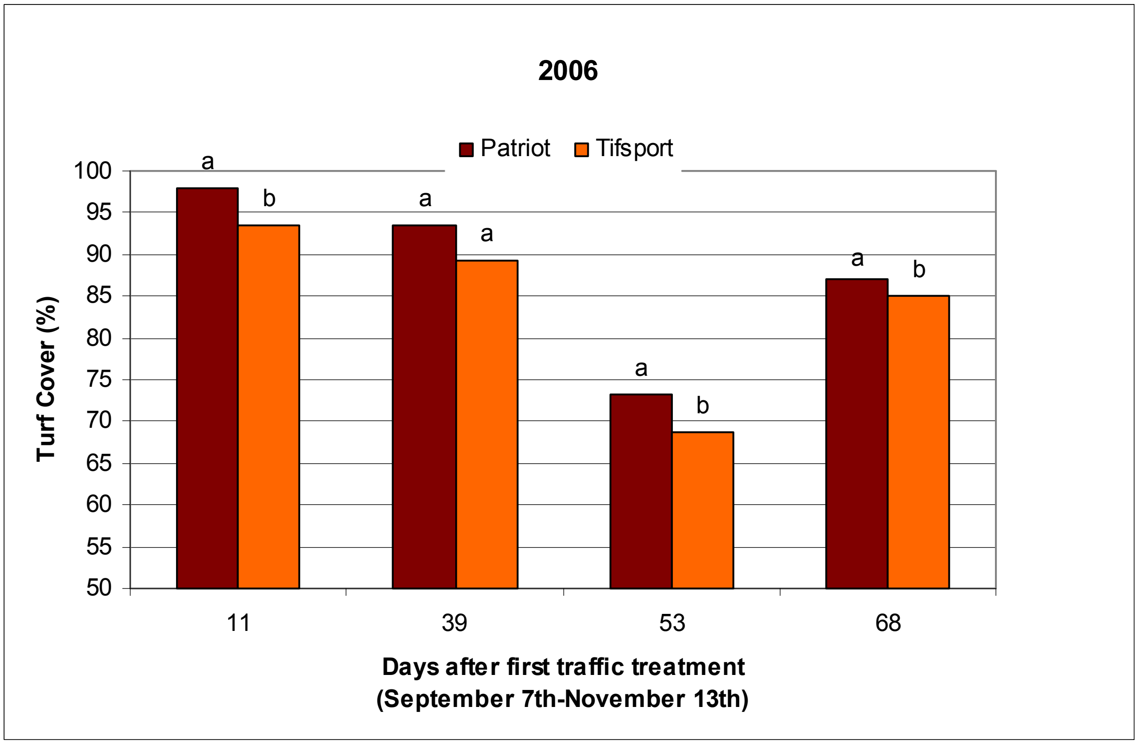


Table 11. Percent turf cover in 2005 and 2006 trials after plant growth regulator and traffic treatments averaged over cultivars.

	2005				2006			
Treatment	Control	6-BAΨ	CPPU	TE	Control	6-BA	CPPU	TE
				% Turf Cover †				
				Traffic 1				
No-Traffic	99.1a	98.7a	99.1a	99.2a	98.7ab	99.1a	99.1a	98.8a
Traffic	92.0bc§	92.3bc	93.1b	91.3c	95.5abc¥	97.6abc	95.0abc	94.9c
				Traffic 2				
No-Traffic	-	-	-	-	98.9a	98.4a	98.7a	97.6a
Traffic	-	-	-	-	93.9b	94.0b	92.3b	92.3b
				Traffic 3				
No-Traffic	98.3a	98.3a	97.8a	98.0a	99.4a	99.3a	99.3a	98.7a
Traffic	91.1b	90.9b	90.4b	87.9c	71.9b	70.7b	70.5b	70.9b
				Traffic 4				
No-Traffic	99.4a	99.3a	98.8a	98.9a	99.4a	99.1a	99.5a	99.1a
Traffic	92.0b	91.6b	91.0b	88.6c	92.7b	93.0b	90.7b	89.6b
				Traffic 5				
No-Traffic	99.7a	99.4a	99.3a	99.2a	99.3a	99.5a	99.3a	99.2a
Traffic	90.8b	91.0b	90.6b	88.6c	85.3bc	87.4b	86.4bc	85.1c

† Means followed by the same letter are not significantly different at the 0.05 probability level. Mean separations are determined on a per traffic basis.

Ψ 6-BA (6-Benzyladenine), CPPU (2-Chloro-4-pyridyl-phenylurea), TE (Trinexapac-ethyl)

§ 2005 Traffic 1 applied from September 8th – September 17th ; Traffic 2 (NA); Traffic 3 applied from September 17th – October 17th; Traffic 4 applied from October 17th – November 1st; Traffic 5 applied from November 1st- November 15th

¥ 2006 Traffic 1 applied from September 7th – September 18th ; Traffic 2 applied from September 18th – October 2nd; Traffic 3 applied from October 2nd – October 16th; Traffic 4 applied from October 16th – October 31st; Traffic 5 applied from October 31st - November 13th

Figure 9. Percent turf cover (2005) after traffic and plant growth regulator treatments averaged over cultivars.

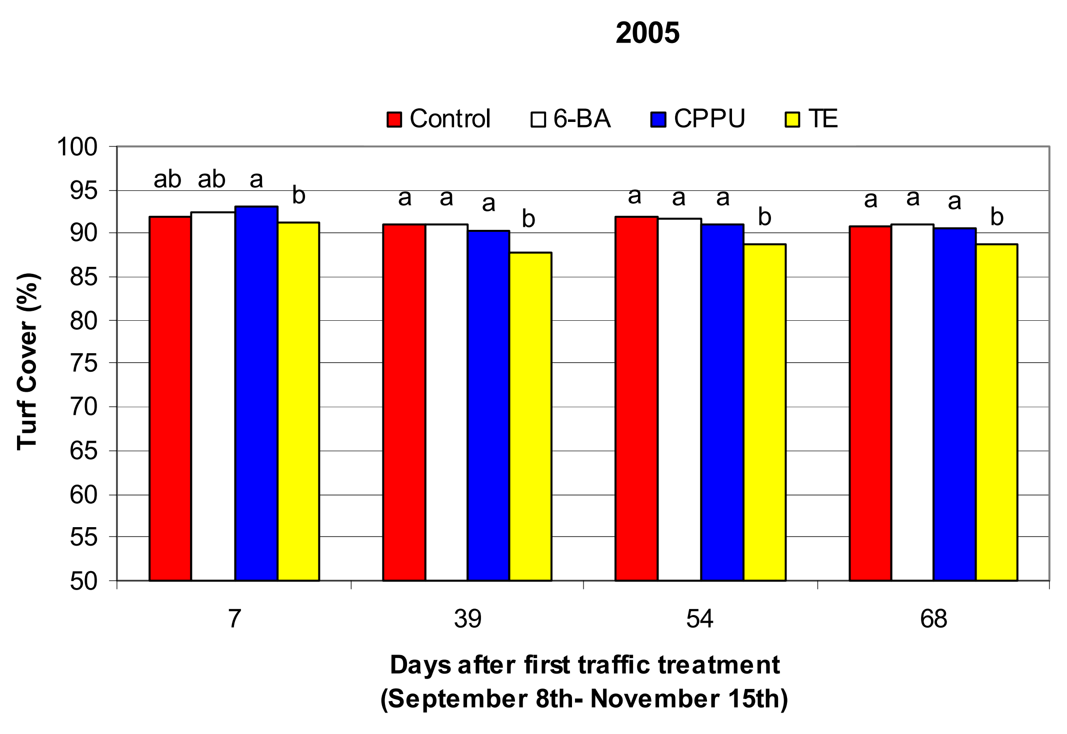
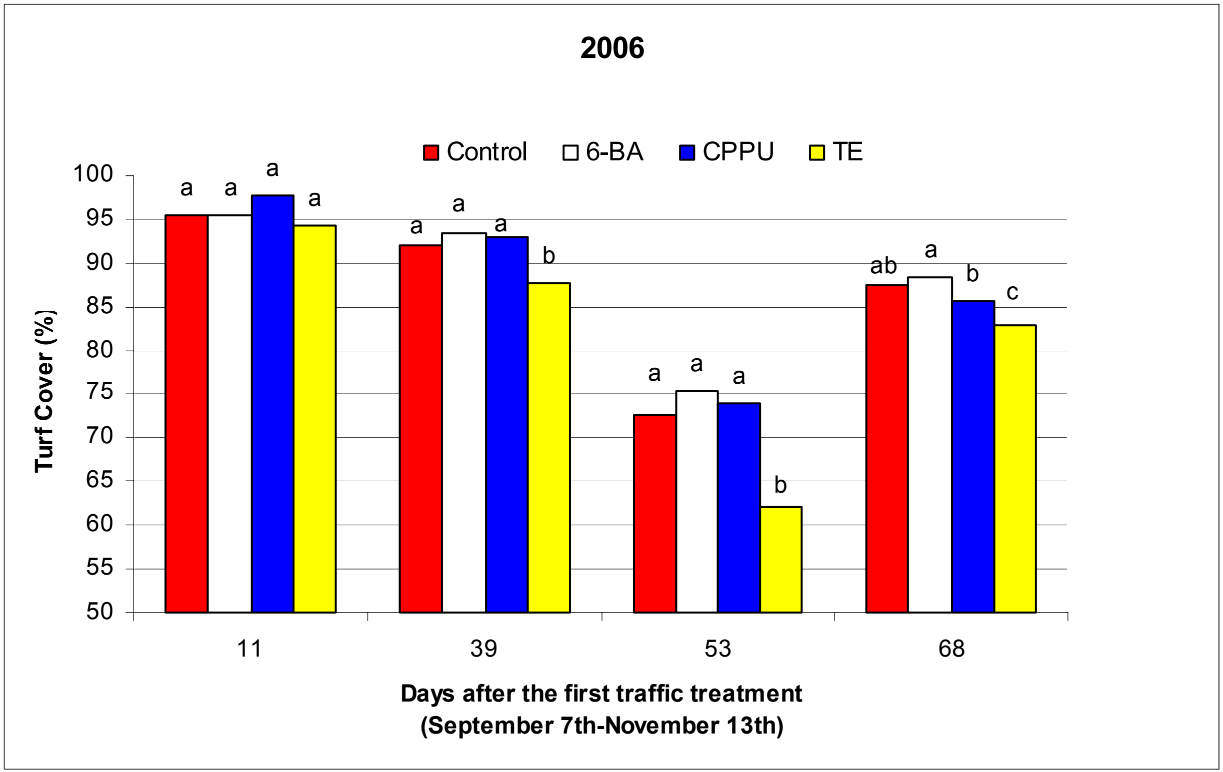


Figure 10. Percent turf cover (2006) after traffic and plant growth regulator treatments averaged over cultivars.



Turf Color

Color ratings were significantly different in 2005 and 2006 (Pr > F of <.0001). Therefore main effects and interactions are presented on a yearly basis (Table 12 and Table 13). The ANOVA showed that there was a high probability of interaction between cultivar x traffic and traffic x PGR. Their interactive effects will be the focus below.

In 2005, Patriot was lighter green than Tifsport after the first traffic treatment (Table 14). The same was seen in 2006. However, Patriot also was seen to have a lighter green color in the no-traffic treatments. Tifsport's color in both years after the third traffic treatment was not significantly different between traffic and no-traffic plots. Patriot, however, showed a difference between traffic treatments. In 2005, after the fourth and fifth traffic treatments, it appears that Patriot was significantly darker green than Tifsport in both traffic and no-traffic plots. In 2006, Patriot and Tifsport no-traffic plots were on average the same color. Patriot traffic plots, on the other-hand, were significantly lighter green than Tifsport.

NTEP reports (Tables 1 and 2) that Patriot and Tifsport have relatively equal turf colors for November, with the exception of the Blacksburg growing location, where Patriot was rated with a significantly lower color rating, probably due to Patriot's ability to go into dormancy earlier than Tifsport. The difference observed between Patriot and Tifsport turf color can be attributed to perennial ryegrass population and mowing frequency. As temperatures cooled in the fall, bermudagrass became dormant and perennial ryegrass became the dominant growing species. This was evident in both years (Figures 11 and 12) by the increase in turf color as the climate transitioned to cooler temperatures that were more conducive to perennial ryegrass growth. In both years, trafficked Patriot plots had a lighter turf color than trafficked Tifsports plots. Patriot's high tiller density and aggressive growth made it difficult to establish perennial ryegrass seed. Because of its aggressive growth, Patriot was more susceptible to scalping, which caused a decrease in turf color and quality. Once Patriot slowed in growth, perennial ryegrass became better established, resulting in a darker green canopy. The difference between 2005 and 2006 color was most likely the result of the reduced ryegrass overseeding amounts applied to the plots in 2006 (Table 13).

All plots showed a general increase in turf color as perennial ryegrass became the active growing species (Figures 13 and 14). In both years, CPPU and 6-BA did not have a consistent effect on turf color. Previous research done by White and Schmidt (1990) revealed bermudagrass treated with BA did not affect fall turf color. In addition, Nakamae and Nakamura (1982), showed Manila grass [*Zoysia matrella* (L.) Merr. var. *matrella*] treated with 6-BA did not increase leaf chlorophyll content. In 2005, after traffic one and traffic two events, plots treated with TE showed a lighter green color compared to the other PGRs and control. It appears that TE applied at 0.097 kg a.i.•ha^{-1} caused the plots to become chlorotic. Fagerness et al. (2002) reported Tifway bermudagrass treated with TE at a rate of 0.11 kg a.i.•ha^{-1}, resulted in chlorotic turf. It was not until perennial ryegrass became the dominant species, after traffic treatments three and four, that there was a significant increase in turf color with plots treated with TE. The explanation is that perennial ryegrass can tolerate TE applications at higher concentrations than hybrid bermudagrass.

Table 12. Turf color Pr > F values for 2005.

	2005 Turf color Pr > F values				
	Traffic Treatments				
Source	Traffic 1§	Traffic 2	Traffic 3	Traffic 4	Traffic 5
Cultivar	0.0765†	NA	0.8719	<0.0001	<0.0001
Traffic	0.0035	NA	0.1536	0.0057	0.0157
PGR	0.7034	NA	0.8189	0.0004	<0.0001
Traffic X PGR	0.8604	NA	0.1458	0.0095	0.3673
Cultivar X PGR	0.2133	NA	0.8691	0.7121	0.2151
Cultivar X Traffic	0.0081	NA	0.1536	0.8439	0.6726
Cultivar X Traffic X PGR	0.1966	NA	0.9611	0.4125	0.8226

† Significant main effects and interactions were determined with the Anova LSD test at an α level of 0.05.

§ Traffic 1 applied from September 8th – September 17th ; Traffic 2 (NA); Traffic 3 applied from September 17th – October 17th; Traffic 4 applied from October 17th – November 1st; Traffic 5 applied from November 1st- November 15th

Table 13. Turf color Pr > F values for 2006.

	2006 Turf color Pr > F values				
	Traffic Treatments				
Source	Traffic 1§	Traffic 2	Traffic 3	Traffic 4	Traffic 5
Cultivar	<0.0001†	<0.0001	<0.0001	0.8471	<0.0001
Traffic	0.2582	0.4634	<0.0001	<0.0001	<0.0001
PGR	0.2220	0.7768	0.0966	<0.0001	<0.0001
Traffic X PGR	0.5805	0.6510	<0.0001	0.0532	0.1505
Cultivar X PGR	0.9532	0.1614	0.1040	0.7250	0.0060
Cultivar X Traffic	0.0072	0.1479	<0.0001	0.0406	0.0013
Cultivar X Traffic X PGR	0.2509	0.3632	0.0009	0.0015	0.4438

† Significant main effects and interactions were determined with the Anova LSD test at an α level of 0.05.

§ Traffic 1 applied from September 7th – September 18th ; Traffic 2 applied from September 18th – October 2nd; Traffic 3 applied from October 2nd – October 16th; Traffic 4 applied from October 16th – October 31st; Traffic 5 applied from October 31st - November 13th

Table 14. Fall turf color in 2005 and 2006 trials for overseeded Patriot and Tifsport bermudagrasses expressed as the interaction between cultivar and traffic treatments averaged over plant growth regulator treatments.

	2005		2006	
	Traffic	No-Traffic	Traffic	No-Traffic
Cultivar	Traffic 1 †			
Patriot	6.0b¥	6.6a	5.7c§	5.9b
Tifsport	6.4a	6.5a	6.1a	6.0ab
	Traffic 2			
Patriot	-	-	6.0b	5.8b
Tifsport	-	-	6.3a	6.4a
	Traffic 3			
Patriot	6.7b	7.0a	6.2b	7.3a
Tifsport	6.8ab	6.8ab	6.4b	6.3b
	Traffic 4			
Patriot	8.2a	8.5a	6.5c	7.2a
Tifsport	7.7b	7.9b	6.7bc	7.0ab
	Traffic 5			
Patriot	8.4ab	8.7a	6.2c	7.4a
Tifsport	8.1c	8.3bc	7.0b	7.7a

† Means followed by the same letter are not significantly different at the 0.05 probability level. Mean separations are determined on a per traffic basis.

¥ 2005 Traffic 1 applied from September 8th – September 17th ; Traffic 2 (NA); Traffic 3 applied from September 17th – October 17th; Traffic 4 applied from October 17th – November 1st; Traffic 5 applied from November 1st- November 15th

§ 2006 Traffic 1 applied from September 7th – September 18th; Traffic 2 applied from September 18th – October 2nd; Traffic 3 applied from October 2nd – October 16th; Traffic 4 applied from October 16th – October 31st; Traffic 5 applied from October 31st - November 13th

Figure 11. Fall turf color ratings (2005) for overseeded Patriot and Tifsport bermudagrasses averaged over plant growth regulators. Means followed by the same letter are not significantly different at the 0.05 probability level. Mean separations are determined on a per traffic basis.

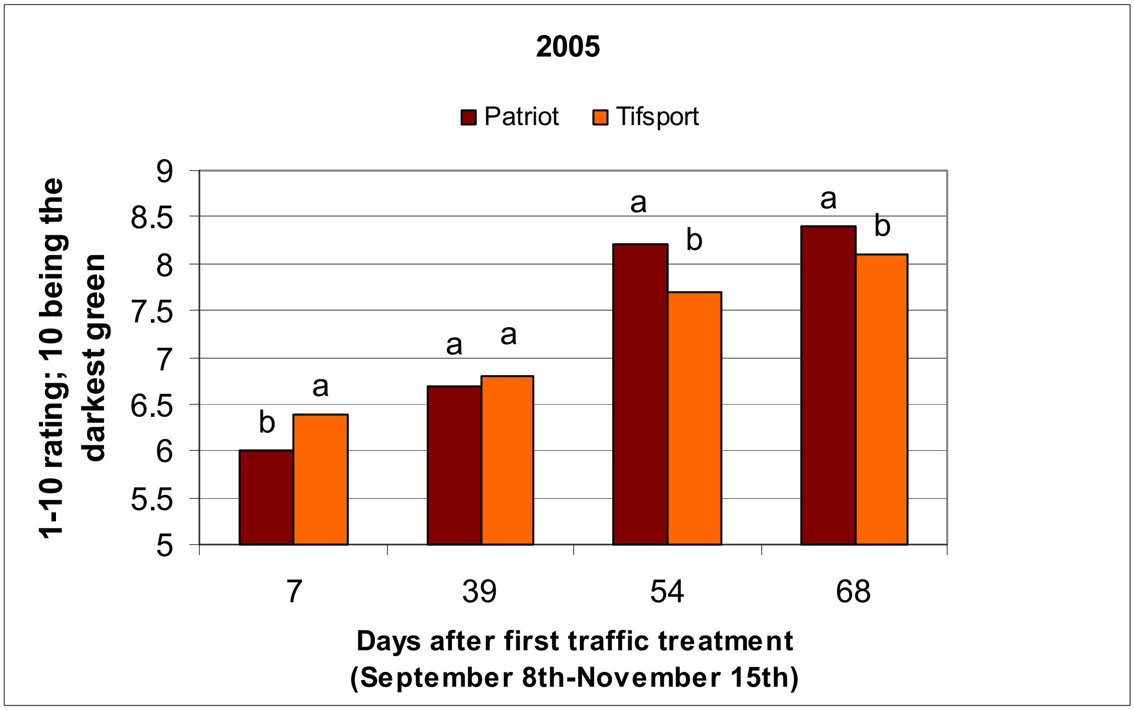


Figure 12. Fall turf color ratings (2006) for overseeded Patriot and Tifsport bermudagrasses after traffic treatments averaged over plant growth regulators. Means followed by the same letter are not significantly different at the 0.05 probability level. Mean separations are determined on a per traffic basis.

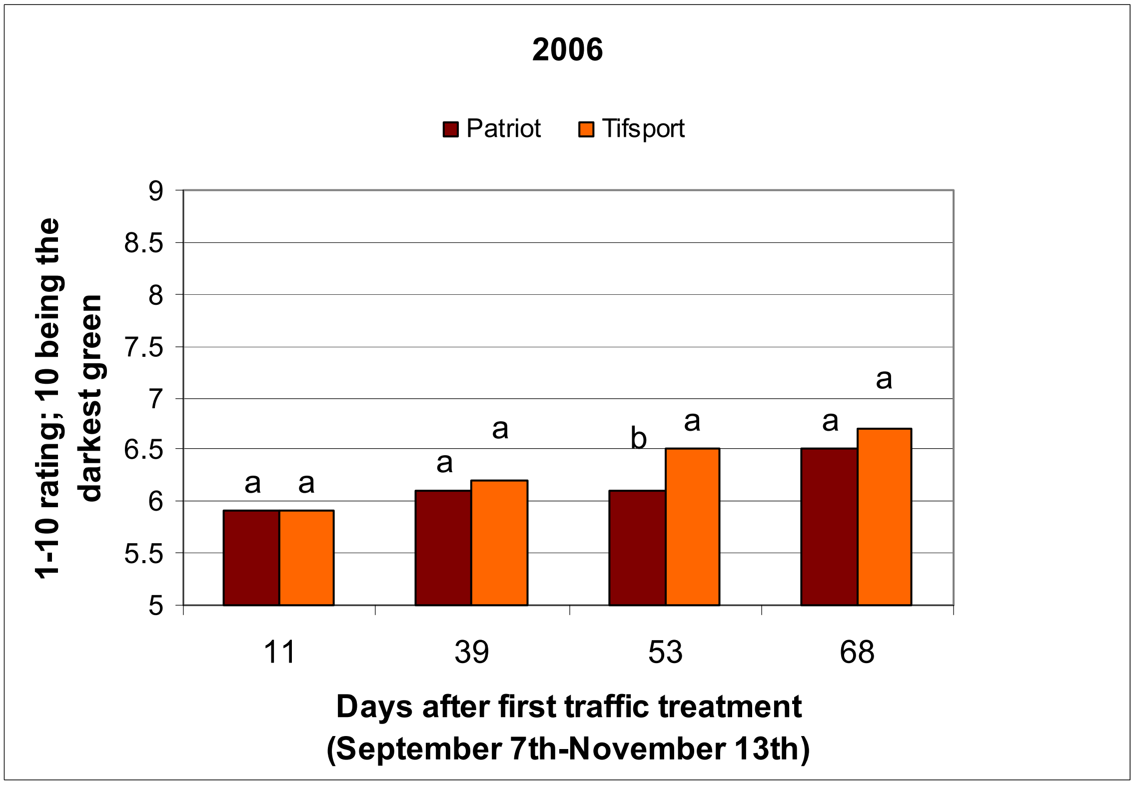


Table 15. Turf color in 2005 and 2006 trials expressed as the interaction between plant growth regulators and traffic treatments averaged over cultivars.

	2005				2006			
	Control	6-BAΨ	CPPU	TE	Control	6-BA	CPPU	TE
Treatment					Traffic 1 †			
No-Traffic	6.5ab¥	6.4ab	6.6a	6.5ab	5.9a§	6.0a	5.9a	6.0a
Traffic	6.3ab	6.2ab	6.3ab	6.1b	5.8a	6.0a	5.9a	5.9a
					Traffic 2			
No-Traffic	-	-	-	-	6.1a	6.1a	6.2a	5.9a
Traffic	-	-	-	-	6.1a	6.1a	6.2a	6.2a
					Traffic 3			
No-Traffic	6.8abc	7.0ab	6.8abc	7.1a	6.9a	6.9a	6.9a	6.6abc
Traffic	6.9abc	6.8abc	6.8abc	6.6c	6.0d	6.3bcd	6.1cd	6.8ab
					Traffic 4			
No-Traffic	8.3b	7.9bc	7.9bc	8.7a	6.9bc	7.1b	6.8bc	7.6a
Traffic	8.1bc	7.8c	8.0bc	8.0bc	6.3d	6.8bc	6.4cd	6.8bcd
					Traffic 5			
No-Traffic	8.3bc	8.4bc	8.3bc	8.9a	7.7ab	7.3bc	7.4bc	7.9a
Traffic	8.1c	8.2c	8.3bc	8.6b	6.5d	6.5d	6.3d	7.0c

† Means followed by the same letter are not significantly different at the 0.05 probability level. Mean separations are determined on a per traffic basis.

Ψ 6-BA (6-Benzyladenine), CPPU (2-Chloro-4-pyridyl-phenylurea), TE (Trinexapac-ethyl)

¥ 2005 Traffic 1 applied from September 8th – September 17th ; Traffic 2 (NA); Traffic 3 applied from September 17th – October 17th; Traffic 4 applied from October 17th – November 1st; Traffic 5 applied from November 1st- November 15th

§ 2006 Traffic 1 applied from September 7th – September 18th; Traffic 2 applied from September 18th – October 2nd; Traffic 3 applied from October 2nd – October 16th; Traffic 4 applied from October 16th – October 31st; Traffic 5 applied from October 31st - November 13th

Figure 13. Fall turf color (2005) after traffic and plant growth regulator treatments averaged over cultivars. Means followed by the same letter are not significantly different at the 0.05 probability level. Mean separations are determined on a per traffic basis.

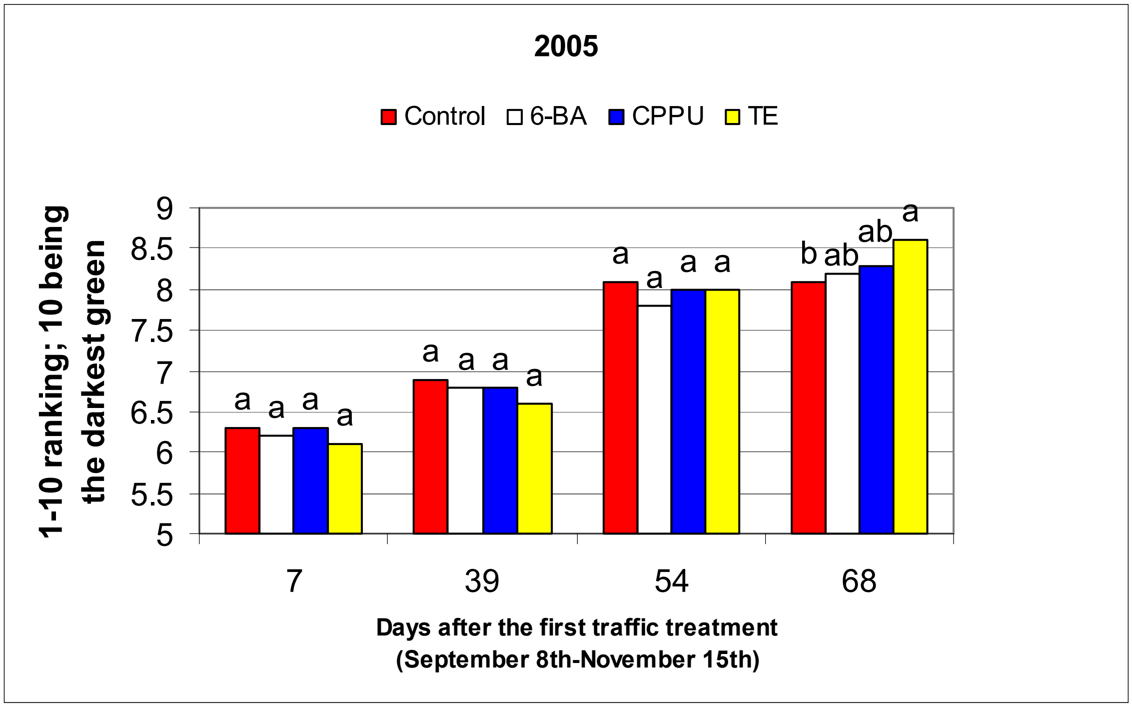
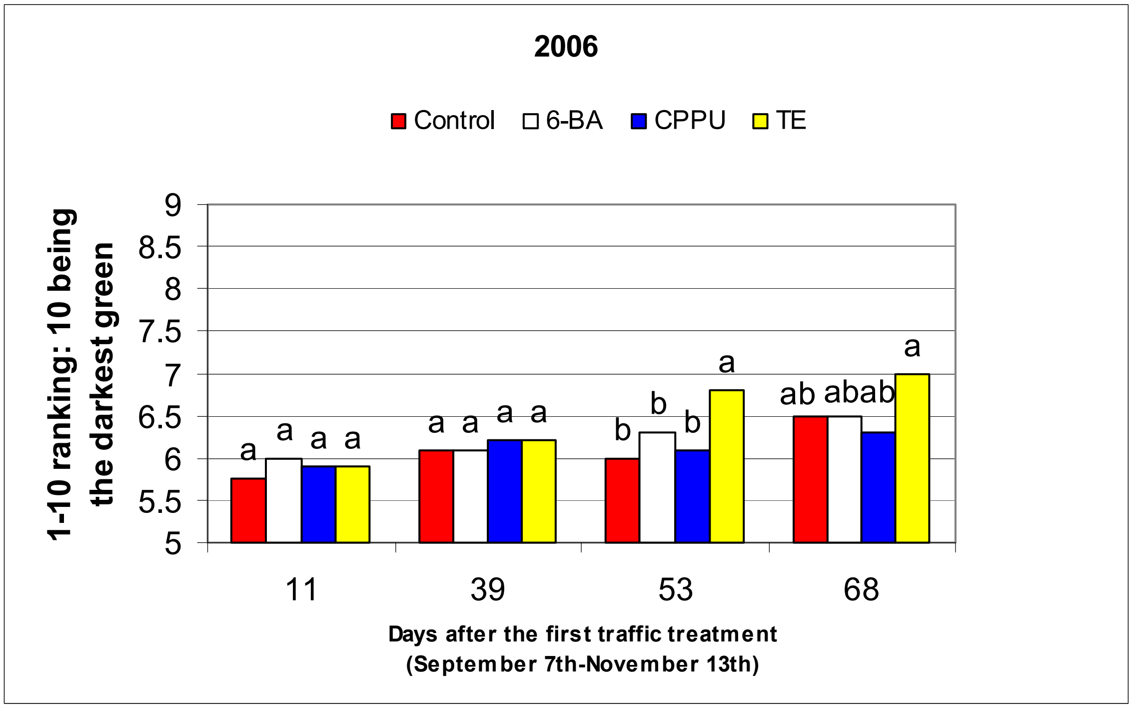


Figure 14. Fall turf color (2006) after traffic and plant growth regulator treatments averaged over cultivars. Means followed by the same letter are not significantly different at the 0.05 probability level. Mean separations are determined on a per traffic basis.



Quality

The ANOVA test for both years showed a high probability of interaction between cultivar x PGR and cultivar x traffic (Table 16 and Table 17). In 2005, quality ratings after the first traffic treatment showed Patriot to have a higher visual quality than Tifsport (Table 18). This trend continued throughout traffic treatments. Patriot plots (trafficked and non-trafficked) consistently received higher quality ratings than Tifsport. In 2006, Patriot and Tifsport non-trafficked plots were on average rated equal. However, as seen in 2005, Tifsport trafficked plots consistently received the lowest quality rating. The difference in quality ratings between the two cultivars was apparently due to Patriot's high tiller density and rapid shoot growth.

CPPU and 6-BA had little effect on turf quality (Table 19). It has been previously reported that BA applied to bermudagrass had little effect on turf color and quality (White et al., 1990). Trinexapac-ethyl, after the fourth traffic treatments for both years, had significantly lower quality ratings than the control and other PGRs. Previous research done with Tifway bermudagrass showed that autumn applications of TE, applied at a rate of 0.11 kg a.i.•ha^{-1}, led to decreased turfgrass density and quality (Fagerness et al., 2002). Reduced quality was due to the suppression of growth and mild leaf chlorosis. Mild chlorosis due to TE was evident in the plots. As perennial ryegrass became the dominant growing species, the effects of TE on turf quality became beneficial as perennial ryegrass responded to the PGR treatment with an increase in turf color.

Table 16. Turf quality Pr > F values for 2005.

	2005 Turf quality Pr > F values				
	Traffic Treatments				
Source	Traffic 1§	Traffic 2	Traffic 3	Traffic 4	Traffic 5
Cultivar	0.0002†	NA	0.0021	<0.0001	<0.0001
Traffic	<0.0001	NA	<0.0001	<0.0001	<0.0001
PGR	0.7745	NA	0.1719	0.0176	0.0361
Traffic X PGR	0.7745	NA	0.6691	0.0831	0.1351
Cultivar X PGR	0.7745	NA	0.0050	0.3752	0.0998
Cultivar X Traffic	0.3958	NA	0.2710	0.0005	0.0782
Cultivar X Traffic X PGR	0.7745	NA	0.6021	0.0125	0.0080

§ Significant main effects and interactions were determined with the Anova LSD test at an α level of 0.05

† Traffic 1 applied from September 8th – September 17th ; Traffic 2 (NA); Traffic 3 applied from September 17th – October 17th; Traffic 4 applied from October 17th – November 1st; Traffic 5 applied from November 1st- November 15th

Table 17. Turf quality Pr > F values for 2006.

	2006 Turf quality Pr > F values				
	Traffic Treatments				
Source	Traffic 1†	Traffic 2	Traffic 3	Traffic 4	Traffic 5
Cultivar	0.0464§	0.0041	<0.0001	<0.0001	<0.0001
Traffic	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
PGR	0.8012	0.0001	<0.0001	0.0053	0.1384
Traffic X PGR	0.4771	0.6403	0.0266	0.1469	0.1384
Cultivar X PGR	0.1501	0.2513	0.1583	0.1586	0.4414
Cultivar X Traffic	<0.0001	<0.0001	<0.0001	<0.0001	0.0141
Cultivar X Traffic X PGR	0.2596	0.3295	0.0031	0.5978	0.1384

§ Significant main effects and interactions were determined with the Anova LSD test at an α level of 0.05.

†Traffic 1 applied from September 7th – September 18th ; Traffic 2 applied from September 18th – October 2nd; Traffic 3 applied from October 2nd – October 16th; Traffic 4 applied from October 16th – October 31st; Traffic 5 applied from October 31st - November 13th

Table 18. Visual quality ratings in 2005 and 2006 trials expressed as the interaction between traffic and cultivar averaged over plant growth regulators.

	2005		2006	
	Traffic	No-Traffic	Traffic	No-Traffic
Cultivar		Traffic 1 †		
Patriot	6.8c¥	7.3a	7.1b§	7.1b
Tifsport	6.4d	7.0b	5.8c	7.8a
		Traffic 2		
Patriot	-	-	5.9a	6.2a
Tifsport	-	-	5.0b	7.6a
		Traffic 3		
Patriot	6.7b	7.3a	5.7b	7.2a
Tifsport	6.4c	7.2a	4.6c	7.4a
		Traffic 4		
Patriot	7.1c	8.6a	6.5b	7.4a
Tifsport	5.7d	7.8b	5.5c	7.4a
		Traffic 5		
Patriot	6.8c	8.4a	6.7b	8.2a
Tifsport	6.0d	7.9b	5.8c	7.8a

† Means followed by the same letter are not significantly different at the 0.05 probability level. Mean separations are determined on a per traffic basis.

¥ 2005 Traffic 1 applied from September 8th – September 17th ; Traffic 2 (NA); Traffic 3 applied from September 17th – October 17th; Traffic 4 applied from October 17th – November 1st; Traffic 5 applied from November 1st- November 15th

§ 2006 Traffic 1 applied from September 7th – September 18th ; Traffic 2 applied from September 18th – October 2nd; Traffic 3 applied from October 2nd – October 16th; Traffic 4 applied from October 16th – October 31st; Traffic 5 applied from October 31st - November 13th

Table 19. Fall quality ratings in 2005 and 2006 trials for overseeded Patriot and Tifsport bermudagrasses expressed as the interaction between cultivar and plant growth regulator treatments averaged over traffic treatments.

	2005				2006			
	Control	6-BAΨ	CPPU	TE	Control	6-BA	CPPU	TE
Cultivar				Traffic 1 †				
Patriot	7.0a¥	7.0a	7.0a	7.0a	7.1a§	6.9a	7.1a	7.3a
Tifsport	6.6a	6.8a	6.6a	6.7a	6.8a	6.8a	7.0a	6.8a
				Traffic 2				
Patriot	-	-	-	-	6.3a	6.2a	6.2a	5.5a
Tifsport	-	-	-	-	6.3a	6.4a	6.5a	6.1a
				Traffic 3				
Patriot	7.2a	6.9ab	7.0ab	6.9ab	6.6a	6.4a	6.5a	6.4a
Tifsport	6.7b	7.0ab	6.8ab	6.7b	6.2a	6.1a	6.2a	5.6a
				Traffic 4				
Patriot	7.9a	7.8ab	7.8ab	7.8ab	6.9ab	7.3a	6.9ab	6.7ab
Tifsport	7.1abc	6.8bc	6.6c	6.6c	6.8ab	6.5ab	6.6ab	6.1b
				Traffic 5				
Patriot	7.6a	7.9a	7.3a	7.8a	7.6a	7.6a	7.2a	7.2a
Tifsport	7.3a	7.0a	6.8a	6.8a	6.9a	6.8a	6.7a	6.8a

† Means followed by the same letter are not significantly different at the 0.05 probability level. Mean separations are determined on a per traffic basis.

Ψ 6-BA (6-Benzyladenine), CPPU (2-Chloro-4-pyridyl-phenylurea), TE (Trinexapac-ethyl)

¥ 2005 Traffic 1 applied from September 8th – September 17th ; Traffic 2 (NA); Traffic 3 applied from September 17th – October 17th; Traffic 4 applied from October 17th – November 1st; Traffic 5 applied from November 1st- November 15th

§ 2006 Traffic 1 applied from September 7th – September 18th ; Traffic 2 applied from September 18th – October 2nd; Traffic 3 applied from October 2nd – October 16th; Traffic 4 applied from October 16th – October 31st; Traffic 5 applied from October 31st - November 13th

Sugars

Starch and total nonstructural carbohydrate (TNC) concentrations were significantly different in 2005 and 2006 (both with a Pr>F of <0.0001). Therefore main effects and interactions are presented on a yearly basis. The ANOVA indicated that Patriot and Tifsport had significantly different TNC concentrations (Tables 20 and 21). In 2006, there are indications that stolon sugar contents were affected by PGRs. It is evident that significant differences exist between cultivar x PGR and traffic x PGR interactions.

During fall acclimation, bermudagrasses store sugars to serve as a reserve for re-growth as temperatures warm in the spring The amount of sugars stored has been directly linked to the winter survival of bermudagrass – the more there are the more likely it will survive the winter. A study using two bermudagrass cultivars, Riveria (cold tolerant) and Princess-77 (cold sensitive), showed the more cold tolerant cultivar to have 86% more carbohydrates than the cold sensitive cultivar (Zhang et al., 2006). Bermudagrasses store most of their carbohydrate reserves in stolon tissues as starch (Figure 15). Soluble sugars, such as glucose, fructose, and sucrose constitute the remaining "energy-storing" sugars. Starch and the soluble sugars collectively constitute TNC. Out of the soluble sugars, sucrose predominantly exists at the highest concentrations (Tables 22 and 23); it is typically the sugar that is transported through the phloem for plant organ uptake. Starch is mobilized to sucrose when the source of photoassimalates is too low to support plant (sink) growth. Up to 50% of assimilated carbon can be stored either as sucrose or as starch (Gifford et al., 1984).

In both years, Patriot had consistently higher winter TNC concentrations than Tifsport (Figure 15 and Table 21) A comparison between years showed TNC concentrations for 2006 to be much higher than 2005, regardless of species or traffic treatment. The difference between years can be linked to the time difference in which samples were taken. In 2005, samples were taken about two months later (February 7, 2006) than the 2006 samples (December 12, 2006). Stolon tissue has a degree of maintenance respiration that occurs during the winter months. Since no photo-assimilates are being made during the dormancy period, stored sugars are being used for respiration.

In both years, non-trafficked TE plots were shown to have significantly higher TNC concentrations compared to the other PGRs and control (Tables 22 and 23). Much of the difference between TNC concentrations was the result of TE increasing starch concentrations.

Previous research done with Tifway bermudagrass showed TE applied at 0.05, 0.10, and 0.15 kg ha^{-1} increased root tissue TNC contents ≥ 38% as compared to the untreated control (Waltz et al., 2005). It was also shown that shoot tissue TNC content was periodically increased by 32% compared to nontreated (Waltz et al., 2005). Furthermore, shaded Diamond zoysiagrass (*Zoysia matrella*), which received monthly and bi-monthly applications of TE at a rate of 0.096 kg a.i. ha^{-1} and 0.192 kg a.i. ha^{-1}, respectively, resulted in a 40 to 38% higher TNC compared to the control (Qian et al., 1999).

Tifsport, for both years, showed a reduction in sugar content as a result of CPPU applications (Table 22). CPPU had no consistent effect on Patriot sugar concentrations. In 2005, Tifsport plots treated with 6-BA showed increased sucrose and starch concentrations compared to the control and other PGRs. Not much interaction was seen for Patriot in 2005. However, in 2006, Patriot plots treated with 6-BA, relative to the other treatments, also showed an increase in soluble sugars. CPPU applied at the rate of 0.025 kg a.i.• ha^{-1} resulted in lower TNC concentrations, indicating that it might have been detrimental to bermudagrass growth. White and Schmidt (1990) found bermudagrass treated with BA did not cause an increase in TNC. Our results support this; neither 6-BA nor CPPU caused a consistent effect on TNC and therefore is thought to have no effect to bermudagrass cold tolerance.

Table 20. Total non-structural carbohydrates Pr > F values for 2005.

	2005 Turf sugar Pr > F values				
	Non-structural carbohydrates				
Source	Glucose	Fructose	Sucrose	Starch	TNC
Cultivar	0.0104§	0.1220	0.0222	<0.0001	<0.0001
Traffic	0.3417	0.0589	0.1681	0.3849	0.1543
PGR	0.0213	0.1944	0.1209	0.6229	0.2417
Traffic X PGR	0.0012	0.0143	0.0332	0.1824	0.0167
Cultivar X PGR	0.0110	0.2231	0.0020	0.0995	0.0273
Cultivar X Traffic	0.3810	0.2715	0.3496	0.9908	0.5832
Cultivar X Traffic X PGR	0.2986	0.8707	0.7673	0.3567	0.4398

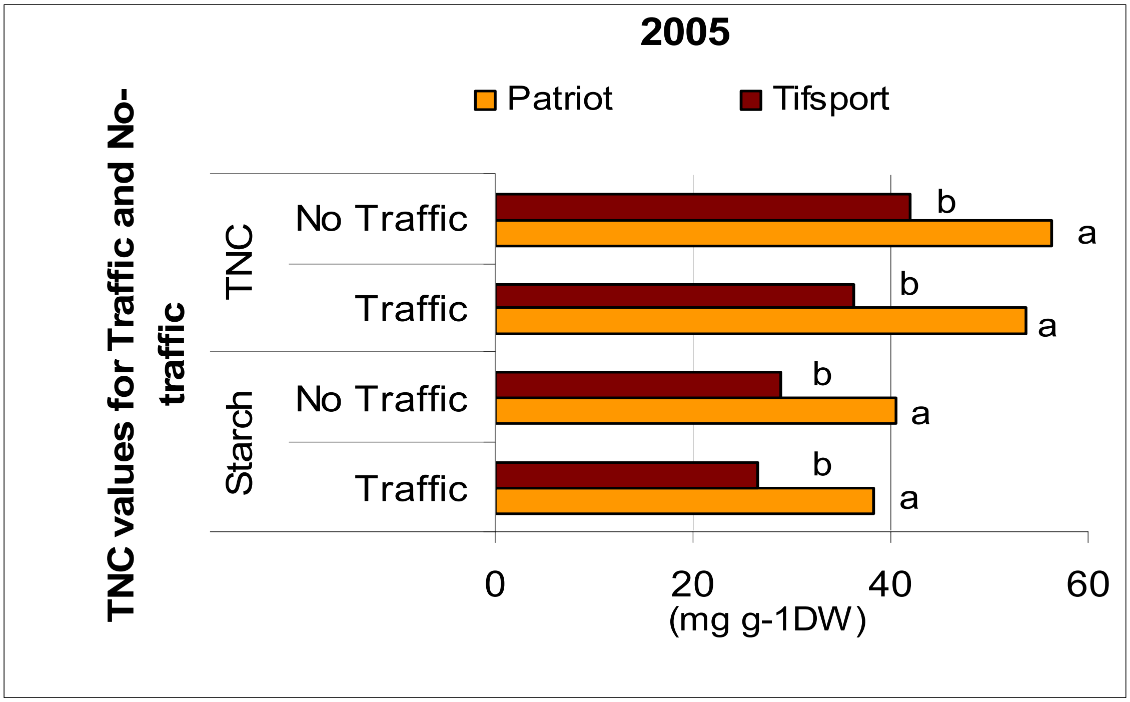
§ Significant main effects and interactions were determined with the Anova LSD test at an α level of 0.05.

Table 21. Total non-structural carbohydrate Pr > F values for 2006.

	2006 Turf sugar Pr > F values				
	Non-structural carbohydrates				
Source	Glucose	Fructose	Sucrose	Starch	TNC
Cultivar	0.0073§	0.0085	0.0003	0.0042	0.0012
Traffic	0.6072	0.4503	0.9718	0.3546	0.4058
PGR	0.1369	0.5390	0.2594	0.0682	0.0884
Traffic X PGR	0.6783	0.2036	0.0735	0.3283	0.2461
Cultivar X PGR	0.4042	0.0636	0.2015	0.6766	0.6413
Cultivar X Traffic	0.5930	0.2895	0.7818	0.3779	0.3953
Cultivar X Traffic X PGR	0.3687	0.3611	0.9402	0.0297	0.0517

§ Significant main effects and interactions were determined with the Anova LSD test at an α level of 0.05.

Figure 15. Total nonstructural carbohydrate (TNC) and starch content in stolons of Patriot and Tifsport bermudagrasses after traffic treatments averaged over plant growth regulators in 2005 and 2006.



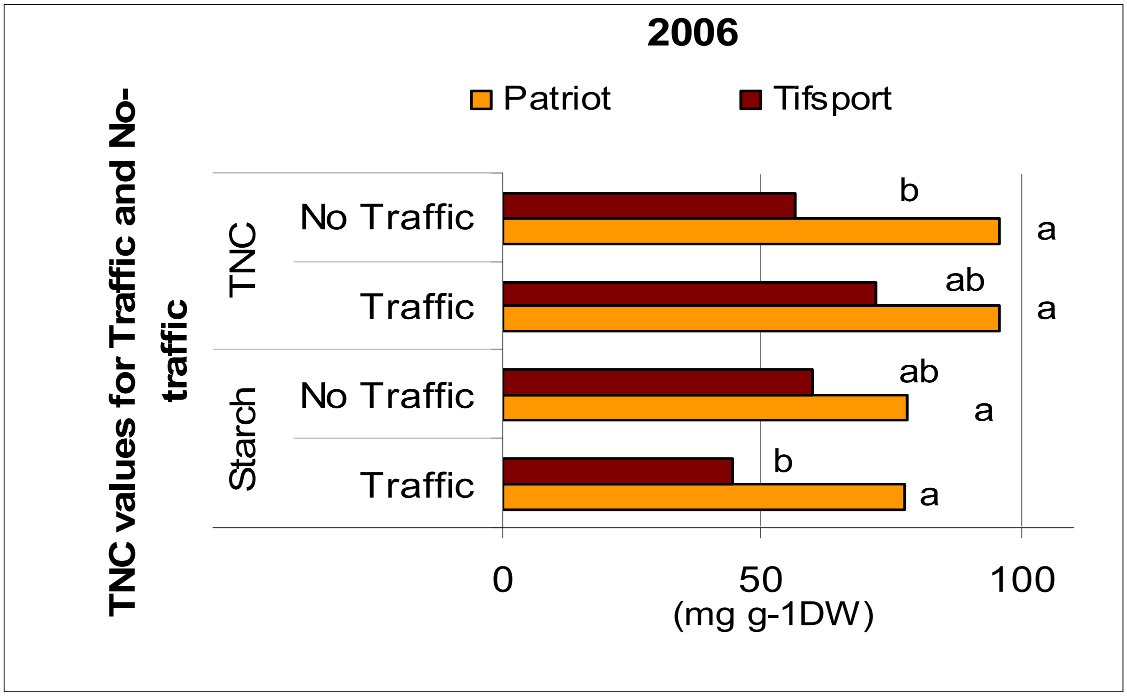


Table 22. Nonstructural carbohydrates content in stolons of Patriot and Tifsport bermudgrasses expressed as the interaction between cultivar and plant growth regulator averaged over traffic treatments.

	2005				2006			
	Control	6-BAΨ	CPPU	TE	Control	6-BA	CPPU	TE
Cultivar	Glucose (mg g^{-1}DW) †							
Patriot	3.25a	2.97ab	3.39a	3.12ab	3.65ab	4.57a	2.99ab	3.47ab
Tifsport	3.50a	2.41bc	2.03c	2.98ab	2.19b	2.85b	2.17b	3.37ab
	Fructose (mg g^{-1}DW)							
Patriot	2.00a	1.93ab	1.72ab	1.68ab	2.87ab	3.91a	2.52b	2.76ab
Tifsport	1.86ab	1.24ab	1.08b	1.97a	2.41b	1.89b	2.18b	2.62ab
	Sucrose (mg g^{-1}DW)							
Patriot	9.2abc	10.19abc	14.33a	7.62bcd	10.87abc	14.13a	11.44ab	9.27abcd
Tifsport	7.25cd	12.73ab	2.36d	6.79cd	5.98cd	8.12bcd	5.94d	8.99bcd
	Starch (mg g^{-1}DW)							
Patriot	36.20ab	39.15a	39.01a	43.73a	88.21ab	74.62abc	54.61bc	92.99a
Tifsport	28.7bc	33.82ab	26.59bc	21.56c	50.44c	42.88c	44.75c	69.21abc
	TNC (mg g^{-1}DW)							
Patriot	50.65ab	54.26ab	58.45a	56.15a	105.6a	97.24ab	71.55abc	108.49a
Tifsport	41.38bc	50.19ab	32.06c	33.31c	61.02bc	55.75c	55.04c	84.2abc

† Means followed by the same letter are not significantly different at the 0.05 probability level.

Ψ 6-BA (6-Benzyladenine), CPPU (2-Chloro-4-pyridyl-phenylurea), TE (Trinexapac-ethyl)

Table 23. Bermudagrass stolon sugar concentrations for 2005 and 2006 trials expressed as the interaction between traffic and plant growth regulators averaged over cultivars.

	2005				2006			
	Control	6-BAΨ	CPPU	TE	Control	6-BA	CPPU	TE
Treatment	Glucose (mg g^{-1}DW) †							
Traffic	3.4a	2.4b	3.0ab	3.3a	2.1bc	2.0bc	2.5abc	3.9a
No-Traffic	2.5ab	2.9ab	3.1ab	2.9ab	2.3abc	3.7ab	1.8c	2.8abc
	Fructose (mg g^{-1}DW)							
Traffic	1.5a	1.4a	1.5a	1.6a	2.8ab	1.35c	2.7ab	3.2a
No-Traffic	1.6a	1.9a	1.9a	2.1a	2.0abc	2.4abc	1.6bc	2.0abc
	Sucrose (mg g^{-1}DW)							
Traffic	7.2ab	9.1ab	8.5ab	6.7b	5.9a	6.6a	7.6a	8.3a
No-Traffic	5.7b	11.0ab	13.0a	9.1ab	6.1a	9.7a	4.3a	9.65a
	Starch (mg g^{-1}DW)							
Traffic	30.0ab	37.0ab	28.6b	34.5ab	52.1b	29.8b	54.9b	39.7b
No-Traffic	30.1ab	35.3ab	31.4ab	42.1a	48.8b	56.0b	34.6b	98.8a
	TNC (mg g^{-1}DW)							
Traffic	42.0ab	49.9ab	41.6ab	46.3ab	62.9b	39.7b	67.7b	55.2b
No-Traffic	39.9b	51.0ab	49.4ab	56.2a	59.1b	71.8b	42.4b	113.2a

† Means followed by the same letter are not significantly different at the 0.05 probability level.

Ψ 6-BA (6-Benzyladenine), CPPU (2-Chloro-4-pyridyl-phenylurea), TE (Trinexapac-ethyl)

Table 24. Bermudagrass stolon sugar concentrations for 2005 and 2006 trials expressed as the interaction between traffic and cultivar averaged over plant growth regulator treatments.

	2005		2006	
	Traffic Treatment			
	Traffic	No-Traffic	Traffic	No-Traffic
Cultivar	Glucose (mg g^{-1}DW) †			
Patriot	3.3a¥	3.0a	3.9a§	3.5ab
Tifsport	2.7a	2.7a	2.6b	2.6b
	Fructose (mg g^{-1}DW)			
Patriot	1.8ab	1.9a	3.0a	3.1a
Tifsport	1.3b	1.8ab	2.5ab	2.0b
	Sucrose (mg g^{-1}DW)			
Patriot	10.0ab	10.6a	11.6a	11.3a
Tifsport	5.8b	8.8ab	7.1b	7.4b
	Starch (mg g^{-1}DW)			
Patriot	38.4a	40.6a	77.4a	77.8a
Tifsport	26.6b	28.8b	44.1b	59.5ab
	TNC (mg g^{-1}DW)			
Patriot	53.6a	56.2a	95.8a	95.6a
Tifsport	36.3b	42.1b	71.6ab	56.4b

† Means followed by the same letter are not significantly different at the 0.05 probability level.

¥ 2005 samples were taken February 7, 2006

§ 2006 samples were taken December 12 , 2006

Stolon Viability

The ANOVA showed Tifsport and Patriot to have significantly different levels of electrolyte leakage (Tables 25 and 26). In 2005, there was a high probability of significance for the traffic x PGR and cultivar x traffic interactions. In 2006, there are some indications that PGRs had a significant effect on stolon viability.

There was little interaction between traffic and no-traffic effects on bermudagrass stolon viability (Table 27). The main effect seen was the difference between cultivars. Patriot consistently had lower electrolyte leakage than Tifsport (Figure 16 and Table 28). Electrolyte leakage has been shown to provide a measure of cold tolerance. A study using the bermudagrass cultivars Riveria (cold tolerant) and Princess-77 (cold sensitive), showed electrolyte leakage to be lower in the more cold tolerant cultivar (Zhang et al., 2006). Patriot has been documented to be more cold tolerant than Tifsport (Anderson et. al, 2003). Therefore, it is not surprising that Patriot would have lower percent electrolyte leakage than Tifsport.

Plant growth regulators for the most part had little effects on stolon viability (Table 28). However, it was seen for both years that trafficked Tifsport plots treated with TE had the highest percentage of electrolyte leakage out of all treatment plots. Previous research done with Kentucky bluegrass, applied with TE at a rate of 0.23 kg a.i.• ha^{-1}, showed a significant increase in electrolyte leakage. TE is classified as being a cyclohexanedione, which is in the same family as sethoxydim {2-[1-(ethylthio)propyl]-3-hydroxy-2-cyclohexen-1-one}. Sethoxydim is a graminicide that inhibits the acetyl coenzyme, restricting the ability to make lipids. Lipids are the main constituent of cell membranes. A reduction in the amount of lipids could result in less stable membranes (Heckman et al., 2001). The decline in membrane stability could have resulted in higher percentages of electrolyte leakage for the bermudagrass treated with TE.

Table 25. Electrolyte leakage Pr > F values for 2005.

	2005 Electrolyte leakage Pr > F values
Source	EL %
Cultivar	<0.0001†
Traffic	0.3325
PGR	0.5038
Traffic X PGR	0.0012
Cultivar X PGR	0.1188
Cultivar X Traffic	0.0170
Cultivar X Traffic X PGR	0.1108

† Significant main effects and interactions were determined with the Anova LSD test at an α level of 0.05.

§ Samples were taken February 7, 2006

Table 26. Electrolyte leakage Pr > F values for 2006.

	2006 Electrolyte leakage Pr > F values
Source	EL %
Cultivar	<0.0001†
Traffic	0.6199
PGR	0.0704
Traffic X PGR	0.3755
Cultivar X PGR	0.0976
Cultivar X Traffic	0.8545
Cultivar X Traffic X PGR	0.2672

† Significant main effects and interactions determined from the Anova LSD test at an α level of 0.05

§ Samples were taken December 12, 2006

Table 27. Bermudagrass stolon electrolyte leakages for 2005 and 2006 trials expressed as the interaction between traffic and cultivar averaged over plant growth regulator treatments.

	2005		2006	
	Traffic	No-Traffic	Traffic	No-Traffic
Cultivar	Electrolyte Leakage (%) †			
Patriot	16.2b	15.8b	17.2b	15.6b
Tifsport	40.5a	39.7a	30.6a	34.4a

† Means followed by the same letter are not significantly different at the 0.05 probability level.

Figure 16. Comparison of stolon viability (2005 and 2006) for Tifsport and Patriot bermudagrasses. Means followed by the same letter are not significantly different at the 0.05 probability level.

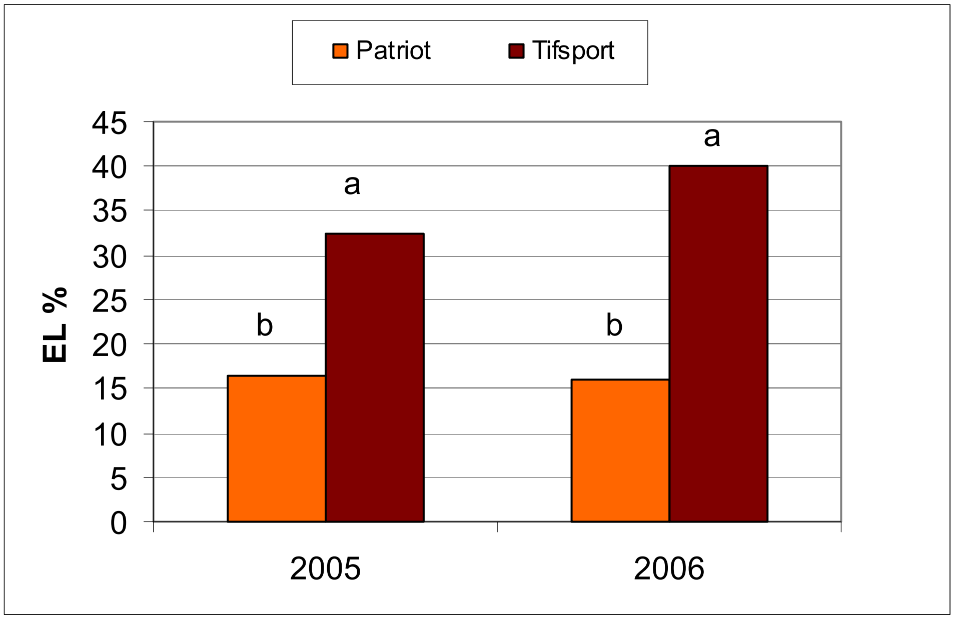


Table 28. Bermudagrass stolon electrolyte leakage for 2005 and 2006 trials expressed as the interaction between traffic, cultivar, and plant growth regulator treatments.

	2005		2006	
	Traffic	No-Traffic	Traffic	No-Traffic
Cultivar	Control †			
Patriot	18.6ef	12.5f	18.4d	16.1d
Tifsport	33.7abc	38.1ab	35.0c	39.8abc
	6-BA Ψ			
Patriot	17.8f	16.4f	16.4d	15.8d
Tifsport	33.1bc	29.7dc	41.8abc	37.2bc
	CPPU			
Patriot	18.3f	15.2f	14.5d	14.1d
Tifsport	30.4dc	29.6dc	40.9abc	36.0bc
	TE			
Patriot	14.3f	18.3f	15.5d	17.3d
Tifsport	25.0ed	40.0a	44.1ab	45.7a

† Means followed by the same letter are not significantly different at the 0.05 probability level. Mean separations are determined on a per traffic basis.

Ψ 6-BA (6-Benzyladenine), CPPU (2-Chloro-4-pyridyl-phenylurea), TE (Trinexapac-ethyl)

Conclusion

Ways to improve the aesthetics and functionality of athletic turfs are continually sought. Improved performance of hybrid bermduagrasses in the transition zone is a testament to the progress being made. Previous research has shown cytokinins or cytokinin-enhancing PGRs to increase tiller density, delay senescence and enhance photochemical efficiency—all factors that could improve the traffic tolerance and playability of overseeded bermudagrass. For these reasons, this study investigated the use of three different PGRs: 6-BA, CPPU, and TE. Our results showed no indication that 6-BA or CPPU improved turf cover. In order for turf cover to be improved, tiller density and anti-senescent properties must be increased. There was no evidence that such beneficial growth changes occurred. Furthermore, the two synthetic cytokinins showed no beneficial effects on turf color or quality. These findings are supported by previous research, but there are still questions that need to be answered regarding the effectiveness of increasing endogenous cytokinin levels. There also continues to be uncertainty as to cytokinins' mode of action. In this study, exogenous cytokinins were foliar applied under the premise that they would increase the biosynthesis of endogenous cytokinins. Future research needs to determine if soil-applied cytokinins near the root zone, an area that has been well documented as being a "hot-spot" for cytokinin biosynthesis and transport, could trigger plant responses. Also, cytokinins have a limited market in the turf industry overall and there is no direct literature that reports the proper application rate of cytokinin to bermudagrass. In order to better predict how turfgrass species will react to cytokinins, response curves specific to this species need to be developed.

Trinexapac ethyl at the level of 0.097 kg a.i.•ha^{-1}, was detrimental to fall-acclimating bermudagrass. Bermudagrass cover, quality, and color were lowered by TE applications. It was not until perennial ryegrass became established that there were beneficial effects seen with TE, in particular, increases in turf color and quality. The application rate of 0.097 kg a.i.•ha^{-1} is what is recommended as the monthly rate; however we were applying it on a two week basis. Past research indicates that this rate would indeed cause detrimental effects to fall-acclimating bermudagrass. However, even when perennial ryegrass became the dominant growing species, a decrease in turf cover was seen. It is unknown if this was caused by the initial effects of TE depleting bermudagrass cover, or if TE was causing a reduction in perennial ryegrass cover as-well. Regardless, sports turf managers should be cautious to use TE at labeled rates specifically for bermudagrass and know that previous research suggests that the application of TE can result in a reduction in turfgrass traffic tolerance.

Sequential applications of 6-BA and CPPU showed no correlation to bermudagrass cold tolerance. Trinexapac ethyl showed some indication that it caused an increase in bermudagrass TNC. However, beneficial effects were not seen in the trafficked plots. TE applied at the recommended rate, might result in improved winter survival of bermudagrass.

There was no correlation between 6-BA and CPPU with stolon viability. However, TE, showed a significantly decrease in stolon viability in Tifsport. Previous research reported that TE causes membrane degradation, also a possibility in this research. Further research on the interaction of the contrasting effects of TE on bermudagrass fitness for winter survival is warranted.

The comparison of the two bermudagrass cultivars showed Patriot to have better traffic tolerance, quality, and winter survival than Tifsport. Patriot's more aggressive growth rate and higher tiller density made it more tolerant to traffic. Also it was shown that Patriot consistently had higher TNC and lower electrolyte leakages than Tifsport, indicating its improved cold tolerance. In the past, sports field managers at Virginia Tech's Worsham Field had difficulties managing Tifsport due to poor spring recuperative potential. Based on our results, renovation to Patriot should result in a field that is more aesthetically acceptable and functional for both fall and spring uses.

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