

Management Practices, Environment, and Spray Adjuvants Influence Efficacy and

Metabolism of Bispyribac-sodium in Turfgrass

by

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## ABSTRACT OF THE DISSERTATION

# MANAGEMENT PRACTICES, ENVIRONMENT, AND SPRAY ADJUVANTS INFLUENCE EFFICACY AND METABOLISM OF BISPYRIBAC-SODIUM IN TURFGRASS

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Bispyribac-sodium has shown potential for selective annual and roughstalk bluegrass control in cool-season turfgrasses but further research is needed for successful integration in to turf management programs. The objectives of this research were to investigate the influence of management practices, environment, and spray adjuvants on efficacy and metabolism of bispyribac-sodium in turfgrass.

In field experiments, sequential bispyribac-sodium applications controlled annual bluegrass 93% but trinexapac-ethyl did not affect efficacy. Tank mixing trinexapac-ethyl with bispyribac-sodium provided similar annual bluegrass control to the herbicide alone. In field experiments, withholding nitrogen increased annual bluegrass and creeping bentgrass sensitivity to bispyribac-sodium while grasses fertilized biweekly generally had darker color. Weekly nitrogen treatments increased <sup>14</sup>C-bispyribac-sodium metabolism in both grasses compared to the unfertilized.

In field experiments, discoloration of creeping bentgrass putting greens was greatest from applications of 37 g/ha every ten days compared to 74, 148, or 298 g/ha applied less frequently. Chelated iron effectively masked discoloration of creeping bentgrass putting greens from bispyribac-sodium while trinexapac-ethyl inconsistently masked these effects. In field experiments, bispyribac-sodium regimes totaling 148, 222, and 296 g/ha controlled annual bluegrass 81, 83, and 91%, respectively, over two years. Pooled over herbicide rates, bispyribac-sodium applied two, three, and six times controlled annual bluegrass 78, 83, and 94%, respectively. Bispyribac-sodium and sulfosulfuron provided substantial reductions (80 to 100%) in roughstalk bluegrass cover in creeping bentgrass fairways by late July but regrowth was detected by October in three years suggesting herbicide applications did not control vegetative stems or crowns.

In growth chamber experiments, annual bluegrass chlorosis and clipping reductions from bispyribac-sodium were exacerbated by increased temperature from 10 to 30° C. Conversely, creeping bentgrass was most sensitive to bispyribac-sodium at 10° C but chlorosis and clipping reductions were less substantial at 20 and 30°. Spray adjuvants increased <sup>14</sup>C-bispyribac-sodium foliar absorption by two to threefold in laboratory experiments and improved annual bluegrass control in field experiments by 25 to 50% from treatments with no adjuvants. In metabolism experiments, half life of <sup>14</sup>C-bispyribac-sodium in annual bluegrass, creeping bentgrass, and perennial ryegrass was estimated at greater than seven days, one day, and two days, respectively.

## **Dedication**

I would like to dedicate this work to my grandfather, John Dullea McCullough, who graduated from Rutgers in 1935. He was born June 9, 1912 in Astoria, Long Island, New York, and attended Rahway High School in New Jersey from 1927 to 1931. He enlisted in the New Jersey National Guard on July 8, 1927 in Elizabeth, New Jersey, and was honorably discharged on July 7, 1930 at Camden, New Jersey with the rank of Corporal, Company C, 114<sup>th</sup> Infantry. He studied at Rutgers University in New Brunswick, New Jersey from 1931 to 1935 receiving his Bachelor of Science in Business on June 8, 1935. His working career is highlighted with jobs at Banker's Trust in New York on Wall Street, the State Department at the American Embassy in Quito, Ecuador, Mallard Pencil Company in Georgetown, Kentucky, and the Credit Rating Service (Credit Bureau) in Mt. Sterling, Kentucky. He died March 20, 1979 and is buried in St. Thomas cemetery, Mt. Sterling, Kentucky.

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## CHAPTER 1: REVIEW OF THE LITERATURE

### ANNUAL BLUEGRASS

Annual bluegrass (*Poa annua* L.) is a major problematic weed in turfgrass management. Annual bluegrass is native to Europe and belongs to the *Poaceae* family, *Festucoideae* subfamily, and *Festuceae* tribe (McCarty and Hale 2005). Annual bluegrass is considered an allotetraploid with 28 chromosomes ( $2n=4x=28$ ) which is believed to have originated from a cross between an annual species, *Poa infirma* H. B. K., and a perennial species, *Poa supina* Schrad. (Koshy 1968, 1969; Tutin 1957). Annual bluegrass is found in subarctic, temperate, and subtropical climates and can be perennially sustained in cool temperate and subarctic regions (Turgeon 1999).

Under routine mowing and maintenance, annual bluegrass may form a uniform turf or constituent of mixed stands with other species (Bogart and Beard 1973; McCarty and Hale 2005). Annual bluegrass has a bunch-type growth habit but perennial biotypes may form a weakly stoloniferous turf (Turgeon 1999). Annual bluegrass is a genetically diverse species with distinct morphological and physiological variation in biotypes found in turf (Lush 1989). Annual bluegrass has flattened sheaths that are bent at the base and roots at the lower sheath joint (Turgeon 1999). Leaf blades are often light green with distinct boat-shaped leaf tips that visually contrast with fine-textured turfgrasses (Uva et al. 1997). Seedling annual bluegrass leaf blade lengths may increase quicker than other grass species which may give competitive growth advantages with other grasses during establishment (Cattani et al. 2002).

Annual bluegrass thrives under moist, fertile soils in cool or shaded environments when pests are adequately controlled (Sprague and Burton 1937; Juska and Hanson 1969;

Beard et al. 1978; Waddington et al. 1978; Varco and Sartain 1986; McCarty et al. 2005). However, annual bluegrass has poor heat, drought, and stress tolerances compared to most turfgrasses, and thus, may require more intensive management in mixed stands for long-term culture (Beard et al. 1978). During summer months, annual bluegrass tends to wilt with inhibited growth from heat stress, which often creates unsightly yellow patches in turf stands (Uva et al. 1997; McCarty et al. 2005). Irrigating annual bluegrass may not improve plant quality under heat stress, and thus, managing turf with annual bluegrass populations presents challenges during summer and periods of drought (Martin and Wehner 1987).

Annual bluegrass is a shallow rooted plant which influences plant stability, stress tolerances, and capacity to secure water and mineral nutrients (Uva et al. 1997; Masaru et al. 2003). Nitrate leaching may be more problematic for annual bluegrass compared to deeper rooted turfgrasses, such as creeping bentgrass (*Agrostis stolonifera* L.), under sand based soils (Pare et al. 2006). Root growth differences among annual bluegrass biotypes have been attributed to differential growth and nitrate uptake levels (Pare et al. 2006). In greenhouse experiments, researchers noted reductions in soil moisture inhibited annual bluegrass shoot growth but creeping bentgrass produced longer plant length as soil moisture level was reduced (Masaru et al. 2003). The researchers concluded that creeping bentgrass was more tolerant of low soil moisture levels than annual bluegrass because of deeper rooting characteristics.

Compared to most turfgrass species, annual bluegrass is generally more sensitive to environmental, mechanical, and physiological stresses. Annual bluegrass has poor heat tolerances and may become dormant or desiccated under extreme temperatures

(Beard et al. 1978). In growth chamber experiments evaluating heat tolerances, Wehner and Watschke (1981) noted annual bluegrass was injured more severely than Kentucky bluegrass from heat but had similar relative growth reductions to perennial ryegrass.

Annual bluegrass is also sensitive to cold stress which may limit the potential for the species to be perennially maintained with turf in cool temperate regions. Tompkins et al. (2000) noted annual bluegrass had higher crown moisture levels than creeping bentgrass which was attributed to greater cold sensitivity. On putting green turf, ice-covered creeping bentgrass had cold-hardiness levels of  $-29^{\circ}$  C after 90 days while annual bluegrass plants were desiccated by 75 DAT (Tompkins et al. 2004).

Perennial biotypes of annual bluegrass may have better stress tolerances than annual biotypes and are often found in intensively managed turfgrasses (Uva et al. 1997; McCarty and Hale 2005). Perennial biotypes root at nodes of tillers and stolons and are often found in patches in mixed turfgrass stands (Hovin 1957). Perennial biotypes of annual bluegrass produce fewer seeds than annual biotypes but may spread and reproduce through vegetative stems (Johnson et al. 1993; Johnson and White 1997a; Uva et al. 1997).

Annual bluegrass produces seedheads in spring and early summer which reduce turfgrass aesthetics and surface uniformity (Beard et al. 1978; Danneberger and Vargas 1984). The inflorescence is a terminal panicle and single plants may produce over 350 viable seeds, even when closely mowed (Uva et al. 1997). Annual bluegrass seedheads generally emerge in spring but formation timing can differ among biotypes. Johnson and White (1998) noted various annual bluegrass biotypes flowered continually, seasonally, or continual to seasonal which was correlated with flowering trait heritability.



Annual bluegrass vernalization patterns, or floral initiation in spring after winter, differ between annual and perennial biotypes. Johnson and White (1997a) noted perennial biotypes of annual bluegrass were vernalized after ten to twelve weeks at 4 to 8° C, but not at 12°, while annual biotypes did not respond to vernalization treatments. Variation in juvenile periods was also detected as perennial biotypes flowered by 25 to greater than 35 days after annual bluegrass germination. In other experiments, Johnson and White (1997b) found perennial biotypes of annual bluegrass had different inflorescence emergence under various photoperiods. The researchers noted one biotype induced inflorescence in sixteen hour photoperiods, two biotypes were induced after eight hours, and another biotype produced no seedheads.

Annual bluegrass seedheads returned to soil with clippings may result in subsequent germination of plants, and thus, increase potential for populations in mixed stands. In field experiments, Gaussoin and Branham (1989) noted returning clippings to soil in a mixed annual bluegrass/creeping bentgrass turf increased annual bluegrass cover by 12% compared to turf with clippings removed. It was also reported that removing clippings reduced viable annual bluegrass seeds in soil by 60%.

Seedhead production directs plant assimilates away from roots, crowns, and leaves which often exacerbates annual bluegrass sensitivity to stresses. In laboratory experiments, Ong and Marshall (1975) found annual bluegrass allocated 55% of total plant assimilates to inflorescence by thirty days after seedhead emergence. The researchers noted inflorescence removal altered carbon allocation back to roots and tillers.

Plant growth regulators that inhibit seedhead formation may improve annual bluegrass quality by redirecting carbohydrates away from inflorescence (Cooper et al. 1987; Watschke et al. 1992; Gelertner and Stowell 2001; Askew et al. 2006). Cooper et al. (1988) noted seedhead suppression from mefluidide resulted in greater concentrations of fructose and glucose in annual bluegrass roots compared to untreated plants during peak seedhead emergence. Ethephon and mefluidide are an ethylene promoter and cell division inhibitor, respectively, that inhibit seedhead formation of annual bluegrass and other grasses when applied prior to emergence (Poovaiah and Leopold 1973; Eggen and Wright 1983, 1985; McCarty et al. 1985; Petrovic et al. 1985; Moore and Tautvydas 1986; Eggen et al. 1989; Serek and Reid 2000). However, application timing is critical and these materials have minimal efficacy when applied after seedhead emergence (Watschke et al. 1992).

Annual bluegrass sexual reproduction creates an abundant seedbank that is attributed to long-term persistence and competition with cool-season turfgrasses. Annual bluegrass germinates in fall but seedlings may establish in spring and early summer under favorable environmental conditions (McCarty et al. 2005). In Maryland, researchers noted the majority (50–70%) of annual bluegrass seedling emergence occurred between late September and mid-October but only 24% of all seedlings emerged between November and May (Kaminski and Deronoden 2007). It was also reported that annual bluegrass peak germination occurred with precipitation and daily temperatures above 20° C.

Researchers in California noted annual bluegrass germinated less than 1% in spring but increased to greater than 95% in fall (Shem-Tov and Fennimore 2003). From

a waveform predicted model over three years, annual bluegrass had peak emergence on November 5 and the lowest germination on June 20. In growth chamber experiments, annual bluegrass had maximum germination at 19/10° C day/night temperatures but germination was restricted under 39/29° day/night temperatures (McElroy et al. 2003). Annual bluegrass establishment in fall is also attributed to better growth and tillering compared to spring and summer. In field experiments, Brede and Duich (1986) noted annual bluegrass had higher rates of tillering than Kentucky bluegrass and perennial ryegrass during early autumn but perennial ryegrass had the highest rate during early summer.

Annual bluegrass seed size also gives the species a competitive growth advantage over other grasses during establishment. Annual bluegrass seeds weigh approximately 0.55 mg compared to 0.07 mg of creeping bentgrass which increases the potential for seedlings to secure water and mineral nutrients (Turgeon 1999; Raley et al. 2005). Additionally, dormant annual bluegrass seeds may remain viable for more than six years which could germinate upon favorable environmental conditions such as adequate moisture and soil temperatures (Allen et al. 1993; McCarty et al. 2005).

Annual bluegrass is generally more susceptible to disease and insect damage than most turfgrasses. Anthracnose (*Colletotrichum graminicola* (Ces.) Wils.) is a major disease of annual bluegrass grown under stressful conditions (Danneberger et al. 1983; Smiley et al. 2005). Anthracnose favors warm, wet, and humid weather and the severity on annual bluegrass may be exacerbated by low mowing heights, traffic, and compacted soils (Danneberger et al. 1984; Inguagiato et al. 2005; Smiley et al. 2005). Annual bluegrass is also susceptible to bacterial wilt (*Xanthomonas campestris*) and other

common turf diseases such as dollar spot (*Sclerotinia homoeocarpa* F.T. Bennett), pythium (*Pythium* spp.), and summer patch (*Magnaporthe poae*) (Turgeon 1999; Smiley et al. 2005).

Annual bluegrass is also highly susceptible to damage by the annual bluegrass weevil (*Listronotus maculicollis* Dietz; Vittum 2005, 2006). Larvae of annual bluegrass weevils commonly damage annual bluegrass around collars and perimeters of golf greens while not harming other turf species (Rothwell and Vittum 2003; Vittum 2006). Annual bluegrass sensitivity to disease, insects, and environmental stress often requires more pesticide use than most turfgrasses for successful culture, and thus, controlling annual bluegrass is often desirable in mixed turfgrass stands.

Preemergence herbicides concentrated in the upper soil layer control annual plants germinating from seed by inhibiting cell division of immature roots and shoots (McCarty et al. 2005). Applying preemergence herbicides, such as bensulide, pendimethalin, or dithiopyr, prior to annual bluegrass germination may control young plants during establishment (Goss 1964; Bingham et al. 1969; Mueller-Warrant and Rosato 2002; McCarty et al. 2005). However, preemergence herbicides often provide inconsistent levels of annual bluegrass control since germination cycles may be altered by temperature, rainfall, soil properties, and other environmental factors (Johnson 1977). Furthermore, dormant annual bluegrass seeds have potential to germinate in spring or under periods of extended rainfall (Peachey et al. 2001).

Selectively controlling annual bluegrass has been traditionally difficult in cool-season turfgrasses. Previous herbicide chemistries generally have erratic efficacy for postemergence control and often cause excessive turfgrass injury (McCarty et al. 2005).

Ethofumesate is a postemergence herbicide labeled for selective annual bluegrass control in creeping bentgrass, Kentucky bluegrass (*Poa pratensis* L.), and perennial ryegrass (*Lolium perenne* L.; Anonymous 2004a). However, labeled rates of ethofumesate for creeping bentgrass (840 g/ha) are lower than perennial ryegrass (1120 to 2240 g/ha) due to injury concerns (Anonymous 2004a). Creeping bentgrass discoloration from ethofumesate at 1120 to 2240 g/ha can be less substantial from single applications in September and October compared to November but injury from sequential applications may persist until spring regardless of timing (Johnson et al. 1989). Registered rates of ethofumesate effectively control annual bluegrass in perennial ryegrass but results are often inconsistent with reduced rates in creeping bentgrass (Dernoeden and Turner 1988; Adams 1989; Dernoeden 1996; Yelverton and McCarty 2001). In multi-year field experiments, ethofumesate at 2020 g/ha only controlled annual bluegrass 47 to 62% while lower rates (1680 g/ha or less) provided minimal to no control (Meyer and Branham 2006).

Postemergence annual bluegrass control with herbicides on creeping bentgrass putting greens is generally more difficult than fairways, tees, and roughs. Creeping bentgrass putting greens are more sensitive to postemergence herbicides than higher mowed turf, and thus, herbicide rates must be significantly reduced from rates used on fairways (McCarty et al. 2005). Selective grassy weed herbicides, such as ethofumesate, are not registered for putting greens due to excessive turfgrass injury (Anonymous 2004a). Furthermore, annual bluegrass decline from herbicides may leave voids in putting green turf that could disrupt surface quality for ball roll.

Spot treating non-selective herbicides, such as glyphosate, controls annual bluegrass plants but may not be applicable when heavy populations are present (McCarty et al. 2005). Additionally, drift from non-selective herbicide applications may injure or kill surrounding turfgrass species which reduces turf recuperation over bare areas left from annual bluegrass desiccation. Unsightly voids following non-selective herbicide applications often reduce turfgrass aesthetics and may increase the potential for crabgrass (*Digitaria* spp.) or other annual weeds to establish.

Rather than applying postemergence herbicides, turf managers have resorted to suppressing annual bluegrass populations with plant growth regulators (PGRs). Early gibberellin synthesis inhibitors, such as paclobutrazol or flurprimidol, are applied throughout fall and spring for annual bluegrass suppression (Gaul and Christians 1988; McCarty et al. 2005). Suppression from these PGRs results from greater annual bluegrass growth inhibition than other grass species, thus creating a competitive growth advantages for turfgrasses (Beasley et al. 2005). Additionally, these materials are applicable for use on creeping bentgrass putting greens (Anonymous 2002b, c). Long-term PGR use (two or more years) has shown to effectively reduce annual bluegrass present in golf course putting greens, tees, and fairways (Johnson and Murphy 1995; Woosley et al. 2004; McCullough et al. 2005). However, PGRs do not completely control annual bluegrass and are generally applied to minimize populations (Gaul and Christians 1988).

## ROUGHSTALK BLUEGRASS

Roughstalk bluegrass (*Poa trivialis* L.) is a major problematic weed of intensively managed turf, especially golf courses, that superintendents have unsuccessfully controlled (Levy 1998; McCarty et al. 2005). Roughstalk bluegrass populations reduce turfgrass aesthetics and functionality due to its light green color and shallow root system (Hurley 1997). More importantly, roughstalk bluegrass has undesirable qualities including poor disease, drought, and wear tolerances that create unsightly patches in golf course turf (Shearman and Beard 1975; Hurley 1982; Hurley and Funk 1985; Peterson et al. 2005). Consequently, golf courses infested with roughstalk bluegrass require more water, fungicides, and intensive management to maintain acceptable turfgrass quality.

Roughstalk bluegrass is a sod forming perennial with aggressive stoloniferous growth during spring and fall (Uva et al. 1997; Hurley 2003). Roughstalk bluegrass thrives under moist, shady conditions and is native to northern Europe, temperate Asia, and Northern Africa (Hurley 1997; Uva et al. 1997). The introduction of roughstalk bluegrass to North America occurred during the colonial period as a contaminant of Kentucky bluegrass seed and has since spread throughout the United States and southern Canada (Levy 1998).

Roughstalk bluegrass is a diploid with 14 chromosomes ( $2n=2x=14$ ) as reported by Ahmed et al. (1972). Roughstalk bluegrass biotypes found in the United States have substantial genetic variability that contribute to differential quality, performance, disease susceptibility, and stress tolerances (Ahmed et al. 1972; Hurley 1982; Hurley and Funk 1985; Liu et al. 1999; Camberato et al. 2000; Rajasekar et al. 2005). Breeders have successfully improved commercial varieties of roughstalk bluegrass for overseeding

dormant bermudagrass putting greens and for use in shade (Dickson et al. 1980; Hurley et al. 1990). However, indigenous roughstalk bluegrass stems and seed in soil produce unsightly patches that may rapidly infest cool-season turfgrasses, especially during turf establishment (Milton 1936; Haggard 1979).

Roughstalk bluegrass initially grows in small patches which may quickly increase up to ten feet in diameter (Hurley 1997, 2003; Uva et al. 1997). During summer months, roughstalk bluegrass infested areas may exhibit substantial decline as plants turn brown and go dormant during extended periods of heat (Hurley 1997, 2003). Roughstalk bluegrass is a perennial malady, and thus, presents significant problems for long-term successful turf management.

Preemergence herbicides control annual grasses germinating from seed but are ineffective in controlling perennial grasses, like roughstalk bluegrass, from reproducing vegetatively by stems and stolons (McCarty et al. 2005). Practitioners have unsuccessfully controlled roughstalk bluegrass due to a lack of selective herbicide chemistries in cool-season turfgrass. Researchers have previously applied dalapon, ethofumesate, and fenoxaprop to roughstalk bluegrass but these herbicides have erratic efficacy and cause unacceptable injury to cool-season turfgrasses at rates required for control (Oswald 1980; Mueller-Warrant 1990; Meyer and Branham 2006). Sulfonylurea herbicides can selectively control overseeded roughstalk bluegrass in bermudagrass during early summer but most of these herbicides cause substantial injury to cool-season grasses such as creeping bentgrass, Kentucky bluegrass, perennial ryegrass, and tall fescue (Oswald 1980; Probst 1994; Barker et al. 2005; McCullough et al. 2006).



## ACETOLACTATE SYNTHASE INHIBITING HERBICIDES

Acetolactate synthase (ALS) inhibitors are important herbicides for weed control in field crops, turf, and ornamentals. In susceptible plants, these herbicides inhibit ALS enzymes which catalyze the synthesis of branch chain amino acids isoleucine, leucine, and valine (LaRossa and Schloss 1984). Plants are controlled through toxic accumulation of substrates from inhibited branch-chain amino acid production, but sequences of phytotoxic responses are unclear (Senseman 2007; Shimizu et al. 2002). Herbicide families that have ALS inhibitors include imidazolinones, pyrimidinyloxybenzoates, sulfonlaminocarbonyltriazolinones, sulfonlureas, and triazolopyrimidines (Senseman 2007).

### **Bispyribac-sodium**

Bispyribac-sodium (2,6-bis(4,6-dimethoxypyrimidin-2-yloxy)benzoate) belongs to the pyrimidinyloxybenzoate family and controls susceptible weeds by inhibiting ALS enzymes involved in branch chain amino acid synthesis (Senseman 2007). Bispyribac-sodium has traditionally been used for grass and broadleaf weed control in dry-seeded or water seeded rice (*Oryza sativa* L.) production (Anonymous 2002). Researchers have noted bispyribac-sodium at 20 to 60 g ai/ha effectively controls alligatorweed (*Alternanthera philoxeroides* (Mart.) Griseb), barnyardgrass (*Echinochloa crus-galli* (L.) Beauv.), ducksalad (*Heteranthera limosa* (Sw.) Willd.) hemp sesbania (*Sesbania exaltata* (Raf.) Cory), Indian jointvetch (*Aeschynomene indica* L.), junglerice (*E. colona* (L.) Link), northern jointvetch (*Aeschynomene virginica* (L.) B.S.P), sedges (*Cyperus* spp.),

and signalgrass (*Brachiaria* spp.) (Braverman and Jordan 1996; Schmidt et al. 1999; Senseman 2007; Shimizu et al. 2002; Webster et al. 1999; Williams 1999, 2000).

Fagerness and Penner (1998) investigated bispyribac-sodium as a Class D plant growth regulator (chemicals with herbicidal activity applied at sublethal rates) for growth suppression in cool-season turfgrasses. In greenhouse experiments, the researchers applied bispyribac-sodium at 0, 15, 29, and 59 g/ha with a non-ionic surfactant at 0.25% (v/v) to creeping bentgrass, Kentucky bluegrass, perennial ryegrass, tall fescue, and creeping red fescue (*Festuca rubra* L.). Bispyribac-sodium discolored all grasses greater than 20% and reduced clipping weight but creeping bentgrass color and growth recovered faster than other species.

Bispyribac-sodium efficacy for annual bluegrass control use in turf was initially discovered in field experiments at Michigan State University (Branham and Calhoun 2005). From 2000 to 2004, extensive regional experiments were conducted to evaluate bispyribac-sodium efficacy for weed control in cool-season turfgrasses and the herbicide was federally registered in the United States for use in turfgrass in 2004 (Anonymous 2004b). Lycan and Hart (2005) investigated responses of field grown Kentucky bluegrass, perennial ryegrass, tall fescue, and chewings fine fescue (*Festuca rubra* L. subsp. *commutata* Gaud.) to single bispyribac-sodium applications at 37 to 296 g/ha. It was noted Kentucky bluegrass was least tolerant to bispyribac-sodium with up to 28% injury but other species had less than 20% injury after thirty days. Bispyribac-sodium reduced Kentucky bluegrass clipping weights by 19 to 35% from the untreated after thirty-five days but other species had similar clippings to the untreated. Similar discoloration and recovery has been reported on creeping bentgrass, colonial bentgrass

(*Agrostis capillaries* L.), perennial ryegrass, and velvet bentgrass (*Agrostis canina* L.) following bispyribac-sodium applications (McCarty and Estes 2005; McDonald et al. 2006a).

Bispyribac-sodium may also severely injure roughstalk bluegrass and have shown to visually eliminate plants in turf stands. In Virginia, three applications of bispyribac-sodium in summer at 74 g ai/ha controlled roughstalk bluegrass 95% by ten weeks after initial treatments (Askew et al. 2004). In regional experiments throughout the U.S. mid-West, researchers noted four bispyribac-sodium applications at 111 g/ha applied biweekly in summer provided 90 to 100% elimination of roughstalk bluegrass in creeping bentgrass fairways after twelve weeks (Morton and Reicher 2007). However, field experiments investigating long-term roughstalk bluegrass control with bispyribac-sodium are limited.

Physiological responses to bispyribac-sodium among grass species may attribute to selectivity for annual bluegrass control in turf. Lycan and Hart (2006b) noted annual bluegrass and creeping bentgrass retained greater than 90% of foliar applied <sup>14</sup>C-bispyribac-sodium in treated leaves but annual bluegrass translocated more herbicide to crowns. It was also noted annual bluegrass translocated more foliar and root applied <sup>14</sup>C-bispyribac-sodium to leaves than creeping bentgrass. In other experiments with ethofumesate, annual bluegrass absorbed more <sup>14</sup>C-herbicide than creeping bentgrass and perennial ryegrass which may be attributed to efficacy in turf (Kohler and Brahnham 2002).

Although herbicide translocation may contribute to efficacy, differential metabolism has been associated with selectivity of ALS-inhibitors. Olson et al. (2000)

noted differential responses of jointed goatgrass (*Aegilops cylindrical* Host. ), downy brome (*Bromus tectorum* L.), and spring wheat (*Triticum aestivum* L.) to sulfosulfuron was attributed to metabolism. Dubelman et al. (1997) noted corn (*Zea mays* L.) and wheat (*Triticum aestivum* L.) tolerance to halosulfuron was due to rapid metabolism while soybean, a sensitive species, had limited metabolism. King et al. (2003) noted less absorption and greater metabolism by wheat and barley (*Hordeum vulgare* L.) of AE-F130060-03 in comparison with Italian ryegrass (*Lolium multiflorum* L.) was correlated with species tolerance and herbicide selectivity. In other experiments, selectivity of halosulfuron, imazethapyr, primisulfuron, and trifloxysulfuron on other grasses has been attributed to metabolism (Cole et al. 1989; Gallaher et al. 1999; Askew and Wilcut 2002).

### Objectives

Bispyribac-sodium has shown potential for selective annual and roughstalk bluegrass control in cool-season turfgrasses but further research is needed for successful integration in turf management programs. The objectives of this research were to investigate efficacy and metabolism of bispyribac-sodium in turfgrass by specifically evaluating:

- 1) the influence of management practices, including mowing height, iron, nitrogen, and plant growth regulators, on bispyribac-sodium efficacy;
- 2) bispyribac-sodium application regimes for annual bluegrass control on creeping bentgrass putting greens and roughstalk bluegrass control in creeping bentgrass fairways.

- 3) bispyribac-sodium metabolism in annual bluegrass, creeping bentgrass, and perennial ryegrass.

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CHAPTER 2: TRINEXAPAC-ETHYL INFLUENCES BISPYRIBAC-SODIUM  
EFFICACY FOR ANNUAL BLUEGRASS CONTROL IN CREEPING BENTGRASS

ABSTRACT

Bispyribac-sodium (BS) selectively controls annual bluegrass in cool-season turf but efficacy may be influenced by management practices, such as plant growth regulator (PGR) use. Experiments were conducted in New Jersey to investigate efficacy and absorption of BS applied with a PGR, trinexapac-ethyl (TE), for annual bluegrass control and turfgrass tolerance. In laboratory experiments with annual bluegrass, creeping bentgrass, and perennial ryegrass, tank mixing TE with  $^{14}\text{C}$ -BS increased foliar absorption from non-TE treated while absorption increased with TE rate. Differences in  $^{14}\text{C}$ -BS absorption were not detected among emulsifiable concentration (EC), microencapsulated concentration (MC), and wettable powder (WP) TE formulations. In field experiments, sequential BS applications controlled annual bluegrass 93% but TE did not affect efficacy. Tank mixing EC, MC, or WP TE formulations with BS provided similar annual bluegrass control and creeping bentgrass quality. Applications of BS reduced dollar spot (DS) cover from non-BS treated in both years while TE reduced DS cover from non-TE treated in 2005.

**Nomenclature:** Bispyribac-sodium; Annual bluegrass, *Poa annua* L.; Creeping bentgrass, *Agrostis stolonifera* L., ‘L-93’, ‘G-2; Perennial ryegrass, *Lolium perenne* L., ‘Applaud’.

## INTRODUCTION

Annual bluegrass (*Poa annua* L.) is a major problematic weed in cool-season turfgrasses (Beard 1970). Annual bluegrass reduces turfgrass aesthetics and functionality due to its lighter green color, unsightly seedheads, and shallow root system (Lush 1989; Sprague and Burton 1937). Annual bluegrass tolerates close mowing, germinates rapidly, and has undesirable qualities including poor disease, drought, and wear tolerances that create unsightly patches in turfgrass stands (Beard et al. 1978; Kaminski and Dernoeden 2002). Consequently, turf infested with annual bluegrass requires more water, fungicides, and intensive management, especially in summer months, to maintain acceptable quality (Sprague and Burton 1937; Beard et al. 1978).

Bispyribac-sodium is a pyrimidinyloxybenzoic herbicide that controls weeds by inhibiting acetolactate synthase (ALS, EC 2.2.1.6), similar to sulfonyleureas (Shimizu et al. 2002). Bispyribac-sodium has been used for selective postemergence control of barnyardgrass (*Echinochloa crus-galli* (L.) Beauv.) and other weeds in rice (*Oryza sativa* L.) (Schmidt et al. 1999; Webster et al. 1999; Williams 1999) but was recently registered for postemergence annual bluegrass control in creeping bentgrass (*Agrostis stolonifera* L.) and perennial ryegrass (*Lolium perenne* L., Anonymous 2004). Previous investigations have noted bispyribac-sodium at 60 to 148 g/ha may reduce annual bluegrass populations but turfgrass discoloration may persist for two to three weeks after applications (Askew et al. 2004; Branham and Calhoun 2005; Lycan and Hart 2006a; Park et al. 2002). Turf discoloration may concern end-users, and thus, management practices to mitigate turf chlorosis from bispyribac-sodium are warranted.



Plant growth regulators (PGRs) are extensively used in turf management to reduce mowing requirements and enhance turfgrass quality. Trinexapac-ethyl is a foliarly absorbed, late gibberellin synthesis inhibitor that is extensively used in golf course management (Rademacher 2000; Anonymous 2002; Tan and Qian 2003). Inhibiting leaf growth with trinexapac-ethyl may increase turfgrass chlorophyll concentration which helps mitigate chlorosis from heat, drought, shade, and environmental stresses (Golembiewski and Dannerbeger 1998; Jiang and Fry 1998; Qian and Engelke 1999; Fagerness and Yelverton 2000; Heckman et al. 2001a,b; Ervin et al. 2001; Goss et al. 2002; Steinke and Stier 2003; McCarty et al. 2004). Enhancing turfgrass color with trinexapac-ethyl may help reduce chlorosis from bispyribac-sodium but applications could affect herbicide efficacy for annual bluegrass control.

To test this hypothesis, laboratory and field experiments were conducted to investigate bispyribac-sodium efficacy and foliar absorption as influenced by trinexapac-ethyl rate, formulation, and application regimen.

## MATERIALS AND METHODS

**Laboratory Experiments.** ‘G-2’ creeping bentgrass, ‘Applaud’ perennial ryegrass, and annual bluegrass were seeded in pots with 1-dm<sup>2</sup> surface area and 10-cm depths in a greenhouse at the New Jersey Agriculture Greenhouse Research Complex. Greenhouse day/night temperatures were set for approximately 25/19 C and natural light was supplemented with high intensity discharge lighting when intensity fell below 600  $\mu\text{mol m}^2/\text{s}$ . Seeding rates were 48 and 288 kg/ha for creeping bentgrass and perennial ryegrass, respectively. Indigenous annual bluegrass seedheads were harvested at Horticultural

Research Farm II in North Brunswick, NJ in spring of 2004 and stored in a growth chamber at 8 C before seeding. Annual bluegrass was seeded by uniformly distributing seedhead material across pots. Soil medium<sup>1</sup> consisted of 80% Canadian Sphagnum peat moss and a 20% mixture of perlite, vermiculite, dolomitic limestone, and calcitic limestone. Grasses were irrigated to prevent plant wilt and mowed at a 1.3-cm height with grass sheers 3 d/wk. Grasses were allowed to grow and tiller in pots before transplanting individual plants into 20.5-cm deep by 3.8-cm diameter conetainers. Soil was similar to that aforementioned. Grasses were allowed to resume active growth in conetainers prior to herbicide treatments.

Treatments included the factorial combination of three trinexapac-ethyl formulations and four trinexapac-ethyl rates, applied with bispyribac-sodium<sup>2</sup> at 74 g/ha. Trinexapac-ethyl formulations included emulsifiable concentration<sup>3</sup> (EC), microencapsulated concentration<sup>4</sup> (MC), and wettable powder<sup>5</sup> (WP). Rates of trinexapac-ethyl tank-mixed with bispyribac-sodium applied to creeping bentgrass and annual bluegrass were 0, 50, 100, or 200 g/ha while trinexapac-ethyl rates for perennial ryegrass were 0, 200, 400, or 800 g/ha. Rates were chosen based on registered rates for respective species (Anonymous 2002). Bispyribac-sodium was also applied with a non-ionic surfactant<sup>6</sup> at 0.25% v/v of spray solution as a separate, standard comparison treatment. Broadcast treatments were applied with a CO<sub>2</sub>-pressued spray chamber<sup>7</sup> equipped with an 8002 EVS spray tip<sup>8</sup> calibrated to deliver 374 L/ha at 290 kPa. Immediately following broadcast applications, a 2 $\mu$ L droplet of spotting solution containing a total of 0.20 kBq of <sup>14</sup>C-bispyribac-sodium [uniformly ring labeled (specific activity, 2.66 MBq/mg; radiochemical purity, 99.1%)] was applied to the second fully

expanded leaf with a microsyringe<sup>9</sup>. Appropriate amount of formulated herbicide with or without trinexapac-ethyl treatments aforementioned or a non-ionic surfactant was added to spotting solutions to simulate 74 g/ha of bispyribac-sodium at a spray volume of 374 L/ha.

Treated leaves were excised eight hours after treatment for foliar absorption evaluations using methods described in previous experiments (Fagerness and Penner, 1998; Young and Hart, 1998; Lycan and Hart, 2006b). This harvest timing was chosen based on previous investigations with <sup>14</sup>C-bispyribac-sodium absorption by Lycan and Hart (2006b). Unabsorbed <sup>14</sup>C-bispyribac-sodium was removed by swirling the treated leaf in a 20-ml scintillation vial containing 2 mls of 10% methanol solution for approximately 45 seconds. Upon removal from vials, leaves were rinsed with 2 ml of washoff solution similar to that aforementioned. Unabsorbed <sup>14</sup>C was quantified by liquid scintillation spectroscopy<sup>10</sup>. Foliar absorption was calculated by subtracting amount of <sup>14</sup>C recovered in rinsate from amount of <sup>14</sup>C applied with the following equation:

$$\text{Percent absorption} = 100 - [100 \times (\text{DPM}_0 - \text{DPM}_x) / (\text{DPM}_0)]$$

$$\text{DPM}_0 = \text{DPM (disintegrations per minute) of } ^{14}\text{C in applied solution}$$

$$\text{DPM}_x = \text{DPM of unabsorbed } ^{14}\text{C}$$

Two experiments were conducted as a randomized complete block with four replications. Data were analyzed using analysis of variance<sup>11</sup> to determine significance of main effects of species, trinexapac-ethyl rate, and formulation. Means were separated using Fisher's Protected LSD test at  $\alpha = 0.05$ . Orthogonal polynomial contrasts were performed to evaluate relationship of trinexapac-ethyl rate with foliar absorption.

Experiment by treatments and species by treatment interactions were not detected, and thus results were pooled and presented by main effects.

**Field Experiments.** Experiments were conducted on creeping bentgrass and annual bluegrass fields at Horticultural Research Farm II, North Brunswick, NJ from May to July 2005 and 2006. Soil on both fields was a Nixon sandy loam (fine-loamy, mixed, semiactive mesic type Hupludults) with a pH of 6.3 with approximately 30 g/kg organic matter. ‘L-93’ creeping bentgrass was established in fall 2002 and plots used in 2006 were adjacent to plots in 2005. Annual bluegrass was established in fall 2004 and 2005 for experiments conducted in 2005 and 2006, respectively. Annual bluegrass fields were prepared by aerification and tillage to allow indigenous seeds to germinate and establish. By May of both years, annual bluegrass coverage ranged 95 to 100%. Beginning late April, both fields were fertilized biweekly at 12 kg N/ha with a 16-1-4 (N-P-K) granular fertilizer<sup>12</sup> and mowed 3 d/wk at 9.5-mm with a reel mower<sup>13</sup>.

Experimental design was a randomized complete block with four replications of 1 x 3-m plots on both fields. Treatments were applied by making two passes per plot in opposite directions with a single nozzle CO<sub>2</sub> pressured sprayer calibrated to deliver a total 374 L/ha. Nozzles<sup>8</sup> used were 9504E and CO<sub>2</sub> regulators were set for 220 kPa.

Bispyribac-sodium<sup>2</sup> was applied at 0 or 111 g/ha and was implemented with three trinexapac-ethyl application regimens: (1) biweekly applications suspended two weeks prior to bispyribac-sodium, (2) biweekly applications suspended one week prior to bispyribac-sodium, and (3) biweekly applications prior to and tank mixed with bispyribac-sodium. Trinexapac-ethyl in EC was applied prior to bispyribac-sodium for

all regimes while EC, MC, or WP formulations were tank mixed with bispyribac-sodium as three separate treatments for regime 3. Trinexapac-ethyl in EC formulation was also applied alone on dates of bispyribac-sodium treatments. All trinexapac-ethyl applications were made at 0 or 50 g/ha.

Leaf color was visually rated weekly on a 1 to 9 scale where 1 equaled brown and 9 equaled dark green turf. Percent annual bluegrass and dollar spot cover were visually rated where 0 equaled no cover and 100 equaled complete plot cover. Annual bluegrass control was calculated with the following equation:

$$\text{Percent control} = [(Poa_0 - Poa_x) / (Poa_0)] \times 100$$

$Poa_0$  = annual bluegrass cover of untreated plots

$Poa_x$  = annual bluegrass cover of treated plots

Data were subjected to analysis of variance and mean separations were made with Fisher's LSD test at  $\alpha = 0.05$ . Year by treatment interactions were detected for creeping bentgrass color, thus results are presented by year. Year by treatment interactions were not detected for annual bluegrass control, therefore results were pooled over years.

## RESULTS AND DISCUSSION

In laboratory experiments, differences among trinexapac-ethyl formulations were not detected for  $^{14}\text{C}$ -bispyribac-sodium foliar absorption (Table 1). Grasses treated with  $^{14}\text{C}$ -bispyribac-sodium alone absorbed 21% of the herbicide after eight hours. Tank-mixing trinexapac-ethyl with  $^{14}\text{C}$ -bispyribac-sodium increased absorption to 30% while a non-ionic surfactant increased absorption to 37%.  $^{14}\text{C}$ -bispyribac-sodium absorption in

Table 1. Influence of trinexapac-ethyl formulation and rate on  $^{14}\text{C}$  bispyribac-sodium foliar absorption in two combined laboratory experiments.

Treatment Tank Mixed with Bispyribac-sodium <sup>a</sup>	Foliar Absorption <sup>b</sup>			
	%			
None	21			
trinexapac-ethyl (EC)	30			
(MC)	28			
(WP)	31			
non-ionic surfactant	37			
LSD <sub>0.05</sub>	3			
Trinexapac-ethyl Rate <sup>c</sup>	Annual Bluegrass	Creeping Bentgrass	Perennial Ryegrass	Mean
0	21	25	16	21
low	21	24	21	22
medium	32	30	41	34
high	29	32	34	32
Linear	*	*	*	*
Quadratic	*	*	*	*
Cubic	NS	NS	NS	NS

<sup>a</sup>EC = trinexapac-ethyl in emulsifiable concentration, MC = trinexapac-ethyl in microencapsulated concentration, WP = trinexapac-ethyl in wettable powder, NIS = non-ionic surfactant applied at 0.25% v/v.

<sup>b</sup>Foliar absorption was measured eight hours after treatments and results were pooled over annual bluegrass, creeping bentgrass, and perennial ryegrass absorption.

<sup>c</sup>Trinexapac-ethyl rates for creeping bentgrass and annual bluegrass were 50 (low), 100 (medium), and 200 (high) g/ha while rates for perennial ryegrass were 200 (low), 400 (medium), and 800 (high) g/ha. Bispyribac-sodium was applied at 74 g/ha.

\*Significant at 0.05 probability level. NS = not significant at 0.05 probability level.

annual bluegrass, creeping bentgrass, and perennial ryegrass increased with trinexapac-ethyl rate.

In field experiments, bispyribac-sodium by trinexapac-ethyl interaction was not detected for annual bluegrass control. Bispyribac-sodium controlled annual bluegrass 93% by July 11 of both years (Table 2) but trinexapac-ethyl did not affect efficacy (not shown). Applying trinexapac-ethyl before bispyribac-sodium treatments did not enhance annual bluegrass tolerance to the herbicide which may be a concern since the PGR often improves grass tolerance to stress and chlorosis (Jiang and Fry 1998; Ervin et al. 2001; Heckman et al. 2001a). Emulsifiers in trinexapac-ethyl formulations increase foliar absorption of suspended materials, and at higher trinexapac-ethyl rates, bispyribac-sodium foliar absorption appears comparable to the addition of a non-ionic surfactant. Although these effects may improve efficacy, bispyribac-sodium applications with trinexapac-ethyl did not improve annual bluegrass control from the herbicide alone.

Bispyribac-sodium by trinexapac-ethyl interactions were not detected for creeping bentgrass color. Differences were also not detected among trinexapac-ethyl formulations tank-mixed with bispyribac-sodium (previously delineated as regimen 3), and thus, results were pooled and presented as a single treatment in Table 3. Color reductions (delineated chlorosis) ranged 5 to 32% and lasted two weeks after initial bispyribac-sodium applications in both years (Table 3). Creeping bentgrass chlorosis was 11% by one week after the second bispyribac-sodium application in 2005 but turf recovered similar to nontreated by the following week. In 2006, initial and sequential bispyribac-sodium applications caused 5 to 18% chlorosis for one to two weeks, but creeping bentgrass recovered similar to the nontreated after three weeks. Similar chlorosis has

*Table 2.* Annual bluegrass control following sequential bispyribac-sodium treatments in field experiments, 2005-2006, North Brunswick.

Bispyribac-sodium <sup>a</sup>	Annual Bluegrass Control <sup>b</sup>
g/ha	%
0	3
111 + 111	93*

\*Significant at the 0.05 probability level.

<sup>a</sup> Bispyribac-sodium was applied June 2 and June 21 in 2005 and May 31 and June 20 in 2006.

<sup>b</sup>Initial annual bluegrass ground cover ranged 95 to 100%. Annual bluegrass control was rated July 11, 2005 and July 11, 2006.



Table 3. Turf color of ‘L-93’ creeping bentgrass treated with bispyribac-sodium and trinexapac-ethyl in field experiments, 2005-2006, North Brunswick, NJ.

Year	TE Application Date <sup>b</sup>	Turf Color <sup>a</sup>							
		5/16	6/1	6/9	6/13	6/21	6/28	7/6	7/11
		----- 1 to 9 -----							
2005	Untreated	7.8	8.0	5.9	6.7	7.1	7.1	7.9	7.8
	5/2 + 5/16	7.5	7.9	6.3	6.9	7.5	7.0	7.9	7.8
	5/9 + 5/27	7.5	7.8	6.8	6.9	7.3	7.0	7.7	7.7
	5/2 + 5/16 + 6/2 + 6/21	7.5	8.1	5.9	6.8	7.6	7.3	7.8	7.9
	LSD <sub>0.05</sub>	NS	NS	0.7	NS	0.4	NS	NS	NS
BS Application Date									
	Untreated			7.6	7.6	7.3	7.8	7.8	7.7
	6/2 + 6/21			5.2	6.4	7.5	6.9	7.9	7.9
	LSD <sub>0.05</sub>			0.5	0.3	NS	0.5	NS	0.2
		----- 1 to 9 -----							
	TE Application Date	5/15	5/31	6/8	6/14	6/20	6/27	7/3	7/11
		----- 1 to 9 -----							
2006	Untreated	7.6	7.3	6.6	6.8	6.9	6.4	7.2	7.0
	5/1 + 5/15	7.5	8.1	7.5	7.6	7.6	6.6	7.4	7.3
	5/8 + 5/25	7.4	7.7	8.0	7.8	7.8	7.1	7.6	7.3
	5/1 + 5/15 + 5/31 + 6/20	7.3	7.9	7.9	8.0	7.8	7.4	7.9	7.4
	LSD <sub>0.05</sub>	NS	0.3	0.3	0.3	0.3	0.4	0.3	0.2
BS Application Date									
	Untreated			8.0	7.8	7.4	7.8	7.8	7.3
	5/31 + 6/20			7.1	7.4	7.6	6.4	7.4	7.2
	LSD <sub>0.05</sub>			0.3	0.2	NS	0.3	0.3	NS

<sup>a</sup>Turf color was rated on a 1 to 9 scale where 1 equaled brown turf and 9 equaled dark green turf.

<sup>b</sup>TE = trinexapac-ethyl. Application rates were 0 or 50 g/ha. BS = bispyribac-sodium. Application rates were 0 or 111 g/ha.

been reported on creeping bentgrass, colonial bentgrass (*Agrostis capillaries* L.), perennial ryegrass, tall fescue (*Festuca arundinacea* (L.) Shreb.), and velvet bentgrass (*Agrostis canina* L.) following bispyribac-sodium applications (Lycan and Hart 2005; McCarty and Estes 2005; Lycan and Hart 2006a; McDonald et al. 2006).

Creeping bentgrass tolerance to bispyribac-sodium was frequently improved with trinexapac-ethyl applications. In 2005, creeping bentgrass treated with trinexapac-ethyl on May 9 and 27 averaged 15% higher color ratings than non-trinexapac-ethyl treated turf on June 9, one week after initial bispyribac-sodium applications (Table 3). On June 21, creeping bentgrass treated with trinexapac-ethyl on May 2, May 16, and June 2 had 7% darker color than non-trinexapac-ethyl treated turf. In 2006, creeping bentgrass treated with all trinexapac-ethyl regimes had higher color ratings than non-trinexapac-ethyl treated turf from May 31 to June 20. Creeping bentgrass treated with trinexapac-ethyl on May 1 and 15 had similar color to non-trinexapac-ethyl treated turf by June 27 while other regimens averaged 6 to 16% darker green color one to two weeks after sequential bispyribac-sodium treatments (June 27 and July 3).

Results suggest color enhancements from trinexapac-ethyl have potential to mitigate chlorosis from bispyribac-sodium applications but may not completely mask these effects. Trinexapac-ethyl suspended one week before or tank-mixed with bispyribac-sodium were the most effective regimens for reducing creeping bentgrass chlorosis from the herbicide alone. In other experiments, chelated iron helped mask chlorosis from bispyribac-sodium but did not affect annual bluegrass control (McDonald et al. 2006).

Creeping bentgrass dollar spot cover was reduced by bispyribac-sodium and trinexapac-ethyl but interactions were not detected with treatments. Dollar spot was present from June 13 to July 25, 2005 and June 14 to July 11, 2006. Creeping bentgrass treated with bispyribac-sodium had dollar spot cover reduced by 63% from nontreated over two years (Table 4). In Virginia, sequential bispyribac-sodium applications at 37 and 74 g/ha reduced 'Penncross' creeping bentgrass dollar spot cover 45 to 69% from nontreated (Askew et al. 2004). Similar results have been reported in South Carolina and Michigan with sequential bispyribac-sodium applications at 74 to 111 g/ha (Branham et al. 2005; McCarty and Estes 2005).

In 2005, trinexapac-ethyl applications in May plus June averaged 50% less dollar spot cover than non-trinexapac-ethyl treated while other regimens were similar (Table 4). Efficacy of trinexapac-ethyl applied in May had probably diminished when dollar spot initially appeared, and thus, sequential trinexapac-ethyl treatments in May plus June were more effective. Golembiewski and Danneberger (1998) noted 'Penncross' creeping bentgrass dollar spot cover averaged 8% for trinexapac-ethyl treated which was reduced from 17% of the nontreated. These effects were not consistent over the two years as creeping bentgrass treated with trinexapac-ethyl had similar dollar spot cover to nontreated turf in 2006. Similar results were reported by Burpee et al. (1996) noting trinexapac-ethyl applications reduced early dollar spot epidemics on creeping bentgrass.

Selective annual bluegrass control with bispyribac-sodium has beneficial agronomic and economic implications for cool-season turfgrass culture. However, routine management regimes may significantly influence herbicide efficacy and turfgrass tolerance. Plant growth regulator use has become an important aspect of numerous

Table 4. Dollar spot cover for ‘L-93’ creeping bentgrass treated with bispyribac-sodium and trinexapac-ethyl, 2005-2006, North Brunswick, NJ.

Trinexapac-ethyl (Application dates) <sup>a</sup>		Pooled Dollar Spot Cover <sup>b</sup>	
2005	2006	2005	2006
		----- % -----	
Untreated	Untreated	6	4
5/2 + 5/16	5/1 + 5/15	5	4
5/9 + 5/27	5/8 + 5/25	8	3
5/2 + 5/16 + 6/1 + 6/21	5/1 + 5/15 + 5/31 + 6/20	3	5
	LSD <sub>0.05</sub>	3	NS
Bispyribac-sodium (Application date)		2005 + 2006	
2005	2006	----- % -----	
Untreated	Untreated	8	
6/2 + 6/21	5/31 + 6/20	3	
	LSD <sub>0.05</sub>	1	

<sup>a</sup>Trinexapac-ethyl and bispyribac-sodium were applied at 50 and 111 g/ha, respectively.

<sup>b</sup>Ratings in 2005 were taken June 13, 21, 28 and July 11, 25. Ratings in 2006 were taken June 14, 20 and July 3, 11. Interactions were detected for year and trinexapac-ethyl so years are presented separately. Results were pooled over years for bispyribac-sodium since an interaction was not detected. Interaction with sampling date and treatments were not detected, so results were pooled and presented as overall means.

management programs to reduce mowing requirements and enhance turfgrass quality. Trinexapac-ethyl has promising implications for use with bispyribac-sodium applications for annual bluegrass control and turfgrass chlorosis mitigation. Overall, routinely using trinexapac-ethyl appears to have beneficial effects when applied with bispyribac-sodium for reducing turf chlorosis, controlling dollar spot, and improving foliar absorption for annual bluegrass control.

### **Source of Materials**

<sup>1</sup> Pro-Mix Soil, Premier Horticulture, Quakertown, PA 18951

<sup>2</sup> Velocity®, 80WP herbicide, Valent U.S.A. Corp. PO Box 8025, Walnut Creek, CA 94596.

<sup>3</sup> Primo, 1EC, Syngenta Crop Protection Inc., Greensboro, NC 27409.

<sup>4</sup> Primo Maxx, 1MC, Syngenta Crop Protection Inc., Greensboro, NC 27409.

<sup>5</sup> Primo 25WP, Syngenta Crop Protection Inc., Greensboro, NC 27409.

<sup>6</sup> X-77® non-ionic surfactant, Loveland Industries Inc., Greeley, CO 80632.

<sup>7</sup> Research Track Sprayers, Devries Manufacturing, Hollandale, MN 56045.

<sup>8</sup> Tee Jet®, Spraying Systems Co. Wheaton, IL 60189-7900.

<sup>9</sup> Microsyringe, Hamilton Co., Reno, NV 89502

<sup>10</sup> Model LS3801, Beckman-Coulter, Inc., Fullerton, CA 92834-3100.

<sup>11</sup> SAS Institute, Cary, NC 27511.

<sup>12</sup> Signature Brand Fertilizer, Greeley, CO 80632-1286.

<sup>13</sup> John Deere 2500B greens mower, Moline, IL 61265.

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CHAPTER 3: NITROGEN INFLUENCES BISPYRIBAC-SODIUM EFFICACY AND  
METABOLISM IN TURFGRASS

ABSTRACT

Field and laboratory experiments were conducted in New Jersey to investigate the influence of nitrogen on annual bluegrass control and creeping bentgrass responses to bispyribac-sodium. In field experiments, withholding nitrogen during the test period increased sensitivity of both grasses to bispyribac-sodium while grasses fertilized biweekly had darker color on the majority of rating dates. Nitrogen generally increased annual bluegrass tolerance to bispyribac-sodium at 74 g/ha but not from 148 g/ha while creeping bentgrass was influenced by nitrogen at both herbicide rates. In laboratory experiments, weekly nitrogen treatments increased <sup>14</sup>C-bispyribac-sodium metabolism in both grasses compared to the unfertilized. Annual bluegrass metabolized approximately 50% less herbicide compared to creeping bentgrass. Overall, routine nitrogen fertilization appears to improve annual bluegrass and creeping bentgrass tolerance to bispyribac-sodium which may be attributed to higher metabolism.

**Nomenclature:** Bispyribac-sodium; Annual bluegrass, *Poa annua* L.; Creeping bentgrass, *Agrostis stolonifera* L., 'L-93'.

## INTRODUCTION

Annual bluegrass (*Poa annua* L.) is a major problematic weed in cool-season turfgrasses (Beard 1970). Annual bluegrass reduces turf aesthetics and functionality due to its lighter green color, unsightly seedheads, and shallow root system (Lush 1989; Sprague and Burton 1937). Annual bluegrass tolerates close mowing, germinates rapidly, and has undesirable qualities including poor disease, drought, and wear tolerances that create unsightly patches in turfgrass stands (Beard et al. 1978; Lush 1989; Kaminski and Dernoeden 2007). Consequently, turf infested with annual bluegrass requires more water, fungicides, and intensive management, especially in the summer months, to maintain acceptable quality (Sprague and Burton 1937; Beard et al. 1978).

Bispyribac-sodium is a pyrimidinyloxybenzoic herbicide that controls weeds by inhibiting acetolactate synthase (ALS, EC 2.2.1.6) enzymes (Shimizu et al. 2002). Bispyribac-sodium has been used for selective postemergence control of barnyardgrass (*Echinochloa crus-galli* (L.) Beauv.) and other weeds in rice (*Oryza sativa* L.) (Schmidt et al. 1999; Webster et al. 1999; Williams 1999) but was recently registered for postemergence annual bluegrass control in creeping bentgrass and perennial ryegrass (Anonymous 2004). Field research with bispyribac-sodium indicates applications at 60 to 148 g/ha can reduce annual bluegrass populations without significant turf injury (Askew et al. 2004; Branham and Calhoun 2005; Lycan and Hart 2006a; Park et al. 2002). Thus, practitioners have a highly efficacious herbicide for selective annual bluegrass control.

Nitrogen is an important macronutrient for turfgrass growth, quality, and leaf color (Bowman 2003; Goss et al. 2002; Landschoot and Waddington 1987; Markland and

Roberts 1969). Nitrogen is a key constituent of chlorophyll, proteins, and amino acids and is needed in greater quantities than other mineral nutrients (Turner and Hummel 1992). Nitrogen has shown to improve turfgrass responses to ethofumesate, MSMA, broadleaf weed herbicides, plant growth regulators, and preemergence herbicides (Dernoeden et al. 1993; Johnson 1984, 1990; Johnson and Burns 1985; McCarty et al. 1984). However, nitrogen fertilization has also shown inconsistent effects on atrazine, mesotrione, nicosulfuron, glufosinate, and glyphosate efficacy for grassy weed control (Cathcart et al. 2004). Thus, routine nitrogen applications may influence annual bluegrass control with bispyribac-sodium. To test this hypothesis, field and laboratory experiments were conducted to investigate the influence of nitrogen on bispyribac-sodium efficacy for annual bluegrass control in creeping bentgrass.

## MATERIALS AND METHODS

**Field Experiments.** Experiments were conducted on an annual bluegrass field and a creeping bentgrass field at Horticultural Research Farm II, North Brunswick, NJ from May to July 2005 and 2006. Soil on both fields was a Nixon sandy loam (fine-loamy, mixed, semiactive mesic type Hupludults) with a pH of 6.3 with approximately 30 g/kg organic matter. ‘L-93’ creeping bentgrass was established in fall 2002. The annual bluegrass field was established in fall 2004 and 2005 for experiments conducted in 2005 and 2006, respectively. Vegetation was controlled with glyphosate and soil was prepared to allow indigenous annual bluegrass establishment. Annual bluegrass was approximately 90% ground cover in spring and the biotype, from visual assessment, was *Poa annua* var. *annua*. Fertilizer was applied at approximately 12 kg N/ha biweekly and

mowed 3 d/wk at 1.3 cm with a reel mower in fall. In April of both years, annual bluegrass and creeping bentgrass fields received two applications of 12 kg N/ha with a granular fertilizer (16N-1P-4K).

Experimental design was a randomized complete block with four replications of 1 x 3-m plots on both fields. Treatments were the factorial combination of four nitrogen regimes with three bispyribac-sodium rates. Bispyribac-sodium<sup>2</sup> was applied at 0, 74, or 148 g/ha with sequential treatments made after three weeks. Nitrogen was applied in ammonium nitrate solution in four regimes: (1) withholding nitrogen four weeks before bispyribac-sodium treatments, (2) withholding nitrogen two weeks before bispyribac-sodium treatments, (3) increasing nitrogen two weeks before bispyribac-sodium treatments, and (4) biweekly applications of nitrogen before and during bispyribac-sodium treatments. Bispyribac-sodium<sup>1</sup> was applied on June 1 and June 21 in 2005 and May 31 and June 20 in 2006. Nitrogen rates and application dates are presented in Tables 1 and 2. Treatments were applied by making two passes per plot in opposite directions with a single nozzle CO<sub>2</sub> pressured sprayer calibrated to deliver a total 370 L/ha. Nozzles<sup>2</sup> used were 9504E and CO<sub>2</sub> regulators were set for 220 kPa.

Annual bluegrass and creeping bentgrass color was visually rated on a 1 to 9 scale weekly where 1 equaled brown and 9 equaled dark green. Annual bluegrass cover was rated visually on a percent scale three weeks after sequential applications where 0 equaled no cover and 100 equaled complete plot cover. Data were subjected to analysis of variance and significance of main effects was analyzed at the 0.05 probability level. Year by treatment interactions were not detected, thus results were pooled over both years.

**Laboratory Experiments.** Annual bluegrass and ‘L-93’ creeping bentgrass were collected from mature stands at Horticulture Research Farm II in North Brunswick, NJ. Annual bluegrass was an indigenous annual biotype (*Poa annua* var. *annua*). Grasses were planted in pots with 1-dm<sup>2</sup> surface area and 10-cm depths in a greenhouse at the New Jersey Agriculture Greenhouse Research Complex. Greenhouse day/night temperatures were set for approximately 25/19 C and natural light was supplemented with high intensity discharge lighting when intensity fell below 600  $\mu\text{mol}/\text{m}^2/\text{s}$ . Soil medium<sup>3</sup> consisted of 80% Canadian Sphagnum peat moss and a 20% mixture of perlite, vermiculite, dolomitic limestone, and calcitic limestone. Pots were irrigated to prevent plant wilt. Once grasses resumed active growth, pots were placed in growth chambers<sup>4</sup> set for 26/20 C (day/night) with 50% relative humidity. Growth chamber photoperiods were set for 12 hrs of 500  $\mu\text{mol}/\text{m}^2/\text{s}$  total radiation from fluorescent lamps<sup>5</sup> and soft light bulbs<sup>5</sup>. Light bulbs were placed parallel to fluorescent lamps every 15 cm and perpendicular to fluorescent lamps every 41 cm.

Grasses were fertilized weekly with ammonium nitrate solution (34N-0P-0K) at 0 or 24 kg N/ha. Nitrogen rates were chosen from growth differences detected in pilot experiments. Grasses received two broadcast applications of bispyribac-sodium<sup>1</sup> at 148 g ai/ha. Sequential treatments were made two weeks after initial applications. Broadcast applications were made with a CO<sub>2</sub>-pressured spray chamber<sup>6</sup> equipped with an 8002 EVS spray tip<sup>2</sup> calibrated to deliver 370 L/ha. Immediately following sequential broadcast applications, a 2 $\mu\text{L}$  droplet of spotting solution containing a total of 2 kBq of <sup>14</sup>C-bispyribac-sodium [uniformly ring labeled (specific activity, 2.66 MBq/mg; radiochemical purity, 99.1%)] was applied to the second fully expanded leaf with a

microsyringe<sup>7</sup>. Appropriate amount of formulated herbicide<sup>2</sup> was added to spotting solutions to simulate 148 g/ha of bispyribac-sodium at a spray volume of 370 L/ha with a non-ionic surfactant<sup>8</sup> at 0.125% v/v to facilitate deposition. Application rate and regimen were chosen based on pilot experiments to mimic plant responses previously noted in field experiments with bispyribac-sodium (Lycan and Hart 2006b). <sup>14</sup>C-bispyribac-sodium was spotted following the second broadcast application since multiple treatments have been shown necessary for annual bluegrass control (McCarty and Estes 2005; Lycan and Hart 2006a; McDonald et al. 2006).

Treated leaves were excised after seven days and unabsorbed <sup>14</sup>C-bispyribac-sodium was removed by swirling the treated leaf in a 20-ml scintillation vial containing 2 mL of 10% methanol solution for approximately 45 seconds. Upon removal from vials, leaves were rinsed with 2 ml of washoff solution similar to that aforementioned and stored at -30 C until further analysis.

For herbicide extraction, methodology was modified in preliminary experiments from procedures provided by the company (Anonymous 2003). Single leaves were placed in 1.5 mL tubes<sup>9</sup>, ground with liquid nitrogen, and then filled with 0.75 mL of acetonitrile<sup>10</sup>:water (4:1) solution. Tubes were then agitated on a rotary shaker<sup>11</sup> for approximately 30 seconds and placed in water sonication<sup>12</sup> for 45 minutes. Samples were then centrifuged<sup>13</sup> for five minutes and solution was transferred to separate tubes. The procedure was repeated with fresh acetonitrile solution added to grounded tissue and the two 0.75 mL samples were combined in a 1.5 mL tube. To evaluate potential effects of extraction on herbicide degradation, fresh grass leaves were harvested, spotted with 2 µL of <sup>14</sup>C herbicide solution, and processed in conjunction with samples. Extraction

techniques were conducted on freshly spotted leaves so procedure effects could be compared to extraction of  $^{14}\text{C}$  bispyribac-sodium solution without leaf tissue.

A 500  $\mu\text{L}$  sample of extract was spotted on 20 sq cm silica gel thin-layer chromatography (TLC) plates<sup>14</sup> and developed to 16 cm with chloroform<sup>15</sup>:methanol<sup>16</sup>:ammonium hydroxide<sup>17</sup>:water (80:30:4:2, v/v). Plates were air dried, partitioned into eight lanes every 2 cm, and silica gel was scraped every 2 cm (2 x 20-cm sample area) into 10 mL scintillation vials. Vials were filled with scintillation fluid and  $^{14}\text{C}$  was quantified by liquid scintillation spectroscopy<sup>18</sup>. Standard  $R_f$  values were identified on silica gel plates from developed 2  $\mu\text{L}$  of stock radiolabeled herbicide solution with and without ground leaf tissue dissolved in 500 mL of acetonitrile. Parent bispyribac-sodium was identified by comparing values from corresponding  $R_f$  standard. Additional plates with extracts from freshly spotted leaves were compared with pure standard  $^{14}\text{C}$  herbicide to confirm extraction procedures did not cause herbicide degradation.

Two experiments were conducted as a randomized complete block with four replications. Metabolite data consisted of percent parent herbicide, sum percentage of metabolites more polar than parent herbicide, and sum percentage of all metabolites less polar than parent herbicide. Data were analyzed using analysis of variance<sup>19</sup> and means were separated using Fisher's Protected LSD test at  $\alpha = 0.05$ . Experiment by treatment interaction was not detected, and thus results were pooled over experiments.

## RESULTS

**Field Experiments.** Bispyribac-sodium by nitrogen interaction was detected by one week after initial herbicide applications for creeping bentgrass color, but interactions were not significant on other dates (Table 1). Bispyribac-sodium discolored creeping bentgrass one and two weeks after initial applications and one week after sequential applications. Discoloration from bispyribac-sodium increased with rate except two weeks after initial applications.

Creeping bentgrass treated with 48 kg N/ha on May 15 had darker color than all other nitrogen treatments by June 1, the day of initial bispyribac-sodium applications. However, creeping bentgrass treated with 48 kg N/ha on May 15 had significant discoloration (up to 18%) by one week after initial bispyribac-sodium treatments (WAIBT). Creeping bentgrass fertilized at 12 kg N/ha on May 1 plus May 15 or biweekly had 12% or less discoloration from bispyribac-sodium by one WAIBT. Non-bispyribac-sodium treated turf fertilized at 12 kg N/ha on May 1 had lower color ratings one WAIBT than other nitrogen regimes but discoloration from bispyribac-sodium at both rates was minimal (5% or less).

Creeping bentgrass was discolored by bispyribac-sodium by two WAIBT but interaction with nitrogen regime was not detected. Turf fertilized biweekly had darker color by two and three WAIBT than turf fertilized in May only. Bispyribac-sodium discolored creeping bentgrass one week after sequential applications (four WAIBT), which increased with rate, but interaction with nitrogen regime was not detected. Creeping bentgrass fertilized only in May had lower color ratings from four to six WAIBT than turf treated biweekly with 12 kg N/ha.



Table 1. 'L-93' creeping bentgrass color following bispyribac-sodium and nitrogen treatments in two combined field experiments, 2005-2006, North Brunswick, NJ.

Treatment <sup>b</sup>	Application Date <sup>c</sup>	Rate	Creeping Bentgrass Color (WAIBT <sup>a</sup> )						
			0	1	2	3	4	5	6
		kg ai/ha	----- 1 to 9 -----						
Bispyribac-sodium	-	0	-	7.0	6.7	6.4	6.8	6.7	6.3
	6/1 + 6/21	0.074 + 0.074	-	6.4	6.4	6.5	6.4	6.8	6.6
	6/1 + 6/21	0.148 + 0.148	-	6.2	6.4	6.5	6.0	6.7	6.6
Nitrogen	5/1	12	6.7	6.2	6.1	6.0	6.1	6.4	6.2
	5/1 + 5/15	12 + 12	6.8	6.5	6.5	6.1	6.3	6.6	6.3
	5/1 + 5/15	12 + 48	7.9	6.5	6.6	6.5	6.3	6.7	6.5
	5/1 + Biweekly	12	7.2	7.0	6.9	7.3	6.9	7.3	7.1
		Bispyribac-sodium (BS)	NS	*	*	NS	*	NS	*
		Nitrogen (N)	*	*	*	*	*	*	*
		BS x N	NS	*	NS	NS	NS	NS	NS

\* and NS represent significant and not significant at the 0.05 probability level, respectively.

<sup>a</sup>WAIBT = week after initial bispyribac-sodium treatment.

<sup>b</sup>Nitrogen source was (34N-0P-0K) ammonium nitrate.

<sup>c</sup>Application dates for nitrogen treatments were 5/1, 5/15, 6/1, 6/15, and 6/28 in 2005 and 5/1, 5/17, 5/31, 6/14, and 6/26 in 2006. Application dates for bispyribac-sodium treatments were 6/1 and 6/21 in 2005 and 5/31 and 6/20 in 2006.

Annual bluegrass was discolored by bispyribac-sodium on all dates while nitrogen significantly influenced these responses (Table 2). Bispyribac-sodium by nitrogen interaction was not detected from one to three WAIBT for annual bluegrass color. Annual bluegrass discoloration increased with bispyribac-sodium rate from initial applications but efficacy of 74 g/ha appeared to diminish from one to three WAIBT. Averaged over bispyribac-sodium rate, annual bluegrass fertilized with 48 kg N/ha on May 15 had darker color than other nitrogen regimes by one WAIBT. Similarly, biweekly fertilizations increased annual bluegrass color by one and two WAIBT more than 12 kg N/ha applied in May only.

Bispyribac-sodium by nitrogen interactions were detected following sequential herbicide applications for annual bluegrass color by 4 and 5 WAIBT. Non-bispyribac-sodium treated annual bluegrass fertilized biweekly with 12 kg N/ha had darker color following sequential bispyribac-sodium applications than annual bluegrass fertilized in May only. Biweekly nitrogen applications at 12 kg/ha provided higher color ratings (4.7 to 4.9) than other nitrogen regimes (2.4 to 3.6) following sequential applications of bispyribac-sodium at 74 g/ha (4 and 5 WAIBT). However, annual bluegrass had substantial discoloration from sequential bispyribac-sodium applications at 148 g/ha and was not influenced by nitrogen regime by 4 and 5 WAIBT.

Bispyribac-sodium by nitrogen interaction was detected for annual bluegrass cover by six WAIBT. Nitrogen regime influenced annual bluegrass cover from sequential bispyribac-sodium applications at 74 g/ha but not from 148 g/ha (Table 3). Annual bluegrass fertilized once or twice at 12 kg N/ha in May averaged 46 to 52% ground cover after sequential bispyribac-sodium treatments at 74 g/ha. Annual bluegrass

Table 2. Annual bluegrass color following bispyribac-sodium and nitrogen treatments in two combined field experiments, 2005-2006, North Brunswick, NJ.

Treatment <sup>b</sup>	Application Date <sup>c</sup>	Rate	Color (WAIBT <sup>a</sup> )				
			1	2	3	4	5
		kg ai/ha	————— 1 to 9 —————				
Bispyribac-sodium	-	0	6.0	6.1	5.7	6.1	5.7
	6/1 + 6/21	0.074 + 0.074	3.8	4.1	5.1	3.7	3.5
	6/1 + 6/21	0.148 + 0.148	3.0	2.7	3.7	2.3	1.6
Nitrogen	5/1	12	3.6	3.8	4.2	3.6	3.4
	5/1 + 5/15	12 + 12	3.7	3.9	4.3	3.9	3.5
	5/1 + 5/15	12 + 48	5.2	4.9	5.1	4.1	3.2
	5/1 + Biweekly	12	4.6	4.8	5.7	4.6	4.3
		Bispyribac-sodium (BS)	*	*	*	*	*
		Nitrogen (N)	*	*	*	*	*
		BS x N	NS	NS	NS	*	*

\* and NS represent significant and not significant at the 0.05 probability level, respectively.

<sup>a</sup>WAIBT = week after initial bispyribac-sodium treatment.

<sup>b</sup>Nitrogen source was (34N-0P-0K) ammonium nitrate.

<sup>c</sup>Application dates for nitrogen treatments were 5/1, 5/15, 6/1, 6/15, and 6/28 in 2005 and 5/1, 5/17, 5/31, 6/14, and 6/26 in 2006. Application dates for bispyribac-sodium treatments were 6/1 and 6/21 in 2005 and 5/31 and 6/20 in 2006.

Table 3. Annual bluegrass cover following bispyribac-sodium and nitrogen (ammonium nitrate) treatments in two combined field experiments, 2005-2006, North Brunswick, NJ.

Bispyribac-sodium <sup>a</sup> kg ai/ha	Nitrogen Regimes <sup>a</sup>		Annual Bluegrass Cover <sup>b</sup> %
	Application Date	Nitrogen Rate kg ai/ha	
0	5/1	12	73
	5/1 + 5/15	12 + 12	84
	5/1 + 5/15	12 + 48	91
	5/1 + Biweekly	12	90
0.074 + 0.074	5/1	12	52
	5/1 + 5/15	12 + 12	46
	5/1 + 5/15	12 + 48	69
	5/1 + Biweekly	12	21
0.148 + 0.148	5/1	12	6
	5/1 + 5/15	12 + 12	8
	5/1 + 5/15	12 + 48	4
	5/1 + Biweekly	12	11
		LSD <sub>0.05</sub>	8
		Bispyribac-sodium (BS)	*
		Nitrogen (N)	*
		BS x N	*

\* and NS represent significant and not significant at the 0.05 probability level, respectively.

<sup>a</sup>Application dates for nitrogen treatments were 5/1, 5/15, 6/1, 6/15, and 6/28 in 2005 and 5/1, 5/17, 5/31, 6/14, and 6/26 in 2006. Application dates for bispyribac-sodium treatments were 6/1 and 6/21 in 2005 and 5/31 and 6/20 in 2006.

<sup>b</sup>Annual bluegrass cover was rated six weeks after initial bispyribac-sodium treatments.

fertilized at 12 kg N/ha biweekly had 69% cover while 48 kg N/ha applied May 15 had 30% cover after sequential bispyribac-sodium applications at 74 g/ha. Annual bluegrass cover following sequential bispyribac-sodium applications at 148 g/ha ranged 4 to 11% and was not influenced by nitrogen.

**Laboratory Experiments.** One distinct metabolite in both grasses was detected at  $R_f$  0.31 from spotted samples. The  $R_f$  value of bispyribac-sodium was 0.56. Radioactivity with  $R_f$  less than 0.56 was pooled and presented as sum percentage of polar metabolites. Radioactivity with  $R_f$  greater than 0.56 was pooled and presented as sum percentage of non-polar metabolites.

Species by nitrogen interaction was not detected for bispyribac-sodium metabolism, and thus, main effects are presented separately (Table 4). Unfertilized grasses averaged 38% polar metabolites which increased to 57% in fertilized grasses. Parent herbicide recovery averaged 40 and 58% of total radioactivity recovered in fertilized and unfertilized grasses, respectively. Non-polar metabolite recovery was low (less than 5%) and was not influenced by nitrogen fertilization.

Parent herbicide recovered in annual bluegrass and creeping bentgrass averaged 60 and 38% of total radioactivity per species, respectively (Table 4). Conversely, annual bluegrass and creeping bentgrass had 36 and 58% polar metabolites recovered per species, respectively. Differences between species were not detected for non-polar metabolite recovery.

*Table 4.* Annual bluegrass and ‘L-93’ creeping bentgrass metabolites seven days after sequential bispyribac-sodium treatments at 148 g ai/ha in two combined laboratory experiments.

Species	Metabolites (% of <sup>14</sup> C recovered)		
	Polar	Parent	Non-polar
Annual Bluegrass	36	60	4
Creeping Bentgrass	58	38	4
Nitrogen			
None	38	58	4
Weekly	57	40	3
Species	*	*	NS
Nitrogen	*	*	NS
Species by Nitrogen	NS	NS	NS

\* and NS represent significant and not significant at the 0.05 probability level, respectively.

## DISCUSSION

Bispyribac-sodium effectively controls annual bluegrass when appropriately applied in creeping bentgrass fairways but turfgrass cultural practices, such as nitrogen fertilization, may influence efficacy (Askew et al. 2004; Branham and Calhoun 2005; Lycan and Hart 2006a; Park et al. 2002). Annual bluegrass was initially discolored by both bispyribac-sodium rates but began to recover from 74 g/ha before sequential applications. Bispyribac-sodium at 148 g/ha caused significantly greater discoloration than the lower rate but nitrogen fertilization regime did not influence annual bluegrass responses from the high rate. Conversely, nitrogen fertilization appeared to have greater influence on annual bluegrass responses to bispyribac-sodium at 74 g/ha which is probably attributed to less phytotoxicity than the higher rate.

Creeping bentgrass discoloration was most notable from bispyribac-sodium when nitrogen was withheld or increased prior to initial herbicide applications. Creeping bentgrass fertilized biweekly consistently had darker color than other regimes from one to six WAIBT although discoloration was observed following initial and sequential bispyribac-sodium applications. In Maryland, researchers noted tank-mixing bispyribac-sodium with a fertilizer containing nitrogen and iron effectively masked creeping bentgrass chlorosis (McDonald et al. 2006). It was also noted that annual bluegrass control from bispyribac-sodium was similar with and without the fertilizer.

Nitrogen fertility has shown to influence turfgrass tolerance to herbicides applied for grassy weed control. Johnson (1990) noted creeping bentgrass tolerance to ethofumesate was improved by increasing total annual nitrogen applications from 98 to 490 kg/ha. Dernoeden et al. (1993) observed tall fescue (*Festuca arundinacea* Schreb.)

treated with 196 kg N/ha annually had better quality following preemergence herbicide applications compared to turf fertilized at 98 kg N/ha. Johnson (1984) noted bermudagrass (*Cynodon dactylon* (L.) Pers.) injury from MSMA and broadleaf weed herbicides was generally less severe when fertilized compared to unfertilized turf. Bermudagrass recovery from herbicides was also quicker when nitrogen was applied. Johnson and Burns (1985) noted bermudagrass receiving a total 400 kg N/ha annually had less injury from preemergence herbicides than turf fertilized at lower rates.

Nitrogen fertility has shown to influence herbicide tolerance of various weeds. Cathcart et al. (2004) noted green foxtail (*Setaria viridis* (L.) Beauv.) grown under low nitrogen (0.7 mM) required approximately six times more nicosulfuron to reduce biomass by 50% compared to plants under high nitrogen fertility (7.7 mM). Similarly, higher doses of nicosulfuron, glufosinate, mesotrione, and glyphosate were required to achieve 50% reduction in redroot pigweed biomass under low nitrogen. However, nitrogen did not influence efficacy of mesotrione, glufosinate, or atrazine on velvetleaf (*Abutilon theophrasti* Medic.). Johnson and Bowyer (1982) noted large crabgrass (*Digitaria sanguinalis* L.) control with monosodium methanearsonate (MSMA) in Kentucky bluegrass (*Poa pratensis* L.) was not influenced by nitrogen fertilization. However, dandelion populations were lower in untreated turf fertilized at 6 kg N/ha compared to 3 kg N/ha.

Creeping bentgrass appears to metabolize bispyribac-sodium greater than annual bluegrass which may explain differential tolerances levels. In previous experiments, parent herbicide recovered in annual bluegrass, creeping bentgrass, and perennial ryegrass was 73, 32, and 39% of total radioactivity per species, respectively (See Chapter



9). It was also noted that polar metabolites recovered after seven days in annual bluegrass, creeping bentgrass, and perennial ryegrass were 24, 59, and 55% of total radioactivity per species, respectively.

Differential herbicide tolerance in grasses has been correlated to metabolism in previous experiments. Olson et al. (2000) noted differential responses of jointed goatgrass (*Aegilops cylindrical* Host. ), downy brome (*Bromus tectorum* L.), and spring wheat (*Triticum aestivum* L.) to sulfosulfuron was attributed to metabolism. Dubelman et al. (1997) noted corn (*Zea mays* L.) and wheat (*Triticum aestivum* L.) tolerance to halosulfuron was due to rapid metabolism while soybean, a sensitive species, had limited metabolism. In other experiments, selectivity of trifloxysulfuron was also attributed to plant metabolism (Askew and Wilcut 2002).

In translocation experiments, Lycan and Hart (2006b) noted annual bluegrass and creeping bentgrass retained greater than 90% of foliar applied <sup>14</sup>C-bispyribac-sodium in treated leaves. Annual bluegrass also translocated more <sup>14</sup>C-bispyribac-sodium to crowns and leaves than creeping bentgrass. Metabolism of selective annual bluegrass herbicides in cool-season turfgrass have received limited investigation but researchers have investigated metabolism of plant growth regulators used for annual bluegrass suppression. Beasley et al. (2005) noted creeping bentgrass metabolized paclobutrazol more rapidly than annual bluegrass. Thus, species metabolism appears important for selectivity of materials used for annual bluegrass control in cool-season turfgrasses.

Increased herbicide metabolism from nitrogen fertilization was probably associated with responses of both grasses in field experiments. Biweekly fertilizations, compared to withholding nitrogen, reduced creeping bentgrass discoloration from initial

and sequential bispyribac-sodium applications. Similarly, annual bluegrass discoloration and cover reductions from the low bispyribac-sodium rate were less substantial when nitrogen was applied biweekly. Results suggest routine nitrogen fertilization may increase bispyribac-sodium metabolism but selectivity for annual bluegrass control in creeping bentgrass is attributed to differential herbicide metabolism regardless of nitrogen input.

### **Source of Materials**

<sup>1</sup> Velocity®, 80WP herbicide, Valent U.S.A. Corp. PO Box 8025, Walnut Creek, CA 94596.

<sup>2</sup> Tee Jet®, Spraying Systems Co. Wheaton, IL 60189-7900.

<sup>3</sup> Pro-Mix Soil, Premier Horticulture, Quakertown, PA 18951

<sup>4</sup> Environmental Growth Chambers, P.O. Box 407, Chagrin Falls, OH 44022

<sup>5</sup> General Electric Lighting, Chicago, IL 60601.

<sup>6</sup> Research Track Sprayers, Devries Manufacturing, Hollandale, MN 56045.

<sup>7</sup> Microsyringe, Hamilton Co., Reno, NV 89502.

<sup>8</sup> X-77® non-ionic surfactant, Loveland Industries Inc., Greeley, CO 80632.

<sup>9</sup> USA Scientific 1.5 mL microcentrifuge tubes, Fisher Scientific, Fair Lawn, NJ 07410.

<sup>10</sup> Acetonitrile, Pesticide Grade, Fisher Scientific, Fair Lawn, NJ 07410.

<sup>11</sup> Vortex Genie 2™, Fisher Scientific, Fair Lawn, NJ 07410.

<sup>12</sup> Sonicater, Solid State/Ultrasonic FS-28, Fisher Scientific, Fair Lawn, NJ 07410.

<sup>13</sup> Marathon Micro A centrifuge, Fisher Scientific, Fair Lawn, NJ 07410.

- <sup>14</sup> Whatman K5 150 A silica gel adsorption preparative plates, Fisher Scientific, Fair Lawn, NJ 07410.
- <sup>15</sup> Chloroform ECD Tested for Pesticide Analysis, Fisher Scientific, Fair Lawn, NJ 07410.
- <sup>16</sup> Methanol, Histological Grade, Fisher Scientific, Fair Lawn, NJ 07410.
- <sup>17</sup> Ammonium hydroxide, A669-212, Fisher Scientific, Fair Lawn, NJ 07410.
- <sup>18</sup> Model LS3801, Beckman-Coulter, Inc., Fullerton, CA 92834-3100.
- <sup>19</sup> SAS Institute, Cary, NC 27511.

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CHAPTER 4: MOWING HEIGHT INFLUENCES CREEPING BENTGRASS  
TOLERANCE TO BISPYRIBAC-SODIUM

ABSTRACT

Bispyribac-sodium is an efficacious herbicide for annual bluegrass control in creeping bentgrass fairways but turf tolerance and growth inhibition from applications may be exacerbated on closer mowed putting greens. To test this hypothesis, field and greenhouse experiments investigated creeping bentgrass putting green tolerance to bispyribac-sodium. In greenhouse experiments, creeping bentgrass discoloration from bispyribac-sodium was exacerbated by reductions in mowing height from 24 to 3 mm but mowing height did not influence clippings or root weight. In field experiments, discoloration of creeping bentgrass putting greens was greatest from applications of 37 g/ha every ten days compared to 74, 111, or 222 g/ha applied less frequently. Chelated iron effectively masked discoloration of creeping bentgrass putting greens from bispyribac-sodium while trinexapac-ethyl inconsistently masked these effects. Overall, creeping bentgrass putting greens appear more sensitive to bispyribac-sodium than higher mowed turf but chelated iron and trinexapac-ethyl could mask discoloration.

**Nomenclature:** Bispyribac-sodium; Creeping bentgrass, *Agrostis stolonifera* L., 'L-93'.

## INTRODUCTION

Annual bluegrass (*Poa annua* L.) is a problematic weed that reduces aesthetics, functionality, and surface quality of creeping bentgrass (*Agrostis stolonifera* L.) putting greens (Beard 1970). Ball roll distances and putting green uniformity are often disrupted by annual bluegrass patches, seedheads, and differential leaf texture compared to creeping bentgrass (Beard et al. 1978; Lush 1989; McCarty et al. 2005). Agronomically, annual bluegrass has undesirable qualities including poor disease, drought, and wear tolerances that create unsightly patches in creeping bentgrass (Beard et al. 1978; Martin and Wehner 1987; Lush 1989; Johnson and White 1997, 1998). Consequently, putting greens infested with annual bluegrass require more water, fungicides, and intensive management, especially in the summer months, to maintain acceptable quality.

Bispyribac-sodium is a pyrimidinyloxybenzoic herbicide that controls weeds by inhibiting acetolactate synthase (ALS, EC 2.2.1.6), similar to sulfonyleureas (Shimizu et al. 2002). Bispyribac-sodium has been used for selective postemergence control of barnyardgrass (*Echinochloa crus-galli* (L.) Beauv.) and other weeds in rice (*Oryza sativa* L.) (Schmidt et al. 1999; Webster et al. 1999; Williams 1999) but was recently registered for use in creeping bentgrass and perennial ryegrass (*Lolium perenne* L.) fairways (Anonymous 2004). Field research with bispyribac-sodium indicates applications at 60 to 148 g/ha can reduce annual bluegrass populations in creeping bentgrass fairways without significant turf injury (Askew et al. 2004; Branham and Calhoun 2005; Lycan and Hart 2006; Park et al. 2002).

Creeping bentgrass golf greens are generally more sensitive to herbicides than higher mowed turf and excessive injury could exacerbate putting green stress from

mowing, traffic, and diseases (Carrow 1996; McCarty et al. 2005). Plant growth regulators have traditionally been used to suppress annual bluegrass growth on creeping bentgrass golf greens but applications generally do not completely eliminate populations (Johnson and Murphy 1995; Bell et al. 2004). In limited research, bispyribac-sodium has shown potential for selective annual bluegrass control on creeping bentgrass putting greens but turf injury was often excessive (Park et al. 2002; Teuton et al. 2007). Severe turf injury could limit the potential to control annual bluegrass on golf greens, and thus, research is warranted to investigate growth, quality, and performance of creeping bentgrass greens treated with bispyribac-sodium.

The objective of greenhouse experiments was to investigate the influence of mowing height on creeping bentgrass quality, clippings, and root growth following bispyribac-sodium treatments. Subsequently, field experiments were conducted to investigate the use of chelated iron and trinexapac-ethyl to mitigate discoloration of creeping bentgrass putting greens following bispyribac-sodium applications.

## MATERIALS AND METHODS

**Greenhouse Experiments.** Two experiments were conducted at the New Jersey Experimental Greenhouse Research Complex from November 2004 to February 2005 to investigate the influence of mowing height on creeping bentgrass tolerance to bispyribac-sodium. Sod was collected from 'L-93' creeping bentgrass established in fall 2002 at Horticulture Research Farm II in North Brunswick, NJ. Roots were washed free of soil and creeping bentgrass was placed in plastic pots with 23 cm depths and 324 cm<sup>2</sup> surface



areas. Soil medium was 80:20 (v/v) sand:peat moss. Fertilizer (10N:4P:8K) was mixed into the soil at 48 kg N/ha before sod was planted.

Greenhouse day/night temperatures were set for approximately 25/19 C and natural light was supplemented by 400 watt high pressure sodium lamps when intensity fell below 600  $\mu\text{mol}/\text{m}^2/\text{s}$ . The experimental design was a randomized complete block with three replications. Blocks were rotated biweekly and re-randomized to minimize local greenhouse light variation. Treatments were the factorial combination of three mowing heights, 3, 12, or 24 mm with four bispyribac-sodium rates, 0, 74, 148, or 296 g ai/ha.

Pots were irrigated to prevent wilt and turf was mowed three days per week with grass sheers<sup>1</sup>. Creeping bentgrass was established at 12 mm mowing height for three weeks before initial mowing height treatments and fertilized (16N-1P-7K) biweekly at 12 kg N/ha throughout the experiment. Mowing heights were set by placing plywood boards, 12 and 24 mm in width, at the soil surface to gauge height of cut below grass sheers. Turf mowed at 3 mm did not have spacer references. Height accuracy was checked after initial mowings with rulers placed at the soil surface. Creeping bentgrass was mowed three days per week at each height and mowing treatments were initiated two weeks before bispyribac-sodium treatments. Bispyribac-sodium<sup>2</sup> was applied with a spray chamber<sup>3</sup> delivering 370 L/ha with an 8002E nozzle<sup>4</sup>.

Creeping bentgrass color was rated seven, fourteen, and twenty-eight days after treatments on a 0 to 10 scale where 0 equaled brown grass and 10 equaled dark green. Results were converted to percent discoloration from the untreated at each mowing height. Data were subjected to regression analysis and herbicide rates that caused 20%

discoloration were determined. This value was chosen because 20% discoloration may be considered unacceptable by creeping bentgrass managers.

Clippings were harvested approximately 48 hours after previous mowing on ten, twenty, and thirty days after treatment. Clippings were harvested by placing plywood boards at the soil surface for mowing height accuracy. Clippings were oven-dried at 55 C for 72 hours, and then weighed. Roots were harvested from the entire pot thirty days after herbicide applications. Roots were thoroughly washed to remove soil and organic matter, oven-dried at 55 C for 72 hours, and then weighed. Data were subjected to analysis of variance and orthogonal contrasts were performed to determine relationship of responses with bispyribac-sodium. Experiment by treatment interaction was not detected, and thus experiments were combined.

**Field Experiments.** Experiments were conducted on creeping bentgrass putting greens at Horticultural Research Farm II, North Brunswick, NJ from June to August 2006 and 2007. Soil on both fields was a Nixon sandy loam (fine-loamy, mixed, semiactive mesic type Hupludults) with approximately 30 g/kg organic matter and 6.0 pH. 'L-93' creeping bentgrass was established in September 2005 and 2006 for experiments conducted in 2006 and 2007, respectively. Turf was fertilized at 9 kg N/ha biweekly and mowed 5 d/wk at 3.2 mm with reel mowers and clippings collected.

Bispyribac-sodium<sup>2</sup> was applied in four regimens, each totaling 222 g/ha over sixty days, including: 37 g/ha every 10 days, 74 g/ha every 20 days, 111 g/ha every 30 days, or 222 g/ha applied once. These rates were chosen because 222 g/ha total bispyribac-sodium following sequential applications is currently the maximum registered

rate in creeping bentgrass fairways (Anonymous 2004). These four regimens were applied alone, with chelated iron<sup>5</sup>, or with trinexapac-ethyl<sup>6</sup>. Bispyribac-sodium applied at 37, 74, 111, or 222 g/ha was tank-mixed with chelated iron at 0.8, 1.6, 2.4, or 4.8 kg/ha on aforementioned application intervals. Chelated iron rates were chosen based on application intervals and single application recommendations (Anonymous 2005). Trinexapac-ethyl was applied separately from chelated iron treatments at 0 or 40 g ai/ha every ten days with and without all bispyribac-sodium treatments. Trinexapac-ethyl was tank-mixed with bispyribac-sodium on dates when both treatments were applied.

Experimental design was a randomized complete block with four replications of 1 x 3-m plots. Initial treatments were made on May 31, 2006 and June 1, 2007. Treatments were applied by making two passes per plot in opposite directions with a single nozzle CO<sub>2</sub> pressured sprayer calibrated to deliver a total 375 L/ha. Nozzles<sup>7</sup> used were 9504E and CO<sub>2</sub> regulators were set for 220 kPa.

Turf color was visually rated on a 0 to 10 scale where 0 equaled brown and 10 equaled dark green. Turf quality was rated on a 1 to 9 scale where 1 equaled dead turf and 9 equaled uniform, dense turf. Ratings were made every five days. Data were subjected to analysis of variance and means were separated with Fisher's LSD test at  $\alpha = 0.05$ . Treatment by year interactions were not detected, and thus, years were combined.

## RESULTS

**Greenhouse Experiments.** Creeping bentgrass treated with bispyribac-sodium had 35 to 50%, 25 to 35%, and 20 to 30% discoloration after one week at 3, 12, and 24 mm mowing heights, respectively (Figure 1). After two weeks, creeping bentgrass treated

with bispyribac-sodium had approximately 10 to 40%, 10 to 24%, and 10 to 24% discoloration when maintained at 3, 12, and 24 mm heights, respectively. Creeping bentgrass maintained at 12 and 24 mm had 5% or less discoloration from bispyribac-sodium by four weeks after treatment but discoloration ranged up to 15% at the 3 mm mowing height.

From regression analysis, creeping bentgrass maintained at 3, 12, and 24 mm required 50, 76, and 117 g/ha of bispyribac-sodium, respectively, to cause 20% discoloration by one week after treatment (Table 1). Turf maintained at 3, 12 and 24 mm required 141, 240, and 259 g/ha of bispyribac-sodium, respectively, to cause 20% discoloration after two weeks. By four weeks after treatments, creeping bentgrass required greater than 296 g/ha of bispyribac-sodium to have 20% discoloration at all mowing heights. There were no meaningful differences in turf color of untreated turf among mowing heights (data not shown).

Mowing height by bispyribac-sodium interaction was not detected for clipping yield or root weight. Clipping yields were similar across mowing heights and thus results were pooled and presented by herbicide rate. Creeping bentgrass clippings were linearly reduced from the untreated by ten and twenty days after treatment, ranging up to 76 and 40%, respectively. By thirty days, creeping bentgrass clippings had a quadratic relationship with bispyribac-sodium rate. Turf treated with 74 and 148 g/ha had 20 and 13% greater clipping yield than the untreated, respectively, but treatments at 296 g/ha averaged 16% less yield. Bispyribac-sodium rate influenced creeping bentgrass root weight but mowing height was not significant. Creeping bentgrass root weight was quadratically reduced with increased bispyribac-sodium rate, ranging 0 to 19% (Table 2).

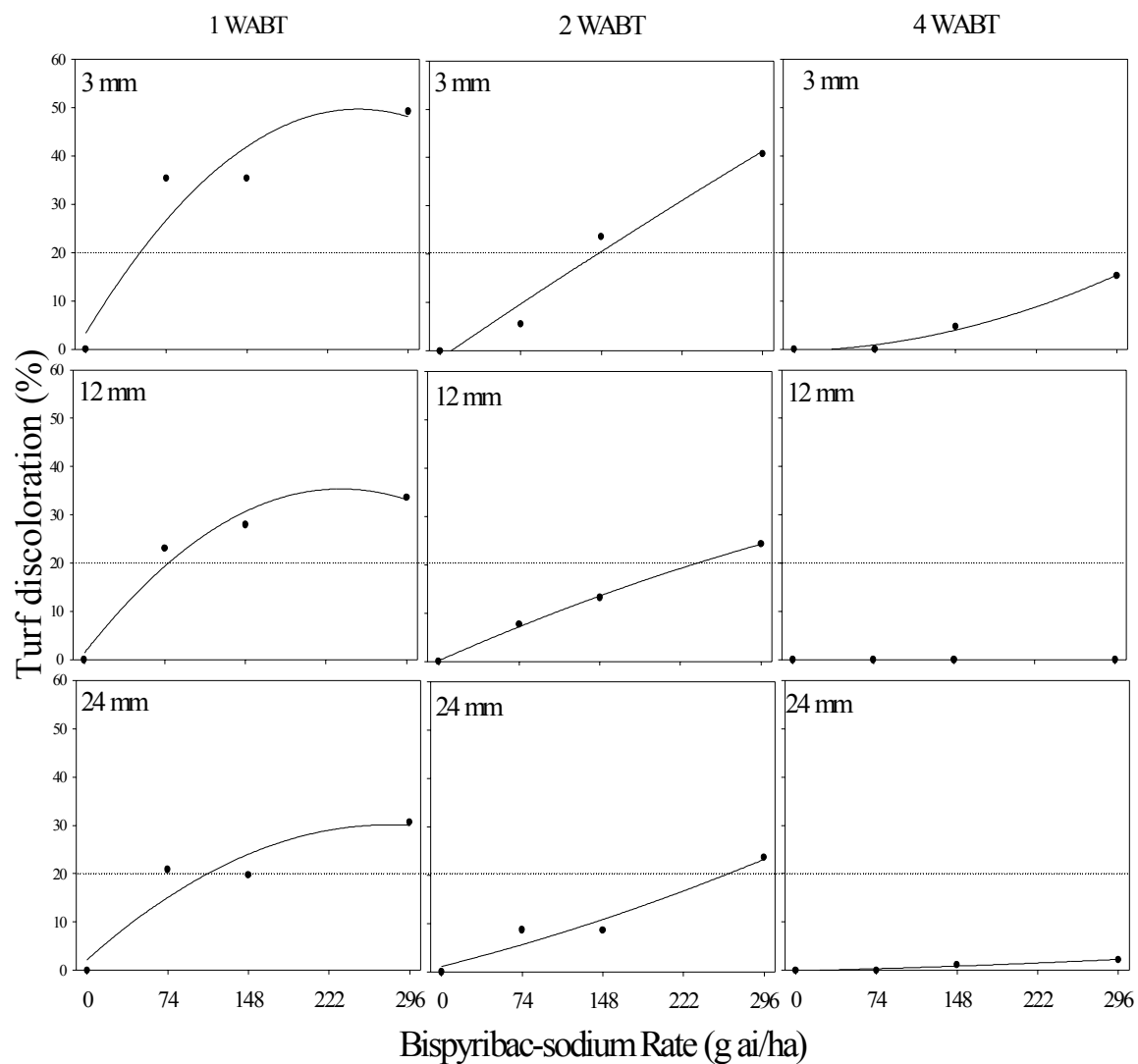


Figure 1. Discoloration of 'L-93' creeping bentgrass treated with bispyribac-sodium and maintained at three mowing heights in two combined greenhouse experiments. WABT = week after bispyribac-sodium treatment.

*Table 1.* Turf discoloration values calculated from regression curves for ‘L-93’ creeping bentgrass maintained at three mowing heights and treated with bispyribac-sodium in two combined greenhouse experiments.

Mowing Height	Turf Discoloration <sub>20</sub> <sup>a</sup>		
	1 WAT <sup>b</sup>	2 WAT	4 WAT
mm	g ai/ ha		
3	50 (± 11)	141 (± 7)	>296 (± 4)
12	76 (± 10)	240 (± 7)	>296 (± 0)
24	117 (± 7)	259 (± 7)	>296 (± 0)

<sup>a</sup>Values are the rate of bispyribac-sodium required to discolor turf by 20% from the untreated determined by the following regression equations where  $x$  = bispyribac-sodium rate in g ai/ha. 1 WAT : (3 mm)  $r^2 = 0.75$ ,  $y = 3.3 + 0.37x - 0.0007x^2$ ; (12 mm)  $r^2 = 0.66$ ,  $y = 1.36 + 0.29x - 0.0006x^2$ ; (24 mm)  $r^2 = 0.69$ ,  $y = 2.15 + 0.2x - 0.0004x^2$ ; 2 WAT : (3 mm)  $r^2 = 0.86$ ,  $y = -1.21 + 0.15x$ ; (12 mm)  $r^2 = 0.61$ ,  $y = 0.83 + 0.08x$ ; (24 mm)  $r^2 = 0.60$ ,  $y = 0.5 + 0.08x$ ; 4 WAT : (3 mm)  $r^2 = 0.76$ ,  $y = -0.4 + 0.01x + 0.0002x^2$ ; (12 mm)  $r^2 = \text{NS}$ ,  $y = 0x$ ; (24 mm)  $r^2 = 0.06$ ,  $y = -0.1 + 0.01x$ . Numbers in parentheses are standard errors.

<sup>b</sup>WAT = week after treatments.

*Table 2.* Relative reductions of clipping yield and root weight of ‘L-93’ creeping bentgrass treated with bispyribac-sodium and pooled over three mowing heights in two combined greenhouse experiments, North Brusniwck, NJ.

Bispyribac-sodium	Clipping yield reduction (DAT <sup>a</sup> )			Root weight reduction (DAT)
	10	20	30	30
g ai/ha	----- % of untreated -----			
0	0	0	0	0
74	37	6	-19	-4
148	54	12	-12	9
296	76	40	16	19
Linear	*	*	*	*
Quadratic	NS	NS	*	*
Cubic	NS	NS	NS	NS

<sup>a</sup>DAT = days after treatment.

**Field Experiments.** Over two years, creeping bentgrass discoloration was most notable from initial bispyribac-sodium applications in June (Table 3). Turf discoloration increased with herbicide rate, ranging 15 to 25% from the untreated, by five and ten days after initial applications. Creeping bentgrass treated with high bispyribac-sodium rates recovered within ten to fifteen days after initial treatments. Creeping bentgrass was discolored on three and four dates from three bispyribac-sodium applications at 74 g/ha and two applications at 111 g/ha, respectively. Applying bispyribac-sodium alone at 37 g/ha every ten days discolored creeping bentgrass on seven dates which was generally most notable by five days after sequential treatments.

Chelated iron significantly alleviated discoloration of creeping bentgrass from bispyribac-sodium (Table 3). Bispyribac-sodium at 37 g/ha applied every ten days with iron only discolored turf on one date. From ten to sixty days after initial treatments, creeping bentgrass color was similar or better than the untreated when chelated iron was tank-mixed with bispyribac-sodium at all rates. Chelated iron effectively reduced discoloration from the two high bispyribac-sodium rates (111 and 222 g ai/ha) but did not completely mask discoloration from the untreated. Iron masked turf discoloration from three bispyribac-sodium applications at 74 g/ha and two applications at 111 g/ha on all dates except five days after initial treatments.

Trinexapac-ethyl applied alone enhanced creeping bentgrass color from the untreated on three dates but these effects were inconsistent and never exceeded 7% improvements (Table 3). Bispyribac-sodium at 37 g/ha applied every ten days with trinexapac-ethyl discolored turf on three dates in early June but discoloration was masked from 25 to 60 days after initial treatments. Trinexapac-ethyl reduced discoloration on



Table 3. 'L-93' creeping bentgrass putting green color following bispyribac-sodium applied with iron or trinexapac-ethyl in field experiments, 2006-2007, North Brunswick, NJ.

Bispyribac-sodium		Turf Color (Days After Initial Treatment)											
Rate	Regimen	5	10	15	20	25	30	35	40	45	50	55	60
g ai/ha		----- 0 to 10 -----											
0	0	7.5	7.1	7.1	7.2	7.4	7.3	6.9	6.9	7.0	7.0	6.9	7.3
37	10 d x 6	6.4	6.8	5.0	6.3	6.5	6.9	6.6	6.9	6.4	6.4	6.4	7.5
74	20 d x 3	6.0	6.9	7.5	7.2	5.8	6.7	6.9	7.0	6.8	6.8	6.9	7.4
111	30 d x 2	5.6	6.6	7.3	7.3	6.7	6.9	6.4	6.8	6.9	6.9	6.9	7.4
222	0 d x 1	5.6	6.3	7.4	7.0	7.7	7.3	6.8	6.9	6.9	6.9	7.1	7.4
Plus Chelated Iron (Tank-Mixed)													
37	10 d x 6	7.4	6.3	6.6	7.2	7.5	7.4	7.4	7.3	7.5	7.5	7.6	7.6
74	20 d x 3	6.9	7.0	7.1	7.3	7.4	7.4	7.5	7.1	7.5	7.5	7.3	7.8
111	30 d x 2	6.8	7.0	7.1	7.3	7.5	7.1	7.5	7.1	7.1	7.1	7.2	7.3
222	0 d x 1	6.3	6.3	7.3	7.4	7.8	7.4	7.4	7.1	7.2	7.2	7.2	7.5
Plus Trinexapac-ethyl (every 10 days)													
0	0	7.6	7.3	7.4	7.2	7.8	7.5	7.3	7.2	7.3	7.3	7.3	7.8
37	10 d x 6	6.8	7.1	5.3	6.6	7.1	7.3	7.3	7.1	7.3	7.3	7.1	7.4
74	20 d x 3	6.5	7.0	7.0	7.4	6.8	7.3	7.4	7.1	7.1	7.1	7.2	7.6
111	30 d x 2	5.8	6.6	7.4	7.3	8.1	7.4	6.2	6.3	7.1	7.1	7.3	7.5
222	0 d x 1	5.5	6.2	7.3	7.2	7.9	7.6	7.5	7.3	7.3	7.3	7.3	7.6
	LSD <sub>0.05</sub>	0.6	0.8	0.5	0.4	0.5	0.3	0.5	0.3	0.4	0.4	0.3	0.3

<sup>a</sup>Chelated iron rate was 0.8, 1.6, 2.4, or 4.8 kg/ha when tank-mixed with bispyribac-sodium at 37, 74, 111, or 222 g/ha, respectively.

<sup>b</sup>Trinexapac-ethyl was applied every ten days at 0 or 0.04 kg/ha.

one date from bispyribac-sodium at 74 and 111 g/ha applied on 20 and 30 day intervals, respectively, but trinexapac-ethyl did not influence turf discoloration from these regimes on other dates. One application of bispyribac-sodium at 222 g/ha alone caused the greatest discoloration (25%) of creeping bentgrass by five and ten days after applications but trinexapac-ethyl did not reduce these effects.

## DISCUSSION

Creeping bentgrass discoloration from bispyribac-sodium was exacerbated by reductions in mowing height in greenhouse experiments suggesting differential tolerances for greens, tees, and fairways. Regardless of mowing height, creeping bentgrass recovered from bispyribac-sodium treatments with acceptable levels of discoloration. Similar discoloration and recovery of higher mowed fairways and roughs have been reported on creeping bentgrass, colonial bentgrass (*Agrostis capillaries* L.), perennial ryegrass, tall fescue (*Festuca arundinacea* (L.) Shreb.), and velvet bentgrass (*Agrostis canina* L.) following bispyribac-sodium applications (Lycan and Hart 2005, 2006; McCarty and Estes 2005; McDonald et al. 2006a).

Bispyribac-sodium caused substantial clipping reductions on creeping bentgrass for ten to twenty days after treatments in the greenhouse. Clipping reductions were consistent over all mowing heights suggesting growth inhibition from bispyribac-sodium could be similar for creeping bentgrass greens, tees, and fairways. Similar clipping weight reductions from bispyribac-sodium have been reported on Kentucky bluegrass (*Poa pratensis* L.), perennial ryegrass, red fescue (*Festuca rubra* L.), and tall fescue (Fagerness and Penner 1998). After thirty days, creeping bentgrass root weight was

reduced from the untreated at high rates (148 and 296 g/ha). Leaf chlorosis and stunted shoot growth from high bispyribac-sodium rates may have inhibited root growth. In field experiments, sequential bispyribac-sodium applications did not exacerbate summer root decline of creeping bentgrass fairways (McCullough and Hart 2008a) but there could be potential for inhibition under controlled greenhouse environments.

In field experiments, creeping bentgrass putting greens were most severely discolored from bispyribac-sodium at 37 g/ha applied every ten days compared to higher rates applied every twenty or thirty days. Although initial discoloration from higher rates was more substantial, creeping bentgrass recovered from exclusive bispyribac-sodium applications after ten days. In Japan, three spring bispyribac-sodium treatments at 60 g/ha applied monthly caused slight discoloration to 'Penncross' creeping bentgrass golf greens after two weeks but turf recovered by four weeks.

In Tennessee (TN), researchers noted weekly bispyribac-sodium applications at 24 g/ha caused 50 to 85% injury to a 'Penncross' creeping bentgrass putting green but dispersing the total 192 g/ha in monthly applications of 48 g/ha caused less than 13% injury (Teuton et al. 2007). Conversely, weekly bispyribac-sodium applications appeared more efficacious than monthly treatments for annual bluegrass control in one of two years. Experiments in TN were initiated on a golf course in April, and thus, potential traffic stress and earlier application timings may have resulted in higher levels of creeping bentgrass phytotoxicity compared to experiments in New Jersey (Lycan and Hart 2006).

Creeping bentgrass tolerance to bispyribac-sodium is highly influenced by application timing, temperature, and soil properties. In field experiments in New Jersey,

creeping bentgrass discoloration from bispyribac-sodium was greater when applied in spring or fall compared to early summer (Lycan and Hart 2006). Similar results were noted in growth chamber experiments where discoloration from bispyribac-sodium was more substantial to creeping bentgrass grown at 10 C compared to 20 or 30 C (McCullough and Hart 2006). Other factors such as soil moisture and foliar absorption influence bispyribac-sodium efficacy (Koger et al. 2007; McCullough and Hart 2008b) which may have also affected creeping bentgrass responses across locations.

Chelated iron appears to effectively mask discoloration of creeping bentgrass golf greens to bispyribac-sodium. Turf color was reduced from the untreated following initial bispyribac-sodium applications at high rates (111 and 222 g/ha) plus chelated iron but discoloration was less substantial than the herbicide alone. From lower rates (37 and 74 g/ha) applied more frequently, creeping bentgrass color was similar or better than the untreated when chelated iron was tank-mixed with bispyribac-sodium on all dates while stand loss was not detected with any treatment.

Chelated iron has shown to mask discoloration from bispyribac-sodium on creeping bentgrass fairways. In Connecticut, researchers noted bispyribac-sodium applied at 74 g/ha discolored creeping and velvet bentgrass maintained at 1.3-cm mowing height for two to three weeks but tank-mixtures with chelated iron masked these effects (McDonald et al. 2006a). In Maryland, chelated iron masked discoloration of creeping bentgrass fairways from sequential bispyribac-sodium applications at 49, 74, or 111 g ai/ha (McDonald et al. 2006b). It was also noted that iron applications did not reduce bispyribac-sodium efficacy for annual bluegrass control.

Trinexapac-ethyl frequently mitigated creeping bentgrass discoloration from bispyribac-sodium but results were not as consistent as chelated iron applications. Conversely, trinexapac-ethyl did not exacerbate creeping bentgrass discoloration from bispyribac-sodium which may be a concern for putting green turf. In previous research on creeping bentgrass fairways, biweekly trinexapac-ethyl applications before and during bispyribac-sodium use, effectively mitigated discoloration from the herbicide without reducing annual bluegrass control (McCullough and Hart 2008c). Trinexapac-ethyl is commonly applied to enhance putting green color, quality, and ball roll and appears compatible with bispyribac-sodium (Fagerness et al. 2000).

Previous investigations with annual bluegrass control on golf greens with bispyribac-sodium are limited but researchers have noted multiple applications are most efficacious (Park et al. 2002; Teuton et al. 2007). In Japan, researchers reported bispyribac-sodium applications in summer followed by preemergence herbicide treatments in fall effectively reduced annual bluegrass populations on creeping bentgrass golf greens (Park et al. 2002). However, inconsistencies with rates and regimens over years have been reported in the United States (Teuton et al. 2007), and thus, further research is needed to refine bispyribac-sodium application programs on creeping bentgrass golf greens. Overall, creeping bentgrass greens may be more sensitive than fairways to bispyribac-sodium but iron and trinexapac-ethyl appear applicable for mitigating turf discoloration from the herbicide.

### Source of Materials

- <sup>1</sup> 3.6 Volt Cordless Grass Shear, Black and Decker®, Towson, MD 21252.
- <sup>2</sup> Velocity®, 80WP, Valent U.S.A. Corp. P. O. Box 8025, Walnut Creek, CA 94596.
- <sup>3</sup> Research Track Sprayers, Devries Manufacturing, Hollandale, MN 56045.
- <sup>4</sup> Tee Jet®, Spraying Systems Co. Wheaton, IL 60189-7900.
- <sup>5</sup> Sprint 330® (chelated iron), Becker Underwood, Inc. Ames, IA 50010.
- <sup>6</sup> Primo® 1EC (trinexapac-ethyl), Syngenta Corp., Greensboro, NC.

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CHAPTER 5: ROUGHSTALK BLUEGRASS (*POA TRIVIALIS*) CONTROL IN  
CREEPING BENTGRASS WITH BISPYRIBAC-SODIUM AND SULFOSULFURON

ABSTRACT

Bispyribac-sodium and sulfosulfuron are registered for roughstalk bluegrass control in creeping bentgrass but comprehensive investigations are limited for long-term control. The objective of these field experiments was to investigate roughstalk bluegrass control with these herbicides on a creeping bentgrass fairway over three years. Bispyribac-sodium was applied twice at 37, 74, or 111 g a.i./ha or thrice at 37 or 74 g/ha. Sulfosulfuron was applied twice or thrice at 6.5, 13, or 26 g a.i./ha or once at 26 g/ha. Creeping bentgrass chlorosis from herbicides was less than 20% by two to three weeks after applications while all treatments generally provided substantial reductions in roughstalk bluegrass cover by late July. However, roughstalk bluegrass regrowth was detected by October in all three years suggesting herbicide applications visually eliminated foliage but did not control vegetative reproductive structures. Overall, bispyribac-sodium and sulfosulfuron effectively suppressed roughstalk bluegrass ground cover in summer months but regrowth during fall months prevented long-term successful control.

**Nomenclature:** Bispyribac-sodium; Creeping bentgrass, *Agrostis stolonifera* L., ‘Penncross’, ‘Seaside’, ‘Southshore’; Roughstalk bluegrass, *Poa trivialis* L.

**Key words:** Chlorosis, efficacy, turfgrass.

## INTRODUCTION

Roughstalk bluegrass (*Poa trivialis* L.) is a problematic perennial weed in cool-season turfgrasses (Haggar 1979; Levy 1998; Hurley 2003). Roughstalk bluegrass has a light green color, medium leaf texture, and prostrate growth habit that reduces turf aesthetics and functionality (Hurley 1997, 2003). More importantly, roughstalk bluegrass has undesirable qualities including poor disease, drought, and wear tolerances that create unsightly patches in golf course turf (Shearman and Beard 1975; Hurley 1982; Hurly and Funk 1985; Peterson et al. 2005). Consequently, golf courses infested with roughstalk bluegrass require more water, fungicides, and intensive management to maintain acceptable turfgrass swards.

Preemergence herbicides do not control perennial plant establishment from vegetative stems and are ineffective for controlling mature plants (McCarty et al. 2005). Researchers have previously applied dalapon, ethofumesate, and fenoxaprop for roughstalk bluegrass control but these postemergence herbicides often have erratic efficacy and cause unacceptable injury to cool-season turfgrasses (Oswald 1980; Mueller-Warrant 1990; Meyer and Branham 2006). Sulfonylurea herbicides, such as chlorsulfuron, formasulfuron, rimsulfuron, and trifloxysulfuron, selectively control overseeded roughstalk bluegrass in bermudagrass (*Cynodon dactylon* x *C. transvaalensis* Burt-Davy) during early summer but most of these herbicides cause substantial injury to cool-season grasses such as Kentucky bluegrass (*Poa pratensis* L.), perennial ryegrass (*Lolium perenne* L.), and tall fescue (*Festuca arundinaceae* L.) (Prostak 1994; Umeda and Towers 2004; McCullough et al. 2006; McCullough and Hart 2008a).

Bispyribac-sodium and sulfosulfuron are acetolactate synthase (ALS, EC 2.2.1.6) inhibitors that were recently registered for roughstalk bluegrass control in creeping bentgrass fairways (Anonymous 2004, 2005). Bispyribac-sodium was previously used for selective postemergence control of barnyardgrass (*Echinochloa crus-galli* (L.) Beauv.) and other weeds in rice (*Oryza sativa* L.) production (Schmidt et al. 1999; Webster et al. 1999; Williams 1999). Sulfosulfuron was previously used in wheat and non-crop areas for control of roughstalk bluegrass, tall fescue, and sedges (*Cyperus* spp.) (Anonymous 2005; Shimizu et al. 2002; Eizenberg et al. 2004; Umeda and Towers 2004). Researchers have noted early summer applications of these herbicides initially discolor creeping bentgrass for seven to ten days but turf generally recovers with acceptable color and quality (Askew et al. 2004; Branham and Calhoun 2005; Lycan and Hart 2006a; McDonald et al. 2006a, b; McCullough and Hart 2008a).

Bispyribac-sodium and sulfosulfuron applications may severely injure roughstalk bluegrass and have shown to visually eliminate plants in turf stands for two to three months. In Virginia, three applications of bispyribac-sodium in summer at 74 g ai/ha controlled roughstalk bluegrass 95% by ten weeks after initial treatments (Askew et al. 2004). In regional experiments throughout the U.S. mid-West, researchers noted three treatments of sulfosulfuron at 26 g ai/ha or four bispyribac-sodium applications at 111 g/ha applied biweekly in summer provided 90 to 100% elimination of roughstalk bluegrass in creeping bentgrass fairways after twelve weeks (Morton and Reicher 2007a). In other regional experiments, researchers noted sequential sulfosulfuron applications at 26 g/ha effectively controlled roughstalk bluegrass in creeping bentgrass fairways and Kentucky bluegrass roughs (Riego et al. 2005; McCullough and Hart 2008b).

While these results appear to have promising implications for temporary desiccation, roughstalk bluegrass has potential for regrowth from vegetative stems in subsequent years. Research suggesting these herbicides control roughstalk bluegrass after two to three months could mislead practitioners applying these herbicides for long-term control or eradication in turf. The objective of these experiments was to investigate efficacy of multi-year bispyribac-sodium and sulfosulfuron treatments for roughstalk bluegrass control in creeping bentgrass fairways.

## MATERIALS AND METHODS

Three experiments were conducted on a fairway at New Jersey National Golf Course in Basking Ridge, NJ from 2005 to 2008. Turf was a blend of ‘Pennncross’, ‘Seaside’, and ‘Southshore’ creeping bentgrass established in fall 1995 on a Wachtung silt loam with a 6.7 pH and 30 g/kg organic matter. Turf was fertilized monthly at approximately 8 kg N/ha and mowed 3 d/wk at 9.5 mm with a reel mower. Indigenous roughstalk bluegrass populations over all plots on day of initial treatments were 29% ( $\pm$  3), 14% ( $\pm$  1), and 11% ( $\pm$  1) in Experiment 1, 2, and 3, respectively.

Experiments 1, 2, and 3 were conducted from June 2005 to October 2006, June 2006 to May 2008, and June 2007 to May 2008, respectively. The three experiments were conducted on separate plots. Experiment 1 application dates were June 10, July 1, and July 19 in 2005 and reapplied to the same plots on June 1, June 26, and July 27 in 2006. Experiment 2 application dates were June 1, June 26, and July 27 in 2006 and reapplied to the same plots on May 30, June 25, and July 24 in 2007. Experiment 3 application dates were May 30, June 25, and July 24 in 2007. All treatment regimens

were initiated on the first date aforementioned with sequential applications made at approximately three week intervals. Bispyribac-sodium<sup>1</sup> was applied twice at 37, 74, or 111 g/ha or thrice at 37 or 74 g/ha. Sulfosulfuron<sup>2</sup> was applied twice or thrice at 6.5 or 13 g/ha and once or twice at 26 g/ha. Sulfosulfuron treatments included a non-ionic surfactant<sup>3</sup> at 0.25% v/v.

Experimental designs were randomized complete blocks with four replications of 1 x 3-m plots. Treatments were applied by making two passes per plot in opposite directions with a single nozzle CO<sub>2</sub> pressured sprayer calibrated to deliver a total 375 L/ha. Nozzles<sup>4</sup> used were 9504E and CO<sub>2</sub> regulators were set for 220 kPa.

Creeping bentgrass discoloration was visually rated on a percent scale where 0 equaled no discoloration from the untreated and 100 equaled brown grass. Turf quality was rated on a 1 to 9 scale where 1 equaled dead turf and 9 equaled ideal, uniform turf. Roughstalk bluegrass cover was visually rated on percent scale before initial applications and monthly throughout experiments. Data were subjected to analysis of variance<sup>5</sup> and means were separated with Fisher's Protected LSD test at  $\alpha = 0.05$ . Experiments are presented separately due to treatment interaction with year.

## RESULTS

**Experiment 1.** Turf treated with herbicides had no meaningful differences in quality compared to the untreated and quality was acceptable (7 or greater) on dates listed in Table 1 (data not shown). Creeping bentgrass discoloration from herbicide treatments was generally minimal (7% or less) on dates listed in Table 1 and meaningful differences were not detected among herbicides, rates, or application regimens (data not shown).

Roughstalk bluegrass cover in untreated plots was 36% on June 10 but declined to 12% by August 15, 2005 (Table 1). However, roughstalk bluegrass in untreated plots increased to 20% cover on October 28. Initial bispyribac-sodium applications at 74 and 111 g/ha reduced roughstalk bluegrass cover to approximately 8% cover on July 1, but differences were not detected between rates. However, initial sulfosulfuron applications at all rates and bispyribac-sodium at 37 g/ha did not reduce roughstalk bluegrass cover from the untreated by July 1. By August 15, all herbicide applications effectively reduced roughstalk bluegrass cover from untreated, averaging 1% cover, except single sulfosulfuron applications at 26 g/ha which averaged 8% cover.

By October 28, 2005 and June 1, 2006, roughstalk bluegrass cover in herbicide treated plots was similar to untreated plots, averaging 13 and 28%, respectively (Table 1). Roughstalk bluegrass cover in untreated plots declined to approximately 10% on July 10 and August 9 but increased to 18% on October 10, 2006. Roughstalk bluegrass cover reductions were inconsistent following single and sequential herbicide applications by July 10 but all herbicide treatments significantly reduced cover to 3% or less by August 9. However, turf treated with herbicides had similar roughstalk bluegrass populations to the untreated, averaging 15% cover, by October 10, 2006.

**Experiment 2.** Turf treated with herbicides had no meaningful differences in quality compared to the untreated (data not shown) and quality was acceptable (7 or greater) on dates listed in Table 2. Creeping bentgrass discoloration from herbicide treatments was generally minimal (5% or less) on dates listed in Table 2 and meaningful differences were not detected among herbicides, rates, or application regimens (data not shown).

Table 1. Roughstalk bluegrass cover in creeping bentgrass following bispyribac-sodium and sulfosulfuron applications in field experiments, 2005-2006, Basking Ridge, NJ.

Treatment	Rate (g ai/ha)	Applications <sup>a</sup>	Roughstalk Bluegrass Cover (%)									
			6/10/05	7/1/05	8/15/05	10/28/05	6/1/06	7/10/06	8/9/06	10/10/06		
Bispyribac-sodium	37	2	29	22	2	13	35	5	2	11		
		3	23	12	1	11	23	0	0	12		
	74	2	24	10	0	9	16	4	1	18		
		3	21	5	0	15	30	3	0	15		
		2	16	10	0	9	20	2	1	12		
Sulfosulfuron	6.5	2	30	12	2	8	28	1	1	13		
		3	31	15	3	13	27	2	1	12		
	13	2	29	13	1	18	47	3	3	19		
		3	28	14	1	18	34	2	2	8		
	26	2	44	20	1	12	29	1	1	12		
Untreated		1	33	15	8	10	29	1	1	30		
			36	30	12	20	28	10	9	18		
		LSD <sub>0.05</sub>	NS	19	6	13	NS	7	4	17		
		Std. Err.	2.6	2.0	0.7	1.3	3.1	0.9	0.5	1.7		

<sup>a</sup> Applications made once, twice, or thrice were applied on 6/10, 6/10 plus 7/1, or 6/10 plus 7/1 plus 7/19 in 2005, respectively, and 6/1, 6/1 plus 6/26, or 6/1 plus 6/26 plus 7/27 in 2006, respectively.

Table 2. Roughstalk bluegrass cover in creeping bentgrass following bispyribac-sodium and sulfosulfuron applications in field experiments, 2006-2007, Basking Ridge, NJ.

Treatment	Rate (g ai/ha)	Applications <sup>a</sup>	Roughstalk Bluegrass Cover (%)									
			6/1/06	7/10/06	8/9/06	10/10/06	5/30/07	7/11/07	8/7/07	10/30/07	5/29/08	
Bispyribac-sodium	37	2	12	2	0	7	7	1	0	13	20	
		3	13	3	2	4	4	1	0	13	15	
	74	2	14	2	0	5	10	1	0	15	15	
Sulfosulfuron	111	3	9	1	2	4	9	1	1	9	18	
		2	8	6	1	9	11	1	1	10	10	
	6.5	2	15	2	1	3	11	2	1	10	11	
Untreated	13	3	21	1	0	5	9	2	3	15	10	
		2	13	1	0	4	11	2	2	18	15	
	26	3	13	1	1	8	11	2	0	17	21	
Untreated		2	12	3	1	11	11	1	1	18	22	
		1	15	4	1	8	11	0	0	13	15	
		14	13	5	7	8	11	3	3	23	15	
		LSD <sub>0.05</sub>	NS	6	8	NS	NS	2	2	NS	NS	
		Std. Err.	1.3	0.6	0.2	0.7	0.7	0.4	0.2	0.9	1.2	

<sup>a</sup> Applications made once, twice, or thrice were applied on 6/1, 6/1 plus 6/26, or 6/1 plus 6/26 plus 7/27 in 2006, respectively, and 5/30, 5/30 plus 6/25, or 5/30 plus 6/25 plus 7/24 in 2007, respectively.



Roughstalk bluegrass cover in untreated plots was reduced from 14% on June 1 to 5% by August 9, 2006 (Table 2). Roughstalk bluegrass in untreated plots increased to approximately 8% cover on October 10, 2006 and May 30, 2007. Initial applications of bispyribac-sodium and sulfosulfuron at all rates reduced roughstalk bluegrass cover to 1 to 6% cover on July 10, 2006 but differences were not detected between rates. By August 9, all herbicide applications effectively reduced roughstalk bluegrass cover from untreated, averaging 1% cover.

By October 10, 2006 and May 30, 2007, roughstalk bluegrass cover in herbicide treated plots was similar to untreated plots (Table 2). Roughstalk bluegrass in untreated plots averaged 11 and 3% cover on July 11 and August 7, respectively, but increased to 23% on October 10, 2007. Roughstalk bluegrass cover was reduced to 3% or less following single and sequential herbicide applications in July and August. However, turf treated with herbicides had similar roughstalk bluegrass cover to the untreated by October 30, 2007 and May 29, 2008 averaging 15 and 16% cover, respectively.

**Experiment 3.** Turf treated with herbicides had no meaningful differences in quality compared to the untreated (data not shown) and quality was acceptable (7 or greater) on dates listed in Table 3. Creeping bentgrass discoloration from herbicide treatments was generally minimal (5% or less) on dates listed in Table 3 and meaningful differences were not detected among herbicides, rates, or application regimens (data not shown).

Roughstalk bluegrass cover in untreated plots was 9 to 11% from May 30 to August 7, 2007 (Table 3). Roughstalk bluegrass in untreated plots averaged 10 and 15% cover on September 25 and October 30, 2007, respectively. Initial applications of

Table 3. Roughstalk bluegrass cover in creeping bentgrass following bispyribac-sodium and sulfosulfuron applications in field experiments, 2007, Basking Ridge, NJ.

Treatment	Rate	Applications <sup>a</sup>	Roughstalk Bluegrass Cover (%)							
			5/30/07	7/11/07	8/7/07	9/25/07	10/30/07	5/29/08		
	g ai/ha									
Bispyribac-sodium	37	2	6	1	0	14	18	15		
		3	8	1	0	5	12	18		
	74	2	15	0	1	7	14	15		
		3	13	1	0	9	14	14		
	111	2	10	5	0	11	13	11		
Sulfosulfuron	6.5	2	10	3	3	10	11	12		
		3	9	5	1	7	18	18		
	13	2	18	4	1	13	20	20		
		3	14	2	1	8	14	14		
	26	2	11	1	1	11	13	13		
Untreated		1	10	1	0	5	10	10		
			9	11	9	10	15	15		
		LSD <sub>0.05</sub>	7	6	4	7	NS	NS		
		Std. Err.	1.1	0.7	0.5	1.1	1.3	1.3		

<sup>a</sup> Applications made once, twice, or thrice were applied on 5/30, 5/30 plus 6/25, or 5/30 plus 6/25 plus 7/24 in 2007.

bispyribac-sodium and sulfosulfuron at all rates reduced roughstalk bluegrass cover to 5% or less cover on July 11, 2007 but differences were not detected between rates and regimens. By August 9, all herbicide applications reduced roughstalk bluegrass cover to 3% or less. However, roughstalk bluegrass cover in herbicide treated plots was similar to untreated by September 25 and October 30, 2007, averaging 9 and 14% cover, respectively. By May 29, 2008, roughstalk bluegrass averaged 15% cover in all plots.

## DISCUSSION

Bispyribac-sodium and sulfosulfuron had substantial activity on roughstalk bluegrass which visually reduced or eliminated populations in creeping bentgrass for two to three months after initial applications which is consistent with previous research. In Virginia, three bispyribac-sodium applications in summer at 74 g/ha controlled roughstalk bluegrass 95% by ten weeks after initial treatments (Askew et al. 2004). In regional experiments throughout the U.S. mid-West, researchers noted three treatments of sulfosulfuron at 26 g/ha or four bispyribac-sodium applications at 74 g/ha applied biweekly in summer provided 90 to 100% elimination of roughstalk bluegrass in creeping bentgrass fairways after twelve weeks (Morton and Reicher 2007). In other regional experiments, researchers noted sequential sulfosulfuron applications at 26 g/ha effectively controlled roughstalk bluegrass in creeping bentgrass fairways and Kentucky bluegrass roughs (Riego et al. 2005; McCullough and Hart 2008b).

Although roughstalk bluegrass leaves were visually eliminated after three months, stems of the plant were probably not controlled from herbicide treatments. Bispyribac-sodium and sulfosulfuron have significant translocation patterns from roots to shoots, and

thus, efficacy for controlling belowground plant tissues may be inhibited by seasonal source to sink patterns of perennial plants (Gallaher et al. 1999; Olson et al. 1999; Lycan and Hart 2006b; Koger et al. 2007). Conversely, herbicide applications in fall may have better translocation to stems and stolons but bispyribac-sodium and sulfosulfuron have shown minimal efficacy on annual and roughstalk bluegrass with this application timing (Askew et al. 2004; Lycan and Hart 2006a). Additionally, creeping bentgrass is more sensitive to these herbicides in fall and turf injury could allow roughstalk bluegrass a competitive growth advantage during periods of active growth (Lycan and Hart 2006a; McCullough and Hart 2006, 2008a).

Inconsistent herbicide efficacy for roughstalk bluegrass may also be attributed to the genetic diversity of roughstalk bluegrass biotypes found in golf course turf (Ahmed et al. 1972; Rajaeskar et al. 2005). Although genetic analyses were not conducted, multiple roughstalk bluegrass biotypes may have been present in the creeping bentgrass fairway which influenced efficacy of single and sequential herbicide applications. In Virginia, bispyribac-sodium applications at 74 g/ha controlled roughstalk bluegrass 95% at one golf course but only 45% control was achieved at another location (Askew et al. 2004). In other experiments, researchers noted bispyribac-sodium and sulfosulfuron reduced cover of seven roughstalk bluegrass cultivars to 6% or less but 'Laser' roughstalk bluegrass was only reduced to 17% ground cover (Morton and Reicher 2007b). Genetic diversity has also been attributed to inconsistent herbicide tolerances of Kentucky bluegrass cultivars to bispyribac-sodium and other ALS inhibitors (Shortell et al. 2005; McCullough et al. 2006).

Bispyribac-sodium and sulfosulfuron may have inconsistent results for long-term roughstalk bluegrass control in creeping bentgrass fairways. However, these herbicides may be valuable tools to superintendents managing significant infestations in cool-season turfgrasses. Creeping bentgrass, Kentucky bluegrass, and perennial ryegrass can be safely reseeded two weeks after a bispyribac-sodium application (Lycan and Hart 2006c; Hart and McCullough 2006). Sulfosulfuron may have longer soil residual activity than bispyribac-sodium but cool-season turfgrasses can be safely reseeded by approximately three weeks after applications (Mitra et al. 2004; Lycan and Hart 2005b; Reicher and Weisenberger 2006). Overall, bispyribac-sodium and sulfosulfuron appear efficacious for roughstalk bluegrass suppression but these herbicides alone may be unreliable for long-term eradication in creeping bentgrass fairways.

#### **Source of Materials**

<sup>1</sup> Velocity®, 80WP herbicide, Valent U.S.A. Corp. PO Box 8025, Walnut Creek, CA 94596.

<sup>2</sup> Certainty®, 75DF herbicide, Monsanto Co., St. Louis, MO 63167.

<sup>3</sup> X-77 non-ionic surfactant, Loveland Industries Inc., Greeley, CO 80632.

<sup>4</sup> Tee Jet® 9504E flat fan nozzles, Spraying Systems Co. Wheaton, IL 60189-7900.

<sup>5</sup> SAS Institute, Cary, NC 27511.

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CHAPTER 6: BISPYRIBAC-SODIUM APPLICATION REGIMES FOR ANNUAL  
BLUEGRASS CONTROL ON CREEPING BENTGRASS PUTTING GREENS

ABSTRACT

Bispyribac-sodium effectively controls annual bluegrass in creeping bentgrass fairways but efficacy on putting greens may be affected by management differences, and thus, application regimes may need to be modified for effective annual bluegrass control. To test this hypothesis, field experiments investigated various bispyribac-sodium application regimens for annual bluegrass control on creeping bentgrass putting greens. Bispyribac-sodium regimes totaling 148, 222, and 296 g/ha controlled annual bluegrass 81, 83, and 91%, respectively, over two years. Pooled over herbicide rates, bispyribac-sodium applied two, three, and six times controlled annual bluegrass 78, 83, and 94%, respectively. Creeping bentgrass discoloration generally increased with total bispyribac-sodium rate while weekly applications discolored turf more frequently than biweekly treatments. After eight weeks, all regimes reduced quality from the untreated, resulting from voids in turf following annual bluegrass control. Overall, bispyribac-sodium appears applicable for use on creeping bentgrass golf greens but turf quality may be compromised following annual bluegrass control.

**Nomenclature:** Bispyribac-sodium; Annual bluegrass, *Poa annua* L.; Creeping bentgrass, *Agrostis stolonifera* L., 'L-93'.

## INTRODUCTION

Annual bluegrass (*Poa annua* L.) is a problematic weed that reduces aesthetics, functionality, and surface quality of creeping bentgrass (*Agrostis stolonifera* L.) putting greens (Beard 1970). Annual bluegrass seedheads and differential leaf texture compared to creeping bentgrass may reduce putting green uniformity and ball roll distances (Beard et al. 1978; Lush 1989; McCarty et al. 2005). Additionally, annual bluegrass has undesirable qualities including poor disease, drought, and wear tolerances that create unsightly patches in creeping bentgrass (Beard et al. 1978; Martin and Wehner 1987; Lush 1989; Johnson and White 1997, 1998). Thus, putting greens infested with annual bluegrass may require more water, fungicides, and intensive management, especially in summer months, to maintain acceptable quality.

Bispyribac-sodium is a pyrimidinyloxybenzoic herbicide that controls weeds by inhibiting acetolactate synthase (ALS, EC 2.2.1.6), similar to sulfonyleureas (Shimizu et al. 2002). Bispyribac-sodium has been used for selective postemergence control of barnyardgrass (*Echinochloa crus-galli* (L.) Beauv.) and other weeds in rice (*Oryza sativa* L.) (Schmidt et al. 1999; Webster et al. 1999) but was recently registered for use in creeping bentgrass and perennial ryegrass (*Lolium perenne* L.) fairways (Anonymous 2004). Field research with bispyribac-sodium indicates applications at 60 to 148 g/ha can reduce annual bluegrass populations in creeping bentgrass fairways without significant turf injury (Askew et al. 2004; Branham and Calhoun 2005; Lycan and Hart 2006; Park et al. 2002).

Creeping bentgrass golf greens are generally more sensitive to herbicides than higher mowed turf and excessive injury could exacerbate stress from mowing, traffic, and

diseases (Carrow 1996; McCarty et al. 2005). Plant growth regulators have traditionally been used to suppress annual bluegrass growth on creeping bentgrass golf greens but applications generally do not completely eliminate populations (Johnson and Murphy 1995; Bell et al. 2004). In limited research, bispyribac-sodium has shown potential for selective annual bluegrass control on creeping bentgrass putting greens but multiple applications were required and turf injury was often excessive (Park et al. 2002; Teuton et al. 2007). Bispyribac-sodium efficacy for annual bluegrass control on putting greens may be affected by closer mowing heights, more frequent mowing, and differential edaphic properties compared to creeping bentgrass fairways. Thus, bispyribac-sodium regimes on putting greens may require modifications from regimes applicable on fairways for effective annual bluegrass control. To test this hypothesis, field experiments investigated bispyribac-sodium application regimens for annual bluegrass control on creeping bentgrass putting greens.

## MATERIALS AND METHODS

Experiments were conducted on creeping bentgrass putting greens at Horticultural Research Farm II, North Brunswick, NJ from May to August in 2007 and 2008. Soil on both fields was a Nixon sandy loam (fine-loamy, mixed, semiactive mesic type Hupludults) with approximately 30 g/kg organic matter and 6.0 pH. 'L-93' creeping bentgrass was established in September 2005 and 2006 for experiments conducted in 2007 and 2008, respectively. Turf was fertilized at 9 kg N/ha biweekly and mowed 5 d/wk at 3.2 mm with reel mowers and clippings collected. Annual bluegrass was transplanted from an indigenous green, with a similar soil to creeping bentgrass, at

Horticultural Research Farm II in April 2007 and 2008 for experiments conducted in respective years. Three annual bluegrass plugs (78 cm<sup>2</sup> by 15 cm) were transplanted approximately 60 cm apart on each plot.

Treatments were the factorial combination of three bispyribac-sodium regimes (two, three, or six applications) at three total rates (148, 222, or 296 g ai/ha). For regimes totaling 148 g/ha, bispyribac-sodium<sup>1</sup> was applied at 74, 49.3, or 24.6 g/ha on two, one, or one week intervals, respectively. For regimes totaling 222 g/ha, bispyribac-sodium was applied at 111, 74, or 37 g/ha on two, two, or one week intervals, respectively. For regimes totaling 296 g/ha, bispyribac-sodium was applied at 148, 98.7, or 49.3 g/ha on two, two, or one week intervals, respectively. Rates were chosen based on the maximum total herbicide rate, 222 g/ha, registered for golf course fairways (Anonymous 2004).

Experimental design was a randomized complete block with four replications of 1 x 3-m plots. Initial treatments were made on May 31, 2006 and June 1, 2007. Treatments were applied by making two passes per plot in opposite directions with a single nozzle CO<sub>2</sub> pressured sprayer calibrated to deliver a total 375 L/ha. Nozzles<sup>2</sup> used were 9504E and CO<sub>2</sub> regulators were set for 220 kPa.

Turf color was visually rated every seven days on a 0 to 10 scale where 0 equaled brown and 10 equaled dark green. Results were converted to percent discoloration from the untreated by the following equation:  $\text{Discoloration} = 100 \times [(\text{Plot}_0 - \text{Plot}_x)/\text{Plot}_0]$ , where Plot<sub>0</sub> was color of untreated turf and Plot<sub>x</sub> was color of treated plots. Turf quality was rated on a 1 to 9 scale where 1 equaled dead turf and 9 equaled uniform, dense turf. Annual bluegrass control was visually rated after eight weeks on each transplanted plug on a percent scale where 0 equaled no control and 100 equaled complete cover reduction.

Data were subjected to analysis of variance and significance of main effects were analyzed at the 0.05 probability level. Year by treatment interactions were not detected, and thus, years were combined.

## RESULTS AND DISCUSSION

Bispyribac-sodium total rate by application number interactions were detected on three dates for creeping bentgrass discoloration (Table 1). Creeping bentgrass discoloration increased with total bispyribac-sodium rate on one and three weeks after initial treatments (WAIT, Table 1). Turf had greater discoloration from regimes with two and three applications compared to six treatments by one WAIT. However, regimes with weekly applications caused more discoloration following sequential treatments by two WAIT than regimes with six applications.

Bispyribac-sodium rate by application number interactions were detected from four to six WAIT for creeping bentgrass discoloration. By four WAIT, discoloration increased with herbicide rate when treated with two or six applications but increased rate did not exacerbate discoloration from regimes with three applications. By five and six WAIT, creeping bentgrass discoloration increased with rate from regimes applied three or six times but not when treated with two applications. Creeping bentgrass was not discolored from any treatments by seven WAIT.

Creeping bentgrass putting greens were more frequently discolored by weekly bispyribac-sodium applications compared to biweekly treatments. However, turf discoloration did not exceed 12% from weekly treatments of 24.6 g/ha while higher rates caused approximately 20% or greater discoloration on several dates. Creeping bentgrass

Table 1. Discoloration of 'L-93' creeping bentgrass putting greens treated with bispyribac-sodium in field experiments, 2007-2008, North Brunswick, NJ.

Total Rate	Treatment <sup>a</sup>		Turf Discoloration (WAIT <sup>b</sup> )						
	Rate	Interval	1	2	3	4	5	6	7
g ai/ha	g ai/ha	weeks	----- % of untreated -----						
148	74 (x 2)	2	17	3	13	2	3	-3	-3
	49.3 (x 3)	1	12	13	20	5	2	-1	-2
	24.6 (x 6)	1	6	5	10	8	12	5	-2
222	111 (x 2)	2	18	4	15	4	0	-3	-1
	74 (x 3)	2	15	2	16	6	27	7	1
	37 (x 6)	1	10	5	15	16	19	13	0
296	148 (x 2)	2	29	5	24	10	2	-3	-4
	98.7 (x 3)	2	15	0	18	6	32	8	-2
	49.3 (x 6)	1	6	13	22	26	28	16	0
LSD <sub>0.05</sub> <sup>c</sup>	Total Rate (TR)		*	NS	*	*	*	*	NS
	Application Number (AN)		*	*	NS	*	*	*	NS
	TR x AN		NS	NS	NS	*	*	*	NS

\*, NS = Significant and not significant at the 0.05 probability level, respectively.

<sup>a</sup>Initial treatments were applied on May 31, 2007 and June 2, 2008.

<sup>b</sup>WAIT = week after initial treatment.

<sup>c</sup>LSD<sub>0.05</sub> values for application number 1 WAIT, total rate 1 WAIT, application number 2 WAIT, and total rate on 3 WAIT was 4, 4, 3, and 4, respectively. Interaction LSD<sub>0.05</sub> values on 4, 5, and 6 WAIT were 6, 5, and 5, respectively.

putting green discoloration was comparable to previous investigations with bispyribac-sodium on higher mowed turf. Discoloration has been reported on creeping bentgrass, perennial ryegrass, and tall fescue (*Festuca arundinacea* (L.) Shreb.) following bispyribac-sodium applications but turf generally recovered after one to two weeks (Lycan and Hart 2005; McCarty and Estes 2005; Lycan and Hart 2006; McDonald et al. 2006).

Bispyribac-sodium total rate by application number interaction was not detected for annual bluegrass control or turf quality, and thus, main effects are presented separately. Annual bluegrass control increased with total rate of bispyribac-sodium after eight weeks (Table 2). Bispyribac-sodium regimes totaling 148, 222, and 296 g/ha controlled annual bluegrass 81, 83, and 91%, respectively. Pooled over herbicide rates, bispyribac-sodium applied two, three, and six times controlled annual bluegrass 78, 83, and 94%, respectively.

Annual bluegrass control was comparable to previous investigations on golf greens and fairways (Park et al. 2002; Teuton et al. 2007). In Japan, researchers reported bispyribac-sodium applications in summer followed by preemergence herbicide treatments in fall effectively reduced annual bluegrass populations on creeping bentgrass golf greens (Park et al. 2002). Inconsistencies with rates and regimens over years have been reported in the United States but bispyribac-sodium applied weekly controlled annual bluegrass more effectively than monthly applications (Teuton et al. 2007).

Annual bluegrass control from bispyribac-sodium left voids in turf and significantly reduced quality from the untreated. Annual bluegrass in untreated plots declined 28% by 8 WAIT which was attributed to summer stress and inhibited growth but



Table 2. Annual bluegrass (AB) control and turf quality of 'L-93' creeping bentgrass putting greens treated with bispyribac-sodium in field experiments, 2007-2008, North Brunswick, NJ.

Total Rate	AB Control <sup>b</sup>	Turf Quality (WAIT <sup>c</sup> )	
	8 WAIT <sup>c</sup>	4	8
g ai/ha	%	----- 1 to 9 -----	
148	81	6.3	5.8
222	83	6.4	5.8
296	91	6.1	5.7
	LSD <sub>0.05</sub>	8	0.3
Application Number			
2	78	6.1	6.0
3	83	6.6	5.8
6	94	6.2	5.5
	LSD <sub>0.05</sub>	8	0.3
Untreated	28	6.9	6.8
Total Rate (TR)	*	NS	NS
Application Number (AN)	*	*	*
TR x AN	NS	NS	NS

\*, NS = Significant and not significant at the 0.05 probability level, respectively.

<sup>a</sup>Initial treatments were applied on May 31, 2007 and June 2, 2008. Bispyribac-sodium was applied in nine treatments of three regimes. For regimes totaling 148 g/ha, bispyribac-sodium was applied at 74, 49.3, or 24.6 g/ha on two, one, or one week intervals, respectively. For regimes totaling 222 g/ha, bispyribac-sodium was applied at 111, 74, or 37 g/ha on two, two, or one week intervals, respectively. For regimes totaling 296 g/ha, bispyribac-sodium was applied at 148, 98.7, or 49.3 g/ha on two, two, or one week intervals, respectively.

<sup>b</sup>Annual bluegrass (10 cm<sup>2</sup> by 15 cm) was transplanted to plots in April of 2007 and 2008. Results are pooled over both years.

<sup>c</sup>WAIT = weeks after initial treatment.

quality ratings were comparable by 4 and 8 WAIT (Table 2; Beard 1970; Beard et al. 1978). Total bispyribac-sodium rate applied did not influence turf quality by four and eight WAIT but application number was significant. By 4 WAIT, turf treated with bispyribac-sodium twice had comparable quality to turf treated six times but reductions were greater than regimes with three applications. By 8 WAIT, turf quality reductions were exacerbated with increased number of bispyribac-sodium applications.

Creeping bentgrass putting green quality reductions resulted from voids in turf following annual bluegrass control rather than bentgrass stand reduction. Additionally, discoloration never exceeded 31% and creeping bentgrass recovered from all treatments unlike reports from experiments in southeastern U.S. In Tennessee, researchers noted weekly bispyribac-sodium applications at 24 g/ha caused 50 to 85% injury to a 'Penncross' creeping bentgrass putting green (Teuton et al. 2007). It was also noted that dispersing the total 192 g/ha in monthly applications of 48 g/ha caused less than 13% injury (Teuton et al. 2007). Experiments in Tennessee were initiated on a golf course in April, and thus, potential traffic stress and earlier application timings may have resulted in higher levels of creeping bentgrass phytotoxicity compared to experiments in New Jersey (Lycan and Hart 2006).

Creeping bentgrass tolerance to bispyribac-sodium is highly influenced by application timing, temperature, and soil properties. In field experiments in New Jersey, creeping bentgrass discoloration from bispyribac-sodium was greater when applied in spring or fall compared to early summer (Lycan and Hart 2006). Similar results were noted in growth chamber experiments where discoloration from bispyribac-sodium was more substantial to creeping bentgrass grown at 10° C compared to 20 or 30°

(McCullough and Hart 2006). Other factors such as soil moisture and foliar absorption influence bispyribac-sodium efficacy (Koger et al. 2007; McCullough and Hart 2008b) which may have also affected creeping bentgrass responses across locations.

Bispyribac-sodium appears applicable for annual bluegrass control when appropriately applied on creeping bentgrass golf greens. However, turf quality was reduced from annual bluegrass control which left open areas in putting green surfaces. Practitioners should assess annual bluegrass populations prior to initiating bispyribac-sodium applications on golf greens especially if voids in turf are unacceptable for player expectation. Creeping bentgrass did not fill in bare areas after eight weeks which may allow the establishment of other weeds and could warrant additional herbicide use. Although less efficacious regimes had reduced levels of annual bluegrass control, practitioners may also incorporate these treatments in management practices to gradually transition annual bluegrass populations from putting greens with less disruption to playing surfaces. Creeping bentgrass may recover quicker over voids in turf by additional management practices, such as aerification, nitrogen applications, or growth regulator use which require further investigation with bispyribac-sodium on putting greens.

#### **Source of Materials**

<sup>1</sup> Velocity®, 80WP herbicide, Valent U.S.A. Corp. PO Box 8025, Walnut Creek, CA 94596.

<sup>2</sup> Tee Jet®, Spraying Systems Co. Wheaton, IL 60189-7900.

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CHAPTER 7: TEMPERATURE INFLUENCES CREEPING BENTGRASS AND  
ANNUAL BLUEGRASS RESPONSE TO BISPYRIBAC-SODIUM

ABSTRACT

Bispyribac-sodium is a postemergence (POST) herbicide that selectively controls annual bluegrass in creeping bentgrass but inconsistent results with seasonal applications are believed to occur from temperature influences on bispyribac-sodium efficacy. Growth chamber experiments investigated three temperature regimes on 'L-93' creeping bentgrass and indigenous annual bluegrass responses to bispyribac-sodium. Annual bluegrass and creeping bentgrass exhibited contrasting responses to bispyribac sodium as temperature increased from 10 to 30° C. Regressions of data after four weeks revealed as temperature increased from 10 to 30°, required bispyribac sodium rates for 50% clipping reduction (CR<sub>50</sub>) of annual bluegrass decreased from 85 to 31 g/ha and required rates for 50% leaf chlorosis decreased from greater than 296 to 98, indicating increased herbicidal efficacy at higher temperatures. In contrast, required bispyribac sodium rates for creeping bentgrass CR<sub>50</sub> increased from 200 to greater than 296 as temperature increased from 10 to 30°. Bispyribac sodium discolored creeping bentgrass 0 to 20% at 20 and 30° and discoloration increased 10 to 50% at 10°. Thus, warmer temperatures (20 and 30°) increase bispyribac sodium efficacy for annual bluegrass control with minimal bentgrass discoloration; however, cooler temperatures (10°) have minimal efficacy on annual bluegrass and increase bentgrass chlorosis.

**Nomenclature:** Bispyribac-sodium; annual bluegrass *Poa annua* L.; creeping bentgrass 'L-93', *Agrostis stolonifera* L.

## INTRODUCTION

Annual bluegrass is one of the most difficult weed species to control in highly maintained creeping bentgrass. Compared to creeping bentgrass, annual bluegrass has a lighter green color, coarser texture, and produces unsightly seedheads which disrupt surface uniformity of golf course greens, tees, and fairways (Beard et al., 1978; Lush, 1989; Sweeney and Danneberger, 1997). Infestations often occur from cultural mismanagement of the turf environment or non-competitiveness of bentgrass cultivars (Beard, 1970). Cultural practices required for bentgrass management such as frequent irrigation, fertilization, and mowing can promote undesirable annual bluegrass populations which may out-compete creeping bentgrass for light, water, and nutrients (Sprague and Burton, 1937). Annual bluegrass has poor heat and drought tolerances, often causing unsightly yellow patches during summer months that reduce aesthetics and functionality of creeping bentgrass turf (Beard, 1973).

Preemergence herbicides generally provide erratic annual bluegrass control and plant growth regulators only suppress its growth (McCarty et al., 2005, McCullough et al., 2005). Controlling established annual bluegrass in cool season turfgrasses has previously been unattainable due to a lack of selective postemergence (POST) herbicides (McCarty et al., 2005). However, a selective POST herbicide, bispyribac-sodium has recently been registered for use in creeping bentgrass at 74 to 111 g ai/ha for selective annual bluegrass control (Anonymous, 2004). Bispyribac-sodium is an acetolactate synthase inhibitor, previously used in rice production for POST control of barnyardgrass, (*Echinochloa crus-galli* (L.) Beauv.), junglerice (*E. colona* (L.) Link), signalgrass (*Brachiaria* spp.), sedges (*Cyperus* spp.) and many other weed species (Shimizu et al.

2002). Field experiments demonstrate bispyribac-sodium applications can selectively control annual bluegrass populations with relative safety to creeping bentgrass (Askew et al., 2004; Fausey, 2004; Lycan and Hart, 2004).

Inconsistencies in annual bluegrass control have been observed, however, with different seasonal bispyribac-sodium application timings. Lycan and Hart (2004) noted bispyribac-sodium controlled annual bluegrass more effectively in summer than fall or spring applications. Conversely, it was noted that creeping bentgrass had greater chlorosis from spring and fall applications compared to summer. Variable results likely occurred from temperature influences on bispyribac-sodium efficacy. To test this hypothesis, growth chamber experiments investigated the influence of three temperatures on creeping bentgrass and annual bluegrass responses to six rates of bispyribac-sodium.

## MATERIALS AND METHODS

Two growth chamber experiments were conducted in 2004 at the New Jersey Experimental Greenhouse Research Complex, New Brunswick, NJ. 'L-93' creeping bentgrass and annual bluegrass were grown in separate pots with a 1 dm<sup>2</sup> surface area and 10 cm depth. Bentgrass was seeded at 48 kg/ha and annual bluegrass was seeded by uniformly distributing seedhead material across the pot. Indigenous annual bluegrass seedheads were harvested at Hort Farm II in North Brunswick, NJ in spring of 2004 and stored in a growth chamber at 8.3° C before seeding. Soil medium consisted of 80% Canadian sphagnum peat moss and a 20% mixture of perlite, vermiculite, dolomitic limestone, and calcitic limestone. Grasses were sufficiently irrigated to prevent plant wilt and mowed at a 1.3 cm height with grass sheers<sup>1</sup> 3 d per wk. Grasses were established



approximately 3 wk after seeding in a greenhouse and then acclimated for one week in respective growth chambers before bispyribac-sodium applications.

Growth chamber temperatures were set at 10, 20, and 30 C with 500  $\mu\text{m}/\text{m}/\text{s}$  of light for 8 hr per d. Pots were arranged in a completely randomized design with four replications. Treatments were the factorial combinations of three temperatures with six bispyribac-sodium rates. Bispyribac-sodium was applied at 0, 37, 74, 148, 222, or 296 g/ha to separate pots with a greenhouse spray chamber<sup>2</sup> delivering 370 L/ha with an 8002E nozzle<sup>3</sup>. Grasses were not clipped or irrigated for 24 hr before or after bispyribac-sodium applications. All possible treatment combinations were used on both species.

Visual quality was rated 2 and 4 WAT on a 1 to 9 scale with 1 equal to completely dead turf and 9 equal to dark green turf. Clippings were harvested approximately 72 hrs after mowing 2 and 4 WAT, oven-dried at 55° C for 72 hrs, and then weighed. Clipping weights were converted to a percentage of clippings from nontreated plants within each temperature regime before statistical analysis. Visual quality results were converted to percent chlorosis relative to untreated plant quality ratings. Data were subjected to regression analysis and fit to a dose equation. From fitted regressions, rates of bispyribac-sodium that caused leaf chlorosis to 20 (LC<sub>20</sub>) and 50% (LC<sub>50</sub>) were determined. These values were chosen because 20% chlorosis to creeping bentgrass may be considered unacceptable by turf managers while 50% annual bluegrass chlorosis indicates significant herbicide activity. Rates of bispyribac-sodium that caused clipping reductions of 50% (CR<sub>50</sub>) were determined from fitted regressions. Data were subjected to analysis of variance with SAS General Linear Model procedure<sup>4</sup>. A combined analysis was conducted to determine significance of experiment repetition,

which was considered random. Data were pooled over experiments since experiment by treatment interaction was not detected.

## RESULTS AND DISCUSSION

Interactions were detected with bispyribac-sodium and temperature in visual quality and clipping weights, and thus results are presented by temperature and species. Visual quality of untreated annual bluegrass was reduced after 4 weeks by 13 and 30% at 20 and 30 C, respectively, relative to 10 C (Table 1). Untreated creeping bentgrass quality was reduced 10% at 30 C relative to turf grown at 10 and 20 C. Clipping yields of untreated annual bluegrass and creeping bentgrass were not significantly different among temperatures (data not shown).

Creeping bentgrass chlorosis ranged 10 to 50% at 10 C but was less than or equal to 20% at 20 and 30 C 4 WAT with BS (Figure 1). Based on  $LC_{20}$  values 4 WAT, 134, 288, and 135 g/ha of bispyribac-sodium required to cause 20% creeping bentgrass chlorosis at 10, 20, and 30 C, respectively (Table 2). Annual bluegrass chlorosis 4 WAT was less than or equal to 20% at 10 C but ranged from 40 to 100% at 20 and 30 C.  $LC_{50}$  values calculated from regression curves indicate greater than 296, 170, and 98 g/ha of bispyribac-sodium were required to cause 50% chlorosis of annual bluegrass 4 WAT at 10, 20, and 30 C, respectively. Creeping bentgrass grown at 20 and 30 C required greater than 296 g/ha of bispyribac-sodium to induce  $LC_{50}$  by 4 WAT while bentgrass grown at 10 C required 281 g/ha.

Creeping bentgrass clippings were reduced approximately 20 to 60% from bispyribac-sodium applied at 10 C (Figure 2). Bentgrass clipping yield reductions ranged

*Table 1.* Visual quality of untreated annual bluegrass and creeping bentgrass .

Species	Temperature C	Visual Quality <sup>a</sup>	
		2 WAT	4 WAT
Annual Bluegrass	10	7.4 a <sup>b</sup>	7.9 a
	20	7.3 a	6.9 b
	30	6.6 a	5.5 c
Creeping Bentgrass	10	7.0 a	6.9 a
	20	7.0 a	6.7 a
	30	6.8 b	6.1 b

<sup>a</sup>Visual quality was rated on a 1 to 9 scale where 1 = dead turf and 9 = ideal, dark green turf.

<sup>b</sup>Different letters indicate a significant difference among species by column at the 0.05 probability level.

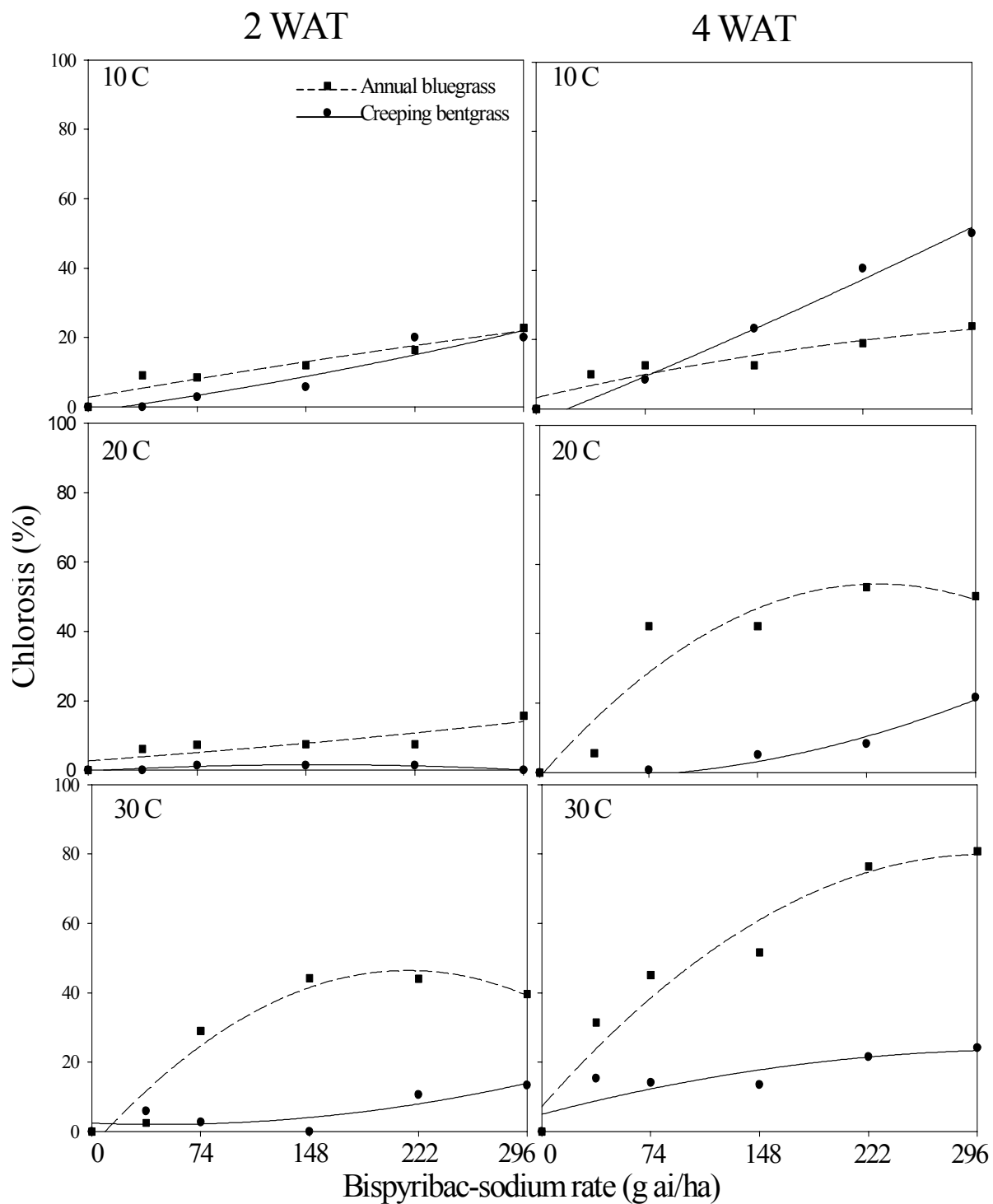


Figure 1. Chlorosis of annual bluegrass and 'L-93' creeping bentgrass grown at three temperatures and treated with bispyribac-sodium in two combined growth chamber experiments. WAT = week after treatments.

0 to 20% and 20 to 40% from bispyribac-sodium applications at 20 and 30 C, respectively.  $CR_{50}$  values indicate 200, >296, and greater than 296 g/ha of bispyribac-sodium were required to reduce creeping bentgrass clippings 50% 4 WAT at 10, 20, and 30 C, respectively (Table 3). Creeping bentgrass receiving bispyribac-sodium at 20 C appears to have less sensitivity and growth reductions compared to 10 and 30 C, likely resulting from better growth following applications at that temperature. Based on clipping yield reductions, creeping bentgrass growth had minimal recovery from 2 to 4 WAT.

Annual bluegrass clipping yield reductions ranged 20 to 60% at 10 C 2 WAT and increased 40 to 80% 4 WAT (Figure 2). In contrast, annual bluegrass clipping yield reductions at 20 C ranged 0 to 20% 2 WAT but increased up to 90% 4 WAT. Similarly at 30 C, bispyribac-sodium reduced clippings 10 to 40% 2 WAT and increased 60 to 100% 4 WAT.  $CR_{50}$  values from regression curves indicate 85, 85, and 31 g/ha of bispyribac-sodium is required to reduce annual bluegrass clippings 50% 4 WAT at 10, 20, and 30 C, respectively (Table 3). However, annual bluegrass grown at 10 C had less than or equal to 20% chlorosis. Results suggest annual bluegrass is susceptible to growth reductions from bispyribac-sodium at all temperatures but higher temperatures (30 C) are required for desiccation.

From these experiments, creeping bentgrass apparently has greater sensitivity to bispyribac-sodium at cooler temperatures (10 C) and greater tolerances at higher temperatures (20 and 30 C). Conversely, annual bluegrass has greater sensitivity to bispyribac-sodium at warmer temperatures (20 and 30 C) and less sensitivity at cooler temperatures (10 C). Lycan and Hart (2004) noted creeping bentgrass had greatest

Table 2. Leaf chlorosis values calculated from regression curves for annual bluegrass and ‘L-93’ creeping bentgrass treated with bispyribac-sodium.

Species	Temperature C	LC <sub>20</sub> <sup>a</sup>		LC <sub>50</sub> <sup>a</sup>	
		2 WAT <sup>b</sup>	4 WAT	2 WAT	4 WAT
		g ai/ha			
Annual Bluegrass	10	>296 (± 1.8)	>296 (± 1.7)	>296 (± 1.8)	>296 (± 1.7)
	20	>296 (± 1.2)	48 (± 4.9)	>296 (± 1.2)	170 (± 4.9)
	30	60 (± 5.5)	28 (± 3.4)	>296 (± 5.5)	98 (± 3.4)
Creeping Bentgrass	10	>296 (± 1.7)	134 (± 4.3)	>296 (± 1.7)	281 (± 4.3)
	20	>296 (± 0.5)	288 (± 2.0)	>296 (± 0.5)	>296 (± 2.0)
	30	>296 (± 2.5)	135 (± 1.9)	>296 (± 2.5)	>296 (± 1.9)

<sup>a</sup>LC<sub>20</sub> and LC<sub>50</sub> values are the amount of bispyribac-sodium required to induce 20 and 50% leaf chlorosis, respectively, from the untreated determined by the following regression equations where  $x$  = bispyribac-sodium rate in g ai/ha. Annual Bluegrass 2 WAT: (10 C)  $r^2 = 0.92$ ,  $y = 2.6115 + 0.0558x + 0.00001x^2$ ; (20 C)  $r^2 = 0.76$ ,  $y = 2.265 + 0.0352x + 0.00001x^2$ ; (30 C)  $r^2 = 0.94$ ,  $y = -1.7688 + 0.5224x - 0.0013x^2$ . Annual Bluegrass 4 WAT: (10 C)  $r^2 = 0.88$ ,  $y = 3.1212 + 0.0978x - 0.0001x^2$ ; (20 C)  $r^2 = 0.89$ ,  $y = -0.7627 + 0.4854x - 0.0011x^2$ ; (30 C)  $r^2 = 0.95$ ,  $y = 4.6164 + 0.5726x - 0.0011x^2$ . Creeping Bentgrass 2 WAT: (10 C)  $r^2 = 0.91$ ,  $y = -1.5049 + 0.0582x + 0.0001x^2$ ; (20 C)  $r^2 = 0.83$ ;  $y = -0.6771 - 0.0536x + 0.0001x^2$ ; (30 C)  $r^2 = 0.71$ ,  $y = 2.9699 + 0.0655x + 0.0004x^2$ . Creeping Bentgrass 4 WAT: (10 C)  $r^2 = 0.94$ ,  $y = -3.3801 + 0.1612x + 0.0001x^2$ ; (20 C)  $r^2 = 0.95$ ,  $y = 0.5612 - 0.0193x + 0.0003x^2$ ; (30 C)  $r^2 = 0.90$   $y = 4.6833 + 0.1407x - 0.0002x^2$ .

<sup>b</sup>WAT = week after treatments.

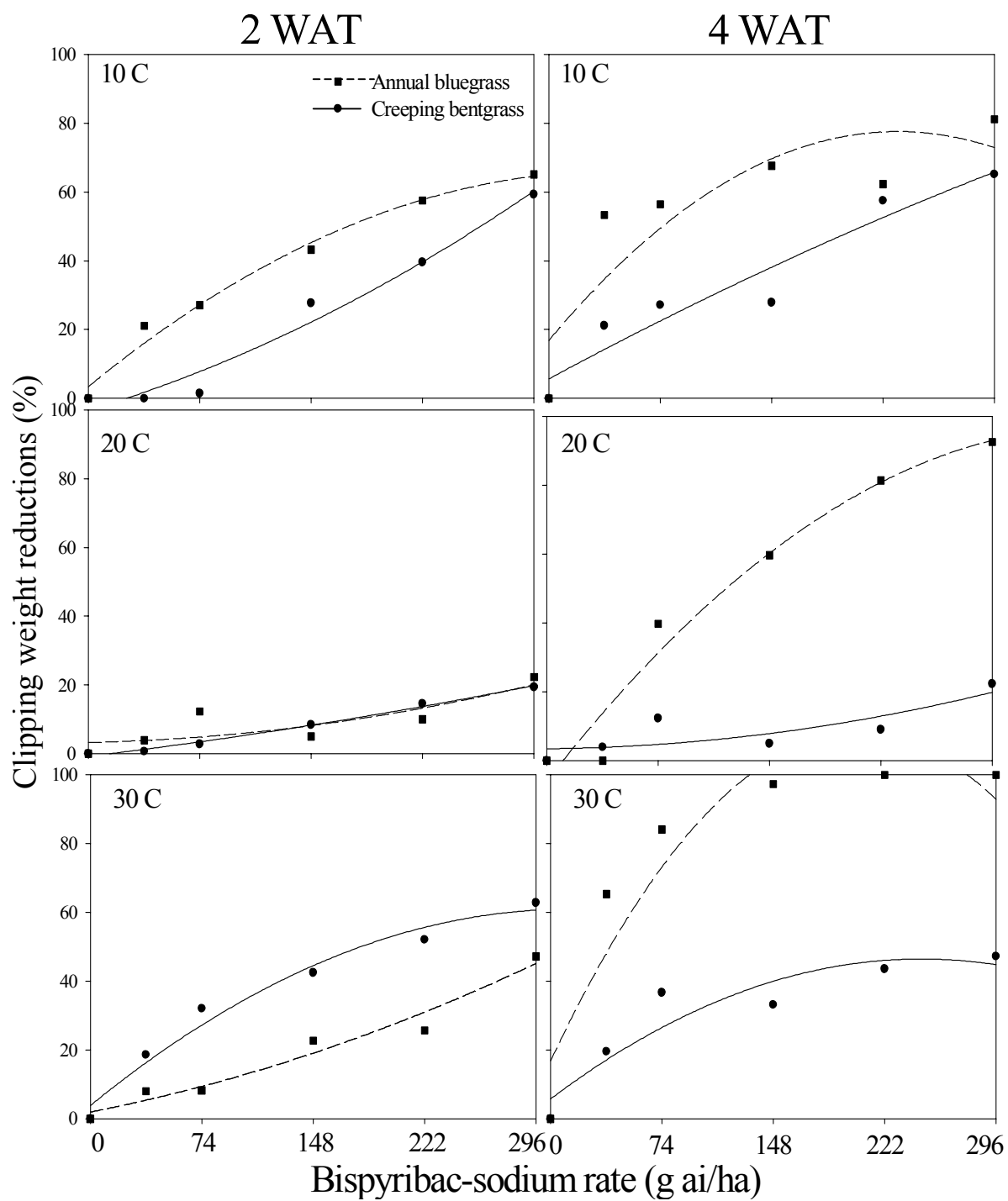


Figure 2. Percent clipping yield reductions relative to untreated creeping bentgrass and annual bluegrass treated with bispyribac-sodium in two combined growth chamber experiments. WAT = week after treatments.

Table 3. Clipping reduction (CR<sub>50</sub>) values calculated from regression curves for annual bluegrass and 'L-93' creeping bentgrass treated with bispyribac-sodium.

Species	Temperature	CR <sub>50</sub> <sup>a</sup>	
		2 WAT <sup>b</sup>	4 WAT
	C	————— g ai/ha —————	
Annual Bluegrass	10	171 (± 6.2)	85 (± 6.7)
	20	>296 (± 7.3)	85 (± 13.2)
	30	>296 (± 6.7)	31 (± 7.5)
Creeping Bentgrass	10	296 (± 6.7)	200 (± 5.3)
	20	>296 (± 2.4)	>296 (± 5.3)
	30	176 (± 6.1)	>296 (± 7.1)

<sup>a</sup>CR<sub>50</sub> value is the amount of bispyribac-sodium required to reduce clippings by 50% from the untreated determined by the following regression equations where  $x$  = bispyribac-sodium rate in g ai/ha. Annual Bluegrass 2 WAT: (10 C)  $r^2 = 0.98$ ,  $y = 5.6401 + 0.2422x - 0.0001x^2$ ; (20 C)  $r^2 = 0.70$ ,  $y = -3.4736 - 0.4721x + 0.0018x^2$ ; (30 C)  $r^2 = 0.96$ ,  $y = 7.5175 + 0.1603x - 0.0001x^2$ . Annual Bluegrass 4 WAT: (10 C)  $r^2 = 0.75$ ,  $y = 16.265 + 0.4814x - 0.0010x^2$ ; (20 C)  $r^2 = 0.96$ ,  $y = 0.6027 + 0.5244x - 0.0007x^2$ ; (30 C)  $r^2 = 0.88$ ,  $y = 18.1352 + 0.9503x - 0.0024x^2$ . Creeping Bentgrass 2 WAT: (10 C)  $r^2 = 0.97$ ,  $y = 2.5176 + 0.1782x - 0.00001x^2$ ; (20 C)  $r^2 = 0.98$ ,  $y = -0.6771 - 0.0536x + 0.0001x^2$ ; (30 C)  $r^2 = 0.97$ ,  $y = 3.3672 + 0.3877x - 0.0007x^2$ . Creeping Bentgrass 4 WAT: (10 C)  $r^2 = 0.94$ ,  $y = 5.6401 + 0.2422x - 0.0001x^2$ ; (20 C)  $r^2 = 0.67$ ,  $y = -3.4736 - 0.4721x + 0.0018x^2$ ; (30 C)  $r^2 = 0.87$ ,  $y = 3.8621 + 0.3675x - 0.0008x^2$ . Numbers in parentheses are standard errors.

<sup>b</sup>WAT = week after treatments.



sensitivity to bispyribac-sodium when applied in spring and fall rather than summer while bispyribac-sodium had minimal efficacy on annual bluegrass when applied in spring and fall but greatest efficacy in summer. Other experiments with acetolactate synthase inhibitors have noted similar results with higher temperatures. Olson et al. (2000) noted control of jointed goatgrass (*Aegilops cylindrica* Host.), cheat (*Bromus secalinus* L.), and downy brome (*Bromus tectorum* L.) with sulfosulfuron was greater at 25/23 C than 5/3 C. The researchers also noted cooler temperatures reduced sulfosulfuron metabolism in all species.

In conclusion, temperature significantly influences bispyribac-sodium efficacy to annual bluegrass and safety to creeping bentgrass. Practitioners will achieve greater annual bluegrass control and minimal bentgrass discoloration at warmer temperatures (20 to 30 C). However, bispyribac-sodium applications significantly injure creeping bentgrass at 10 C and have minimal to no annual bluegrass control with single applications. Applying bispyribac-sodium at 20 C does not injure creeping bentgrass as significantly as 10 C. Further research should investigate bispyribac-sodium absorption, translocation, and metabolism as influenced by various temperatures in creeping bentgrass and annual bluegrass.

#### **Source of Materials**

<sup>1</sup> 3.6 Volt Cordless Grass Shear Model GS300, Black and Decker, Towson, MD 21252.

<sup>2</sup> Research Track Sprayers, Devries Manufacturing, Hollandale, MN 56045.

<sup>3</sup> Tee Jet®, Spraying Systems Co. Wheaton, IL 60189-7900.

<sup>4</sup> SAS Institute, Cary, NC 27511.

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CHAPTER 8: SPRAY ADJUVANTS INFLUENCE BISPYRIBAC-SODIUM  
EFFICACY FOR ANNUAL BLUEGRASS CONTROL IN TURF

ABSTRACT

Field and laboratory experiments were conducted in New Jersey to investigate the influence of spray adjuvants on foliar absorption and efficacy of bispyribac-sodium on annual bluegrass, creeping bentgrass, and perennial ryegrass. In laboratory experiments on annual bluegrass,  $^{14}\text{C}$ -bispyribac-sodium without an adjuvant had 25% foliar absorption by eight hours after treatment while absorption increased to 45, 46, and 75% when applied with crop oil concentrate, non-ionic surfactant, and methylated seed oil, respectively. In creeping bentgrass fairways, sequential bispyribac-sodium applications at 37 g ai/ha with spray adjuvants controlled annual bluegrass similarly to 74 g ai/ha applied sequentially without adjuvants. In perennial ryegrass, treatments with methylated seed oil and non-ionic surfactant required 25 and 41% lower bispyribac-sodium rates, respectively, to obtain annual bluegrass control levels comparable to bispyribac-sodium rates without adjuvants. Spray adjuvants did not exacerbate turfgrass discoloration from bispyribac-sodium. Overall, spray adjuvant use with bispyribac-sodium may allow practitioners to reduce application rates and enhance efficacy for annual bluegrass control.

**Nomenclature:** Bispyribac-sodium; Annual bluegrass, *Poa annua* L.; Creeping bentgrass, *Agrostis stolonifera* L., ‘L-93’, ‘Penncross’; Perennial ryegrass, *Lolium perenne* L., ‘Manhattan 4’.

## INTRODUCTION

Annual bluegrass (*Poa annua* L.) is a major problematic weed in cool-season turfgrasses (Beard 1970). Annual bluegrass reduces turfgrass aesthetics and functionality due to its lighter green color, unsightly seedheads, and shallow root system (Lush 1989; Sprague and Burton 1937). Annual bluegrass tolerates close mowing, germinates rapidly, and has undesirable qualities including poor disease, drought, and wear tolerances that create unsightly patches in turfgrass stands (Beard et al. 1978; Lush 1989; Kaminski and Dernoeden 2002). Consequently, turf infested with annual bluegrass require more water, fungicides, and intensive management than creeping bentgrass, especially in the summer months, to maintain acceptable quality (Sprague and Burton 1937; Beard et al. 1978).

Bispyribac-sodium is a pyrimidinyloxybenzoic herbicide that controls weeds by inhibiting acetolactate synthase (ALS, EC 2.2.1.6), similar to sulfonylureas (Shimizu et al. 2002). Bispyribac-sodium has been used for selective postemergence control of barnyardgrass (*Echinochloa crus-galli* (L.) Beauv.) and other weeds in rice (*Oryza sativa* L.) (Schmidt et al. 1999; Webster et al. 1999; Williams 1999) but was recently registered for postemergence annual bluegrass control in creeping bentgrass and perennial ryegrass. Field research with bispyribac-sodium indicates applications at 60 to 148 g/ha can reduce annual bluegrass populations without significant turf injury (Askew et al. 2004; Branham and Calhoun 2005; Lycan and Hart 2006a; Park et al. 2002). Thus, practitioners have a highly efficacious herbicide for selective annual bluegrass control.

Spray adjuvants are tank-mixed with herbicides to increase absorption through leaf surfaces into plant cells (Wanamarta and Penner 1989). These materials, including crop oils, surfactants, and ammonium based fertilizers, reduce surface tension of spray

droplets to enhance herbicide retention and spread over leaf surfaces (Sun 1996; Sanyal et al. 2006a; Wanamarta and Penner 1989). Spray adjuvants improve herbicide foliar absorption to enhance activity, rainfastness, and reduce antagonism from other applied materials (Ferrell and Vencill 2003; Holshouser and Coble 1990; Jordan et al. 1989; Sanyal et al. 2006a). Spray adjuvants are applied with many ALS-inhibiting herbicides, such as chlorsulfuron, halosulfuron, primisulfuron, rimsulfuron, and sulfosulfuron, to improve efficacy for grassy weed control (Larocque and Christians 1985; Bruce and Kells 1997; Hart 1997; Gallaher et al. 1999; Hutchison et al. 2004; Sanyal et al. 2006b). Similarly, adjuvants may enhance bispyribac-sodium efficacy, like other ALS-inhibitors, and improve herbicide consistency for weed control.

To test this hypothesis, field and laboratory experiments were conducted to investigate the use of spray adjuvants on bispyribac-sodium efficacy for annual bluegrass control in creeping bentgrass and perennial ryegrass.

## MATERIALS AND METHODS

**Laboratory Experiments.** Experiments investigated the influence of spray adjuvants on annual bluegrass foliar absorption of  $^{14}\text{C}$ -bispyribac-sodium. Annual bluegrass was seeded in pots with 1 dm<sup>2</sup> surface area and 10-cm depths in a greenhouse at the New Jersey Agriculture Greenhouse Research Complex. Greenhouse day/night temperatures were set for approximately 25/19 C and natural light was supplemented by 400 watt high pressure sodium lamps when intensity fell below 600  $\mu\text{mol}/\text{m}^2/\text{s}$ . Indigenous annual bluegrass seedheads were harvested at Horticultural Research Farm II in North Brunswick, NJ in spring of 2004 and stored in a growth chamber at 8 C for

approximately one year before seeding. Annual bluegrass was seeded by uniformly distributing seedhead material across pots. Soil medium<sup>1</sup> consisted of 80% Canadian Sphagnum peat moss and a 20% mixture of perlite, vermiculite, dolomitic limestone, and calcitic limestone. Pots were irrigated to prevent plant wilt and mowed at a 1.3-cm height with grass sheers 3 d/wk. Annual bluegrass was allowed to grow and tiller in pots before transplanting individual plants into 20.5 cm deep by 3.8 cm diameter containers. Soil was the same to that aforementioned. Annual bluegrass was allowed to resume active growth in containers prior to herbicide treatments.

Bispyribac-sodium<sup>2</sup> at 74 g ai/ha was applied with no adjuvant, crop oil concentrate<sup>3</sup>, methylated seed oil<sup>4</sup>, or a non-ionic surfactant<sup>5</sup> at 0, 1, 1, or 0.25% vol/vol, respectively. Broadcast treatments were applied with a CO<sub>2</sub>-pressured spray chamber<sup>6</sup> equipped with an 8002 EVS spray tip<sup>7</sup> calibrated to deliver 375 L/ha at 290 kPa. Immediately following broadcast applications, a 2 $\mu$ L droplet of spotting solution containing a total of 0.20 kBq of <sup>14</sup>C-bispyribac-sodium [uniformly ring labeled (specific activity, 2.66 MBq/mg; radiochemical purity, 99.1%)] was applied to the second fully expanded leaf with a microsyringe<sup>8</sup>. Leaf tissue was wet from broadcast treatments when <sup>14</sup>C-bispyribac-sodium was applied. Appropriate amount of formulated herbicide<sup>2</sup> with or without adjuvants was added to spotting solutions to simulate 74 g/ha of bispyribac-sodium at a spray volume of 374 L/ha.

Treated leaves were excised eight hours after treatment for foliar absorption evaluations using methods described in previous experiments (Fagerness and Penner, 1998; Young and Hart, 1998; Lycan and Hart, 2006b). This harvest timing was chosen based on previous investigations with <sup>14</sup>C-bispyribac-sodium absorption by Lycan and

Hart (2006b) that noted significant absorption after eight hours. Unabsorbed  $^{14}\text{C}$ -bispyribac-sodium was removed by swirling the treated leaf in a 20 ml scintillation vial containing 2 mls of 10% methanol solution for approximately 45 s. Upon removal from vials, leaves were rinsed with 2 ml of the same washoff solution to that aforementioned. Unabsorbed  $^{14}\text{C}$  was quantified by liquid scintillation spectroscopy<sup>9</sup>. Foliar absorption was calculated by subtracting amount of  $^{14}\text{C}$  recovered in rinsate from amount of  $^{14}\text{C}$  applied with the following equation:

$$\text{Percent absorption} = 100 - [100 \times (\text{DPM}_0 - \text{DPM}_x) / (\text{DPM}_0)]$$

$\text{DPM}_0$  = DPM (disintegrations per minute) of  $^{14}\text{C}$  in applied solution

$\text{DPM}_x$  = DPM of unabsorbed  $^{14}\text{C}$

Two experiments were conducted as a randomized complete block with four replications. Data were analyzed using analysis of variance and means were separated using Fisher's Protected LSD test at  $\alpha = 0.05$ . Experiment by treatment interaction was not detected, and thus results were pooled over experiments.

**Field Experiment 1.** Experiments were conducted on two perennial ryegrass fields at Horticultural Research Farm II, North Brunswick, NJ from May to July 2006 and 2007 to investigate annual bluegrass control from bispyribac-sodium with spray adjuvants. Soil on both fields was a Nixon sandy loam (fine-loamy, mixed, semiactive mesic type Hupludults) with a pH of 6.3 with approximately 30 g/kg organic matter. 'Manhattan 4' perennial ryegrass was established in fall 2005 and 2006 for experiments conducted in 2006 and 2007, respectively. Fertilizer was applied at approximately 12 kg N/ha biweekly and mowed 3 d/wk at 1.3 cm with a reel mower. Annual bluegrass populations



on day of initial treatments were 39% ( $\pm$  4) and 31% ( $\pm$  8) in 2006 and 2007, respectively. From visual assessments, annual bluegrass biotype was predominately *Poa annua* var. *annua*.

Experimental design was a randomized complete block with four replications of 1 x 3-m plots on both fields. Treatments were applied June 13 and June 28 in 2006 and May 29 and June 11 in 2007. Treatments were applied by making two passes per plot in opposite directions with a single nozzle CO<sub>2</sub> pressured sprayer calibrated to deliver a total 375 L/ha. Nozzles<sup>7</sup> used were 9504E and CO<sub>2</sub> regulators were set for 220 kPa.

Bispyribac-sodium<sup>2</sup> was applied at 0, 37, 74, 111, or 148 g/ha with no adjuvant, methylated seed oil<sup>4</sup>, or non-ionic surfactant<sup>5</sup> at 0, 1, or 0.25 % vol/vol, respectively. Sequential applications were made two weeks after initial treatments (WAIT). Perennial ryegrass injury was rated weekly on a percent scale where 0 equaled no injury and 100 equaled completely dead turf. Percent annual bluegrass cover was visually rated one month after sequential applications where 0 equaled no cover and 100 equaled complete plot cover. Annual bluegrass control was calculated with the following equation:

$$\text{Percent control} = [(Poa_0 - Poa_x)/(Poa_0)] \times 100 \quad [1]$$

$Poa_0$  = annual bluegrass cover of untreated plots

$Poa_x$  = annual bluegrass cover of treated plots

Data were subjected to analysis of variance. Year by treatment interactions were not detected, thus results were pooled over both years. Regression analysis was performed by adjuvant to predict bispyribac-sodium rates that would control annual bluegrass by 75%.

**Field Experiment 2.** Two experiments were conducted on a creeping bentgrass fairway at Riverton Country Club Golf Course, Riverton, NJ, in 2006 and 2007 to investigate annual bluegrass control from bispyribac-sodium with adjuvants. Plots used in 2007 were adjacent to plots used in 2006. Creeping bentgrass was a blend of ‘Penncross’ and ‘L-93’ mowed at 1 cm height 3 d/wk. Soil medium was a Sassafras loamy sand with a pH of 6.5. Turf was irrigated sufficiently to prevent plant wilt and received 12 kg N/ha biweekly. Annual bluegrass populations on day of initial treatments were 26% ( $\pm 2$ ) and 31% ( $\pm 8$ ) in 2006 and 2007, respectively. From visual assessments, annual bluegrass biotype was estimated at half *Poa annua* var. *annua* and half *Poa annua* var. *reptans*. Treatments were applied June 5 and June 22 in 2006 and May 31 and June 14 in 2007. Bispyribac-sodium<sup>2</sup> was applied at 37 or 74 g/ha with and without five spray adjuvant treatments. Adjuvant treatments included no adjuvant, methylated seed oil<sup>4</sup> at 1% vol/vol, non-ionic surfactant<sup>5</sup> at 0.25% vol/vol, methylated seed oil plus ammonium sulfate<sup>10</sup> at 2 kg/L, and non-ionic surfactant plus ammonium sulfate. An untreated check was also included in field plots. Experimental design, application methods, and ratings were made similar to methodology in Field Experiment 1. Year by treatment interaction was not detected for annual bluegrass control, and thus, years were combined. Year by treatment interactions were detected for creeping bentgrass discoloration, and thus, years are presented separately.

## RESULTS AND DISCUSSION

In laboratory experiments, annual bluegrass treated with <sup>14</sup>C-bispyribac-sodium without an adjuvant had 25% foliar absorption by eight hours after treatment (Table 1).

*Table 1.* Annual bluegrass foliar absorption of  $^{14}\text{C}$ -bispyribac-sodium as influenced by spray adjuvants in two combined laboratory experiments.

Adjuvant <sup>a</sup>	Rate (% vol/vol)	Foliar Absorption <sup>b</sup> (%)
None	0	25
Crop Oil Concentrate	1	45
Methylated Seed Oil	1	75
Non-ionic Surfactant	0.25	46
	LSD <sub>0.05</sub>	6

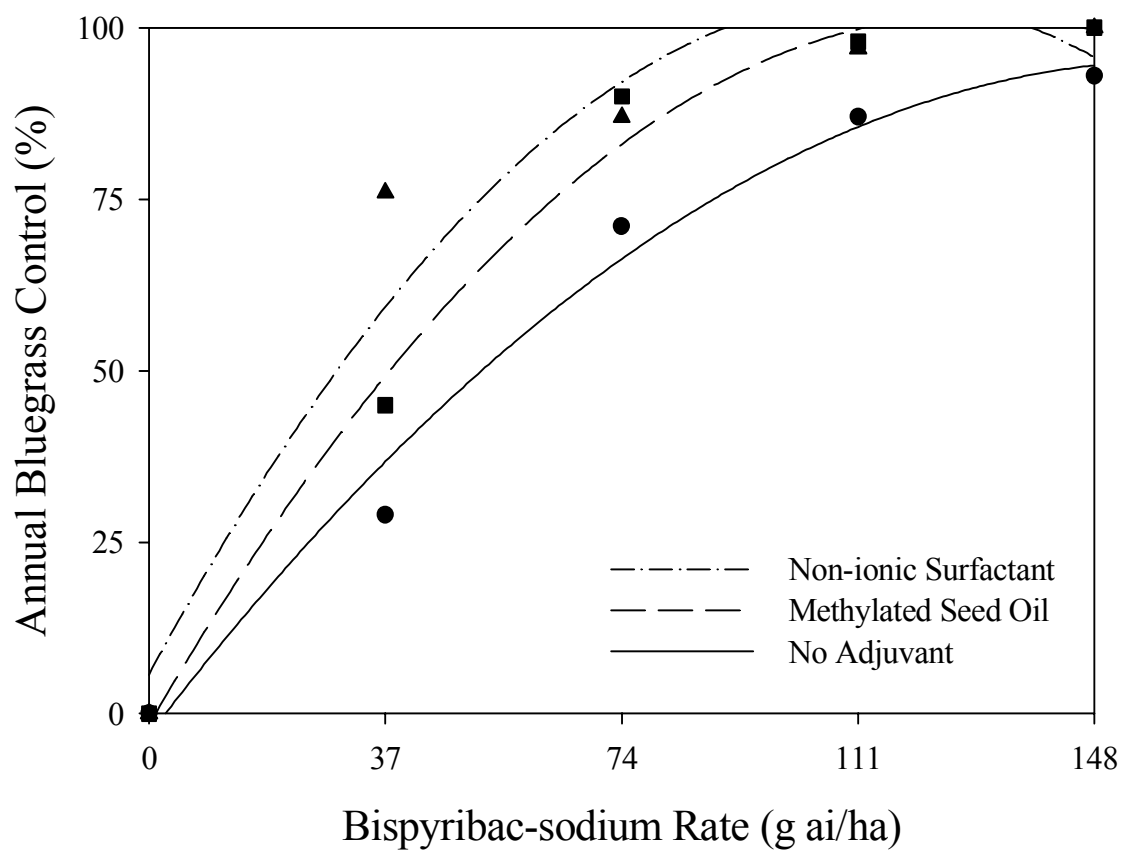
<sup>a</sup>Adjuvants applied were Agri-Dex® crop oil concentrate, Helena Chemical Company, 225 Schilling Blvd., Collierville, TN 38017, methylated seed oil, Destiny methylated soy, Agriliance, P.O. Box 64089, St. Paul, MN 55164-0089, X-77 non-ionic surfactant, Loveland Industries Inc., Greeley, CO 80632.

<sup>b</sup>Bispyribac-sodium was applied at 74 g/ha. Absorption was measured eight hours after treatment and is presented as percentage of applied. Herbicide used was Velocity®, 80WP herbicide, Valent U.S.A. Corp. PO Box 8025, Walnut Creek, CA 94596.

<sup>14</sup>C-bispyribac-sodium absorption increased to 45, 46, and 75% when applied with crop oil concentrate, non-ionic surfactant, and methylated seed oil, respectively. <sup>14</sup>C-bispyribac-sodium foliar absorption levels are comparable to previous investigations by Lycan and Hart (2006b) in cool-season turfgrasses. Koger et al. (2007a) noted applying non-ionic surfactant or methylated seed oil with urea ammonium nitrate enhanced bispyribac-sodium absorption and efficacy on barnyardgrass.

In perennial ryegrass field experiments, annual bluegrass control ranged 29 to 93%, 45 to 100%, and 76 to 100% from five rates of bispyribac-sodium applied with no adjuvant, methylated seed oil, and a non-ionic surfactant, respectively (Figure 1). From regression analysis, predicted bispyribac-sodium rates for 75% annual bluegrass control was 85, 64, and 50 g/ha when applied with no adjuvant, methylated seed oil, and non-ionic surfactant, respectively (Table 2).

In creeping bentgrass experiments, bispyribac-sodium rate by adjuvant interaction was detected for annual bluegrass control (Table 3). Spray adjuvants enhanced annual bluegrass control from bispyribac-sodium at 37 g/ha but not from 74 g/ha. Bispyribac-sodium applied at 37 g/ha controlled annual bluegrass 53% while adjuvants enhanced efficacy similarly to approximately 80% control. Furthermore, bispyribac-sodium applied at 37 g/ha with adjuvants provided similar annual bluegrass control to treatments at 74 g/ha with and without adjuvants. Although bispyribac-sodium at 74 g/ha alone controlled annual bluegrass 83%, spray adjuvants did not improve efficacy of this rate. Ammonium sulfate added to methylated seed oil or non-ionic surfactant treatments did not improve annual bluegrass control from bispyribac-sodium plus adjuvants alone.



*Figure 1.* Annual bluegrass control in perennial ryegrass one month after sequential treatments of bispyribac-sodium with adjuvants in field experiments, 2006-2007, North Brunswick, NJ.

Table 2. Predicted rates of bispyribac-sodium required for annual bluegrass control in perennial ryegrass from regression analysis in field experiments, 2006-2007, North Brunswick, NJ.

Adjuvant	Adjuvant Rate	CL <sub>75</sub> <sup>a</sup> (Bispyribac-sodium <sup>b</sup> Rate)
	% vol/vol	g ai/ha
None	0	85
Methylated Seed Oil	1	64
Non-ionic Surfactant	0.25	50
Equations <sup>b</sup>		
None	$r^2 = 0.78, y = -1.61 + 1.24x - 0.004x^2, SE = 19.4$	
Methylated Seed Oil	$r^2 = 0.90, y = -1.28 + 1.64x - 0.007x^2, SE = 13.7$	
Non-ionic Surfactant	$r^2 = 0.88, y = 6.68 + 1.77x - 0.008x^2, SE = 13.9$	

<sup>a</sup>CL<sub>75</sub> is annual bluegrass control level of 75%. SE = standard error.

<sup>b</sup>Bispyribac-sodium was applied at 0, 37, 74, 111, or 148 g ai/ha with or without adjuvants. Herbicide applied was Velocity®, 80WP herbicide, Valent U.S.A. Corp. PO Box 8025, Walnut Creek, CA 94596. Adjuvants applied were methylated seed oil, Destiny® methylated soy, Agrilience, P.O. Box 64089, St. Paul, MN 55164-0089, X-77® non-ionic surfactant, Loveland Industries Inc., Greeley, CO 80632.

<sup>c</sup>SE = standard error of estimate.

Table 3. Annual bluegrass control in creeping bentgrass with bispyribac-sodium applied with spray adjuvants in field experiments, 2006-2007, Riverton, NJ.

Adjuvant <sup>b</sup>	Bispyribac-sodium Rate (g ai/ha) <sup>a</sup>	
	37 + 37	74 + 74
	----- (Control, %) <sup>c</sup> -----	
None	53	83
MSO	71	84
NIS	85	84
MSO + AMS	85	80
NIS + AMS	75	92
LSD <sub>0.05</sub>	15	
	Rate	*
	Adjuvant	*
	Rate x Adjuvant	*

\*Significant at 0.05 probability level.

<sup>a</sup>Treatments were applied June 5 and June 22 in 2006 and May 31 and June 14 in 2007. Herbicide applied was Velocity®, 80WP herbicide, Valent U.S.A. Corp. PO Box 8025, Walnut Creek, CA 94596.

<sup>b</sup>Methylated seed oil (MSO) was included at 1% volume per volume (vol/vol) of spray solution. Non-ionic surfactant (NIS) was included at 0.25% vol/vol of spray solution. Ammonium sulfate (AMS) was included at 2 kg/L of product. Products applied were methylated seed oil, Destiny® methylated soy, Agrilience, P.O. Box 64089, St. Paul, MN 55164-0089, X-77 non-ionic surfactant, Loveland Industries Inc., Greeley, CO 80632, and ammonium sulfate, Actimaster Soluble Crystal Spray Adjuvant, Platte Chemical, 150 S. Main St., Fremont, NE 68025.

<sup>c</sup>Annual bluegrass control was rated July 13, 2006 and July 12, 2007. Results were pooled over both years.

Results suggest adjuvants have potential to enhance bispyribac-sodium efficacy, and thus, practitioners may reduce rates without compromising annual bluegrass control. In perennial ryegrass, treatments with methylated seed oil and non-ionic surfactant required 25 and 41% less bispyribac-sodium, respectively, to obtain annual bluegrass control levels comparable to treatments without adjuvants. Similarly, bispyribac-sodium at the half rate (37 g/ha) with adjuvants in creeping bentgrass provided similar annual bluegrass control to the registered rate (74 g/ha) without adjuvants.

Bispyribac-sodium is highly efficacious for annual bluegrass control when applied under appropriate environmental conditions (Anonymous 2004; Lycan and Hart 2006a; McCullough and Hart 2006). Lycan and Hart (2006b) noted <sup>14</sup>C-bispyribac-sodium was primarily translocated to shoots suggesting herbicide concentration in leaf tissue is likely associated with herbicide activity. Similar retention has been noted with <sup>14</sup>C-bispyribac-sodium in barnyardgrass leaf tissue (Koger et al. 2007b). Thus, increased foliar absorption of bispyribac-sodium with adjuvants appears beneficial for enhancing control of annual bluegrass and other grassy weeds.

Previous experiments noted similar efficacy improvements of bispyribac-sodium and grassy weed herbicides applied with methylated seed oil and non-ionic surfactants. Koger et al. (2007a) noted bispyribac-sodium applied with non-ionic surfactant or methylated seed oil increased barnyardgrass foliar absorption which reduced rainfast period by 88 and 50%, respectively. Sanvel et al. (2006) noted primisulfuron applied with a non-ionic surfactant had more spread area and retention than without a surfactant on barnyardgrass and green foxtail (*Setaria faberi* Hermm.). Other studies noted methylated seed oil improved foliar absorption and efficacy of imazethapyr,



primisulfuron, quinclorac, rimsulfuron, and several graminicides for grassy weed control (Nalewaja 1986; Hart et al. 1992; Hart and Wax 1996; Zawierucha and Penner 2001; Hutchison et al. 2004).

Ammonium sulfate did not improve efficacy on annual bluegrass when bispyribac-sodium was applied with methylated seed oil and non-ionic surfactant. However, ammonium based fertilizers have shown to improve efficacy and foliar absorption of bispyribac-sodium on other grassy weeds. Koger et al. (2007b) noted urea ammonium nitrate with methylated seed oil and non-ionic surfactant increased foliar absorption four to fivefold compared to treatments without urea ammonium nitrate (Koger et al. 2007b). Hart and Wax (1996) noted ammonium sulfate enhanced <sup>14</sup>C-imazethapyr foliar absorption which prevented antagonism from dicamba on giant foxtail and large crabgrass (*Digitaria sanguinalis* (L.) Scop.).

Bispyribac-sodium by adjuvant interactions were not detected for perennial ryegrass or creeping bentgrass discoloration (Table 4). Perennial ryegrass discoloration increased with bispyribac-sodium rate one week after initial and sequential applications in both years but turf recovered similar to untreated turf after one week. Creeping bentgrass discoloration increased with bispyribac-sodium rate one WAIT in 2006 and one week after sequential treatments in 2007 but recovered one week after applications. Spray adjuvants did not affect creeping bentgrass or perennial ryegrass discoloration from bispyribac-sodium. Similar discoloration and recovery have been reported on creeping bentgrass, perennial ryegrass, and tall fescue (*Festuca arundinacea* (L.) Shreb.) from bispyribac-sodium (Lycan and Hart 2005; McCarty and Estes 2005; Lycan and Hart 2006a; McDonald et al. 2006).

Table 4. Turfgrass discoloration from bispyribac-sodium applied with spray adjuvants in creeping bentgrass and perennial ryegrass field experiments, 2006-2007, Riverton, NJ and North Brunswick, NJ, respectively.

Species	Bispyribac-sodium Rate <sup>b</sup> g ai/ha	Turf Discoloration (WAIT) <sup>a</sup>									
		2006				2007					
		1	2	3	4	1	2	3	4	5	
		%									
Creeping Bentgrass	37	5	2	3	0	8	1	15	6	0	
	74	9	3	5	0	10	1	27	7	0	
	Adjuvant <sup>c</sup>										
	None	9	2	6	0	7	0	16	6	0	
	MSO	8	2	4	0	8	0	23	7	0	
	NIS	6	3	3	0	12	0	19	7	0	
	MSO + AMS	6	2	4	0	11	3	22	8	0	
	NIS + AMS	6	3	5	0	6	3	22	6	0	
	Rate		*	—	NS	—	NS	NS	*	NS	NS
	Adjuvant		— NS —								
Rate x Adjuvant		— NS —									
Perennial Ryegrass	Bispyribac-sodium Rate <sup>b</sup>										
	37	8	0	8	1	1	0	0	0		
	74	10	0	9	3	4	0	1	0		
	111	12	0	14	5	8	0	1	0		
	148	13	0	20	9	12	0	3	0		
	Adjuvant <sup>c</sup>										
	None	8	0	9	2	5	0	1	0		
	MSO	11	0	14	6	5	0	1	0		
	NIS	11	0	12	4	7	0	1	0		
	Rate		*	NS	*	NS	*	NS	*	NS	
Adjuvant		— NS —									
Rate x Adjuvant		— NS —									

(Table 4 continued)

\*Significant at 0.05 probability level.

<sup>a</sup>WAIT = Week after initial treatment.

<sup>b</sup>Treatments were applied June 5 and June 22 in 2006 and May 31 and June 14 in 2007 for creeping bentgrass experiments. Treatments were applied June 13 and June 28 in 2006 and May 29 and June 11 in 2007 for perennial ryegrass experiments.

<sup>c</sup>Methylated seed oil (MSO) was included at 1% volume per volume (vol/vol) of spray solution. Non-ionic surfactant (NIS) was included at 0.25% vol/vol of spray solution. Ammonium sulfate (AMS) was included at 2 kg/L of product. Products applied were methylated seed oil, Destiny® methylated soy, Agriliance, P.O. Box 64089, St. Paul, MN 55164-0089, X-77 non-ionic surfactant, Loveland Industries Inc., Greeley, CO 80632, and ammonium sulfate, Actimaster Soluble Crystal Spray Adjuvant, Platte Chemical, 150 S. Main St., Fremont, NE 68025.

Overall, spray adjuvants have potential to improve bispyribac-sodium efficacy for annual bluegrass control in cool-season turfgrasses. Improved efficacy is likely attributed to increased foliar penetration of bispyribac-sodium in grass tissues. Selective annual bluegrass control from bispyribac-sodium may result from differential translocation (Lycan and Hart 2006a) or metabolism compared to creeping bentgrass and perennial ryegrass. Mechanisms of bispyribac-sodium selectivity in cool-season turfgrasses have received limited investigation and warrant further research.

### **Source of Materials**

<sup>1</sup> Pro-Mix Soil, Premier Horticulture, Quakertown, PA 18951.

<sup>2</sup> Velocity®, 80WP, Valent U.S.A. Corp. P. O. Box 8025, Walnut Creek, CA 94596.

<sup>3</sup> Agri-Dex® crop oil concentrate, Helena Chemical Company, 225 Schilling Blvd., Collierville, TN 38017.

<sup>4</sup> Destiny® methylated soy adjuvant, Agriliance, P.O. Box 64089, St. Paul, MN 55164-0089.

<sup>5</sup> X-77® non-ionic surfactant, Loveland Industries Inc., Greeley, CO 80632.

<sup>6</sup> Research Track Sprayers, Devries Manufacturing, Hollandale, MN 56045.

<sup>7</sup> Tee Jet®, Spraying Systems Co. Wheaton, IL 60189-7900.

<sup>8</sup> Microsyringe, Hamilton Co., Reno, NV 89502

<sup>9</sup> Model LS3801, Beckman-Coulter, Inc., Fullerton, CA 92834-3100.

<sup>10</sup> Ammonium sulfate, Actimaster Soluble Crystal Spray Adjuvant, Platte Chemical, 150 S. Main St., Fremont, NE 68025.

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CHAPTER 9: BISPYRIBAC-SODIUM METABOLISM IN ANNUAL BLUEGRASS,  
CREEPING BENTGRASS, AND PERENNIAL RYEGRASS

ABSTRACT

Bispyribac-sodium selectively controls annual bluegrass in creeping bentgrass and perennial ryegrass which may be attributed to differential metabolism among species. To test this hypothesis, experiments investigated metabolism of  $^{14}\text{C}$ -bispyribac-sodium in annual bluegrass, creeping bentgrass, and perennial ryegrass. Creeping bentgrass and perennial ryegrass metabolized  $^{14}\text{C}$ -bispyribac-sodium by approximately 50% after one day while annual bluegrass metabolized less than 20%. After seven days, parent herbicide recovered in annual bluegrass, creeping bentgrass, and perennial ryegrass was 73, 32, and 39% of total radioactivity per species, respectively. Polar metabolites recovered after seven days in annual bluegrass, creeping bentgrass, and perennial ryegrass were 24, 59, and 55% of total radioactivity per species, respectively. Half life of  $^{14}\text{C}$ -bispyribac-sodium in annual bluegrass, creeping bentgrass, and perennial ryegrass was estimated at greater than seven days, one day, and two days, respectively. Results support the hypothesis that differential tolerances of these grasses are attributed to herbicide metabolism.

**Nomenclature:** Bispyribac-sodium; annual bluegrass, *Poa annua* L.; Creeping bentgrass, *Agrostis stolonifera* L., 'L-93'; Perennial ryegrass, *Lolium perenne* L., 'Manhattan 4'.



## INTRODUCTION

Annual bluegrass (*Poa annua* L.) is a problematic weed in creeping bentgrass and perennial ryegrass turf (Beard, 1970). Annual bluegrass reduces turfgrass aesthetics and functionality due to its lighter green color, unsightly seedheads, and shallow root system (Lush 1989; Sprague and Burton 1937). Annual bluegrass tolerates close mowing, germinates rapidly, and has undesirable qualities including poor disease, drought, and wear tolerances that create unsightly patches in turfgrass stands (Beard et al. 1978; Lush 1989; Kaminski and Dernoeden 2002). Consequently, turfgrasses infested with annual bluegrass require more water, fungicides, and intensive management, especially in the summer months, to maintain acceptable quality.

Bispyribac-sodium is a pyrimidinyl carboxy herbicide that controls weeds by inhibiting acetolactate synthase (ALS, EC 2.2.1.6), similar to sulfonylureas (Shimizu et al. 2002). Bispyribac-sodium has been used for selective postemergence control of barnyardgrass (*Echinochloa crus-galli* (L.) Beauv.) and other weeds in rice (*Oryza sativa* L.) (Schmidt et al. 1999; Webster et al. 1999; Williams 1999) but was recently registered for use in creeping bentgrass and perennial ryegrass (Anonymous 2004). Field experiments have noted bispyribac-sodium applications at 60 to 148 g/ha can control annual bluegrass populations without significant injury to creeping bentgrass and perennial ryegrass (Askew et al. 2004; Branham and Calhoun 2005; Lycan and Hart 2006a; Park et al. 2002). Thus, practitioners have a highly efficacious herbicide for selective annual bluegrass control.

Physiological responses to bispyribac-sodium among grass species may attribute to selectivity for annual bluegrass control in turf. Lycan and Hart (2006b) noted annual

bluegrass and creeping bentgrass retained greater than 90% of foliar applied  $^{14}\text{C}$ -bispyribac-sodium in treated leaves but annual bluegrass translocated more herbicide to crowns. It was also noted annual bluegrass translocated more foliar and root applied  $^{14}\text{C}$ -bispyribac-sodium to leaves than creeping bentgrass.

Although herbicide translocation may contribute to efficacy, differential metabolism has been associated with selectivity of ALS-inhibiting herbicides. Olson et al. (2000) noted differential responses of jointed goatgrass (*Aegilops cylindrical* Host. ), downy brome (*Bromus tectorum* L.), and spring wheat (*Triticum aestivum* L.) to sulfosulfuron was attributed to differences in metabolism. Dubelman et al. (1997) noted corn (*Zea mays* L.) and wheat tolerance to halosulfuron was due to rapid metabolism while soybean, a sensitive species, had limited metabolism. In other experiments, selectivity of imazethapyr, primisulfuron, and trifloxysulfuron was attributed to species differential metabolism (Cole et al. 1989; Gallaher et al. 1999; Askew and Wilcut 2002). Thus, differential metabolism of bispyribac-sodium by grasses may be associated with herbicide selectivity for annual bluegrass control in cool-season turf. To test this hypothesis, experiments were conducted to investigate bispyribac-sodium metabolism in annual bluegrass, creeping bentgrass, and perennial ryegrass.

## MATERIALS AND METHODS

Annual bluegrass, 'L-93' creeping bentgrass, and 'Manhattan IV' perennial ryegrass plants were collected from mature stands at Horticulture Research Farm II in North Brunswick, NJ. Annual bluegrass was an indigenous annual biotype (*Poa annua* var. *annua*). Grasses were planted in pots with 1-dm<sup>2</sup> surface area and 10-cm depths in a

greenhouse at the New Jersey Agriculture Greenhouse Research Complex. Greenhouse day/night temperatures were set for approximately 25/19 C and natural light was supplemented with high intensity discharge lighting when intensity fell below 600  $\mu\text{mol}/\text{m}^2/\text{s}$ . Soil medium<sup>1</sup> consisted of 80% Canadian Sphagnum peat moss and a 20% mixture of perlite, vermiculite, dolomitic limestone, and calcitic limestone. Pots were irrigated to prevent plant wilt. Once grasses resumed active growth, pots were placed in growth chambers<sup>2</sup> set for 26/20 C (day/night) with 50% relative humidity. Growth chamber photoperiods were set for 12 hrs of 500  $\mu\text{mol}/\text{m}^2/\text{s}$  total radiation from fluorescent lamps<sup>3</sup> and soft light bulbs<sup>3</sup>. Light bulbs were placed parallel to fluorescent lamps every 15 cm and perpendicular to fluorescent lamps every 41 cm.

Grasses received two broadcast applications of bispyribac-sodium<sup>4</sup> at 148 g ai/ha. Sequential treatments were made two weeks after initial applications. Broadcast applications were made with a CO<sub>2</sub>-pressured spray chamber<sup>5</sup> equipped with an 8002 EVS spray tip<sup>6</sup> calibrated to deliver 370 L/ha. Immediately following sequential broadcast applications, a 2 $\mu\text{L}$  droplet of spotting solution containing a total of 2 kBq of <sup>14</sup>C-bispyribac-sodium [uniformly ring labeled (specific activity, 2.66 MBq/mg; radiochemical purity, 99.1%)] was applied to the second fully expanded leaf with a microsyringe<sup>7</sup>. Appropriate amount of formulated herbicide<sup>2</sup> was added to spotting solutions to simulate 148 g/ha of bispyribac-sodium at a spray volume of 370 L/ha with a non-ionic surfactant<sup>8</sup> at 0.125% vol/vol to facilitate deposition. Application rate and regimen were chosen based on pilot experiments to mimic plant responses previously noted in field experiments with bispyribac-sodium (Lycan and Hart 2006b). <sup>14</sup>C-bispyribac-sodium was spotted following the second broadcast application since multiple

treatments have been shown necessary for annual bluegrass control (Askew et al. 2004; McCarty and Estes 2005; Lycan and Hart 2006a; McDonald et al. 2006).

Treated leaves were excised one, three, and seven days after treatment. Unabsorbed  $^{14}\text{C}$ -bispyribac-sodium was removed by swirling the treated leaf in a 20-ml scintillation vial containing two mLs of 10% methanol solution for approximately 45 seconds. Upon removal from vials, leaves were rinsed with 2 ml of washoff solution similar to that aforementioned and stored at  $-30\text{ C}$  until further analysis.

For herbicide extraction, methodology was modified in preliminary experiments from procedures provided by Valent U.S.A. (Anonymous 2003). Single leaves were placed in 1.5-mL tubes<sup>9</sup>, ground with liquid nitrogen, and then filled with 0.75-mL of acetonitrile<sup>10</sup>:deionized water (4:1) solution. Tubes were then agitated on a rotary shaker<sup>11</sup> for approximately 30 seconds and placed in water sonication<sup>12</sup> for 45 minutes. Samples were then centrifuged<sup>13</sup> for five minutes and solution was transferred to separate tubes. The procedure was repeated with fresh acetonitrile solution added to grounded tissue and the two 0.75 mL samples were combined in a 1.5-mL tube. To evaluate potential effects of extraction on herbicide degradation, fresh grass leaves of each species were harvested, spotted with 2-ml of  $^{14}\text{C}$  herbicide solution, and processed in conjunction with samples. Extraction techniques were conducted on freshly spotted leaves so procedure effects could be compared to extraction techniques with  $^{14}\text{C}$  bispyribac-sodium alone, without leaf tissue.

A 500  $\mu\text{L}$  of extract was spotted on 20 sq cm silica gel thin-layer chromatography (TLC) plates<sup>14</sup> and developed to 16 cm with chloroform<sup>15</sup>:methanol<sup>15</sup>:ammonium hydroxide<sup>17</sup>:water (80:30:4:2, v/v). Plates were air dried, partitioned into eight lanes

every 2-cm, and silica gel was scraped every 2-cm (2 x 20-cm sample area) into 10-mL scintillation vials. Vials were filled with scintillation fluid and  $^{14}\text{C}$  was quantified by liquid scintillation spectroscopy<sup>18</sup>. Standard  $R_f$  values were identified on silica gel plates from developed 2-mL of stock radiolabeled herbicide solution with and without ground leaf tissue dissolved in 500 mL of acetonitrile. Parent bispyribac-sodium was identified by comparing values from corresponding  $R_f$  standard values after plate development. Additional plates with extracts from freshly spotted leaves were compared with pure standard  $^{14}\text{C}$  herbicide to confirm extraction procedures did not cause herbicide degradation.

Two experiments were conducted as a randomized complete block with four replications. Metabolite data consisted of percent parent herbicide, sum percentage of metabolites more polar than parent herbicide, and sum percentage of all metabolites less polar than parent herbicide. Data were analyzed using analysis of variance<sup>19</sup> and means were separated using Fisher's Protected LSD test at  $\alpha = 0.05$ . Experiment by treatment interaction was not detected, and thus results were pooled over experiments.

## RESULTS AND DISCUSSION

One distinct metabolite in all grasses was detected at  $R_f$  0.31 from spotted samples. The  $R_f$  value of bispyribac-sodium was 0.56. Radioactivity with  $R_f$  less than 0.56 was pooled and presented as sum percentage of polar metabolites. Radioactivity with  $R_f$  greater than 0.56 was pooled and presented as sum percentage of non-polar metabolites.

Metabolism of  $^{14}\text{C}$ -bispribac-sodium varied among species from one to seven days after treatments (Table 1). After one day, annual bluegrass, creeping bentgrass, and perennial ryegrass had 79, 49, and 59% parent herbicide recovered, respectively, and 19, 46, and 34% polar metabolites, respectively. Annual bluegrass had significantly more parent herbicide recovered after one day than creeping bentgrass and perennial ryegrass while creeping bentgrass had less parent herbicide recovered than perennial ryegrass.

Creeping bentgrass and perennial ryegrass had similar metabolism after three and seven days but both species metabolized more herbicide than annual bluegrass. Pooled over creeping bentgrass and perennial ryegrass, parent herbicide recovered was 43 and 36% of total radioactivity after three and seven days, respectively. Conversely, creeping bentgrass and perennial ryegrass polar metabolites averaged 50 and 57% of total radioactivity recovered after three and seven days, respectively. Parent herbicide recovered in annual bluegrass after three and seven days was 82 and 73% of total radioactivity, respectively, while polar metabolites were 14 and 24% of the total, respectively.

Non-polar metabolite recovery was generally low ( $< 9\%$ ) and was similar across all species after one day. Perennial ryegrass had more non-polar metabolites than annual bluegrass after three days but both species were similar to creeping bentgrass. However, creeping bentgrass and perennial ryegrass averaged 9% non-polar metabolite recovery after seven days which was significantly greater than 3% of total in annual bluegrass. Bispribac-sodium applications to annual bluegrass under warm temperatures (20 to 30 C) cause substantial growth inhibition and desiccation following sequential applications (Askew et al. 2004; Lycan and Hart 2006a; McCullough and Hart 2006;

Table 1. Metabolism of  $^{14}\text{C}$ -bispyribac-sodium one, three, and seven days after sequential treatments at 148 g ai/ha in two combined growth chamber experiments.

Species	Metabolites (% of total $^{14}\text{C}$ recovered) <sup>a</sup>								
	1 DAT <sup>b</sup>			3 DAT			7 DAT		
	Polar	Parent	Nonpolar	Polar	Parent	Nonpolar	Polar	Parent	Nonpolar
Annual Bluegrass	19 c	79 a	2 a	14 b	82 a	4 b	24 b	73 a	3 b
Creeping Bentgrass	46 a	49 c	5 a	47 a	47 b	6 ab	59 a	32 b	9 a
Perennial Ryegrass	34 b	59 b	7 a	53 a	38 b	9 a	55 a	39 b	8 a

<sup>a</sup>Single leaves of grasses were treated with a 2- $\mu\text{L}$  droplet of  $^{14}\text{C}$ -bispyribac-sodium dissolved in water and 0.125% (v/v) nonionic surfactant, and containing approximately 2 kBq radioactivity plus non-labeled bispyribac-sodium. “Polar metabolites” indicates percentage sum of all metabolites more polar than bispyribac-sodium, and “nonpolar metabolites” indicates percentage sum of all metabolites less polar than bispyribac-sodium. “Parent” indicates percentage sum of all metabolites similar in polarity to bispyribac-sodium.

McDonald et al. 2006). Conversely, creeping bentgrass and perennial ryegrass have better tolerance to bispyribac-sodium under warm temperatures and generally recover from growth inhibition after approximately fourteen days (Lycan and Hart 2005; Lycan and Hart 2006a; McCullough and Hart 2006; McDonald et al. 2006). Half life of  $^{14}\text{C}$ -bispyribac-sodium appears greater than seven, one, and two days for annual bluegrass, creeping bentgrass, and perennial ryegrass, respectively. Creeping bentgrass and perennial ryegrass appear to rapidly metabolize bispyribac-sodium within one to three days while annual bluegrass metabolism was substantially lower than tolerant grasses.

Efficacy of other grassy weed herbicides has been attributed to differential species metabolism. Dubelman et al. (1997) noted corn and wheat tolerance to halosulfuron was due to rapid metabolism while little metabolism was detected in soybean. Olson et al. (2000) noted sensitivity of downy brome and wild oat (*Avena fatua* L.) to MON 37500 was attributed to slower metabolism than wheat. King et al. (2003) noted less absorption and greater metabolism by wheat and barley (*Hordeum vulgare* L.) of AE-F130060-03 in comparison with Italian ryegrass (*Lolium multiflorum* L.) was correlated with species tolerance and herbicide selectivity.

In other experiments, Lycan and Hart (2006b) noted annual bluegrass and creeping bentgrass retained greater than 90% of foliar applied  $^{14}\text{C}$ -bispyribac-sodium in treated leaves. It was also noted that annual bluegrass translocated more  $^{14}\text{C}$ -bispyribac-sodium to crowns and leaves than creeping bentgrass. Metabolism of selective annual bluegrass herbicides in cool-season turfgrass have received limited investigation. However, researchers have investigated metabolism of plant growth regulators used for annual bluegrass suppression. Beasley et al. (2005) noted creeping bentgrass



metabolized paclobutrazol more rapidly than annual bluegrass. Thus, differential species metabolism appears important for selectivity of materials used for annual bluegrass control in cool-season turfgrasses.

### Source of Materials

<sup>1</sup> Pro-Mix Soil, Premier Horticulture, Quakertown, PA 18951

<sup>2</sup> Environmental Growth Chambers, P.O. Box 407, Chagrin Falls, OH 44022

<sup>3</sup> General Electric Lighting, Chicago, IL 60601.

<sup>4</sup> Velocity®, 80WP herbicide, Valent U.S.A. Corp. PO Box 8025, Walnut Creek, CA 94596.

<sup>5</sup> Research Track Sprayers, Devries Manufacturing, Hollandale, MN 56045

<sup>6</sup> Tee Jet®, Spraying Systems Co. Wheaton, IL 60189-7900.

<sup>7</sup> Microsyringe, Hamilton Co., Reno, NV 89502

<sup>8</sup> X-77® non-ionic surfactant, Loveland Industries Inc., Greeley, CO 80632.

<sup>9</sup> USA Scientific 1.5 mL microcentrifuge tubes, Fisher Scientific, Fair Lawn, NJ 07410.

<sup>10</sup> Acetonitrile, Pesticide Grade, Fisher Scientific, Fair Lawn, NJ 07410.

<sup>11</sup> Vortex Genie 2™, Fisher Scientific, Fair Lawn, NJ 07410.

<sup>12</sup> Sonicater, Solid State/Ultrasonic FS-28, Fisher Scientific, Fair Lawn, NJ 07410.

<sup>13</sup> Marathon Micro A centrifuge, Fisher Scientific, Fair Lawn, NJ 07410.

<sup>14</sup> Whatman K5 150 A silica gel adsorption preparative plates, Fisher Scientific, Fair Lawn, NJ 07410.

<sup>15</sup> Chloroform ECD Tested for Pesticide Analysis, Fisher Scientific, Fair Lawn, NJ 07410.

- <sup>16</sup> Methanol, Histological Grade, Fisher Scientific, Fair Lawn, NJ 07410.
- <sup>17</sup> Ammonium hydroxide, A669-212, Fisher Scientific, Fair Lawn, NJ 07410.
- <sup>18</sup> Model LS3801, Beckman-Coulter, Inc., Fullerton, CA 92834-3100.
- <sup>19</sup> SAS Institute, Cary, NC 27511.

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Williams, B. J. 1999. Barnyardgrass (*Echinochloa crus-galli*) control in dry-seeded rice with V-10029. *Proc. South. Weed Sci. Soc.* 52: 50.

**Vita**  
**Patrick E. McCullough**

**Education**

Rutgers, The State University of New Jersey

Doctor of Philosophy: Plant Biology, 2009

Dissertation: Management Practices, Environment, and Spray Adjuvants  
Influence Bispyribac-Sodium Efficacy and Metabolism in Turf

Clemson University

Master of Science: Plant and Environmental Sciences, 2004

Minors: Experimental Statistics and Biological Sciences

Thesis: Physiological Response of 'TifEagle' Bermudagrass to Nitrogen and  
Trinexapac-ethyl

Western Kentucky University

Bachelor of Science: Agriculture, 2002

**Professional Experience**

Rutgers University, New Brunswick, NJ

Program Associate (2004 – 2008)

Clemson University, Clemson, SC

Graduate Research Assistant (2002 – 2004)

**Internships**

Fenway Park, Boston, MA (5/2002 – 8/2002)

Western Kentucky University Sports Fields (8/2001 – 5/2002)

Hartland Golf Course, Bowling Green, KY (3/2001 – 8/2001)

Legends Club of Tennessee, Franklin, TN (5/2000 – 8/2000)

**Selected Honors & Awards**

ASA Graduate Student Oral Presentation Contest Winner (1<sup>st</sup> place), 2008

ASA Graduate Student Oral Presentation Contest Winner (3<sup>rd</sup> place), 2007

Spencer Davis Plant Biology Research Award, 2005

Wilbur M. Runk Award Recipient, 2005

Turf and Ornamental Communicators Association Scholarship, 2004

Dean's List, Western Kentucky University, 2002

**Selected Journal Publications**

McCullough, P.E., and S.E. Hart 2008. Roughstalk bluegrass and tall fescue control in Kentucky bluegrass with sulfosulfuron. Online. Applied Turfgrass Science. doi:10.1094/ATS-2008-0625-01-RS.

McCullough, P.E. and S.E. Hart. 2008. Spray adjuvants influence bispyribac-sodium efficacy for annual bluegrass control in cool-season turfgrasses. *Weed Technol.* 22:257-262.

McCullough, P.E., H. Liu, L.B. McCarty, and J.E. Toler. 2007. Trinexapac-ethyl application regimens influence growth, quality and performance of creeping bentgrass and dwarf bermudagrass golf greens. *Crop Science* 47:2138-2144.

Hart, S.E. and P.E. McCullough. 2007. Annual bluegrass control in Kentucky bluegrass with bispyribac-sodium, primisulfuron, and sulfosulfuron. *Weed Technology* 21:251-254.

McCullough, P.E., H. Liu, L.B. McCarty, T. Whitwell, and J.E. Toler. 2006. Bermudagrass putting green quality, growth, and nutrient partitioning influenced by nitrogen and trinexapac-ethyl. *Crop Sci.* 46:1515-1525.

McCullough, P.E., S.E. Hart, S. Askew, P.H. Dernoeden, Z. Reicher, and D. Weisenberger. 2006. Kentucky bluegrass control with postemergence herbicides. *HortSci.* 41(1):255-258.

McCullough, P.E., H. Liu, and L.B. McCarty. 2005. Trinexapac-ethyl application regimens influence creeping bentgrass putting green performance. *HortSci.* 40(6):2168-2169.

McCullough, P.E., H. Liu, and L.B. McCarty. 2005. Response of six dwarf-type bermudagrasses to trinexapac-ethyl. *Hort Sci.* 40(2):460-462.

McCullough, P.E., S.E. Hart, and D.W. Lycan. 2005. Plant growth regulator regimens reduce *Poa annua* populations in creeping bentgrass. Online. *Applied Turfgrass Science*. doi: 10.1094/ATS-2005-0304-01-RS.

McCullough, P.E., H. Liu, L.B. McCarty, and T. Whitwell. 2004. Response of 'TifEagle' bermudagrass to seven plant growth regulators. *HortSci.* 39(7):1759-1762.

### **Selected Trade Publications**

McCullough, P.E. 2006. Annual bluegrass control: implications for eliminating a historic weed in golf course turfgrass. *Golf Course Management* (Web Exclusive Article). <http://www.gcsaa.org/GCM/2006/oct/WebEx.asp>. Published 2 Oct. 2006.

McCullough, P., H. Liu, and B. McCarty. 2006. TifEagle putting green management: nitrogen and PGR applications. *Golf Course Management.* 15(8):72-75.

McCullough, P., H. Liu, and B. McCarty. 2006. PGR programs for bermudagrass greens. *Golf Course Management.* 15(7):84-88.

McCullough, P., S. Hart, and D. Lycan. 2005. PGR programs for annual bluegrass suppression. *Golf Course Management.* 14(10):78-81.

### **Extension Fact Sheets**

Hart, S.E., P.E. McCullough, and J.A. Murphy. 2006. Annual and roughstalk bluegrass management in New Jersey home lawns. Extension Fact Sheet FS072. Rutgers University, New Brunswick, NJ.

McCullough, P.E. and S.E. Hart. 2006. Perennial grassy weed control in New Jersey turfgrass. Extension Fact Sheet FS1050. Rutgers University Extension Fact Sheet.

### **Selected Funded Proposals**

Hart, S., P. McCullough, and J. Murphy. 2005. Renovating golf course fairways and putting greens to creeping bentgrass. Funded by the Rutgers Center for Turfgrass Science. \$41,407.

McCullough, P.E. and H. Liu. 2003. Physiological Response of 'TifEagle' Bermudagrass to Trinexapac-ethyl. Syngenta Corporation. \$3000

McCullough, P.E. and H. Liu. 2002. Reducing Nitrogen Applications with Trinexapac-ethyl on Bermudagrass Greens. South Carolina Turf and Landscape Association. \$3000.

Liu, H., L.B. McCarty, and P.E. McCullough. 2002. Evaluating Foliar Versus Granular Fertilizers on Bentgrass and Bermudagrass Putting Greens. USGA. \$25,000.

Liu, H. and P.E. McCullough. 2002. Reducing Annual Nitrogen Fertilizer with Growth Regulators on Bermudagrass Putting Greens. 2002. Clemson Research Initiative Grant. \$2000.