

## Flucarbazone–Sodium Absorption, Translocation, and Metabolism in Bermudagrass, Kentucky Bluegrass, and Perennial Ryegrass

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Perennial ryegrass is overseeded in bermudagrass and Kentucky bluegrass to improve turf quality, but selective control may be warranted for transition back to monostand turfgrass. Flucarbazone–sodium controls perennial ryegrass in bermudagrass and Kentucky bluegrass, but the physiological basis of selectivity has received limited investigation. Greenhouse and laboratory experiments were conducted to evaluate efficacy, absorption, translocation, and metabolism of flucarbazone–sodium in these grasses. Flucarbazone–sodium reduced perennial ryegrass shoot mass from the nontreated an average  $\approx 22$  times and 3 times more than bermudagrass and Kentucky bluegrass at 4 wk after treatment, respectively. In laboratory experiments, foliar and root absorption of  $^{14}\text{C}$ –flucarbazone–sodium were similar among species. Bermudagrass distributed  $\approx 25\%$  more foliar-absorbed  $^{14}\text{C}$  to nontreated shoots than Kentucky bluegrass and perennial ryegrass. From root applications, all grasses averaged 84% distribution of  $^{14}\text{C}$  to shoots. Bermudagrass and Kentucky bluegrass metabolized 100% and 74% of  $^{14}\text{C}$ –flucarbazone–sodium at 1 d after treatment (DAT), whereas perennial ryegrass metabolism measured 44, 58, and 65% at 1, 3, and 7 DAT, respectively. Bermudagrass, Kentucky bluegrass, and perennial ryegrass had 4, 4, and 2 metabolites after 7 d, respectively. Results suggest differential metabolism of flucarbazone–sodium is attributed to selectivity for controlling perennial ryegrass in bermudagrass and Kentucky bluegrass.

**Nomenclature:** Flucarbazone–sodium; bermudagrass [*Cynodon dactylon* (L.) Pers.  $\times$  *Cynodon transvaalensis* Burtt-Davy] ‘Princess-77’; Kentucky bluegrass (*Poa pratensis* L.) ‘Midnight’; perennial ryegrass (*Lolium perenne* L.) ‘Manhattan V’.

**Key words:** Efficacy, herbicide fate, overseeding, selectivity, turfgrass, herbicide uptake.

Bermudagrass and Kentucky bluegrass are the most widely planted turfgrasses in the world. Bermudagrass is a warm-season species used for lawns, golf courses, and athletic fields in arid, tropical, and warm–humid regions. Kentucky bluegrass is a cool-season species planted throughout the northern U.S. transition zone and cool–humid regions for similar uses. These grasses have high densities, fine leaf textures, and good recovery from traffic, wear, and environmental stresses (Shearman and Beard 1975; Trenholm et al. 1999, 2000). However, physiological limitations of these grasses often warrant overseeding with perennial ryegrass to improve turf quality.

Perennial ryegrass is a cool-season species with desirable color, quality, and texture for overseeding (Bonos et al. 2004). Practitioners overseed dormant bermudagrass with perennial ryegrass in fall to enhance turfgrass aesthetics and functionality in winter and spring months. Perennial ryegrass establishment provides winter color to bermudagrass and may also suppress winter annual weed populations (Elford et al.

2008). Turf managers overseed Kentucky bluegrass with perennial ryegrass to enhance surface uniformity, reduce erosion, and suppress pest populations (Niehaus 1976; Richmond et al. 2000). Because perennial ryegrass germinates quickly, overseeding Kentucky bluegrass may provide timely turf restoration in order to meet expectations of end users (Allen et al. 1993; Anslow 1962).

Recently developed perennial ryegrass cultivars have improved tolerances to disease, heat, and drought stress compared to traditional cultivars (Bonos et al. 2004; Jiang and Huang 2001; Wehner and Watschke 1981). However, the presence of perennial ryegrass in bermudagrass or Kentucky bluegrass may be undesirable in areas unintended for overseeding or during periods that warrant transition back to monostand turfgrass. As bermudagrass resumes active growth in spring, overseeded perennial ryegrass must be controlled with POST herbicides to minimize competition and improve quality of the desirable turfgrass. Perennial ryegrass overseeding in Kentucky bluegrass may provide temporary turf cover, especially during periods of active turf use. However, managing mixtures of turfgrass species is often undesirable for long-term culture, and practitioners must control overseeded

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perennial ryegrass with POST herbicides to allow Kentucky bluegrass recovery or establishment in thinned areas.

Bermudagrass managers use sulfonylureas, triazines, and other herbicides to control perennial ryegrass in spring (Johnson 1976; Harrell et al. 2005). These herbicides are generally safe and effective for transitioning overseeded areas back to bermudagrass without reducing turf quality. However, there are limited POST herbicides available for controlling perennial ryegrass in Kentucky bluegrass lawns, golf courses, or sports turf. Cultural practices, such as low mowing and interseeding Kentucky bluegrass, have potential to reduce perennial ryegrass populations, but results are often inconsistent and warrant nonselective herbicide use (Kraft et al. 2004).

Flucarbazone–sodium (hereafter delineated flucarbazone) is an acetolactate synthase (ALS) inhibitor used for POST grassy and broadleaf weed control in wheat (*Triticum aestivum* L.) and Kentucky bluegrass seed production. In turfgrass, flucarbazone has shown potential to control perennial ryegrass, and has good safety in bermudagrass, Kentucky bluegrass, and other turfgrass species (McCullough et al. 2012; McElroy et al. 2013). However, the behavior of flucarbazone has received limited investigation in turfgrass, and the physiological basis associated with selectivity warrants further evaluations. The objective of this research was to evaluate efficacy, uptake, translocation, and metabolism of flucarbazone in bermudagrass, Kentucky bluegrass, and perennial ryegrass.

## Materials and Methods

**Response of Grasses to Flucarbazone.** Experiments were conducted from May to July 2013 in Griffin, GA. ‘Princess-77’ bermudagrass (Pennington Seed, Inc., Madison, GA 30650), ‘Midnight’ Kentucky bluegrass (Seedland, Inc., Wellborn, FL 32094) and ‘Manhattan V’ perennial ryegrass (Pure Seed Testing, Inc., Hubbard, OR 97032) were seeded in pots with 3.8-cm diameter and 20-cm depth, filled with sand : peat moss (80 : 20 v/v). Bermudagrass was established in a greenhouse set for 32/25 C (day/night). Kentucky bluegrass and perennial ryegrass were established in a greenhouse set for 23/17 C. Grasses were irrigated to prevent wilting and trimmed weekly at 5-cm height with shears. After reaching a three- to five-tiller stage, individual plants were transplanted to pots with 79 cm<sup>2</sup> and 10-cm depths filled with sand : peat moss (80 : 20 v/v). Pots were placed in a

greenhouse set for 23/17 C at 1 wk before treatments.

A broadcast application of flucarbazone (Everest 70WG, flucarbazone–sodium, Arysta LifeSciences, Cary, NC) was made to grasses at 0, 30, or 60 g ai ha<sup>-1</sup>. Rates were chosen based on recommendations for use in tolerant turfgrasses (Anonymous 2010). Treatments were applied with a CO<sub>2</sub>-pressured sprayer calibrated to deliver 374 L ha<sup>-1</sup> with a single 9504E flat-fan nozzle (Tee Jet, Spraying Systems Co., Roswell, GA 30075). Grasses were not irrigated for 24 HAT, but received irrigation after this period to prevent moisture deficiency.

Experimental design was a randomized complete block with four replications. After 4 wk, shoots were harvested, oven dried at 60 C for 72 h, and weighed. Data were subjected to analysis of variance at the 0.05 probability level. Means were separated with Fisher’s Protected LSD test at  $\alpha = 0.05$ . Two separate experiments were conducted. Experiment by treatment interaction was not detected, and thus, experiments were combined.

**Foliar Absorption and Translocation.** Experiments were conducted in Griffin, GA in February 2013. ‘Princess-77’ bermudagrass, ‘Midnight’ Kentucky bluegrass, and ‘Manhattan V’ perennial ryegrass were established with aforementioned materials and methods. Grasses were thinned to one plant per container (3.8-cm diameter by 20-cm depth) after emergence and allowed to develop three to five tillers before treatments. Grasses were selected based on size and uniformity of populations, and placed in a growth chamber (Percival Scientific, Inc., 505 Research Drive, Perry, IA 50220) set for 27/22 C (day/night) with 12-h photoperiod of 350  $\mu\text{mol m}^{-2} \text{s}^{-1}$  and approximately 50% relative humidity. Grasses were watered as needed to prevent wilt and acclimated in the growth chamber for 3 d before treatments.

A broadcast application of flucarbazone was made to grasses at 30 g ha<sup>-1</sup> with a single-nozzle CO<sub>2</sub>-pressured sprayers at 374 L ha<sup>-1</sup>. Immediately following the broadcast application, a fully expanded leaf of each grass was spotted with a 2- $\mu\text{l}$  droplet of <sup>14</sup>C–flucarbazone–sodium (specific activity: 64.9 mCi/mmol; labeled at phenyl ring) containing a total 250 Bq with the use of a 10- $\mu\text{l}$  microsyringe (Hamilton Co., Reno, NV 89502). Formulated product was added to the spotting solution to simulate spray droplets of 30 g ha<sup>-1</sup> at 374 L ha<sup>-1</sup>. A nonionic surfactant (Activator 90, Loveland Products, Inc., Greeley, CO 80632) was added at 0.125% v/v to facilitate deposition on the leaf surface. This

methodology is similar to previous research with ALS inhibitor foliar uptake in turfgrasses (Lycan and Hart 2006).

Plants (roots + shoots) were harvested at 1, 4, 8, 24, 48, and 72 h after treatment (HAT). The treated leaf was excised from the base of the plant and swirled in a 20-ml glass scintillation vial with 2 ml of 20% methanol solution for 30 s to remove unabsorbed  $^{14}\text{C}$ . The treated leaf was then rinsed with an additional 2 ml of methanol solution. Roots and shoots of plants harvested at 1, 4, 8, and 48 HAT were oxidized to quantify absorption. Grasses harvested at 24 and 72 HAT were sectioned into treated leaf, nontreated shoots, and roots for determining  $^{14}\text{C}$  distribution. Samples were oven dried at 60 C and then combusted in a biological oxidizer (Model OX-500, R.J. Harvey Instrument Corp., Hillsdale, NJ 076742). Radioactivity was then quantified with liquid scintillation spectroscopy (Model LS6500, Beckman-Coulter, Inc., Fullerton, CA 92834-3100). Percent absorption was determined by dividing total radioactivity in samples by the amount applied. Percent  $^{14}\text{C}$  distribution was calculated by dividing the radioactivity recovered in samples by total radioactivity recovered in each plant.

The experiment was conducted as a completely randomized design with four replications, and the experiment was repeated. Data were subjected to analysis of variance, and significance of main effects was analyzed at the 0.05 probability level. Absorption data were subjected to regression analysis, and time required for 50% foliar absorption was calculated for each species. For  $^{14}\text{C}$  distribution data, means were separated with the use of Fisher's Protected LSD test at  $\alpha = 0.05$ . Experiment by species interaction was not detected, and thus, experiments were combined.

**Root Absorption and Translocation.** Experiments were conducted in Griffin, GA in February 2013, with the use of previously described plant materials and greenhouses. Grasses were removed from pots after reaching three to five tillers, and selected based on population uniformity. Potting medium was washed from roots, and plants were grown hydroponically in a 4-L plastic tank filled with half-strength Hoagland solution (Hoagland and Arnon 1950). Roots were suspended in solution by placement through holes in a floating Styrofoam board (Styrofoam, 12961 San Fernando Road, Sylmar, CA 91342) and aluminum foil covered the sides of the tank to shield roots from light. An aquarium pump (Shkerry Aqua<sup>®</sup>, Shanghai Uni-Aqua Co., Ltd., Chang Shou Road, Shanghai,

China 200042) was used to provide oxygen to the solution.

After 7 d, grasses were placed individually into 50-ml plastic tubes filled with 25-ml half-strength Hoagland solution spiked with 8 kBq L<sup>-1</sup> of  $^{14}\text{C}$ -flucarbazone. Formulated flucarbazone was added to tubes to simulate a 30 g ha<sup>-1</sup> surface area application rate. Plant roots were submerged in the herbicide solution by placing cotton balls around the base of shoots and tubes were covered with aluminum foil. The cotton balls were placed above the solution to facilitate root submergence without soaking additional solution in the plant.

Plants were harvested at 72 HAT. Unabsorbed  $^{14}\text{C}$  was removed by rinsing roots with 5 ml of 20% methanol solution and then blotting on paper towels. Grasses were then separated to shoots and roots, oven-dried at 60 C, and combusted in a biological oxidizer (Model OX-500, R.J. Harvey Instrument Corp., Hillsdale, NJ 076742). Radioactivity in each sample was determined with the use of liquid scintillation spectroscopy (Model LS6500, Beckman-Coulter, Inc., Fullerton, CA 92834-3100).

The experiment was conducted in a completely randomized design with four replications, and was repeated. Percent absorption was determined by dividing the amount of radioactivity in the whole plant from total radioactivity applied. Percent  $^{14}\text{C}$  distribution was calculated by dividing radioactivity recovered in samples by total radioactivity in the plant. Data were subjected to analysis of variance at the 0.05 probability level. Means were separated using Fisher's Protected LSD test at  $\alpha = 0.05$ . Experiment by treatment interactions were not detected, and thus, experiments were combined.

**Metabolism.** Experiments were conducted in Griffin, GA with aforementioned grasses, materials, and methods. Once grasses reached three to five tillers in pots, plants were selected for treatments based on size and population uniformity. A broadcast treatment of flucarbazone at 30 g ha<sup>-1</sup> was applied to grasses with aforementioned CO<sub>2</sub>-pressured sprayers calibrated to deliver 374 L ha<sup>-1</sup>. Immediately after broadcast applications, a 2- $\mu\text{l}$  droplet of  $^{14}\text{C}$ -flucarbazone containing 1.7 kBq was applied to the adaxial surface of a single leaf on each grass with a 10- $\mu\text{l}$  microsyringe (Hamilton Co., Reno, NV 89502). The spotting solution contained formulated flucarbazone to simulate 30 g ha<sup>-1</sup> at 374 L ha<sup>-1</sup>. A nonionic surfactant was added to the spotting solution at 0.125% v/v to facilitate deposition on the leaf surface. Grasses were placed

in a growth chamber with previously described settings. Treated leaves were harvested at 1, 3, and 7 d after treatment (DAT) and unabsorbed  $^{14}\text{C}$  was removed with the use of a procedure described in foliar absorption experiments.

Treated leaves were placed in 1.5-ml microcentrifuge tubes (Fisher Scientific, Fair Lawn, NJ 07410) and ground with liquid nitrogen. Tubes were then filled with 500  $\mu\text{l}$  of acetonitrile with 0.2 M ammonium acetate, agitated with the use of a rotary shaker for 30 s, and placed in a sonication bath (Fisher Scientific 300, Industry Drive, Pittsburgh, PA 15275) for 45 m. Tubes were then centrifuged for 10 min and extract solution was transferred to separated tubes. This procedure was repeated three times and samples were combined in a 1.5-ml tube. Extraction efficiency from these methods measured  $> 85\%$  after residue oxidation.

Extraction solution was then transferred to glass vials (Thermo Scientific, 320 Rolling Ridge Drive, Bellefonte PA 16823) and evaporated on a heating block set for 50 C in a fume hood. Samples were resuspended in 300  $\mu\text{l}$  of acetonitrile and spotted on 20 by 20-cm thin-layer chromatography (TLC) plates. Samples were then developed to 16 cm in a glass chamber with the use of 16.5 : 3 dichloromethane : methanol solution. Plates were air dried and metabolites were identified with a radiochromatogram scanner (BioScan AR-2000, Bioscan, 4590 MacArthur Boulevard NW, Washington, DC 20007) connected to a computer equipped with Laura Chromatography Data Collection and Analysis Software<sup>®</sup> (LabLogic System, Inc., 1040 East Brandon Blvd Brandon, FL 33511). The  $R_f$  value of the parent herbicide was identified by spotting  $^{14}\text{C}$ -flucarbazone in acetonitrile on TLC plates and running samples through the analysis. Data included number of metabolites; sum percentage of metabolites more polar than the parent herbicide, sum percentage of metabolites less polar than the parent herbicide, and parent herbicide. Data were subjected to analysis in SAS, and means were separated with the use of Fisher's Protected LSD test at  $\alpha = 0.05$ . Two separate experiments were conducted. Experiment by treatment interactions were not detected, and thus, data were pooled over experiments.

## Results and Discussion

**Response of Grasses to Flucarbazone.** Rate by species interaction was not detected for shoot mass reductions, and thus, main effects are discussed.

Compared to nontreated controls, the shoot biomass reduction of perennial ryegrass, 4 WAT, was 22 and 3 times higher than bermudagrass and Kentucky bluegrass, respectively (data not shown). Furthermore, no differences were detected between flucarbazone rates.

Perennial ryegrass has shown susceptibility to shoot biomass reductions by flucarbazone treatments. In previous work, flucarbazone efficacy increased on perennial ryegrass as temperatures increased from 10 to 30 C (McCullough et al. 2012). Perennial ryegrass shoot biomass was reduced 44% from the nontreated, but efficacy could have increased with higher greenhouse temperatures or with sequential applications. Tolerant turfgrasses had negligible reductions in biomass from the nontreated, and supports the efficacy of flucarbazone for selective perennial ryegrass control. Bermudagrass shoot biomass was reduced by  $< 5\%$  from nontreated, and Kentucky bluegrass had only 16% reductions.

Bermudagrass has high tolerance levels to ALS inhibitors labeled for perennial ryegrass control including flazasulfuron, foramsulfuron, rimsulfuron, and trifloxysulfuron (Harrell et al. 2005; Toler et al. 2007). Chlorsulfuron is currently the only POST herbicide labeled for perennial ryegrass control in Kentucky bluegrass, and applications may cause excessive ( $> 20\%$ ) turfgrass injury (Dernoe-den 1990; McCullough and Hart 2008). In previous research, Kentucky bluegrass has shown acceptable tolerances to primisulfuron and sulfosulfuron for selective grassy weed control (Hart and McCullough 2007). However, Kentucky bluegrass tolerances to ALS inhibitors may vary by cultivar (Shortell et al. 2008), and further research is needed to evaluate flucarbazone use in varieties with genetic and morphological differences that may influence herbicide injury. Bermudagrass and perennial ryegrass cultivars are also genetically diverse, and further research is needed to evaluate differential tolerances to flucarbazone applications.

**Absorption and Translocation.** Foliar absorption of  $^{14}\text{C}$ -flucarbazone was similar among species at all harvests (data not shown). Foliar absorption increased from 19 to 96% of the applied over the 72-h period, and grasses reached 50% absorption at 8 HAT (data not shown). Differences in foliar absorption do not appear associated with tolerance levels to flucarbazone in these grasses, and results are similar to reports with other ALS inhibitors. Researchers have noted tolerant and susceptible

Table 1. Radioactivity distribution from foliar applications of  $^{14}\text{C}$ -flucarbazone to 'Princess-77' bermudagrass, 'Midnight' Kentucky bluegrass, and 'Manhattan V' perennial ryegrass.

Species	$^{14}\text{C}$ distribution					
	24 HAT <sup>a</sup>			72 HAT		
	Treated leaf	Nontreated shoots	Roots	Treated leaf	Nontreated shoots	Roots
	%					
Bermudagrass	75	25	0	68	32	0
Kentucky bluegrass	80	18	2	94	6	0
Perennial ryegrass	84	16	0	90	10	0
LSD <sub>0.05</sub>	NS	NS	NS	11	11	NS

<sup>a</sup> HAT = hours after treatment.

grasses had similar foliar absorption levels of ALS inhibitors, including chlorsulfuron, nicosulfuron, propoxycarbazone, rimsulfuron, and sulfometuron (Baird et al. 1989; Christopher et al. 1992; Mekki and Leroux 2006; Park et al. 2004).

Foliar absorption levels of flucarbazone were comparable to previous reports with ALS inhibitors applied to grasses. Fandrich et al. (2001) noted jointed goatgrass (*Aegilops cylindrica* Host) and downy brome (*Bromus tectorum* L.) absorbed 90 and 89% of applied  $^{14}\text{C}$ -propcarbazone-sodium at 48 HAT, respectively. Pester et al. (2001) reported foliar absorption of imazamox measured > 90% of the applied at 48 HAT in jointed goatgrass, downy brome, and feral rye (*Secale cereale* L.). In two *Kyllinga* species, McElroy et al. (2004) noted the majority of  $^{14}\text{C}$ -halosulfuron and  $^{14}\text{C}$ -trifloxysulfuron was absorbed at 4 HAT, and absorption was similar between species.

At 24 HAT, all grasses retained  $\geq 75\%$  of absorbed  $^{14}\text{C}$  in treated leaves and differences were not detected for distribution to nontreated shoot or roots (Table 1). At 72 HAT, Kentucky bluegrass and perennial ryegrass recovered  $\geq 90\%$  of absorbed  $^{14}\text{C}$  in treated leaves and averaged 8% distribution to nontreated shoots. Bermudagrass distributed 32% of foliar-absorbed  $^{14}\text{C}$  to nontreated shoots at 72 HAT and was significantly greater than other species. All grasses had no radioactivity recovered in roots at 72 HAT, suggesting flucarbazone or metabolites are concentrated in shoots after foliar uptake.

Differences were not detected among species for absorption or  $^{14}\text{C}$  distribution from root-applied flucarbazone (data not shown). Root absorption measured 13% of the applied across species, and the majority of radioactivity was distributed to shoots in all grasses. Distribution of root-absorbed  $^{14}\text{C}$  to shoots averaged 85% of the absorbed in all grasses at 72 HAT and is similar to reports with other ALS-

inhibiting herbicides. Lycan and Hart (2006) noted annual and roughstalk bluegrass (*Poa annua* L. and *P. trivialis* L.) distributed  $\approx 80\%$  of root-absorbed  $^{14}\text{C}$ -bispyribac-sodium to shoots at 72 HAT, but creeping bentgrass and Kentucky bluegrass averaged 66%. Novosel and Renner (1995) reported 57% of root-absorbed nicosulfuron translocated to shoots of sugarbeet (*Beta vulgaris* L.) at 12 HAT, but an equal concentration of primisulfuron was translocated to shoots at 48 HAT.

Flucarbazone appears to have acropetal movement in turfgrasses, with limited distribution from treated leaves. Grasses readily absorbed flucarbazone through shoots, and retained  $\geq 75\%$  of  $^{14}\text{C}$  in treated leaves. Uptake of flucarbazone in roots and shoots appears similar in these species, but bermudagrass may have slightly more translocation after foliar absorption than Kentucky bluegrass and perennial ryegrass.

**Metabolism.** Parent  $^{14}\text{C}$ -flucarbazone was detected at  $R_f$  0.5 and metabolite recovery varied across species. Two major metabolites were identified in all species at  $R_f$  0.19 and 0.81. Bermudagrass had these major metabolites and two other metabolites were detected at 7 DAT at  $R_f$  0.06 and 0.66. Parent herbicide was not detected in bermudagrass on any date, suggesting flucarbazone was completely metabolized in < 1 d (Table 2). After 7 d, 45% and 55% of metabolites recovered in bermudagrass were polar and nonpolar, respectively. Rapid metabolism of flucarbazone in bermudagrass is probably associated with distribution of  $^{14}\text{C}$  to nontreated shoots from the treated leaf at 72 HAT in absorption experiments.

Kentucky bluegrass had the major metabolites formed at 1 and 3 DAT, but two additional metabolites were detected at 7 DAT at  $R_f$  0.06 and 0.38. The metabolite at  $R_f$  0.38 was not detected in bermudagrass, and the bermudagrass metabolite at

Table 2. Metabolism of  $^{14}\text{C}$ -flucarbazone in 'Princess-77' bermudagrass, 'Midnight' Kentucky bluegrass, and 'Manhattan V' perennial ryegrass in two combined experiments, 2013, Griffin, GA.

Species	Metabolites recovered								
	1 DAT <sup>a</sup>			3 DAT			7 DAT		
	Polar <sup>b</sup>	Parent	Nonpolar	Polar	Parent	Nonpolar	Polar	Parent	Nonpolar
	—% of $^{14}\text{C}$ extracted—								
Bermudagrass	32	0	68	55	0	45	45	0	55
Kentucky bluegrass	41	26	33	64	12	24	65	0	35
Perennial ryegrass	21	56	23	37	42	21	30	35	35
LSD <sub>0.05</sub>	12	13	8	14	12	11	8	12	10
	Metabolite number								
	Total	Polar	Nonpolar	Total	Polar	Nonpolar	Total	Polar	Nonpolar
Bermudagrass	3	1	2	3	1	2	4	2	2
Kentucky bluegrass	2	1	1	2	1	1	4	3	1
Perennial ryegrass	2	1	1	2	1	1	2	1	1
LSD <sub>0.05</sub>	0	NS	0	0	NS	0	0	0	0

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

<sup>b</sup> Polar and nonpolar delineate sum percentage of metabolites from the total recovered.

$R_f$  0.66 was not detected in Kentucky bluegrass. Results suggest different metabolites of  $^{14}\text{C}$ -flucarbazone may be produced by warm- and cool-season turfgrasses, but the two major metabolites were consistent across species. Parent herbicide levels in Kentucky bluegrass measured 26, 12, and 0% at 1, 3, and 7 DAT, respectively (Table 2). Kentucky bluegrass had 20% more total polar metabolites than bermudagrass at 7 DAT, but nonpolar metabolite levels were 20% greater in bermudagrass.

Perennial ryegrass had less metabolism of flucarbazone than bermudagrass and Kentucky bluegrass on all dates (Table 2). Perennial ryegrass only had the two major metabolites during the 7-d period. Levels of flucarbazone ranged 56, 42, and 35% in perennial ryegrass at 1, 3, and 7 DAT, respectively. By 7 DAT, perennial ryegrass had 15 and 35% less polar metabolites than bermudagrass and Kentucky bluegrass, respectively, but nonpolar metabolite levels were similar to Kentucky bluegrass.

Differential metabolism is the physiological basis for selectivity of many ALS inhibitors and is clearly associated with flucarbazone efficacy for perennial ryegrass control. Metabolic rate of flucarbazone appears substantially slower in perennial ryegrass than bermudagrass and Kentucky bluegrass. Moreover, perennial ryegrass only metabolized flucarbazone to two metabolites, and bermudagrass and Kentucky bluegrass developed four within a 7-d period. Bermudagrass rapidly metabolized flucarbazone and was comparable to previous reports with corn (*Zea mays* L.) treated with the ALS inhibitors, nicosulfuron, and primisulfuron (Gallaher et al. 1999).

Metabolism of ALS-inhibiting herbicides has been previously attributed to efficacy in grasses. For example, Baird et al. (1989) reported centipede-grass [*Eremochloa ophiuroides* (Munro) Hack.] metabolized twice as much  $^{14}\text{C}$ -sulfometuron than bahiagrass (*Paspalum notatum* Flueggé) at 72 HAT. Gallaher et al. (1999) reported broadleaf signalgrass [*Urochloa platyphylla* (Nash.) R.D. Webster] metabolized > 20 times more primisulfuron than nicosulfuron at 4 HAT, and was associated with differential tolerances. In other experiments, Park et al. (2004) found 80% of the absorbed  $^{14}\text{C}$ -propoxycarbazine was metabolized in susceptible and resistant biotypes of cheatgrass, but resistant biotypes had more rapid metabolism. Similar differences in metabolism have been noted in tolerant and susceptible species for other ALS inhibitors including chlorsulfuron, nicosulfuron, procarbazine-sodium, primisulfuron, sulfosulfuron, and trifloxysulfuron (Carey et al. 1997; Christopher et al. 1992; Fandrich et al. 2001; Hoseini et al. 2011; Mekki and Leroux 2006; Richardson et al. 2003).

Overall, flucarbazone has potential for controlling perennial ryegrass in bermudagrass and Kentucky bluegrass. The introduction of this chemistry to lawns, golf, or sports turf could provide a tool for controlling perennial ryegrass in Kentucky bluegrass. The physiological mechanism attributed to selectivity of flucarbazone in turfgrass appears to be differential metabolism. Future work should evaluate application rates and regimens of flucarbazone to help maximize efficacy for perennial ryegrass control in these turfgrasses.

## Literature Cited

- Allen PS, White DB, Markhart AH (1993) Germination of perennial ryegrass and annual bluegrass seeds subjected to hydration–dehydration cycles. *Crop Sci* 33:1020–1025
- Anonymous (2010) Align herbicide label. Cary, NC: Arysta LifeScience North America. Pp 1–4
- Anslow RC (1962) A quantitative analysis of germination and early seedling growth in perennial ryegrass. *Grass and Forage Sci* 17:260–263
- Baird JH, Wilcut JW, Wehjte GR, Dickens R, Sharpe S (1989) Absorption, translocation, and metabolism of sulfometuron in centipedegrass (*Eremochloa ophiuroides*) and bahiagrass (*Paspalum notatum*). *Weed Sci* 37:42–46
- Bonos SA, Kubik C, Clarke BB, Meyer WA (2004) Breeding perennial ryegrass for resistance to gray leaf spot. *Crop Sci* 44:575–580
- Carey JB, Penner D, Kells JJ (1997) Nicosulfuron and primisulfuron selectivity in five species. *Weed Sci* 45:22–30
- Christopher JT, Powles SB, Holtum JAM (1992) Resistance to acetolactate synthase-inhibiting herbicides in annual ryegrass (*Lolium rigidum*) involves at least two mechanisms. *Plant Phys* 100:1909–1913
- Dernoeden PH (1990) Comparison of three herbicides for selective tall fescue control in Kentucky bluegrass turf. *Agron J* 82:278–282
- Elford EMA, Tardif FJ, Robinson DE, Lyons EM (2008) Effect of perennial ryegrass overseeding on weed suppression and sward composition. *Weed Technol* 22:231–239
- Fandrich L, McDonald SK, Nissen SJ, Westra P, Santel HJ (2001) Absorption and fate of BAY MKH 6561 in jointed goatgrass and downy brome. *Weed Sci* 49:717–722
- Gallaher K, Mueller TC, Hayes RM, Schwartz O, Barrett M (1999) Absorption, translocation, and metabolism of primisulfuron and nicosulfuron in broadleaf signalgrass (*Brachiaria platyphylla*) and corn. *Weed Sci* 47:8–12
- Harrell MS, Williams DW, Brecke BJ (2005) Evaluation of sulfonylurea herbicides on cool and warm season turf species. *Appl Turf Sci*. DOI:10.1094/ATS-2005-1121-01-RS
- Hart SE, McCullough PE (2007) Annual bluegrass (*Poa annua*) control in Kentucky bluegrass (*Poa pratensis*) with bispyribac–sodium, primisulfuron, and sulfosulfuron. *Weed Technol* 21:702–708
- Hoagland DR, Arnon DI (1950) The Water-Culture Method for Growing Plants Without Soil. Berkeley, CA: California Agricultural Experiment Station Cir. No. 347. 31 p
- Hoseini SA, Mohassel MHR, Spliid NH, Mathiassen SK, Kudsk P (2011) Response of wild barley (*Hordeum spontaneum*) and winter wheat (*Triticum aestivum*) to sulfosulfuron: the role of degradation. *Weed Biol Manage* 11:64–71
- Jiang Y, Huang B (2001) Physiological response to heat stress alone or in combination with drought: a comparison between tall fescue and perennial ryegrass. *HortScience* 36:682–686
- Johnson BJ (1976) Transition from overseeded cool-season grass to warm-season grass with pronamide. *Weed Sci* 24:309–311
- Kraft RW, Keeley SJ, Su K (2004) Conversion of fairway-height perennial ryegrass turf to Kentucky bluegrass without nonselective herbicides. *Agron J* 96:576–579
- Lycan DW, Hart SE (2006) Foliar and root absorption and translocation of bispyribac–sodium in cool-season turfgrass. *Weed Technol* 20:1015–1022
- McCullough PE, Hart SE (2008) Roughstalk bluegrass and tall fescue control in Kentucky bluegrass with sulfosulfuron. *Appl Turf Sci*. DOI:10.1094/ATS-2008-0625-01-RS
- McCullough PE, Yu J, Brosnan JT, Breeden GK (2012) Relative tolerance of perennial ryegrass and tall fescue to flucarbazone. *Weed Technol* 26:673–678
- McElroy JS, Flessner ML, Murphy TR (2013) Bermudagrass growth regulation with flucarbazone. *Int Turf Res J* 12:197–200
- McElroy JS, Yelverton FH, Burke IC, Wilcut JW (2004) Absorption, translocation, and metabolism of halosulfuron and trifloxysulfuron in green kyllinga (*Kyllinga brevifolia*) and false-green kyllinga (*K. gracillima*). *Weed Sci* 52:704–710
- Mekki M, Leroux GD (2006) Foliar absorption and translocation of nicosulfuron and rimsulfuron in five annual weed species. *Weed Res* 35:377–383
- Niehaus ME (1976) Effect of cultivar, seeding rate, and fertilization on Kentucky bluegrass–perennial ryegrass turf mixtures. *Agron J* 68:955–957
- Novosel KM, Renner KA (1995) Nicosulfuron and primisulfuron root uptake, translocation, and inhibition of acetolactate synthase in sugarbeet (*Beta vulgaris*). *Weed Sci* 43:342–346
- Park KW, Fandrich L, Mallory-Smith CA (2004) Absorption, translocation, and metabolism of propoxycarbazon–sodium in ALS-inhibitor resistant *Bromus tectorum* biotypes. *Pest Biochem Phys* 79:18–24
- Pester TA, Nissen SJ, Westra P (2001) Absorption, translocation, and metabolism of imazamox in jointed goatgrass and feral rye. *Weed Sci* 49:607–612
- Richardson RJ, Hatzios KK, Wilson HP (2003) Absorption, translocation, and metabolism of CGA 362622 in cotton and two weeds. *Weed Sci* 51:157–162
- Richmond DS, Niemczyk HD, Shetlar DJ (2000) Overseeding endophytic perennial ryegrass stands of Kentucky bluegrass to manage bluegrass billbug (*Coleoptera: Curculionidae*). *J Econ Ent* 93:1662–1668
- Shearman RC, Beard JB (1975) Turfgrass wear tolerance mechanisms: I. wear tolerance of seven turfgrass species and quantitative methods for determining turfgrass wear injury. *Agron J* 67:208–211
- Shortell RR, Hart SE, Bonos SA (2008) Response of Kentucky bluegrass (*Poa pratensis* L.) cultivars and selections to bispyribac–sodium herbicide. *HortScience* 43:2252–2255
- Toler JE, Willis TG, Estes AG, McCarty LB (2007) Postemergent annual bluegrass control in dormant nonoverseeded bermudagrass turf. *HortScience* 42:670–672
- Trenholm LE, Carrow RN, Duncan RR (2000) Mechanisms of wear tolerance in seashore paspalum and bermudagrass. *Crop Sci* 40:1350–1357
- Trenholm LE, Duncan RR, Carrow RN (1999) Wear tolerance, shoot performance, and spectral reflectance of seashore paspalum and bermudagrass. *Crop Sci* 39:1147–1152
- Wehner DJ, Watschke TL (1981) Heat tolerance of Kentucky bluegrasses, perennial ryegrasses, and annual bluegrass. *Agron J* 73:79–84

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