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Short title: Dodhvlex™ Active Use in Turf

Turfgrass Tolerance to Tetflupyrolimet Applications for Preemergence Grassy Weed

Control

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Abstract

Tetflupyrolimet (DodhylexTM Active. FMC Corporation) is a novel herbicide inhibiting de novo pyrimidine biosynthesis that controls grassy weeds preemergence in rice (Oryza sativa L.) production. Field trials were conducted from 2021-2024 to evaluate turfgrass tolerance to tetflupyrolimet applications for annual bluegrass (Poa annua L.) and smooth crabgrass [Digitaria ischaemum (Schreb.) Schreb. ex Muhl.] control. Tolerance was evaluated on seven turfgrass species, including creeping bentgrass (Agrostis stolonifera L.), Kentucky bluegrass (Poa pratensis L.), tall fescue [Schedonorus arundinaceus (Schreb.) Dumort.; syn. Festuca arundinacea Schreb.], hybrid bermudagrass [C. dactylon (L.) Pers. x C. transvaalensis Burtt-Davy], and manilagrass [Zoysia matrella (L.) Merr.] at various mowing heights ranging from 3.8 to 12.5 mm. Separate experiments were conducted on each turfgrass species to evaluate tolerance in both fall and spring. Tetflupyrolimet was applied at rates of 0, 25, 50, 100, 200, 400, 800, 1600, 3200, or 6400 g ha⁻¹. No injury was observed on any warm-season turfgrass species in either season, whereas cool-season grass tolerance varied among species each season; however, cool-season turfgrass tolerance for all species was greater in spring than fall. While efficacy of tetflupyrolimet (400 g ha⁻¹) for preemergence D. ischaemum control varied among years, mixtures of tetflupyrolimet (400 g ha⁻¹), pyroxasulfone (128 g ha⁻¹), and rimsulfuron (35 g ha⁻¹) applied preemergence or early postemergence effectively controlled multiple resistant P. annua in both seasons. Overall, these findings highlight that warm-season turfgrasses are highly tolerant of tetflupyrolimet applications for *P. annua* or *D. ischaemum* control.

Keywords: Tetflupyrolimet; turfgrass tolerance; herbicide; efficacy; grass weed control.

Introduction

Tetflupyrolimet is a novel mode-of-action herbicide inhibiting de novo pyrimidine biosynthesis, a process essential to the production of both DNA and RNA, as well as phospholipids and glycoproteins (Kang et al. 2023). Tetflupyrolimet binds to dihydroorotate dehydrogenase (DHODH), an enzyme located on the outer surface of the inner mitochondrial membrane (Reis et al. 2017), impeding ubiquinone-mediated oxidation of dihydroorate to orotate (Zrenner et al. 2006).

Pyrimidine biosynthesis inhibition has been explored as a treatment for both cancers and viruses that are characterized by rapid cell proliferation (Zhou et al. 2021; Luban et al. 2021). However, chemical inhibitors of human-DHODH are not active in plants due to differences in amino acid sequence and structure that affect binding (Ullrich et al. 2002). For example, DHODH sequences from *Homo sapiens* and *Arabidopsis thaliana* were only 63.6% similar (Ullrich et al. 2002). In regards to tetflupyrolimet, Kang et al. (2023) reported *in vitro* I₅₀ values of 4.3 and 380 nM for foxtail millet [*Setaria italica* (L.) P. Beauv.] and *Homo sapiens*, respectively. Similarly, fungicides targeting DHODH, particularly ipflufenoquin, have little effect on pyrimidine biosynthesis in plants given less than 40% sequence identity between orthologs from *S. italica* and fungal pathogens such as *Aspergillus fumigatus* or *Cryptococcus* spp. (Kang et al. 2023). In the field, ipflufenoquin is not an effective herbicide whereas tetflupyrolimet offers minimal efficacy against fungal pathogens (Kang et al. 2023; van Rhijn et al. 2024).

Tetflupyrolimet is an effective preemergence herbicide to control troublesome grass weeds in rice (*Oryza sativa* L.), particularly barnyardgrass [*Echinochloa crus-galli* (L.) P. Beauv.] and junglerice [*Echinochloa colona* (L.) Link] (Castner et al. 2024a; Whitt et al. 2024). In rice, tetflupyrolimet has been mixed with other active ingredients (e.g., clomazone) to expand weed control spectrum, particularly versus dicot weeds (Acrement 2024a). However, a mixture of tetflupyrolimet and saflufenacil (70 g ha⁻¹) was antagonistic on spreading dayflower [*Commelina diffusa* Burm. f.]. When applied to silt loam, carryover potential with tetflupyrolimet is low (Castner et al. 2024b), in part due to its chemical properties [pKa = 12.8, $K_{SP} = 4.7 \text{ mg L}^{-1}$, $K_{OC} = 658 \text{ to } 1131 \text{ mL g}^{-1}$ (A. Puri, personal communication)].

Similar to *Echinochloa* spp. in rice, herbicide resistance in annual bluegrass (*Poa annua* L.) populations of turfgrass is widespread (Heap 2024; Brosnan et al. 2020a). A national survey

of herbicide resistance within *P. annua* populations of turfgrass reported that 42% of 1,349 populations collected exhibited putative resistance to at least one mode-of-action (McCurdy et al. 2023). At the state level, 25% of *P. annua* collected from golf courses across Tennessee was resistant to at least two herbicides (Brosnan et al. 2020b), with certain populations resistant to multiple mode-of-action groups including HRAC Groups #2, #3, #5, #29 (Brosnan et al. 2020c). Similar to previous reports in commodity agriculture (Schroeder et al. 2018), there is a strong desire amongst turfgrass managers for new herbicides to control herbicide-resistant *P. annua* (Allen et al. 2022), particularly in the southeastern United States where the issue is most prevalent (McCurdy et al. 2023; Rutland et al. 2023).

The first novel mode-of-action commercialized in decades, tetflupyrolimet could provide turfgrass managers a valuable tool for managing herbicide resistance in *P. annua*. However, there is minimal information available regarding the tolerance of commonly used warm- and cool-season turfgrass to tetflupyrolimet applications for controlling *P. annua* and *Digitaria* spp. which are both regarded as troublesome grassy weeds (Van Wychen 2020). This paper presents the results of several replicated trials conducted in Tennessee with a central objective of evaluating turfgrass tolerance and weed control efficacy following tetflupyrolimet applications for grassy weed control in fall and spring.

Materials and Methods

Turfgrass Tolerance

Field experiments to evaluate the tolerance of five turfgrass species to tetflupyrolimet (DodhylexTM Active. FMC Corporation. Philadelphia, PA) were conducted at the East Tennessee AgResearch and Education Center – Plant Sciences Unit (Knoxville, TN) during both fall and spring. Experiments exploring turfgrass tolerance to tetflupyrolimet in fall were initiated on 28 September 2022 and 30 September 2023. Experiments exploring turfgrass tolerance to tetflupyrolimet in spring were initiated on 28 February 2023 and 29 February 2024. Plots (1.2 by 1.2 m) in all studies were arranged in a randomized complete block design with four replications. Growing conditions during these experiments are presented in Table 1.

Separate experiments were conducted on cool- and warm-season turfgrass species commonly used on golf courses throughout the United States (Shaddox et al. 2023). These species included Kentucky bluegrass (*Poa pratensis* L., cv., 'HGT'), creeping bentgrass

(Agrostis stolonifera L., cv. 'L-93XD'), tall fescue (Schedonorus arundinaceus (Schreb.) Dumort.; syn. Festuca arundinacea Schreb., cv. 'Lebanon Winning Colors Plus'), manilagrass (Zoysia matrella L. Merr., cv. 'Trinity') and hybrid bermudagrass [C. dactylon (L.) Pers. x C. transvaalensis Burtt-Davy, cv. 'Latitude 36']. These species were established atop a clay loam (fine-loamy, siliceous, semiactive, thermic humic Hapludult) that is characterized in detail in Table 2.

The Kentucky bluegrass and tall fescue sites were maintained with a rotary mower (ZTrakTM Z915E. John Deere. Moline, Il 1.5 cutting width) at 75 and 100 mm, respectively. Slow-release nitrogen (GAL-Xe^{ONE®} Polymer Coated Controlled Release Fertilizer. J.R. Simplot Company. Boise, ID) was applied to these sites at 73 kg N ha⁻¹ in spring and 49 kg N ha⁻¹ in fall. Cyazofamid (Segway[®] SC. PBI-Gordon Corporation. Shawnee, KS) was applied at 1150 g ha⁻¹ to the tall fescue site to manage *Pythium aphanidermatum* during summer both years. Once soil temperatures (5 cm depth) measured \geq 18.3°C, pyraclostrobin (Insignia[®] SC Intrinsic Brand Fungicide. BASF Corporation. Research Triangle Park, NC) was applied at 556 g ha⁻¹ every 28 d through August to manage *Rhizoctonia solani*. Plant growth regulators were not applied to the tall fescue and Kentucky bluegrass sites during these experiments.

The creeping bentgrass, manilagrass, and hybrid bermudagrass sites were maintained as golf course fairways using a reel mower (7700A PrecisionCut™. John Deere. Moline, II 2.54 cutting width) at a 12.7 mm height of cut. All fairways received slow-release nitrogen (73 kg N ha⁻¹) in the spring. Additionally, creeping bentgrass received 12 kg N ha⁻¹ (Turf & Ornamentals ™ 18 N − 3 P₂O₅ 6 K₂O UMAXX®. J.R. Simplot Company. Boise, ID) from April through August, as well as 49 kg N ha⁻¹ (GAL-XeONE® Polymer Coated Controlled Release Fertilizer. J.R. Simplot Company. Boise, ID) in the fall. Isofetamid (Kabuto® Fungicide SC. PBI-Gordon Corporation. Shawnee, KS) was applied at 4100 g ha⁻¹ on manilagrass and hybrid bermudagrass sites once soil temperatures (5 cm depth) were < 18.3°C to prevent spring dead spot (*Ophiosphaerella herpotricha*). The creeping bentgrass site was treated with a preventative fungicide program that delivered two applications per month from April through August. Aluminum-tris (Signature™ XTRA Stressguard®. Envu Turf and Ornamentals. Cary, NC) and triadimefon1-(4-Chlorophenoxy)-3,3-dimethyl-1-(1H-1,2,4-triazol-1-yl)-2-butanone (Bayleton® Flo. Envu Turf and Ornamentals. Cary, NC) were applied in April and May at 1200 g ha⁻¹ and 1590 g ha⁻¹, respectively. Aluminum-tris (1200 g ha⁻¹) and cyazofamid (1150 g ha⁻¹) + a

commercially formulated mixture of fluopyram + trifloxystrobin [Exteris® Stressguard®. Envu Turf and Ornamentals. Cary, NC (38.9 and 62.4 g ha⁻¹, respectively)] were applied in June, July, and August. Plant growth regulators were not applied to the creeping bentgrass, manilagrass, or hybrid bermudagrass sites during these experiments.

Tolerance of manilagrass (cv. 'Prizm') and hybrid bermudagrass (cv. 'Mach 1') maintained as golf course putting greens was also evaluated in fall and spring each year. These species were maintained using a reel mower (Greensmaster® TriFlex™ 3300. The Toro Company. Bloomington, MN 1.5 m cutting width) at 3.8 mm on sand-based rootzones (Table 2). These sites received 12.2 kg N ha⁻¹ from a complete fertilizer (Turf & Ornamentals™ 18 N − 3 P₂O_{5 −} 6 K₂O UMAXX®. J.R. Simplot Company. Boise, ID) every 14 d from April through October each year. Both putting green sites were treated with a preventative fungicide program that delivered two applications per month. From April through August, commercially formulated mixtures of fluopyram (38.9 g ha⁻¹) + trifloxystrobin (62.4 g ha⁻¹) or pyraclostrobin (556 g ha⁻¹) + aluminum-tris (1200 g ha⁻¹) were applied, whereas cyazofamid (1150 g ha⁻¹) + aluminum-tris (1200 g ha⁻¹) or aluminum-tris (1200 g ha⁻¹). Syngenta Crop Protection. Greensboro, NC (12.5 and 42.2 g ha⁻¹, respectively) was applied from September through November. Plant growth regulators were not applied to the manilagrass or hybrid bermudagrass putting green sites during these experiments.

In each experiment, turfgrass was subjected to increasing doses of tetflupyrolimet using a CO₂-pressurized sprayer at 374 L ha⁻¹ via flat-fan nozzle tips (XR8002. TeeJet Technologies. Wheaton, IL). Tetflupyrolimet (Dodhylex[™] Active. FMC Corporation. Philadelphia, PA) application rates were 0, 25, 50, 100, 200, 400, 800, 1600, 3200, or 6400 g ha⁻¹. Overhead irrigation (5 mm) was applied within one hour of application in both fall and spring to ensure herbicide activation in the soil. Turfgrass injury was visually evaluated 14, 35, 95, and 180 days after treatment (DAT) relative to non-treated check plots (i.e., 0 g ha⁻¹ tetflupyrolimet) in each replication using a 0 (i.e., no phytotoxicity) to 100% (i.e., complete plant kill) scale.

Separate analyses were conducted to determine turfgrass tolerance to tetflupyrolimet in fall and spring on each species. Within a season (i.e., fall or spring trials), turfgrass injury data from a single species were subjected to combined analysis of variance (ANOVA) in R (v.4.2.2) using expected mean squares of McIntosh (1983) to determine if years could be combined. Non-

linear regression analysis was conducted in GraphPad Prism (v.10.1.1. Graphpad Prism. LaJolla, CA) using injury data collected 95 DAT. An 'EC anything' model was used to calculate the dose of tetflupyrolimet required to generate 25% turfgrass injury (EC₂₅ values) on each turfgrass species in fall or spring. EC₂₅ values were compared using 95% confidence intervals.

Controlling Herbicide-Resistant Poa annua

Field experiments were conducted on a hybrid bermudagrass (cv. 'Tifway') golf course fairway at Montgomery Bell State Park Golf Course (Burns, TN) during 2022 and 2023. Nontarget site resistance is suspected in *P. annua* from this location as resistance to multiple mode-of-action groups has been associated with elevated cytochrome P450 expression (Brosnan et al. 2024). Hybrid bermudagrass at this location was maintained at 1.3 cm during periods of active growth each year. Plots (measuring 1.5 by 1.5 m) were arranged in a randomized complete block design with four replications. The soil series at this location was 9.6% Hawthorne (loamy-skeletal, siliceous, semiactive, thermic Typic Dystrudepts)—Sulphura (loamy-skeletal, siliceous, semiactive, thermic Typic Dystrudepts) association + 90.4% Sengtown gravelly silt loam (fine, mixed, semiactive, thermic Typic Paleudalfs) with a pH of 5.5 (Table 2). Supplemental nutrition was not applied during the course of the study and the site was only irrigated via rainfall. This site received 70.4 cm of rainfall during the data collection period in 2022 compared to 63.7 cm in 2023.

A *P. annua* emergence model developed by Taylor et al. (2021) was used to time preemergence and early-postemergence herbicide applications each year. In 2022, treatments were applied preemergence on 14 September and early postemergence on 8 November. The 7-d average soil temperature (5 mm depth) on these dates was 23.6°C and 17.1°C, respectively. In 2023, treatments were applied preemergence on 26 September and early postemergence on 24 October. The 7-d average soil temperatures were 18.5°C and 14.8°C, respectively. *Poa annua* plants were not visible above the hybrid bermudagrass canopy when early postemergence treatments were applied either year.

Herbicides were applied with a CO₂-pressurized sprayer at 374 L ha⁻¹ via flat-fan nozzle tips (XR8002. TeeJet Technologies, Wheaton, IL). Preemergence treatments included tetflupyrolimet at 400 g ha⁻¹, pyroxasulfone (Zidua SC. BASF Corporation. Research Triangle Park, NC) at 128 g ha⁻¹, tetflupyrolimet (400 g ha⁻¹) + pyroxasulfone (128 g ha⁻¹), and indaziflam

(Specticle Flo. Envu Turf and Ornamentals. Cary, NC) at 45 g ha⁻¹. Early-postemergence treatments included tetflupyrolimet (400 g ha⁻¹), pyroxasulfone (128 g ha⁻¹), tetflupyrolimet + pyroxasulfone (400 g ha⁻¹ + 128 g ha⁻¹), and indaziflam + simazine (Princep 4FL. Syngenta Crop Protection. Greensboro, NC) + a commercially formulated mixture of thiencarbazone-methyl + foramsulfuron + halosulfuron (Tribute[®] Total. Envu Turf and Ornamentals. Cary, NC). Simazine and this commercially formulated mixture were combined with indaziflam (32.5 g ha⁻¹) at rates of 26.2 and 27.3 g ha⁻¹, respectively. All early postemergence herbicide applications were mixed with a non-ionic surfactant (Induce. Helena Agri-Enterprises. Memphis, TN) at 0.25%.

Hybrid bermudagrass injury data were evaluated using a 0 (i.e., no control) to 100 (i.e., complete kill) percent scale relative to non-treated check plots in each replication. *Poa annua* control data were collected using a similar percentage-based scale at 11, 19, 23, and 27 weeks after initial treatment (WAIT) each year. Similar to turfgrass tolerance trials, a combined ANOVA using expected mean squares of McIntosh (1983) was conducted in R to determine if years could be combined. No significant year-by-treatment interactions were detected; therefore, the combined data set was subjected to ANOVA with means separated using the LSD.test function found within the 'agricolae' package (Mendiburu & Simon, 2015).

Controlling Digitaria ischaemum

Field experiments were conducted on a bermudagrass [*Cynodon dactylon* (L.) Pers., cv. 'Yukon'] stand, maintained as a golf course fairway at East Tennessee AgResearch and Education Center – Plant Sciences Unit (Knoxville, TN) during 2021 and 2022 that was naturally infested with smooth crabgrass [*Digitaria ischaemum* (Schreb.) Schreb. ex Muhl.]. Bermudagrass at this location was maintained at 1.3 cm throughout active growth periods. Soil at this location was a Sequatchie loam (fine-loamy, siliceous, semiactive, thermic Humic Hapludults) with a pH of 5.7 (Table 2). Plots (1.5 by 1.5 m) were arranged in a randomized complete block design with four replications. Slow-release nitrogen (GAL-Xe^{ONE®} Polymer Coated Controlled Release Fertilizer. J.R. Simplot Company. Boise, ID) was applied at 73 kg N ha⁻¹ in April 2021 and 2022.

Herbicides were applied preemergence on 11 March 2021 and 11 March 2022 with a CO₂-pressurized sprayer at 374 L ha⁻¹ via flat-fan nozzle tips (XR8002. TeeJet Technologies, Wheaton, IL). Seven-day average soil temperatures (5 cm depth) on the date of study initiation

each year, were 14° C and 14.3° C, respectively. Treatments included tetflupyrolimet at 250, 300, and 400 g ha⁻¹, prodiamine (Barricade® 4FL. Syngenta Crop Protection. Greensboro, NC) at 840 g ha⁻¹, and indaziflam (45 g ha⁻¹). Within 24 hours of application, ≥ 0.6 cm of precipitation fell each year and 0.5 cm of irrigation was also applied via an in-ground irrigation system at this location. In 2021 and 2022, total precipitation during the data collection period was 45.8 cm and 54.8 cm, respectively.

Digitaria ischaemum control was evaluated using a 0 (i.e., no control) to 100% (i.e., complete plant death) scale with a non-treated check plot in each replication through 145 DAT. A combined ANOVA using expected mean squares of McIntosh (1983) was conducted in R to determine if years could be combined. A significant year-by-treatment interaction was detected; therefore, data from each year were subjected to ANOVA with means separated using the LSD.test function found within the 'agricolae' package (Mendiburu & Simon, 2015).

Results and Discussion

Turfgrass Tolerance

For several of the tested turfgrass species, significant year-by-treatment interactions were detected in both fall and spring datasets; therefore, years were analyzed separately. However, no injury was detected on hybrid bermudagrass or manilagrass (regardless of mowing height) with fall or spring applications of tetflupyrolimet applications up to 4800 g ha⁻¹ in either year (data not shown).

Differential responses to tetflupyrolimet applications in fall were detected among coolseason turfgrasses each year (Table 3, Figure 1). The dose of tetflupyrolimet to induce 25% injury to tall fescue (EC₂₅) in fall was 956 g ha⁻¹ in 2022 and 904 g ha⁻¹ in 2023 at 95 DAT (Table 3). EC₂₅ values following tetflupyrolimet applications to Kentucky bluegrass and creeping bentgrass in fall varied among years. For example, EC₂₅ values for Kentucky bluegrass were 704 g ha⁻¹ in 2022 and 1407 g ha⁻¹ in 2023. On creeping bentgrass, EC₂₅ values were 806 g ha⁻¹ in 2022 and 465 g ha⁻¹ in 2023. Differences were also detected among cool-season turfgrasses in response to tetflupyrolimet applications in spring each year (Table 3, Figure 1). Using EC₂₅ values, tolerance of cool-season turfgrass to tetflupyrolimet in spring 2023 was greatest for tall fescue, lowest for creeping bentgrass, with Kentucky bluegrass ranking intermediate. In 2024,

Kentucky bluegrass exhibited the greatest tolerance to tetflupyrolimet of all cool-season turfgrass species treated in spring.

Differential tolerance among cool-season turfgrass species has been reported and attributed to biokinetics (Yu et al. 2013; McCullough et al. 2009). For example, amicarbazone absorption and translocation are greater in *P. annua* than in creeping bentgrass or tall fescue; metabolism is also decreased in *P. annua* compared to those species as well (Yu et al. 2013). Differential metabolism of bispyribac-sodium contributes to its selectivity for *P. annua* control and tolerance among creeping bentgrass and perennial ryegrass (McCullough et al. 2009). In cereals, herbicide tolerance has also been associated with herbicide metabolism as well (Pester et al. 2000). It is possible that tetflupyrolimet absorption, translocation, or metabolism may vary among tall fescue, Kentucky bluegrass, and creeping bentgrass. Further research with ¹⁴C-tetflupyrolimet to better understand these relationships is warranted.

Interestingly, cool-season turfgrass tolerance to tetflupyrolimet was greater in spring than fall. Across all cool-season species, EC₂₅ values following fall applications of tetflupyrolimet ranged from 465 to 1407 g ha⁻¹ compared to 1415 to 6655 g ha⁻¹ in spring (Table 3). Increased growth potential of cool-season turfgrasses in spring may explain the increased tolerance to tetflupyrolimet compared to applications made in fall. Growth potential is a metric used to quantify potential growing conditions for warm- and cool-season turfgrass based on air temperature (Gelernter and Stowell 2005). Units range from 0 to 100% with scores of 100% equating to optimal growing conditions, > 50% representing adequate growing conditions, and values ≤ 10% indicating that conditions would severely limit growth (Gelernter and Stowell 2005). Growth potential values during fall and spring tolerance trials are presented in Figure 2. Applications of tetflupyrolimet in fall were made following a summer stress period where growth potential was $\leq 71\%$, and aside from a one-month recovery period in September (when treatments were applied), declined thereafter. In spring, growing conditions linearly increased prior to applying treatments in March for several weeks thereafter (Figure 2). While not directly linked to growth potential, the varying efficacy of amicarbazone and bispyribac-sodium in fall and spring has been associated with changes in air temperature (Yu et al. 2015). Methiozolin dissipation following applications to actively growing cool-season turfgrass is also more rapid

than bare soil further illustrating the effects of turfgrass growing conditions on herbicide activity (Peppers et al. 2024).

Controlling Herbicide-Resistant Poa annua

Significant differences in P. annua control were detected among treatments from 11 to 27 WAIT (Table 4). When applied preemergence, all herbicides provided effective ($\geq 89\%$) control of this resistant population 11 WAIT with differences manifesting thereafter. This response aligns with those observed in rice where tetflupyrolimet applications at 100 g ha⁻¹ followed by carfentrazone-ethyl (530 g ha⁻¹) controlled E. crus-galli and bearded sprangletop [Leptochloa fusca (L.) Kunth; syn. Diplachne fusca (L.) P. Beauv.] to ≥ 99% for 4 weeks (Lombardi and Al-Khatib 2023). Given that *Poa annua* control was assessed in dormant hybrid bermudagrass that offered little plant competition, the efficacy of tetflupyrolimet applied alone at 400 g ha-1 declined to 69% control by 19 WAIT. By 27 WAIT, indazflam only controlled this resistant Poa annua population 3% compared to 40% for tetflupyrolimet. It should be noted that differential expression of a cytochrome P450 ortholog similar to CYP81A10v7 found in non-target site resistant rigid ryegrass (Lolium rigidum L.) from Australia (Han et al. 2021) has been identified in P. annua from this location (Brosnan et al. 2024). PRE applications tetflupyrolimet + pyroxasulfone controlled this resistant population 89% by 23 WAIT. Effective preemergence control of *Poa annua* for a 23-week period is similar to previous reports with industry standards applied to herbicide-susceptible populations (Brosnan et al. 2014). As an inhibitor of very-long chain fatty acid synthesis (HRAC Group #15), pyroxasulfone exhibits activity on grass and dicot weeds (Anonymous 2017). Including pyroxasulfone in mixture with tetflupyrolimet would expand the spectrum of weeds controlled via a single application and steward against the evolution of resistance to this novel Group #28 herbicide (Busi et al. 2020).

No significant differences were detected between early postemergence treatments in this study (Table 4). Tetflupyrolimet + rimsulfuron and tetflupyrolimet + pyroxasulfone + rimsulfuron controlled resistant *P. annua* at this location 89 to 95% by 23 WAIT, similar to indaziflam + simazine + a commercially formulated mixture of thiencarbazone-methyl + foramsulfuron + halosulfuron (88%). This level of *P. annua* control is similar to previous reports with early postemergence applications of pendimethalin + dimethenamid-P (Carroll et al. 2021), highlighting that tetflupyrolimet could be effectively incorporated into a resistance management

program at either preemergence or early postemergence timings when combined with herbicides from other mode-of-action groups.

Controlling Digitaria ischaemum

Year-by-treatment interactions were detected in *D. ischaemum* control data; therefore, data from each year were analyzed and are presented separately (Table 5). *Digitaria ischaemum* control efficacy with tetflupyrolimet 8 WAIT in this study aligns with reports of bearded sprangletop control in rice 7 WAIT with tetflupyrolimet (100 g ha⁻¹) in mixture with carfentrazone-ethyl (530 g ha⁻¹) (Lombardi and Al-Khatib 2023). Efficacy for *D. ischaemum* control declined beyond the 8 WAIT assessment each year. When applied at 400 g ha⁻¹, tetflupyrolimet only controlled *D. ischaemum* \geq 81% for 13 weeks in 2021. However, in 2022, the same treatment controlled *D. ischaemum* \geq 88% for 21 weeks. A similar relationship was observed with tetflupyrolimet rates \leq 300 g ha⁻¹ as well; however, overall efficacy with these applications tended to be lower than the 400 g ha⁻¹ rate in 2021. Prodiamine and indaziflam performed similarly each year with *D. ischaemum* control ranging from 89 to 98%, similar to previous reports (Brosnan et al. 2011).

Differences in *D. ischaemum* control between years could be attributed to differences in moisture availability. In 2021, this site received 11.3 cm of rainfall during the first 14 d of the trial compared to only 2.2 cm in 2022. Labeling for industry standard herbicides used for preemergence *D. ischaemum* control in turfgrass recommends ≤ 1.3 cm of irrigation or rainfall for activation (Anonymous 2021). It is possible that increased rainfall during the initial 14 d of this study in 2022 may have compromised tetflupyrolimet efficacy. In wheat (*Triticum* spp.), simulated rainfall as low as 0.5 mm 14 d after treatment resulted in leaching of prosulfocarb, pyroxasulfone, and trifluralin, with this response being affected by rainfall timing after application more than rainfall intensity (Khalil et al. 2019). Additional research to better understand effects of soil moisture on tetflupyrolimet efficacy is warranted.

Overall, our findings highlight that tetflupyrolimet has the utility of controlling problematic weeds of warm and cool-season turfgrass, particularly herbicide-resistant *P. annua*. We observed no injury on hybrid bermudagrass or manilagrass cultivars with tetflupyrolimet applications up to 4800 g ha⁻¹ maintained at mowing heights ranging from 3.8 to 12.5 mm. This response suggests that the warm-season turfgrasses in this study are tolerant to tetflupyrolimet

applications for controlling *P. annua* or *D. ischaemum*. On the cool-season turfgrasses in this

study, tolerance to tetflupyroliment was greater in spring than fall and is likely affected by

growth potential during the time period preceding and after treatment. Further research to better

understand this phenomenon is warranted, along with efforts to understand effects of post-

application moisture on tetflupyrolimet efficacy given year-by-treatment interactions observed

herein. Efficacy for controlling herbicide-resistant P. annua in this study suggests that

evaluations on other resistant biotypes are warranted, as well as efforts to understand tolerance

following applications to turfgrass species and cultivars not evaluated herein.

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Competing Interests

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Table 1. Growing conditions during fall and spring experiments evaluating tolerance of five turfgrass species to tetflupyrolimet (Dodhylex[™] Active. FMC Corporation. Philadelphia, PA) conducted in Knoxville, TN. Fall experiments were initiated on 28 September 2022 and 30 September 2023. Spring experiments were initiated on 28 February 2023 and 29 February 2024. In all studies, data were collected for 180 days. All weather data obtained from the mesur.io Earthstream[®] platform (mesur.io, Yanceyville, NC).

	Fall		Spring	
	2022	2023	2023	2024
Air temperature (°C)	9.3	9.2	19.5	18.1
Soil temperature (°C)	10.7	10.7	19.6	17.8
Humidity (%)	75	75	74	74
Precipitation (cm)	73.7	63.1	81.3	85.6

Table 2. Physical and chemical properties of the soils in place for all experimental trial work evaluating efficacy and tolerance of tetflupyrolimet (DodhylexTM Active. FMC Corporation. Philadelphia, PA) at Montgomery Bell State Park Golf Course (Burns, TN) and the East Tennessee Research and Education Center (ETREC) – Plant Science Unit (Knoxville, TN).

	Soil analysis							
Location	pН	TEC ^z	OM % ^z	% Sand	% Silt	% Clay		
Montgomery Bell State Park Golf Course	5.5	14.04	4.72	21.2	61.5	17.3		
ETREC – Plant Science Unit ^x	5.7	6.71	2.20	38.6	35.5	25.9		
Sand-based rootzone ^w	6.5	2.74	1.35	98.7	0.3	1.0		

^z Abbreviation: TEC = total exchange capacity (ME / 100 g), OM % = organic matter % (360° LOI), BD = bulk density (g cm⁻³), TP = total porosity, CP = capillary porosity.

^y Bulk density (BD) calculated at 75% of the maximum density according to ASTM D698-12 2021. Standard test methods for laboratory compaction characteristics of soil using standard effort (12 400 ft-lbf/ft3 (600 kN-m/ m3)). West Conshohocken, PA: ASTM International.).

^x Soil type during all turfgrass tolerance and efficacy trials conducted at the East Tennessee AgResearch and Education Center-Plant Sciences Unit on plots maintained at heights of cut > 12.5 mm.

^w Soil type during all turfgrass tolerance trials at the East Tennessee AgResearch and Education Center-Plant Sciences Unit on plots maintained as golf course putting greens (3.8 mm height-of-cut).

Table 3. Rate of tetflupyrolimet (Dodhylex[™] Active. FMC Corporation. Philadelphia, PA) to induce 25% injury (EC₂₅) at 95 days after treatment on Kentucky bluegrass (*Poa pratensis* L., cv., 'HGT'), creeping bentgrass (*Agrostis stolonifera* L., cv. 'L-93XD'), and tall fescue (*Festuca arundinacea* Schreb., cv. 'Lebanon Winning Colors Plus') during fall and spring. Fall tolerance experiments were initiated on 28 September 2022 and 30 September 2023, while spring experiments were initiated on 28 February 2023 and 29 February 2024. All studies were collected at the East Tennessee AgResearch and Education Center – Plant Sciences Unit (Knoxville, TN).

	Tetflupyrolimet rate for 25% injury at 95 DAT (EC ₂₅)									
	Fall 2022		Fall 2023		Spring 2023		Spring 2024			
	EC_{25}	CI	EC_{25}	CI	EC_{25}	CI	EC_{25}	CI		
Creeping bentgrass	806	662 to 963	465	433 to 497	1415	1112 to 1771	2200	1719 to 2751		
Kentucky bluegrass	702	578 to 842	1407	1306 to 1500	1724	1485 to 1990	6655	5987 to 8580		
Tall fescue	956	813 to 1114	904	849 to 963	2556	undefined	1581	1410 to 1764		

^z Abbreviation: CI = 95% confidence interval.

Table 4. *Poa annua* control with preemergence and early postemergence applications of tetflupyrolimet (Dodhylex[™] Active. FMC Corporation. Philadelphia, PA) alone and in mixtures. Applications made to a hybrid bermudagrass (*C. dactylon* x *C. transvaalensis*, cv. 'Tifway') golf course fairway at Montgomery Bell State Park Golf Course (Burns, TN) infested with herbicide-resistant *Poa annua*.

			Poa annua control					
Timing	Herbicide	Rate	11 WAIT ^z	19 WAIT	23 WAIT	27 WAIT		
		g ha ⁻¹		%	%			
PRE	Tetflupyrolimet	400	98	69	57	40		
	Pyroxasulfone	128	100	86	68	57		
	Tetflupyrolimet + pyroxasulfone	400 + 128	99	89	89	68		
	Indaziflam	45	89	48	14	3		
EPOST	Tetflupyrolimet + rimsulfuron	400 + 35	100	87	89	71		
	Tetflupyrolimet + pyroxasulfone + rimsulfuron	400 + 128 + 35	99	94	95	83		
	Tribute [®] Total + simazine + indazflam	42 + 1120 + 26	100	88	88	66		
	P > F		*** ^X	***	***	***		
	$\mathrm{LSD}_{0.05}$		2	15	20	20		

² Abbreviations: WAIT = weeks after initial treatment.

***, **, * significant at $p \leq 0.001$, and 0.05, respectively.

^y Tribute[®] Total (Envu Turf and Ornamentals. Cary, NC) is a commercialized formulation of the following active ingredients: thiencarbazone-methyl + foramsulfuron + halosulfuron.

Table 5. *Digitaria ischaemum* control with preemergence applications of tetflupyrolimet (DodhylexTM Active. FMC Corporation. Philadelphia, PA) compared to industry standards. Applications made to a bermudagrass (*Cynodon dactylon*, cv. 'Yukon') stand, maintained as a golf course fairway at East Tennessee Research and Education Center – Plant Science Unit (Knoxville, TN).

	Rate	Digitaria ischaemum control									
		2021					2022				
Herbicide		8 WAIT ^z	13 WAIT	17 WAIT	21 WAIT	_	8 WAIT	13 WAIT	17 WAIT	21 WAIT	
	g ha ⁻¹					%					
Tetflupyrolimet	250	96	56	28	8		100	89	79	75	
Tetflupyrolimet	300	98	66	23	8		100	95	90	89	
Tetflupyrolimet	400	100	81	44	40		100	96	95	88	
Prodiamine	840	100	99	96	96		100	96	99	98	
Indaziflam	45	100	98	92	89		100	96	90	90	
P > F		ns	** ^y	***	***		ns	ns	ns	*	
$LSD_{0.05}$		ns	25	35	26		ns	ns	ns	14	

² Abbreviations: WAIT = weeks after initial treatment; ns = non-significant.

y ***, **, * significant at p \leq 0.001, 0.01, and 0.05, respectively.

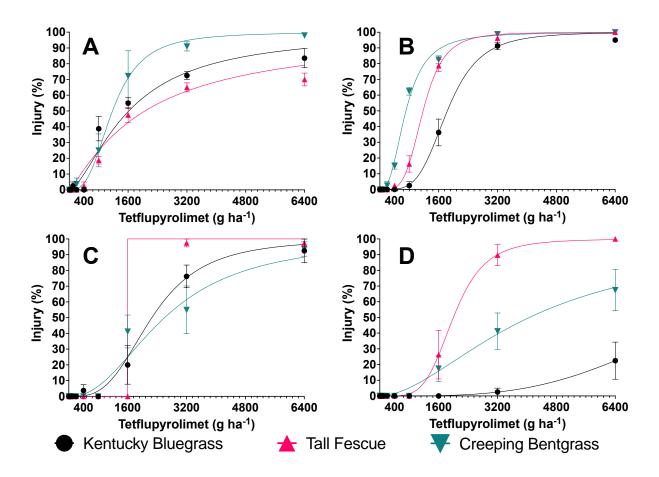


Figure 1. Dose response curves for Kentucky bluegrass (*Poa pratensis* L., cv., 'HGT'), creeping bentgrass (*Agrostis stolonifera* L., cv. 'L-93XD'), and tall fescue (*Festuca arundinacea* Schreb., cv. 'Lebanon Winning Colors Plus') treated with increasing rates of tetflupyrolimet (Dodhylex™ Active. FMC Corporation. Philadelphia, PA). Panels A and B present responses from applications made in fall of 2022 and 2023, respectively, in Knoxville, TN. Panels C and D present responses from applications made in spring of 2023 and 2024, respectively, in Knoxville, TN.

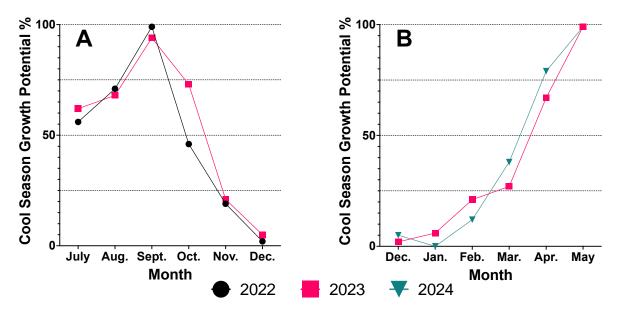


Figure 2. Temperature based growth potential (%) for cool-season turfgrasses during trials evaluating tolerance to increasing rates of tetflupyrolimet (Dodhylex™ Active. FMC Corporation. Philadelphia, PA) at East Tennessee Research and Education Center − Plant Science Unit (Knoxville, TN) in fall and spring. Fall tolerance trials on these cool-season grasses were initiated on 28 September 2022 and 30 September 2023; spring tolerance trials were initiated on 28 February 2023 and 29 February 2024. Panel A presents growth potential (%) three months prior to and after applications of tetflupyrolimet in fall 2022 and 2023; Panel B presents growth potential (%) three months prior to and after applications of tetflupyrolimet in spring 2023 and 2024.