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DOI: 10.1017/wet.2024.112

Soil Residual Activity of Tetflupyrolimet and the Influence of Soil Moisture and Flood Timeliness on Barnyardgrass Efficacy

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Running title: Tetflupyrolimet Soil Activity

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ABSTRACT

Tetflupyrolimet is the first herbicide with a novel site of action to be commercialized for use in agronomic crops in three decades. Direct-seed rice field experiments were conducted at research facilities near Stuttgart (silt loam), AR, and Keiser (clay), AR, to evaluate tetflupyrolimet as a preemergence herbicide versus commercial standards. Greenhouse experiments determined the influence of soil moisture on pre- and postemergence (POST) barnyardgrass control with tetflupyrolimet and clomazone, and the impact of a delayed flood on efficacy when POST-applied. For the field experiments, clomazone, tetflupyrolimet, and quinclorac were applied individually PRE at 336 and 560, 134 and 224, and 336 and 560 g ai ha⁻¹, respectively, on a silt loam and clay soil, along with clomazone plus tetflupyrolimet and clomazone plus quinclorac at the same rates. The soil moisture experiment included a single PRE and POST application of clomazone at 336 g ai ha⁻¹, tetflupyrolimet at 134 g ai ha⁻¹, and a mixture at the respective rates, on a silt loam soil at 50, 75, and 100% of field capacity. For the flood timing experiment, tetflupyrolimet was applied to 2- to 3-leaf barnyardgrass at 134 g ai ha⁻¹ and a flood was established at 4 hr after treatment (HAT) and 5 and 10 d after treatment (DAT). Barnyardgrass control with a tetflupyrolimet and clomazone mixture was comparable to clomazone plus quinclorac when averaged over all evaluations on silt loam and clay texture soils (≥91%). Soil moisture interacted with herbicide treatments for PRE and POST barnyardgrass efficacy when averaged over DAT, with tetflupyrolimet plus clomazone generally providing the greatest and most consistent control across regimes. Flooding barnyardgrass at 4 HAT provided superior control to later flood timings. Tetflupyrolimet is an effective residual barnyardgrass herbicide, and the addition of clomazone will aid in providing consistent control across varying soil moisture conditions.

Nomenclature: Clomazone; quinclorac; tetflupyrolimet; barnyardgrass, *Echinochloa crus-galli* (L.) P. Beauv.; rice, *Oryza sativa* L.

Key words: Flood timing, postemergence, preemergence, soil moisture

INTRODUCTION

Tetflupyrolimet is the first herbicide with a novel site of action (SOA) for use in agronomic crops since the commercialization of hydroxyphenylpyruvate dioxygenase- and glutamine synthetase-inhibiting herbicides 30 years ago (Duke 2012; Duke and Dayan 2022). Tetflupyrolimet is a Herbicide Resistance Action Committee (HRAC)/Weed Science Society of America (WSSA) Group 28, de novo pyrimidine biosynthesis inhibitor (orotate pathway), which targets the dihydroorotate dehydrogenase (DHODH) enzyme. The DHODH enzyme facilitates and catalyzes the oxidation reaction of dihydroorotate to orotate (Zrenner et al. 2006; Dayan 2019; Duke and Dayan 2022). Inhibition of DHODH prevents the downstream formation of uridine monophosphate from several precursors in the orotate pathway. It is lethal to most organisms due to the critical role of nucleotide production in plant growth and development. Inhibition activity of tetflupyrolimet on DHODH enzyme was approximately 10-fold greater on *Setaria* sp. in comparison to rice; however, the selectivity for the latter is magnitudes greater than sensitive weed species, suggesting that differences in metabolism may confer tolerance (Dayan 2019; Selby et al. 2023). The evaluated compounds of this new SOA have been documented to be specifically active toward monocotyledon weeds, with tetflupyrolimet having a high level of crop safety and effectiveness in paddy rice systems (Selby et al. 2023).

An extensive volume of direct-seeded and transplanted rice field experiments with tetflupyrolimet have been conducted in Brazil, India, Indonesia, Japan, the United States, and Vietnam with success in controlling economically important grass weed genus' (*Echinochloa*, *Leptochloa*, and *Monochoria*) (Selby et al. 2023). Tetflupyrolimet exhibits preemergence (PRE) and postemergence (POST) control of sensitive grass species, although PRE applications are generally more effective. Tetflupyrolimet provides a high level of PRE weed control in paddy rice at 125 g ai ha⁻¹ and up to 90% POST efficacy at 250 g ai ha⁻¹ with no visible injury to rice (Selby et al. 2023). Furthermore, field and greenhouse data confirm the effectiveness of tetflupyrolimet on barnyardgrass and its excellent crop safety margin that is not necessarily exclusive to rice (Castner, unpublished data).

FMC Corporation will likely position tetflupyrolimet as a PRE herbicide to be used in a mixture with clomazone to preserve the longevity of both SOAs and broaden the spectrum of grass weed control. Mixing different SOAs has been proven to be one of the most effective chemical-based management strategies for mitigating the evolution of herbicide-resistant weeds

by alleviating selection pressure often imposed by a single SOA (Norsworthy et al. 2012; Barbieri et al. 2022). Since its commercialization in the early 2000s, clomazone has been widely adopted as a soil-applied herbicide in mid-southern United States rice production for effective control of imidazolinone-, propanil-, and quinclorac-resistant barnyardgrass populations despite a few fields having confirmed resistance to the herbicide (Baltazar and Smith 1994; Carey et al. 1995; Scherder et al. 2004; Norsworthy et al. 2007; Heap 2024). In addition to barnyardgrass, clomazone can effectively control broadleaf signalgrass (*Urochloa platyphylla* Munro ex Wright) *Digitaria* spp., and *Panicum* spp. (Anonymous 2021). Unlike clomazone, a caveat to tetflupyrolimet is that its activity is primarily confined to, or is most effective on *Echinochloa* species, which further identifies the need and advantage of mixing with clomazone from an efficacy and herbicide-resistance management perspective.

A major disadvantage of direct-seeded paddy rice systems is the length of time irrigation can take to uniformly cover an entire field, which may be 10 or more days, depending upon rainfall, field size, and pumping capacity (Norsworthy, Distinguished Professor, personal communication). In some instances, rainfall is limiting, and irrigation must be used to activate soil-applied herbicides or allow the herbicide to be placed into the soil solution to become bioavailable. Suppose a field takes up to 10 d to receive adequate irrigation during dry weather. In that case, there may be variation in weed control in areas where activation was immediate, as opposed to those delayed in being activated. The field experiments mentioned in the manuscript by Selby et al. (2023) indicate that the efficacy of tetflupyrolimet was not compromised under a variety of conditions, although not specific in terms of the range of environmental parameters. It is common for delayed activation (generally less than 1.3 cm of irrigation or rainfall within 7 d after planting) to reduce the efficacy of soil-applied herbicides (Barnes and Oliver 2004), especially if weeds begin to germinate before irrigation or rainfall occurs (Anonymous 2022), although there are some exceptions, such as dicamba (Shaner et al. 2014; Anonymous 2022).

Furrow-irrigated rice (FIR) has become an increasingly popular alternative production system in Arkansas to simplify crop rotation and management practices, accounting for approximately 18% of hectares as of 2022 (Hardke 2022). However, approximately 78% of the rice area is produced in a direct-seeded, delayed-flood system, with silt loam soils contributing to 56.9% of those hectares in Arkansas. Weed management programs can be particularly more challenging in FIR systems than a direct-seeded, delayed-flood system. Frequent wetting and the

absence of a permanent flood create an ideal environment for weed management to become more synonymous with row crop production and is, therefore, more reliant on residual herbicides than a typical paddy rice system (Norsworthy et al. 2008). The shift in the weed spectrum can make management more challenging, often leading to more herbicide applications. Because such an emphasis is placed on soil-applied herbicides in a FIR system, consistent performance is imperative across various soil moisture regimes.

As tetflupyrolimet nears commercial launch, it is important to understand the general efficacy of the herbicide as an individual component as well as the advantages of mixing with clomazone, specifically on barnyardgrass where management across the mid-southern U.S. can be challenging due to the prevalence of herbicide resistance (Talbert and Burgos 2007). Mixing tetflupyrolimet serves two purposes: mitigating herbicide resistance risk by combining two effective SOAs and increasing the spectrum of grass weed control. In addition to establishing the level of expected control with tetflupyrolimet as a standalone herbicide or in a mixture with clomazone, the impact of environmental conditions and management practices on efficacy must be evaluated to define expectations. Therefore, experiments were designed 1) to evaluate the efficacy of tetflupyrolimet on barnyardgrass and other grass species compared to commercial standards based on a single PRE application, 2) to determine if soil moisture influences the level of control of tetflupyrolimet, clomazone, and the mixture on barnyardgrass PRE and POST, and 3) to determine if the variability of water movement across a paddy rice field influences POST performance of tetflupyrolimet.

MATERIALS AND METHODS

Soil Residual Activity Experiment. To determine the effectiveness of tetflupyrolimet in comparison to other commercial PRE standards (clomazone, quinclorac, and clomazone + quinclorac) and to quantify the length of residual control over time, three field experiments were conducted from 2021 to 2023 on a clay and a silt loam soil. All silt loam experiments were conducted at the Rice Research and Extension Center, near Stuttgart, AR, on a Dewitt silt loam soil (19% sand, 64% silt, and 17% clay with 1.1% organic matter) with a pH of 5.7. Experiments on the clay soil were at the Northeast Research and Extension Center, in Keiser, AR, on a Sharkey clay (41% sand, 1% silt, 58% clay, with 2.8% organic matter) with a pH of 5.5. Each field experiment was conducted once per year at the respective location and included four replications. Before planting, each field was subjected to conventional tillage events to

prepare the seedbed. The experiment was arranged as a single-factor randomized complete block design with four replications, and each plot measured 1.8 m wide by 5.2 m long. Herbicide treatments on the silt loam soil consisted of tetflupyrolimet at 134 g ai ha⁻¹, clomazone at 336 g ai ha⁻¹, quinclorac at 336 g ai ha⁻¹, tetflupyrolimet plus clomazone (134 and 336 g ai ha⁻¹, respectively), and clomazone plus quinclorac (336 and 336 g ai ha⁻¹, respectively). The herbicide treatments were adjusted to the recommended rates for clay soil, where each respective rate was increased by a factor of 1.7.

The rice cultivar, “Diamond,” (conventional, very short-season, long-grain, inbred) (University of Arkansas System Division of Agriculture, Little Rock, AR) was planted at the silt loam site on May 14, April 30, and May 10, in 2021, 2022, and 2023, respectively. The same rice cultivar was planted at the clay site on May 20, May 10, and May 4 in 2021, 2022, and 2023, respectively. All applications were made immediately after planting using a hand-held backpack sprayer calibrated to deliver 140 L ha⁻¹ at 4.8 km hr⁻¹ equipped with AIXR 110015 nozzles (TeeJet, Glendale Heights, IL). Soil test potassium and phosphorus concentrations were addressed with samples collected in the fall before the start of each growing season and were amended before planting for each site-year. Immediately prior to flood establishment, the silt loam site received 168 kg ha⁻¹ of nitrogen, and an additional 30 kg ha⁻¹ was applied at the clay site in the form of urea (Roberts et al. 2016). Once rice in each experiment reached the 5-leaf growth stage or tillering, a permanent flood was established until harvest maturity. Non-target broadleaf and sedge weeds were managed with the recommended rate of conventional rice herbicides with no activity on grass weed species, such as bentazon, halosulfuron, halosulfuron plus prosulfuron, or 2,4-D.

Visible rice injury, visible barnyardgrass and broadleaf signalgrass control, and weed density (two 0.25 m² quadrats per experimental unit) assessments were collected at 21, 28, 35, and 42 d after treatment (DAT) following the PRE applications on the silt loam soil. The same assessments were collected at 14, 28, and 42 DAT on the clay soil; however, barnyardgrass was the only grass weed species evaluated on the clay soil. Each visible assessment was evaluated on a 0 to 100% scale, with 0% representing no injury or control and 100% representing crop death or complete control (Frans and Talbert 1977). Additionally, rice maturity was assessed by recording when at least 50% of the panicles within a plot were present. At full maturity, a 1.8-m-

wide swath was harvested using a small-plot combine (Almaco, Nevada, IA), and grain yield was adjusted to 12% moisture.

For each location, site-year and replication were considered random to allow for generalizations to be drawn for visible rice injury, barnyardgrass and broadleaf signalgrass control, cumulative weed density, and rice grain yield. The silt loam and clay soil were analyzed independently due to differences in soil texture, weed species present, and herbicide rates. Excluding grain yield, a repeated measures analysis, including herbicide and DAT as fixed effects, was conducted for all response variables at each location and was only significant for broadleaf signalgrass cumulative density (silt loam), and barnyardgrass cumulative density (clay). Values were averaged over all evaluation dates for response variables that had a main effect of herbicide treatment and did not have an interaction between herbicide treatment and DAT. A single-factor analysis (herbicide treatment) was used for rice grain yield because the response could only be assessed at a single point in time. All distributions were analyzed using the JMP PRO 17.1 (SAS Institute Inc., Cary, NC) distribution platform (Avent et al. 2022) and residuals of the injury, weed control, and rice grain yield data assumed a normal distribution. Weed densities assumed a Poisson distribution (Gbur et al. 2012). All data were analyzed in JMP PRO 17.1 and subjected to analysis of variance (ANOVA) using the fit model platform. Means were separated using Tukey's honest significant difference (HSD) ($\alpha=0.05$).

Soil Moisture Experiment. To evaluate the influence of soil moisture on the PRE and POST efficacy of tetflupyrolimet and clomazone individually, as well as the mixture of the two herbicides, two separate PRE and POST greenhouse experiments were initiated and repeated three times on a silt loam soil with three replications. Each experiment was conducted at the Milo J. Shult Research and Extension Center, in Fayetteville, AR, in 2023 and 2024. A Captina silt loam soil (Fine-silty, siliceous, active, mesic Typic Fragiudults) (USDA-NRCS 2022) with 20% sand, 66% silt, 14% clay, and 2.3% organic matter (Arkansas Agricultural Diagnostic Laboratory, Fayetteville, AR 72701) was collected and sieved to remove large pieces of residue and reduce the size of soil aggregates. The sieved soil was dried at 65 C for two weeks until no moisture was present. Once dried, 4,500 g of soil was added to 3.8 L buckets (no drainage). Soil bulk density and volumetric field capacity were calculated using Soil Plant Air Water software (SPAW) (USDA ARS, Washington DC) using soil texture and organic matter inputs to determine

the appropriate volume of water to maintain 50, 75, and 100% field capacity of the soil (Equation 1).

$$MW = \frac{VFC}{BD} \times \%M \times MS$$

The methodology used for establishing the desired soil moisture regimes is directly adapted from Avent et al. (2023), where *MW* represents the estimated mass of water required (1 g water = 1 ml water), *VFC* is the volumetric field capacity generated from SPAW, *BD* is the bulk density computed by SPAW using texture and organic matter, *%M* is the percent soil moisture established as the testing parameter, and *MS* is the mass of dried soil (31.5% ÷ 1.42 x 100% x 4,500 g = 998 g of water).

For each experiment, the greenhouse was set to provide a 14-h photoperiod with day and night temperatures of 32 C and 24 C, respectively. The 50, 75, and 100% of field capacity soil moisture regimes should reflect paddy or furrow-irrigated rice production before the establishment of a permanent flood and provide an extreme scenario in instances where irrigation is limited or delayed reaching certain areas of a field.

The buckets were filled with 4,250 g of soil brought to the appropriate moisture regime (based on 4,500 g of soil) and seeded with barnyardgrass at approximately 130 seeds per bucket. The remaining 250 g of soil was used to cover the exposed seed. Tetflupyrolimet and clomazone were applied individually and in a mixture at 134 and 336 g ai ha⁻¹ at the PRE and POST (2- to 3-leaf barnyardgrass) application timings. For POST applications, nonionic surfactant was added at 0.25% v/v to reduce the surface tension of the spray droplet. The PRE and POST experiments were conducted simultaneously for each run but were considered independent for statistical analysis. All PRE buckets received activating irrigation (1.3 cm) immediately following the herbicide application and were irrigated up to the appropriate moisture regime daily. The soil in buckets designated for POST treatments was also maintained daily in the respective moisture regimes. Each experiment was terminated 28 DAT. All herbicide treatments were applied using a motorized spray chamber calibrated to deliver 187 L ha⁻¹ with two 110067 flat-fan nozzles (TeeJet, Glendale Heights, IL).

Visible estimations of barnyardgrass control were recorded at 14 and 28 DAT on a 0 to 100% scale, with 0% representing no control and 100% representing complete control. The number of barnyardgrass plants in each bucket were recorded 28 DAT for PRE treatments. For

POST treatments, barnyardgrass counts were recorded immediately before the herbicide application and again 28 DAT. The height of three barnyardgrass plants per bucket was recorded at 28 DAT, and above-ground biomass was harvested, oven-dried to constant mass, and weighed.

Visual barnyardgrass control, density, biomass, and height data for the PRE and POST experiments were averaged over the three independent runs conducted from 2022 to 2024 and analyzed. Each experiment was initiated as a completely randomized design, and experimental run was considered a random effect, where block was nested within run. Herbicide treatment, moisture regime, and DAT were included in the model as fixed effects for a repeated measures analysis of percent barnyardgrass control. Barnyardgrass height, biomass, and mortality were analyzed as a two-factor factorial (herbicide treatment by soil moisture) because data collection of each response was assessed at a single point in time. All distributions were analyzed using the JMP PRO 17.1 distribution platform (Avent et al. 2022) and the residuals of all data assumed a normal distribution, excluding barnyardgrass densities, which assumed a Poisson distribution (Gbur et al. 2012). All data were analyzed in JMP PRO 17.1 and subjected to ANOVA using the fit model platform. Means were separated using Tukey's honest significant difference (HSD) ($\alpha=0.05$).

Flood Timing Experiment. A greenhouse experiment was conducted and replicated three times to assess the impact of flood timing on POST-applied tetflupyrolimet in scenarios where the timeliness of water movement across a field can be variable (up to 10 d). The experiment was arranged as a single-factor, completely randomized design with three replications conducted on silt loam soil (same soil, location, and greenhouse parameters as the previous experiment). Barnyardgrass was planted into pots filled with sieved soil and thinned to five plants. The soil in pots was maintained at 80% of field capacity until a permanent flood was established. Tetflupyrolimet at 134 g ai ha⁻¹ plus nonionic surfactant at 0.25% v/v was simultaneously applied POST (2- to 3-lf) to all pots. Large metal containers were used for permanent flood establishment that maintained 5-cm of clearance between the soil surface and the top of the container. Flood depth was monitored and replenished daily. When One set of pots was flooded after allowing the herbicide to dry (approximately 4 hr after application). The other two flood timing treatments were submersed to a 5-cm depth at 5 and 10 DAT until 28 d after the initial herbicide application.

Visible estimates of barnyardgrass control were recorded at 14 and 28 DAT, as described previously. At 28 DAT, live plants were counted in each pot, and above-ground biomass was harvested, oven-dried to constant mass, and weighed. Percent mortality was calculated because each pot was thinned to 5 plants.

Visible barnyardgrass control, biomass, and percent mortality were averaged over the three independent runs, with flood timing considered as the only fixed effect for percent mortality and biomass. A repeated measures analysis was conducted for visible barnyardgrass control at 14 and 28 DAT to determine if efficacy increased over time, where flood timing and DAT were included in the model as fixed effects. Run was considered as a random effect and was not included in the model. Distributions were confirmed in JMP PRO 17.1 using the distribution platform (Avent et al. 2022), where the residuals assumed normality, and all data were subjected to ANOVA in JMP PRO 17.1 using the fit model platform. Means were separated using Fisher's protected least significant difference ($\alpha=0.05$).

RESULTS AND DISCUSSION

Soil Residual Activity Experiment.

Silt loam soil. Visible injury to rice was comparable for each herbicide treatment except for tetflupyrolimet (3%) and clomazone plus quinclorac (9%) when averaged over 21, 28, 35, and 42 DAT for the repeated measures analysis (Table 1). Visible injury from clomazone or mixtures that include clomazone, typically manifests as transient bleaching (Zhang et al. 2005), and it is not surprising that greater numerical injury was observed on a silt loam soil in comparison to other treatments as has been previously observed by others (Jordan et al. 1998). When averaged over DAT, all other herbicide treatments shared a similar level of damage to rice that ranged from 7 to 9%, indicating that injury remained minimal and was not different across evaluation dates. Injury to rice caused by tetflupyrolimet, if any, should exhibit a lack of root and shoot growth development (stunting) without the presence of chlorosis or necrosis (Selby et al. 2023); however, the only discernable symptomology was a negligible degree of stunting that was likely due to variability in the field.

According to the repeated measures analysis, there were no differences in visible control of broadleaf signalgrass and barnyardgrass from 21 to 42 DAT, suggesting that efficacy remained consistent throughout the experiment for each weed species and could be averaged over DAT (Table 1). When averaged over DAT, the two mixtures that included clomazone provided the

greatest broadleaf signalgrass control, where both were equal to 97%. Individual applications of tetflupyrrolimet (90%) and clomazone (88%) were comparable, but quinclorac (92%) provided better control of broadleaf signalgrass than clomazone. Like what was observed with broadleaf signalgrass, the repeated measures analysis concluded that barnyardgrass control was consistent from 21 to 42 DAT (Table 1). The same trend was observed, where the mixture of clomazone and tetflupyrrolimet, or clomazone and quinclorac, provided 97% control of barnyardgrass. However, an individual application of tetflupyrrolimet (93%) was comparable to the mixtures, which may allude to its specificity towards barnyardgrass, albeit the herbicide provided 90% visible control of broadleaf signalgrass at the silt loam location.

The repeated measures analysis of cumulative broadleaf signalgrass densities further confirmed and reiterated the importance of mixing at least two compatible SOAs to increase weed control (Table 2). The two mixtures reduced the emergence of broadleaf signalgrass compared to any individual component by at least 4, 6, and 8 plants per m² across all evaluation dates. Unlike broadleaf signalgrass, barnyardgrass cumulative densities did not increase or decrease over time but demonstrated an overall herbicide treatment effect when averaged across evaluation dates (Table 2). The individual applications of each herbicide ranged from 20 to 28 barnyardgrass escapes on average as opposed to the two mixtures ranging from 8 to 10 m⁻². However, tetflupyrrolimet was comparable to the clomazone plus quinclorac mixture. Having eight barnyardgrass escapes per square meter, the mixture of tetflupyrrolimet and clomazone was more effective than all other individual treatments. Grain yield was similar for all treatments ($\geq 8,440$ kg ha⁻¹), excluding quinclorac (7,120 kg ha⁻¹), which may reflect poorer early-season barnyardgrass control relative to the other treatments in this experiment (Table 1).

This research indicates that quinclorac-resistant barnyardgrass was likely not prevalent during the three site-years on the silt loam soil. At this time, there is no supporting evidence from this experiment to conclude that a mixture of clomazone and tetflupyrrolimet would be advantageous over a mixture of clomazone and quinclorac. However, given the current resistance status of barnyardgrass to quinclorac, there may be a benefit of mixing the novel SOA with clomazone. These potential implications underscore the importance of our research in the field of herbicide efficacy and weed control.

Clay soil. Visible injury to rice differed amongst herbicide treatments when averaged over 14, 28, and 42 DAT evaluations, although no estimation exceeded 9% and is likely biologically

insignificant (Table 1). The mixture of clomazone plus tetflupyrolimet or clomazone alone caused the highest levels of injury to rice at 14 DAT (9%) on the clay soil, comparable to clomazone alone at 6%. The visible injury caused by the mixture of clomazone and tetflupyrolimet was not greater than clomazone alone, indicating that the damage was likely associated with bleaching. The repeated measures analysis (averaged over DAT) emphasized that tetflupyrolimet caused less numerical damage or was comparable to current PRE-applied commercial standards used in this experiment.

An initial concern was adequately adjusting the rate of tetflupyrolimet to optimize barnyardgrass control on clay soils since approximately 19% of rice hectareage is produced on a clay or clay loam soil in Arkansas (Hardke 2022). However, adjusting the rate of clomazone to 670 g ai ha⁻¹ has proven to be sufficient on a clay soil without compromising weed control or increasing injury to rice (Zhang et al. 2005), which is above the rate of the herbicide used in this experiment. Adjusting the rate of tetflupyrolimet by a factor of 1.7 from a silt loam soil (228 g ai ha⁻¹) provided a comparable level of barnyardgrass control to clomazone at 560 g ai ha⁻¹ and the mixture with clomazone at the respective rates when averaged over DAT (Table 1). Shrinking and swelling of the clay soil surface often translates to a loss in residual control due to a comprised herbicide-rich barrier, where there is no longer contact with germinating weeds and an opportunity for seeds to germinate at greater depths within soil openings. Albeit this phenomenon rarely translated into end-of-season escapes in plots treated with tetflupyrolimet based on early- and late-season visual observations. Cumulative barnyardgrass densities emphasized that more consistent control could be achieved over time (14 to 42 DAT) when mixing two SOAs, such as clomazone and tetflupyrolimet or clomazone and quinclorac (Table 3). Excluding clomazone alone, by 42 DAT, plots treated with tetflupyrolimet and quinclorac had greater barnyardgrass densities than as tank-mix partners with clomazone by 14 and 50 plants per square meter, respectively. Similar to the silt loam site, plots treated with quinclorac as a standalone herbicide had a lower grain yield on average than all other treatments, with higher infestations of barnyardgrass likely contributing to the observed reduction.

Soil Moisture Experiment.

Preemergence. An interaction of herbicide and soil moisture regime was observed for barnyardgrass when averaged over the 14 and 28 DAT evaluation dates (Table 4). Barnyardgrass control with clomazone numerically decreased from 92 to 86% as soil moisture increased from

50 to 100% of field capacity, respectively, but was not significantly reduced. Plots treated with tetflupyrolimet demonstrated a contrasting effect compared to clomazone, where increased moisture increased barnyardgrass control. However, an increase in barnyardgrass control with tetflupyrolimet could only be observed between 50% (85% control) and 100% (93% control) of field capacity. A similar effect has been documented with PRE applications of amiben, atrazine, and *N, N*-dipropylthiocarbamate on a clay loam, where an increase in soil moisture resulted in greater performance (Stickler et al. 1969). Mixing clomazone and tetflupyrolimet provided effective and consistent barnyardgrass control, never falling below 98% at the evaluated soil moisture regimes when averaged over 14 and 28 DAT. Repeated measures analysis showed that there were differences in barnyardgrass control from 14 to 28 DAT when averaged over the soil moisture regime (Table 5). Barnyardgrass control decreased from 92 to 85% for clomazone from 14 to 28 DAT, respectively, while control increased by 10 percentage points from 84% for tetflupyrolimet in the same period. Mixing the two herbicides resulted in $\geq 98\%$ barnyardgrass control at each evaluation date and continued to emphasize the importance of using them together in field situations for greater initial efficacy and persistence.

The same herbicide and soil moisture regime interaction was observed for barnyardgrass percent mortality, where percent mortality appeared to be reflective of visible estimations (Table 6). Tetflupyrolimet was the only herbicide treatment in which increasing soil moisture likewise led to an increase in barnyardgrass mortality. A 2-fold increase in soil moisture (50% to 100% of field capacity) resulted in a 20-percentage point differential in PRE barnyardgrass control (76% vs. 96% control). Despite increased PRE barnyardgrass control with tetflupyrolimet, the mixture of tetflupyrolimet and clomazone consistently remained above 98% barnyardgrass mortality across the three soil moisture regimes. If tetflupyrolimet is commercialized and applied as a standalone product, rice producers need to be mindful that maintaining a high level of soil moisture, if possible, may aid in barnyardgrass management. However, mixing with clomazone would be an ideal tank-mix partner due to the increase in grass weed spectrum coupled with the consistent performance in the evaluated moisture regimes that are representative of field scenarios. In comparison to clomazone, barnyardgrass height and biomass were reduced by approximately 6-fold when tetflupyrolimet was applied alone or in a mixture (Table 6). Visually, plots treated with the mixture of clomazone and tetflupyrolimet displayed a combination of bleaching and stunting from the inhibition of 1-deoxy-D-xyulose 5-phosphate synthase and

DHODH, where the latter explains the lack of growth and development of barnyardgrass (Selby et al. 2023).

Combining clomazone and tetflupyrolimet displayed consistency across all moisture regimes for each response, which again, highlighted the advantage of mixing the two SOAs. The high level of barnyardgrass control and overall consistency would allow greater flexibility in FIR, where fields are divided into top, middle, and bottom (flooded) management zones with varying degrees of soil moisture (Chlapecka et al. 2021). If soil-applied herbicides needed to be activated in a conventional paddy rice system, it would likely take longer for each bay to receive adequate irrigation than the management zones in FIR. However, irrigation from flushing or flooding would potentially be more effective at distributing the herbicide in the surface layers of the soil and increasing efficacy opposed to moistened soil (Russell et al. 1990).

Postemergence. Clomazone, tetflupyrolimet, and the combination of the two herbicides displayed POST efficacy on barnyardgrass, albeit not to the extent of PRE applications (Table 4) (Selby et al. 2023). Although the herbicides evaluated in this experiment will not be advocated for POST applications as standalone treatments (Atul Puri, Global Research and Development Product Manager of FMC, personal communication), it is important to identify the effectiveness of each on emerged weeds and if differing soil moisture regimes further influence efficacy. Tetflupyrolimet alone or in combination with clomazone will likely be mixed with other POST grass products to extend residual control of non-emerged weeds, and to date, there have been no incompatible herbicides identified that adversely impact the efficacy of actively growing barnyardgrass (Castner, unpublished data).

An interaction of herbicide treatment and soil moisture regime was also present for POST applications when averaging over the 14 and 28 DAT evaluation dates (Table 4). Barnyardgrass control with the individual herbicides or the mixture was not influenced by any change in soil moisture regime, although mixing clomazone and tetflupyrolimet was superior to clomazone by 21, 16, and 34 percentage points at the 50, 75, and 100% of field capacity, respectively. Under greenhouse conditions, changes in soil moisture did not influence the control of red rice (*Oryza sativa* L.) or barnyardgrass from POST applications of imazethapyr (Zhang et al. 2001). From a POST standpoint, barnyardgrass control will be consistent across differing moisture regimes with a mixture of tetflupyrolimet and clomazone, which would provide greater flexibility in irrigation

practices and ensure that efficacy would not be reduced if applied prior to establishment of a permanent flood.

Barnyardgrass height, biomass, and mortality were influenced by the POST herbicide applied, where the mixture of clomazone and tetflupyrolimet outperformed the individual components in two out of the three measured responses at 28 DAT (Table 7). Mixing the two SOAs reduced plant biomass by approximately 3- and 2-fold and increased percent mortality by a factor of roughly 3 and 6 over clomazone and tetflupyrolimet, respectively. Barnyardgrass height was similar between tetflupyrolimet alone and the mixture with clomazone, which is not surprising due to the downstream inhibition of pyrimidines needed for cellular reproduction caused by the new SOA. In PRE and POST applications, the addition of clomazone to tetflupyrolimet appeared to visibly compound symptomology from each respective SOA, where plants were stunted, bleached, and demonstrated more rapid necrosis from loss of carotenoids leading to photooxidation.

Flood Timing Experiment. Postemergence barnyardgrass control was improved by a range of 9 to 16 percentage points by establishing a permanent flood at 4 HAT as opposed to delaying until 5 or 10 DAT (Table 8). By 28 DAT, barnyardgrass control increased for all treatments; however, the 4 HAT flood was not improved statistically, but was superior to the later flood timings. From 14 to 28 DAT evaluation dates, the 5 DAT flood timing saw the most improvement (16 percentage points) and was comparable to barnyardgrass in pots flooded at 10 DAT. At 28 DAT, the 4 HAT flood timing provided the highest level of POST barnyardgrass control (79%), which translated to the greatest percentage of plant mortality (26%) though comparable to flooding at 10 DAT (10%).

Despite the inconsistencies of POST barnyardgrass control at the 5 and 10 DAT flood timings, areas in the field where a permanent flood can be established at an earlier time appear to demonstrate a beneficial relationship and, at the least, should not adversely impact efficacy. Similar results have been documented with imazethapyr, where flooding 1 to 14 DAT maintained a higher level of red rice control at 28 d following a POST application (Avila et al. 2005). Although the flood timing experiment with tetflupyrolimet focused on POST efficacy, soil-applied herbicides generally improve performance when the activation is immediate and not excessive (Stewart et al. 2012). Further experiments will need to be conducted to determine if delaying herbicide activation influences PRE activity of tetflupyrolimet.

PRACTICAL IMPLICATIONS

With any herbicide there are limitations, especially given the novelty of the SOA and the lack of published research specific to tetflupyrolimet. Results from these experiments demonstrate the effectiveness of tetflupyrolimet on barnyardgrass as a soil-applied residual herbicide in comparison to commercial standards, with the potential to aid in management of other grass weed species. Performance of tetflupyrolimet remained consistent in most instances compared to the evaluated commercial standards when adjusting the rate from a silt loam to a clay soil with minimal visible injury to rice. Mixing tetflupyrolimet with clomazone improved PRE and POST barnyardgrass efficacy across a range of dry to saturated environments that can be expected to occur in paddy rice and FIR systems. Furthermore, the addition of clomazone to tetflupyrolimet offers rice producers two effective SOAs to manage barnyardgrass, increase grass weed spectrum, and minimize the selection placed on POST grass products in all available technologies. The consistency provided by a clomazone and tetflupyrolimet mixture should allow producers flexibility in time when using irrigation to activate the herbicides without compromising efficacy. Despite the effectiveness of both herbicides, making timely applications, using the appropriate rates, and incorporating a systems approach for weed management will aid in preserving longevity as new chemistries become available.

Acknowledgements: The staff at the Rice Research and Extension Center and the Northeast Research and Extension Center are appreciated for their assistance with plot establishment and maintenance.

Funding: FMC Corporation provided funding for this research.

Competing Interest: Mason Castner is currently an employee of FMC Corporation.

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TABLES AND FIGURES

Table 1. Broadleaf signalgrass and barnyardgrass percent control, rice visible injury, and rice grain yield collected at harvest. All data were averaged over the 2021, 2022, and 2023 site-years at the silt loam and clay sites.^{ad}

Herbicide	Rate	Broadleaf signalgrass		Barnyardgrass		Rice		Grain yield
	g ai ha ⁻¹	-----% control ^b -----		-----		% injury		kg ha ⁻¹
<i>Silt loam</i>								
nontreated								4,800
tetflupyrolimet	134	90	bc	93	ab	3	b	8,440 ab
clomazone	336	88	c	90	b	7	ab	8,440 ab
quinclorac	336	92	b	89	b	8	ab	7,120 b
clomazone + tetflupyrolimet	134 + 336	97	a	97	a	7	ab	9,300 a
clomazone + quinclorac	336 + 336	97	a	97	a	9	a	9,550 a
<i>P</i> -value		<.0001		<.0001		0.0200		0.0076
<i>Clay^c</i>								
nontreated								5,500
tetflupyrolimet	228			87	ab	4	b	8,890 ab
clomazone	570			91	ab	6	ab	8,640 ab
quinclorac	570			86	b	4	b	7,980 b
clomazone + tetflupyrolimet	228 + 570			91	ab	9	a	9,300 a
clomazone + quinclorac	570 + 570			94	a	6	ab	9,250 a
<i>P</i> -value				0.0211		0.0020		0.0069

^aMeans within a column followed by the same letter are not different according to Tukey's HSD ($\alpha=0.05$).

^bPercent control analyzed using repeated measures and averaged over 21, 28, 35, and 42 d after treatment.

^cBroadleaf signalgrass was not present at the clay site.

^dNontreated control was not included in statistical analysis, but means were provided for grain yield.

Table 2. Cumulative broadleaf signalgrass density at 21, 28, 35, and 42 d after treatment at the silt loam site. All data were averaged over the 2021, 2022, and 2023 site-years.^{ab}

Herbicide	Rate	Broadleaf signalgrass ^c								Barnyardgrass	
		21 DAT ^e		28 DAT		35 DAT		42 DAT			
		plants 1 m ⁻²									
nontreated ^d		230		270		294		294		272	
tetflupyrolimet	134	34	bcd	42	bc	58	a	58	a	20	ab
clomazone	336	38	bc	40	bc	46	ab	46	ab	22	a
quinclorac	336	20	efg	30	cde	40	bc	40	bc	28	a
clomazone + tetflupyrolimet	134 + 336	12	h	12	gh	20	efg	20	efg	8	c
clomazone + quinclorac	336 + 336	10	h	18	fgh	24	def	24	def	10	bc
<i>P</i> -value		0.0032								<.0001	

^aMeans within a column followed by the same letter are not different according to Tukey's HSD ($\alpha=0.05$) for barnyardgrass.

^bBarnyardgrass averaged over d after treatment.

^cRepeated measures analysis was significant for interaction of density and d after treatment; therefore, means can be compared across columns and rows according to Tukey's HSD ($\alpha=0.05$).

^dNontreated control was not included in statistical analysis but means provided to show density.

^eAbbreviation: DAT, d after treatment.

Table 3. Cumulative barnyardgrass density at 14, 28, and 42 d after treatment at the clay site. All data were averaged over the 2021, 2022, and 2023 site-years.^{ab}

Herbicide	Rate	Barnyardgrass					
		14 DAT ^c		28 DAT		42 DAT	
	g ai ha ⁻¹	-----plants 1 m ⁻² -----					
nontreated		254		456		480	
tetflupyrolimet	228	22	cd	58	a	60	a
clomazone	570	12	e	42	b	48	b
quinclorac	570	30	bc	68	a	80	a
clomazone + tetflupyrolimet	228 +570	16	de	42	b	46	b
clomazone + quinclorac	570 + 570	4	f	24	c	30	c
<i>P</i> -value		0.0031					

^aRepeated measures analysis was significant for interaction of density and d after treatment; therefore, means can be compared across columns and rows according to Tukey's HSD ($\alpha=0.05$).

^bNontreated control was not included in statistical analysis but means provided to show density.

^cAbbreviation: DAT, d after treatment.

Table 4. Interaction of herbicide treatment and moisture regime at the preemergence and postemergence timings on barnyardgrass control. All data were averaged over the three runs conducted in the greenhouse at the Milo J. Shult Research and Extension Center, in Fayetteville, AR, in 2023 and 2024. The preemergence and postemergence experiments were analyzed independently.^a

Herbicide	Rate	Moisture regime	Control ^b	
<i>Preemergence</i>	g ai ha ⁻¹	% of field capacity	%	
clomazone	336	50	92	abcd
tetflupyrolimet	134	50	85	d
clomazone + tetflupyrolimet	336 + 134	50	99	a
clomazone	336	75	87	cd
tetflupyrolimet	134	75	90	bcd
clomazone + tetflupyrolimet	336 + 134	75	98	ab
clomazone	336	100	86	cd
tetflupyrolimet	134	100	93	abc
clomazone + tetflupyrolimet	336 + 134	100	99	a
<i>P</i> -value			0.0051	
<i>Postemergence^c</i>				
clomazone	336	50	57	cde
tetflupyrolimet	134	50	64	bcde
clomazone + tetflupyrolimet	336 + 134	50	78	ab
clomazone	336	75	58	cde
tetflupyrolimet	134	75	71	abcd
clomazone + tetflupyrolimet	336 + 134	75	74	ab
clomazone	336	100	51	e
tetflupyrolimet	134	100	73	abc
clomazone + tetflupyrolimet	336 + 134	100	85	a
<i>P</i> -value			0.04811	

^aMeans within a column followed by the same letter are not different according to Tukey's HSD ($\alpha=0.05$).

^bPercent control was analyzed using repeated measures and averaged over 14 and 28 d after treatment.

^cIncluded nonionic surfactant at 0.25% v/v.

Table 5. Interaction of herbicide treatment and moisture regime at the preemergence timing on barnyardgrass mortality collected at 28 d after treatment. Data were averaged over the three experimental runs conducted in the greenhouse at the Milo J. Shult Research and Extension Center, in Fayetteville, AR, in 2023 and 2024.^a

Herbicide	Rate	Moisture regime	Mortality	
	g ai ha ⁻¹	% of field capacity	% of nontreated	
clomazone	336	50	92	ab
tetflupyrolimet	134	50	76	c
clomazone + tetflupyrolimet	336 + 134	50	98	ab
clomazone	336	75	87	abc
tetflupyrolimet	134	75	84	bc
clomazone + tetflupyrolimet	336 + 134	75	98	ab
clomazone	336	100	87	abc
tetflupyrolimet	134	100	96	ab
clomazone + tetflupyrolimet	336 + 134	100	99	a
<i>P</i> -value			0.0073	

^aMeans within a column followed by the same letter are not different according to Tukey's HSD ($\alpha=0.05$).

Table 6. Visible barnyardgrass control at 14 and 28 d after treatment (DAT) including height and biomass collected at 28 DAT for the preemergence experiment. Means are the average of three runs conducted in the greenhouse at the Milo J. Shult Research and Extension Center, in Fayetteville, AR, in 2023 and 2024.

Herbicide	Rate	Control ^a		Height ^{bc}	Biomass ^{bd}
		14 DAT	28 DAT		
	g ai ha ⁻¹	---% of nontreated---		-% reduction of nontreated-	
clomazone	336	92 b	85 c	58 a	78 a
tetflupyrolimet	134	84 c	94 ab	93 b	96 b
clomazone + tetflupyrolimet	336 + 134	98 a	99 a	95 b	99 b
<i>P</i> -value		<.0001		<.0001	0.0005

^aRepeated measures analysis was significant for interaction of control and d after treatment; therefore, means can be compared across columns and rows according to Tukey's HSD ($\alpha=0.05$).

^bMeans within a column followed by the same letter are not different according to Tukey's HSD ($\alpha=0.05$).

^cMean barnyardgrass height of the nontreated control at 28 DAT was 37 cm.

^dMean barnyardgrass biomass of the nontreated control at 28 DAT was 12 g.

Table 7. Barnyardgrass height, biomass, and mortality collected at 28 d after treatment for the postemergence experiment. Means are the average of three runs conducted in the greenhouse at the Milo J. Shult Research and Extension Center, in Fayetteville, AR, in 2023 and 2024.^{ab}

Herbicide	Height		Biomass		Mortality	
	-----% reduction of nontreated-----				% of nontreated	
clomazone	50	a	66	a	21	b
tetflupyrolimet	80	b	74	a	10	b
clomazone + tetflupyrolimet	85	b	88	b	59	a
<i>P</i> -value	<0.0001		0.0002		<0.0001	

^aMeans within a column followed by the same letter are not different according to Tukey's HSD ($\alpha=0.05$).

^bIncluded nonionic surfactant at 0.25% v/v.

^cMean barnyardgrass height of the nontreated control at 28 DAT was 49 cm.

^dMean barnyardgrass biomass of the nontreated control at 28 DAT was 21 g.

Table 8. Repeated measures analysis of percent visible control of barnyardgrass at 14 and 28 days after treatment, as well as mortality and biomass as a percentage and percent reduction of the nontreated control, respectively, at each flood timing. All data were averaged over the three runs conducted in the greenhouse at the Milo J. Shult Research and Extension Center, in Fayetteville, AR, in 2023.^{ab}

Herbicide	Flood timing	Control		Mortality	Biomass
		14 DAT	28 DAT		
		-----% of nontreated-----			% reduction of nontreated
tetflupyrolimet	4 HAT	70 a	79 a	26 a	90
tetflupyrolimet	5 DAT	54 d	70 b	2 b	89
tetflupyrolimet	10 DAT	61 c	69 b	10 ab	92
<i>P</i> -value		0.0058		0.0108	0.2681

^aRepeated measures analysis was significant for the interaction of percent barnyardgrass control and d after treatment, therefore means can be compared across columns and rows according to Fisher's protected LSD ($\alpha=0.05$).

^bMeans within a column followed by the same letter are not different according to Fisher's protected LSD ($\alpha=0.05$).