


Grapevine, stone fruit, and tree nut crop response to simulated tetflupyrolimet drift

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Note

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Nomenclature:

Tetflupyrolimet; almond, *Prunus dulcis* (Mill.) D.A. Webb; grapevine, *Vitis vinifera* L.; peach, *Prunus persica* (L.) Batsch; pistachio, *Pistacia vera* L.; plum, *Prunus domestica* L.; rice, *Oryza sativa* L.; walnut, *Juglans regia* L.

Keywords:

Aryl pyrrolidinone anilide; de novo pyrimidine nucleotide biosynthesis; dihydroorotate dehydrogenase inhibitor; nontarget crop; off-target drift; rice herbicide

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Abstract

Off-target herbicide drift away from rice is historically a concern in California, where susceptible crops such as orchards and vineyards are nearby. Tetflupyrolimet is a potent inhibitor of dihydroorotate dehydrogenase that provides excellent grass weed control in rice cropping systems. In efforts to steward tetflupyrolimet before its registration in California, this research was conducted to compare the onset of foliar symptoms from tetflupyrolimet applications onto almond, grapevine, peach, pistachio, plum, and walnut. Tetflupyrolimet was applied to these tree and vine crops at fractional rates of 1/200×, 1/100×, 1/33×, and 1/10× of the 125 g ai ha⁻¹ recommended use rate on rice. Almond, pistachio, and walnut trees also received 1× of the use rate. Tetflupyrolimet treatments were applied on one side of 3- to 4-yr-old almond, peach, pistachio, plum, and walnut trees, and on one side of 25- to 26-yr-old grapevines in 2022 and 2023. Visible injury ratings were carried out weekly to assess symptomology throughout the growing seasons and at leaf-out the following springs. Tree trunk diameter was recorded before and after herbicide applications. No injury was observed to any tested crops, regardless of the tetflupyrolimet application rate. In all orchard crops, tree trunk diameter was not affected by tetflupyrolimet treatments. Likewise, grape yield was not reduced even at the 1/10× tetflupyrolimet fractional rate. Since no injury symptoms were recorded, this research suggests that tetflupyrolimet can be safely used on nearby rice fields and might be a target for future registration consideration for use on orchard and vineyard crops.

Introduction

Tetflupyrolimet [CAS: 2053901-33-8; (3S,4S)-*N*-(2-fluorophenyl)-1-methyl-2-oxo-4-[3-(trifluoromethyl)phenyl]pyrrolidine-3-carboxamide] is an aryl pyrrolidinone anilide herbicide that was developed for selective control of grass, sedge, and broadleaf weeds in rice cropping systems. As the first novel mode-of-action Group 28 herbicide (as categorized by the Herbicide Resistance Action Committee and Weed Science Society of America) in the last three decades, tetflupyrolimet is a potent inhibitor of dihydroorotate dehydrogenase (DHODH), which is involved in the de novo pyrimidine nucleotide biosynthesis (Maienfisch and Mangelinckx 2021). The DHODH is the fourth enzyme of the pyrimidine de novo biosynthesis pathway that is localized to the mitochondria and catalyzes the conversion of dihydroorotate to orotate (Zrenner et al. 2006). In plants, the de novo pyrimidine nucleotide biosynthesis pathway is a vital process for metabolism; gene expression; and the production of substrates for DNA, RNA, and multiple biosynthesis pathways such as polysaccharides, glycoproteins, and phospholipids (Kang et al. 2023; Zrenner et al. 2006).

Owing to the central role of nucleotides, inhibition of DHODH is lethal to most organisms (Dayan 2019). Tetflupyrolimet has been shown to have high levels of activity against grass species such as watergrasses (*Echinochloa* P. Beauv.), sprangletops (*Leptochloa* P. Beauv.) (Lombardi and Al-Khatib 2024), giant foxtail (*Setaria faberi* Herrm.), and hairy crabgrass [*Digitaria sanguinalis* (L.) Scop.] (Selby et al. 2023). Moreover, tetflupyrolimet's activity is at least 10-fold greater on weeds such as foxtail than rice, which suggests it is selective in rice because the crop can metabolize tetflupyrolimet (Dayan 2019; Selby et al. 2023). Therefore, the discovery of tetflupyrolimet is important and promising for weed management in rice cropping systems.

California is the second largest rice producer in the United States with approximately 220,000 ha under cultivation (Galvin et al. 2022), which accounts for more than US\$1 billion in farmgate value (CDFA 2024). The primary rice production area is in the Sacramento and Northern San Joaquin valleys, and the crop typically is water-seeded and grown under continuously flooded conditions during the growing season (Inci et al. 2024d). Competitive grass weeds in rice systems such as barnyardgrass [*Echinochloa crus-galli* (L.) P. Beauv.], early watergrass [*E. oryzoides* (Ard.) Fritsch], late watergrass [*E. phyllopopon* (Stapf) Koso-Pol.], and

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bearded sprangletop [*Leptochloa fusca* (L.) Kunth ssp. *fascicularis* (Lam.) N. Snow] can significantly reduce rice yields (Brim-DeForest et al. 2017). Alongside cultural management methods such as planting certified weed-free rice seed, high seeding rates, and continuous water management, herbicides are crucial for weed management in rice (Inci and Al-Khatib 2024).

Beyond rice, California is the primary producer of many specialty crop commodities, including production of more than 99% of the nation's almonds, nectarines, peaches, pistachios, plums, raisin grapes, and walnuts (CDFA 2024). Among those, grape is the most valued crop in California, generating more than US\$5.5 billion in cash receipts from 350,000 ha of wine, table, and raisin grapes. Tree nuts are produced on 1 million ha in the Sacramento and San Joaquin valleys, and these regions account for 85% of the world's almond production. Collectively, grapes, stone fruits, and tree nuts are significant crops grown on ~1.5 million ha with a value of more than US\$12 billion. With such diverse cropping systems in California, rice is often grown adjacent to orchards and vineyards (Inci et al. 2024b, 2024c).

California's unique crop diversity, paired with strict regulatory structures, has limited the number of herbicide active ingredients available to rice growers because of the potential for off-target herbicide drift to nearby orchards and vineyards (Hill et al. 2006). Research on tetflupyrolimet in the complex herbicidal programs of California rice suggests its utility as both a preemergence and early postemergence herbicide (Lombardi and Al-Khatib 2024). The application timing of tetflupyrolimet is from the day-of-seeding to the 2-leaf rice growth stage. This herbicide application timing usually occurs from May to June depending on the planting date, rice variety, and environmental conditions.

During May and June, orchard crops and grapes are in relatively vulnerable growth stages for off-target herbicide exposures. Almond, pistachio, and walnut trees are in vigorous growth periods and actively developing terminal and lateral buds, leaves, spurs, and shoots and accumulating assimilates for kernels (Galla et al. 2018a, 2018b). Grapevine phenological stages at this time range from bloom to veraison (Bettiga 2013), and stone fruits are at a stage when the endocarp (pit) hardening process begins, and the fruit size increases (Strand 1999).

Herbicide drift is the physical movement of spray droplets through the air at the time of application or soon thereafter to any site other than the intended target (Whithaus 2016). Under most circumstances, off-target herbicide exposures are similar to herbicide fractional rates from below 1/100× up to 1/33× of the field application rates of herbicides (Al-Khatib and Peterson 1999). Significant drift events are most frequently associated with relatively high air temperature and wind speed, low relative humidity, small spray droplet size, and short distances to nearby nontarget crops (Whithaus 2016). The concerns of rice herbicide drift to off-target crops in the Sacramento and Northern San Joaquin valleys are common among growers, crop consultants, and researchers.

Tetflupyrolimet is anticipated to be a useful and widely used herbicide in rice cropping systems (Lombardi and Al-Khatib 2024). It is important to understand the relative sensitivity of crops subjected to fractional rates of tetflupyrolimet, particularly considering the economic impact of California grape, stone fruit, and tree nut industries. To steward tetflupyrolimet prior to its registration, this research was conducted to compare the onset of foliar symptoms from fractional tetflupyrolimet rates onto six tree and vine crops. We analyzed the growth response of almond, peach, pistachio, plum, and walnut trees to tetflupyrolimet at

different rates, and evaluated grapevine yield in response to tetflupyrolimet exposure.

Materials and Methods

Study Sites

Six herbicide experiments were conducted in 2022 and 2023 in a 3- to 4-yr-old almond (38.539°N, 121.794°W), peach (38.539°N, 121.794°W), pistachio (38.539°N, 121.793°W), plum (38.538°N, 121.794°W), and walnut (38.539°N, 121.794°W) orchards (elev. 18 m asl) at the University of California–Davis Plant Sciences Field Facility orchards; and in a 25- to 26-yr-old wine grape vineyard (38.525°N, 121.788°W) at the University of California–Davis Department of Viticulture and Enology Tyree Vineyard. The orchards were established in March 2020 with 'Nonpareil' almond on 'Empyrean 1', 'Coralstar' peach on 'Krymsk 86', 'Kerman' pistachio on 'UCB 1', 'French Improved' plum on 'Krymsk 86', and 'Chandler' walnut on 'clonal RX1'. All almond, peach, plum, and walnut trees were planted 6 m apart within rows and 4.2 m apart between rows, while pistachio trees were 6 m apart within rows and 7 m apart between rows. The vineyard was established in 1998 with a bilateral double-cordon-trained 'Grenache' wine grape planted 1.8 m apart within rows and 3.6 m apart between rows.

The soil was classified as a Yolo silt loam with NO₃-N 57 ppm, Olsen-P 26 ppm, K 351 ppm, Na 21 ppm, Ca 8 meq 100 g⁻¹, Mg 10 meq 100 g⁻¹, cation exchange capacity (CEC) 19 meq 100 g⁻¹, organic matter 2.7%, and pH 6.7 in the orchards; and NO₃-N 23 ppm, Olsen-P 12 ppm, K 288 ppm, Na 12 ppm, Ca 11 meq 100 g⁻¹, Mg 9 meq 100 g⁻¹, CEC 21 meq 100 g⁻¹, organic matter 2.5%, and pH 7.1 in the vineyard (soils were analyzed at the University of California–Davis Analytical Laboratory). Irrigation was applied in all crops through a single-line drip irrigation system with emitters spaced every 30 cm during the growing seasons. All trees and vines were maintained free of diseases and insects following standard commercial practices (Bettiga 2013; Strand 1999). In all experiments, weeds between rows were managed with regular mowing and within rows with a mixture of rimsulfuron at 70 g ai ha⁻¹, indaziflam at 50 g ai ha⁻¹, oxyfluorfen at 560 g ai ha⁻¹, and glufosinate-ammonium at 450 g ai ha⁻¹ plus manufacturer recommended surfactants.

Herbicide Applications

Tetflupyrolimet (Dodhylex™, 400 g ai L⁻¹; FMC Corporation, Philadelphia, PA) herbicide as a suspension concentrate (SC) formulation was applied on June 6, 2022, in all experiments. Tetflupyrolimet was applied at concentrations of 1/200×, 1/100×, 1/33×, 1/10×, and 1× the use rate of 125 g ai ha⁻¹ on almond, pistachio, and walnut trees. The concentrations actually represent percentages of the use rate as 0.5%, 1%, 3%, 10%, and 100% (Inci et al. 2024b). Due to a limited number of trees and vines, the 1× tetflupyrolimet treatment was not included for grapevines, or peach and plum trees. Plots with nontreated trees or vines were also included for comparison in each experiment.

All herbicide treatments were applied to one side of the tree or vine canopy as one pass (top to bottom for trees and side to side for cordon-trained vines) with a handheld, carbon dioxide-propelled backpack sprayer calibrated to deliver 187 L ha⁻¹ at 206 kPa pressure through XR 8004-VS nozzle tips (TeeJet Technologies, Glendale Heights, IL). The sprayer boom had two nozzles spaced 50 cm apart and spray was delivered based on a 3-s pass per tree or vine. Plots were sprayed early in the morning when winds were

calm to avoid herbicide cross-contamination to adjacent trees or vines. At the time of the orchard and vineyard applications on June 6, 2022, the air temperature was 16 C, with 58% relative humidity (RH) and 0.4 m s⁻¹ wind speed. All experiments were repeated on May 31, 2023, with a different set of trees or vines in the same orchards and vineyard. At the time of second-year application the air temperature was 18 C, with 50% RH and 0.6 m s⁻¹ wind speed. Because the trees were relatively young, no yield data were taken for orchard crops; however, grapevines had 5–10 mm berries present at the time of herbicide application in both 2022 and 2023.

Data Collection and Experimental Design

Experiments were arranged in a randomized complete block design with four replications, where an individual tree or vine was an experimental unit. Trees and vines were observed for visible injury symptoms at 6, 12, 24, 48, and 72 h after herbicide treatment; and 7, 14, 21, 28, 35, 42, and 90 d after treatment (DAT). Visible injury was rated on a scale on which 0 = no injury and 100 = plant death.

Trees and vines treated with tetflupyrolimet were compared with nontreated control plants at each observation. Furthermore, trunk diameters of almond, peach, pistachio, plum, and walnut trees were measured using a digital caliper with ±25 µm accuracy at approximately 25 cm above the ground on April 15 and October 20, 2022, and on April 23 and October 20, 2023. The timing of these measurements correspond with the beginning of spring growth, which starts in April (pretreatment), and after the growing season, which ends in October (posttreatment) (Inci et al. 2024b, 2024c). To maintain the consistency of assessments, the trunk diameter of all trees was measured four times regardless of whether they were treated in 2022 or 2023 experiments. Tree growth was expressed through trunk diameter growth as a percentage increase based on the following formula:

$$y = \{[x_{n+1}/x_n] - 1\} \times 100 + 100$$

where y is the percent relative change of trunk diameter, x_n = trunk diameter at pretreatment measurements in spring, and x_{n+1} = trunk diameter at posttreatment measurements approximately 140 DAT in fall. Thereby, the relative change in trunk diameter of herbicide-treated trees was compared with the relative change in the diameter of nontreated control trees.

Grapes were hand-harvested when berries on the vines of nontreated control plants reached ~20°Bx (1% soluble solids), a common practice for the Northern San Joaquin and Sacramento Valleys grapevine industry (Bettiga 2013). Grape clusters were harvested from all treated vines and from nontreated control vines, and weighed for total fruit yield and sugar content from a fruit subsample determined with a handheld refractometer.

Statistical Analysis

Trunk diameter data were subjected to analysis of covariance using AGRICOLAE (de Mendiburu 2024) and DPLYR (Wickham et al. 2024a, 2024b) packages to characterize the growth of the orchard crops with equation $Y = A + BX$, where Y is the predicted value, A is the y-intercept; B is the slope of the line, and X is the observation time. These analyses were conducted using RStudio v. 2024.09.1+394 (R Core Team 2024). Means were separated using Tukey's honestly significant difference post hoc test at significance level of $P \leq 0.05$, when applicable. The MULTCOMP (Bretz et al. 2010) package was used to generate multiple comparisons among means.

Tetflupyrolimet fractional rates were considered fixed factors, while crop, year, and replication were considered random factors. Grape yield and brix were analyzed with ANOVA at $\alpha = 0.05$ (Kniss and Streibig 2018). The Type II Wald F -tests with the Kenward-Roger degrees-of-freedom method and Type III with the Satterthwaite method were used when the confidence level was 0.95 for both ANOVA types. Graphical illustration was generated using the GGPlot2 package v. 3.5.1 in RStudio (Wickham et al. 2024c).

Results and Discussion

Tetflupyrolimet did not cause any distinguishable injury symptoms to any crop at any rating time or at any herbicide treatment, including up to the 1× rate of 125 g ai ha⁻¹ on almond, pistachio, and walnut (data not shown). The lack of any observed injury could be because established plants are not dependent on the pyrimidine nucleotide biosynthesis as much as developing plants because the pyrimidine nucleotide biosynthesis is energetically expensive, cells utilize pyrimidine nucleotides only if they are rapidly growing and dividing. In established plants, mature cells can meet their metabolic needs through a salvage pathway, a reutilization mechanism, to break down cellular components that are no longer needed and that do not metabolize the DHODH enzyme (Zrenner et al. 2006). Tetflupyrolimet, as a DHODH inhibitor, is therefore most active against weed seedlings and did not cause any injury symptoms to established trees and vines. No fruit yield data were taken in the orchard crops, fruit that were present appeared normal and consistent among treatments.

Tree trunk diameter change is a common parameter for interpreting orchard crop growth (Inci et al. 2024b, 2024c). The percent of relative change data for 2022 and 2023 were combined ($n = 8$) for tree crops because there were no significant interactions between year and treatment (Wickham et al. 2024a). In all orchard crops, tree trunk diameter change was variable for almond, peach, pistachio, plum, and walnut trees. From April 2022 to October 2023, the relative trunk diameter of all tested crops increased substantially, and the growth was not different ($P < 0.05$) from that of nontreated control trees (Figures 1 and 2). Almond, peach, and plum trees exhibited an average of ~30 mm trunk diameter increase across all treatments in 2022, whereas the average diameter of walnut and pistachio trees increased by 22 mm and 12 mm, respectively (data not shown). In 2023, the diameter of almond increased by 17 mm, 10 mm in peach and pistachio, 5 mm in plum, and 7 mm in walnut. At the fall 2023 observations, the total of two seasons of growth, almond, peach, pistachio, plum, and walnut trees showed cumulative diameter increases of 70 mm, 50 mm, 35 mm, 50 mm, and 45 mm, respectively. Together, these data corresponded to an average trunk diameter of almonds increased by 230%, pistachios by 292%, walnuts by 210% (Figure 1), peaches by 220%, and plums by 241% (Figure 2) at the end of two seasons.

The yield response of grapevines treated with tetflupyrolimet fractional rates did not differ ($P < 0.05$) from the nontreated control vines, which was approximately 23.6 kg vine⁻¹ in 2022 and 16.6 kg vine⁻¹ in 2023 (Table 1). Grape yield was approximately 6.85 kg vine⁻¹ lower overall in 2023 than 2022, including nontreated control vines, possibly related to a cooler (average air temperature ~1.5 C lower) season in 2023. Likewise, grape sugar content was similar among all treatments and ranged at approximately 20–21°Bx in 2022 and 2023 harvest (Table 1). The insignificant differences overall indicate that tetflupyrolimet drift events did not cause meaningful damage to grape yield or brix levels.

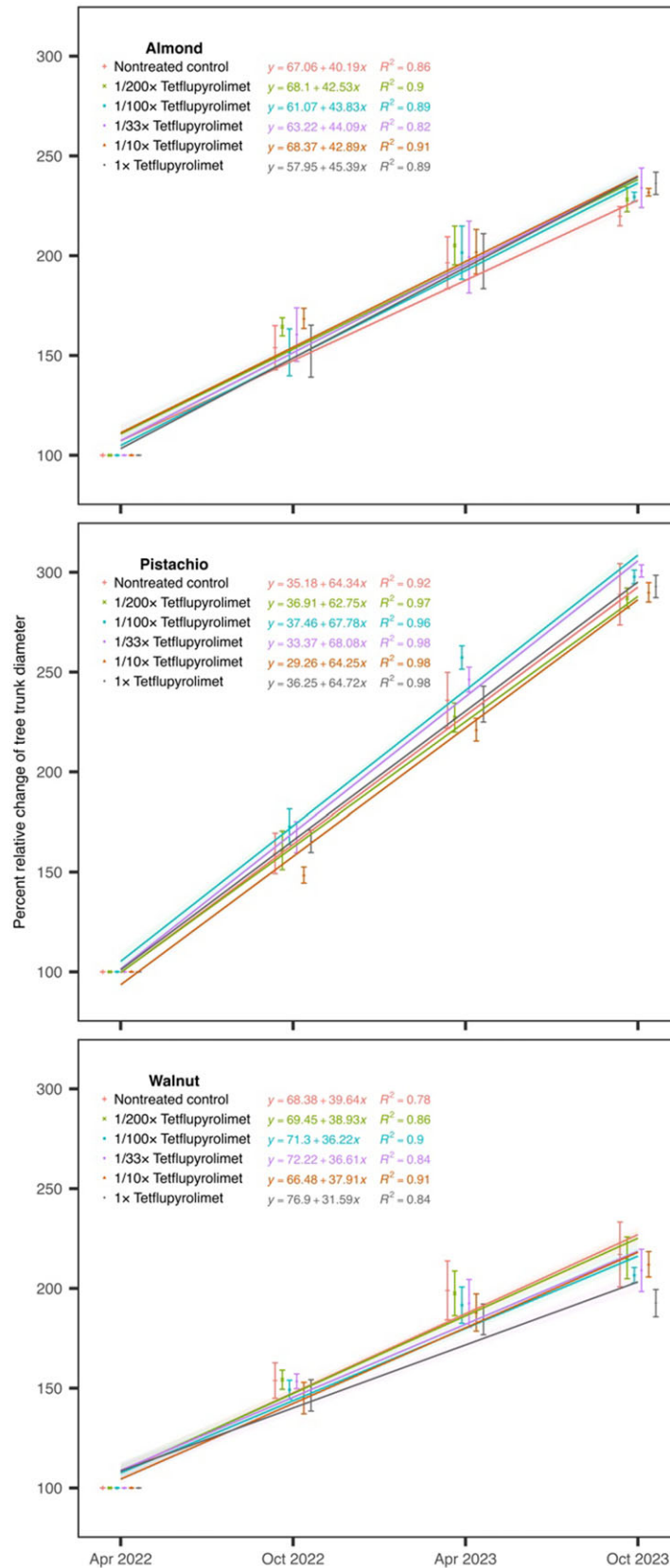


Figure 1. Trunk diameter measurements ($n = 8$) of almond (top), pistachio (middle), and walnut (bottom) trees before initiation of tetflupyrolimet treatments (April 2022 and 2023) and after treatments (October 2022 and 2023). Tree trunk diameter change was expressed as percent relative change with a linear model $Y = A + BX$, where Y is the predicted value, A is the y-intercept; B is the slope of the line, and X is the observation time. Tetflupyrolimet simulated drift rates were expressed as a fraction of rice use rate of 125 g ai ha^{-1} .

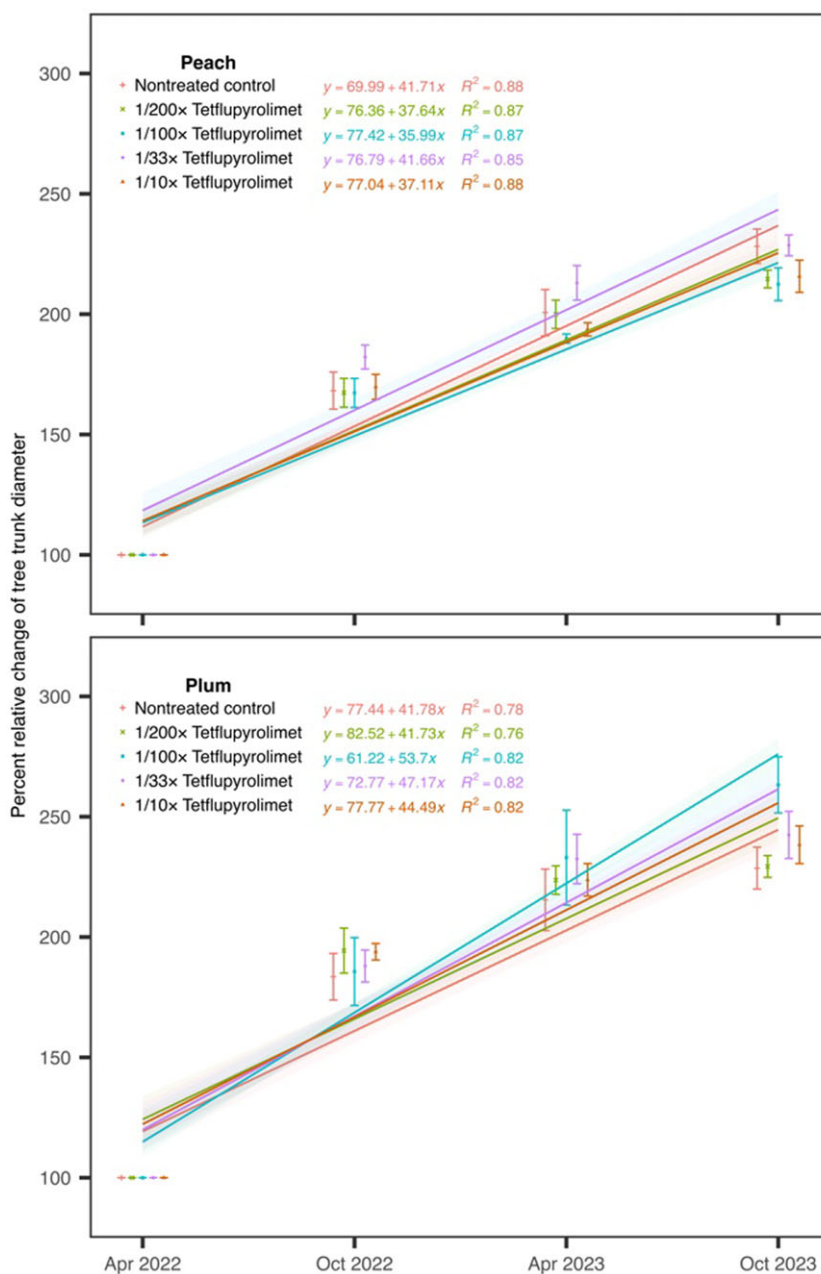


Figure 2. Trunk diameter measurements ($n = 8$) of peach (top) and plum (bottom) trees before initiation of tetflupyrolimet treatments (April 2022 and 2023) and after treatments (October 2022 and 2023). Tree trunk diameter change was expressed as percent relative change with a linear model $Y = A + BX$, where Y is the predicted value, A is the y-intercept; B is the slope of the line, and X is the observation time. Tetflupyrolimet simulated drift rates were expressed as a fraction of rice use rate of 125 g ai ha^{-1} .

Under most conditions, realistic herbicide drift rates range from below 1/100 \times up to 1/33 \times of the field use rates of herbicides (Al-Khatib and Peterson 1999; Inci et al. 2024a). This research included tetflupyrolimet at rates up to 1/10 \times or 1 \times in these crops to evaluate a worst-case scenario such as consecutive drift events, an accidental herbicide application, or herbicide-contaminated tank, events that are unlikely to happen in a typical drift situation. Even at these exaggerated rates, almond, grape, peach, pistachio, plum, and walnut crops were not injured by tetflupyrolimet exposure. This simulated drift research was conducted using a constant spray volume with variable rates including field use rate on tree nut crops, which is different from actual drift scenarios where both concentration and volume change as herbicides move off target. Banks and Schroeder (2002) suggested that droplet concentration can affect crop injury;

however, understanding the relative sensitivity of the stone fruit, tree nut, and vine crops species grown near California rice fields is highly relevant to stewardship of tetflupyrolimet in the Sacramento and San Joaquin valleys (Inci et al. 2024b). Anticipated application advisories for tetflupyrolimet in rice systems should also help prevent the off-target movement of herbicides to nontarget crops (Anonymous 2024). Together, these datasets suggest that tetflupyrolimet can be safely used for early-season weed management in rice fields with normal off-target herbicide drift precautions, as noted on the herbicide labels.

Practical Implications

Owing to its outstanding grass activity and crop safety on rice, tetflupyrolimet is expected to be widely used in rice cropping

Table 1. Grape yield and sugar concentration response to tetflupyrolimet simulated drift rates in 2022 and 2023 growing seasons near Davis, California.^{a,b}

Treatment	Rate ^c g ai ha ⁻¹	2022		2023	
		Yield ^d kg vine ⁻¹	Brix ^e °Bx	Yield	Brix
NTC	—	23.58 (1.3)	20.67 (0.5)	16.60 (0.2)	20.87 (0.9)
1/200×	0.625	23.34 (1.7)	20.27 (0.4)	16.30 (0.2)	21.25 (0.4)
1/100×	1.25	23.23 (0.8)	20.65 (1.1)	16.37 (0.5)	21.22 (0.7)
1/33×	3.75	23.17 (0.8)	20.82 (1.1)	16.52 (1.1)	21.22 (0.5)
1/10×	12.5	23.01 (1.3)	20.87 (0.9)	16.29 (1.4)	21.05 (0.4)

^aAbbreviation: NTC, nontreated control treatment.

^bThere were no significant differences within each column at $P < 0.05$ according to Tukey's honestly significant difference post hoc test.

^cTetflupyrolimet rate is expressed as a fraction of the rice use rate, 125 g ai ha⁻¹.

^dYield is reported as average mean, where parentheses represent SE.

^eOne degree °Bx is 1 g of sucrose in 100 g of solution, where parentheses represent SE.

systems (Lombardi and Al-Khatib 2024). Anticipated rice field applications for tetflupyrolimet are as pre-plant, preemergence, or early postemergence up to the 3-leaf stage for grasses (Anonymous 2024; Lombardi and Al-Khatib 2024). This research suggests that tetflupyrolimet is not likely to cause significant injury to nearby tree and vine crops if off-target drift occurs. Given the apparent low risk of crop injury to tree crops, tetflupyrolimet might be of interest for future registration for use on these crops. As a new site of action, tetflupyrolimet could help manage weeds in orchards and vineyards that are difficult to control, such as glyphosate-resistant grasses. Further studies should investigate the use of tetflupyrolimet in floor management of orchard and vineyard production systems.

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Competing Interests. The authors declare they have no competing interests.

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