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Characterization of rice cultivar response to florpyrauxifen-benzyl

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Research Article

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Abstract

Many factors such as environment, herbicide rate, growth stage at application, and days between sequential applications can influence the response of a crop to herbicides. Florpyrauxifen-benzyl is a new broad-spectrum, POST herbicide that was commercialized for use in U.S. rice production in 2018. Field experiments were conducted in 2018 at the Pine Tree Research Station (PTRS) near Colt, AR, and the Rice Research and Extension Center (RREC), near Stuttgart, AR, to evaluate crop injury and yield response of three rice cultivars to sequential applications of florpyrauxifen-benzyl. Greenhouse and growth chamber experiments were conducted at the Altheimer Laboratory in Fayetteville, AR, to evaluate cultivar responses when florpyrauxifen-benzyl was applied at 30 or 60 g ae ha⁻¹ to rice exposed to different temperature regimes or at various growth stages. Three rice cultivars were used in all experiments: long-grain variety ‘CL111’, medium-grain variety ‘CL272’, and long-grain hybrid cultivar ‘CLXL745’. CL111 exhibited sufficient tolerance to florpyrauxifen-benzyl with only 10% visible injury and no effect on yield. CL272 showed 15% injury 3 wk after the second application in the field experiment when applications were made 14 d apart. Additionally, 12% injury was observed in greenhouse studies when florpyrauxifen-benzyl was applied at 30 g ae ha⁻¹ averaged over various growth stages at application. Florpyrauxifen-benzyl did not reduce the yield of CL272 in field experiments, indicating that CL272 can recover from florpyrauxifen-benzyl injury. As much as 64% injury was observed for CLXL745 at 3 wk after application (WAA) when sequential herbicide applications were made 4 d apart. High levels of injury occurred in the growth chamber and greenhouse studies for this cultivar as well. Sequential applications of florpyrauxifen-benzyl reduced yields of CLXL745 in nearly all treatments. Data from these experiments suggest that CL272 and CLXL745 are sensitive to sequential applications of florpyrauxifen-benzyl. Growers must follow the prescribed guidelines for using florpyrauxifen-benzyl in these cultivars and others like it.

Introduction

Florpyrauxifen-benzyl is a new synthetic auxin herbicide (Weed Science Society of America [WSSA] Group 4) released for commercial use in rice in 2018 by Corteva Agriscience. Previous research has explored both the weed control spectrum and the chemical properties of the herbicide, including residual activity and translocation. Florpyrauxifen-benzyl controls many troublesome weeds in rice production, including yellow nutsedge (*Cyperus esculentus* L.), hemp sesbania [*Sesbania herbacea* (Mill.) McVaugh], and barnyardgrass [*Echinochloa crus-galli* (L.) P. Beauv.] when used at the labeled rate of 30 g ae ha⁻¹ (Miller and Norsworthy 2018a). Florpyrauxifen-benzyl has a site of action that is different than that of quinclorac (WSSA Group 4), favoring the AFB5 IAA co-receptor instead of the TIR1 co-receptor, which allows florpyrauxifen-benzyl to have activity on quinclorac-resistant barnyardgrass (Lee et al. 2014; Miller et al. 2018; Walsh et al. 2006). Florpyrauxifen-benzyl has minimal residual activity and should therefore be used in conjunction with a herbicide with residual activity to control weeds with prolonged emergence (Miller and Norsworthy 2018b). Florpyrauxifen-benzyl can be applied at 30 g ae ha⁻¹, with a maximum of two applications per growing season (Anonymous 2017).

Much of the research conducted with florpyrauxifen-benzyl to date has focused on weed control and characterization of its chemical properties. Thus far little research has been conducted to determine differences in cultivar responses to an application of the herbicide. Injury symptoms from florpyrauxifen-benzyl can be in the form of leaf malformations, reduced

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height, and reduced biomass (JKN, personal communication). The herbicide label also warns of potential risk for rice injury to long-grain hybrid cultivars and medium-grain varieties (Anonymous 2017).

Arkansas is the top producer of rice in the United States, producing roughly half of all rice hectares harvested in the country in 2018 (USDA-NASS 2019). Of the rice grown in Arkansas, 49% is planted with long-grain hybrid cultivars; 39% is planted with long-grain, pureline cultivars; and 13% is planted with medium-grain, pureline cultivars (Hardke 2018). Understanding cultivar tolerances to new herbicides is crucial to reducing the risk of yield loss and to make appropriate rate and timing recommendations for producers.

Tolerance of crops to herbicides is due in part to the ability of a crop to metabolize and detoxify the herbicide (Cole 1994). Different cultivars of the same crop can exhibit different levels of tolerance to a herbicide. A good example of this is the differential tolerance of soybean [*Glycine max* (L.) Merr.] varieties to metribuzin (Hardcastle 1979). Rice varieties also exhibit differential responses to some herbicides. For example, the imidazolinone-tolerant rice cultivars differ in tolerance to imazamox (Bond and Walker 2011), wherein two rice hybrid cultivars exhibited more injury than a long-grain pureline cultivar to an application of imazamox.

Many factors can affect crop tolerance to a herbicide, including herbicide rate, crop growth stage at application, and environmental factors near the time of application. The study by Bond and Walker (2011) demonstrated cultivar differences in response to a herbicide and the effect of growth stage on injury and yield. Grain yield of hybrid cultivars was reduced 9% to 21% when imazamox was applied 14 d after panicle initiation and at boot, but not when the herbicide was applied at panicle initiation. Differential tolerance of rice cultivars is well documented, with numerous examples of growth stage, herbicide rate, and cultivar.

Environmental conditions surrounding the time of application can influence injury following a herbicide application. In a growth chamber experiment, corn (*Zea mays* L.) growth was reduced following treatment with thiocarbamate herbicides at a higher temperature regime (Burt and Akinsorotan 1976). Plant growth was slowed following herbicide treatment, regardless of herbicide rate or soil moisture, at 30 C compared with 20 C. Conversely, in a different study by Wright and Rieck (1974), dry weights from various corn hybrids were reduced following an application of butylate when the temperature was 20 C compared with 33 C. Both experiments demonstrate that environmental conditions can influence crop response to a herbicide. Because of the limited knowledge of the response of rice cultivars to florypyrauxifen-benzyl, the objective of these experiments was to consider the effect of florypyrauxifen-benzyl rate, days between sequential applications, temperature (environmental conditions), and growth stage on three rice cultivars.

Materials and Methods

Sequential Applications of Florypyrauxifen-Benzyl

Field experiments were conducted in 2018 at the Pine Tree Research Station (PTRS) near Colt, AR, and the Rice Research and Extension Center (RREC) in Stuttgart, AR, to evaluate the effect of florypyrauxifen-benzyl rate and number of days between applications on injury and grain yield of three rice cultivars. The soil at PTRS was a Calloway silt loam (fine-silty, mixed, active,

thermic Aquic Fraglossudalf) with 1.3% organic matter, 10.6% sand, 68.6% silt, and 20.8% clay, pH 7.5. The soil at RREC was a DeWitt silt loam (fine, smectic, thermic typic Albaqualf) with 1.8% organic matter, 8.4% sand, 71.4% silt, and 20.2% clay, pH 6.0.

This experiment was set up as a randomized complete block, two-factor factorial with four replications. The first factor was florypyrauxifen-benzyl rate and the second factor was the number of days between sequential applications. Long-grain pureline cultivar CL111, medium-grain pureline cultivar CL272, and long-grain hybrid cultivar CLXL745 were selected for these studies due to their significant planted acreage in 2016. Rice was drill-seeded on April 19, 2018, at both locations using a 10-row cone drill with 18-cm row spacing. The plots were 5.2 m long. Pureline cultivars were seeded at 72 seeds m^{-1} per row and the hybrid cultivar was seeded at 26 seeds m^{-1} per row. A nontreated control in each block was included for each cultivar. These trials were maintained weed-free using labeled herbicides, hand-weeded as necessary, and were managed according to University of Arkansas System Division of Agriculture recommendations for direct-seeded, delayed-flood rice production (Roberts et al. 2018). Preflood applications of nitrogen in the form of urea totaling 130 kg N ha^{-1} were applied at the 5-leaf growth stage at both locations. Muriate of potash (MOP) was applied immediately following emergence at PTRS, and preplant MOP and phosphorous were applied and incorporated prior to planting at RREC.

Florypyrauxifen-benzyl (Loyant™ Herbicide, Dow AgroSciences LLC, Indianapolis, IN) was applied at 30 or 60 g $ae\ ha^{-1}$ early POST on 2- to 3-leaf rice. Second applications of florypyrauxifen-benzyl were targeted for 7, 10, 14, and 21 d after the first application, but were actually made 5, 13, 18, and 21 d after the initial application at PTRS and 4, 11, 14, and 20 d after the initial application at RREC. Application dates at PTRS were May 17 for the first application, and May 22, May 30, June 4, and June 7 for the sequential applications. At RREC, the first application was made on May 17 and sequential applications were made on May 21, May 28, May 31, and June 6. The same florypyrauxifen-benzyl rates were used in the sequential application such that plots that received 30 g $ae\ ha^{-1}$ early POST also received 30 g $ae\ ha^{-1}$ in the sequential application and plots that received 60 g $ae\ ha^{-1}$ early POST also received 60 g $ae\ ha^{-1}$ in the sequential application. Methylated seed oil was added to all florypyrauxifen-benzyl treatments at 0.6 L ha^{-1} . Herbicide treatments were made using a CO₂-pressurized backpack sprayer at 140 L ha^{-1} with 110015 AIXR nozzles (TeeJet Technologies, Springfield, IL). The flood was established at PTRS on June 2, 2018, and at RREC on May 31, 2018. Visible injury was assessed 2 and 3 wk after the second application on a scale of 0% to 100%, where 0 equals no injury and 100 equals crop death (Frans and Talbert 1977). Visible injury included leaf malformations, reduced height, and decreased biomass. Additionally, groundcover and yield can be correlated (Donald 1998); thus, groundcover was assessed using drone images taken with a DJI Phantom 4 Pro drone equipped with a multispectral camera (Sentra, Minneapolis, MN). Images were taken 1 and 3 wk after the last application at each location from a height of approximately 60 m. Those images consisted of approximately 6.7 million pixels and were analyzed using Field Analyzer (Turf Analyzer, Fayetteville, AR) to calculate the percent of groundcover. Rough rice grain yield was harvested from the center of each plot using a small-plot combine and adjusted to 12% moisture. Groundcover is reported as a percentage of the weed-free nontreated control.

Florypyrauxifen-Benzyl Rate and Temperature

A growth chamber experiment was conducted at the University of Arkansas Altheimer Laboratory in Fayetteville, AR, in autumn 2018 and repeated twice in spring 2019 to determine the effect of different day/night temperatures on injury of rice cultivars following an application of florypyrauxifen-benzyl. Rice was seeded into 10-cm-diameter pots filled with a 50:50 (vol/vol) mixture of field soil and potting mix (Metro-Mix®, Sun Gro Horticulture, Agawam, MA), thinned to one plant per pot, and grown in the greenhouse until plants reached the 2-leaf growth stage. Field soil was a Captina silt loam (Fine-silty, siliceous, active, mesic Typic Fragiudults) with 1.7% organic matter, pH 6.1. The same cultivars in the field experiments were used in this experiment, and nontreated plants were included. This experiment was set up as a split-plot design with temperature as the whole-plot factor and florypyrauxifen-benzyl rate as the split-plot factor. There were three runs in time and five replications per run. Plants were placed in their respective growth chambers at the 2- to 3-leaf growth stage 3 d before herbicide application to allow acclimation. Each chamber was set to 27/18 C and 32/24 C day/night temperature respectively, with a 16-h photoperiod. Light quantity at plant height in both growth chambers was approximately 500 $\mu\text{mol m}^{-2} \text{s}^{-1}$. Florypyrauxifen-benzyl was applied at a rate of 0 (nontreated), 30, and 60 g ae ha⁻¹ using a two-nozzle boom equipped with 800067 flat-fan nozzles in a spray chamber calibrated to deliver 187 L ha⁻¹. Methylated seed oil was added to all florypyrauxifen-benzyl treatments at 0.6 L ha⁻¹. Plants were returned to their respective growth chamber following application. Injury was assessed 14 and 28 d after application (DAA) on a scale of 0% to 100%, where 0 is no injury and 100 is crop death. Plant heights were recorded 14 and 28 DAA by stretching the plant and measuring the tallest leaf. Tillers were counted and aboveground biomass collected at 28 DAA, then oven dried at 66 C for 4 d and weighed. Heights, tillers, and dry biomass are reported relative to the nontreated plants.

Florypyrauxifen-Benzyl Rate and Growth Stage

A greenhouse experiment was conducted in autumn 2018 and spring 2019 at the University of Arkansas Altheimer Laboratory in Fayetteville, AR, to evaluate the effect of florypyrauxifen-benzyl rate and growth stage on rice injury. The three cultivars used in field and growth chamber experiments were seeded into 10-cm-diameter pots with a 50:50 mixture of field soil and potting mix (Metro-Mix®, Sun Gro Horticulture) and thinned to one plant per pot. Field soil was a Captina silt loam (Fine-silty, siliceous, active, mesic Typic Fragiudults) with 1.7% organic matter, pH 6.1. Plants were grown in the greenhouse at a 32 C daytime and 22 C nighttime (± 3 C) temperature regime with a 16-h photoperiod. This experiment was established as a completely randomized design, two-factor factorial, with florypyrauxifen-benzyl rate being one factor and growth stage as the second factor. There were three runs in time with five replications per run, and nontreated plants were included for each cultivar at each growth stage. Florypyrauxifen-benzyl was applied at 30 or 60 g ae ha⁻¹ at the 1-, 3-, or 5-leaf stage and immediately returned to the greenhouse. Applications were made as described previously using the same setup as noted for the growth chamber experiment. Visible injury was assessed 14 and 28 DAA on a 0% to 100% scale for each application timing. Heights were also measured 14 and 28 DAA. Tillers were counted and aboveground biomass was collected at 28 DAA.

Heights, tillers, and biomass are reported relative to the nontreated plants.

Statistical Analyses

All data were analyzed using the GLIMMIX procedure in SAS 9.4 (SAS Institute, Cary, NC). Each cultivar in the field experiment was analyzed separately by location because of differences in environmental conditions during and after application, and differences in number of days between sequential applications for both locations. A beta distribution was assumed for injury data (Gbur et al. 2012). Due to a significant Shapiro-Wilk test, a gamma distribution was assumed for yield and percent groundcover (Gbur et al. 2012). Because of the large number of zero days delayed in 50% heading, formal analysis was not performed on heading data. Thus, delays in 50% heading data are reported with mean and standard error. Cultivars were also analyzed separately in the greenhouse and growth chamber experiments. Again, a beta distribution was assumed for injury and a gamma distribution was assumed for height, tiller number, and biomass. Replication and runs were considered random effects with replication nested within run. All data were subject to analysis of variance using Fisher's protected least significant difference ($\alpha = 0.05$) to separate means when appropriate. P-values from the field experiment are provided in Table 1 and P-values from the growth chamber and greenhouse experiments are provided in Table 2.

Results

Long-Grain Pureline Cultivar

The field experiment conducted on CL111 showed florypyrauxifen-benzyl had little effect on this cultivar. In the field experiment, CL111 was injured less than 10% at 3 wk after sequential applications (WAA) of florypyrauxifen-benzyl, regardless of herbicide rate (Table 3). Florypyrauxifen-benzyl did not reduce groundcover relative to the nontreated control at 1 or 3 WAA at either location. The delay in 50% heading was no more than 1.5 d and there was no reduction in yield (Table 4).

Visible injury in the growth chamber was consistent with the field experiment, with no more than 9% injury 14 DAA (Table 5). Plant heights recorded 28 DAA indicate plants treated with florypyrauxifen-benzyl at 60 g ae ha⁻¹ constituted 16% of the nontreated plants, whereas plants treated with 30 g ae ha⁻¹ constituted 13% of the nontreated plants, averaged over temperature (Table 5). Neither high temperature nor florypyrauxifen-benzyl rate reduced number of tillers or biomass collected 28 DAA (Table 5). This suggests applications of florypyrauxifen-benzyl to CL111 will not have significant or lasting net negative effects.

In the growth stage experiment, 17% injury was observed 14 DAA when florypyrauxifen-benzyl was applied to 1-leaf rice, averaged over rates (Table 5). This was the highest injury observed in the growth stage experiment. The sensitivity of young plants to this herbicide justifies the label restriction of florypyrauxifen-benzyl applications allowed to 2-leaf or larger rice (Anonymous 2017). The levels of injury associated with applications made to 3- and 5-leaf rice are consistent with both the sequential application field experiment and the growth stage experiment, suggesting that florypyrauxifen-benzyl causes low risk for high levels of visible injury to CL111.

There was a reduction from the nontreated plants in height, tiller production, and biomass associated with florypyrauxifen-benzyl treatments in the growth stage experiment (Table 5).

Table 1. P-values for the long-grain pureline cultivar CL111, the medium-grain pureline cultivar CL272, and the long-grain hybrid cultivar CLXL745 in 2018 for florypyrauxifen-benzyl rate and number of days between sequential applications.^{a,b}

Cultivar	Factor	PTRS					RREC				
		Injury		Yield	Relative groundcover		Injury		Yield	Relative groundcover	
		2 WAA	3 WAA		1 WAA	3 WAA	2 WAA	3 WAA		1 WAA	3 WAA
P-value											
CL111	Rate	0.1769	0.1616				0.0477*	0.0142*			
	Days	0.1881	0.1651				0.8193	0.0018*			
	Rate × days	0.3915	0.4472	0.5021	0.1158	0.1647	0.5097	0.3957	0.6533	0.6577	0.9043
CL272	Rate	<0.0001*	0.2138				0.1051	0.0423*			
	Days	<0.0001*	0.0026*				0.0363*	<0.0001*			
	Rate × days	0.1047	0.4488	0.2397	0.2451	0.6408	0.4368	0.2153	0.2385	0.1691	0.2702
CLXL 745	Rate	0.0024*	0.0012*				0.0029*	0.0522			
	Days	0.1103	<0.0001*				0.0002*	<0.0001*			
	Rate × days	0.3853	0.2459	0.0366*	0.0008*	0.1593	0.1325	0.9458	0.0254*	<0.0001*	0.5508

^aAbbreviations: PTRS, Pine Tree Research Station; RREC, Rice Research and Extension Center; WAA, weeks after application.

^bAsterisks (*) indicates significance at P = 0.05.

Table 2. P-values for the long-grain pureline cultivar CL111, medium-grain pureline cultivar CL272, and long-grain hybrid cultivar CLXL745 for injury, height, tiller number, and biomass for the growth chamber and greenhouse experiments.^{a,b}

Cultivar	Experiment	Factor	Injury		Height		Tillers	Biomass
			14 DAA	28 DAA	14 DAA	28 DAA		
			P-value					
CL111	Growth chamber	Temperature	0.3680	0.4191	0.3416	0.0658	0.4074	0.2259
		Rate	0.0016*	0.0367*	0.0004*	<0.0001*	0.0329*	0.6354
		Temperature × rate	0.4808	0.0401*	0.8486	0.0750	0.6126	0.6509
	Greenhouse	Stage	<0.0001*	0.0021*	0.0006*	0.0112*	<0.0001*	0.1156
		Rate	<0.0001*	0.0347*	0.0021*	0.2763	0.3897	0.0007*
		Stage × rate	0.3901	0.0015*	0.0186*	0.0635	0.0138*	0.0198*
CL272	Growth chamber	Temperature	0.0669	0.1915	0.0061*	<0.0001*	0.1496	0.0004*
		Rate	0.0406*	0.0695	0.5096	<0.0001*	0.3754	0.0008*
		Temperature × rate	0.0008*	0.0279*	0.4517	<0.0001*	0.5201	0.0217*
	Greenhouse	Stage	<0.0001*	0.4594	<0.0001*	0.0390*	0.5314	0.2121
		Rate	0.0359*	<0.0001*	<0.0001*	0.0003*	0.1382	<0.0001*
		Stage × rate	0.7766	0.5063	<0.0001*	0.1889	0.9559	0.7015
CLXL745	Growth chamber	Temperature	0.1223	0.9371	0.0115*	<0.0001*	0.1197	0.5640
		Rate	0.0027*	0.1610	0.4894	0.4192	0.3496	0.0056*
		Temperature x rate	0.0038*	0.0458*	0.2956	0.0080*	0.6941	0.2013
	Greenhouse	Stage	<0.0001*	0.0004*	0.0086*	<0.0001*	0.2520	0.9701
		Rate	0.0002*	0.0117*	<0.0001*	0.0090*	0.1526	0.0003*
		Stage x rate	0.0025*	0.2828	0.1552	0.0094*	0.0549	0.0935

^aAbbreviation: DAA, days after application.

^bAsterisks (*) indicate significance at P = 0.05.

Table 3. Injury for CL111 in 2018 as influenced by florypyrauxifen-benzyl rate and number of days between sequential applications.^a

		Injury RREC ^b	
Factor		2 WAA	3 WAA
		%	
Rate ^c	30 fb 30	5 b	1 b
	60 fb 60	11 a	4 a
Days	4		7 a
	11		3 b
	14		3 b
	20		<1 c

^aAbbreviations: fb, followed by; RREC, Rice Research and Extension Center; WAA, weeks after second application.

^bMeans are separated using Fisher's protected least significant difference ($\alpha = 0.05$). Means with the same letter within a factor and column are not significantly different.

^cFlorypyrauxifen-benzyl rate is reported in g ae ha⁻¹.

Height 14 DAA was reduced from that of nontreated plants when florypyrauxifen-benzyl was applied at the 1-leaf growth stage (Table 5). There was a greater than 20% reduction in tillers associated with applications made to 1-leaf plants, regardless of florypyrauxifen-benzyl rate (Table 5). Florypyrauxifen-benzyl at 60 g ae ha⁻¹ reduced biomass at all growth stages tested, indicating that injury may not always be detected visually (Table 5). Florypyrauxifen-benzyl applied at 30 g ae ha⁻¹ to 1-leaf rice plants also reduced biomass. Due to label restrictions the reductions in height, tillers, and biomass observed when the herbicide is applied at the 1-leaf stage are unlikely to be problematic in production scenarios.

It is possible that differences in the level of injury observed in the temperature and growth stage experiments versus the field experiment are the result of lower light quantity and quality.

Table 4. Heading and grain yield of the long-grain pureline cultivar CL111.^a

Location	Florypyrauxifen-benzyl rate ^b	Days between sequential applications	Delay in 50% heading ^{c,d,e}		Grain yield ^c
			— days —		kg ha ⁻¹
PTRS	Nontreated	—	—	—	7,490
		5	1.0	(1.2)	8,500
		13	0	(0)	8,410
		18	1.5	(1.9)	7,110
		21	1.3	(1.9)	7,660
	60 fb 60	5	1.0	(0.8)	7,940
		13	0.5	(0.6)	7,370
		18	1.5	(0.6)	6,990
		21	1.3	(1.9)	7,480
		—	—	—	8,510
RREC	Nontreated	4	1.0	(1.4)	7,690
		11	0.8	(1.0)	7,470
		14	1.0	(0.8)	7,760
		20	1.0	(1.4)	8,030
		—	—	—	8,510
	60 fb 60	4	1.0	(1.2)	8,260
		11	1.0	(0.8)	8,640
		14	1.5	(1.0)	8,440
		20	1.3	(0.5)	7,040
		—	—	—	8,510

^aAbbreviations: fb, followed by; PTRS, Pine Tree Research Station; RREC, Rice Research and Extension Center.^bFlorypyrauxifen-benzyl rate is reported in g ae ha⁻¹.^cThere were no significant differences in delay in 50% heading or grain yield.^dMean followed by standard error in parenthesis.^eHyphens (-) represent nontreated delay in heading as zero.**Table 5.** Injury, height, tiller number, and biomass for the long-grain pureline cultivar CL111 as influenced by day/night temperature regime and florypyrauxifen-benzyl rate for the growth chamber experiment and growth stage at application and florypyrauxifen-benzyl rate for the greenhouse experiment.^{a,b}

		Injury ^c		Height ^e			
Factor		14 DAA	28 DAA	14 DAA	28 DAA	Tiller ^e	Biomass ^e
Growth chamber		% of nontreated					
Temperature ^d	27/18						
	32/24						
Rate ^d	Nontreated	—		100 b	100 a	100 b	
	30	3 b	2 b	114 a	87 b	104 ab	
	60	9 a	4 a	112 a	84 c	114 a	
Temperature × rate ^d	27/18 nontreated	—					
	27/18 × 30		1 b				
	27/18 × 60		6 a				
	32/24 nontreated	—					
	32/24 × 30		3 b				
	32/24 × 60		3 b				
Greenhouse		% of nontreated					
Stage	1-leaf	17 a		81 b	97 ab	85 c	
	3-leaf	12 b		99 a	102 a	110 a	
	5-leaf	6 c		93 a	91 b	95 b	
Rate ^d	Nontreated	—		100 a			100 a
	30	8 b		84 b			81 b
	60	15 a		89 b			71 b
Stage × rate ^d	1-leaf nontreated	—		100 a		100 abc	100 ab
	1-leaf × 30		15 a	66 c		76 e	55 d
	1-leaf × 60		9 ab	81 b		79 ed	74 bcd
	3-leaf nontreated	—		100 a		100 abc	100 ab
	3-leaf × 30		5 cd	99 a		118 a	107 a
	3-leaf × 60		11 ab	98 a		113 ab	67 dc
	5-leaf nontreated	—		100 a		100 abc	100 ab
	5-leaf × 30		4 d	91 ab		90 cde	89 abc
	5-leaf × 60		8 bc	89 ab		94 bcd	71 dc

^aAbbreviation: DAA, d after application.^bMeans are separated using Fisher's protected least significant difference ($\alpha = 0.05$). Means with the same letter within a factor and column are not significantly different. Data not shown for some treatments and variables indicate data is not significant.^cHyphens (-) represent nontreated data as 0% injury.^dTemperature is reported in celsius (C); florypyrauxifen-benzyl rate is reported in g ae ha⁻¹.^eHeight 14 DAA for the nontreated in the 27/18 C growth chamber was 30 cm and 36 cm for the nontreated in the 32/24 C growth chamber. Average number of tillers for the nontreated in both growth chambers was 4. Height at 14 DAA was 38, 41, and 59 cm and at 28 DAA was 45, 47, and 75 cm for 1-, 3-, and 5-leaf growth stages, respectively. Additionally, average number of tillers for the nontreated of all growth stages was 2. Biomass of the nontreated treatments for 1-, 3-, and 5-leaf growth stages at 28 DAA was 0.35 g, 0.60 g, and 1.80 g, respectively.

Table 6. Injury for the medium-grain pureline cultivar CL272 in 2018 as influenced by florpyrauxifen-benzyl rate and number of days between sequential applications.^a

Location	Factor	Treatment	Injury ^b	
			2 WAA	3 WAA
			%	
PTRS	Rate ^c	30 fb 30	6 b	
		60 fb 60	15 a	
	Days	5	5 b	11 a
		13	14 a	6 b
		18	17 a	6 b
RREC	Rate ^c	21	6 b	3 c
		30 fb 30		10 b
	Days	60 fb 60		21 a
		4	30 a	50 a
		11	31 a	23 b
		14	25 a	15 b
		20	2 b	2 c

^aAbbreviations: fb, followed by; PTRS, Pine Tree Research Station; RREC, Rice Research and Extension Center; WAA, weeks after second application.

^bMeans are separated using Fisher's protected least significant difference ($\alpha = 0.05$). Means with the same letter within a factor and column are not significantly different.

^cFlorpyrauxifen-benzyl rate is reported in g ae ha⁻¹.

Sunlight on a clear day in the summer months often exceeds 2,000 $\mu\text{mol m}^{-2} \text{s}^{-1}$; however, this level of light intensity drastically decreases on a cloudy day, which in turn decreases photosynthesis and herbicide metabolism (Bazzaz and Carlson 1982). Light quantity in the temperature experiment conducted in the growth chambers was near 500 $\mu\text{mol m}^{-2} \text{s}^{-1}$. Although light in the greenhouse was supplemented with a light-emitting diode system on a 16-h photoperiod, low light quantity and quality was likely in the greenhouse because the experiment was conducted in winter months. Bond and Walker (2012) attributed the decrease in the translocation and metabolism of quinclorac in rice to lower solar radiation, or light intensity. Furthermore, Pritchard and Warren (1980) documented a reduction in tomato (*Solanum lycopersicum* L.), velvetleaf (*Abutilon theophrasti* Medik.), and jimsonweed (*Datura stramonium* L.) tolerance to metribuzin when plants were shaded prior to herbicide application. In that study, light intensity was reduced 76% by using a shade cloth, which simulates light conditions on a moderately cloudy day (Pritchard and Warren 1980). The study by Prichard and Warren (1980) also attributed reduced herbicide metabolism to a reduction in photosynthesis. Thus, the low light intensity conditions observed in the greenhouse and growth chamber studies conducted in this research likely affected florypyrauxifen-benzyl metabolism, leading to a decrease in height and biomass.

Based on these three experiments, we conclude that the long-grain pureline cultivar CL111 exhibits sufficient tolerance to florypyrauxifen-benzyl. Low levels of visible injury, little impact on groundcover, no more than a 1.5-d delay in heading, and no yield loss lead to the conclusion that sequential applications of florypyrauxifen-benzyl do not cause serious, lasting impacts on CL111 when applied at the 2-leaf growth stage with a minimum of 14 d between applications.

Medium-Grain Pureline Cultivar

Injury to CL272 was no more than 11% at PTRS at 3 WAA, whereas up to 50% injury was observed at RREC (Table 6). At RREC, applications made 14 d apart, the minimum length of

time required by the label, resulted in only 15% injury (Table 6). Florypyrauxifen-benzyl applied more than 20 d apart injured rice by less than 5%. Yield of CL272 was not reduced at either site, which is not surprising considering there was no more than a 3-d delay in 50% heading (Table 7).

Injury of CL272 rice in the temperature experiment was consistent with injury at PTRS in the field experiment (Table 8). Although little injury was observed, height and biomass were reduced by florypyrauxifen-benzyl. Generally, height and biomass were reduced at least 20% and 13%, respectively, compared with control plants, regardless of rate and temperature (Table 8).

In the greenhouse experiment, injury to CL272 at 14 DAA was highest when applied to 1-leaf plants (46%) when averaged over florypyrauxifen-benzyl rate, with injury generally decreasing as applications were delayed (Table 8). Although injury in this experiment was higher than observed in the field and growth chamber experiments, it was expected because applications in those experiments were made to 2- to 3-leaf rather than 1-leaf rice. In a weed control experiment by Teló et al. (2018), fall panicum control (*Panicum dichotomiflorum* Michx.) was higher when florypyrauxifen-benzyl was applied at 30 g ae ha⁻¹ to 3- to 4-leaf rice as opposed to larger, 1- to 2-tiller rice. This may be applied to rice as well, although rice is more tolerant to florypyrauxifen-benzyl than fall panicum. When rice plants are small, they will be more affected by an application of florypyrauxifen-benzyl than when plants are larger, likely due to an increase in plant growth and metabolism.

Additionally, there was no reduction in rice height 14 DAA from any treatment, except those applied to 1-leaf rice (Table 8). As in the temperature experiment, biomass of plants treated with florypyrauxifen-benzyl was significantly reduced from the nontreated plants; biomass of plants treated with 30 g ae ha⁻¹ was 74% of nontreated plants, and biomass from plants treated with 60 g ae ha⁻¹ was 62% of nontreated plants, averaged over growth stage (Table 8).

These results indicate that CL272 can potentially recover from early-season injury. However, considering the high level of injury at RREC, growers should abide by the 14-d interval between applications to minimize potential negative effects on the rice crop. Based on these data, injury appears to be exacerbated by warmer temperatures; however, plants recover more quickly under relatively warmer compared to cooler temperatures. This was observed in the field experiment at RREC, a period of warm temperatures between May 29 and June 4 (average high temperature 33 °C) caused greater than expected injury from applications made May 28 and 31 (Table 6; Figure 1A and 1B).

Long-Grain Hybrid Cultivar

At PTRS, CLXL745 injury 3 wk after florypyrauxifen-benzyl was applied 5 d apart was 34%, averaged over herbicide rate. However, applications made 4 d apart at RREC resulted in 64% injury when averaged over herbicide rate (Table 9). Injury decreases as there is more time between sequential applications; however, at RREC 17% injury was still observed 3 WAA when applications were made 14 d apart, averaged over rate (Table 9).

There was a significant reduction in CLXL745 rice groundcover relative to the nontreated control 1 WAA at both locations ($P = 0.0008$ at PTRS, $P < 0.0001$ at RREC). At PTRS, sequential applications of florypyrauxifen-benzyl at 60 g ae ha⁻¹ made 5 d and 13 d apart resulted in 47% and 76% groundcover relative to the nontreated control, respectively, whereas sequential applications

Table 7. Heading and grain yield of the medium-grain pureline cultivar CL272.^a

Location	Floryprauxifen-benzyl rate ^b	Days between sequential applications	Delay (in days) in 50% heading ^{c,d,e}		Grain yield ^{d,f}
PTRS	Nontreated	5	–	–	8,040
		13	0.8	(0.5)	7,840
		18	1.3	(1.9)	6,750
		21	0.8	(1.0)	8,170
	60 fb 60	5	1.0	(2.0)	8,190
		13	3.0	(0.8)	8,020
		18	2.8	(1.5)	8,450
		21	2.3	(1.7)	7,380
	Nontreated	21	2.5	(2.1)	7,550
		4	–	–	6,400
		11	1.5	(1.0)	6,290
		14	1.0	(0.8)	6,560
RREC	30 fb 30	14	1.3	(1.3)	6,270
		20	2.3	(2.8)	5,990
		4	2.3	(1.5)	6,660
		11	2.5	(1.9)	7,190
	60 fb 60	14	2.0	(2.7)	5,840
		20	1.5	(2.4)	5,880

^aAbbreviations: fb, followed by; PTRS, Pine Tree Research Station; RREC, Rice Research and Extension Center.

^bFloryprauxifen-benzyl rate is reported in g ae ha⁻¹.

^cMean followed by standard error in parenthesis.

^dThere were no significant differences in delay in 50% heading or grain yield.

^eHyphens (-) represent nontreated delay in heading as zero.

^fGrain yield is reported in kg ha⁻¹.

Table 8. Injury and height for the medium-grain pureline cultivar CL272 in the grow chamber experiment as influenced by day/night temperature regime and floryprauxifen-benzyl rate and by growth stage and floryprauxifen-benzyl rate in the greenhouse experiment.^a

		Injury ^{b,c}		Height ^{b,d}		
Factor		14 DAA	28 DAA	14 DAA	28 DAA	Biomass ^{b,d}
Growth chamber		% of nontreated				
Temperature ^e	27/18			104 a	79 b	82 b
	32/24			98 b	85 a	95 a
Rate ^e	Nontreated	–			100 a	100 a
	30	9 b			74 b	81 b
	60	13 a			72 c	86 b
	Nontreated	–			100 a	100 a
Temperature × rate	27/18 × 30	11 b	4 b		73 d	76 b
	27/18 × 60	8 b	5 b		64 e	72 b
	Nontreated	–			100 a	100 a
	32/24 × 30	8 b	2 b		76 c	86 ab
	32/24 × 60	18 a	15 a		80 b	100 a
Greenhouse		% of nontreated				
Stage	1-leaf	46 a		67 b	96 ab	
	3-leaf	24 b		96 a	98 a	
	5-leaf	14 c		92 a	89 b	
Rate	Nontreated	–		100 a	100 a	100 a
	30	22 b	12 b	77 b	96 a	74 b
	60	30 a	26 a	77 b	87 b	62 b
Stage × rate	1-leaf nontreated			100 a		
	1-leaf × 30			52 b		
	1-leaf × 60			58 b		
	3-leaf nontreated			100 a		
	3-leaf × 30			98 a		
	3-leaf × 60			90 a		
	5-leaf nontreated			100 a		
	5-leaf × 30			91 a		
	5-leaf × 60			87 a		

^aAbbreviation: DAA, d after application.

^bMeans are separated using Fisher's protected least significant difference ($\alpha=0.05$). Means with the same letter within a factor and column are not significantly different. Data not shown for some treatments and variables indicate data is not significant.

^cHyphens (-) represent nontreated data as 0% injury.

^dHeight and biomass collected at 28 DAA for the nontreated of the 27/18 C growth chamber was 35 cm, 4, and 1.80 g, respectively, and for the nontreated of the 32/24 C growth chamber 44 cm, 3, and 1.60 g, respectively. Height 14 DAA was 51, 55, and 67 cm and 28 DAA was 60, 61, and 82 cm for the nontreated of 1-, 3-, and 5-leaf growth stages. Biomass for the nontreated at the 1-, 3-, and 5-leaf growth stages was 0.40 g, 0.95 g, and 1.60 g at 28 DAA, respectively in the greenhouse experiment.

^eTemperature is reported in celsius (C); floryprauxifen-benzyl rate is reported in g ae ha⁻¹

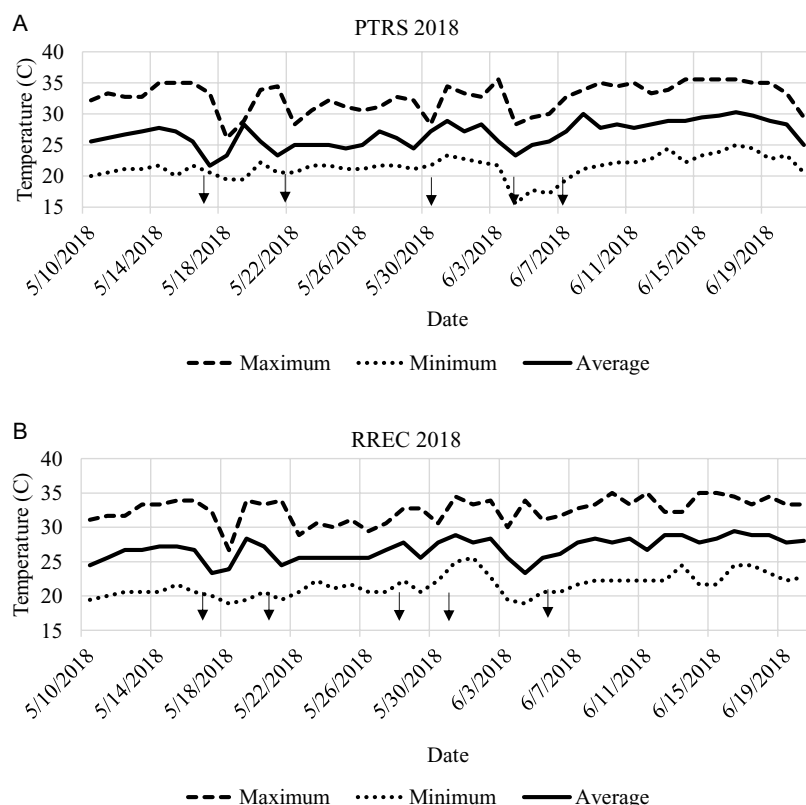


Figure 1. Daily minimum, maximum, and average temperatures (A) at the Pine Tree Research Station (PTRS) near Colt, AR, in 2018 and (B) the Rice Research and Extension Center (RREC) near Stuttgart, AR, in 2018 for dates ranging from 7 d before first florypyrauxifen-benzyl application to 14 d after the second application. Rice cultivars CL111, CL272, and CLXL745 were planted on April 19, 2018, at PTRS and RREC. The first florypyrauxifen-benzyl application was made to 2- to 3-leaf rice on May 17, 2018, at both locations. Sequential applications were made May 22, May 30, June 4, and June 7 at PTRS and May 21, May 28, May 31, and June 6 at RREC. Application dates are indicated by an arrow.

Table 9. Injury for the long-grain rice hybrid cultivar CLXL745 in 2018 as influenced by florypyrauxifen-benzyl rate and number of days between sequential applications.^{a,b}

			Injury ^b	
Location	Factor	Treatment ^c	2 WAA	3 WAA
			%	
PTRS	Rate	30 fb 30	5 b	9 b
		60 fb 60	24 a	22 a
	Days	5		34 a
		13		17 b
		18		10 bc
21			6 c	
RREC	Rate	30 fb 30	24 b	
		60 fb 60	46 a	
	Days	4	66 a	64 a
		11	24 b	13 b
		14	30 b	17 b
20		22 b	1 c	

^aAbbreviations: fb, followed by; PTRS, Pine Tree Research Station; RREC, Rice Research and Extension Center; WAA, weeks after second application.

^bMeans are separated using Fisher's protected least significant difference ($\alpha = 0.05$). Means with the same letter within a factor and column are not significantly different.

^cFlorypyrauxifen-benzyl rate is reported in g ae ha⁻¹.

of 60 g ae ha⁻¹ made 4 d apart resulted in 29% groundcover (Table 10).

Rough rice grain yields for most treatments were significantly reduced from the nontreated control, even in treatments where label directions were followed (Table 10). At PTRS, CLXL745 grain

yields following treatments where florypyrauxifen-benzyl was applied sequentially at 30 g ae ha⁻¹ 18 d apart or 60 g ae ha⁻¹ 21 d apart were not significantly less than those of the nontreated controls. Additionally, at RREC, applications made 4 d apart at both rates were not significantly less than those applied to the nontreated controls. This could be due to rice plants having additional time to recover from injury before harvest compared to the other treatments, however because this is inconsistent with results from PTRS, further research is needed to confirm this. Yield reduction from herbicide applications has been observed in other studies as well. Mid-season herbicide injury from quinclorac has been shown to reduce yield up to 19% for another hybrid cultivar, 'XL723' (Bond and Walker 2012). Yield reductions coupled with injury data lead to the conclusion that CLXL745 is particularly sensitive to sequential applications of florypyrauxifen-benzyl.

In the growth chamber experiment, injury was 25% at 14 DAA for florypyrauxifen-benzyl at 60 g ae ha⁻¹ applied to plants maintained at 32/24 C (Table 11). By 28 DAA, no more than 10% injury was observed, indicating that plants had begun to recover. Recovery is also reflected in heights of CLXL745 at 28 DAA, when heights of plants treated with florypyrauxifen-benzyl in the 32/24 C growth chamber were not different than those of nontreated plants (Table 11). Heights of treated plants, regardless of rate, were reduced from nontreated plants in the 27/18 C growth chamber; however, this difference was numerically small (Table 11). This suggests that plants may recover more quickly under warmer growing conditions. Because differences were slight, temperature appears to have a minimal effect on hybrid recovery from

Table 10. Heading and grain yield of the long-grain rice hybrid cultivar CLXL745.^a

Location	Florpyrauxifen-benzyl rate ^b	Days between sequential applications	Delay in 50% heading ^{c,d,e}		Grain yield ^d	Relative groundcover 1 WAA ^d
	g ae ha ⁻¹		—d—		kg ha ⁻¹	%
PTRS	Nontreated		—	—	11,180 a	100 a
	30 fb 30	5	1.0	(1.4)	9,910 bc	90 ab
		13	0.8	(1.0)	9,910 bc	101 a
		18	0	(0)	10,530 ab	107 a
		21	0.3	(0.5)	9,080 c	89 ab
	60 fb 60	5	7.0	(1.4)	9,180 c	47 c
		13	4.0	(2.3)	9,930 bc	76 b
		18	2.3	(3.2)	9,460 bc	88 ab
		21	3.0	(2.2)	10,270 abc	85 ab
					P = 0.0366	P = 0.0008
	Nontreated		—	—	12180 a	100 a
	30 fb 30	4	4.0	(2.9)	10730 ab	76 a
RREC		11	1.0	(1.4)	8500 c	107 a
		14	0.8	(0.5)	9130 bc	111 a
		20	1.8	(2.2)	9590 bc	110 a
	60 fb 60	4	6.5	(3.4)	10370 ab	29 b
		11	3.3	(4.6)	9780 bc	106 a
		14	3.0	(2.0)	9600 bc	96 a
		20	3.3	(4.6)	9980 bc	106 a
					P = 0.0254	P < 0.0001

^aAbbreviations: fb, followed by; PTRS, Pine Tree Research Center; RREC, Rice Research and Extension Center.

^bPercent groundcover for the nontreated was 82% and 88%, respectively, at PTRS and RREC.

^cMeans followed by standard error in parenthesis.

^dMeans are separated using Fisher's protected least significant difference ($\alpha = 0.05$). Means with the same letter within a factor and column are not significantly different.

^eHyphens (-) represent nontreated delay in heading as zero.

florpyrauxifen-benzyl injury. Additionally, biomass was reduced for plants treated with florpyrauxifen-benzyl for both rates when averaged over temperature, further suggesting that CLXL745 is sensitive to florpyrauxifen-benzyl.

Injury in the greenhouse experiment was highest 14 DAA when florpyrauxifen-benzyl was applied at 60 g ae ha⁻¹ to 1- and 3-leaf rice plants, (43% and 30% injury, respectively; Table 11). By 28 DAA, injury was highest for plants treated at 1-leaf and 3-leaf stages averaged over rate, with 27% and 18%, respectively, whereas injury to plants treated at the 5-leaf stage was only 5% (Table 11). There was a height reduction 28 DAA in plants treated at the 5-leaf growth stage, which can be attributed to a shift in resources due to tillering (Table 11). There was also a reduction in height for plants treated with the 60 g ae ha⁻¹ rate at the 1-leaf and 3-leaf stages. Biomass was reduced only in rice treated with florpyrauxifen-benzyl applied at 60 g ae ha⁻¹ averaged over growth stage ($P = 0.0003$; Table 11). These data suggest that larger rice plants can metabolize and detoxify florpyrauxifen-benzyl better than when plants are smaller; however, florpyrauxifen-benzyl is still injurious to CLXL745, resulting in height and biomass reduction.

These experiments lead to the conclusion that a single application of florpyrauxifen-benzyl, especially at the standard field rate of 30 g ae ha⁻¹, can be utilized on CLXL745 when plants are larger than 1 leaf. However, florpyrauxifen-benzyl applied sequentially to CLXL745 can reduce grain yields.

Discussion

It is important to note that most biomass collected from plants treated with florpyrauxifen-benzyl was reduced from that of nontreated plants, which is seemingly in contrast to data from the field experiment. Plants had several months to recover from

florpyrauxifen-benzyl injury, whereas plants in the growth chamber had only 1 mo to recover from the herbicide application before biomass was collected. Additionally, the herbicide may have been metabolized more slowly in these the growth chamber and greenhouse experiments because of the low light. Research conducted using 2,4-D, another auxinic herbicide, showed that by increasing the light intensity, translocation of the herbicide increased (Schultz and Burnside 1980). Further research is needed to evaluate the effect of light quantity and quality on the propensity of florpyrauxifen-benzyl to injure rice. This could be significant because several cloudy days immediately followed an application of florpyrauxifen-benzyl to rice, which could have reduced plant vigor, resulting in lower yields and greater weed pressure due to reduced groundcover (Norsworthy 2004).

Because a symptom of florpyrauxifen-benzyl injury is leaf malformation, this may be a contributing factor to the reduction in biomass we observed. Additionally, stalk strength is an important factor in rice lodging resistance (Kashiwagi et al. 2008; Zuber et al. 2001), and damaged stems could cause rice to lodge. Further research is necessary to explore whether there is an increased risk of lodging when florpyrauxifen-benzyl is applied to CLXL745, since this cultivar was most injured by the herbicide.

These experiments explored various factors that could affect injury and rice yield; however, the herbicide label indicates that florpyrauxifen-benzyl may be applied at 30 g ae ha⁻¹ per application, with two applications allowed per season to 2-leaf or larger rice, and a minimum of 14 d between applications (Anonymous 2017). The label also warns that medium-grain varieties and long-grain hybrid cultivars are more sensitive to florpyrauxifen-benzyl, and the findings in these experiments for the medium-grain pureline cultivar CL272 and long-grain hybrid cultivar CLXL745 support the label recommendations. This was also

Table 11. Injury and height for the long-grain rice hybrid cultivar CLXL745 in the growth chamber experiment as influenced by day/night temperature regime and florpyrauxifen-benzyl rate and florpyrauxifen-benzyl rate and by growth stage and florpyrauxifen-benzyl rate in the greenhouse experiment.^a

		Injury ^{b,c}		Height ^{b,d}		
	Factor	14 DAA	28 DAA	14 DAA	28 DAA	Biomass ^{b,d}
Growth chamber		% of nontreated				
Temperature ^e	27/18			96 b	96 b	
	32/24			101 a	102 a	
	Nontreated	–				100 a
	30	6 b				87 b
Rate ^e	60	17 a				81 b
	27/18 nontreated	–		100 ab		
	27/18 × 30	9 b	8 ab	93 c		
	27/18 × 60	9 b	8 ab	96 bc		
Temperature × rate	32/24 nontreated	–		100 ab		
	32/24 × 30	4 b	6 b	102 a		
	32/24 × 60	25 a	10 a	104 a		
	Greenhouse		% of nontreated			
Stage	1-leaf		27 a	85 ab	100 a	
	3-leaf		18 a	78 b	100 a	
	5-leaf		5 b	93 a	79 b	
Rate	Nontreated	–		100 a	100 a	100 a
	30		10 b	82 b	92 ab	92 a
	60		20 a	75 b	85 b	76 b
Stage × rate	1-leaf nontreated	–			100 a	
	1-leaf × 30	13 cd			108 a	
	1-leaf × 60	43 a			92 a	
	3-leaf nontreated	–			100 a	
	3-leaf × 30	18 c			102 a	
	3-leaf × 60	30 a			97 a	
	5-leaf nontreated	–			100 a	
	5-leaf × 30	11 cd			71 b	
	5-leaf × 60	10 d			70 b	

^aAbbreviation: DAA, d after application.^bMeans are separated using Fisher's protected least significant difference ($\alpha = 0.05$). Means with the same letter within a factor and column are not significantly different. Data not shown for some treatments and variables indicate data is not significant.^cHyphens (–) represent nontreated data as 0% injury.^dHeight 28 DAA for the nontreated of the 27/18 C growth chamber was 49 cm and 42 cm for the nontreated of the 32/24 C growth chamber. Height 14 DAA was 45, 54, and 64 cm and 28 DAA was 56, 61, and 78 cm for the nontreated s of 1-, 3-, and 5-leaf growth stages. Biomass for the nontreated s at the 1-, 3-, and 5-leaf growth stages was 0.70 g, 1.50 g, and 2.10 g at 28 DAA, respectively in the greenhouse experiment.^eTemperature is reported in Celsius (C); florpyrauxifen-benzyl rate is reported in g ae ha⁻¹.

observed by Pantone and Baker (1992) with injury from triclopyr at 800 g ha⁻¹ applied to different cultivars. Montgomery et al. (2014) reported that hybrid cultivar CLXL745 and medium-grain varieties 'Caffey' and 'CL261' were more sensitive to saflufenacil and carfentrazone than two long-grain cultivars.

Rice growers should expect some level of injury when florpyrauxifen-benzyl is applied sequentially to CL272 and CLXL745. Based on the findings in these experiments for CLXL745, sequential applications of florpyrauxifen-benzyl are not recommended. Because CLXL745 is particularly sensitive to florpyrauxifen-benzyl, further research should be conducted on other hybrid cultivars.

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