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#### Nomenclature:

acetochlor; anilofos; butachlor; clomazone; oxadiazon; pendimethalin; pretilachlor; pyraclonil; thiobencarb; barnyardgrass; *Echinochloa crus-galli* (L.) Beauv.; rice; *Oryza sativa* L.

#### Keywords:

ALS-resistant; control efficacy; PRE herbicide; rice varieties; soil-applied herbicides

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# Rice safety and control of penoxsulam-resistant and -susceptible barnyardgrass (*Echinochloa crus-galli*) populations with soil-applied herbicides

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## Abstract

Resistance to penoxsulam among barnyardgrass populations is prevalent in rice fields in China. Seeds of penoxsulam-resistant (AXXZ-2) and penoxsulam-susceptible (JLGY-3) barnyardgrass populations, as well as the seeds of two rice varieties, including Wuyungeng32 (WY) and Liangyou669 (LY), were planted in plastic pots and then treated with a rate titration of acetochlor, anilofos, butachlor, clomazone, oxadiazon, pendimethalin, pretilachlor, pyraclonil, or thiobencarb. The two barnyardgrass populations exhibited similar susceptibility to acetochlor, anilofos, butachlor, oxadiazon, pretilachlor, or pyraclonil. However, the susceptibility differed between the barnyardgrass populations in response to clomazone, pendimethalin, and thiobencarb. For AXXZ-2, herbicide rates that caused 50% reduction in shoot biomass from the nontreated control (GR<sub>50</sub>) were 179, >800, and 1,798 g ha<sup>-1</sup> for clomazone, pendimethalin, and thiobencarb, respectively; whereas JLGY-3 GR<sub>50</sub> values were 61, 166, and 552 g ha<sup>-1</sup>, respectively. Both rice varieties demonstrated excellent tolerance to acetochlor, butachlor, oxadiazon, pretilachlor, and thiobencarb. However, substantial rice damage was observed when anilofos and clomazone were used. Anilofos at 352 g ha<sup>-1</sup> and clomazone at 448 g ha<sup>-1</sup> reduced rice shoot biomass by 41% and 50% from the nontreated, respectively. Averaged across herbicide rates, clomazone use resulted in a reduction in rice shoot biomass from that of the nontreated control by 52% and 34% for WY and LY, respectively; and pendimethalin use resulted in a reduction in rice shoot biomass from the nontreated control by 25% and 9% for WY and LY, respectively.

#### Introduction

Barnyardgrass, an allohexaploid grass, has always been considered the most troublesome weed species in rice production in China (Fang et al. 2018, 2019a, 2019b). Season-long competition of barnyardgrass can cause as much as 80% rice yield reduction (Ntanos and Koutroubas 2000; Smith 1988). A single barnyardgrass plant growing 40 cm from a rice plant can reduce rice yield by 27% (Stauber et al. 1991). The competitive ability of barnyardgrass is largely due to its extended emergence throughout the growing season, high photosynthetic capacity, rapid growth habit, and prolific seed production (Chon et al. 1994; Kennedy et al. 2010; Zhang et al. 2015). Therefore, controlling barnyardgrass is believed to be essential for optimum rice production.

Five structurally distinct chemical classes of acetolactate synthase (ALS) inhibitors, including imidazolinones, triazolopyrimidines, pyrimidinylthiobenzoates, and sulfonylamino-carbonyl-triazolinones, have been commercialized (Shaner 2014). These herbicides inhibit the first enzyme in the biosynthesis pathway of branched-chained amino acids including isoleucine, leucine, and valine (LaRossa and Schloss 1984). ALS-inhibiting herbicides are well known for their high efficacy, low use rate, potency, and multicrop selectivity (Shaner 2014). In addition, various formulations have proved to be active on both foliage and in soil, with very low mammalian toxicity (Shaner 2014). These positive attributes have ensured the intensive use of ALS inhibitors over huge farmland areas worldwide for weed control (Powles and Yu 2010). ALS-inhibiting herbicides such as bispyribac, imazethapyr, and penoxsulam, are used for POST control of barnyardgrass in rice (Martini et al. 2015; Ottis et al. 2003; Pellerin and Webster 2004; Rouse et al. 2017). Unfortunately, the chance is high that barnyardgrass will become resistant to ALS

inhibitors (Powles and Yu 2010). During the last decade, resistance to ALS-inhibiting herbicides has increased drastically in various cropping systems (Fang et al. 2019a, 2019b; Heap 2020; McCullough et al. 2016a, 2016b; Powles and Yu 2010; Yu et al. 2020a). To date, a total of 165 weed species worldwide have been found to be resistant to at least one of the five ALS inhibitor chemical families (Heap 2020).

Penoxsulam is one of the commonly used ALS-inhibiting herbicides for POST control of barnyardgrass in rice fields in China (Anonymous 2020). At recommended use rates, penoxsulam demonstrated excellent safety on both *indica* and *japonica* rice varieties (Bond et al. 2007) and provided broad-spectrum control of various weed species including barnyardgrass, various broadleaves, and sedges (*Cyperus* spp.; Ottis et al. 2003; Khare et al. 2014; Willingham et al. 2008). Unfortunately, the extensive use of this herbicide in rice has caused resistant barnyardgrass populations in China (Chen et al. 2016; Fang et al. 2019a), South Korea (Won et al. 2014), the United States (Norsworthy et al. 2014; Wilson et al. 2014), and Vietnam (Duy et al. 2018).

Herbicide resistance mechanisms can be classified to target-site resistance (TSR) and nontarget-site resistance (NTSR). Recent investigations revealed that TSR and NTSR can concurrently confer resistance to ALS inhibitors in barnyardgrass (Fang et al. 2019a). Fang et al. (2019a) reported that amino acid substitution at Trp-574-Arg in the ALS was the basis of TSR mechanisms conferring penoxsulam resistance in a barnyardgrass population, while metabolism mediation by cytochrome P450 monoxygenases (P450s) and glutathione S-transferases (GSTs) also contributed to penoxsulam resistance. This population of barnyardgrass displayed cross-resistance to POST applications of other ALS-inhibiting herbicides including flucarbazone-sodium, pyroxsulam, pyribenzoxim, flucetosulfuron, and propyrisulfuron, and exhibited multiple resistance to three ACCase-inhibiting herbicides including cyhalofop-butyl, metamifop, and pinoxaden, as well as two synthetic auxin herbicides including florpyrauxifen-benzyl and quinclorac.

In a POST control scenario, applying a premix or tank-mix of herbicides with two effective modes of action are recommended to mitigate the evolution and spread of resistant weeds (Norsworthy et al. 2012; Powles and Yu 2010; Vencill et al. 2012; Yu et al. 2017). Although rice growers in China adopted such recommendation and applied a mixture of penoxsulam and cyhalofop-butyl or quinclorac for POST control of barnyardgrass (Anonymous 2020), multiple resistance to POST herbicides in barnyardgrass populations has increased drastically in recent years (Chen et al. 2016; Dong et al. 2018; Fang et al. 2019a, 2019b).

A barnyardgrass population in Anhui province, China, has evolved resistance to all five chemical families of ALS-inhibiting herbicides because of a target site mutation (Fang et al. 2019b). This population of barnyardgrass has shown resistance to multiple POST herbicides, including flucarbazone-sodium, imazapic, pyroxsulam, and propyrisulfuron, which had never been previously used for weed control in rice fields in China (Fang et al. 2019b). At present, rice growers have limited choices for POST control of this barnyardgrass population. Therefore, soil-applied PRE herbicides are considered for resistance management. Nevertheless, limited information exists on rice safe and efficacy using soil residual herbicides for controlling penoxsulam-resistant barnyardgrass. Thus, the objectives of this research were to evaluate rice safety and control efficacy of soil-applied herbicides for controlling penoxsulam-resistant and -susceptible barnyardgrass populations.

#### **Materials and Methods**

#### Plant Materials

The barnyardgrass seeds used were a known penoxsulam-resistant (AXXZ-2) population collected in 2012 in Anhui, China (110.98°E, 30.93°N), whereas the seeds of a penoxsulam-susceptible (JLGY-3) population were collected in 2012 from a recreational field in Jiangsu, China (119.12°E, 34.83°N), which has no history of having had herbicides applied. In previous research, Fang et al. (2019b) confirmed that the resistant AXXZ-2 population exhibited 33-fold resistance to penoxsulam compared with the susceptible JLGY-3 population. In this paper, the penoxsulam-resistant and penoxsulam-susceptible barnyardgrass populations were renamed to be consistent with the terminology used in the previous publication (Fang et al. 2019b). Rice tolerance to soil-applied herbicides was evaluated with Wuyungeng32 (WY, *Oryza sativa* L. ssp. *japonica*) and Liangyou669 (LY, *Oryza sativa* L. ssp. *indica*) varieties of rice.

Barnyardgrass Control Experiments

Greenhouse experiments were established in Nanjing Agricultural University, Nanjing, China (118.46°E, 32.03N), in 2019, to evaluate the efficacy of soil-applied herbicides for controlling AXXZ-2 and JLGY-3 barnyardgrass populations. Pots with a 49-cm<sup>2</sup> surface areas and 7-cm depth were filled a mixture of sand and potting soil (2:1 by weight). Twenty seeds from each barnyardgrass population were planted in each plastic pot and then covered with a thickness of 0.3 to 0.5 cm of soil mixture. The soil mixture contained 1.4% organic matter, pH 6.1, that had no history of herbicide use or barnyardgrass infestation.

The pots were placed in a greenhouse set for 27/23 C (day/night temperatures) with a 12-h photoperiod, 9,000 lux light intensity, and 85% relative humidity. The pots were treated with a rate titration of acetochlor, anilofos, butachlor, clomazone, oxadiazon, pendimethalin, pretilachlor, pyraclonil, and thiobencarb (Table 1). A nontreated check was included in each replication. Herbicide treatments were sprayed in a sprayer chamber (machine model 3WP-2000, Nanjing Research Institute for Agricultural Mechanization, Nanjing, National Ministry of Agriculture of China) calibrated to deliver 280 L ha<sup>-1</sup> with an even flat-fan nozzle (Boyuan<sup>®</sup> Nozzle Technologies Inc., Guangdong, China). The pots were returned to the greenhouse at 1 h after herbicide treatment and then received 0.6 cm of irrigation. The pots were overhead-watered as needed to prevent soil moisture deficiencies. Herbicide rate, product, and manufacturer information are shown in Table 1. At 30 d after treatment (DAT), shoots were harvested, and fresh biomass was weighed.

## **Rice Study**

Greenhouse experiments were conducted in Nanjing Agricultural University in 2019 to evaluate rice safety following the application of PRE herbicides. Twenty rice seeds of WY (ssp. *japonica*) or LY (ssp. *indica*) were planted with previously described plastic pot and soil. Seeds were covered with a thickness of 0.3 to 0.5 cm soil. At 24 h after planting, pots were sprayed with a rate titration of acetochlor, anilofos, butachlor, clomazone, oxadiazon, pendimethalin, pretilachlor, pyraclonil, and thiobencarb using the aforementioned spray chamber. A nontreated check was included in each replication. Herbicide rate, product, and manufacturer information are identical to the barnyardgrass control experiments (Table 1). At 1 h after treatment, pots were returned to the greenhouse and received 0.6 cm of irrigation. Temperature, light intensity, and relative humidity of the greenhouse were described in the

Table 1. Herbicide rate, product, and manufacturer information.

WSSA group number <sup>a</sup>	Common name	Labeled rate	Rates applied to each population	Manufacturer	Manufacturer's address
-			g ai ha <sup>-1</sup>		
15	Acetochlor 900 g $L^{-1}$ EC	2,025	0, 150, 300, 600, 1,200, 2,400	Jiangsu Lvlilai Co, Ltd.	Yancheng, Jiangsu, China
15	Anilofos 30% EC	350	0, 44, 88, 176, 352, 704	Liaoning Province Dalian Songliao Chemical Co, Ltd.	Dalian, Liaoning, China
15	Butachlor 60% EC	800	0, 100, 200, 400, 800, 1,600	Jiangsu Lvlilai Co, Ltd.	Yancheng, Jiangsu, China
13	Clomazone 480 g $L^{-1}$ EC	1,200	0, 56, 112, 224, 448, 896	Zhejiang Zhongshan Chemical Group Co, Ltd.	Huzhou, Zheijang, China
14	Oxadiazon 250 g $L^{-1}$ EC	460	0, 58, 116, 232, 464, 928	Jiangsu Longdeng Chemical Co, Ltd.	Suzhou, Zhejiang, China
3	Pendimethalin 30% SC	720	0, 50, 100, 200, 400,800	Jiangsu Futian Agrochemical Co, Ltd.	Nanjing, Jiangsu, China
15	Pretilachlor 30% EC	675	0, 42, 84, 168, 336, 672	Jiangsu Futian Agrochemical Co, Ltd.	Nanjing, Jiangsu, China
14	Pyraclonil 2% GR	200	0, 25, 50, 100, 200, 400	Hubei Xianghe Precision Chemical Co, Ltd.	Qianjiang, Hubei, China
8	Thiobencarb 90% EC	1,600	0, 200, 400, 800, 1,600, 3,200	Beijing Birongda Biochemical Technology Development Co, Ltd.	Beijing, China

<sup>a</sup>Weed Science Society of America group numbers listed represent the following herbicide mechanisms of action: (15) inhibitors of synthesis of very long-chain fatty acids; (13) inhibitor of 1deoxy-D-xyulose 5-phosphate synthetase; (14) inhibitor of protoporphyrinogen oxidase (Protox, PPO); (3) inhibitor of microtubule assembly; (8) inhibitor of lipid synthesis. <sup>b</sup>Abbreviations: EC, emulsifiable concentrates; GR, granules.

barnyardgrass control study. The pots were overhead-watered as needed to prevent soil moisture stress. At 30 DAT, rice shoots were harvested, and fresh shoot biomass was measured.

# Experimental Design and Statistical Analysis

Each herbicide was considered to be a separate trial. The experiment designed as a randomized complete block and was a twoway factorial with six herbicide rates by two barnyardgrass populations or two rice varieties with two experimental runs and four replications in each run. All experiments were conducted twice over time.

For barnyardgrass control experiments, data were subjected to nonlinear regression analysis using SigmaPlot (version 12, Systat Software, San Jose, CA). Treatment means were plotted on figures and regressed against the following three-parameter exponentialgrowth function model:

$$y = \beta_0 + \beta_1 * \exp(-\beta_2 * x)$$

where y is percent barnyardgrass shoot biomass reduction from the nontreated control, x is the herbicide rate (g ai ha<sup>-1</sup>),  $\beta_0$  is the lower asymptote,  $\beta_1$  is the upper asymptote, and  $\beta_2$  is the slope. This equation was used because it described the relationship between the percentage of shoot mass reduction and herbicide rate. Herbicide rate that caused 50% reduction in fresh shoot biomass from the nontreated control (GR<sub>50</sub>) was calculated based on the regression curves. The 95% confidence limits for GR<sub>50</sub> values were calculated.

For rice safety study, data were subjected to ANOVA using the MIXED procedure in SAS (version 9.3, SAS Institute, Cary, NC). Rice variety and herbicide rate were considered as the fixed factors, whereas replications and experimental runs were considered as the random factors. Treatment means were separated with Tukey adjustment means comparison at  $P \le 0.05$ .

## **Results and Discussion**

The percentage of shoot biomass reduction, with standard errors of the means, is presented in Figure 1. Control of AXXZ-2 and JLGY-3 barnyardgrass populations increased with increased herbicide rate for all herbicides (Figure 1). According to 95% confidence limits, the two barnyardgrass populations were equally susceptible to acetochlor, anilofos, butachlor, oxadiazon, pretilachlor, or pyraclonil, with statistically equivalent GR<sub>50</sub> or GR<sub>90</sub> values (Table 2). However, JLGY-3 exhibited significantly lower GR<sub>50</sub> or GR<sub>90</sub> values and was more susceptible to clomazone, pendimethalin, and thiobencarb than AXXZ-2. JLGY-3 GR<sub>50</sub> values were 61, 166, and 552 g ha<sup>-1</sup> for clomazone, pendimethalin, and thiobencarb, whereas GR<sub>50</sub> values for AXXZ-2 were 179, >800, and 1,798 g  $ha^{-1}\!,$  respectively. GR\_{90} values for JLGY-3 were 96, >800, and 1,988 g ha<sup>-1</sup> for clomazone, pendimethalin, and thiobencarb; whereas GR<sub>90</sub> values for AXXZ-2 were 624, >800, and >3,200 g ha<sup>-1</sup>, respectively.

Of the herbicides included in this study, the labeled rates of acetochlor, clomazone, oxadiazon, pretilachlor, and pyraclonil were effective at controlling both barnyardgrass populations and resulted in reduced shoot biomass from the nontreated control by at least 90% (Table 2; Figure 1). However, at the labeled use rates, neither pendimethalin nor thiobencarb effectively controlled AXXZ-2. The highest rate of pendimethalin at 800 g ha<sup>-1</sup> reduced AXXZ-2 and JLGY-3 shoot biomass by 10% and 60% compared with the nontreated, respectively, whereas thiobencarb (1,600 g ha<sup>-1</sup>) reduced AXXZ-2 and JLGY-3 shoot biomass by 65% and 89%, respectively. These findings suggest that AXXZ-2 has reduced susceptibility to pendimethalin and thiobencarb.

The effectiveness of evaluated herbicides on barnyardgrass has been well documented (Akkari et al. 1986; Balyan et al. 1996; Juraimi et al. 2013; Kirkwood and Fletcher 1984; Ryang 1998; Shaner 2014; Ushiguchi et al. 2014; Wilson et al. 2014). Soilapplied PRE herbicides need to be applied prior to weed emergence to maximize weed control (Shaner 2014). However, environmental factors can influence weed seed germination and seedling emergence (Sharpe and Boyd 2019; Yu et al. 2020b). As a summer

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**Figure 1.** Penoxsulam resistant- and penoxsulam-susceptible barnyardgrass shoot biomass reductions compared with the nontreated control at 30 d after herbicide applications in two combined greenhouse experiments, 2019, in Nanjing, Jiangsu, China. Vertical bars represent standard errors of the mean (n = 8). Results were pooled over experimental runs. AXXZ-2 represents the penoxsulam-resistant barnyardgrass population, JLGY-3 represents the penoxsulam-susceptible barnyardgrass population.

annual weed species, barnyardgrass may have several peaks of seed germination during the rice production season (Kennedy et al. 2010). Because of the herbicide degradation and dissipation, a

single application of residual herbicides may not be able to provide season-long control of barnyardgrass (Shaner 2014). Field studies conducted in Arkansas revealed that sequential herbicide



Figure 1. Continued

application programs with clomazone, thiobencarb, pendimethalin, and quinclorac applied as a delayed PRE in different combinations followed by imazethapyr early POST followed by preflood controlled ALS-resistant barnyardgrass above  $\geq$ 96% in rice fields (Wilson et al. 2014).

The widespread resistance of barnyardgrass to POST herbicides is a concern to rice producers in China and other countries as well (Chen et al. 2016; Fang et al. 2019a, 2019b; Heap 2020; Rouse et al. 2017). Barnyardgrass with resistance to ALS-inhibiting herbicides has been frequently reported in China in recent years (Fang et al. 2019a, 2019b; Liu et al. 2015; Wu et al. 2012) and may continuously cause severe problems for rice production due to competition with crops in seasons and weed seed bank management for the following production seasons. Therefore, it is important to determine the effectiveness of soil-applied herbicides for controlling ALS-resistant barnyardgrass. In the present research, except for pendimethalin and thiobencarb, the labeled rates of the herbicides we evaluated effectively controlled both barnyardgrass populations. Those herbicides differ in their mechanisms of action with ALS inhibitors and can control penoxsulam-resistant barnyardgrass.

In addition to tank-mixture or sequential application of herbicides with different mechanisms of action, other integrated weed management strategies such as crop rotation, selection of competitive rice variety, high seedling rate; and the use of clean tillage equipment, narrow row spacing, proper weed identification, and scouting of fields, can influence the degree of herbicide resistance management (Norsworthy et al. 2012; Vencill et al. 2012). The integration of these weed management practices in rice production may significantly slow down the herbicide resistance evolution in barnyardgrass populations (Norsworthy et al. 2012; Vencill et al. 2012).

Except for pyraclonil, the effect of herbicide rate by rice variety interaction on rice shoot biomass was significant for all herbicides (Table 3). The effect of herbicide rate was significant for acetochlor, anilofos, clomazone, pendimethalin, and pretilachlor, while the effect of herbicide rate was not significant for butachlor, oxadiazon, and thiobencarb. For both rice varieties, acetochlor, butachlor, oxadiazon, pretilachlor, and thiobencarb at the rates tested caused  $\leq 20\%$  shoot biomass reduction from the nontreated control. Anilofos at 44 and 88 g ha<sup>-1</sup>, clomazone at 56 g ha<sup>-1</sup>, and pendimethalin at 50 and 100 g ha<sup>-1</sup> caused < 20% reduction in rice shoot biomass; however, these herbicides applied at higher rates caused substantial reduction in shoot biomass. At the highest rate, anilofos at 704 g ha<sup>-1</sup>, clomazone at 896 g ha<sup>-1</sup>, and pendimethalin at 800 g ha<sup>-1</sup> caused 57\%, 75\%, and 18\% reduction in rice shoot biomass, respectively.

The two rice varieties, *japonica* and *indica*, differed significantly in their response to clomazone or pendimethalin (Table 3).

# $\label{eq:table 2. Values of parameters for regression equation data presented in figures.^{a,c}$

					P-	Standard error of		95% CI for		95% CI for
Herbicide	Population <sup>b</sup>	Parameter	Estimate (±SE)	R <sup>2</sup>	value	estimate	GR <sub>50</sub>	GR <sub>50</sub>	GR <sub>90</sub>	GR <sub>90</sub>
						g ai ha <sup>-1</sup>				
Acetochlor	AXXZ-2	$\beta_1$	99.2073	0.9372	0.0158	13.30	396	242-638	1.557	659-2,216
		62	(±14.7352)							
		μz	(±0.0006)							
	JLGY-3	$\beta_1$	101.6614	0.9307	0.0182	14.28	359	206-565	1.272	542-1,814
		62	(±14.7243)							
		μz	(±0.0007)							
Anilofos	AXXZ-2	$\beta_1$	126.477	0.9014	0.0310	17.84	147	100-247	427	301-728
		02	(±48.4176)							
		pz	(+0.0018)							
	JLGY-3	$\beta_1$	98.4501	0.9922	0.0007	4.20	108	95-203	313	217-530
		0.5	(±4.3169)							
		β2	0.006							
Butachlor	AXXZ-2	β1	91.5126	0.9771	0.0035	6.88	192	103-295	993	613-1,606
		, -	(±5.6000)							
		β2	0.0039							
	JII GY-3	ß1	(±0.0007) 107.8779	0.9783	0.0032	8.15	280	190-470	745	520-1.265
	02010	P1	(±9.365)	010100	010002	0.20	200	200		020 1,200
		β2	0.0023							
Clomazono	AVV7 2	ß	(±0.0005)	0 0000	0.0012	4 92	170	120 209	624	121 1 059
ciomazone	AVVT-7	P1	(±17.78)	0.9890	0.0012	4.52	115	129-308	024	434-1,038
		β2	0.0042							
		0	(±0.0006)							=0.400
	JLGY-3	$\beta_1$	102.7482 (+7.0364)	0.9593	0.0082	10.98	61	41-102	96	72-168
		β2	0.0139							
			(±0.0037)							
Oxadiazon	AXXZ-2	$\beta_1$	97.8884	0.9073	0.0282	14.62	96	50-146	364	134-498
		62	(±11.6908) 0.0071							
		P2	(±0.0026)							
	JLGY-3	$\beta_1$	96.0027	0.9734	0.0043	7.91	34	24-58	266	177-343
		82	(±4.7674)							
		μz	(±0.0039)							
Pendimethalin	AXXZ-2	$\beta_1$	10.3858	0.9759	0.0037	0.82	>800	NA	>800	NA
		02	(±0.9453)							
		βZ	0.0073							
	JLGY-3	$\beta_1$	64.2369	0.8877	0.0377	12.40	166	111-277	>800	NA
		·	(±12.9535)							
		β2	0.0051							
Pretilachlor	AXXZ-2	β1	(±0.0027) 96.1774	0.9828	0.023	6.07	126	87-213	500	305-805
		11	(±6.5721)							
		β2	0.0058							
	II GV-3	в.	(±0.0010)	0 9720	0.0047	8 59	122	81-204	373	226-599
	JE01-5	Ρ1	(±10.3855)	0.5120	0.0047	0.55	125	01-204	515	220-333
		β2	0.0051							
Duradanil		0	(±0.0012)	0.0204	0.0140	12.05	50	20.00	175	00.255
Pyracionii	AXXZ-Z	p1	(+13,2083)	0.9394	0.0149	13.05	28	30-88	175	80-255
		β2	0.114							
		~	(±0.0039)							
	JLGY-3	$\beta_1$	109.6917	0.9720	0.0047	9.38	76	47-123	180	124-304
		β2	0.0083							
		r	(±0.0021)							
Thiobencarb	AXXZ-2	$\beta_1$	130.2123	0.9683	0.0057	6.18	1,798	1,429-3,227	>3,200	NA
		ß2	(±9.1559) 0.0002							
		P <del>~</del>	(±0.0002)							
		$\beta_1$								
										(Continued)

#### Table 2. (Continued)

Herbicide	Population <sup>b</sup>	Parameter	Estimate (±SE)	R <sup>2</sup>	P- value	Standard error of estimate	GR <sub>50</sub>	95% CI for GR <sub>50</sub>	GR <sub>90</sub>	95% CI for GR <sub>90</sub>
	JLGY-3	β2	101.2774 (±11.4253) 0.001 (±0.0003)	0.9657	0.0064	9.76	552	384-936	1,988	1,100-3,088

<sup>a</sup>Shoot mass reductions were regressed against a three-parameter exponential-growth function model:  $y = \beta_0 + \beta_1 \exp(-\beta_2 \star x)$ , where y is percent barnyardgrass shoot biomass reduction from the nontreated control, x is the herbicide rate (g ai ha<sup>-1</sup>),  $\beta_0$  is the lower asymptote,  $\beta_1$  is the upper asymptote, and  $\beta_2$  is the slope.

<sup>b</sup>AXXZ-2 is the resistant barnyardgrass population, JLGY-3 is the susceptible barnyardgrass population.

<sup>c</sup>Abbreviations: CI, confidence interval; GR<sub>50</sub>, effective herbicide rate that causes 50% reduction in fresh shoot biomass from the nontreated control; GR<sub>90</sub>, effective herbicide rate that causes 90% reduction in fresh shoot biomass from the nontreated control; NA, not available; SE, standard error.

#### Table 3. Effect of soil-applied herbicides on rice shoot biomass.<sup>a</sup>

Acetochlor		Anilo	ofos	Butac	hlor	Clomazone	
Rate	Shoot biomass reduction	Rate	Shoot biomass reduction	Rate	Shoot biomass reduction	Rate	Shoot biomass reduction
g ai ha <sup>-1</sup>	%	g ai ha <sup>1</sup>	%	g ai ha <sup>-1</sup>	%	g ai ha <sup>-1</sup>	%
150	2c	44	2c	100	2	56	18c
300	6c	88	13bc	200	4	112	35bc
600	8bc	176	32a-c	400	6	224	40bc
1,200	13b	352	41ab	800	8	448	50b
2,400	20a	704	57a	1600	12	896	75a
Rice variety <sup>b</sup>		Rice variety		Rice variety		Rice variety	
WY	10	WY	20	WY	6	WY	52a
LY	10	LY	39	LY	8	LY	34b
Herbicide rate	***	Herbicide rate	***	Herbicide rate	NS	Herbicide rate	***
Rice variety	NS	Rice variety	NS	Rice variety	NS	Rice variety	**
Herbicide rate ×	NS	Herbicide rate $\times$	NS	Herbicide rate $\times$	NS	Herbicide rate $\times$	NS
rice variety		rice variety		rice variety		rice variety	
Oxadiazon		Pendimethalin		Pretilachlor		Thiobencarb	
Rate	Shoot biomass reduction	Rate	Shoot biomass reduction	Rate	Shoot biomass reduction	Rate	Shoot biomass reduction
g ai ha <sup>-1</sup>	%	g ai ha <sup>-1</sup>	%	g ai ha <sup>-1</sup>	%	g ai ha <sup>-1</sup>	%
58	3	50	12ab	42	1b	200	2
116	3	100	10b	84	1b	400	4
232	5	200	21ab	168	3ab	800	5
464	6	400	23a	336	4ab	1,600	5
928	9	800	18ab	672	5a	3,200	6
Rice variety		Rice variety		Rice variety		Rice variety	
WY	4	WY	25a	WY	2	WY	5
LY	7	LY	9b	LY	3	LY	4
Herbicide rate	NS	Herbicide rate	**	Herbicide rate	**	Herbicide rate	NS
Rice variety	NS	Rice variety	***	Rice variety	NS	Rice variety	NS
Herbicide rate $\times$ rice variety	NS	Herbicide rate × rice variety	NS	Herbicide rate × rice variety	NS	Herbicide rate × rice variety	NS

<sup>a</sup>Means within columns followed by the same letter are not significantly different according to Tukey adjusted mean comparisons at the 0.05 probability level.

<sup>b</sup>WY represents the rice variety of Wuyungeng 32 (ssp. *japonica*), LY represents the rice variety of Liangyou 669 (ssp. *indica*).

\*\*Significant at the 0.01 probability level.

\*\*\*Significant at the 0.001 probability level.NS, not significant at the 0.05 probability level.

Clomazone and pendimethalin caused significantly greater shoot biomass reduction on WY than LY. Averaged across herbicide rates, clomazone caused 52% and 34% shoot biomass reduction for WY and LY, respectively; and pendimethalin caused 25% and 9% shoot biomass reduction for WY and LY, respectively. The two rice varieties exhibited similar susceptibility to acetochlor, anilofos, butachlor, oxadiazon, pretilachlor, or thiobencarb.

The effects of herbicide rate and herbicide by rice variety interaction on rice shoot biomass were significant for pyraclonil, and as a result, data are presented across all treatment combinations (Table 4). At the rates tested, pyraclonil at rates ranging from 25 to 200 g ha<sup>-1</sup> were safe to LY and caused  $\leq 8\%$  shoot biomass reduction; however, pyraclonil at 400 g ha<sup>-1</sup> caused 32% shoot biomass reduction. At the rates tested, pyraclonil caused  $\leq$ 3% shoot biomass reduction for WY.

Rice tolerance to acetochlor, butachlor, oxadiazon, pretilachlor, and thiobencarb is consistent with previous reports (Fogleman et al. 2018; Kirkwood and Fletcher 1984; Wilson et al. 2014). In the present study, however, substantial rice damage on both rice varieties was observed with anilofos and clomazone. Pyraclonil ( $400 \text{ g ha}^{-1}$ ) also caused significant damage to LY. Soil-applied herbicides can offer residual weed control, but the length of control and crop safety are often dependent upon multiple factors, such as herbicide application methods, rates, soil characteristics, and environmental conditions, particularly soil moisture (Racke et al. 1999; Shaner 2014; Taylor-Lovell et al. 2002; Wilson et al.

Table 4. Effect of pyraclonil on rice shoot biomass.<sup>a</sup>

	Pyraclonil					
Rice variety <sup>b</sup>	Pyraclonil rate	Shoot biomass reduction				
	g ai ha <sup>-1</sup>	%				
WY	25	1				
	50	2				
	100	2				
	200	3				
	400	3				
LY	25	1b				
	50	1b				
	100	2b				
	200	8b				
	400	32a				
	Herbicide rate	**				
	Rice variety	NS				
	Herbicide rate $\times$ rice variety	**				

<sup>a</sup>Means within columns followed by the same letter are not significantly different according to Tukey adjusted mean comparisons at the 0.05 probability level.

<sup>b</sup>WY represents the rice variety of Wuyungeng 32 (ssp. *japonica*), LY represents the rice variety of Liangyou 669 (ssp. *indica*).

\*\*Significant at the 0.01 probability level.NS, not significant at the 0.05 probability level.

2014). In the present study, rice damage caused by clomazone may be related to the high content of sand in the soil. Although anilofos and clomazone are used for weed control in rice (Scherder et al. 2004; Zhao et al. 2009), caution needs to be taken when using them to avoid potential rice damage.

The two rice varieties in this study exhibited similar tolerance to all herbicides evaluated with the exception of clomazone and pendimethalin. In other studies, Zhang et al. (2004) found that a longgrain rice variety, 'Drew', exhibited significantly greater tolerance to clomazone compared with medium-grain rice varieties 'Earl' and 'LL-401'. Koger et al. (2006) noted differential tolerance to pendimethalin among rice varieties. The authors suggested that seeds of rice varieties that are sensitive to soil-applied herbicides should be planted more deeper, which may enable the seed to absorb soil moisture before contacting with herbicides and thus reducing potential rice injury.

In summary, penoxsulam-resistant and -susceptible barnyardgrass populations demonstrated similar susceptibility to acetochlor, anilofos, butachlor, oxadiazon, pretilachlor, or pyraclonil. However, penoxsulam-susceptible barnyardgrass exhibited greater susceptibility to clomazone, pendimethalin, and thiobencarb than penoxsulam-resistant populations. At labeled rates, acetochlor, anilofos, butachlor, clomazone, oxadiazon, pretilachlor, and pyraclonil can effectively control penoxsulam-resistant and -susceptible barnyardgrass populations; however, pendimethalin and thiobencarb are ineffective at controlling penoxsulam-resistant barnyardgrass. Except for anilofos and clomazone, the rice varieties are tolerant to the evaluated herbicides. WY is more tolerant to clomazone and pendimethalin than LY. Field research is needed to further confirm the effectiveness of these herbicides for controlling penoxsulam-resistant barnyardgrass.

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