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Evaluation of Very-Long-Chain Fatty Acid-Inhibiting Herbicides in Arkansas Rice Production

A thesis submitted in partial fulfillment of the requirements for the degree of Master of Science in Crop, Soil, and Environmental Science

by

## John A. Godwin Jr. University of Arkansas Bachelor of Science in Agriculture, Food, and Life Sciences, 2015

December 2017 University of Arkansas

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#### ABSTRACT

Because of the evolution of herbicide-resistant weeds, such as barnyardgrass and red rice in rice, there is a need for alternative herbicide sites of action. Very-long-chain fatty acid (VLCFA)-inhibiting herbicides are not labeled for use in U.S. rice production; however, this site of action (SOA) has been used with success in Asian rice. The VLCFA-inhibiting herbicides pethoxamid, pyroxasulfone, acetochlor, and S-metolachlor were evaluated for rice tolerance and control of commonly problematic weeds in Arkansas rice at various rates and application timings. Pyroxasulfone and S-metolachlor were deemed unfit for use in rice production because of negative effects on rice visual injury, rough rice yield, height, shoot density, and heading. Pethoxamid and acetochlor were used with little detriment to the rice crop when applied no earlier than the 1-lf growth stage. Along with minimal rice injury, pethoxamid controlled barnyardgrass all season when used in a program with other common rice herbicides such as clomazone, imazethapyr, or quinclorac. Considering the minimal injury observed, pethoxamid and acetochlor should be considered for integration into U.S. rice production to represent a unique herbicide SOA to use in rotation, sequential application, or tank mixtures with other rice herbicides.

Nomenclature: Acetochlor; pethoxamid; pyroxasulfone; S-metolachlor; barnyardgrass, Echinochloa crus-galli (L.) Beauv.; red rice, Oryza sativa var. sylvatica L.; rice, Oryza sativa L.

Key words: weed control, herbicide-resistant weeds, herbicide sites of action

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#### Chapter 1

#### **General Introduction and Review of Relevant Literature**

#### Weed Control in Rice

Along with nutrient management, disease control, and several others, weed control is one of the most important and most difficult to accomplish aspects of rice production. Weeds directly compete with rice and will reduce yield if not controlled (Perez de Vida et al. 2006). Weed interference has been estimated to reduce rice yield by up to 96% (Ampong-Nyarko and Datta 1991). Along with direct competition, weeds indirectly serve as hosts for pests that can subsequently infest the rice crop (Ampong-Nyarko and Datta 1991). Harm to a rice crop from weeds depends on the weed species, duration of interference, weed density, rice cultivar characteristics, and other cultural management dynamics (Smith 1968).

Weeds can be controlled using cultural, mechanical, chemical, and biological methods. Cultural control methods in rice include increasing the competitive ability of rice to suppress weeds. Rice can become more competitive by excluding weed introduction through land preparation, crop rotation, cultivar selection, time of seeding, planting method, plant population, fertilization, and water management (Ampong-Nyarko and Datta 1991). Mechanical control directly removes the weeds from the soil surface with a piece of machinery such as a disk harrow, moldboard plow, field cultivator, etc. In order to destroy as many weeds as possible through tillage, the interval between tilling events should be long enough to allow many seeds to germinate and seedlings be later killed by tillage (Ampong-Nyarko and Datta 1991). Tillage is used to start the growing season weed free and is a common practice in Arkansas rice production.

Along with tillage, water-seeded rice is an effective method for controlling weeds. Although not a common practice, water-seeded rice can suppress weed emergence due to replacement of oxygen in the soil by water at establishment of a permanent flood. Water-seeding can reduce red rice (*Oryza sativa* var. *sylvatica*) germination by 70% alone (Smith 1988). Even if the rice is not water-seeded, applying a flood to the rice as soon as it reaches the 5- to 6-leaf growth stage plays a major role in weed reduction.

An additional cultural practice that provides effective weed control is crop rotation. Weeds in North America have evolved resistance where there has been monoculture crop production and use of a single family of herbicides (Gressel and Segel 1990; Heap (2006). Crop rotation gives farmers the ability to break that monoculture and apply different herbicide sites of action (SOA), which control the same weeds in different crops. Weeds that may be difficult to control in one crop can be successfully controlled in another (Hauser et al. 1974). Growing a broadleaf crop in rotation with rice allows growers to use herbicides that are effective against difficult to control grassy weeds in rice (Ampong-Nyarko and Datta 1991).

Herbicides are the most common weed control method in U.S. weed management programs. Herbicides in rice can be applied to the soil or directly over the top of the crop. In most weed management practices, a preplant application of a non-selective herbicide is applied to remove weedy vegetation prior to planting, especially in a minimum tillage system. After planting, a preemergence (PRE) herbicide is generally applied prior to crop and weed emergence. Subsequently, herbicides can then be applied postemergence (POST) to specific troublesome weeds. Ground equipment is often used to apply herbicides prior to establishing the permanent flood in drill-seeded rice; however, after flooding, aerial applications become standard.

#### Herbicide Resistance

Herbicide resistance is arguably the top concern in weed science today. Currently, there are 471 unique cases of herbicide-resistant weeds among 250 species (Heap 2017). Of these, 152 unique cases of weed resistance have been confirmed in rice alone. Additionally, weeds have evolved resistance to 23 of the 26 known SOA and to 160 different herbicides.

Several weeds found in rice are resistant to commonly used SOA such as the acetolactate synthase (ALS) inhibitors, acetyl CoA carboxylase (ACCase) inhibitors, TIR1 auxin receptors, photosystem II (PSII) inhibitors, and diterpene biosynthesis inhibitors. These weeds include barnyardgrass (*Echinochloa crus-galli* L. Beauv.), yellow nutsedge (*Cyperus esculentus* L.), rice flatsedge (*Cyperus iria* L.), California arrowhead (*Safittaria montevidensis* Cham. & Schlecht.), red rice, and many others (Heap 2017).

Resistance is driven by the expression of single or multiple resistance genes through mutation or preexistence or by selection pressure (Rotteveel et al. 1997). Following repeated applications of the same herbicide SOA, the frequency of resistance alleles increases within weed populations (Jasieniuk et al. 1996). When a genetic shift occurs, these plants can become more adaptable by increasing their ability to compete and produce more seeds than their counterparts (UAEX 2013).

Resistance can be categorized several ways including: target site resistance vs. nontarget-site resistance and cross resistance vs. multiple resistance. Target-site resistance results from modification of the herbicide-binding site and is the most studied type of resistance (Powles and Shaner 2001). In contrast, non-target-site resistance is due to mechanisms such as enhanced metabolism, reduced rates of herbicide translocation, and sequestration. Within these categories, cross-resistance consists of a single resistance mechanism providing the plant with the ability to

survive herbicide applications from different herbicide families, whereas multiple-resistance involves more than one resistance mechanism.

Lower than labeled rates of herbicides can result in the evolution of herbicide resistance. Low herbicide rates can be the result of any scenario that reduces the lethal amount of herbicide on a weed such as applying a reduced rate or applying to weeds beyond the recommended weed size. If reduced herbicide rates result in weed survivors, then resistance evolution can occur, especially in cross-pollinated species (Manalil et al. 2011). The treatment of wild-type plants with sublethal concentrations of herbicide can induce increased defense response by the plant resulting in enhanced tolerance to a particular herbicide (Molina et al. 1999). This resistance may spread over several generations of a weed species, resulting in the inability to control that particular plant.

One of the most effective ways to avoid the onset of herbicide resistance is to use multiple herbicide SOAs that are effective against weeds most prone to evolve resistance (Norsworthy et al. 2012). Reports suggest that the most important reason for the evolution of herbicide resistance is reliance upon one particular herbicide or herbicide SOA. The use of herbicides with different SOAs simultaneously, sequentially, or annually, greatly reduces the survival and reproduction of resistant weed species (Norsworthy et al. 2012).

Combating herbicide resistance is crucial to producing high crop yields. Herbicide sequences and rotations, mixtures, application rates, site-specific application, and use of herbicide-resistant crops are all tactics used to mitigate resistance (Beckie 2006). A combination of all of these can improve herbicide efficacy, reduce resistance, decrease weed pressure, and lead to increased crop yield.

#### **Current Herbicide Options in Rice**

Several herbicide options among numerous SOAs are available for use in Arkansas rice today. However, due to the evolution of resistance to many of these herbicides, options are becoming more limited each year. Grass control, with emphasis on barnyardgrass and red rice, may be the most daunting management challenge in rice today. Herbicides that can be used to control barnyardgrass in conventional rice include thiobencarb, cyhalofop, quinclorac, bispyribac, penoxsulam, propanil, pendimethalin, and fenoxaprop (Scott et al. 2013). Imidazolinone herbicides can also be used in ALS-resistant (Clearfield<sup>™</sup> BASF Corporation, Research Triangle Park, NC) rice to control troublesome grasses such as red rice and barnyardgrass; however, resistance to ALS-inhibiting herbicides is becoming increasingly common (Burgos et al. 2008).

**Propanil.** Propanil is a PSII-inhibiting herbicide and was introduced in the early 1960's to control dicotyledonous weeds and grasses. It has been used extensively in U.S. rice production since its introduction, and due to its overuse, several weeds have evolved resistance (Hoagland et al. 2004). Outside of the realm of resistance, propanil controls weeds such as barnyardgrass, broadleaf signalgrass (*Urochloa platyphylla* Nash), fall panicum (*Panicum dichotomiflorum* Michx.), Amazon sprangletop (*Leptochloa panicoides* J. Presl), eclitpa (*Eclipta prostrata* L.), hemp sesbania (*Sesbania herbacea* P. Mill.), northern jointvetch (*Aeschynomene virginica* L.), rice flatsedge, and several others (Scott et al. 2015). The continual use of propanil led to the resistance of barnyardgrass in 1989, and by 1992 resistance had spread to 16 of the 38 rice-producing counties in Arkansas (Talbert and Burgos 2007). Today, the weeds resistant to propanil in U.S. rice include barnyardgrass, junglerice (*Echinochloa colona* L.), smallflower

umbrellasedge (*Cyperus difformis* L.), and ricefield bulrush (*Schoenoplectus mucronatus* L.) (Heap 2017).

**Quinclorac.** Quinclorac is a synthetic auxin herbicide that belongs to the carboxylic acid family. Quinclorac is used to control weeds such as barnyardgrass, broadleaf signalgrass, large crabgrass (*Digitaria sanguinalis* L.), and fall panicum (Scott et al. 2015). Quinclorac can be applied PRE, delayed PRE, or early POST (Scott et al 2013). Quinclorac was introduced in 1992 and soon became the replacement for controlling propanil-resistant barnyardgrass. Soon after, in 1999, a biotype of barnyardgrass that was resistant to both propanil and quinclorac was found in Craighead County, Arkansas (Tablert and Burgos 2007). Today, barnyardgrass resistance to quinclorac is widespread in Midsouth rice (Bagavathiannan et al. 2011).

**Cyhalofop and Fenoxaprop.** Cyhalofop and fenoxaprop are WSSA Group 1 ACCase-inhibiting herbicides, belonging to the aryloxyphenoxypropionate family, and are effective in controlling barnyardgrass, broadleaf signalgrass, Amazon sprangletop, and several other grass weeds (Scott et al. 2015). Cyhalofop and fenoxaprop control quinclorac- and propanil-resistant barnyardgrass (Scott et al. 2013); however, there are cases of cyhalofop- and fenoxaprop-resistant barnyardgrass and Amazon sprangletop in the U.S. (Heap 2017).

ALS-Inhibiting Herbicides. The imidazolinone herbicides inhibit the ALS enzyme in plants and are primarily used for weed control in Clearfield<sup>™</sup> rice. In 2002, Clearfield<sup>™</sup> cultivars became commercially available, which gave growers the option to use imazethapyr to control red rice and other resistant weeds in rice (Burgos et al. 2008).

In a study conducted by Ottis et al. (2005), rice yields were reduced by 755 kg ha<sup>-1</sup> for every red rice plant m<sup>-2</sup>. Therefore, the Clearfield<sup>™</sup> technology was rapidly and widely adopted to control red rice. In 2011, 64% of Arkansas and Mississippi rice fields were seeded to imidazolinone-resistant varieties, and of this 64%, 42% were treated with ALS-inhibiting herbicides alone (Norsworthy et al. 2013).

The introduction of Clearfield<sup>™</sup> rice led to the concern of gene flow to red rice (Rajguro et al. 2005). Gene flow from Clearfield<sup>™</sup> rice to red rice is a two-part process that involves pollination of sexually compatible species and integration of the trait into the weed (Gealy et al. 2003). Gene flow has created ALS-resistant biotypes of red rice, once again, making them impossible to control with imidazolinone herbicides.

**Clomazone.** Clomazone is a WSSA Group 13 diterpene biosynthesis inhibitor belonging to the isoxazolidinone family. Clomazone was first labeled for use in soybean in 1985 for control of grasses and small-seeded broadleaves (Webster et al. 1999), but since has become the most frequently used PRE herbicide in rice (Norsworthy et al. 2007).

Clomazone provides above average residual activity and is among the lowest-cost rice herbicides. It can be applied conveniently immediately post-plant as a true PRE application (Smith and Dilday 2003). Because of the repetitive use of clomazone in rice, the evolution of barnyardgrass resistance to clomazone has occurred (Norsworthy et al. 2008).

**VLCFA-inhibiting Herbicides.** The VLCFA-inhibiting herbicides consist of three families: chloroacetamide, oxyacetamide, and pyrazole. Several herbicides found within these families include acetochlor (chloroacetamide), metolachlor (chloroactemide), pyroxasulfone (pyrazole),

and pethoxamid (chloroacetamide). VLCFA-inhibiting herbicides (WSSA Group 15) prevent cell division primarily in developing shoots and root tips of germinating weeds (Anonymous 2015). Group 15 herbicides typically affect susceptible weeds after germination but before emergence, which results in these herbicides being used for residual weed control (LeBaron and Wilcut 2014). Currently, no VLCFA-inhibiting herbicides are registered for use in U.S. rice production; however, pretilachlor and butachlor are commonly used in Asian dry- and wet-seeded rice as PRE and POST herbicides (Chauhan 2012).

Butachlor applications alone resulted in up to 59% weed control when applied 1, 3, 5, and 7 days after broadcasting sprouted rice seed in India (Mutnal et al. 1998). In a study conducted in Sri Lanka (Chauhan et al. 2013), a herbicide program consisting of pretilachlor (Group 15) + pyribenzoxim followed by (fb) MCPA was compared to several other herbicides, including cyhalofop-butyl fb MCPA, thiobencarb + propanil fb MCPA, propanil fb MCPA, and bispyribac-sodium + metamifop fb MCPA. These herbicides were applied 8 days after sowing rice in a dry-seeded system and were evaluated for control of barnyardgrass, Chinese sprangletop (*Leptochloa chinensis* L.), and knotgrass (*Paspalim distichum* L.). The results showed that the pretilachlor-containing herbicide program was the most effective based on total weed control by reducing weed density up to 99%. Because of the lack of weed competition, rice treated with pretilachlor had the highest yield among all herbicide programs evaluated. Since VLCFA-inhibiting herbicides are effectively used in Asian rice culture, it is likely that they could be effectively used in U.S. rice and may improve control of many weeds to which herbicide resistance has evolved.

No new herbicide SOA has been commercialized in U.S. rice within the last 25 years (Duke 2011). This may be in part to the high cost associated with the research, development,

and marketing of a new herbicide, or the fact that many herbicide target sites have already been utilized (Duke 2011). Due to the success of VLCFA-inhibiting herbicides in Asian rice culture, it is believed that Group 15 herbicides can be used effectively to control barnyardgrass and red rice in rice. This assumption is partly based on the fact that metolachlor, pyroxasulfone, and acetolachlor are currently used in soybean (*Glycine max* L. Merr.), a rotational crop with rice, to control both of these weeds. VLCFA-inhibiting herbicides would not be a newly developed SOA; however, they would be new to U.S. rice production. The incorporation of a new SOA into current herbicide programs should delay the onset of herbicide resistance of red rice and barnyardgrass to more common rice herbicides and provide another option for controlling other weeds of rice.

*Pyroxasulfone*. Pyroxasulfone is a VLCFA-inhibiting herbicide in the pyrazole family. Pyroxasulfone is currently labeled in corn (*Zea mays* L.), wheat (*Triticum aestivum* L.), soybean, and cotton (*Gossypum hirsutum* L.). Pyroxasulfone is a selective herbicide used to control grasses such as ryegrass (*Loilium* spp. L.), foxtails (*Setaria* spp. (L.) P. Beauv.), and barnyardgrass, along with small-seed broadleaves. The herbicide can be used preplant to early POST in labeled crops. Pyroxasulfone inhibits many cell elongation steps catalyzed by very-long-chain fatty acids. Susceptible weeds fail to emerge because the growth of the meristem and coleoptile are interrupted after germination (LeBaron and Wilcut 2014).

<u>Acetochlor</u>. Acetochlor is a VLCFA-inhibiting herbicide belonging to the chloroacetamide family. Acetochlor is labeled in cotton, soybean, and grain sorghum (*Sorghum bicolor* L. Moench.) and can also be used in corn production if a safener is incorporated. The herbicide can

be applied preplant incorporated or PRE for control of most annual grasses, yellow nutsedge, and certain small-seeded broadleaf weeds. Acetochlor is absorbed primarily through seedling shoots and secondarily through seedling roots. Plants larger than seedlings absorb acetochlor through the roots and translocate it throughout the shoots, where it accumulates in the nonreproductive parts of the plant (LeBaron and Wilcut 2014).

*Metolachlor*. Metolachlor is a VLCFA-inhibiting herbicide belonging to the chloracetamide family and is labeled in cotton, soybean, peanut (*Arachis hypogaea* L.), potato (*Solanum tuberosum* L.), pumpkin (*Cucurbita pepo* L.), tomato (*Solanum lycopersicum* L.), sunflower (*Helianthus annus* L.), sugar beet (*Beta vulgaris subsp. vulgaris* L.), and several other crops. *S*metolachlor is an isomeric form of metolachlor that is more active than metolachlor products containing both isomers (LeBaron and Wilcut 2014). Metolachlor can be applied preplant incorporated, PRE, or POST in most of these crops for the control of yellow nutsedge, smallseeded broadleaves, and annual grasses including barnyardgrass and red rice. Metolachlor is absorbed and acts within plants similar to acetochlor (LeBaron and Wilcut 2014).

**<u>Pethoxamid</u>**. Pethoxamid is a VLCFA-inhibiting herbicide in the chloroacetamide family. Studies have revealed that pethoxamid is active against several annual grasses and small-seeded broadleaves in corn and soybean (Dhareesank et al. 2005). Pethoxamid, along with other Group 15 herbicides, is systemic and is absorbed by plant roots and shoots. Pethoxamid is not currently labeled in rice nor is it labeled in other crops in the U.S., although the registration process for pethoxamid in rice, soybean, and corn in ongoing.

#### Past Research on VLCFA-Inhibiting Herbicides in U.S. Rice

There has been a limited amount of preliminary research conducted on the use of VLCFA-inhibiting herbicides in U.S. rice systems. In 2011 and 2012, the influence of herbicide rate and application timing (spiking, 2-lf, and 4-lf stages) was evaluated for two different VLCFA-inhibiting herbicides: *S*-metolachlor and acetochlor (Bararpour et al. 2012). Acetochlor (Warrant, Monsanto Company, St. Louis, MO) was applied at 420, 840, and 1,260 g ai ha<sup>-1</sup>, while *S*-metolachlor (Dual II Magnum, Syngenta Crop Protection LLC, Greensboro, NC) was applied at 840 and 1,400 g ai ha<sup>-1</sup>. At 5 wk after treatment, no injury was observed for the 2-lf application of acetochlor, and rice injury was only 4% and 3% for the highest rate of acetochlor at the spiking and 4-lf rice stage, respectively. Yield of rice treated with acetochlor was similar to the nontreated control for all application rates and timings except for 1,260 g ai ha<sup>-1</sup> applied to spiking rice. The rice injury caused by *S*-metolachlor was unacceptable for all rates and timings, with as much as 89% injury observed; however, rice yield from all applications except for 1,400 g ai ha<sup>-1</sup> applied to spiking rice was similar to the nontreated control.

The use of pyroxasulfone in rice was also evaluated in 2011 (Bararpour et al. 2013). Pyroxasulfone (Zidua, BASF Corporation, Research Triangle Park, NC) was applied to rice at rates of 50, 75, and 90 g ai ha<sup>-1</sup> at the spiking, 2-lf, and 4-lf rice stages on a silt loam and clay soil. Pyroxasulfone applied to rice grown on a silt loam soil caused 75% to 81% injury when applied at spiking, 69% to 76% injury when applied to 2-lf rice, and 6% to 31% injury when applied to 4-lf rice. Rice treated with pyroxasulfone on a clay soil resulted in 8% to 29%, 0% to 3%, and 3% to 21% rice injury for the various rates applied at the spiking, 2-lf, and 4-lf application timings, respectively. Rice yield was not affected by pyroxasulfone rate or application timing on the clay soil; however, yield was statistically less than the nontreated

control across all treatments on the silt loam soil (Bararpour et al. 2013).

Considering the similarities in rice yield to the nontreated control following applications of acetochlor, *S*-metolachlor, and pyroxasulfone on a clay soil, when applied at appropriate rates and timings, rice may tolerate several VLCFA-inhibiting herbicides. In order to combat herbicide-resistant weeds in rice production, it is imperative that an alternative herbicide SOA be used whenever possible. Pending further research, VLCFA-inhibiting herbicides may provide an additional herbicide SOA to be integrated into rice production.

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#### **Chapter 2**

# Efficacy and Selectivity of Pethoxamid Alone and in Mixture as a Delayed Preemergence Application to Rice

The evolution of herbicide resistance is making chemical control of weed species such as barnyardgrass and red rice extremely difficult in U.S. rice production. In order to combat herbicide resistance, it is imperative that alternative herbicide sites of action (SOA) be incorporated into rice whenever possible. There are currently no very-long-chain fatty acid (VLCFA)-inhibiting herbicides (WSSA Group 15) labeled for use in U.S. rice production; however, pethoxamid is one such herbicide currently under development. If appropriate rice tolerance and weed control can be established, pethoxamid would represent a unique herbicide SOA for use in U.S. rice. Field trials were conducted at the Rice Research and Extension Center (RREC) near Stuttgart, AR, in 2015 and the Pine Tree Research Station (PTRS) near Colt and the University of Arkansas Pine Bluff Farm (UAPB) near Lonoke, AR, in 2016, to assess pethoxamid applied alone and in combination with other herbicides as a delayed preemergence (DPRE) application in drill-seeded rice. Pethoxamid was applied at 0, 420, or 560 g ai ha<sup>-1</sup> alone and in combination with clomazone, imazethapyr, pendimethalin, and quinclorac. Minimal injury was seen with any treatment assessed. A reduction in shoot density and height compared to the nontreated control occurred in association with the use of pethoxamid; however, no decrease in yield resulted. The highest levels of barnyardgrass control followed the use of imazethapyr (91%) and quinclorac (89%) regardless of the presence of pethoxamid near Lonoke; however, pethoxamid applied at both rates in combination with clomazone and quinclorac increased barnyardgrass efficacy compared to when clomazone and quinclorac were applied alone. Near Colt, barnyardgrass control of 92 and 96% resulted from pethoxamid alone,

averaged over both rates. Based on these data, rice can tolerate pethoxamid when applied DPRE, and adequate levels of barnyardgrass control can be achieved at the rates evaluated; hence, pethoxamid appears to be a viable option for use in rice to allow for increased rotation of herbicide SOA to combat herbicide-resistant and difficult-to-control weeds.

**Nomenclature:** Pethoxamid; barnyardgrass, *Echinochloa crus-galli* (L.) Beauv.; red rice, *Oryza sativa* var. *sylvatica* L.; rice, *Oryza sativa* L.

**Key words:** weed control, herbicide-resistant weeds, herbicide site of action, delayed preemergence

#### Introduction

Due to the repetitive use of the same herbicide SOA year after year across vast portions of the U.S. rice acreage, evolution of herbicide resistance has occurred to many of the common weeds found in rice (Heap 2014). The evolution of resistance has limited the number of herbicides that can be used today for effective weed control in U.S. rice. Several of the most difficult-to-control, herbicide-resistant weeds in U.S. rice include barnyardgrass and red rice. In a survey of certified crop advisors in Arkansas in 2006, barnyardgrass and red rice were the most problematic weed species found in rice (Norsworthy et al. 2007). Cases of barnyardgrass with resistance to seven different herbicides among four herbicide SOA have been confirmed including: propanil (Weed Science Society of America [WSSA] Group 7); quinclorac (WSSA Group 4); clomazone (WSSA Group 13); and imazethapyr, imazamox, penoxsulam, and bispyribac (WSSA Group 2) (Heap 2017). Several populations of barnyardgrass were even found to have resistance to multiple SOA (Miller et al. 2015).

Red rice has been an extremely difficult-to-control weed species in rice for many years. The only way to chemically control red rice in cultivated rice is through use of imidazolinoneresistant rice (Clearfield<sup>TM</sup>, BASF Corporation, Research Triangle Park, NC). Imidazolinoneresistant rice enables the use of imazethapyr and imazamox in the crop (Croughan et al. 1996). Imidazolinone-resistant rice was introduced in 2002, and since then has been planted extensively. In 2014, approximately 49% of Arkansas rice acreage was planted to the imidazolinone-resistant technology (Hardke 2015).

The poor stewardship associated with imidazolinone-resistant rice and repetitive use of the same herbicides in turn caused an increased occurrence of red rice with resistance to both imazethapyr and imazamox (Burgos et al. 2008). Cultivated rice and red rice are sexually compatible, which allows for natural hybridization between the two. The outcrossing associated with cultivated rice and red rice is the primary reason for the rapid buildup of red rice with resistance to acetolactate synthase (ALS)-inhibiting herbicides (Shivrain et al. 2007).

In order to combat herbicide resistance, multiple herbicide SOA should be used annually, rather than reliance on one particular herbicide (Norsworthy et al. 2012). From 2004 to 2007, a study was conducted by Beckie (2009) to determine the impact of herbicide rotation on the evolution of resistance to ALS-inhibiting herbicides in field pennycress (*Thlaspi arvense* L.) in Canada. Resistance of plants in the soil seedbank increased from 29% to 85% after four applications of the ALS-inhibiting herbicide, ethanmetsulfuron. In contrast, the frequency of resistance to the ALS-inhibiting herbicide ethanmetsulfuron, when applied in rotation with a bromoxynil + MCPA formulated product (WSSA Group 6/WSSA Group 4), was similar to the nontreated control. Hence, the use of multiple herbicide SOA can delay the onset of herbicide resistance for many of the problematic weed species found in crops.

Currently, there are no VLCFA-inhibiting herbicides (WSSA Group 15) registered for use in U.S. rice (Anonymous 2017). Pethoxamid (FMC Corporation, Philadelphia, PA), a WSSA Group 15 herbicide, belongs to the chloroacetamide family and is currently under development in canola (*Brassica napus* L.), corn (*Zea mays* L.), cotton (*Gossypium hirsutum* L.), rice, soybean (*Glycine max* (L.) Merr.), and sunflower (*Helianthus annuus* L.). If the appropriate rice tolerance and weed efficacy can be achieved, pethoxamid would represent a unique SOA to be used in U.S. rice.

Pethoxamid is a root and shoot inhibitor; therefore, it must be applied prior to weed germination in order to be effective (Anonymous 2015). A potential application timing for pethoxamid would be delayed PRE (DPRE) to control weeds that germinate soon after the rice is

seeded. Pethoxamid has a spectrum of activity similar to acetochlor, metolachlor, alachlor, and dimethanamid-P, also chloroacetamides, which control small-seeded broadleaves and grasses such as barnyardgrass and red rice (O'Connell et al. 1998).

Chloroacetamides are used with success in Asian rice culture. Herbicides such as pretilochlor and butachlor are used to control troublesome grasses in Asian dry- and wet-seeded rice. Butachlor has been used for weed control in India by applying the herbicide after broadcasting sprouted rice (Mutnal et al. 1998). A study was conducted comparing the use of pretilochlor + pyribenozaxim followed by (fb) MCPA to several other common rice herbicide regimes applied 8 days after rice sowing in Sri Lanka. The program containing pretilochlor resulted in the highest level of weed control at 99% and highest yield compared to other treatments (Chauhan 2012). Because herbicides belonging to the chloroacetamide family have been successfully used in Asian rice culture, pethoxamid may be a viable option for use in U.S. rice.

Current soil-applied herbicide options in the U.S. prior to weed germination in rice include clomazone, quinclorac, pendimethalin, imazethapyr, and thiobencarb (Scott 2013); however, barnyardgrass resistance to clomazone, quinclorac, and imazethapyr has been reported (Heap 2017). Group 15 herbicides, such as pethoxamid, are at a relatively low risk for resistance, having only five resistant species worldwide.

Pethoxamid was evaluated for barnyardgrass control in sunflower under irrigated and non-irrigated conditions. Herbicide efficacy was evaluated at the four-leaf sunflower stage and just before canopy closure. Barnyardgrass control with pethoxamid was 90 to 97% with a pethoxamid rate of 1,200 g ai ha<sup>-1</sup>, regardless of irrigation condition (Jursik et al. 2015).

If appropriate rice tolerance can be established, it is believed that pethoxamid may present an alternative herbicide SOA for use in U.S. rice. Therefore, the objective of this research was to assess the efficacy and rice tolerance following DPRE-applied pethoxamid alone and in combination with other rice herbicides in drill-seeded rice. It was hypothesized that rice would display adequate tolerance to pethoxamid applied alone and in combination with other soil-applied herbicides when applied DPRE while providing acceptable early-season weed control.

#### **Materials and Methods**

Field trials were conducted in 2015 at the Rice Research and Extension Center (RREC) near Stuttgart, AR, and in 2016 at the Pine Tree Research Station (PTRS) near Colt, AR, and the University of Arkansas Pine Bluff Farm (UAPB) near Lonoke, AR. The soils at the RREC, PTRS, and UAPB, respectively, were a Dewitt silt loam (fine, smectic, thermic typic albaqualf), Calloway silt loam (fine-silty, mixed, active, thermic aquic fraglossudalfs), and Immanuel silt loam (fine-silty, mixed, active, thermic oxyaquic glossudalfs). Imidazolinone-resistant (Clearfield<sup>™</sup>, BASF Corporation, Research Triangle Park, NC) inbred rice cultivars CL111 (RREC) and CL151 (PTRS and UAPB) were drill-seeded into 1.8- by 5.2-m plots at a seeding rate of 72 seed m<sup>-1</sup> of row, and row width was 18 cm. Rice was planted on May 5, 2015, at the RREC, May 9, 2016, at the PTRS, and May 18, 2016, at the UAPB. Rice fertility programs were based on University of Arkansas Division of Agriculture Research and Extension recommendations (Norman et al. 2013).

The experiment was set up as a factorial arrangement of treatments in a randomized complete block design with four blocks, with the rate of pethoxamid and additional herbicide as

the two factors. The pethoxamid rates were 0 (none), 420, and 560 g ai ha<sup>-1</sup>. Each rate of pethoxamid was applied alone and in tank mixture with clomazone at 336 g ai ha<sup>-1</sup> (Command 3ME, FMC Corporation, Philadelphia, PA), imazethapyr at 71 g ai ha<sup>-1</sup> (Newpath, BASF Corporation, Research Triangle Park, NC), pendimethalin at 1,120 g ai ha<sup>-1</sup> (Prowl H<sub>2</sub>O, BASF Corporation, Research Triangle Park, NC), and quinclorac at 420 g ai ha<sup>-1</sup> (Facet L, BASF Corporation, Research Triangle Park, NC). All treatments were applied DPRE at five to seven days after rice planting (DAP).

The DPRE applications were made May 11, 2015 (6 DAP) at the RREC, May 23, 2016 (5 DAP) at the UAPB, and May 16, 2016 (7 DAP) at the PTRS. Data collection included a visual assessment of crop injury (0 to 100%, with 100 being crop death) every 2 weeks after application until physiological maturity, the average of three crop canopy heights (cm) per plot 2 weeks after treatment (WAT), shoot density (number of shoots m<sup>-1</sup> of row), rough rice yield (kg ha<sup>-1</sup>) (RREC and PTRS), and estimates of barnyardgrass control (0 to 100%, with 100 being complete control) (UAPB and PTRS). Rice injury was evaluated for percentage visible reduction in rice height, stand, and tillering. Data collection were intended to be identical at each location; however, due to the lack of natural weed pressure at the RREC, barnyardgrass control was not assessed. Also, yield was only reported for the RREC and PTRS due to the inconsistent results caused by panicle blast (*Magniporthe grisea* (Hebert) Barr [teleom]) at the UAPB.

Parameters including shoot density and rice height were normalized for each location by converting them to a percentage relative to the average of the nontreated control plots. Data were analyzed in JMP 12 Pro (SAS Institute Inc, Cary, NC).. Site years were analyzed separately using analysis of variance (ANOVA) with block included as a random effect. The no pethoxamid rate  $\times$  no additional herbicide (none  $\times$  none) treatment was removed from the

analysis for the rice tolerance and barnyardgrass control assessments. All means were separated using Fisher's protected LSD ( $\alpha$ =0.05).

#### **Results and Discussion**

Injury ratings were low across all locations and rating dates. Up to 19% injury was observed at the RREC 2 WAT following the application of a high rate of pethoxamid + clomazone (Table 2.1). The injury was due to the bleaching symptomology caused by clomazone in the mixture, considering that injury from pethoxamid alone was only 6%. Injury to grasses by VLCFA-inhibiting herbicides include leafing-out underground or leaves not properly unfurling (Gunsolus and Curran 1999). By 5 WAT, however, this injury dissipated to 4% (Table 2.1). By 9 WAT, injury from all treatments was  $\leq$  3% (data not shown).

Rice shoot densities were slightly reduced when pethoxamid at either rate was mixed with some of the herbicides evaluated at RREC, whereas pethoxamid alone did not cause reduced density (Table 2.2). Pethoxamid alone or in combination with other herbicides had no negative impact on shoot density at UAPB. At the PTRS, the low and high rate of pethoxamid averaged over other herbicides caused a decrease in shoot density of only 4 percentage points compared to the nontreated control.

Plant height reductions caused by pethoxamid-containing treatments occurred at PTRS but not at RREC or UAPB (Table 2.2). At the PTRS, the only pethoxamid-containing treatments in which a reduction in plant height was not observed included the low rate of pethoxamid alone (94% of nontreated), the low rate of pethoxamid plus clomazone (100% of nontreated), and the high rate of pethoxamid plus quinclorac (97% of nontreated). All treatments from which pethoxamid was excluded had similar or greater plant heights than the nontreated

control. Based on data from the PTRS, a slight reduction in shoot density and height may be associated with the use of pethoxamid at either rate evaluated; albeit, this was not observed at the RREC or UAPB.

An interaction was observed between pethoxamid rate and additional herbicide for percent barnyardgrass control at the UAPB at 2 and 10 WAT (Table 2.3). At 5 WAT, use of the high rate of pethoxamid with clomazone, pendimethlin, or quinclorac improved barnyardgrass control over thethree herbicides alone (Table 2.3). Barnyardgrass control at 10 WAT ranged from 83 to 95% when the high pethoxamid rate was applied with clomazone, imazethapyr, pendimethalin, or quinclorac. In a standard production system, a subsequent POST application would be needed to control the few escaped barnyardgrass plants that exist immediately prior to establishing a permanent flood. At the PTRS, barnyardgrass control ranged from 89 to 100% at 5 WAT when pethoxamid was applied with clomazone, imazethapyr, pendimethalin, or quinclorac. By 10 WAT, the same treatments still maintained a high level of barnyardgrass control ( $\geq$  85%). Averaged over herbicides added to pethoxamid, the high rate of pethoxamid at PTRS resulted in a slight improvement in barnyardgrass control over the low rate at 10 WAT, with an average of 98% barnyardgrass control.

Because of panicle blast, yield data at the UAPB was inconsistent and is not reported. At the RREC, rough rice yield was solely a function of rice tolerance due to lack of weeds present at this location. All treatments at the RREC resulted in comparable or greater rough rice yields than the nontreated control, indicating pethoxamid did not have a deleterious effect on yield (Table 2.4). At the PTRS location, only the main effects of pethoxamid rate and herbicide additive were significant (Tables 2.4). The highest rate of pethoxamid resulted in greater rough rice yield than did the lower rate or the absence of pethoxamid, averaged over herbicide

additives. The improved yield is partly attributed to barnyardgrass control but may also be a result of other weeds at the experimental site that existed at lower densities. Yellow nutsedge (*Cyperus esculentus* L.), which can be suppressed by chloroacetamide herbicides (Zimdahl 2007), was present but not at a consistent enough density throughout the test to rate; however, this weed may have reduced yield in some plots.

Based on these results, adequate rice tolerance and adequate early-season barnyardgrass control can be achieved with pethoxamid-containing weed control programs applied DPRE; however, the addition of other herbicides to pethoxamid applied DPRE are deemed necessary. These results coincide with Doherty et al. (2016) who saw the highest levels of barnyardgrass control with pethoxamid applied in combination with imazethapyr. Pethoxamid in combination with clomazone or pendimethalin sometimes improved barnyardgrass control compared to clomazone or pendimethalin applied alone; therefore, pethoxamid can have a positive impact on barnyardgrass control as a soil-applied herbicide in combination with other rice herbicides. The addition of pethoxamid to current weed control programs would likely increase barnyardgrass control and provide an alternative SOA to protect against further evolution of resistance in this as well as other weeds of rice.

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		Injury								
				2 W	AT				5 WAT	
Factor		RR	EC	UA	PB	P	ΓRS	RREC	UAPB	PTRS
							%			
Pethoxamid Rate										
	None	2	b	2	c	1	b	3	1	2
	Low	9	a	5	b	2	b	2	1	1
	High	10	а	10	a	4	a	1	1	1
Additional Herbicide										
	None	8	b	9		3	b	1	1	1
	Clomazone	13	а	7		5	a	3	2	2
	Imazethapyr	4	с	6		2	bc	2	2	1
	Pendimethalin	5	bc	4		2	bc	1	0	0
	Quinclorac	5	bc	4		1	с	2	1	1
Pethoxamid Rate $\times$										
Additional Herbicide	None $\times$ None	-		-		-		-	-	-
	None × Clomazone	5		5		4		1	1	1
	None × Imazethapyr	1		1		0		0	0	0
	None × Pendimethalin	0		2		0		0	0	0
	None × Quinclorac	0		0		0		0	1	1
	$Low \times None$	9		8		2		1	1	1
	Low × Clomazone	16		3		4		4	1	0
	$Low \times Imazethapyr$	4		8		2		1	3	1
	Low × Pendimethalin	8		2		3		1	1	0
	Low × Quinclorac	6		3		2		1	0	0
	$High \times None$	6		11		3		0	1	1
	High × Clomazone	19		13		6		4	4	5
	$High \times Imazethapyr$	6		8		4		4	1	2
	High $\times$ Pendimethalin	8		9		3		2	0	0
	High $\times$ Quinclorac	10		10		2		5	1	1

Table 2.1. Percent rice injury ratings 2 and 5 WAT for pethoxamid alone and in combination at RREC, UAPB, and PTRS.<sup>a,b,c,d</sup>

Table 2.1 Cont.

P Values from ANOVA				
Pethoxamid Rate (P)	< 0.0001	< 0.0001	0.0043	
Additional Herbicide (P)	< 0.0001	NS	<.0001	
Pethoxamid Rate $\times$ Addittional Herbicide (P)	NS	NS	NS	

<sup>a</sup> Abbreviations: WAT, weeks after treatment; RREC, Rice Research and Extension Center; UAPB,

University of Arkansas Pine Bluff Lonoke Farm; PTRS, Pine Tree Research Station; Peth, pethoxamid <sup>b</sup> Means within a column followed by the same lowercase letter are not different according to Fisher's protected LSD ( $\alpha = 0.05$ ).

<sup>c</sup> The inability to collect percent injury ratings for none  $\times$  none is represented by (-).

<sup>d</sup> 5 WAT data were not amenable to formal statistical analysis.

		Shoot density					Height			
Factor		RF	REC	UAPB	PTR	S	RREC	UAPB	PTI	RS
					%	of nor	ntreated			
Pethoxamid Rate										
	None	103		101	97	a	101	101	105	
	Low	91		100	93	b	100	98	90	
	High	90		97	93	b	101	98	83	
Additional Herbicide										
	None	101		98	93		100	99	89	
	Clomazone	85		104	94		101	93	94	
	Imazethapyr	93		97	95		101	101	88	
	Pendimethalin	91		100	94		101	105	91	
	Quinclorac	101		100	94		100	95	98	
Pethoxamid Rate × Additional Herbicide										
	None × None	100	bcde	100	100		100	100	100	bc
	None × Clomazone	92	efg	113	96		100	94	102	bc
	None $\times$ Imazethapyr	108	ab	99	97		101	106	107	ab
	None × Pendimethalin	103	abc	99	96		101	112	105	ab
	None × Quinclorac	111	а	97	97		100	96	111	а
	Low × None	102	abcd	99	94		99	100	94	cd
	Low × Clomazone	81	h	103	93		101	94	100	bc
	$Low \times Imazethapyr$	82	gh	100	94		101	100	85	e
	Low × Pendimethalin	89	fgh	99	93		100	100	85	e
	Low × Quinclorac	98	cdef	102	92		101	94	87	de
	High $\times$ None	100	bcde	95	91		101	99	84	e
	High $\times$ Clomazone	83	gh	96	92		101	92	80	ef
	High × Imazethapyr	89	fgh	92	93		101	99	74	f
	High × Pendimethalin	81	h	103	93		101	103	81	ef
	High × Quinclorac	93	def	101	94		101	95	97	с

Table 2.2. Rice shoot density and height at RREC, UAPB, and PTRS.<sup>a,b,c</sup>

Table 2.2 Cont.

<i>P</i> Values from ANOVA						
Pethoxamid Rate (P)	< 0.0001	NS	0.0043	NS	NS	<.0001
Additional Herbicide (P)	< 0.0001	NS	< 0.0001	NS	NS	0.0028
Pethoxamid Rate $\times$ Additional Herbicide (P)	0.0086	NS	NS	NS	NS	<.0001

<sup>a</sup> Abbreviations: RREC, Rice Research and Extension Center near Stuttgart; UAPB, University of Arkansas Pine Bluff Lonoke Farm near Lonoke; PTRS, Pine Tree Research Station near Colt; Peth, pethoxamid <sup>b</sup> Means within a column followed by the same lowercase letter are not different according to Fisher's protected LSD ( $\alpha = 0.05$ ).

<sup>c</sup> Average shoot density and height for the nontreated plots was 88 shoots per m<sup>-1</sup> of row and 86 cm at the RREC, 52 shoots per m<sup>-1</sup> of row and 45 cm at the UAPB, and 98 shoots m<sup>-1</sup> of row and 43 cm at the PTRS

Factor				Barny	ardgr	ass coi	ass control					
Factor			2 W.	AT			10 V	VAT				
		UA	APB	PTRS	5	UA	PB	PTF	RS			
					%							
Pethoxamid Rate												
	None	74		96		77		97	al			
	Low	82		96		78		95	b			
	High	82		98		81		98	a			
Additional Herbicide												
	None	74		92	b	92		96	b			
	Clomazone	76		98	а	98		100	a			
	Imazethapyr	89		99	а	10 0		100	a			
	Pendimethalin	68		96	a	96		99	al			
	Quinclorac	88		96	a	96		98	al			
Pethoxamid Rate $\times$												
Additional Herbicide	None $\times$ None	-		-		-		-				
	None × Clomazone	65	g	99		74	f	98				
	None $\times$ Imazethapyr	91	а	100		91	a-c	100				
	None $\times$ Pendimethalin	50	h	95		76	ef	94				
	None × Quinclorac	89	ab	91		89	a-d	96				
	$Low \times None$	79	de	99		81	d-f	85				
	Low × Clomazone	80	cde	89		83	c-e	99				
	$Low \times Imazethapyr$	87	abc	100		93	ab	100				
	$Low \times Pendimethalin$	74	ef	96		76	ef	95				
	Low × Quinclorac	89	ab	96		89	a-d	96				
	$High \times None$	70	fg	96		76	ef	93				
	$High \times Clomazone$	82	bcd	98		86	b-d	99				
	High  imes Imazethapyr	90	a	99		95	a	99				
	High $\times$ Pendimethalin	80	cde	96 100		83	c-e	98				
	High × Quinclorac	87	abc			89	a-d	100				

# Table 2.3. Percent barnyardgrass control 2 and 10 WAT at UAPB and PTRS.<sup>a,b,c</sup>

<sup>a</sup> Abbreviations: WAT, weeks after treatment; UAPB, University of Arkansas Pine Bluff Lonoke Farm near Lonoke; PTRS, Pine Tree Research Station near Colt; Peth, pethoxamid <sup>b</sup> Means within a column followed by the same lowercase letter are not different according to Fisher's protected LSD at ( $\alpha = 0.05$ )

<sup>c</sup> The inability to collect percent injury ratings for none  $\times$  none is represented by (-).

		Yield							
Factor		RF	REC	PTR	S				
Pethoxamid Rate			—kg h	a <sup>-1</sup>					
Peuloxannu Kale	None	7760		5860	b				
	Low	8010		5810	b				
		8230		6570					
Additional Herbicide	High	8230		0370	а				
Additional Herbicide	None	7730		5460	с				
	Clomazone	8020		6320	ab				
	Imazethapyr	8020 7860		6070	ab				
	Pendimethalin	8570		6020	ab b				
	Quinclorac	7810		6620					
Pethoxamid Rate $\times$	Quinciorae	/010		0020	а				
Additional Herbicide									
	None $\times$ None	7810	cdef	4750					
	None × Clomazone	7630	ef	6520					
	None $\times$ Imazethapyr	7160	f	5910					
	None × Pendimethalin	8440	abc	5610					
	None $\times$ Quinclorac	7780	cdef	6670					
	Low × None	7580	ef	4950					
	Low × Clomazone	8290	abcd	5560					
	$Low \times Imazethapyr$	7780	cdef	6120					
	Low × Pendimethalin	8440	ab	6270					
	Low × Quinclorac	7950	bcde	6270					
	$High \times None$	7780	def	6620					
	High $\times$ Clomazone	8140	abcde	6920					
	High $\times$ Imazethapyr	8590	а	6170					
	High $\times$ Pendimethalin	8790	а	6220					
	$High \times Quinclorac$	7730	def	6920					
P Values from ANOV	A								
Peth Rate (P)	0.0117		0.0018						
Additional Herbicide (	0.0003	0.0025							
Peth Rate $\times$ Additiona	l Herbicide ( <i>P</i> )	0.0482	2	NS					

2.4 Rough rice yield at RREC and PTRS. <sup>a,b,c</sup>

<sup>a</sup> Abbreviations: PTRS, Pine Tree Research Station near Colt; Peth, pethoxamid <sup>b</sup> Means within a column followed by the same lowercase letter are not different according to Fisher's protected LSD at ( $\alpha = 0.05$ )

<sup>c</sup> Average rough rice yield for the nontreated control was 7,370 kg ha<sup>-1</sup> at the RREC and 7,120 kg ha<sup>-1</sup> at the PTRS.

#### Chapter 3

# Effect of Application Timing and Rate of Very-Long-Chain Fatty Acid-Inhibiting Herbicides on Rice Tolerance

Integrating herbicide sites of action (SOA) used in other cropping systems into rice production provides a unique opportunity to control weeds that are resistant to current rice herbicides. Today, there are no very-long-chain fatty acid (VLCFA)-inhibiting herbicides labeled for use in U.S. rice. Field trials were conducted in 2015 and 2016 near Stuttgart, Arkansas, to assess rice tolerance to four VLCFA-inhibiting herbicides. Individual field trails were conducted for rice tolerance to pyroxasulfone, S-metolachlor, pethoxamid, and acetochlor. In each trial, two rates of the particular herbicide were applied to rice at the delayed preemergence (DPRE), spiking, 1to 2-leaf (lf), and 3- to 4-lf timings. Rice injury ranging from 20 to 96% was observed 5 weeks after flooding (WAF) following the use of pyroxasulfone at all rates and application timings, which caused a decrease in rice yield relative to the nontreated control in both years. Rice injury caused by S-metolachlor 5 WAF was reduced the further the application timings were from planting in 2016; however, injury ratings ranged from 19 to 100% over the growing season. Overall, acetochlor and pethoxamid caused the lowest injury with little or no reduction in yield compared to the nontreated control. Because of rice tolerance to acetochlor and pethoxamid in both 2015 and 2016 at the 1- to 2-lf and 3- to 4-lf application timings, it appears that these herbicides can safely be applied in rice production.

Nomenclature: Acetochlor; pethoxamid; pyroxasulfone; *S*-metolachlor; rice, *Oryza sativa* L. Key words: Herbicide-resistant weeds, very-long-chain fatty acid-inhibiting herbicides, weed control

#### Introduction

VLCFA-inhibiting herbicides are currently labeled in U.S. row crop production for the control of grasses and small-seeded broadleaf weeds (Knowles 1998). VLCFA-inhibiting herbicides include metolachlor, alachlor, dimethenamid-P, flufenacet, pyroxasulfone, acetochlor, and pethoxamid among others. Several families within the VLCFA-inhibiting SOA are the chloroacetamides, pyrazoles, and oxyacetamides (Gibson 2004).

VLCFA-inhibiting herbicides are soil applied, and when taken up by the plant have an effect on meristem-bearing cell division in the developing root and shoot tips; however, these herbicides do not damage preexisting tissues (Babczinski 2011). Very-long-chain fatty acids are essential biological components of sphingolipids, which are used for effective eukaryotic cell function, and are also constituents of cellular waxes and serve as seed storage triacylglycerols. Biosynthesis of very-long-chain fatty acids is inhibited through a reaction involving covalent binding between herbicide and a cysteine residue in the reactive site of the target enzyme. The damage in the roots, shoots, and coleoptiles caused by VLCFA-inhibiting herbicides are due to inhibitions of cell enlargement and mitosis (Babczinski 2011).

Barnyardgrass (*Echinochloa crus-galli* (L.) Beauv.) and red rice (*Oryza sativa* L. var. *sylvatica*) have become increasingly difficult to control because of herbicide resistance to several of the commonly used herbicide sites of action (SOA) in U.S. rice such as acetolactate synthase (ALS) inhibitors, acetyl CoA carboxylase (ACCase) inhibitors, auxin receptors (synthetic auxins), photosystem II (PSII) inhibitors, and diterpene biosynthesis inhibitors (Heap 2017). Resistance of barnyardgrass and red rice has occurred in great part due to the repetitive use of the same herbicide SOA in rice. Repetitive applications of herbicides containing the same SOA can lead to the rapid evolution of herbicide resistance (Norsworthy et al. 2012).

Under selection pressure, dominant alleles increase in frequency faster than recessive alleles in random mating weed populations; however, dominant and recessive alleles increase at similar rates in self-fertilizing species. Moreover, movement of pollen or seeds throughout a population of weeds can provide gene flow, which spreads resistant genes much faster than rates of mutation (Jasieniuk et al. 1996). The evolution of herbicide resistance through selection pressure in rice has limited effective herbicide SOA; therefore, it is imperative that alternative herbicides be evaluated in rice.

Only five weed species have evolved resistance to VLCFA-inhibitors worldwide, and Italian ryegrass (*Lolium multiflorum* Lam.) is the only weed with confirmed resistance to VLCFA-inhibitors in the U.S. (Heap 2017). Furthermore, the VLCFA-inhibiting herbicides pretilachlor and butachlor have been used with success in Asian dry- and wet-seeded rice production as preemergence (PRE) and postemergence (POST) herbicides (Rao et al. 2007). Butachlor is used for the control of annual grasses PRE, but can be applied POST for control of 1- to 2-lf grasses. Butachlor is generally applied 3 to 7 days after transplanting or 10 to 12 days after emergence for direct-seeded rice. Pretilachlor is a selective herbicide that can be applied prior to transplanting or between transplanting and weed emergence. It is generally applied to transplanted rice; however, in direct-seeded rice systems, fenclorim must be applied one day prior to pretilachlor applications to act as a safener (Ampong-Nyarko and Datta 1991).

The following experiments will focus on the use of VLCFA-inhibiting herbicides including pyroxasulfone, *S*-metolachlor, pethoxamid, and acetaochlor in rice. Pyroxasulfone belongs to the pyrazole family and has been shown to control barnyardgrass effectively at 31.25 g ai ha<sup>-1</sup> (Nurse et al. 2011). *S*-metolachlor, a chloroacetamide, has been used as a soil-applied herbicide to control grasses and small-seeded broadleaf weeds in agronomic crops other than rice

(PPDB 2016). In soybean (*Glycine max* (L.) Merr.), *S*-metolachlor at 1,680 g ha<sup>-1</sup> provided up to 90% control of red rice 14 days after application (Zemolin et al. 2014). Pethoxamid, a chloroacetamide, is currently under development in rice. Pethoxamid provided 90 to 97% control of barnyardgrass in sunflower (*Helianthus annuus* L.) (Jursik et al. 2015). Pethoxamid has also shown adequate barnyardgrass control and rice tolerance when applied alone and in combination with other rice herbicides (Doherty et al. 2016). Acetochlor, also a chloroacetamide, is available as a microencapsulated, slow-release formulation. Encapsulated formulations enclose the herbicide molecule in microscopic, porous polymer particles. The herbicide is released from the microencapsulation slowly by the dissolution of the polymer, generally in the presence of moisture. The slow release of the herbicide lengthens the time of activity and can play a significant role in weed control and crop tolerance (Rao 2000).

A microencapsulated formulation of acetochlor has been used with success for weed control, including barnyardgrass, in various row crops. In 2011, microencapsulated acetochlor was evaluated for weed control in cotton (*Gossypium hirsutum* L.) on a silt loam soil. Acetochlor applied 14 days prior to cotton planting at 1,262 g ha<sup>-1</sup> provided barnyardgrass control of 93% at planting, 64% 1 week after planting, and 43% 2 weeks after planting (Riar et al. 2012).

These studies suggest that at appropriate herbicide rates and application timings, rice may exhibit tolerance to one or more VLCFA-inhibiting herbicides, providing an alternative herbicide SOA. It was hypothesized that at least one VLCFA-inhibiting herbicide may be applied to rice with minimal crop injury dependent upon herbicide rate and application timing. The objective of these experiments is to determine the effect of application timing and rate of VLCFA-inhibiting herbicides on rice tolerance.

#### **Materials and Methods**

Field trials were conducted in 2015 and 2016 at the Rice Research and Extension Center (RREC), near Stuttgart, Arkansas, on a Dewitt silt loam soil (fine, smectic, thermic type Albaqualf). In each year, imidazolinone-resistant (Clearfield<sup>™</sup>, BASF Corporation, Research Triangle Park, NC) CL111 rice was drill seeded at 72 seeds per m<sup>-1</sup> of row into 1.8 by 5.2 m plots at a drill spacing of 18 cm. Rice was planted for all trials at the RREC on May 5, 2015, and on April 25, 2016. Rice fertility programs were based on University of Arkansas Division of Agriculture Research and Extension recommendations (Norman et al. 2013).

Separate field trials were conducted for pyroxasulfone, S-metolachlor, pethoxamid, and acetochlor. Each experiment was designed as a two-factor randomized complete block with eight treatments as well as a nontreated control. Factor A (fixed) included herbicide rate and factor B (fixed) consisted of application timing for each experiment. A high and low rate of each herbicide were applied at DPRE, spiking, 1- to 2-, and 3- to 4-leaf (lf) timings. Actual rice stages at the time of herbicide application for 1- to 2-lf and 3- to 4-lf timings were 2-lf in 2015 and 1-lf in 2016, and 3-lf in 2015 and 4-lf in 2016, respectively. The rates for each herbicide included: pyroxasulfone (Zidua, BASF Corporation, Research Triangle Park, NC) at 90 g ha<sup>-1</sup> (low) and 150 g ha<sup>-1</sup> (high), S-metolachlor (Dual II Magnum, Syngenta Crop Protection, LLC, Greensboro, NC) at 535 g ha<sup>-1</sup> (low) and 1,070 g ha<sup>-1</sup> (high), pethoxamid (FMC Corporation, Philadelphia, PA) at 420 g ha<sup>-1</sup> (low) and 840 g ha<sup>-1</sup> (high), and acetochlor (Warrant, Monsanto Company, St. Louis, MO) at 630 g ha<sup>-1</sup> (low) and 1,050 g ha<sup>-1</sup> (high). Application dates for all trials are presented in Table 3.1. All herbicides were applied with a CO<sub>2</sub>-pressurized backpack sprayer through 110015 AIXR (TeeJet) nozzles calibrated to deliver 143 L ha<sup>-1</sup> using a threenozzle boom at 51 cm spacing at 4.83 km h<sup>-1</sup>.

All trials were focused strictly on rice tolerance; therefore, plots were kept weed free with clomazone (Command 3 ME, FMC Corporation, Philadelphia, PA) (336 g ha<sup>-1</sup>) PRE on May 5, 2015, imazethapyr (Newpath, BASF Corporation, Research Triangle Park, NC) (70 g ha<sup>-1</sup>) on June 4, 2015, clomazone (336 g ha<sup>-1</sup>) + quinclorac (Facet L, BASF Corporation, Research Triangle Park, NC) (420 g ha<sup>-1</sup>) on June 20, 2015, clomazone PRE on April 25, 2016, imazethapyr (70 g ha<sup>-1</sup>) on May 23, 2016, and propanil (SuperWham, RiceCo LLC, Memphis TN) (3.36 kg ha<sup>-1</sup>) + thiobencarb (Bolero 8 EC, Valent U.S.A. Corporation, Walnut Creek, CA) (3.36 kg ha<sup>-1</sup>) + halosulfuron (Permit 75WG, Gowen Company, Yuma, AZ) (52 g ha<sup>-1</sup>) on June 14, 2016.

Data collection for all trials included a visual assessment of crop injury compared to the nontreated control approximately every two weeks after herbicide application until physiological maturity. Visual crop injury was assessed on a scale of 0 to 100%, with 0% being no injury and 100% being crop death. Visual injury assessment included reduction in rice height, stand counts, and tillering. Along with percent injury, assessment parameters included the average of three crop canopy heights (cm) per plot, rice shoot density (number of shoots per m<sup>-1</sup> row), days to 50% heading, and rough rice yield. Parameters including crop density, height, and rough rice yield were normalized for each test by converting them to a percentage relative to the average of each parameter in the nontreated control. Rainfall data were collected from a weather station located at the RREC near the trials evaluated.

Data were analyzed in JMP 12 Pro (SAS Institute Inc, Cary, NC). Data were analyzed separately by year because of rainfall differences between years (Figures 3.1 and 3.2) and were analyzed using analysis of variance (ANOVA) with block included as a random effect. All means were separated using Fisher's protected LSD ( $\alpha$ =0.05).

#### **Results and Discussion**

**Rainfall.** More injury and reduction in shoot density, height, and yield were seen in 2016 than in 2015 for most experiments and evaluation parameters due to differences in timing and amount of rainfall in relation to the application dates. For instance, at 2 weeks after the low rate of pyroxasulfone was applied DPRE in 2015, 33% injury was observed; however, in 2016, 68% injury was observed for the same treatment (Table 3.2).

In 2015, rice was planted into a dry seedbed on May 5 (Table 3.1), which was not conducive for germination. A rainfall event of 5.5 cm occurred May 11 (Figure 3.1), which enabled the rice seed to imbibe water and germinate immediately after receiving the DPRE herbicide applications. VLCFA-inhibiting herbicides are soil-applied root and shoot inhibitors; therefore, they require rainfall for herbicidal activation (Rao 2000) and affect root and shoots after germination (Anonymous 2015). The 5.5 cm rainfall event activated the soil-applied herbicides as the rice seed began to imbibe water and germinate, causing significant injury from several of the DPRE herbicides across all trials.

In 2016, minor rainfall events occurred prior to planting; however, rice was still planted into a relatively dry seedbed on April 24 (Table 3.1) and prevented consistent germination of the rice seed. A rainfall event greater than 1 cm did not occur after planting until May 2 (Figure 3.1), which was the same day the spiking treatments were applied for all trials. Even though, spiking rice was present, a lack of consistency in stand due to the dry conditions was noted. A rainfall event of 10.4 cm occurred soon after the spiking treatments were applied. The moist conditions enhanced activation of both the spiking and DPRE treatments. The initially dry conditions followed by saturated conditions likely enhanced injury to rice from the VLCFA-inhibiting herbicides in 2016.

Research has been conducted assessing the effect of soil moisture on the phytotoxicity of pethoxamid to rice. In a study conducted in Japan, soil moisture was adjusted to 50, 60, 70, and 80% directly after pethoxamid applications (Dhareesank et al. 2006). Rice seedlings were then transplanted 0, 1, 3, 5, 10, 15, and 20 days later. Results showed that as soil moisture increased, the phytotoxicity associated with pethoxamid to rice increased, which was induced by the availability of pethoxamid in the soil water. Results also showed that a decrease in phytotoxicity occurred as time increased between pethoxamid application and planting (Dhareesank et al. 2006). The results from the trial support observations of an increase in injury to rice with the use of VLCFA-inhibiting herbicides prior to a significant rainfall event.

**Pyroxasulfone.** Generally, pyroxasulfone caused an unacceptable level of visible injury when applied to rice across all rates and application timings. The highest visible rice injury was 55 and 100% in 2015 and 2016, respectively, 2 WAT following an application of the high rate of pyroxasulfone DPRE (Table 3.2). In both 2015 and 2016, injury 2 WAT generally decreased as pyroxasulfone treatments were applied to later rice growth stages. These results coincide with previous findings on the use of pyroxasulfone in rice, when a decrease in injury was seen from a DPRE application to 4-lf rice on both silt loam and clay soils (Bararpour et al. 2013).

In both 2015 and 2016, similar trends in rice injury as a function of rate and application timing were observed at 5 WAF (Table 3.2). Rice treated with pyroxasulfone often had reduced shoot density and height compared to the nontreated control. The highest injury at 5 WAF was caused by the high rate of pyroxasulfone applied DPRE, similar to the 2 WAT rating. In 2015 and 2016, injury to rice caused by pyroxasulfone at 5 WAF was  $\geq$ 20% and  $\geq$ 40%, respectfully, which is considered unacceptable.

An interaction between pyroxasulfone rate and application timing occurred for shoot density in both 2015 and 2016 (Table 3.2). In 2015, the shoot density ranged from 99 to 107% of the nontreated control with either rate of pyroxasulfone applied to 2- and 3-lf rice. In 2016, shoot density was  $\leq$  80% of the nontreated control following any treatment, which suggests a reduction in shoot density from pyroxasulfone compared to nontreated rice at all rates and timings. The lower shoot densities observed in 2016 than in 2015 can be attributed to the increased rainfall mentioned previously. In both years, the height of rice was reduced by pyroxasulfone, at all timings and rates. Additionally, there was a delay to 50% heading of  $\geq$ 8 days for all treatments in both years (Table 3.3). Furthermore, pyroxasulfone had a negative impact on rough rice yield. Yield for all pyroxasulfone-treated plots was  $\leq$ 75% of the nontreated control in both years evaluated.

Pyroxasulfone at any rate or application timing in both 2015 and 2016 had a negative effect on rice yield, days to 50% heading, shoot density, and percent visible injury. The negative impact associated with the use of pyroxasulfone on the majority of the parameters assessed, with an emphasis on yield, demonstrate that pyroxasulfone is not safe when applied to rice at the rates and application timings evaluated.

*S*-metolachlor. *S*-metolachlor had a negative impact on most rice parameters assessed. Similar to pyroxasulfone, injury generally decreased as *S*-metolachlor was applied later in the growing season (Table 3.4). Injury in 2016 caused by *S*-metolachlor increased greatly from 2 WAT to 5 WAF for treatments receiving the high rate of *S*-metolachlor at all application timings. Any treatment of *S*-metolachlor at the high rate at 5 WAF caused  $\geq$ 16% visible injury.

When averaged across herbicide rates, the lowest shoot density as a percentage of the nontreated control occurred following the DPRE applications in 2015 (84%) and 2016 (2%)

(Table 3.4). Rice receiving 1- to 2- or 3- to 4-lf applications of *S*-metolachlor contained shoot densities equal to or greater than other application timings, ranging from 95 to 96% of the control in 2015 and 91 to 96% of the control in 2016. Height as a percentage of the nontreated control for all application timings in 2015 was similar to the DPRE timing when averaged over *S*-metolachlor rates (Table 3.4). In 2016, rice height was similar for all application timings when averaged over both herbicide rates, ranging from 89 to 95% of the nontreated control, except for rice receiving DPRE applications, which averaged 61% of the nontreated control.

Days to 50% heading of rice receiving DPRE applications was delayed 31 days in 2016; however, all other treatments reached 50% heading on dates comparable to the nontreated control (Table 3.5). This delay in heading is caused by the early-season injury from *S*-metolachlor applied DPRE.

Yield differed only as a function of *S*-metolachlor rate in 2015 (Table 3.5). Yield was significantly higher for rice receiving a low rate of *S*-metolachlor (97%) compared to the high rate (89%) as a percentage of the nontreated control yield of 7,670 kg ha<sup>-1</sup> (Table 3.5). In 2016, yield was 98% of the nontreated control or greater following the high rate of *S*-metolachlor to 4-lf rice and the high rate of *S*-metolachlor to 1-lf rice (98%). The lowest yields in 2016 as a percentage of the nontreated control occurred following both DPRE applications at 7% with the low rate, and 0% with the high rate of *S*-metolachlor.

These data confirm that rice treated with a high rate of *S*-metolachlor can reach yields equivalent to that of the nontreated control when treated at the 4-lf growth stage as seen in 2016; however, the visible injury of 40% associated with the same treatment 5 WAF would indicate that there is low potential for commercialization of this herbicide in rice. Similar results have been seen where the use of *S*-metolachlor in rice resulted in substantial injury with little

reduction in yield (Bararpour et al. 2012). *S*-metolachlor applied at 840 g ha<sup>-1</sup> and 1,400 g ha<sup>-1</sup> to 4-lf rice caused 23% and 35% injury five weeks after emergence, respectively, with no reduction in yield compared to the nontreated control (Bararpour et al. 2012).

In general, delayed application timing played an important role in minimizing percent injury, shoot density reduction, height reduction, delayed heading, and yield loss associated with the use of *S*-metolachlor in rice. Because of the high injury observed with *S*-metolachlor, regardless of application timing, it is unlikely that this herbicide has a fit in dry-seeded rice. **Pethoxamid.** Little to no injury or reduction in yield occurred following later treatments in both years, making pethoxamid one of the most promising VLCFA-inhibiting herbicides assessed for use in rice. In 2016, 100% injury was observed 2 WAT following both DPRE applications; however, injury ratings of 9, 13, 16% were observed for rice after receiving the high rate of pethoxamid at 4-lf, low rate of pethoxamid at 1-lf, and high rate of pethoxamid at 1-lf, respectively (Table 3.6). Complete control of rice with the DPRE application is actually a positive aspect of this herbicide because the results would indicate that later application timings for which tolerance was observed should result in a high degree of weedy rice control if plants have not germinated prior to treatment. At 5 WAF, no more than 3% injury to rice was observed in 2015 (Table 3.6).

Shoot density as a percentage of the nontreated control in 2015 increased with time between planting and herbicide application from 89% and 84% following the DPRE and spiking treatments to 103% and 110% following the 2- and 3--If treatments, respectively, when averaged over both rates (Table 3.6). Similarly, in 2016, rice shoot density was greater than 96% of the nontreated control following both rates of pethoxamid applied at 1- and 4-lf, and the low rate of pethoxamid applied at spiking. Yield was equal to or greater than the nontreated control for all

the rate and timing main effects in 2015 (Table 3.7). Yield decreased in 2016 following the DPRE and spiking treatments when averaged over both rates of pethoxamid compared to the 1-and 4-lf applications, which yielded 103% and 99% of the nontreated control, respectively.

Considering these data, rice displays adequate tolerance to pethoxamid at the two rates evaluated when applied at the 1- to 2- or 3- to 4-lf stage. Pethoxamid caused no negative affect on yield, shoot density, height, or maturity when applied at the 1- to 2- or 3- to 4-lf rice growth stage (Tables 3.6 and 3.7). Rice was sometimes negatively impacted by earlier applications of pethoxamid. While in the drier year of 2015, DPRE and spiking applications generally did not deleteriously affect rice whereas the same timings caused unacceptable damage to rice in the wetter 2016. One concern with these early applications, especially a DPRE application is that the total loss of rice as observed in 2016 would prevent replanting of the crop because pethoxamid would already be present in soil. For this reason, it is imperative that an adequate stand be established prior to applying pethoxamid if registered in rice.

Acetochlor. Similar to pethoxamid, several treatments of the microencapsulated acetochlor formulation caused little negative impact to rice (Tables 3.8 and 3.9). No visible injury greater than 3% was observed for any treatment at any rating in 2015. In 2016, <10% injury was seen 2 WAT following the low rate of acetochlor at 1-lf (4%) and at 4-lf (1%), and high rate of acetochlor at 4-lf (3%) stages (Table 3.8). At 2 WAT, injury ranging from 34 to 89% occurred following the low and high rate of acetochlor applied DPRE and spiking in 2016.

Shoot density in 2015 differed as a function of application timing averaged over acetochlor rate as the 2-lf and 3-lf timings resulted in the highest shoot density at 95% of the nontreated control (Table 3.8). In 2016, the highest rice shoot density occurred following the 4-lf applications at both the high (96%) and low (92%) rates. Rice shoot density in 2016 often

decreased as acetochlor was applied earlier in the growing season. This damage from the DPRE and spiking applications translated to delayed maturity of 8 days compared to nontreated control (Table 3.9). The high rate of acetochlor applied to 1--lf rice also caused a 6-day delay in heading compared to the nontreated control; however, no delay in heading was seen for any other treatment.

Neither a main effect nor an interaction for acetochlor rate or application timing on rough rice yield occurred in 2015, indicating that the herbicide had no negative affect on yield (Table 3.9). In 2016, rough rice yield for all 1- to 2- and 3- to 4-lf applications was at least 97% of the nontreated control. Considering the parameters assessed for acetochlor applied at 630 and 1,050 g ha<sup>-1</sup>, the microencapsulated formulation appears to have great potential for use in rice from the 1- to 4-lf stage based on the tolerance exhibited in this research.

**Conclusions.** This research leads to the conclusion that pethoxamid and acetochlor should be further examined for use in rice. The manufacturer of pethoxamid is currently pursuing registration of the herbicide in U.S. rice, and based on this research, the best application timing to ensure adequate tolerance appears to begin no earlier than the 1-lf stage of rice. As observed in other research (Dhareesank et al. 2006), the rice injury is likely to be a function of soil moisture, with greater risk for injury when conditions are moist at application or immediately following application. It is also concluded that *S*-metolachlor and pyroxasulfone, at least at the rates tested, are injurious to rice, regardless of application timing, and should not be further pursued, especially considering the crop tolerance demonstrated with pethoxamid and acetochlor, two herbicides having a similar SOA.

## **Literature Cited**

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		D	ates of signi	ficance	
Year	Planting	DPRE	Spiking	1- to 2-lf	3- to 4-lf
2015	May 5	May 11	May 13	May 18 (2-lf)	May 27 (3-lf)
2016	April 24	April 29	May 3	May 12 (1-lf)	June 1 (4-lf)

Table 3.1. Planting and herbicide application dates for trials in 2015 and 2016.<sup>a,b</sup>

<sup>a</sup> Abbreviations: DPRE, delayed preemergence; lf, leaf.

					Inj	ury											
			2 V	VAT		-	5 W	ΆF			Shoo	t dens	ity		Hei	ght	
Factor		20	15	20	16	20	)15	20	16	20	15	20	16	20	15	20	)16
						%						9	6 of no	ntreated	1		
Rate <sup>e</sup>																	
	Low	16		29		52		52		91		57		51	a*	80	
]	High	27		35		88		71		90		42		43	b*	54	
Timing	C																
-	DPRE	44		84		87		87		104		16		47	ab*	77	
	Spiking	30		18		82		65		103		41		46	b*	66	
	1-2 LF	8		16		60		48		79		60		53	a*	69	
	3-4 LF	4		10		49		44		74		81		42	b*	54	
Rate $\times$																	
Timing																	
	$Low \times DPRE$	33	b	68	b	80	bc	79	b	84	b*	31	c*	49		94	a*
]	Low × Spiking	20	с	19	с	73	с	45	cd	78	bc*	56	b*	50		81	b*
]	$Low \times 1-2$ lf	8	d	23	с	20	e	43	d	99	а	60	b*	59		83	b*
J	$Low \times 3-4 lf$	4	d	9	c	34	d	40	d	102	a	83	a*	46		60	c*
l	High  imes DPRE	55	a	100	а	95	a	96	a	75	bc*	1	d*	44		60	c*
l	High × Spiking	40	b	18	c	93	a	85	b	71	c*	26	c*	42		51	d*
]	High× 1-2 lf	9	d	10	c	78	bc	46	cd	107	a	60	b*	46		55	cd*
]	High $\times$ 3-4 lf	5	d	11	с	86	ab	55	с	106	а	80	a*	38		48	d*
P Values fr	rom ANOVA																
Rate (P)		< 0.0	0001	Ν	S	<0.	0001	<0.	0001	N	S	<0.0	0001	0.0	261	<0.	0001
Timing (P)	)	< 0.0	0001	< 0.0			0001	<0.	0001	<0.0		<0.0	0001		010		0001
Rate × tim	ing(P)	0.0	029	0.00	)18	<0.	0001	0.0	)005	0.0	447	0.0	002	Ν	S	0.0	0007

Table 3.2. Effect of pyroxasulfone rate and application timing on rice injury, shoot density, and height in 2015 and 2016.<sup>a,b,c,d</sup>

Table 3.2 Cont.

<sup>a</sup> Abbreviations: WAT, weeks after treatment; WAF, weeks after rice flooding; DPRE, delayed preemergence; lf, leaf; NS, not significant.

<sup>b</sup> Means within a column followed by the same lowercase letter are not different according to Fisher's protected LSD ( $\alpha = 0.05$ ).

<sup>c</sup> Letters followed by asterisks (\*) for height and yield denote a statistical decrease from the nontreated control based on the confidence intervals obtained for each interaction or main effect.

<sup>d</sup> Average shoot density and height for nontreated plots were 49 and 47 shoots m<sup>-1</sup> of row and 80 cm and 95 cm in 2015 and 2016, respectively.

<sup>e</sup> The high and low rate applied for pyroxasulfone was 150 and 90 g ha<sup>-1</sup>, respectively.

		50% Hea	ding delay	Yie	ld		
Factor		2015	2016	2015	2016		
		days after	nontreated	% of nontreated			
Rate <sup>e</sup>							
	Low	16	10	33 a*	61		
	High	30	14	13 b*	31		
Timing	U						
C	DPRE	25	10	11 c*	25		
	Spiking	24	12	19 bc*	35		
	1-2 lf	19	12	37 a*	68		
	3-4 lf	25	14	24 b*	56		
Rate $\times$ Timing							
	$Low \times DPRE$	17 c	8 e	20	47 c*		
	Low × Spiking	16 c	11 d	31	54 bc*		
	$Low \times 1-2 lf$	13 d	12 cd	47	75 a*		
	$Low \times 3-4 lf$	17 c	11 d	33	68 ab*		
	High  imes DPRE	32 a	13 bc	3	3 d*		
	High × Spiking	32 a	14 b	6	16 d*		
	High× 1-2 lf	25 b	13 bc	26	61 b*		
	High $\times$ 3-4 lf	32 a	18 a	18	45 c*		
P Values from ANO	VA						
Rate (P)		< 0.0001	< 0.0001	< 0.0001	< 0.0001		
Timing $(P)$		< 0.0001	< 0.0001	< 0.0001	< 0.0001		
Rate $\times$ timing (P)		< 0.0001	0.0002	NS	0.0186		

Table 3.3. Effect of pyroxasulfone rate and application timing on rice heading date and yield in 2015 and 2016.<sup>a,b,c,d</sup>

<sup>a</sup> Abbreviations: DPRE, delayed preemergence; lf, leaf; NS, not significant.

<sup>b</sup> Means within a column followed by the same lowercase letter are not different according to Fisher's protected LSD ( $\alpha = 0.05$ ).

<sup>c</sup> Letters followed by asterisks (\*) for height and yield denote a statistical decrease from the nontreated control based on the confidence intervals obtained for each interaction or main effect.

<sup>d</sup> Average days to 50% heading and rough rice yield for the nontreated control was 87 and 89 days and 7,240 and 6,290 kg ha<sup>-1</sup> in 2015 and 2016, respectively.

<sup>e</sup> The high and low rate applied for pyroxasulfone was 150 and 90 g ha<sup>-1</sup>, respectively.

			Inj	ury					
		2 W	'AT	5 W	VAF	Shoot	density	Hei	ght
Factor		2015	2016	2015	2016	2015	2016	2015	2016
			9	6			% of no	ontreated	
Rate <sup>e</sup>									
	Low	8	41	8 b	40	91	64	93	88
	High	13	37	20 a	66	97	59	91	82
Timing	U								
e	DPRE	23	100	12	100	84 b*	2 c*	94 ab*	61 b*
	Spiking	16	30	16	49	100 a	57 b*	91 b*	95 a
	1-2 LF	2	16	14	36	96 a	91 a*	89 b*	94 a
	3-4 LF	1	9	14	28	95 a	96 a	98 a	89 a*
Rate $\times$									
Timing									
-	$Low \times DPRE$	16 b	100 a	8	99 a	88	5	95	65
	Low × Spiking	10 c	31 b	8	28 e	97	59	95	95
	$Low \times 1-2 lf$	1 d	25 b	6	19 f	92	98	87	97
	$Low \times 3-4 lf$	3 d	7 c	11	16 f	85	96	95	94
	$High \times DPRE$	30 a	100 a	16	100 a	80	0	93	58
	High × Spiking	21 b	29 b	25	70 b	103	55	86	95
	High× 1-2 lf	3 d	6 c	23	53 c	99	85	83	91
	High $\times$ 3-4 lf	0 d	11 c	18	40 d	104	97	101	84
P Values fro	om ANOVA								
Rate (P)		0.0005	0.0207	< 0.0001	< 0.0001	NS	NS	NS	NS
Timing (P)		< 0.0001	< 0.0001	NS	< 0.0001	0.0150	< 0.0001	0.0109	< 0.000
Rate × timir	ng $(P)$	0.0015	0.0005	NS	< 0.0001	NS	NS	NS	NS

Table 3.4. Effect of *S*-metolachlor rate and application timing on rice injury, shoot density, and height in 2015 and 2016.<sup>a,b,c,d</sup>

Table 3.4 Cont.

<sup>a</sup> Abbreviations: WAT, weeks after treatment; WAF, weeks after rice flooding; DPRE, delayed preemergence; lf, leaf; NS, not significant.

<sup>b</sup> Means within a column followed by the same lowercase letter are not different according to Fisher's protected LSD ( $\alpha = 0.05$ ).

<sup>c</sup> Letters followed by asterisks (\*) for height and yield denote a statistical decrease from the nontreated control based on the confidence intervals obtained for each interaction or main effect.

<sup>d</sup> Average shoot density and height for nontreated plots were 55 and 50 shoots m<sup>-1</sup> of row and 83 cm and 88 cm in 2015 and 2016, respectively.

<sup>e</sup> The high and low rate applied for *S*-metolachlor was 1070 and 535 g ha<sup>-1</sup>, respectively.

		50% He	ading delay	Yi	eld
Factor		2015	2016	2015	2016
		days afte	er nontreated	——% of n	ontreated—
Rate <sup>f</sup>					
	Low	0	8	97 a	63
	High	1	8	89 b*	70
Timing					
	DPRE	0	31	92	3
	Spiking	2	0	92	76
	1-2 LF	1	0	90	91
	3-4 LF	0	0	97	96
Rate $\times$ Timing					
	$Low \times DPRE$	0	31	100	7 d*
	Low × Spiking	0	0	97	68 c*
	$Low \times 1-2 lf$	0	0	94	85 b*
	$Low \times 3-4 lf$	0	0	96	93 ab
	$High \times DPRE$	0	31	84	0 d*
	High × Spiking	1	0	87	84 b*
	High× 1-2 lf	4	0	86	98 a
	High $\times$ 3-4 lf	0	0	99	100 a
P Values from ANO	VA				
Rate (P)				0.0072	0.0137
Timing $(P)$				NS	< 0.0001
Rate $\times$ timing (P)				NS	0.0329

Table 3.5. Effect of *S*-metolachlor rate and application timing on rice heading date and yield in 2015 and 2016.<sup>a,b,c,d,e</sup>

<sup>a</sup> Abbreviations: DPRE, delayed preemergence; lf, leaf; NS, not significant.

<sup>b</sup> Means within a column followed by the same lowercase letter are not different according to Fisher's protected LSD ( $\alpha = 0.05$ ).

<sup>c</sup> Letters followed by asterisks (\*) for height and yield denote a statistical decrease from the nontreated control based on the confidence intervals obtained for each interaction or main effect.

<sup>d</sup> Average days to 50% heading and rough rice yield for the nontreated control was 85 and 88 days and 7,670 and 7,130 kg ha<sup>-1</sup> in 2015 and 2016, respectively

<sup>e</sup> 50% heading delay data were not amenable to formal statistical analysis.

<sup>f</sup> The high and low rate applied for *S*-metolachlor was 1070 and 560 g ha<sup>-1</sup>, respectively.

				Injury	y									
		2 V	VAT		5	WAF		S	hoot d	lensity		H	eight	
Factor		2015	201	16	2015	201	6	20	15	201		2015	20	16
				%-						%	of no	ontreated—		
Rate <sup>e</sup>														
	Low	6	50		2	36		106	а	78		95	91	
	High	8	53		1	43		88	b*	56		93	90	
Timing	-													
_	DPRE	10 a	100		3	98		89	b*	1		97	68	b*
	Spiking	9 a	76		1	41		84	b*	57		93	99	a
	1-2 LF	4 b	15		0	7		103	а	102		96	96	a
	3-4 LF	4 b	15		3	13		110	а	108		90	98	a
Rate $\times$														
Timing														
	$Low \times DPRE$	9	100	a	3	97	а	104		1	c*	103	71	
	Low × Spiking	6	64	c	1	26	c	97		96	a	98	101	
	$Low \times 1-2$ lf	2	13	de	0	4	e	111		108	а	89	93	
	$Low \times 3-4 lf$	5	21	d	3	16	d	110		108	а	89	99	
	High  imes DPRE	11	100	a	3	98	a	75		1	c*	91	67	
	High × Spiking	11	89	b	0	56	b	71		19	b*	88	98	
	High× 1-2 lf	5	16	de	0	9	de	95		96	a	103	99	
	High $\times$ 3-4 lf	3	9	e	3	10	de	110		108	а	91	98	
P Values	s from ANOVA													
Rate $(P)$		NS	NS	5	NS	0.00	18	0.00	002	< 0.0	001	NS	N	S
Timing (		0.0069	< 0.00	001	NS	< 0.0	001	0.00	)05	< 0.0	001	NS	< 0.0	001
U	Timing (P)	NS	< 0.00	001	NS	< 0.0	001	N	S	< 0.0	001	NS	N	S

Table 3.6. Effect of pethoxamid rate and application timing on rice injury, shoot density, and height in 2015 and 2016.<sup>a,b,c,d</sup>

Table 3.6 Cont.

<sup>a</sup> Abbreviations: WAT, weeks after treatment; WAF, weeks after rice flooding; DPRE, delayed preemergence; lf, leaf; NS, not significant.

<sup>b</sup> Means within a column followed by the same lowercase letter are not different according to Fisher's protected LSD ( $\alpha = 0.05$ ).

<sup>c</sup> Letters followed by asterisks (\*) for height and yield denote a statistical decrease from the nontreated control based on the confidence intervals obtained for each interaction or main effect.

<sup>d</sup> Average shoot density and height for nontreated plots were 46 and 42 shoots m<sup>-1</sup> of row and 83 cm and 91 cm in 2015 and 2016, respectively.

<sup>e</sup> The high and low rate applied for pethoxamid was 840 and 420 g ha<sup>-1</sup>, respectively.

		50% Hea	ading delay	Y	ïeld
Factor		2015	2016	2015	2016
		days after	r nontreated	% of n	ontreated-
Rate <sup>f</sup>					
	Low	0	1	114 a	79 a*
	High	0	1	109 b	70 b*
Timing					
	DPRE	0	1	113 a	10 c*
	Spiking	0	0	101 b	89 b*
	1-2 LF	0	0	113 a	103 a
	3-4 LF	0	0	118 a	99 a
Rate × Timing					
	$Low \times DPRE$	0	1	113	20
	Low × Spiking	0	0	106	96
	$Low \times 1-2 lf$	0	1	119	105
	$Low \times 3-4 lf$	0	1	119	96
	High  imes DPRE	0	1	112	0
	High × Spiking	0	1	97	78
	High× 1-2 lf	0	0	107	101
	$High \times 3-4 lf$	0	0	118	102
P Values from ANO	VA				
Rate (P)				0.0424	0.0209
Timing (P)				0.0009	< 0.0001
Rate $\times$ Timing (P)				NS	NS

Table 3.7. Effect of pethoxamid rate and application timing on rice heading date and yield in 2015 and 2016.<sup>a,b,c,d,e</sup>

<sup>a</sup> Abbreviations: DPRE, delayed preemergence; lf, leaf; NS, not significant.

<sup>b</sup> Means within a column followed by the same lowercase letter are not different according to Fisher's protected LSD ( $\alpha = 0.05$ ).

<sup>c</sup> Letters followed by asterisks (\*) for height and yield denote a statistical decrease from the nontreated control based on the confidence intervals obtained for each interaction or main effect.

<sup>d</sup> Average days to 50% heading and rough rice yield for the nontreated control was 88 and 90 days and 7,430 and 6,700 kg ha<sup>-1</sup> in 2015 and 2016, respectively.

<sup>e</sup> The 50% heading delay data were not amendable to formal statistical analysis.

<sup>f</sup> The high and low rate applied for pethoxamid was 840 and 420 g ha<sup>-1</sup>, respectively.

			Inju	ıry						
		2 W	VAT	5	WAF	Shoot o	lensity	He	ight	
Factor		2015	2016	2015	2016	2015	2016	2015	2016	
		%%					——% of no	ontreated		
Rate <sup>f</sup>										
Rate	Low	1	27	0	9	94	68	93 b*	98 b*	
	High	1	36	0	35	92	47	101 a	100 a	
Timing	mgn	1	50	Ū	55	2	• /	101 u	100 u	
8	DPRE	2	79	0	51	90 b*	33	96	99	
	Spiking	1	38	0	26	91 b*	31	101	100	
	1-2 lf	0	7	0	7	95 a*	73	98	100	
	3-4 lf	0	2	0	4	95 a*	94	94	98	
Rate $\times$										
Timing										
U	$Low \times DPRE$	3	69 b	0	18 c	93	56 c*	91	98	
	Low × Spiking	1	34 d	0	13 cd	92	52 c*	96	99	
	$Low \times 1-2 lf$	0	4 f	0	3 e	96	71 b*	94	99	
	$Low \times 3-4 lf$	0	1 f	0	5 e	94	92 a*	92	97	
	$\operatorname{High}  imes \operatorname{DPRE}$	1	89 a	0	85 a	87	10 d*	100	100	
	High  imes Spiking	1	43 c	0	40 b	89	10 d*	107	100	
	High× 1-2 lf	0	10 e	0	7 de	94	74 b*	102	100	
	High $\times$ 3-4 lf	0	3 f	0	9 de	96	96 a	98	99	
P Values	from ANOVA									
Rate (P)			< 0.0001		< 0.0001	NS	< 0.0001	0.0047	0.0069	
Timing (P)			< 0.0001		< 0.0001	0.0123	< 0.0001	NS	NS	
Rate × Ti			0.0019		< 0.0001	NS	< 0.0001	NS	NS	

Table 3.8. Effect of acetochlor rate and application timing on rice injury, shoot density, and height in 2015 and 2016.<sup>a,b,c,d,f</sup>

Table 3.8 Cont.

<sup>a</sup> Abbreviations: WAT, weeks after treatment; WAF, weeks after rice flooding; DPRE, delayed preemergence; lf, leaf; NS, not significant.

<sup>b</sup> Means within a column followed by the same lowercase letter are not different according to Fisher's protected LSD ( $\alpha = 0.05$ ).

<sup>c</sup> Letters followed by asterisks (\*) for height and yield denote a statistical decrease from the nontreated control based on the confidence intervals obtained for each interaction or main effect.

<sup>d</sup> Average shoot density and height for nontreated plots were 49 and 50 shoots m<sup>-1</sup> of row and 86 cm and 95 cm in 2015 and 2016, respectively.

<sup>e</sup> The 2015 2 WAT and 5 WAF injury data were not amenable to formal statistical analysis.

<sup>f</sup>The high and low rate applied for acetochlor was 1,050 and 630 g ha<sup>-1</sup>, respectively.

		50% Hea	ading delay	Yield		
Factor		2015	2016	2015	2016	
		days after	r nontreated	% of nontreated		
Rate <sup>f</sup>						
	Low	0	4	97	93	
	High	0	6	97	85	
Timing	-					
-	DPRE	0	8	98	73	
	Spiking	1	8	96	84	
	1-2 lf	0	3	97	98	
	3-4 lf	0	0	97	101	
Rate $\times$ Timing						
	$Low \times DPRE$	0	8 a	98	88 bc*	
	Low × Spiking	1	8 a	95	82 c*	
	$Low \times 1-2 lf$	0	0 c	98	99 ab	
	$Low \times 3-4 lf$	0	0 c	97	104 a	
	$High \times DPRE$	1	8 a	99	58 d*	
	High × Spiking	0	8 a	96	86 bc*	
	High× 1-2 lf	0	6 b	97	97 ab	
	High $\times$ 3-4 lf	0	0 c	96	99 ab	
<i>P</i> Values from ANO	VA					
Rate (P)			< 0.0001	NS	0.0214	
Timing (P)			< 0.0001	NS	< 0.0001	
Rate $\times$ Timing (P)			< 0.0001	NS	0.0095	

Table 3.9. Effect of acetochlor rate and application timing on rice heading date and yield in 2015 and 2016.<sup>a,b,c,d,e</sup>

<sup>a</sup> Abbreviations: DPRE, delayed preemergence; lf, leaf; NS, not significant.

<sup>b</sup> Means within a column followed by the same lowercase letter are not different according to Fisher's protected LSD ( $\alpha = 0.05$ ).

<sup>c</sup> Letters followed by asterisks (\*) for height and yield denote a statistical decrease from the nontreated control based on the confidence intervals obtained for each interaction or main effect. <sup>d</sup> Average days to 50% heading and rough rice yield for the nontreated control was 86 and 91 days and 7,150 and 6,380 kg ha<sup>-1</sup> in 2015 and 2016, respectively.

<sup>e</sup> The 2015 50% heading delay data were not amenable to formal statistical analysis.

<sup>f</sup> The high and low rate applied for acetochlor was 1,050 and 630 g ha<sup>-1</sup>, respectively.

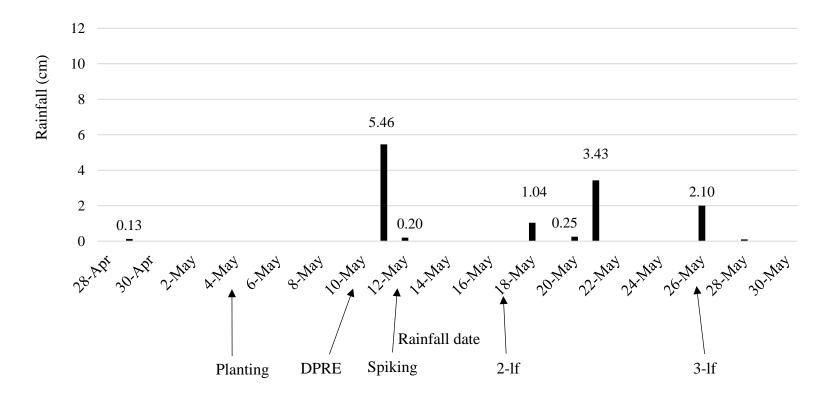


Figure 3.1. Rice Research and Extension Center (RREC) rainfall amount and dates in 2015.

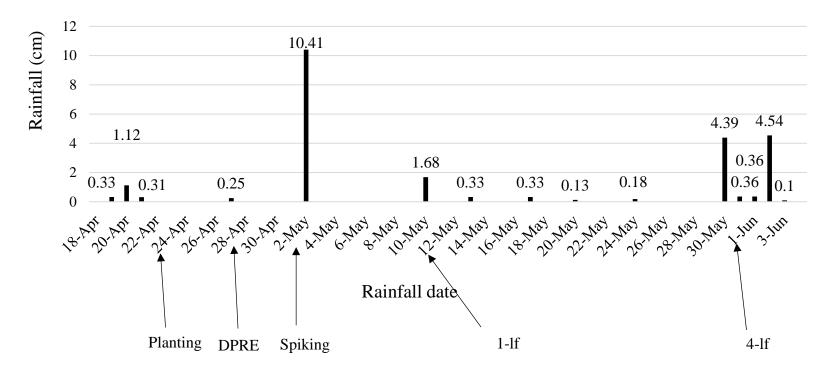


Figure 3.2. Rice Research and Extension Center (RREC) rainfall amount and dates in 2016.

#### **Chapter 4**

### **Evaluation of Pethoxamid-Containing Weed Control Programs in Arkansas Rice**

Herbicide resistance to several of the most common weed species in U.S. rice production, such as barnyardgrass and red rice, has made weed control extremely difficult with current herbicide options. Currently, no very-long-chain fatty acid)-inhibiting herbicides are labeled for use in U.S. rice; however, pethoxamid is one such herbicide currently under development for soil-applied use to control grasses and small-seeded broadleaves in rice and various row crops. Field trials were conducted in 2015 and 2016 near Stuttgart, AR, for rice tolerance and in 2016 near Colt, AR, and Lonoke, AR, for weed control with the use of pethoxamid-containing rice herbicide programs. Pethoxamid was applied alone and in a program at 420 and 560 g ai ha<sup>-1</sup> with other herbicides labeled in rice including clomazone, quinclorac, propanil, imazethapyr, and carfentrazone postemergence. Injury less than 10% was seen for all treatments 2 weeks after treatment in 2015 and 2016, except for pethoxamid at 420 g ha<sup>-1</sup> + clomazone to 1-leaf rice. Rice injury dissipated to less than 5% following all treatments by 4 weeks after flood establishment. Barnyardgrass was controlled  $\geq$ 95% near Colt and  $\geq$ 93% near Lonoke for herbicide programs including clomazone preemergence followed by pethoxamid + quinclorac or imazethapyr at 3- to 4-leaf rice. Considering the minimal injury and high levels of barnyardgrass control associated with pethoxamid-containing weed control programs, pethoxamid provides a unique and effective site of action for use in U.S. rice production.

Nomenclature: Carfentrazone; clomazone; imazethapyr; pethoxamid; propanil; quinclorac; barnyardgrass, *Echinochloa crus-galli* (L.) Beauv.; red rice *Oryza sativa* var. *sylvatica* L.; rice, *Oryza sativa* L.

**Key words:** Barnyardgrass control, herbicide resistance, herbicide site of action, rice injury, rice production

### Introduction

Pethoxamid (FMC Corporation, Philadelphia, PA) is a very-long-chain fatty acid (VLCFA)-inhibiting herbicide, which belongs to the chloroacetamide family. Pethoxamid is currently under development in the U.S. for use in canola (*Brassica napus* L.), corn (*Zea mays* L.), cotton (*Gossypium hirsutum* L.), rice, soybean (*Glycine max* (L.) Merr.), and sunflower (*Helianthus annuus* L.). Currently there are no VLCFA-inhibiting herbicides labeled for use in U.S. rice production; therefore, pethoxamid would present a unique site of action (SOA) to combat herbicide-resistant weeds in rice.

Pethoxamid is currently used as a PRE herbicide for European corn and soybean (Schlosser et al. 2016) and is a soil-applied, systemic herbicide with activity on many annual grasses and small-seeded broadleaves including barnyardgrass and red rice (Anonymous 2016). Pethoxamid, like other VLCFA-inhibiting herbicides, is a root and shoot inhibitor; therefore, it controls germinating seeds (Anonymous 2015). Being a chloroacetamide, pethoxamid has similar activity to commonly used herbicides in U.S. row crops such as acetochlor, alachlor, and metolachlor.

Previous research has shown that the overreliance of one particular herbicide or herbicide SOA is the leading cause for the evolution of herbicide resistance (Norsworthy et al. 2012). This has led to the evolution of barnyardgrass resistance to several of the most commonly used herbicides in rice. In a survey of crop consultants in Arkansas and Mississippi, clomazone was listed as the most often recommended PRE herbicide by 91% of respondents (Norsworthy et al. 2012). The most commonly used POST herbicides included imazethapyr, quinclorac, fenoxaprop, and propanil (Norsworthy et al. 2012). Barnyardgrass resistance has been confirmed by the University of Arkansas resistance screening program to propanil, quinclorac,

imazethapyr, penoxsulam, bispyribac, imazamox, and clomazone; and fenoxaprop and cyhalofop-resistant barnyardgrass populations have been confirmed in Mississippi (Heap 2017; Miller et al. 2015).

Targeting the most resistant-prone weeds by rotation, mixing, and sequential applications of differing herbicide SOA can delay the onset of resistance by reducing selection pressure (Norsworthy et al. 2012). Although yearly herbicide rotations and sequential applications of herbicides with different SOA can reduce selection on weed species compared to a monoherbicidal program, there is a possibility that multiple resistance may be selected. Tank mixing herbicides with multiple SOA applies simultaneous selection, which reduces the risk for herbicide resistance evolution (Norsworthy et al. 2012).

Herbicide SOA sequences influence the evolution of herbicide resistance. Research was conducted on three different application sequences including alternating herbicide SOA by year, changing herbicide SOA when the initial SOA became ineffective, and tank-mixing two herbicide SOA (Diggle et al. 2003). The only sequence where herbicide resistance did not occur was the application of herbicides with differing SOA in tank mixtures (Diggle et al. 2003). The integration of a new herbicide SOA in rice production broadens the grower's ability to tank-mix pethoxamid with other rice herbicides in order to combat herbicide-resistant weed species.

VLCFA-inhibiting herbicides are not currently labeled in U.S. rice, but have been used with success in U.S. row crops such as corn, cotton, and soybean for the control of red rice and barnyardgrass. Studies were conducted from 1992 to 1994 evaluating the use of metolachlor and alachlor preplant incorporated (PPI), PRE, and POST in soybean (Noldin et al. 1998). Results showed that alachlor provided late-season, red rice control from 85 to 91% at 4.5 kg ai ha<sup>-1</sup> when applied PPI. Metolachlor also provided adequate late-season red rice control from 90 to 92% at

3.4 kg ai ha<sup>-1</sup> when applied PPI. In general, barnyardgrass control was lower than red rice control for all herbicides evaluated; however, late-season barnyardgrass control of up to 84 and 89% following applications of alachlor and metolachlor, respectively, was observed (Noldin et al. 1998).

VLCFA-inhibiting herbicides have also been used in Asian dry- and wet-seeded rice culture as both PRE and POST herbicides. Herbicides such as pretilachlor and butachlor are the most commonly used VLCFA inhibitors in Asian rice production. Butachlor alone was shown to control barnyardgrass up to 59% when applied 1, 3, 5, and 7 days after broadcasting sprouted rice seed in India (Mutnal et al. 1998). In a separate study conducted in Sri Lanka, pretilachlor + pyribenzoxim followed by (fb) MCPA was applied to rice 8 days after sowing in a dry-seeded system. Pretilachlor + pyribenzoxim fb MCPA reduced the weed densities of barnyardgrass, Chinese sprangletop (*Leptochloa chinensis* L.), and knotgrass (*Paspalim distichum* L.) by 99%, which was the highest of any treatment evaluated (Chauhan et al. 2012).

Some preliminary research has been conducted on the tolerance of rice to applications of pethoxamid. In 2015, pethoxamid was applied to rice at the delayed preemergence (DPRE), spiking, and 1- to 2-leaf rice stages. Rice treated with pethoxamid showed no more than 5% injury and there was no reduction in yield when compared to the nontreated control (Godwin et al. 2016). In 2015, pethoxamid was also evaluated for rice tolerance and weed control at 420 and 560 g ai ha<sup>-1</sup> when applied to spiking rice alone and in combination with other commonly use rice herbicides including: clomazone, imazethapyr, pendimethalin, and quinclorac. Rice was tolerant to all treatments and weed control was adequate (Doherty et al. 2016). At 66 days after application, pethoxamid at 560 g ha<sup>-1</sup> alone provided 80% barnyardgrass control and 86% Amazon sprangletop (*Leptichloa panicoides* J Presl) control. The highest level of weed control

observed consisted of 96% control of barnyardgrass and 98% control of Amazon sprangletop with pethoxamid at 560 g ha<sup>-1</sup> in combination with imazethapyr (Doherty et al. 2016).

Many of the most commonly used herbicides in U.S. rice production today are those that inhibit acetolactate synthase (ALS) and acetyl CoA carboxylase (ACCase). The ALS-inhibiting herbicides, such as imazethapyr, imazamox, and bispyribac, have confirmed resistance from 51 different weed species in the U.S. alone. Likewise, ACCase-inhibiting herbicides, such as cyhalofop and fenoxaprop, have confirmed resistance from 15 weed species in the U.S. (Heap 2017). Low risk for the evolution of weed resistance is associated with VLCFA-inhibiting herbicides compared to many commonly used rice herbicides. VLCFA-inhibiting herbicides have only five confirmed resistant species worldwide, and only one in the U.S., that being Italian ryegrass (*Lolium multiflorum* Lam.) (Heap 2017).

Considering the tolerance of rice to pethoxamid, the adequate levels of barnyardgrass control with the use of pethoxamid, and the success of VLCFA-inhibiting herbicides in Asian rice and U.S. row crop production, it is believed that pethoxamid may provide an effective and alternative herbicide SOA for use in U.S. rice production. It was hypothesized that rice would display adequate tolerance to pethoxamid applied alone and in combination with other commonly used rice herbicides at various application timings, while providing an effective level of weed control. The objective of this research was to evaluate efficacy and rice tolerance following pethoxamid-containing weed control programs in drill-seeded rice.

### **Materials and Methods**

Field trials were conducted in 2015 and 2016 at the Rice Research and Extension Center (RREC) near Stuttgart, AR, and in 2016 at the Pine Tree Research Station (PTRS) near Colt, AR,

and the University of Arkansas Pine Bluff Lonoke Farm (UAPB) near Lonoke, AR. The soils at the RREC, PTRS, and UAPB were a Dewitt silt loam (fine, smectic, thermic typic albaqualf), Calloway silt loam (fine-silty, mixed, active, thermic aquic fraglossudalfs), and Immanuel silt loam (fine-silty, mixed, active, thermic oxyaquic glossudalfs), respectively. Imidazolinone-resistant (Clearfield<sup>TM</sup>, BASF Corporation, Research Triangle Park, NC) inbred rice cultivars CL111 (RREC) and CL151 (PTRS and UAPB) were drill-seeded into 1.8- by 5.2-m plots at a seeding rate of 72 seeds m<sup>-1</sup> of row, at a row width of 18 cm. Rice was planted on May 5, 2015, and April 29, 2016, at the RREC, May 9, 2016, at the PTRS, and May 18, 2016, at the UAPB. Rice fertility programs were based on University of Arkansas Division of Agriculture Research and Extension recommendations (Norman et al. 2013).

At each location the experiment was a randomized complete block design with four blocks. In total, twelve different herbicide treatments were evaluated along with a nontreated control. The herbicide programs included pethoxamid applied alone at 420 and 560 g ha<sup>-1</sup> to 1-lf rice, pethoxamid at 420 and 560 g ha<sup>-1</sup> + clomazone (Comand 3ME, FMC Corporation, Philadelphia, PA) at 336 g ai ha<sup>-1</sup> to 1-lf rice, and clomazone applied PRE at 336 g ha<sup>-1</sup> followed by (fb) pethoxamid at 420 and 560 g ha<sup>-1</sup> in combination with quinclorac (Facet L, BASF Corporation, Research Triangle Park, NC) at 420 g ai ha<sup>-1</sup>, propanil (STAM M4, RiceCo USA, Fair Oaks, CA) at 4.5 kg ai ha<sup>-1</sup>, imazethapyr (Newpath, BASF Corporation, Research Triangle Park, NC) at 71 g ai ha<sup>-1</sup>, or carfentrazone (AIM EC, FMC Corporation, Philadelphia, PA) at 18 g ai ha<sup>-1</sup> to 3- to 4-lf rice (Table 4.1). A nonionic surfactant (NIS) was used at 0.25% v/v in combination with the pethoxamid + quinclorac, pethoxamid + imazethapyr, and pethoxamid + carfentrazone applied with a CO<sub>2</sub>-pressurized

backpack sprayer through 110015 AIXR (TeeJet) nozzles calibrated to deliver 143 L ha<sup>-1</sup> using a three-nozzle boom at 51-cm nozzle spacing at 4.83 km h<sup>-1</sup>.

At the RREC, the PRE herbicide applications were applied on May 6, 2015, and April 29, 2016; the 1-lf rice applications were applied on May 19, 2015, and May 12, 2016; and the 3- to 4-lf rice applications were applied on May 27, 2015, and June 1, 2016. At the PTRS, the PRE applications were applied on May 10, 2016; the 1-lf rice applications were applied on May 25, 2016; and the 3- to 4-lf rice applications were applied on June 2, 2016. At the UAPB, the PRE applications were applied on May 19, 2016; the 1-lf rice applications were applied on May 31, 2016; and the 3- to 4-lf rice applications were applied on June 13, 2016.

PRE applications at the UAPB and the PTRS were made with no weeds present because preplant tillage was used to prepare the test site for planting. All 1-If herbicide applications were made with an average of 11 barnyardgrass plants m<sup>-2</sup> ranging from 2 to 5 cm at UAPB and 1 barnyardgrass plant m<sup>-2</sup> ranging from 2 to 4 cm at PTRS. The 3-If applications were applied to an average of two barnyardgrass plants per m<sup>-2</sup> ranging from 6 to 9 cm at UAPB and four barnyardgrass plants per m<sup>-2</sup> ranging from 5 to 8 cm at PTRS. Weed densities at the 1-If and 3-If applications are not comparable considering all herbicide programs contained a treatment prior to 3-If rice.

The trials located at the RREC were strictly rice tolerance trials, while the trials located at the PTRS and the UAPB assessed weed control. Data collection at the RREC included a visual assessment of crop injury every 2 weeks from the first application until physiological maturity. Visual injury was estimated on a scale of 0 to 100%, with 0 being no injury and 100 being complete crop death. Visible injury assessments included reduction in crop density, height, and overall plant vigor. Other parameters evaluated at the RREC included shoot density (number of

shoots m<sup>-1</sup> of row) 5 weeks after planting (WAP), the average of three crop canopy heights per plot (cm) 8 WAP, days to 50% heading relative to the nontreated control, and rough rice yield (kg ha<sup>-1</sup>) adjusted to 12.5% moisture. Parameters such as shoot density and crop height were normalized for each site year by converting them to a percentage of the nontreated control plots. A visual assessment of barnyardgrass control (0 to 100%) compared to the nontreated control was assessed every 2 weeks following the first herbicide application until physiological maturity at the PTRS and the UAPB.

Data were analyzed in JMP Pro 12 (SAS Institute Inc, Cary, NC). Data were analyzed separately by site-year using with block included as a random effect. The nontreated control was removed from the analysis for the rice tolerance and barnyardgrass control assessments. All means were separated using Fisher's protected LSD ( $\alpha$ =0.05). In order to determine the effect of pethoxamid rate, the use of a PRE herbicide, and the use of an additional herbicide with pethoxamid on barnyardgrass control, contrasts were conducted for the rate of pethoxamid applied (420 g ha<sup>-1</sup> vs. 560 g ha<sup>-1</sup>); the use of a PRE herbicide compared to no PRE; and pethoxamid + clomazone, pethoxamid + quinclorac, pethoxamid + propanil, pethoxamid + imazethapyr, or pethoxamid + carfentrazone compared to pethoxamid applied alone for barnyardgrass control 3 weeks after pethoxamid treatment (WAT) and 4 weeks after rice flooding (WAF) at the PTRS and the UAPB.

#### **Results and Discussion**

At the RREC, rice injury ratings were less than 10% for all treatments, except pethoxamid at 560 g ha<sup>-1</sup> + clomazone applied to 1-lf rice 2 WAT in both 2015 and 2016 (Table 4.1). However, by 4 WAF, rice injury had dissipated to less than 5% following all treatments

evaluated (data not shown). This indicates that pethoxamid can be safely applied at either 1-lf or 3- to 4-lf rice alone or in combination with other rice herbicides. These results coincide with previous research where a higher rate of pethoxamid applied at 840 g ha<sup>-1</sup> at delayed PRE (DPRE), spiking, and 1- to 2-lf rice caused less than 5% injury 3 WAT (Godwin et al. 2016).

Even though little visible injury was observed and no height reduction occurred for any treatment in 2015 or 2016 (data not shown), differences in shoot density occurred at the RREC in 2015 as a result of herbicide treatment. In 2015, clomazone fb pethoxamid at 420 g ha<sup>-1</sup> + carfentrazone or clomazone fb pethoxamid at 560 g ha<sup>-1</sup> + propanil had shoot densities less than the nontreated control (Table 4.1). Clomazone PRE fb pethoxamid + propanil had visual injury less than 5% following the same application in both 2015 and 2016. Even though a reduction in shoot density was observed, yield was not reduced (Table 4.2).

Barnyardgrass was controlled  $\geq$  97% up to 4 WAF at the PTRS with PRE-applied clomazone-containing herbicide programs that were fb pethoxamid + another herbicide to 3- to 4-lf rice (Table 4.3). At the UAPB, barnyardgrass was controlled  $\geq$  93% 3 WAT and 4 WAF following applications of clomazone PRE fb pethoxamid + quinclorac or imazethapyr. The reduction in control with propanil-containing programs was attributed to the population of barnyardgrass at the UAPB being confirmed resistant to propanil (J.K. Norsworthy, personal communication). Propanil resistance is a common problem in Arkansas considering from 2006 to 2012 nearly 50% of barnyardgrass samples submitted to the University of Arkansas herbicide resistance screening program were documented as resistant to propanil (Norsworthy et al. 2013b).

Contrasts revealed differences in barnyardgrass control between the rate of pethoxamid used, the use of pethoxamid in combination with other rice herbicides vs. the use of pethoxamid

alone, and the use of a PRE vs. the absence of a PRE (Table 4.4). At the UAPB, barnyardgrass control was improved with pethoxamid at 560 g ha<sup>-1</sup> compared to 420 g ha<sup>-1</sup> at both 3 WAT and 4 WAF (Table 4.4). Also, pethoxamid in combination with all herbicides evaluated, except propanil or carfentrazone at the UAPB, enhanced late-season barnyardgrass control 4 WAF (Table 4.4). Likewise, the use of a PRE herbicide resulted in higher levels of barnyardgrass control at the PTRS, 3 WAT and 4 WAF, when compared to treatments without a PRE. Hence, for season-long barnyardgrass control, it is imperative that pethoxamid be used in a herbicide program rather than applied alone to 1-lf rice.

Herbicide-based weed control approaches using multiple effective SOA have been documented to be more effective than relying upon a single herbicide SOA in rice (Wilson et al. 2010). At the UAPB, clomazone + quinclorac applied PRE fb imazethapyr EPOST fb imazethapyr + fenoxaprop preflood (PREFLD) provided 100% control of ALS-resistant barnyardgrass 10 wk after planting whereas two passes of imazethapyr resulted in only 44% barnyardgrass control (Wilson et al. 2010). Clomazone + quinclorac applied PRE fb imazethapyr early postemergence (EPOST) fb imazethapyr + fenoxaprop preflood (PREFLD) contains four different SOA, which are effective for barnyardgrass, outside of the realm of resistance, including a diterpene biosynthesis inhibitor (clomazone), a synthetic auxin (quinclorac), an ALS-inhibitor (imazethapyr), and an ACCase-inhibitor (fenoxaprop).

All treatments at the PTRS and UAPB containing clomazone PRE fb pethoxamid in a tank-mixture with another herbicide to 3- to 4-lf rice provided three different herbicide SOA, including a diterpene biosynthesis inhibitor (clomazone), a VLCFA-inhibitor (pethoxamid), and the corresponding SOA for the pethoxamid tank-mix partner. The three effective herbicide SOA at the PTRS resulted in an increase in weed control both 3 WAT and 4 WAF when compared to

pethoxamid applied alone (Table 4.3). Hence, pethoxamid in a program can provide residual control until permanent flood is established, resulting in season-long barnyardgrass control.

**Practical Implications.** The minimal rice injury or reduction in yield shows that rice can tolerate the use of pethoxamid POST. Chloroacetamides, such as pethoxamid, have activity on weeds prior to emergence and inhibit early seedling growth (Fuerst 1987); therefore, pethoxamid would only control weeds that germinate after the herbicide application. In order for pethoxamid to be used POST in rice, it is necessary to tank-mix pethoxamid with a rice herbicide that has activity on weeds that have already emerged. Chloroacetamides are considered relatively low risk herbicides for the evolution of weed resistance (Powles and Shaner 2001); therefore, pethoxamid used in a herbicide program offers an alternative and effective herbicide SOA to control many of the resistant weed species found in rice such as barnyardgrass. Considering the tolerance of rice and high levels of barnyardgrass control associated with the use of pethoxamid-containing rice herbicide programs, an early POST application of pethoxamid provides a unique SOA and aids residual weed control until the permanent rice flood is established.

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Program PRE POST herbicide						jury VAT		Shoot density			
		POST	Pethoxamid					~	1000000		
herbicide	1 0 0 1 1010100	timing	rate	2015 2			16	201	5	2016	
		8	g ai ha <sup>-1</sup>			-%				treated—	
Nontreated								100	abcd	100	
	Pethoxamid	1-lf	420	1	с	7	bc	99	abcd	89	
	Pethoxamid	1-lf	560	3	bc	5	cd	104	a	93	
	Pethoxamid + clomazone	1-lf	420	8	ab	8	b	102	abc	95	
	Pethoxamid + clomazone	1-lf	560	13	а	11	а	104	а	89	
Clomazone	Pethoxamid + quinclorac	3-4 lf	420	3	bc	1	e	87	ef	90	
Clomazone	Pethoxamid + quinclorac	3-4 lf	560	2	с	3	de	89	ef	89	
Clomazone	Pethoxamid + propanil	3-4 lf	420	5	bc	2	de	91	def	92	
Clomazone	Pethoxamid + propanil	3-4 lf	560	4	bc	3	de	75	g	86	
Clomazone	Pethoxamid + imazethapyr	3-4 lf	420	1	с	3	de	107	a	90	
Clomazone	Pethoxamid + imazethapyr	3-4 lf	560	1	с	2	de	95	bcde	90	
Clomazone	Pethoxamid + carfentrazone	3-4 lf	420	1	c	2	de	82	fg	92	
Clomazone	Pethoxamid + carfentrazone	3-4 lf	560	1	c	3	de	93	cde	89	
			<i>P</i> -value	0.0	033	<0.0	0001	<0.00	001	NS	

4.1. Rice injury ratings 2 weeks after treatment and shoot density at the Rice Research and Extension Center near Stuttgart, AR, in 2015 and 2016.<sup>a,b</sup>

Table 4.1 Cont.

<sup>a</sup> Abbreviations: PRE, preemergence; POST, postemergence; lf, leaf; NS, not significant

<sup>b</sup> Means within a column followed by the same lowercase letter are not different according to Fisher's protected LSD ( $\alpha = 0.05$ ).

				50% H	leading	Yield					
PRE herbicide	POST herbicide	POST timing	Pethoxamid rate	2015	2016	20	2015		16		
licibicide		unning	g ai ha <sup>-1</sup>		layed—		kg h		-		
			g ai na	days de	layea		ĸgii	iu			
Nontreated				0	0	8289	efg	7080	def		
	Pethoxamid	1-lf	420	0	0	7680	g	6370	f		
	Pethoxamid	1-lf	560	0	0	9810	a	6870	def		
	Pethoxamid + clomazone	1-lf	420	0	0	8640	cdefg	6420	ef		
	Pethoxamid + clomazone	1-lf	560	0	3	8540	defg	7280	cdef		
Clomazone	Pethoxamid + quinclorac	3-4 lf	420	0	4	9550	abcd	8290	ab		
Clomazone	Pethoxamid + quinclorac	3-4 lf	560	0	5	9090	abcde	7330	cd		
Clomazone	Pethoxamid + propanil	3-4 lf	420	1	0	9350	abcd	8740	а		
Clomazone	Pethoxamid + propanil	3-4 lf	560	0	4	8790	bcdef	7990	abc		
Clomazone	Pethoxamid + imazethapyr	3-4 lf	420	0	3	7730	fg	7680	bcd		
Clomazone	Pethoxamid + imazethapyr	3-4 lf	560	0	3	9600	abc	7730	bcd		
Clomazone	Pethoxamid + carfentrazone	3-4 lf	420	0	3	9700	ab	8340	ab		
Clomazone	Pethoxamid + carfentrazone	3-4 lf	560	0	3	9650	abc	8090	abc		
			<i>P</i> -value			0.00	001	< 0.0	001		

Table 4.2. Days delayed to 50% heading and rough rice yield at the Rice Research and Extension Center near Stuttgart, AR, in 2015 and 2016.<sup>a,b,c</sup>

Table 4.2 Cont.

<sup>a</sup> Abbreviations: PRE, preemergence; POST, postemergence; lf, leaf; NS, not significant

<sup>b</sup> Means within a column followed by the same lowercase letter are not different according to Fisher's protected LSD ( $\alpha = 0.05$ ).

<sup>c</sup> The 50% heading ratings were not amenable to formal statistical analysis.

				Barnyardgrass control										
					3 W.	AT			4 W.	AF				
PRE herbicide	POST herbicide	POST timing	Pethoxmid rate	PTRS		UAPB		PTRS		UAI	PB			
			g ai ha <sup>-1</sup>				%							
Nontreated														
	Pethoxamid	1-lf	420	94	c	84	d	81	d	71	С			
	Pethoxamid	1-lf	560	94	c	94	ab	88	c	90	a			
	Pethoxamid +	1-lf	420	97	abc	91	abc	90	c	91	a			
	clomazone													
	Pethoxamid +	1-lf	560	95	bc	95	а	93	bc	93	a			
	clomazone													
Clomazone	Pethoxamid +	3-4 lf	420	100	a	94	ab	100	а	94	a			
	quinclorac													
Clomazone	Pethoxamid +	3-4 lf	560	100	a	96	а	98	ab	93	a			
	quinclorac													
Clomazone	Pethoxamid +	3-4 lf	420	100	a	90	abcd	100	а	79	b			
	propanil													
Clomazone	Pethoxamid +	3-4 lf	560	100	a	91	abc	99	а	81	b			
	propanil													
Clomazone	Pethoxamid +	3-4 lf	420	97	ab	93	abc	100	а	93	а			
	imazethapyr													
Clomazone	Pethoxamid +	3-4 lf	560	100	a	94	ab	100	а	93	а			
	imazethapyr													
Clomazone	Pethoxamid +	3-4 lf	420	100	a	86	cd	98	ab	78	bo			
	carfentrazone													
Clomazone	Pethoxamid +	3-4 lf	560	100	а	87	bcd	100	а	78	bo			
	carfentrazone									. 0	-			
			<i>P</i> -value	0.00	005	0.0	297	< 0.00	01	< 0.0	001			

Table 4.3. Barnyardgrass control 3 weeks after treatment (WAT) and 4 weeks after permanent flood (WAF) at UAPB and PTRS in 2016.<sup>a,b</sup>

Table 4.3 Cont.

<sup>a</sup> Abbreviations: PTRS, Pine Tree Research Station near Colt, AR; UAPB, University of Arkansas Pine Bluff Farm near Lonoke, AR; PRE, preemergence; POST, postemergence; lf, leaf; NS, not significant

<sup>b</sup> Means within a column followed by the same lowercase letter are not different according to Fisher's protected LSD ( $\alpha = 0.05$ ).

		Barnyardg	grass control	
	3 W	/AT	4 W	VAF
Contrast	PTRS	UAPB	PTRS	UAPB
		p-v	value	
420 vs. 560 g ha <sup>-1</sup>	NS	0.0339	NS	0.0105
PRE vs. no PRE	< 0.0001	NS	< 0.0001	NS
Peth $+$ clom vs. peth	NS	NS	0.0008	< 0.0001
Peth + quin vs. peth	< 0.0001	0.0136	< 0.0001	< 0.0001
Peth $+$ prop vs. peth	< 0.0001	NS	< 0.0001	NS
Peth + imazeth vs peth	0.0003	NS	< 0.0001	< 0.0001
Peth $+$ carf vs. peth	< 0.0001	NS	< 0.0001	NS

Table 4.4. Contrasts for percentage barnyard grass control 3 WAT and 4 WAF at PTRS and UAPB in 2016.  $^{\rm a,b}$ 

<sup>a</sup> Abbreviations: WAT, weeks after treatment; WAF, weeks after permanent rice flood; PTRS, Pine Tree Research Station near Colt, AR; UAPB, University of Arkansas Pine Bluff Farm near Lonoke, AR; NS, not significant; PRE, preemergence herbicide; Peth, pethoxamid; clom, clomazone; quin, quinclorac; prop, propanil; imazeth, imazethapyr; carf, carfentrazone

### Chapter 5

# Selectivity of Very-Long-Chain Fatty Acid-Inhibiting Herbicides in Rice as Influenced by Application Timing and Soil Texture

Very-long-chain fatty acid (VLCFA)-inhibiting herbicides include pyroxasulfone, S-metolachlor, acetochlor, and pethoxamid. Currently, no VLCFA-inhibiting herbicides are labeled for use in U.S. rice; however, if rice tolerance can be established they would provide an alternative herbicide site of action. In 2015 and 2016, pyroxasulfone at 150 g ai ha<sup>-1</sup>, S-metolachlor at 1,070 g ai ha<sup>-1</sup>, acetochlor at 1,050 g ai ha<sup>-1</sup>, and pethoxamid at 840 g ai ha<sup>-1</sup> were applied delayed preemergence (DPRE) and to spiking and 1- to 2-leaf (lf) rice on a Dewitt silt loam and a Sharkey clay soil. Trials were conducted at separate locations to determine the effect differing soil properties may have on the tolerance of rice to VLCFA-inhibiting herbicides. Substantial rice injury was observed on the Dewitt silt loam soil following all pyroxasulfone and Smetolachlor treatments, ranging from 20 to 100% injury 3 weeks after rice flooding. On the Dewitt silt loam, treatments containing acetochlor and pethoxamid applied to 1- to 2-lf rice resulted in minimal injury and the highest yields observed among VLCFA-inhibiting herbicides. In general, rice appeared to be injured less on the Sharkey clay soil. Pethoxamid and acetochlor applied to rice at any application timing resulted in  $\leq 1\%$  injury 2 weeks after treatment. Hence, contingent upon a label, pethoxamid can be applied under similar conditions as early as DPRE on the Sharkey clay soil and not until the 1- to 2-lf rice stage on the Dewitt silt loam at the rates assessed.

Nomenclature: Acetochlor; pethoxamid; pyroxasulfone; S-metolachlor; rice, Oryza sativa L.

**Key words:** Delayed preemergence, Dewitt silt loam, Sharkey clay, spiking rice, very-longchain fatty acid-inhibiting herbicides

### Introduction

Currently, there are no VLCFA-inhibiting herbicides labeled for use in U.S. rice production. However, due to the evolution of resistant weed species such as barnyardgrass (*Echinochloa crus-galli* (L.) Beauv.) and red rice (*Oryza sativa* var. *sylvatica* L.), it is imperative that alternative herbicide sites of action (SOA) are integrated into rice production whenever possible. VLCFA-inhibiting herbicides are currently labeled in the US for use in various row crops for control of grasses and small-seeded broadleaf weeds (Knowles 1998). VLCFA herbicides would include acetochor, alachlor, butachlor, metolachlor, pyroxasulfone, and pethoxamid, which is currently under development for use in the US by FMC (FMC Corporation, Philadelphia, PA) (Mallory-Smith et al. 2003; Kraehmer et al. 2014).

Herbicides used to control barnyardgrass in rice include pendimethalin (Weed Science Society of America [WSSA] Group 3), quinclorac (WSSA Group 4), thiobencarb (WSSA Group 8), clomazone (WSSA Group 13), cyhalofop (WSSA Group 1), fenoxaprop (WSSA Group 1), penoxsulam (WSSA Group 2), bispyribac (WSSA Group 2), imazethapyr (WSSA Group 2), imazapyr (WSSA Group 2), and propanil (WSSA Group 7) (Heap 2017; Scott et al. 2016). Currently, populations of barnyardgrass have been confirmed resistant to every SOA in the Midsouth used to control barnyardgrass in rice, except for pendimethalin and thiobencarb (Heap 2017). Targeting the most troublesome weeds and using multiple herbicide SOA through annual rotations, tank-mixtures, and sequential applications is an important way to combat herbicide resistance (Norsworthy et al. 2012); therefore, the integration of effective alternative herbicide SOAs into rice would provide growers an opportunity to combat resistant weed species.

No new SOA has been introduced commercially in any agronomic crop in over 20 years (Duke 2011). This is due to several factors, one being that overreliance on glyphosate-resistant

crops devalued the use of herbicides other than glyphosate. Another factor concerns the consolidation of the majority of the pesticide discovery industry along with the high cost associated with pesticide development and increasing regulation (Duke 2011). Because the development of a new herbicide SOA is difficult, integrating a SOA currently used in other cropping systems into rice, such as VLCFA-inhibiting herbicides, may provide a simple yet effective benefit to rice weed control.

VLCFA-inhibiting herbicides such as pretilachlor and butachlor are used successfully in Asian dry- and wet-seeded rice at both preemergence (PRE) and postemergence (POST) application timings (Rao et al. 2007). VLCFA herbicides are soil-applied, root- and shootinhibiting herbicides, which have a strong effect on meristem-bearing cell division; however, they have little effect on preexisting plant tissues (Babczinski et al. 2012). Therefore, in order to maximize rice tolerance and weed control, these herbicides should be applied after rice germination but prior to weed germination.

VLCFA herbicides are currently used to control weeds such as barnyardgrass and red rice in row crop production including soybean (*Glycine maxx* (L.) Merr.), corn (*Zea mays* L.), cotton (*Gossypium hirsutum* L.), and various other crops. In Brazil, *S*-metolachlor has provided up to 90% control of red rice 14 days after a preemergence (PRE) application of 1,680 g ai ha<sup>-1</sup> in soybean (Zemolin et al. 2014). Season-long control of barnyardgrass, up to 63%, has been observed 95 to 140 days after a single PRE application of a microencapsulated formulation of acetochlor at 1,270 g ha<sup>-1</sup> in corn (Janak and Grichar 2016). Another VLCFA-inhibiting herbicide that has displayed high levels of barnyardgrass control is pyroxasulfone. In 2009, PRE applications of pyroxasulfone at 150 g ha<sup>-1</sup> controlled barnyardgrass up to 100% in potato (*Solanum tuberosum* L.) 66 days after treatment in Ontario (Boydston et al. 2012). The final

VLCFA-inhibiting herbicide evaluated is pethoxamid, which has also displayed levels of barnyardgrass control up to 91% as a PRE application at 1,200 g ha<sup>-1</sup> in sunflower (*Helianthus annuus* L.) (Jursik et al. 2012). The red rice and barnyardgrass control associated with the VLCFA-inhibiting herbicides in these studies indicate that this SOA may provide weed control in rice if tolerance can be established.

Because VLCFA-inhibiting herbicides are soil-applied, several factors have a significant impact on herbicidal activity including soil composition and soil chemistry (Curran 2001). The most important soil properties that affect activity of a soil-applied herbicide are organic matter (OM), clay content, and pH (Eberlein et al. 1984). All these factors interact to influence the activity and persistence of soil-applied herbicides.

Adsorption of herbicides in the soil is closely associated with the inorganic and organic colloids of the soil (Rao 2000). Most inorganic soil colloids are composed of clay, which contains three major minerals, including montmorillonite, illite, and kaolinite. In mineral soils, clay and organic matter are ultimately bound together; therefore, there are two major types of adsorbing surfaces available to the herbicide: clay-humus and clay alone (Stevenson 1972).

A study was conducted determining the relationship between clay content with herbicide adsorption (Villaverde et al. 2008). Dicamba, 2,4-D, metsulfuron-methyl, and flupyrsulfuronmethyl-sodium were all applied to five different soils with textures ranging from sand to clay and clay contents ranging from 2.5 to 65.9%. A strong correlation was observed between clay content and the adsorption coefficient for each herbicide evaluated; hence, higher clay content results in higher adsorption, which may lead to decreased phytotoxicity to the crop and lessened weed control.

Research has shown that OM is the main component responsible for herbicide adsorption (Rao 2000). Humic acids present in soil OM are responsible for the stable bonding of herbicides. Humic substances can be described as highly acidic, yellow to black colored, high molecular weight polyelectrolytes with a high content of oxygen-containing functional groups, which give them the ability to readily combine with organic molecules such as herbicides (Stevenson 1972). Decomposing plant residues have a greater adsorptive capability than the soil itself, which occurs mostly through hydrogen bonding. Due to the increased adsorptive capability of soils high in OM, it is sometimes necessary to increase the application rates of some soil-applied herbicides (Rao 2000).

Along with clay content and soil OM, soil pH also affects herbicide adsorption. Herbicides respond differently to changes in soil pH. For instance, herbicides such as triazines develop more cationic characteristics as soil pH decreases, which leads to higher adsorption (Rao 2000). Soil pH mainly affects herbicides belonging to the triazine and sulfonylurea families as chemical breakdown becomes slower in soils with higher pH, resulting in prolonged activity (Curran 2001).

When assessing herbicides, such as VLCFA-inhibiting herbicides, to be integrated into a crop such as rice, rice tolerance must be established. Characterizing herbicides over various soils allows for the determination of appropriate application timings based on rice tolerance. Considering the success of VLCFA-inhibiting herbicides in Asian rice production and the effect of soil properties on herbicidal activity, it is believed that rice tolerance to VLCFA-inhibiting herbicides will differ between a Sharkey clay soil and a Dewitt silt loam soil; however, these herbicides may provide a safe and alternative SOA for use in U.S. rice. It was hypothesized that rice would be tolerant to at least one VLCFA-inhibiting herbicide based on application timing, and that rice would be more tolerant to these herbicides on a Sharkey clay than a Dewitt silt loam. The objective of these experiments was to assess the tolerance of rice to VLCFAinhibiting herbicides on two different soils applied over multiple application timings.

### **Materials and Methods**

Field trials were conducted on the tolerance of rice to VLCFA-inhibiting herbicides in 2015 and 2016 at the Rice Research and Extension Center (RREC) near Stuttgart, Arkansas, and the Northeast Research and Extension Center (NEREC) in Keiser, Arkansas. Soil at the RREC was a Dewitt silt loam soil (fine, smectic, thermic typic Albaqualf) with a pH of 6.0, OM of 1.8%, 8.4% sand, 71.4% silt, and 20.2% clay. The NEREC contained a Sharkey clay soil (very-fine, smectic, thermic chromic Epiaquerts) with a pH of 7.3, organic matter of 3.4%, 17.8% sand, 28.5% silt, and 53.7% clay. Each year CL 111 rice (Clearfield<sup>TM</sup>, BASF Corporation, Research Triangle Park, NC) was drill seeded at 72 seed m<sup>-1</sup> of row into 1.8- by 5.2-m plots at a drill spacing of 18 cm. Rice was planted on May 5, 2015, and April 25, 2016, at the RREC and on June 11, 2015, and May 9, 2016, at the NEREC (Table 5.1). Rice fertility programs were based on University of Arkansas Division of Agriculture Research and Extension recommendations (Norman et al. 2013).

Each experiment was designed as a two-factor randomized complete block with twelve treatments along with a nontreated control. The first factor consisted of four herbicides (fixed): acetochlor (Warrant, Monsanto Company, St. Louis, MO), pyroxasulfone (Zidua, BASF Corporation, Research Triangle Park, NC), *S*-metolachlor (Dual II Magnum, Syngenta Crop Protection, LLC, Greensboro, NC), and pethoxamid (FMC Corporation, Philadelphia, PA).The

second factor (fixed) was application timing: delayed preemergence [(DPRE) (4 to 6 days after planting)], spiking, and 1- to 2-lf rice. Each herbicide was applied at the following rate: acetochlor at 1,050 g ha<sup>-1</sup>, pyroxasulfone at 150 g ha<sup>-1</sup>, *S*-metolachlor at 1,070 g ha<sup>-1</sup>, and pethoxamid at 840 g ha<sup>-1</sup>. Application dates for each timing and location are shown in Table 5.1. All herbicides were applied with a CO<sub>2</sub>-pressurized backpack sprayer through 110015 AIXR (TeeJet) nozzles calibrated to deliver 143 L ha<sup>-1</sup> using a three-nozzle boom at 51 cm spacing at 4.83 km h<sup>-1</sup>. All trials were focused strictly on rice tolerance; therefore, plots were kept weed free at both locations using herbicides recommended for weed control in Arkansas rice (Scott et al. 2016).

Data collection for all trials consisted of a visual assessment of crop injury 2 WAT and 3 WAF compared to the nontreated control on a 0 to 100 scale, with 0 representing no crop injury and 100 representing complete crop death. The aspects associated with the visual assessment of crop injury included a visible reduction in rice stand, tillering, and height, and overall plant vigor. Other parameters evaluated were rice shoot density (shoots m<sup>-1</sup> of row), rice canopy height at three locations per plot (cm), days to 50% heading relative to the nontreated control, and rough rice yield (kg ha<sup>-1</sup>) corrected to 14% moisture. Rice heights were assessed 4 weeks after planting (WAP) in 2015 and 10 WAP in 2016 at the RREC and 8 WAP in 2015 and 9 WAP in 2016 at the NEREC. Rice shoot densities were assessed 4 WAP in 2015 and 5 WAP in 2016 at the RREC and 4 WAP in 2016 at the NEREC. Rice shoot density was not evaluated at the NEREC in 2015. Shoot density, height, and rough rice yield were all normalized as a percentage of the nontreated control.

Data were analyzed in JMP 12 Pro (SAS Institute Inc, Cary, NC). Data for each year were analyzed separately because of differences in rainfall patterns between years. All data were

subjected to analysis of variance with block included as a random effect. All means were separated using Fisher's protected LSD ( $\alpha$ =0.05).

### **Results and Discussion**

Silt Loam Soil. Overall, more injury was observed across all application timings following pyroxasulfone and *S*-metolachlor when compared to acetochlor and pethoxamid (Tables 5.2). In 2015, visible injury 2 WAT was  $\leq$  9% following applications of acetochlor at all timings assessed, the 1- to 2-lf application of *S*-metolachlor, and the spiking and 1- to 2-lf applications of pethoxamid (Table 5.2). Although not compared directly between years, injury was generally greater in 2016 than in 2015 due to a rainfall event of 10.4 cm on May 3, 2016, which was immediately after the spiking treatments were applied in the same day. Even though spiking rice was present, an inconsistent stand occurred due to the dry conditions between planting on April 25 and the 10.4 cm of rainfall on May 3 (Figure 5.1). The 10.4 cm of rainfall was likely sufficient to activate the herbicides and cause some downward movement into the soil, which caused an increase in overall injury compared to 2015 when no rainfall event greater than 5.5 cm was recorded between planting and the final herbicide application (Figure 5.2). The lowest injury in 2016, however, was following acetochlor and pethoxamid applications to 1- to 2-lf rice (Table 5.2).

By 3 WAF, visible injury was  $\leq$  3% following applications of acetochlor or pethoxamid when averaged over all application timings in 2015 (Table 5.2). Similar to 2 WAT, injury was lowest following the 1- to 2-lf applications of acetochlor and pethoxamid at 3 WAF in 2016. Relatively high levels of visible injury were observed 3 WAF for all applications of pyroxasulfone and *S*-metolachlor.

Rice shoot densities and height were similar to the nontreated control in 2015 following applications of pethoxamid and acetochlor when averaged over all application timings and in 2016 following applications of acetochlor and pethoxamid at 1- to 2-lf rice (Tables 5.2 and 5.3). Delays to 50% heading in 2015 compared to the nontreated control were observed following all applications of pyroxasulfone and *S*-metolachlor; however, no delay in heading resulted from any application of acetochlor or pethoxamid (Table 5.3). Different from 2015, delays to 50% heading did occur in 2016 following the DPRE application of acetochlor and the spiking and 1- to 2-lf applications of pethoxamid, ranging from 8 to 11 days. The delay to 50% heading associated with pethoxamid applied at spiking and 1- to 2-lf rice did not have a deleterious effect on rough rice yield (Table 5.3). No interaction occurred for rough rice yield in 2015; however, the highest yields compared to the nontreated control were for the acetochlor and pethoxamid-treated plots, when averaged over all application timings (Table 5.3).

Rice generally exhibited adequate tolerance to acetochlor and pethoxamid in 2015 and 2016 on the silt loam soil whereas the crop was often negatively affected by *S*-metolachlor and pyroxasulfone based on yield, height, shoot density, and visible injury. These findings lead to the conclusion that the VLCFA-inhibiting herbicides pethoxamid and acetochlor can safely be applied to rice on a silt loam soil under the environmental conditions evaluated; however, it is important to make sure at least one true leaf is present to ensure rice tolerance.

**Clay Soil.** Similar to results in the silt loam soil, rice injury was less with acetochlor and pethoxamid than with pyroxasulfone and *S*-metolachlor (Table 5.4). Unlike the RREC, however, rainfall events were not substantial enough to have an impact on rice injury in either 2015 or 2016 at the NEREC (Figures 5.3 and 5.4). Injury ratings of up to 48% and 28% at 2 WAT were observed in 2016 following applications of pyroxasulfone and *S*-metolachlor, respectively;

however, injury was  $\leq 1\%$  with acetochlor or pethoxamid (Table 5.4). At 3 WAF in 2015, there was no significant interaction between factors for rice injury (Table 5.4). However, pyroxasulfone injured rice more than did acetochlor, *S*-metolachlor, or pethoxamid, which was minimal. Injury levels  $\geq 10\%$  were observed following all applications of pethoxamid in 2016; however, the injury observed was a function of overall plant vigor since no reduction in shoot density or rice height was noticed compared to the nontreated control (Tables 5.4, 5.5).

There was no appreciable delay in heading ( $\leq 1$  day) and no reduction in yield compared to the nontreated control following applications of pethoxamid or acetochlor at any timing (Table 5.5). Hence, on the clay soil, pethoxamid and acetochlor were the only VLCFA-inhibiting herbicides to which rice exhibited adequate tolerance across all parameters assessed. Even though visible injury ranging from 10 to 23% was observed following applications of pethoxamid 3 WAF in 2016, no reduction in shoot density, days to 50% heading, or rough rice yield occurred when compared to the nontreated control (Tables 5.4, 5.5). Visible injury was  $\leq$ 4% following any application of acetochlor at 2 WAT and 3 WAF in 2015 and 2016. Similarly, yield of rice treated with acetochlor was not reduced compared to the nontreated control. Considering these data, acetochlor and pethoxamid can safely be applied at any timing assessed to rice on a clay soil under the conditions present in this study with minimal rice injury. Soil Properties. The contrasting soil properties between the Dewitt silt loam at the RREC and the Sharkey clay at the NEREC likely contributed to some of the differences observed in rice tolerance to VLCFA-inhibiting herbicides. The Sharkey clay soil at the NEREC has a higher OM and clay content than the Dewitt silt loam soil at the RREC. The NEREC has an OM content of 3.4% and a clay content of 53.7% compared to the OM content of 1.8% and the clay content of 20.2% present at the RREC.

Organic matter content is the main component responsible for herbicide adsorption (Rao 2000). Research has shown that two to three times more herbicide may be required for 80% weed control on a soil with an OM of 19.3% compared to a soil with an OM of 8% due to the adsorptive properties of OM (Rahman et al. 1978). The soils at the NEREC with an OM of 3.4% would be more adsorptive than the soil at the RREC with an OM of 1.8%, resulting in less rice injury from VLCFA-inhibiting herbicides. It should be noted that rates of the herbicides evaluated may need to be increased on the clay soil in order to provide a similar level of weed control as that observed on the silt loam soil.

Along with OM, the increase in clay content at the NEREC compared to the RREC would allow more herbicide adsorption and less rice injury. There was reduction in rice injury from VLCFA-inhibiting herbicides on the Sharkey clay soil with a clay content of 53.7% compared to the Dewitt silt loam soil with a clay content of 20.2%. This coincides with research conducted where the strongest sorbent of acetochlor was the soil highest in clay content on soils ranging from 23.6 to 3.4% clay (Durovic et al. 2009).

Although weed control was not assessed, the differences in rice tolerance to VLCFAinhibiting herbicides could theoretically correlate with weed control. Rahman et al. (1978) showed that more herbicide is needed for 80% weed control on soils with greater OM; hence, higher rates of pethoxamid and acetochlor may be needed on the Sharkey clay soil for sufficient levels of weed control.

**Practical Implications.** VLCFA-inhibiting herbicides may provide an alternative herbicide option for rice weed control. As the evolution of herbicide resistance continues, it is important that growers be presented with new herbicide options to alternate and tank-mix in order to combat resistant weed species such as red rice and barnyardgrass. Considering the low amounts

of injury and absence of yield reduction associated with the use of acetochlor and pethoxamid in rice at the 1- to 2-lf application timing on a silt loam soil and at all timings assessed on a clay soil, these VLCFA-inhibiting herbicides may be used safely in rice.

Differences in injury were noticed with the same treatments on differing soil textures. In general, numerically less injury was seen on the Sharkey clay soil than on the Dewitt silt loam soil. The increase in clay content and OM associated with the clay soil caused more herbicide adsorption and less phytotoxicity than that observed on the silt loam soil. At the rates assessed, pethoxamid and acetochlor can be applied safely to rice at application timings as early as DPRE on a Sharkey clay soil, whereas on the Dewitt silt loam soil the safest timing is at the 1- to 2-lf stage. However, for acceptable levels of weed control, higher rates of pethoxamid and acetochlor than those used on the silt loam may be required on the Sharkey clay soil. Additional research is necessary to establish the rates of acetochlor and pethoxamid necessary for effective weed control on these two soils.

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				Herbicide applications										
	RREC May 5 Apr	nting	DI	PRE	Sp	iking	-	1-2 lf						
Location	2015	2016	2015	2016	2015	2016	2015	2016						
RREC	May 5	Apr 25	May 11	Apr 26	May 13	May 3	May 18	May 12						
NEREC	Jun 11	May 9	Jun 15	May 14	June 17	May 19	June 25	May 24						

Table 5.1. Planting and herbicide application dates at RREC and NREC in 2015 and 2016. <sup>a</sup>

<sup>a</sup> Abbreviations: DPRE, delayed preemergence; NEREC, Northeast Research and Extension Center in Keiser, AR; RREC, Rice Research and Extension Center near Stuttgart, AR

		Injury											
		2 V		/AT			3 V	WAF		Shoot		density <sup>c,d</sup>	
Factor		202	2015		2016		15	2016		20	15	2	2016
						·%				%	of n	ontrea	nted-
Herbicide													
	Acetochlor	4		58		3	С	27		92	a	67	
	Pyroxasulfone	15		69		68	a	67		73	b*	48	
	S-metolachlor	17		90		30	b	83		66	b*	26	
	Pethoxamid	11		50		1	с	37		96	a	54	
Timing													
-	DPRE	21		98		28		91		71	c	8	
	Spiking	10		54		26		50		82	b	54	
	1-2 lf	4		40		23		20		93	а	85	
Herbicide × Timing													
	Acetochlor $\times$ DPRE	7	d-g	92	b	5		66	с	88		28	d*
	Acetochlor × Spiking	3	fg	36	f	4		11	f	95		84	a*
	Acetochlor $\times$ 1-2 lf	1	g	15	g	1		2	g	94		90	a
	Pyroxasulfone × DPRE	23	b	100	a	70		99	a	56		3	ef*
	Pyroxasulfone × Spiking	13	cd	58	d	65		82	b	77		52	c*
	Pyroxasulfone× 1-2 lf	10	c-e	49	e	68		20	e	88		90	a
	S-metolachlor $\times$ DPRE	33	a	100	a	36		100	а	55		1	f*
	S-metolachlor $\times$ Spiking	14	c	88	bc	32		95	a	55		14	e*
	S-metolachlor $\times$ 1-2 lf	4	e-g	82	c	23		55	d	88		63	bc*
	Pethoxamid × DPRE	22	b	100	a	1		98	а	85		2	ef*
	Pethoxamid × Spiking	9	c-f	36	f	1		13	ef	103		65	b*
	Pethoxamid $\times$ 1-2 lf	3	fg	13	g	1		1	g	102		96	а
<i>P</i> -Values from ANO	VA												
Herbicide ( <i>P</i> )		< 0.0	001	< 0.00	001	< 0.00	01	< 0.0	001	< 0.0	001	<0	.000
Timing $(P)$		< 0.0		< 0.0		NS		< 0.0		0.00			.000
Herbicide × timing (	<i>P</i> )	0.00		< 0.0		NS		< 0.0		N			.000

Table 5.2. Rice injury ratings 2 weeks after treatment and 3 weeks after rice flooding and shoot density at RREC in 2015 and 2016.<sup>a,b</sup>

Table 5.2 Cont.

<sup>a</sup> Abbreviations:; RREC, Rice Research and Extension Center near Stuttgart, AR; DPRE, delayed preemergence; lf, leaf; NS, not significant

<sup>b</sup> Means within a column followed by the same lowercase letter are not different according to Fisher's protected LSD ( $\alpha = 0.05$ ).

<sup>c</sup> Letters followed by asterisks (\*) for height and yield denote a statistical decrease from the nontreated control based on the confidence intervals obtained for each interaction or main effect.

<sup>d</sup> The nontreated control average shoot density at the RREC was 92 m<sup>-1</sup> of row in 2015 and 75 m<sup>-1</sup> of row in 2016.

		Height <sup>c,d</sup>				Headin	ng delay <sup>e</sup>	Yield <sup>f</sup>		
Factor	-	201			16	2015	2016	2015	2016	
		% of nontreated			c	lays	% of nontreated			
Herbicide										
	Acetochlor	99	a	93		0	4	95 a	80	
	Pyroxasulfone	56	c*	65		14	14	52 c*	36	
	S-metolachlor	91	b*	72		3	16	80 b*	43	
	Pethoxamid	101	a	87		0	12	94 a	66	
Timing										
-	DPRE	87		65		5	15	77 b*	23	
	Spiking	88		79		3	11	80 ab*	61	
	1-2 lf	85		93		5	8	84 a*	84	
Herbicide × Timing										
-	Acetochlor × DPRE	100		91	bc*	0 d	11 c	90	73 b*	
	Acetochlor × Spiking	97		88	cd*	0 d	0 e	93	74 b*	
	Acetochlor $\times$ 1-2 lf	99		101	a	0 d	0 e	103	92 a	
	Pyroxasulfone $\times$ DPRE	54		52	f*	17 a	17 a	46	8 e*	
	Pyroxasulfone × Spiking	60		58	f*	11 b	17 a	58	23 d*	
	Pyroxasulfone× 1-2 lf	53		84	cd*	15 a	8 d	52	77 b*	
	S-metolachlor $\times$ DPRE	91		49	f*	2 c	17 a	79	7 e*	
	S-metolachlor × Spiking	95		79	d*	3 c	15 b	77	54 c*	
	S-metolachlor $\times$ 1-2 lf	88		86	cd*	4 c	15 b	84	69 b*	
	Pethoxamid $\times$ DPRE	101		70	e*	0 d	17 a	93	4 e*	
	Pethoxamid × Spiking	102		92	bc*	0 d	10 c	94	95 a	
	Pethoxamid $\times$ 1-2 lf	101		99	ab	0 d	8 d	96	99 a	
<i>P</i> -Values from ANO	VVA									
Rate (P)		< 0.00	01	< 0.0001		< 0.0001	< 0.0001	< 0.0001	< 0.000	
Timing (P)		NS		<0.0	0001	0.0028	< 0.0001	0.0196	< 0.000	
Herbicide $\times$ timing (P)		NS		< 0.0001		0.0002	< 0.0001	NS	< 0.000	

Table 5.3. Height, days delayed to 50% heading, and rough rice yield as a percentage of the nontreated at RREC in 2015 and 2016.<sup>a,b</sup>

Table 5.3 Cont.

<sup>a</sup> Abbreviations: RREC, Rice Research and Extension Center near Stuttgart, AR; DPRE, delayed preemergence; lf, leaf; NS, not significant.

<sup>b</sup> Means within a column followed by the same lowercase letter are not different according to Fisher's protected LSD ( $\alpha = 0.05$ ).

<sup>c</sup> Letters followed by asterisks (\*) for height and yield denote a statistical decrease from the nontreated control based on the confidence intervals obtained for each interaction or main effect.

<sup>d</sup> The nontreated control average height at the RREC was 86 cm in 2015 and 94 cm in 2016.

<sup>e</sup> Delay in heading is reported as average days to 50% heading after the nontreated control.

<sup>f</sup> The nontreated control yield at the RREC was 8,850 kg ha<sup>-1</sup> in 2015 and 8,930 kg ha<sup>-1</sup> in 2016.

			Inju	ıry			
		2 W	AT	3 V	VAF	Shoot density	
Factor		2015	2016	2015	2016	2015	2016
			%	)		% of nor	treated
Herbicide							
	Acetochlor	1	0	1 b	3	-	96
	Pyroxasulfone	3	24	25 a	33	-	96
	S-metolachlor	2	15	2 b	24	-	91
	Pethoxamid	0	0	0 b	16	-	93
Timing							
C	DPRE	0	12	7	25	-	92
	Spiking	1	16	9	26	-	97
	1-2 LF	3	2	7	5	-	93
Herbicide ×							
Timing	Acetochlor × DPRE	0 c	0 e	1	4 fg	-	93
C	Acetochlor × Spiking	1 c	0 e	0	4 fg	-	97
	Acetochlor $\times$ 1-2 lf	0 c	0 e	0	0 g	-	99
	Pyroxasulfone $\times$ DPRE	0 c	18 c	24	36 c	-	96
	Pyroxasulfone × Spiking	1 c	48 a	29	51 a	-	100
	Pyroxasulfone $\times$ 1-2 lf	7 a	6 d	24	11 e	-	94
	S-metolachlor $\times$ DPRE	0 c	28 b	2	44 b	-	88
	S-metolachlor $\times$ Spiking	1 c	17 c	2	29 d	-	96
	S-metolachlor $\times$ 1-2 lf	3 b	0 e	0	0 g	-	90
	Pethoxamid × DPRE	0 c	1 de	0	15 e	-	91
	Pethoxamid × Spiking	0 c	0 e	1	23 d	-	98
	Pethoxamid $\times$ 1-2 lf	0 c	0 e	0	10 ef	-	91
<i>P</i> -Values from A	NOVA						
Herbicide ( <i>P</i> )		< 0.0001	< 0.0001	< 0.0001	< 0.0001		NS
Timing $(P)$		< 0.0001	< 0.0001	NS	< 0.0001		NS
Herbicide × timi	ng ( <i>P</i> )	< 0.0001	< 0.0001	NS	< 0.0001		NS

Table 5.4. Rice injury ratings 2 weeks after treatment and 3 weeks after rice flooding and shoot density at
NEREC in 2015 and 2016. <sup>a,b</sup>

Table 5.4 Cont.

<sup>a</sup>Abbreviations: WAT, weeks after treatment; WAF, weeks after permanent rice flooding; NEREC, Northeast Research and Extension Center near Keiser, AR; DPRE, delayed preemergence; lf, leaf; NS, not significant <sup>b</sup> Means within a column followed by the same lowercase letter are not different according to Fisher's protected LSD ( $\alpha = 0.05$ ).

<sup>c</sup> Letters followed by asterisks (\*) for height and yield denote a statistical decrease from the nontreated control based on the confidence intervals obtained for each interaction or main effect.

 $^{\rm d}$  The nontreated control average shoot density at the NREC was 74  $\rm m^{-1}$  of row in 2015 and 58  $\rm m^{-1}$  of row in 2016.

Factor		Height <sup>c,d</sup>			Heading delay <sup>e</sup>			Yield <sup>f</sup>			
		2015		2016		2015	2016	2015		2016	
		—% of nontreated—			days			% of nontreated-			nted
Herbicide											
	Acetochlor	93 a*	97		0		0	93		100	
	Pyroxasulfone	62 c*	86		2		1	59		71	
	S-metolachlor	89 b*	92		1		1	78		85	
	Pethoxamid	96 a	95		0		0	95		101	
Timing											
-	DPRE	86	92		0		1	80		88	
	Spiking	84	95		1		0	78		99	
	1-2 lf	85	90		1		1	85		81	
Herbicide ×											
Timing		05	100		0		0	0.0		104	1
	Acetochlor × DPRE	95	100	a	0	d	0 c	93	а	104	ab
	Acetochlor × Spiking	91	94	b-e	0	d	0 c	91	а	104	ab
	Acetochlor $\times$ 1-2 lf	93	95	a-d	0	d	0 c	91	a	93	a-d
	Pyroxasulfone $\times$ DPRE	63	86	fg*	1	с	1 b	60	cd*	83	cd*
	Pyroxasulfone × Spiking	60	91	c-f*	3	a	1 b	56	d*	88	b-d
	Pyroxasulfone× 1-2 lf	64	81	g*	2	b	1 b	61	cd*	44	e*
	$S$ -metolachlor $\times$ DPRE	90	88	ef*	1	с	2 a	72	b*	75	d*
	S-metolachlor × Spiking	89	98	ab	1	с	1 b	66	bc*	92	a-d
	S-metolachlor $\times$ 1-2 lf	88	89	d-f*	1	с	1 b	95	а	89	b-d
	Pethoxamid × DPRE	96	95	a-d	0	d	0 c	95	а	91	a-d
	Pethoxamid × Spiking	95	96	a-c	0	d	1 b	98	a	111	а
	Pethoxamid $\times$ 1-2 lf	96	95	a-d	0	d	0 c	93	a	101	a-c
P-Values from	m ANOVA										
Rate (P)		< 0.000	l <0.0	001	<(	0.0001	< 0.0001	<0.	0001	<(	).0001
Timing (P)		NS	0.01	113	<(	0.0001	NS	0.0	011	0	.0063
Rate × timing	g(P)	NS	0.01	121	<(	0.0001	0.0056	<0.0	0001	0	.0114

Table 5.5. Height, days delayed to 50% heading, and rough rice yield as a percentage of the nontreated at NEREC in 2015 and 2016.<sup>a,b</sup>

<sup>a</sup>Abbreviations: NEREC, Northeast Research and Extension Center near Keiser, AR; DPRE, delayed preemergence; NS, not significant

<sup>b</sup> Means within a column followed by the same lowercase letter are not different according to Fisher's protected LSD ( $\alpha = 0.05$ ).

<sup>c</sup> Letters followed by asterisks (\*) for height and yield denote a statistical decrease from the nontreated control based on the confidence intervals obtained for each interaction or main effect.

<sup>d</sup> The nontreated control average height at the NEREC was 62 cm in 2015 and 56 cm in 2016.

<sup>e</sup> Delay in heading is reported as average days to 50% heading after the nontreated control.

<sup>f</sup> The nontreated control yield at the NEREC was 7,430 kg ha<sup>-1</sup> in 2015 and 7,910 kg ha<sup>-1</sup> in 2016.

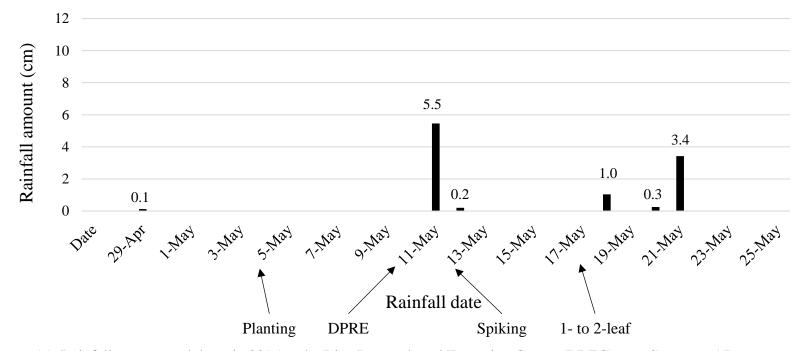


Figure 5.1. Rainfall amount and dates in 2015 at the Rice Research and Extension Center (RREC) near Stuttgart, AR .

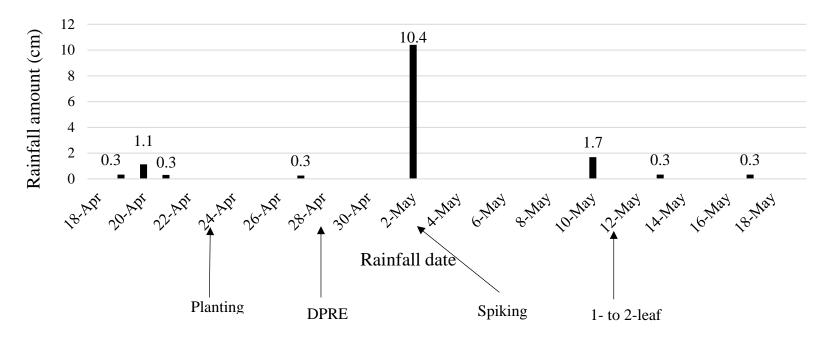


Figure 5.2. Rainfall amount and dates in 2016 at the Rice Research and Extension Center (RREC) near Stuttgart, AR.

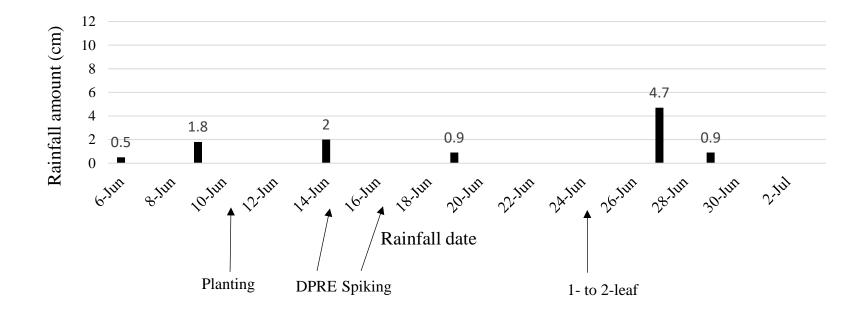


Figure 5.3. Rainfall amount and dates in 2015 at the Northeast Research and Extension Center (NEREC) in Keiser, AR.

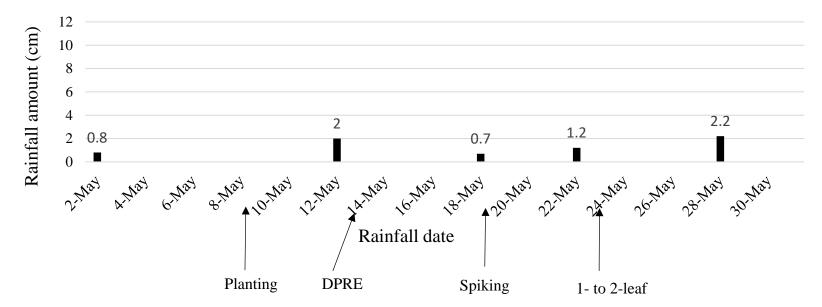


Figure 5.4. Rainfall amount and dates in 2016 at the Northeast Research and Extension Center (NEREC) in Keiser, AR.

## **General Conclusions**

In order to combat herbicide resistance in U.S. rice production it is essential that alternate herbicide sites of action (SOA) are utilized. Considering the minimal injury or reduction in yield to the rice crop following the use of pethoxamid or acetochlor at application timings after the 1leaf rice stage, pethoxamid and acetochlor may provide an additional SOA for use in rice. If labeled in rice, pethoxamid and acetochlor could offer growers the opportunity to use VLCFAinhibiting herbicides in a program based approach for control of barnyardgrass and red rice. Phytotoxicity associated with pethoxamid or acetochlor varied between rice on a Dewitt silt loam soil and a Sharkey clay soil. Application timings of pethoxamid or acetochlor as early as delayed preemergence (DPRE) on the Sharkey clay soil showed minimal injury and no reduction in yield; however, it was determined that rice must reach the 1-leaf growth stage in order to safely apply pethoxamid or acetochlor to rice on a Dewitt silt loam soil. Considering the generally lower amounts of injury associated with pethoxamid acetochlor on the Sharkey clay soil, similar results may be seen for weed control at the rates assessed. More research must be done in order to determine the proper rates to achieve adequate levels of weed control and low levels of rice phytotoxicity to rice on various soil types. On the other hand, VLCFA-inhibiting herbicides such as S-metolachlor and pyroxasulfone were considered unfit for use in rice production due to the vast amount of rice injury and reduction in yield associated with these herbicides at any rate, timing, and soil type assessed. Due to the lack of new herbicide SOA being registered for use in U.S. crops, it is important that we integrate herbicide SOA used in other crops whenever possible. The success shown with the VLCFA-inhibiting herbicides pethoxamid and acetochlor shows promise for the integration of new SOA into U.S. rice production.