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Effects of simulated drift of glyphosate, imazethapyr, glufosinate, and imazamox to non-transgenic rice

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EFFECTS OF SIMULATED DRIFT OF GLYPHOSATE, IMAZETHAPYR, GLUFOSINATE, AND
IMAZAMOX TO NON-TRANSGENIC RICE

A Dissertation

Submitted to the Graduate Faculty of the
Louisiana State University and
Agriculture and Mechanical College
in partial fulfillment of the
requirements for the degree of
Doctor of Philosophy

In

The School of Plant, Environmental, and Soil Sciences

By

Justin Brian Hensley
B.S., University of Arkansas, 1999
M.S., University of Arkansas, 2004
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Abstract

Four studies were conducted at the LSU AgCenter Rice Research Station near Crowley, Louisiana to evaluate the effects of simulated herbicide drift on 'Cocodrie' rice. Herbicides were applied at 6.3 and 12.5% of the labeled usage rate of 863 g ae/ha of glyphosate, 70 g ai/ha of imazethapyr, 493 g ai/ha of glufosinate, and 44 g ai/ha of imazamox. Herbicides were applied to rice at the 1-tiller, panicle differentiation (PD), boot, and physiological maturity growth stages. Spray volume varied proportionally to herbicide dosage and was 15 L/ha for the 6.3% rate and 29 L/ha for the 12.5% rate using 234 L/ha as the target spray volume and were applied with a tractor-mounted CO₂-pressurized sprayer.

Glyphosate reduced plant height and primary and total crop yield, with the greatest reduction in primary crop yield resulting from glyphosate applied at boot. Primary crop rice seed germination was reduced by glyphosate. Glyphosate reduced ratoon crop rice seed weight; however, ratoon crop seed germination was not reduced.

Imazethapyr reduced plant height and primary and total crop yield, with the greatest reduction in primary crop yield resulting from imazethapyr applied at boot. Primary crop rice seed weight was reduced by imazethapyr applied at boot. Primary crop rice seed germination was reduced by imazethapyr. Ratoon crop rice seed germination was reduced by imazethapyr applied at PD.

Glufosinate reduced rice plant height and, when applied at boot, reduced primary and total crop yield. Primary crop rice seed germination and primary crop seedling vigor were reduced by glufosinate.

Imazamox reduced plant height and primary and total crop yield with the greatest reduction in yield observed from rice treated at boot. Primary crop rice seed germination was reduced by imazamox. Primary crop seedling vigor

was reduced with imazamox applied at boot. Ratoon crop rice seed weight and germination was not affected by imazamox.

Simulated glyphosate, imazethapyr, glufosinate, and imazamox drift applications did not affect rice when applied at maturity. The greatest reduction in primary crop yield was observed when glyphosate, imazethapyr, glufosinate, and imazamox were applied to rice at boot and they all reduced primary crop rice seed germination.

Chapter 1

Literature Review

Rice (*Oryza sativa* L.) is a major crop produced in the four state region of Arkansas, Louisiana, Mississippi, and Texas, with these states accounting for 76% of the 1.2 million total hectares of rice planted in the U.S. and 70% of the \$3.4 billion of total value of rice produced in the U.S. in 2008 (NASSa 2009; NASSb 2009). Louisiana planted approximately 184,160 hectares of rice in 2008 with approximately 59% planted in imidazolinone-resistant rice cultivars and hybrids (LSUA 2009). That same year, approximately 98% of Louisiana's 424,920 hectares of soybean [*Glycine max* (L.) Merr.] were planted in glyphosate-resistant soybean cultivars and, in 2009, glufosinate-resistant soybean cultivars became available to commercial soybean producers in Louisiana (Ronald J. Levy, Jr.¹, personal communication).

Averaged over the ten year period 1999 to 2008, the Louisiana Department of Agriculture and Forestry (LDAF) processed 76 Pesticide Investigation Reports per year listing ground or aerial applicators in Louisiana, and, on average, six reports per year involved rice (Lisa Gautreaux², personal communication). However, it was reported that, in 2009, at least 50 rice fields were suspected of being affected by glyphosate drift and at least 25 rice fields were suspected of being affected by imazethapyr drift, many of which were not reported to the LDAF (Ronald J. Levy, Jr.¹, personal communication; John K. Saichuk³, personal communication). Therefore, the number of rice fields actually affected by a drift event each year may be underrepresented by the number of official complaints processed by the LDAF.

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Since many of the rice producing parishes in Louisiana also produce glyphosate-resistant corn (*Zea mays* L.), cotton (*Gossypium hirsutum* L.) and soybean, glufosinate-resistant corn, cotton, and soybean, and imidazolinone-resistant rice, the potential exists for off-target drift from these crops to rice (LSUA 2009; NASSc 2009).

It has been reported that fine spray droplets less than 150 µm in size have a greater potential to drift off-target (Hanks 1995; SDTF 1997). The use of adjuvants and selection of proper spray nozzle type, size, and application pressure can be beneficial in reducing the amount of fine spray droplets in the spray cloud (Hanks 1995; Jones et al. 2007; Nuyttens et al. 2007; VanGessel and Johnson 2005). This increase in droplet size can reduce the potential for off-target drift from droplets larger than 150 µm; however, environmental conditions at the time of herbicide application can also impact the off-target drift of spray solutions (Bouse et al. 1976; Crabbe et al. 1994; Thistle 2004).

Wind speed and direction may be considered the two most important factors affecting spray droplets in the atmosphere, a stable atmosphere may be the third most important factor (Thistle 2004). A stable atmosphere, commonly referred to as an inversion, is an atmosphere that has a change of temperature with a change in elevation in the atmosphere. In a stable atmosphere, warm air overlies cool air. If air in a particular layer is displaced upward or downward it will be colder or warmer, respectively, than the immediately adjacent layer it enters and thus return to its layer of origin. If a herbicide application is made during an inversion scenario, the fine droplets that do not succumb to gravity will remain in the air layer in which they are applied due to the lack of layers mixing. The droplets in this layer can be very concentrated and horizontally may move off-target great distances.

Ultra-low-volume applications made during a stable atmosphere produced 35% more herbicide drift than applications made during turbulent atmosphere with light wind speeds (Crabbe et al. 1994). The conditions in which the greatest drift occurred were in moderately stable conditions with wind at 3 m/s resulting in off-target drift 71% at 400 m and 50% at 2200 m from the application site and in slightly stable conditions with wind at 5 m/s resulting in off-target drift 77% at 400 m and 27% at 2200 m from the application site. It is recommended that herbicide applications should be avoided during the early morning and late evening as these times are most favorable for the development of inversion conditions (Crabbe et al. 1994; Thistle 2004).

Through the use of simulated herbicide drift studies, the potential effects of herbicide drift to rice can be evaluated. In previous research, simulated drift studies varying the spray volume proportionally with reduced herbicide rates to simulate herbicide drift have resulted in increased crop injury compared with the same lower herbicide rates at a constant high spray volume (Banks and Schroeder 2002; Ellis et al. 2002; Ramsdale et al. 2003; Roeder et al. 2008). Banks and Schroeder (2002) reported varying spray volume proportionally with herbicide dosage, thus maintaining constant herbicide concentration in the spray, would change the response of sweet corn to glyphosate when compared with a constant spray volume where herbicide rate would vary and be more dilute in the carrier. The no-effect glyphosate rate for sweet corn was 0.046 kg ae/ha when using a spray volume proportional to the reduced glyphosate rate; however, the no-effect glyphosate rate was four times greater when glyphosate was applied in a constant spray volume. Ellis et al. (2002) reported glyphosate applied to corn at 12.5 and 6.3% of the labeled use rate in a proportional spray volume and a constant spray volume produced results similar to those observed by Banks and Schroeder (2002).

The use of a constant spray volume in drift research may underestimate the effects of off-target drift to susceptible crops.

Glyphosate is a nonselective, foliar applied, postemergence herbicide used to control annual and perennial weeds in preplant burndown applications and for weed control in glyphosate-resistant crops (Senseman 2007c). The use of glyphosate has greatly increased with the introduction and extensive acceptance of glyphosate-resistant canola (*Brassica napus* L.), cotton (*Gossypium hirsutum* L.), corn, and soybean (Shaner 2000).

The mechanism of action of glyphosate is the inhibition of 5-enolpyruvylshikimate-3-phosphate (EPSP) synthase (EC 2.5.1.19) which produces EPSP from shikimate-3-phosphate and phosphoenolpyruvate in the shikimic acid pathway (Amrhein et al. 1980; Amrhein et al. 1983; Boocock and Coggins 1983; Herrmann and Weaver 1999; Hollander-Czytko and Amrhein 1987; Jakeman et al. 1998; Schonbrunn et al. 2001; Senseman 2007c). The inhibition of EPSP synthase is accomplished by glyphosate competing with phosphoenolpyruvate to bind with the shikimate-3-phosphate:EPSP synthase complex to form EPSP (Amrhein et al. 1980; Boocock and Coggins 1983; Herrmann and Weaver 1999; Jaworski 1972; Rubin et al. 1982; Schonbrunn et al. 2001). This outcompeting of phosphoenolpyruvate inhibits the formation of EPSP, which in turn inhibits the formation of the aromatic amino acids tryptophan, tyrosine, and phenylalanine which are needed for protein synthesis.

The symptoms expressed in plants from the inhibition of EPSP synthase are that growth is inhibited soon after application followed by general foliar chlorosis and necrosis within 4 to 7 days in highly susceptible species (Senseman 2007c). Visual symptoms occur within 10 to 20 days for less susceptible species, appearing first and most pronounced in immature leaves and growing points in the form of chlorosis. Also, regrowth of treated perennial and woody species often appears deformed and multiple shoots may develop at the nodes.

Simulated drift applications of glyphosate applied to rice at the two- to three-leaf and panicle differentiation growth stages reduced rice yield 67 to 99% and 29 to 54%, respectively (Ellis et al. 2003). Kurtz and Street (2003) reduced rice yield with simulated glyphosate drift applied to rice at mid-tiller, panicle initiation, and boot growth stages. Applications of glyphosate to red rice (*Oryza sativa* L.) in the two- to three-tiller, boot, and bloom growth stages reduced red rice seed germination, regardless of timing (Brommer et al. 1998). The use of glyphosate as a preharvest desiccant in grain sorghum [*Sorghum bicolor* (L.) Moench.] reduced grain sorghum seed germination (Baur et al. 1977).

In 1993, imidazolinone-resistant rice was developed and exhibited tolerance to the imidazolinone class of herbicides (Croughan 1994; Pellerin et al. 2004; Webster and Masson 2001). Imazethapyr and imazamox are selective imidazolinone herbicides used to control annual and perennial weeds in soybean, edible legumes, and imidazolinone-resistant crops (Senseman 2007a, 2007b).

The mechanism of action for imazethapyr and imazamox is inhibition of acetolactase synthase (ALS) (EC 4.1.3.18) also called acetohydroxyacid synthase (AHAS), a key enzyme in the biosynthesis of the branched-chain amino acids isoleucine, leucine, and valine (Muhitch et al. 1987; Senseman 2007a, 2007b; Shaner 1991; Shaner et al. 1984; Stidham 1991; Stidham and Singh 1991). Plant death results from events occurring in response to ALS inhibition, specifically the inhibition of isoleucine, leucine and valine, but the actual sequence of phytotoxic processes is unclear (Shaner 1991; Stidham and Singh 1991). Some secondary effects may include disruption of photosynthate translocation, hormone imbalance due to interruption of source/sink relationships, and interference in DNA synthesis and cell growth.

The symptoms expressed from this toxicity are growth inhibition within a few hours of herbicide application and meristematic areas becoming

chlorotic, followed by a slow general foliar chlorosis and necrosis (Shaner 1991). This injury to meristematic areas can be attributed to inhibition of branched-chain amino acids in the meristematic region. Even though plants have the ability to scavenge amino acids from pre-existing proteins, the meristematic region lacks the protein reserve pools that are available in the mature regions of the plant. Injury symptoms usually appear within 7 to 14 d for susceptible species.

A simulated drift application of the commercial herbicide premix of imazethapyr plus imazapyr affected rice plant height and yield; however, simulated drift of the imazethapyr plus imazapyr premix did not affect yield when applied to corn (Bond et al. 2006). Al-Khatib et al. (2003) reported imazethapyr applied at various times within 30 d of planting resulted in reduced grain sorghum yield. Deeds et al. (2006) reported imazamox applied to wheat (*Triticum aestivum* L.) at the flowering and jointing growth stages at 33% of the labeled use rate reduced wheat yield more than 90%. Applications of imazethapyr to red rice in the two to three-tiller, boot, and bloom growth stages reduced red rice seed germination, regardless of timing (Brommer et al. 1998). Simulated imazamox drift applications applied to wheat at the jointing and flowering growth stages had no effect on wheat seed germination (Deeds et al. 2006).

Glufosinate is a nonselective, foliar applied, postemergence herbicide used to control annual and perennial weeds in non-crop areas and for weed control in glufosinate-resistant crops (Senseman 2007d). The mechanism of action of glufosinate is the inhibition of the enzyme glutamine synthetase (EC 6.3.1.2) that converts glutamate and ammonia to glutamine (Lea et al. 1984; Senseman 2007d). This inhibition of glutamine synthetase results in a toxic accumulation of ammonia in treated plants and inhibition of photosystems I and II (Sauer et al. 1987; Senseman 2007d; Tachibana et al. 1986; Wild et al. 1987).

The symptoms expressed in plants from the inhibition of glutamine synthase are chlorosis and wilting within 3 to 5 d followed by necrosis in 7 to 14 d after application to susceptible species (Senseman 2007d). The rate of symptom development is increased in bright sunlight, high humidity, and moist soil.

Glufosinate applied to rice at a simulated drift rate of 53 g/ha reduced rice yield 30% (Ellis et al. 2003). When glufosinate was applied to grain sorghum at 1, 3, 10, and 33% of its labeled use rate only the 10 and 33% rates resulted in reduced grain sorghum yield (Al-Khatib et al. 2003). Rice seed germination was reduced by simulated glufosinate drift when evaluated at 16 C (Ellis et al. 2003). Glufosinate applied for preharvest desiccation of grain sorghum did not effect grain sorghum seed germination (Bovey et al. 1999). Bennett and Shaw (2000) found that glufosinate applied preharvest in soybean reduced seed germination of sicklepod (*Senna obtusifolia* (L) Irwin and Barnaby) and pitted morningglory (*Ipomoea lacunosa* L.).

A need exists to evaluate the effects of a glufosinate, glyphosate, imazamox, or imazethapyr drift event on rice. The objectives of this research were to evaluate the effects of simulated drift of these herbicides applied to rice during the primary rice crop on the crop response and impact on the seed produced on treated rice in the primary and ratoon rice crops.

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Chapter 2

Response of Rice to Glyphosate Drift

Introduction

Glyphosate is a nonselective, foliar applied, postemergence herbicide used to control annual and perennial weeds in preplant burndown applications and for weed control in glyphosate-resistant crops (Senseman 2007). The use of glyphosate has greatly increased with the introduction and extensive acceptance of glyphosate-resistant canola (*Brassica napus* L.), cotton (*Gossypium hirsutum* L.), corn (*Zea mays* L.), and soybean [*Glycine max* (L.) Merr.] (Shaner 2000).

The mechanism of action of glyphosate is the inhibition of 5-enolpyruvylshikimate-3-phosphate (EPSP) synthase (EC 2.5.1.19) which produces EPSP from shikimate-3-phosphate and phosphoenolpyruvate in the shikimic acid pathway (Amrhein et al. 1980; Amrhein et al. 1983; Boocock and Coggins 1983; Herrmann and Weaver 1999; Hollander-Czytko and Amrhein 1987; Jakeman et al. 1998; Schonbrunn et al. 2001; Senseman 2007). The inhibition of EPSP synthase is accomplished by glyphosate competing with phosphoenolpyruvate to bind with the shikimate-3-phosphate:EPSP synthase complex to form EPSP (Amrhein et al. 1980; Boocock and Coggins 1983; Herrmann and Weaver 1999; Jaworski 1972; Rubin et al. 1982; Schonbrunn et al. 2001). This outcompeting of phosphoenolpyruvate inhibits the formation of EPSP, which in turn inhibits the formation of the aromatic amino acids tryptophan, tyrosine, and phenylalanine which are needed for protein synthesis.

The symptoms expressed in plants from the inhibition of EPSP synthase are that growth is inhibited soon after application followed by general foliar chlorosis and necrosis within 4 to 7 days in highly susceptible species (Senseman 2007). Visual symptoms occur within 10 to 20 days for less susceptible species, appearing first and most pronounced in immature leaves and growing points in the form of chlorosis. Also, regrowth of treated

perennial and woody species often appears deformed and multiple shoots may develop at the nodes.

Rice (*Oryza sativa* L.) is a major crop produced in the four state region of Arkansas, Louisiana, Mississippi, and Texas, with these states accounting for 76% of the 1.2 million total hectares of rice planted in the U.S. and 70% of the \$3.4 billion of total value of rice produced in the U.S. in 2008 (NASSa 2009; NASSb 2009). That same year, approximately 98% of Louisiana's 424,920 hectares of soybean were planted in glyphosate-resistant soybean cultivars (Ronald J. Levy, Jr.¹, personal communication). Since many of the rice producing parishes in Louisiana also produce glyphosate-resistant soybean, corn, and cotton (NASSc 2009), the potential exists for off-target herbicide drift from one of these crops to rice.

In 2003 to 2005, glyphosate ranked second among herbicides listed in all Pesticide Investigation Reports processed by the Louisiana Department of Agriculture and Forestry (LDAF) and was the most listed herbicide in reports processed in 2006 to 2008 (Lisa Gautreaux², personal communication). Averaged over the ten year period 1999 to 2008, the LDAF processed 76 Pesticide Investigation Reports per year listing ground or aerial applicators in Louisiana, and, on average, six reports per year involved rice. However, it was reported that, in 2009, at least 50 rice fields were suspected of being affected by glyphosate drift and at least 25 rice fields were suspected of being affected by imazethapyr drift, many of which were not reported to the LDAF (Ronald J. Levy, Jr.¹, personal communication; John K. Saichuk³, personal communication). Therefore, the number of rice fields actually affected by a

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drift event each year may be underrepresented by the number of official complaints processed by the LDAF.

It has been reported that fine spray droplets less than 150 μm in size have a greater potential to drift off-target (Hanks 1995; SDTF 1997). The use of adjuvants and selection of proper spray nozzle type, size, and application pressure can be beneficial in reducing the amount of fine spray droplets in the spray cloud (Hanks 1995; Jones et al. 2007; Nuyttens et al. 2007; VanGessel and Johnson 2005). This increase in droplet size can reduce the potential for off-target drift from droplets larger than 150 μm ; however, environmental conditions at the time of herbicide application can also impact the off-target drift of spray solutions (Bouse et al. 1976; Crabbe et al. 1994; Thistle 2004).

Wind speed and direction may be considered the two most important factors affecting spray droplets in the atmosphere, a stable atmosphere may be the third most important factor (Thistle 2004). A stable atmosphere, commonly referred to as an inversion, is an atmosphere that has a change of temperature with a change in elevation in the atmosphere. In a stable atmosphere, warm air overlies cool air. If air in a particular layer is displaced upward or downward it will be colder or warmer, respectively, than the immediately adjacent layer it enters and thus return to its layer of origin. If a herbicide application is made during an inversion scenario, the fine droplets that do not succumb to gravity will remain in the air layer in which they are applied due to the lack of layers mixing. The droplets in this layer can be very concentrated and horizontally may move off-target great distances.

Ultra-low-volume applications made during a stable atmosphere produced 35% more herbicide drift than applications made during turbulent atmosphere with light wind speeds (Crabbe et al. 1994). The conditions in which the greatest drift occurred were in moderately stable conditions with wind at 3

m/s resulting in off-target drift 71% at 400 m and 50% at 2200 m from the application site and in slightly stable conditions with wind at 5 m/s resulting in off-target drift 77% at 400 m and 27% at 2200 m from the application site. It is recommended that herbicide applications should be avoided during the early morning and late evening as these times are most favorable for the development of inversion conditions (Crabbe et al. 1994; Thistle 2004).

Through the use of simulated herbicide drift studies, the potential effects of glyphosate drift to rice can be evaluated. In previous research, simulated drift studies varying the spray volume proportionally with reduced herbicide rates to simulate herbicide drift resulted in increased crop injury compared with the same herbicide rate applied in a constant spray volume (Banks and Schroeder 2002; Ellis et al. 2002; Ramsdale et al. 2003; Roider et al. 2008). Banks and Schroeder (2002) reported varying spray volume proportionally with herbicide dosage, thus maintaining constant herbicide concentration in the spray, would change the response of sweet corn to glyphosate when compared with a constant spray volume where herbicide rate would vary and be more dilute in the carrier. The no-effect glyphosate rate for sweet corn was 0.046 kg ae/ha when using a spray volume proportional to the reduced glyphosate rate; however, the no-effect glyphosate rate was four times greater when glyphosate was applied in a constant spray volume. Ellis et al. (2002) reported glyphosate applied to corn at 12.5 and 6.3% of the labeled use rate in a proportional spray volume and a constant spray volume produced results similar to those observed by Banks and Schroeder (2002). The use of a constant spray volume in drift research may underestimate the effects of off-target drift to susceptible crops.

Simulated drift applications of glyphosate applied to rice at the two- to three-leaf and panicle differentiation growth stages in a constant spray volume reduced rice yield 67 to 99% and 29 to 54%, respectively (Ellis et al.

2003). Kurtz and Street (2003) reduced rice yield with simulated glyphosate drift applied to rice at mid-tiller, panicle initiation, and boot growth stages.

Some studies evaluating the effects of glyphosate on seed weight, germination, and viability have been conducted. Brommer et al. (1998) reported applications of glyphosate and imazethapyr to red rice (*Oryza sativa* L.) reduced red rice seed germination, regardless of timing. The use of glyphosate as a preharvest desiccant in grain sorghum [*Sorghum bicolor* (L.) Moench.] reduced grain sorghum seed germination when planted after treatment (Baur et al. 1977). Glyphosate applied during reproductive growth stages to giant foxtail (*Setaria faberi* Herrm.) and velvetleaf (*Abutilon theophrasti* Medik.) resulted in reduced seed germination of both species (Biniak and Aldrich 1986). Roider et al. (2007) reported glyphosate applied to wheat at first node, boot, and early flowering growth stages resulted in 16 to 36 percent reductions in seed weight. A need exists to evaluate the possible effects of a glyphosate drift event on rice crop seed germination and seedling vigor.

Even though published studies evaluating the effects of simulated glyphosate drift on rice exist (Ellis et al. 2003; Koger et al. 2005; Kurtz and Street 2003), none of these studies were conducted using spray volumes that vary proportionally with reduced herbicide dosage. The objectives of this research were to evaluate the effects of simulated glyphosate drift applied to rice during the primary rice crop on the crop response and impact on the seed produced on treated rice in the primary and ratoon rice crops.

Materials and Methods

Simulated Glyphosate Drift Field Study. A study was conducted on rice grown in 2005 through 2007 at the LSU AgCenter Rice Research Station near Crowley, Louisiana on a Crowley silt loam (fine montmorillonitic, thermic Typic Albaqualf) with pH 5.5 and 1.2% organic matter. Field preparation consisted

of a fall and spring disking and two passes in opposite directions with a two-way bed conditioner equipped with rolling baskets and S-tine harrows 15 cm deep. The long grain rice cultivar 'Cocodrie' was drill-seeded March 28 to April 17 in 2005 through 2007. Plots consisted of twelve-18 cm spaced rows 6 m long.

The experimental design was an augmented two-factor factorial arrangement of treatments in a randomized complete block with four replications. Factor A consisted of glyphosate applied at simulated drift rates of 6.3 and 12.5% of the labeled usage rate of 863 g ae/ha, or 54 and 108 g/ha, respectively. Factor B consisted of application timings at different growth stages: one-tiller, panicle differentiation (PD), boot, and physiological maturity. Each herbicide application was made with the spray volume varying proportionally to herbicide dosage based on a constant spray volume of 234 L/ha. The 12.5% herbicide rate was applied at a spray volume of 29 L/ha and the 6.3% herbicide rate was applied at a spray volume of 15 L/ha. Each application was made with a tractor-mounted CO₂-pressurized sprayer calibrated to deliver a constant carrier volume with speed adjusted to vary application rate and equipped with Teejet^{®4} TX-2 Conejet[®] 800033 nozzles. A ratoon rice crop was not produced in 2006 due to unfavorable weather following primary crop harvest.

The study area was maintained weed-free using clomazone at 420 g ai/ha applied preemergence followed by propanil at 4483 g ai/ha plus halosulfuron at 53 g ai/ha applied postemergence. For the primary rice crop a preplant application of 280 kg/ha of 8-24-24 (N-P₂O₅-K₂O) fertilizer and a pre-flood application of 365 kg/ha 46-0-0 urea fertilizer were applied to the study area and for the ratoon rice crop a pre-flood application of 100 kg/ha 46-0-0 urea fertilizer was applied to the study area to maintain proper fertility

⁴ Spraying Systems Co., P. O. Box 7900, Wheaton, IL 60187.

and to maximize yields in the primary and ratoon crops. Standard agronomic and pest management practices were implemented throughout the growing season to maximize yield.

Rice plant height and rice injury in the primary rice crop were obtained 7 days after herbicide treatment (DAT) and continued weekly for 28 DAT. Rice plant height was obtained by measuring four plants per plot from the soil surface to the tip of the extended uppermost emerged leaf or extended rice panicle. Rice injury was evaluated based on chlorosis and necrosis of foliage and reduced plant height using a scale of 0 to 100% where 0 = no injury and 100 = plant death. Rice plant height at primary crop harvest and rough rice yield, 100-count seed weight, and stem and panicle counts for the primary and ratoon crop were also obtained. Whole plots were harvested using a mechanical plot harvester and rough rice yield was adjusted to 12% moisture. Stem and panicle counts were calculated by hand harvesting a 0.46 m section of row and determining the number of stems present at the mid-height of the plants and the number of panicles with bases emerged beyond the sheath of the flag leaf or the last leaf to emerge prior to the panicle.

All data were subjected to the Mixed Procedure of SAS (SAS 2003). Year, replications (nested within year), and all interactions containing either of these effects were considered random effects. Application timing and rate were considered fixed effects. Considering year or combination of year as random effects permits inferences about treatments over a range of environments (Carmer et al. 1989; Leon et al. 2008). Type III statistics were used to test all possible effects of fixed factors (application timing and rate) and least square means were used for mean separation at the 5% probability level ($p \leq 0.05$).

Seed Germination Study. The germination potential of seed collected from grain harvested in the simulated glyphosate drift field study at primary crop harvest, 2005 through 2007, and at ratoon crop harvest, 2005 and 2007, was

evaluated at multiple temperatures. Seed collected from each plot were air-dried and stored at 8 C. Germination temperatures evaluated were 13, 16, 19, 22, and 25 C. Temperature selection and germination testing procedure for this study were based on procedures previously described by Webster et al. (2003) and follow germination procedures recommended by Association of Official Seed Analysts (AOSA) (AOSA 2006). Temperature selection was based on 19 C being the historical mean 10-cm soil temperature in Crowley, LA on April, 1, which corresponds to 50% of the rice being planted across the state (Webster et al. 2003).

One hundred seeds from each field plot were prepared by soaking for 30 min in a 50:50 (v/v) solution of chlorine bleach and distilled water to decrease seedling diseases. After soaking, seeds were triple rinsed with distilled water. After seed preparation, seeds were placed in a 10 cm plastic Petri dish between two 9 cm germination blotters⁵. Next, 10 ml of carboxin (5,6-dihydro-2-methyl-N-phenyl-1,4-oxathiin-3-carboxamide) plus thiram (tetramethylthiuram disulfide) plus distilled water solution (52 ml of a 10% carboxin and 10% thiram premix liquid fungicide combined with 948 ml distilled water) was applied in each Petri dish to reduce seedling diseases. Petri dishes were sealed with Parafilm M⁶ to prevent moisture loss and placed in a constant-temperature growth chamber in total darkness. Germination counts were taken 5, 9, and 14 d after initiation (DAI) of the study. A seed was considered germinated if the radical had reached a length of 1 mm.

Seed germination data were arranged as repeated measures and subjected to the Mixed Procedure of SAS (SAS 2003). Year, replications (nested within year), DAI (nested within replications), and all interactions containing either of these effects were considered random effects. Application timing

⁵ Anchor Steel Blue Seed Germination Blotter®, SDB 3.5. Anchor Paper Company, 480 Broadway, St. Paul, MN 55101.

⁶ Parafilm M®. Pechiney Plastic Packaging, Menash, WI 54952.

and rate and germination temperature were considered fixed effects. Considering year or combination of year as random effects permits inferences about treatments over a range of environments (Carmer et al. 1989; Leon et al. 2008). Type III statistics were used to test all possible effects of fixed factors (application timing and rate and germination temperature) and least square means were used for mean separation at the 5% probability level ($p \leq 0.05$).

Seedling Vigor Study. Vigor of seedlings from grain collected at primary crop harvest in the simulated glyphosate drift field study in 2006 and 2007 was examined. Seedling vigor, as defined by AOSA (AOSA 2002), is "seedling vigor comprises those seed properties which determine the potential for rapid, uniform emergence, and development of normal seedlings under a wide range of field conditions" and it is used as a measure of seed quality by producers. Since there is no accepted standard vigor test for rice, vigor testing procedures for this study were based on procedures previously described by Webster et al. (2003).

Approximately 100 seeds from each field plot were prepared by soaking for 30 min in a 50:50 (v/v) solution of chlorine bleach and distilled water to decrease seedling diseases. After soaking, seeds were triple rinsed with distilled water. Following seed preparation, seeds were pre-germinated by soaking in distilled water for 24 h. Ten pre-germinated seeds from each field plot were placed on a single sheet of nontreated germination paper⁷ cut to fit a 12 by 23 by 0.3 cm acrylic sheet. Germination paper was moistened by submerging in distilled water for 5 seconds to facilitate adherence to the acrylic sheet and provide residual moisture to rice seeds. Seeds were placed along the center of germination paper oriented with the radical end of the seed toward the lower half of the sheet. A one-ply paper towel strip was

⁷ Anchor Steel Blue Seed Germination Blotters®, SDB 1924. Anchor Paper Company, 480 Broadway, St. Paul, MN 55101.

placed over the seed, and 5 ml of a mancozeb [ethylene (bis)-dithiocarbamate] plus distilled water solution (dry formulation of mancozeb at 1640 mg ai/L distilled water) was applied on top of the strip to reduce seedling diseases. The plated seeds were then placed vertically in a rack and then placed in a 30 by 51 by 5 cm dish with 1,420 ml of distilled water to allow for evaporation. The dish and racks of plates were wrapped in plastic wrap to prevent desiccation. The glass dish was placed in a constant temperature growth chamber at 21 C for 12 d in total darkness. At the end of 12 d, shoot lengths were measured and an average of the 10 shoot lengths was obtained for data analysis.

Seedling vigor data were subjected to the Mixed Procedure of SAS (SAS 2003). Year, replications (nested within year), and all interactions containing either of these effects were considered random effects. Application timing and rate were considered fixed effects. Considering year or combination of year as random effects permits inferences about treatments over a range of environments (Carmer et al. 1989; Leon et al. 2008). Type III statistics were used to test all possible effects of fixed factors (application timing and rate) and least square means were used for mean separation at the 5% probability level ($p \leq 0.05$).

Results and Discussion

Simulated Glyphosate Drift Field Study. A crop injury response was observed in the primary crop (Table 2.1). When applied at an equal rate, rice crop injury was higher when applied to the one-tiller stage of rice, the earliest timing evaluated in this study. At the one-tiller stage some recovery or regrowth was observed when treated with the low rate of glyphosate; however, the 108 g/ha rate of glyphosate had a greater impact on one-tiller rice. When treatments were delayed to the PD and boot timings visual injury was below 20% except for the 21 and 28 DAT evaluation at the PD timing when

Table 2.1. Effects of simulated glyphosate drift application rate and timing on primary rice crop injury 7, 14, 21, and 28 days after treatment (DAT), 2005 through 2007, Crowley, Louisiana.^a

Glyphosate		Injury			
		7 DAT	14 DAT	21 DAT	28 DAT
rate ^b	Timing	%			
54	1-tiller	32 a	45 a	31 b	33 b
	PD	6 bc	12 bc	14 c	14 c
	Boot	5 bc	10 bc	10 cd	10 cd
	Maturity	0 c	0 c	0 d	0 d
108	1-tiller	37 a	56 a	49 a	52 a
	PD	9 b	19 b	30 b	35 b
	Boot	7 bc	11 bc	11 cd	11 cd
	Maturity	0 c	0 c	0 d	0 d
Nontreated		0 c	0 c	0 d	0 d

^a Means within a column followed by the same letter were not statistically different according to the t-test on difference of least square means at P = 0.05.

^b The 54 and 108 g/ha glyphosate rates were applied at spray volumes of 15 and 29 L/ha, respectively.

treated with 108 g/ha of glyphosate. This indicated that drift can be rate dependant when applied to earlier growth stages. As with actual drift events, identifying drift based on visual injury is more difficult as rice matures (Eric P. Webster⁸, personal communication). Similar findings were reported by Ellis et al. (2003) and Kurtz and Street (2003) where visual injury symptoms were more severe in rice treated with glyphosate during early vegetative growth stages than rice treated during late reproductive growth stages.

This reduction in visual injury during reproductive growth stages may be due to the translocation of glyphosate to meristematic tissue (Martin and Edgington 1981). This tissue is located in the internal portions of the rice plant during the reproductive stages of growth and would not be expressed on foliar tissue.

The injury symptoms observed in this study on plants treated at the one-tiller timing were a general chlorosis in the uppermost leaves to plant death. The newest leaf to emerge following treatment often emerged tightly rolled. Also, an overall stunting of plants was observed on plants treated at the one-tiller and PD timings (Table 2.2).

Visual symptomology observed on plants treated with glyphosate at PD and boot, often beyond the rating dates evaluated in this study, were various forms of foliar and inflorescence malformations. Foliar symptoms were plants having multiple shoots arising from the secondary nodes of the main stem and the flag leaf on the main stem and secondary shoots would often appear wrinkled, contorted, or rolled (Figure 2.1, 2.2). In some instances secondary shoots were stunted or both stunted and malformed. At maturity some panicles failed to fully exert beyond the flag leaf sheath or emerged

⁸ Eric P. Webster, Louisiana State University AgCenter Rice Weed Specialist, 104 M.B. Sturgis Hall, Baton Rouge, LA 70803.

Table 2.2. Effects of simulated glyphosate drift application rate and timing on primary crop rice plant height at 7, 14, 21, and 28 days after treatment (DAT) and at harvest, 2005 through 2007, as percent of the nontreated, Crowley, Louisiana.^a

Glyphosate		Rice plant height				
		7 DAT ^c	14 DAT ^d	21 DAT ^e	28 DAT ^f	Harvest ^g
rate ^b	Timing	% of nontreated				
g ae/ha						
54	1-tiller	69 b	75 e	82 c	79 cd	88 bc
	PD	95 a	87 cd	83 c	81 c	90 b
	Boot	97 a	94 ab	94 b	89 b	89 bc
	Maturity	102 a	97 ab	99 ab	102 a	100 a
108	1-tiller	70 b	65 f	74 d	75 cd	88 bc
	PD	95 a	85 d	75 d	73 d	85 c
	Boot	98 a	93 bc	93 b	90 b	88 bc
	Maturity	102 a	102 a	101 a	100 a	102 a
Nontreated		100 a	100 ab	100 a	100 a	100 a

^a Means within a column followed by the same letter were not statistically different according to the t-test on difference of least square means at P = 0.05.

^b The 54 and 108 g/ha glyphosate rates were applied at spray volumes of 15 and 29 L/ha, respectively.

^c Actual heights of nontreated rice for the 1-tiller, PD, boot, and maturity timings were 41, 64, 92, and 50 cm, respectively.

^d Actual heights of nontreated rice for the 1-tiller, PD, boot, and maturity timings were 54, 72, 99, and 56 cm, respectively.

^e Actual heights of nontreated rice for the 1-tiller, PD, boot, and maturity timings were 65, 79, 98, and 60 cm, respectively.

^f Actual heights of nontreated rice for the 1-tiller, PD, boot, and maturity timings were 70, 88, 97, and 64 cm, respectively.

^g Actual height of nontreated rice at primary crop harvest was 94 cm.



Figure 2.1. Symptoms observed with a boot application of 108 g ae/ha glyphosate.



Figure 2.2. Symptoms observed with a boot application of 54 g ae/ha glyphosate.



Figure 2.3. Symptoms observed with a boot application of 108 g ae/ha glyphosate.



Figure 2.4. Symptoms observed with a boot application of 108 g ae/ha glyphosate.

from the side of the sheath (Figure 2.3). Some of the inflorescence malformations were due to malformed panicle axis and partially emerged panicles due to fusing with the flag leaf sheath. Individual florets malformations that were observed were florets that were void of a developing grain with only a bleached lemma and palea remaining and individual florets with tips of the lemma excessively curved toward the palea (Figure 2.4) causing an appearance often referred to as "parrot beaked" when observed in association with the straighthead physiological disorder of rice (Groth et al. 2009).

A plant height response was observed in the primary rice crop with glyphosate applications (Table 2.2). When evaluated at 7, 14, 21, and 28 DAT, glyphosate applied to rice at the one-tiller stage resulted in reduced rice plant height, compared with the nontreated. This is similar to results observed through visual injury. Rice plant height was reduced at 14, 21, and 28 DAT when glyphosate was applied to rice at PD and at 21 and 28 DAT when glyphosate was applied at boot, regardless of rate. These findings support the trend of increased crop injury at earlier application timings. Similar findings were reported by Ellis et al. (2003) and Kurtz and Street (2003). However, glyphosate applied to rice at one-tiller, PD, and boot timings resulted in reduced rice plant height at primary crop harvest, 85 to 90% of the nontreated. Glyphosate applied to mature rice had no effect on rice plant height.

Stem and panicle counts in the primary and ratoon crops were affected by glyphosate applications (Table 2.3). Glyphosate applied at one-tiller, PD, and boot increased secondary plant stems in the primary crop resulting in an increase in stem count compared with the nontreated. However, panicle count was only increased in the primary crop when glyphosate was applied at both rates at PD and at the 108 g/ha rate at one-tiller. In

Table 2.3. Effects of simulated glyphosate drift application rate and timing on primary crop rice stem and panicle counts, 2005 through 2007, and ratoon crop rice stem and panicle counts, 2005 and 2007, as percent of the nontreated, Crowley, Louisiana.^a

Glyphosate		Primary crop counts		Ratoon crop counts	
		Stem	Panicle	Stem	Panicle
rate ^b	Timing	% of nontreated ^c			
g ae/ha		Stem	Panicle	Stem	Panicle
54	1-tiller	98 c	105 c	83 bc	76 b
	PD	147 b	161 b	96 bc	73 b
	Boot	161 b	104 c	198 a	242 a
	Maturity	100 c	106 c	106 b	96 b
108	1-tiller	139 b	150 b	67 c	65 b
	PD	196 a	197 a	113 b	103 b
	Boot	196 a	98 c	215 a	235 a
	Maturity	97 c	103 c	104 b	94 b
Nontreated		100 c	100 c	100 bc	100 b

^a Means within a column followed by the same letter were not statistically different according to the t-test on difference of least square means at P = 0.05.

^b The 54 and 108 g/ha glyphosate rates were applied at spray volumes of 15 and 29 L/ha, respectively.

^c Actual nontreated primary crop stem and panicle counts were 39 and 35 per 0.46 m of row, respectively, and actual nontreated ratoon crop stem and panicle counts were 44 and 32 per 0.46 m of row, respectively.

the ratoon crop, an increase in stem and panicle counts was only observed in rice treated at the boot stage (Table 2.3).

Glyphosate applied at one-tiller, PD, and boot reduced primary crop yield, compared with the nontreated (Table 2.4). The primary crop yield reduction resulting from an application of glyphosate at the boot timing was more severe than when applied to the earlier growth stages of rice evaluated in this study. Glyphosate applied at 54 and 108 g/ha at boot resulted in a primary crop yield 54 and 36% of the nontreated, respectively. However, the ratoon crop yield was 149 and 148% of the nontreated. This increase was due to glyphosate causing an excess of secondary stems to be produced on the upper plant nodes in the primary rice crop (Table 2.3). This excess of secondary stems did not produce panicles in the primary crop but did produce panicles in the ratoon crop. This response was not observed with rice treated at the other timings. However, when primary and ratoon crop yields were combined, the increase in ratoon crop yield did not compensate for the primary crop yield loss. These data indicate a drift event at the growth stages evaluated in this study reduced total crop yield regardless of glyphosate rate. Total yield was reduced when glyphosate was applied at the one-tiller, PD, and boot timings, compared to the nontreated (Table 2.4). Glyphosate applications at maturity had no effect on primary, ratoon, or total crop rough rice yield.

Though primary crop rice yield was reduced by simulated glyphosate drift applications at the one-tiller, PD, and boot timings, it appears that rice is most susceptible to glyphosate during the boot growth stage, which is similar to results reported by Kurtz and Street (2003). Rice producers in Louisiana may have the ability to recover some yield loss from a drift event occurring to rice during the boot growth stage by increasing ratoon crop yield; however, the reduction in total crop yield from a glyphosate drift

Table 2.4. Effects of simulated glyphosate drift application rate and timing on primary crop rice yield, 2005 through 2007, and ratoon and total crop rice yield, 2005 and 2007, as percent of the nontreated, Crowley, Louisiana.^a

Glyphosate rate ^b		Yield		
		Primary crop	Ratoon crop	Total crop
g ae/ha		% of nontreated ^c		
54	1-tiller	66 bc	62 c	54 bc
	PD	75 b	66 c	70 b
	Boot	54 c	149 a	57 bc
	Maturity	99 a	85 bc	93 a
108	1-tiller	55 c	86 bc	66 b
	PD	55 c	86 bc	52 bc
	Boot	36 d	148 a	44 c
	Maturity	96 a	92 b	93 a
Nontreated		100 a	100 b	100 a

^a Means within a column followed by the same letter were not statistically different according to the t-test on difference of least square means at P = 0.05.

^b The 54 and 108 g/ha glyphosate rates were applied at spray volumes of 15 and 29 L/ha, respectively.

^c Actual nontreated yield for the primary, ratoon, and total crops were 7000, 1300, and 8300 kg/ha, respectively.

event at the one-tiller, PD, or boot growth stages of rice can be significant.

Seed Germination and Seedling Vigor Studies. Simulated glyphosate drift applications did not affect rice seed weight in the primary rice crop; however, ratoon crop rice seed weight was affected (Table 2.5). Averaged across application rates, glyphosate applied at one-tiller reduced ratoon crop rice seed weight to 91% of the nontreated. It is expected that any unfilled or malformed grain observed on rice panicles on treated plants was separated and expelled by the mechanical plot harvester. This separation is similar to a commercial harvesting operation so any affect on seed weight, germination, or seedling vigor of harvested grain observed in this study is reflective of the impact expected on commercial seed rice producers. Studies conducted using hand-harvesting of seeds, such as Walker and Oliver (2008), which bypass a separation process, may misrepresent the impact of herbicides on seed in mechanically harvested grain crops.

Averaged across application rates, primary crop rice seed germination was not effected at 13, 16, or 19 C, compared with the nontreated (Table 2.6). Germination of primary crop rice seed at 22 C was reduced to 93 and 88% of the nontreated when glyphosate was applied at PD and boot, respectively. When evaluated at 25 C, primary crop rice seed germination was reduced to 92% of the nontreated when glyphosate was applied to rice at one-tiller and PD and to 82% of the nontreated when applied at boot.

Ratoon crop rice seed germination was not reduced, compared to the nontreated, at any temperature evaluated in this study (Table 2.7). Ellis et al. (2003) also observed a decrease in rice seed germination at various temperatures for seed collected from plants treated with a simulated glyphosate drift application. Glyphosate drift applications did not reduce primary crop seedling vigor (data not shown). These data indicate that rice seed weight may not be affected but seed germination may be, especially if

Table 2.5. Effects of simulated glyphosate drift application timing on primary crop seed weight, 2005 through 2007, and ratoon crop rice seed weight, 2005 and 2007, as percent of the nontreated, Crowley, Louisiana.^a

Glyphosate timing	100-seed weight	
	Primary crop	Ratoon crop
	————— % of nontreated ^{b,c} —————	
1-tiller	100 a	91 b
PD	98 a	100 a
Boot	99 a	101 a
Maturity	101 a	100 a
Nontreated	100 a	100 a

^a Means within a column followed by the same letter were not statistically different according to the t-test on difference of least square means at P = 0.05.

^b Data averaged across application rates of 54 and 108 g ai/ha glyphosate applied at spray volumes of 15 and 29 L/ha, respectively.

^c Actual nontreated 100-seed weight for the primary and ratoon crop were 2300 and 2000 mg, respectively.

Table 2.6. Effects of simulated glyphosate drift application timing on primary crop rice seed germination at various temperatures, 2005 through 2007, as percent of the nontreated, Crowley, Louisiana.^a

Glyphosate timing	Temperatures				
	13 C	16 C	19 C	22 C	25 C
	% of nontreated ^{b,c}				
1-tiller	89 a	98 a	93 a	97 a	92 b
PD	78 a	83 a	89 a	93 b	92 b
Boot	89 a	91 a	85 a	88 c	82 c
Maturity	89 a	98 a	91 a	100 a	100 a
Nontreated	100 a	100 a	100 a	100 a	100 a

^a Means within a column followed by the same letter were not statistically different according to the t-test on difference of least square means at P = 0.05.

^b Data averaged across application rates of 54 and 108 g ai/ha glyphosate applied at spray volumes of 15 and 29 L/ha, respectively.

^c Actual germination of the nontreated seed at 13, 16, 19, 22, and 25 C was 9, 42, 54, 73, and 66%, respectively.

Table 2.7. Effects of simulated glyphosate drift application timing on ratoon crop rice seed germination at various temperatures, 2005 and 2007, as percent of the nontreated, Crowley, Louisiana.^a

Glyphosate timing	Temperatures				
	13 C	16 C	19 C	22 C	25 C
	% of nontreated ^{b,c}				
1-tiller	167 a	150 a	116 a	115 a	108 a
PD	67 c	100 b	105 ab	104 c	102 b
Boot	133 ab	117 b	116 a	119 a	105 ab
Maturity	100 bc	89 b	92 b	108 bc	102 b
Nontreated	100 bc	100 b	100 b	100 c	100 b

^a Means within a column followed by the same letter were not statistically different according to the t-test on difference of least square means at P = 0.05.

^b Data averaged across application rates of 54 and 108 g ai/ha glyphosate applied at spray volumes of 15 and 29 L/ha, respectively.

^c Actual germination of the nontreated seed at 13, 16, 19, 22, and 25 C was 3, 18, 38, 48, and 60%, respectively.

the drift event occurs to the primary crop during the reproductive growth stages. However, it appears that if seeds germinate seedling vigor will not be adversely affected. If seed rice is affected by a drift event, extra caution should be taken before that seed is sold to producers.

In conclusion, simulated glyphosate drift applications at the one-tiller, PD, and boot timings resulted in reduced plant height and primary and total crop yield losses, with the greatest reduction in primary crop yield resulting from a simulated glyphosate drift application applied at the boot growth stage. Glyphosate applications to mature rice had no effect on rice plant height or yield. Seed weight and seed vigor of primary crop rice seed was not affected by simulated glyphosate drift applications; however, primary crop seed germination was reduced when glyphosate was applied at one-tiller, PD, and boot, with increased susceptibility at the boot growth stage. A reduction in ratoon crop rice seed weight by glyphosate applications was observed; however, ratoon crop rice seed germination was not reduced.

The ability to identify glyphosate drift on rice can be helpful to producers, Cooperative Extension Service personnel, crop consultants, and state regulatory agencies in distinguishing between herbicide drift and injury associated with soil fertility issues, diseases, and other disorders affecting rice. Misidentification of herbicide drift symptoms as injury associated with these factors can lead to loss in profitability if growers apply unnecessary applications of inputs to correct these factors when the symptoms present are actually a result of herbicide drift. The ability to correlate the symptoms observed to glyphosate drift also may assist state regulatory agencies in identifying the source of a herbicide drift event. If glyphosate can be identified by observation of plant symptoms this can reduce the cost associated with confirmation of a herbicide drift event through the use of diagnostic testing of foliar residue since most analytical facilities

charge per evaluation and the diagnostic tests involved are often herbicide specific.

A glyphosate drift event occurring to a producer's field at the one-tiller, PD, or boot growth stages of rice can reduce rice yield; however, this study indicates that a drift event occurring at the boot stage may be the most detrimental to yield. Rice receiving a drift event in vegetative growth stages, one-leaf to one-tiller, can often recover if stand is maintained at recommended densities (Eric P. Webster⁸, personal communication). However, a glyphosate drift event occurring to rice in the reproductive stage of growth may have little to no visual foliar injury and often symptoms may not appear until rice plants near crop maturity. This may lead to loss of yield and profitability due to continuing to supply crop inputs, such as increased fertilizer, insecticide, and fungicide applications, to a crop that has an already reduced yield potential. The negative effects of a glyphosate drift event occurring to a seed producer's field to rice in the PD or boot growth stages has the potential to be two-fold. The reduction in profitability the year of the event from reduced yield in combination with the reduction in seed germination has the ability to reduce profitability in the subsequent year's crop due to an increase in seeding rate to offset the reduced seed germination.

Caution should be taken when applying glyphosate near commercial rice fields, especially when making applications near rice in the reproductive growth stages. Though the effects of glyphosate drift on rice may not be immediately apparent by visual observation, the potential affect on grain yield and the germination potential of the harvested grain could be highly detrimental to rice producers.

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Chapter 3

Effects of Simulated Imazethapyr Drift on Non-Clearfield Rice Grown for Grain and Seed Rice

Introduction

In 1993, imidazolinone-resistant rice (*Oryza sativa* L.) was developed and exhibited tolerance to the imidazolinone class of herbicides (Croughan 1994; Pellerin et al. 2004; Webster and Masson 2001). Imazethapyr is a selective herbicide used to control annual and perennial weeds in soybean [*Glycine max* (L.) Merr.], edible legumes, and imidazolinone-resistant crops (Senseman 2007).

The mechanism of action for imazethapyr is inhibition of acetolactase synthase (ALS) (EC 4.1.3.18) also called acetohydroxyacid synthase (AHAS), a key enzyme in the biosynthesis of the branched-chain amino acids isoleucine, leucine, and valine (Muhitch et al. 1987; Senseman 2007; Shaner 1991; Shaner et al. 1984; Stidham 1991; Stidham and Singh 1991). Plant death results from events occurring in response to ALS inhibition, specifically the inhibition of isoleucine, leucine and valine, but the actual sequence of phytotoxic processes is unclear (Shaner 1991; Stidham and Singh 1991). Some secondary effects may include disruption of photosynthate translocation, hormone imbalance due to interruption of source/sink relationships, and interference in DNA synthesis and cell growth.

The symptoms expressed from this toxicity are growth is inhibited within a few hours of herbicide application, meristematic areas become chlorotic, followed by a slow general foliar chlorosis and necrosis (Shaner 1991). This injury to meristematic areas can be attributed to inhibition of branched-chain amino acids in the meristematic region. Even though plants have the ability to scavenge amino acids from pre-existing proteins, the meristematic region lacks the protein reserve pools that are available in the

mature regions of the plant. Injury symptoms usually appear within 7 to 14 d for susceptible species.

Rice is a major crop produced in the four state region of Arkansas, Louisiana, Mississippi, and Texas, with these states accounting for 76% of the 1.2 million total hectares of rice planted in the U.S. and 70% of the \$3.4 billion of total value of rice produced in the U.S. in 2008 (NASSa 2009; NASSb 2009). Louisiana planted approximately 184,000 hectares of rice in 2008 with approximately 59% planted to imidazolinone-resistant rice cultivars or hybrids (LSUA 2009). Since many of the rice producing parishes in Louisiana produce imidazolinone-resistant and conventional rice, the potential exists for off-target drift of imazethapyr to conventional rice.

Averaged over the ten year period 1999 to 2008, the Louisiana Department of Agriculture and Forestry processed 76 Pesticide Investigation Reports per year listing ground or aerial applicators in Louisiana, and, on average, six reports per year involved rice (Lisa Gautreaux¹, personal communication). However, it was reported that, in 2009, at least 50 rice fields were suspected of being affected by glyphosate drift and at least 25 rice fields were suspected of being affected by imazethapyr drift, many of which were not reported to the LDAF (Ronald J. Levy, Jr.², personal communication; John K. Saichuk³, personal communication). Therefore, the number of rice fields actually affected by a drift event each year may be underrepresented by the number of official complaints processed by the LDAF.

It has been reported that fine spray droplets less than 150 μm in size have a greater potential to drift off-target (Hanks 1995; SDTF 1997). The

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use of adjuvants and selection of proper spray nozzle type, size, and application pressure can be beneficial in reducing the amount of fine spray droplets in the spray cloud (Hanks 1995; Jones et al. 2007; Nuyttens et al. 2007; VanGessel and Johnson 2005). This increase in droplet size can reduce the potential for off-target drift from droplets larger than 150 μm ; however, environmental conditions at the time of herbicide application can also impact the off-target drift of spray solutions (Bouse et al. 1976; Crabbe et al. 1994; Thistle 2004).

Wind speed and direction may be considered the two most important factors affecting spray droplets in the atmosphere, a stable atmosphere may be the third most important factor (Thistle 2004). A stable atmosphere, commonly referred to as an inversion, is an atmosphere that has a change of temperature with a change in elevation in the atmosphere. In a stable atmosphere, warm air overlies cool air. If air in a particular layer is displaced upward or downward it will be colder or warmer, respectively, than the immediately adjacent layer it enters and thus returns to the original layer. If a herbicide application is made during an inversion scenario, the fine droplets that do not succumb to gravity will remain in the layer in which they are deposited due to the lack of layers mixing. The droplets in this layer can be very concentrated and horizontally may move off-target great distances.

Ultra-low-volume applications made during a stable atmosphere produced 35% more herbicide drift than applications made during a turbulent atmosphere with light wind speeds (Crabbe et al. 1994). The conditions in which the greatest drift occurred were in moderately stable conditions with wind at 3 m/s resulting in off-target drift 71% at 400 m and 50% at 2200 m from the application site and in slightly stable conditions with wind at 5 m/s resulting in off-target drift 77% at 400 m and 27% at 2200 m from the application site. It is recommended that herbicide applications should be

avoided during the early morning and late evening because these times are most favorable for the development of inversion conditions (Crabbe et al. 1994; Thistle 2004).

Through the use of simulated herbicide drift studies, the potential effects of imazethapyr drift to rice can be evaluated. In previous research, simulated drift studies varying the spray volume proportionally with reduced herbicide rates to simulate herbicide drift resulted in increased crop injury compared with the same herbicide rate applied in a constant spray volume (Banks and Schroeder 2002; Ellis et al. 2002; Ramsdale et al. 2003; Roider et al. 2008). Banks and Schroeder (2002) reported varying spray volume proportionally with herbicide dosage, thus maintaining constant herbicide concentration in the spray, would change the response of sweet corn (*Zea mays* L.) to glyphosate when compared with a constant spray volume where herbicide rate would vary and be more dilute in the carrier. The no-effect glyphosate rate for sweet corn was 0.046 kg ae/ha when using a spray volume proportional to the reduced glyphosate rate; however, the no-effect glyphosate rate was four times greater when glyphosate was applied in a constant spray volume.

A simulated drift application of the commercial herbicide premix of imazethapyr plus imazapyr affected rice plant height and yield; however, simulated drift of the imazethapyr plus imazapyr premix did not affect yield when applied to corn (Bond et al. 2006). Al-Khatib et al. (2003) reported imazethapyr applied at various times within 30 d of planting resulted in reduced grain sorghum (*Sorghum bicolor* L.) yield.

Rapid rice seed germination and seedling growth can enhance plant stand establishment in commercial fields (Krishnasamy and Seshu 1989; Pollock and Roos 1972; Wright 1980). Clay and Griffin (2000) reported late season applications of glyphosate to common cocklebur (*Xanthium strumarium* L.), hemp sesbania [*Sesbania herbacea* (Mill.) McVaugh], and sicklepod [*Senna obtusifolia* (L.) Irwin and Barneby] reduced 100-seed weight and seedling

emergence when applied to plants at initial seed set but had no effect when applied to plants at physiological maturity. Glyphosate applied to wheat at first node, boot, and early flowering growth stages resulted in 16 to 36% reductions in seed weight (Roider et al. 2007). A negative correlation between 100-count seed weight and rice seed germination has been observed (Krishnasamy and Seshu 1989). Applications of glyphosate and imazethapyr to red rice (*Oryza sativa* L.) in the two- to three-tiller, boot, and bloom growth stages reduced red rice seed germination, regardless of timing (Brommer et al. 1998). The use of glyphosate as a preharvest desiccant in grain sorghum reduced grain sorghum seed germination (Baur et al. 1977). A need exists to evaluate the possible effects of an imazethapyr drift event on rice crop seed germination and seedling vigor.

Even though published studies evaluating the effects of simulated imazethapyr drift exist (Al-Khatib et al. 2003; Bond et al. 2006), none of these studies were conducted using spray volumes proportional with reduced herbicide dosage. The objectives of this research were to evaluate the effects of simulated imazethapyr drift applied to rice during the primary rice crop on the crop response and impact on the seed produced on treated rice in the primary and ratoon rice crops.

Materials and Methods

Simulated Imazethapyr Drift Field Study. A study was conducted on rice grown in 2005 through 2007 at the LSU AgCenter Rice Research Station near Crowley, Louisiana on a Crowley silt loam (fine montmorillonitic, thermic Typic Albaqualf) with pH 5.5 and 1.2% organic matter. Field preparation consisted of a fall and spring disking and two passes in opposite directions with a two-way bed conditioner equipped with rolling baskets and S-tine harrows 15 cm deep. The long grain rice cultivar 'Cocodrie' was drill-seeded March 28 to April 17 in 2005 through 2007. Plots consisted of twelve-18 cm spaced rows 6 m long.

The experimental design was an augmented two-factor factorial arrangement of treatments in a randomized complete block with four replications. Factor A consisted of imazethapyr applied at simulated drift rates of 6.3 and 12.5% of the labeled usage rate of 70 g ai/ha, or 4.4 and 8.7 g/ha, respectively. Factor B consisted of application timings at different growth stages: one-tiller, panicle differentiation (PD), boot, and physiological maturity. Each herbicide application was made with the spray volume varying proportionally to herbicide dosage based on a constant spray volume of 234 L/ha. The 12.5% herbicide rate was applied at a spray volume of 29 L/ha and the 6.3% herbicide rate was applied at a spray volume of 15 L/ha. Each application was made with a tractor-mounted CO₂-pressurized sprayer calibrated to deliver a constant carrier volume with speed adjusted to vary application rate and equipped with Teejet^{®4} TX-2 Conejet[®] 800033 nozzles. A ratoon rice crop was not produced in 2006 due to unfavorable weather following primary crop harvest.

The study area was maintained weed-free using clomazone at 420 g ai/ha applied preemergence followed by propanil at 4483 g ai/ha plus halosulfuron at 53 g ai/ha applied postemergence. For the primary rice crop a preplant application of 280 kg/ha of 8-24-24 (N-P₂O₅-K₂O) fertilizer and a pre-flood application of 365 kg/ha 46-0-0 urea fertilizer were applied to the study area and for the ratoon rice crop a pre-flood application of 100 kg/ha 46-0-0 urea fertilizer was applied to the study area to maintain proper fertility and to maximize yields in the primary and ratoon crops. Standard agronomic and pest management practices were implemented throughout the growing season to maximize yield.

Rice plant height and rice injury in the primary rice crop were obtained 7 days after herbicide treatment (DAT) and continued weekly for 28

⁴ Spraying Systems Co., P. O. Box 7900, Wheaton, IL 60187.

DAT. Rice plant height was obtained by measuring four plants per plot from the soil surface to the tip of the extended uppermost emerged leaf or extended rice panicle. Rice injury was evaluated based on chlorosis and necrosis of foliage and reduced plant height using a scale of 0 to 100% where 0 = no injury and 100 = plant death. Rice plant height at primary crop harvest and rough rice yield, 100-count seed weight, and stem and panicle counts for the primary and ratoon crop were also obtained. Whole plots were harvested using a mechanical plot harvester and rough rice yield was adjusted to 12% moisture. Total stem and panicle counts were calculated by hand harvesting a 0.46 m section of row and determining the number of stems present at the mid-height of the plants and the number of panicles with bases emerged beyond the sheath of the flag leaf, the last leaf to emerge prior to the panicle.

All data were subjected to the Mixed Procedure of SAS (SAS 2003). Year, replications (nested within year), and all interactions containing either of these effects were considered random effects. Application timing and rate were considered fixed effects. Considering year or combination of year as random effects permits inferences about treatments over a range of environments (Carmer et al. 1989; Leon et al. 2008). Type III statistics were used to test all possible effects of fixed factors (application timing and rate) and least square means were used for mean separation at the 5% probability level ($p \leq 0.05$).

Seed Germination Study. The germination potential of seed collected from grain harvested in the simulated imazethapyr drift field study at primary crop harvest, 2005 through 2007, and at ratoon crop harvest, 2005 and 2007, was evaluated at multiple temperatures. Seed collected from each plot was air-dried and stored at 8 C. Germination temperatures evaluated were 13, 16, 19, 22, and 25 C. Temperature selection and germination testing procedure for this study were based on procedures previously described by Webster et

al. (2003) and follow standard germination procedures recommended by Association of Official Seed Analysts (AOSA) (AOSA 2006). Temperature selection was based on 19 C being the historical mean 10-cm soil temperature in Crowley, LA on April, 1, which corresponds to 50% of the rice being planted across the state (Webster et al. 2003).

One hundred seeds from each field plot were prepared by soaking for 30 min in a 50:50 (v/v) solution of chlorine bleach and distilled water to decrease seedling diseases. After soaking, seeds were triple rinsed with distilled water. After seed preparation, seeds were placed in a 10 cm plastic Petri dish between two 9 cm germination blotters⁵. Next, 10 ml of carboxin (5,6-dihydro-2-methyl-*N*-phenyl-1,4-oxathiin-3-carboxamide) plus thiram (tetramethylthiuram disulfide) plus distilled water solution (52 ml of a 10% carboxin and 10% thiram premix liquid fungicide combined with 948 ml distilled water) was applied in each Petri dish to reduce seedling diseases. Petri dishes were sealed with Parafilm M⁶ to prevent moisture loss and placed in a constant-temperature growth chamber in total darkness. Germination counts were taken 5, 9, and 14 d after initiation (DAI) of the study. A seed was considered germinated if the radical had reached a length of 1 mm.

Seed germination data were arranged as repeated measures and subjected to the Mixed Procedure of SAS (SAS 2003). Year, replications (nested within year), DAI (nested within replications), and all interactions containing either of these effects were considered random effects. Application timing and rate and germination temperature were considered fixed effects. Considering year or combination of year as random effects permits inferences about treatments over a range of environments (Carmer et al. 1989; Leon et al. 2008). Type III statistics were used to test all possible effects of

⁵ Anchor Steel Blue Seed Germination Blotter®, SDB 3.5. Anchor Paper Company, 480 Broadway, St. Paul, MN 55101.

⁶ Parafilm M®. Pechiney Plastic Packaging, Menash, WI 54952.

fixed factors (application timing and rate and germination temperature) and least square means were used for mean separation at the 5% probability level ($p \leq 0.05$).

Seedling Vigor Study. Vigor of seedlings from grain collected at primary crop harvest in the simulated imazethapyr drift field study in 2006 and 2007 was examined. Seedling vigor, as defined by AOSA (AOSA 2002), is "seedling vigor comprises those seed properties which determine the potential for rapid, uniform emergence, and development of normal seedlings under a wide range of field conditions" and it is used as a measure of seed quality by producers. Since there is no accepted standard vigor test for rice, vigor testing procedures for this study were based on procedures previously described by Webster et al. (2003).

Approximately 100 seeds from each field plot were prepared by soaking for 30 min in a 50:50 (v/v) solution of chlorine bleach and distilled water to decrease seedling diseases. After soaking, seeds were triple rinsed with distilled water. Following seed preparation, seeds were pre-germinated by soaking in distilled water for 24 h. Ten pre-germinated seeds from each field plot were placed on a single sheet of nontreated germination paper⁷ cut to fit a 12 by 23 by 0.3 cm acrylic sheet. Germination paper was moistened by submerging in distilled water for 5 seconds to facilitate adherence to the acrylic sheet and provide residual moisture to rice seeds. Seeds were placed along the center of germination paper oriented with the radical end of the seed toward the lower half of the sheet. A one-ply paper towel strip was placed over the seed, and 5 ml of a mancozeb [ethylene (bis)-dithiocarbamate] plus distilled water solution (dry formulation of mancozeb at 1640 mg ai/L distilled water) was applied on top of the strip to reduce seedling diseases. The plated seeds were then placed vertically in a rack and then placed in a

⁷ Anchor Steel Blue Seed Germination Blotters®, SDB 1924. Anchor Paper Company, 480 Broadway, St. Paul, MN 55101.

30 by 51 by 5 cm dish with 1,420 ml of distilled water to allow for evaporation. The dish and racks of plates were wrapped in plastic wrap to prevent desiccation. The glass dish was placed in a constant temperature growth chamber at 21 C for 12 d in total darkness. At the end of 12 d, shoot lengths were measured and an average of the 10 shoot lengths was obtained for data analysis.

Seed vigor data were subjected to the Mixed Procedure of SAS (SAS 2003). Year, replications (nested within year), and all interactions containing either of these effects were considered random effects. Application timing and rate were considered fixed effects. Considering year or combination of year as random effects permits inferences about treatments over a range of environments (Carmer et al. 1989; Leon et al. 2008). Type III statistics were used to test all possible effects of fixed factors (application timing and rate) and least square means were used for mean separation at the 5% probability level ($p \leq 0.05$).

Results and Discussion

Simulated Imazethapyr Drift Field Study. A crop injury response was observed in the primary crop (Table 3.1). Imazethapyr applied at 4.4 and 8.7 g/ha at one-tiller resulted in crop injury of 28 to 39% at 7, 14, 21, and 28 DAT. When applications were delayed to the PD and boot stages crop injury was 20% or less except for rice treated at the boot stage at 28 DAT. An increase in injury from 21 to 28 DAT with rice treated with imazethapyr at boot was noted because necrosis of the flag leaf was observed at 28 DAT that was not present at 21 DAT. No response was observed on rice treated with imazethapyr at maturity. These data indicate that visual injury to rice is more severe when imazethapyr was applied during the early, vegetative growth stage of rice. As with actual drift events, identifying drift based on visual injury is more

Table 3.1. Effects of simulated imazethapyr drift application rate and timing on primary rice crop injury 7, 14, 21, and 28 days after treatment (DAT), 2005 through 2007, Crowley, Louisiana.^a

Imazethapyr rate ^b		Injury			
		7 DAT	14 DAT	21 DAT	28 DAT
g ai/ha	Timing	%			
4.4	1-tiller	32 a	33 a	32 a	28 ab
	PD	10 bc	8 bc	8 c	8 cd
	Boot	6 cd	8 bc	10 bc	22 b
	Maturity	0 d	0 c	0 c	0 d
8.7	1-tiller	32 a	39 a	38 a	37 a
	PD	15 b	20 b	20 b	20 bc
	Boot	8 bc	9 bc	12 b	27 ab
	Maturity	0 d	0 c	0 c	0 d
Nontreated		0 d	0 c	0 c	0 d

^a Means within a column followed by the same letter were not statistically different according to the t-test on difference of least square means at P = 0.05.

^b The 4.4 and 8.7 g/ha imazethapyr rates were applied at spray volumes of 15 and 29 L/ha, respectively.

difficult as rice matures (Eric P. Webster⁸, personal communication). When evaluating the efficacy of imazethapyr on selected weed species, visual injury symptoms were more severe on plants treated at earlier timings (Hoss et al. 2003; Shaw et al. 1990).

This reduction in visual injury during reproductive growth stages may be due to the translocation of imazethapyr to meristematic tissue (Shaner et al. 1984). This tissue is located in the internal portions of the rice plant during the reproductive stages of growth and would not be expressed on foliar tissue.

The injury symptoms observed in this study on plants treated at the one-tiller timing were an interveinal chlorosis in the uppermost leaves (Figure 3.1) to plant death. Leaves of treated plants often exhibited small, narrow reddish-brown leaf lesions similar to those associated with leaf blast disease of rice (Groth et al. 2009). Subsequent tillers on recovering treated plants often emerged along a single plane resulting in a flat, fan-shaped appearance in plants. Also, an overall stunting of plants was observed on plants treated at the one-tiller and PD timings (Table 3.2).

Visual symptomology observed on plants treated with imazethapyr at PD and boot, often beyond the rating dates evaluated in this study, were various forms of foliar and inflorescence malformations. Foliar symptoms were plants having multiple shoots arising from the secondary nodes of the main stem (Figure 3.2). The flag leaf on the main stem and secondary shoots would often appear malformed wrinkled, contorted, or rolled. In some instances secondary shoots were stunted or both stunted and malformed. Panicles may partially exert beyond the flag leaf sheath or emerged from the side of the sheath (Figure 3.3). Often panicles failed to initiate emersion from the

⁸ Eric P. Webster, Louisiana State University AgCenter Rice Weed Specialist, 104 M.B. Sturgis Hall, Baton Rouge, LA 70803.



Figure 3.1. Symptoms observed with a one-tiller application of 4.4 g ai/ha imazethapyr.

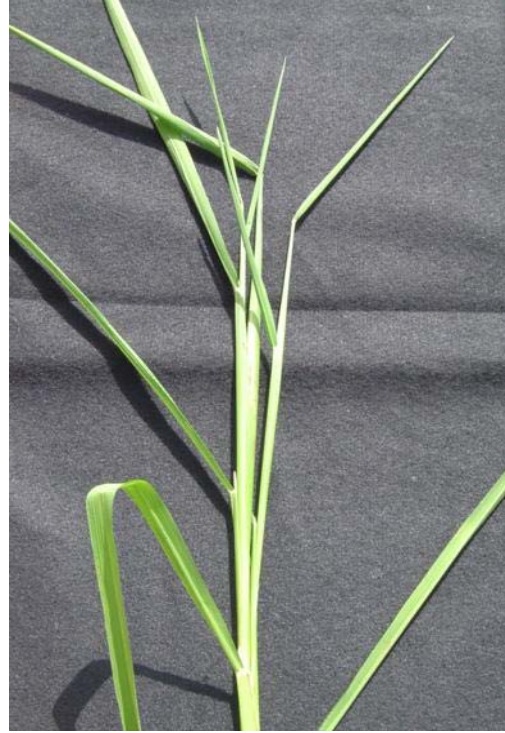


Figure 3.2. Symptoms observed with a boot application of 8.7 g ai/ha imazethapyr.



Figure 3.3. Symptoms observed with a boot application of 8.7 g ai/ha imazethapyr.



Figure 3.4. Symptoms observed with a boot application of 8.7 g ai/ha imazethapyr.

Table 3.2. Effects of simulated imazethapyr drift application rate and timing on primary crop rice plant height at 7, 14, 21, and 28 days after treatment (DAT) and at harvest, 2005 through 2007, as percent of the nontreated, Crowley, Louisiana.^a

Imazethapyr		Rice plant height				
rate ^b	Timing	7 DAT ^c	14 DAT ^d	21 DAT ^e	28 DAT ^f	Harvest ^g
g ai/ha		% of nontreated				
4.4	1-tiller	73 d	84 d	84 cd	80 d	92 c
	PD	95 bc	88 cd	87 c	96 ab	96 b
	Boot	95 bc	94 b	92 b	92 bc	91 c
	Maturity	103 a	101 a	102 a	101 a	100 a
8.7	1-tiller	71 d	74 e	78 e	76 d	90 c
	PD	92 c	83 d	80 de	80 d	90 c
	Boot	94 bc	91 bc	92 b	89 c	91 c
	Maturity	100 ab	96 ab	102 a	100 a	100 a
Nontreated		100 ab	100 a	100 a	100 a	100 a

^a Means within a column followed by the same letter were not statistically different according to the t-test on difference of least square means at P = 0.05.

^b The 4.4 and 8.7 g/ha imazethapyr rates were applied at spray volumes of 15 and 29 L/ha, respectively.

^c Actual heights of nontreated rice for the 1-tiller, PD, boot, and maturity timings were 40, 66, 94, and 47 cm, respectively.

^d Actual heights of nontreated rice for the 1-tiller, PD, boot, and maturity timings were 52, 73, 100, and 55 cm, respectively.

^e Actual heights of nontreated rice for the 1-tiller, PD, boot, and maturity timings were 65, 78, 99, and 58 cm, respectively.

^f Actual heights of nontreated rice for the 1-tiller, PD, boot, and maturity timings were 72, 86, 98, and 63 cm, respectively.

^g Actual height of nontreated rice at primary crop harvest was 96 cm.

flag leaf sheath and decomposed in the leaf sheath causing necrosis of the flag leaf if the plants were treated at the boot stage (Figure 3.4). Some of the inflorescence malformations were due to a malformed panicle axis and partially emergence of the panicle due to fusing of the panicle with the flag leaf sheath. Individual florets malformations that were observed were florets with the tips of the lemma excessively curved toward the palea (Figure 3.3) causing an appearance often referred to as "parrot beaked" when observed in association with the straighthead physiological disorder of rice (Groth et al. 2009).

A plant height response was observed in the primary rice crop when imazethapyr was applied to rice (Table 3.2). At 7 DAT, rice plant height was 71 to 73% of the nontreated when either imazethapyr rate was applied at one-tiller and 92% of the nontreated when 8.7 g/ha imazethapyr was applied at PD. Imazethapyr at 8.7 g/ha was more detrimental to rice plant height applied at one-tiller and PD, 71 to 83% of the nontreated, than applications at boot, 89 to 92% of the nontreated, at 14, 21, and 28 DAT. A similar trend was observed at 14, 21, and 28 DAT with imazethapyr applied 4.4 g/ha. At primary crop harvest, applications at one-tiller, PD, and boot resulted in rice plant height 90 to 92% of the nontreated, with the exception of 4.4 g/ha imazethapyr applied at PD, 96% of the nontreated. Imazethapyr applications at maturity had no effect on primary crop rice plant height. Generally, imazethapyr applied at equal rates was more detrimental to rice plant height when applied at one-tiller and PD than when applied at boot. These findings support the trend of increased crop injury at the one-tiller application timing. Similar findings were reported by Ellis et al. (2003) and Kurtz and Street (2003).

Stem and panicle counts in the primary and ratoon crops were affected by imazethapyr applications (Table 3.3). Imazethapyr applied at PD and boot increased secondary plant stems in the primary crop resulting

Table 3.3. Effects of simulated imazethapyr drift application rate and timing on primary crop rice stem and panicle counts, 2005 through 2007, and ratoon crop rice stem and panicle counts, 2005 and 2007, as percent of the nontreated, Crowley, Louisiana.^a

Imazethapyr rate ^b		Primary crop counts		Ratoon crop counts	
		Stem	Panicle	Stem	Panicle
g ai/ha		————— % of nontreated ^c —————			
4.4	1-tiller	97 d	95 b	105 b	127 b
	PD	136 c	116 b	100 b	110 b
	Boot	189 ab	102 b	177 a	269 a
	Maturity	107 cd	94 b	115 b	112 b
8.7	1-tiller	89 d	95 b	114 b	119 b
	PD	179 b	172 a	127 b	133 b
	Boot	212 a	106 b	184 a	318 a
	Maturity	112 cd	104 b	97 b	103 b
Nontreated		100 d	100 b	100 b	100 b

^a Means within a column followed by the same letter were not statistically different according to the t-test on difference of least square means at P = 0.05.

^b The 4.4 and 8.7 g/ha imazethapyr rates were applied at spray volumes of 15 and 29 L/ha, respectively.

^c Actual nontreated primary crop stem and panicle counts were 33 and 32 per 0.46 m of row, respectively, and actual nontreated ratoon crop stem and panicle counts were 39 and 27 per 0.46 m of row, respectively.

in an increase in stem count compared with the nontreated. This increase was due to imazethapyr causing an excess of secondary stems to be produced on the upper plant nodes. However, panicle count was only increased in the primary crop when imazethapyr was applied at PD at the 8.7 g/ha rate. In the ratoon crop, an increase in stem and panicle counts was only observed in rice treated at the boot stage (Table 3.3).

A primary crop rice yield response was observed (Table 3.4). Imazethapyr applied at 8.7 g/ha at one-tiller and PD and 4.4 g/ha at one-tiller resulted in a primary crop rice yield 62 to 74% of the nontreated. The primary crop yield reduction resulting from an application of imazethapyr at the boot timing is more severe than when applied to the earlier growth stages of rice evaluated in this study. Regardless of rate, imazethapyr applied at boot resulted in a primary crop yield 31 to 44% of the nontreated. However, the ratoon crop yield was 131 to 137% of the nontreated with the same boot timing. This increase was due to imazethapyr causing an excess of secondary stems to be produced on the upper plant nodes in the ratoon rice crop (Table 3.3). This excess of secondary stems did not produce panicles in the primary crop but did produce panicles in the ratoon crop. This response was not observed with rice treated at the other timings. However, when primary and ratoon crop yields were combined, the increase in ratoon crop yield did not compensate for the primary crop yield loss. Either rate of imazethapyr applied at boot and 8.7 g/ha of imazethapyr applied at one-tiller resulted in a total crop yield 41 to 53% of the nontreated (Table 3.4). Imazethapyr applied at 4.4 g/ha at one-tiller and at 8.7 g/ha at PD reduced total crop yield to 63 and 80% of the nontreated, respectively. Imazethapyr applied to rice at maturity and at 4.4 g/ha at PD had no effect on primary, ratoon, or total crop rice yield, compared with the nontreated.

Table 3.4. Effects of simulated imazethapyr drift application rate and timing on primary crop rice yield, 2005 through 2007, and ratoon and total crop rice yield, 2005 and 2007, as percent of the nontreated, Crowley, Louisiana.^a

Imazethapyr rate ^b	Timing	Yield		
		Primary crop	Ratoon crop	Total crop
g ai/ha		% of nontreated ^c		
4.4	1-tiller	64 b	83 bc	63 c
	PD	90 a	86 bc	91 ab
	Boot	44 c	131 a	51 cd
	Maturity	96 a	87 bc	96 ab
8.7	1-tiller	62 b	82 bc	53 cd
	PD	74 b	77 c	80 b
	Boot	31 c	137 a	41 d
	Maturity	98 a	93 bc	98 a
Nontreated		100 a	100 b	100 a

^a Means within a column followed by the same letter were not statistically different according to the t-test on difference of least square means at P = 0.05.

^b The 4.4 and 8.7 g/ha imazethapyr rates were applied at spray volumes of 15 and 29 L/ha, respectively.

^c Actual nontreated yield for the primary, ratoon, and total crops were 6800, 1300, and 8100 kg/ha, respectively.

Though primary crop rice yield was reduced by simulated imazethapyr drift applications at the one-tiller, PD, and boot timings, it appears that rice is most susceptible to imazethapyr during the boot growth stage. Similar results were reported by Hensley et al. (2009) when evaluating simulated glyphosate drift on rice. Rice producers in Louisiana may have the ability to recover some yield loss from an imazethapyr drift event occurring to rice during the boot growth stage by increasing ratoon crop yield; however, the reduction in total crop yield from an imazethapyr drift event at the boot growth stage of rice has the potential to be significant. These data also indicate an increased susceptibility to imazethapyr drift occurring at the one-tiller timing compared to the PD timing. This may be due to the reduced plant biomass at this growth stage compared to the later PD growth stage. Shaw et al. (1990) reported an increased susceptibility to imazethapyr in smaller plants when evaluating its effects on johnsongrass (*Sorghum halepense* L.) at 15, 30, and 60 cm plant heights. Though rice has the ability to recover from imazethapyr drift occurring at the vegetative one-tiller stage, if a combination of herbicide drift and climatic conditions unsuitable for growth hinder recovery, yield losses may be significant (Eric P. Webster⁸, personal communication).

Seed Germination and Seedling Vigor Studies. Simulated imazethapyr drift applications did affect primary crop rice seed weight; however, ratoon crop rice seed weight was not affected (Table 3.5). An application of 8.7 g/ha imazethapyr at boot reduced primary crop rice seed weight to 93% of the nontreated. These data indicate that the grain that remains after mechanical harvesting can be impacted by imazethapyr applications. It is expected that any unfilled or malformed grain observed on rice panicles on treated plants would have been separated and expelled by the mechanical plot harvester. This separation is similar to a commercial harvesting operation so any reduction in seed weight, germination, or seedling vigor of harvested grain

Table 3.5. Effects of simulated imazethapyr drift application rate and timing on primary crop rice seed weight, 2005 through 2007, and ratoon crop rice seed weight, 2005 and 2007, as percent of the nontreated, Crowley, Louisiana.^a

		100-seed weight	
Imazethapyr			
rate ^b	Timing	Primary crop	Ratoon crop
g ai/ha		% of nontreated ^c	
4.4	1-tiller	100 ab	108 a
	PD	100 ab	98 a
	Boot	101 a	101 a
	Maturity	103 a	99 a
8.7	1-tiller	100 ab	96 a
	PD	97 b	98 a
	Boot	93 c	100 a
	Maturity	102 a	98 a
Nontreated		100 ab	100 a

^a Means within a column followed by the same letter were not statistically different according to the t-test on difference of least square means at P = 0.05.

^b The 4.4 and 8.7 g/ha imazethapyr rates were applied at spray volumes of 15 and 29 L/ha, respectively.

^c Actual nontreated 100-seed weight for the primary and ratoon crop were 2320 and 2014 mg, respectively.

observed in this study is reflective of the impact expected on commercial seed rice producers. Studies conducted using hand-harvesting of seeds, such as Walker and Oliver (2008), which bypass a separation process, may misrepresent the impact of herbicides on seed in mechanically harvested grain crops.

Imazethapyr applications did not affect primary crop rice seed germination at 13 or 16 C (Table 3.6). Primary crop seed germination at 19 and 22 C was reduced by imazethapyr applied at one-tiller, PD, and boot, compared with the nontreated. At these temperatures, when applied at PD and boot, 8.7 g/ha imazethapyr resulted in a greater reduction in germination than 4.4 g/ha. When evaluated at 25 C, primary crop rice seed germination was reduced, compared with the nontreated, when imazethapyr was applied at 8.7 g/ha at one-tiller, PD, and boot, and 4.4 g/ha at boot. The greatest reduction in primary crop seed germination at 19, 22, and 25 C was observed from an imazethapyr application at 8.7 g/ha at boot resulting in germination 53, 69, and 62% of the nontreated, respectively. Reductions in germination of this magnitude can lead to a significant increase in seed cost to rice producers. Compared with the nontreated at 19 C, a producer using seed affected by the 8.7 g/ha imazethapyr rate at boot would have to approximately double their seeding rate to achieve a rice seed germination comparable to that of the nontreated seed. Ratoon crop rice seed germination was reduced at 19 and 22 C when 4.4 g/ha imazethapyr was applied at PD (Table 3.7).

Imazethapyr applications did not reduce primary crop seedling plant vigor (data not shown). These data indicate that when 8.7 g/ha imazethapyr is applied at boot, there is a correlation between reduced rice seed weight and seed germination, also observed by Krishnasamy and Seshu (1989). However, a lack of reduction in seed weight was not an indicator of seed germination for imazethapyr applications at one-tiller and PD or the 4.4

Table 3.6. Effects of simulated imazethapyr drift application rate and timing on primary crop rice seed germination at various temperatures, 2005 through 2007, as percent of the nontreated, Crowley, Louisiana.^a

Imazethapyr rate ^b		Temperatures				
		13 C	16 C	19 C	22 C	25 C
g ai/ha		% of nontreated ^c				
4.4	1-tiller	43 a	95 a	79 b	89 bc	97 ab
	PD	71 a	95 a	81 b	90 b	100 a
	Boot	100 a	93 a	71 bc	87 bc	82 d
	Maturity	100 a	102 a	100 a	99 a	103 a
8.7	1-tiller	57 a	91 a	74 bc	91 b	91 bc
	PD	100 a	79 a	66 c	83 c	85 cd
	Boot	100 a	74 a	53 d	69 d	62 e
	Maturity	100 a	105 a	98 a	95 ab	97 ab
Nontreated		100 a	100 a	100 a	100 a	100 a

^a Means within a column followed by the same letter were not statistically different according to the t-test on difference of least square means at P = 0.05.

^b The 4.4 and 8.7 g/ha imazethapyr rates were applied at spray volumes of 15 and 29 L/ha, respectively.

^c Actual germination of the nontreated seed at 13, 16, 19, 22, and 25 C was 7, 42, 58, 78, and 68%, respectively.

Table 3.7. Effects of simulated imazethapyr drift application rate and timing on ratoon crop rice seed germination at various temperatures, 2005 and 2007, as percent of the nontreated, Crowley, Louisiana.^a

Imazethapyr rate ^b		Temperatures				
		13 C	16 C	19 C	22 C	25 C
g ai/ha		% of nontreated ^c				
4.4	1-tiller	125 a	213 a	126 a	127 a	103 a
	PD	50 a	67 c	78 d	87 d	93 a
	Boot	100 a	120 b	113 b	109 bc	91 a
	Maturity	100 a	113 bc	96 c	95 cd	96 a
8.7	1-tiller	100 a	200 a	122 ab	129 a	96 a
	PD	75 a	120 b	100 c	96 cd	93 a
	Boot	125 a	133 b	128 a	116 b	99 a
	Maturity	100 a	127 b	89 cd	91 cd	94 a
Nontreated		100 a	100 bc	100 c	100 c	100 a

^a Means within a column followed by the same letter were not statistically different according to the t-test on difference of least square means at P = 0.05.

^b The 4.4 and 8.7 g/ha imazethapyr rates were applied at spray volumes of 15 and 29 L/ha, respectively.

^c Actual germination of the nontreated seed at 13, 16, 19, 22, and 25 C was 4, 15, 46, 55, and 69%, respectively.

g/ha rate at boot in this study. These data also indicate that rice seed germination may be affected by an imazethapyr drift even if there is no notable reduction in seed weight. However, it appears that if seeds germinate, seedling vigor will not be adversely affected. If seed rice is affected by a drift event, extra caution should be taken before that seed is sold to producers.

In conclusion, simulated imazethapyr drift applications at the one-tiller, PD, and boot timings result in reduced plant height and primary and total crop yield losses, with the greatest reduction in primary crop yield resulting from imazethapyr applied at the boot growth stage. Imazethapyr applications to mature rice had no effect on rice plant height or yield. Primary crop rice seed weight was reduced by an imazethapyr application to rice at boot. Primary crop rice seed germination was reduced when imazethapyr was applied at one-tiller, PD, and boot, with increased susceptibility at the boot growth stage. A reduction in ratoon crop rice seed weight was not observed; however, ratoon crop rice seed germination was reduced by an imazethapyr application at the PD growth stage.

The ability to identify imazethapyr drift on rice can be helpful to producers, Cooperative Extension Service personnel, crop consultants, and state regulatory agencies in distinguishing between herbicide drift and injury associated with soil fertility issues, diseases, and other disorders affecting rice. Misidentification of herbicide drift symptoms as injury associated with these factors can lead to loss in profitability if growers apply unnecessary applications of inputs to correct these factors when the symptoms present are actually a result of herbicide drift. The ability to correlate the symptoms observed to imazethapyr drift also may assist state regulatory agencies in identifying the source of a herbicide drift event. If imazethapyr can be identified by observation of plant symptoms this can reduce the cost associated with confirmation of a herbicide drift event

through the use of diagnostic testing of foliar residue since most analytical facilities charge per evaluation and the diagnostic tests involved are often herbicide specific.

An imazethapyr drift event occurring to a producer's field at the one-tiller, PD, or boot growth stages of rice can reduce yield; however, this study indicates that a drift event occurring at the boot stage may be the most detrimental to yield. Rice receiving a drift event in vegetative growth stages, one-leaf to one-tiller, can often recover if stand is maintained at recommended densities (Eric P. Webster⁸, personal communication). However, an imazethapyr drift event occurring to rice in the reproductive stage of growth may have little to no visual foliar injury and often symptoms may not appear until rice plants near crop maturity. This may lead to loss of yield and profitability due to continuing to supply crop inputs, such as increased fertilizer, insecticide, and fungicide applications, to a crop that has an already reduced yield potential. The negative effects of an imazethapyr drift event occurring to a seed producer's field to rice in the PD or boot growth stages has the potential to be two-fold. The reduction in profitability the year of the event from reduced yield and a reduction in seed germination has the ability to reduce profitability in the subsequent year's crop due to an increase in seeding rate to offset the reduced seed germination.

Caution should be taken when applying imazethapyr near adjacent susceptible rice fields, especially when making applications near rice in the reproductive growth stages. Though the effects of imazethapyr drift on rice may not be immediately apparent by visual observation, the potential affect on grain yield and the germination potential of the harvested grain could be highly detrimental to rice producers.

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Chapter 4

Impact of Off-site Deposition of Glufosinate to Rice

Introduction

Glufosinate is a nonselective, foliar applied, postemergence herbicide used to control annual and perennial weeds in non-crop areas and for weed control in glufosinate-resistant crops (Senseman 2007). The mechanism of action of glufosinate is the inhibition of the enzyme glutamine synthetase (EC 6.3.1.2) that converts glutamate and ammonia to glutamine (Lea et al. 1984; Senseman 2007). This inhibition of glutamine synthetase results in a toxic accumulation of ammonia in treated plants and inhibition of photosystems I and II (Sauer et al. 1987; Senseman 2007; Tachibana et al. 1986; Wild et al. 1987).

The symptoms expressed in plants from the inhibition of glutamine synthase are that chlorosis and wilting usually occur within 3 to 5 d followed by necrosis in 7 to 14 d after application to susceptible species (Senseman 2007). The rate of symptom development is increased in bright sunlight, high humidity, and moist soil.

Rice (*Oryza sativa* L.) is a major crop produced in the four state region of Arkansas, Louisiana, Mississippi, and Texas, with these states accounting for 76% of the 1.2 million total hectares of rice planted in the U.S. and 70% of the \$3.4 billion of total value of rice produced in the U.S. in 2008 (NASSa 2009; NASSb 2009). Glufosinate-resistant rice has been evaluated and approximately 320 ha of glufosinate-resistant rice was produced commercially in Louisiana in 2000 (Lanclos et al. 2002; Lanclos et al. 2003; Steven D. Linscombe¹, personal communication; Zhang et al. 2003). Glufosinate-resistant corn (*Zea mays* L.) has been researched and evaluated for more than a decade and glufosinate-resistant soybean [*Glycine max* (L.)

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Merr.] cultivars became available for commercial soybean producers in Louisiana in 2009 (Krausz et al. 1999; Ronald J. Levy, Jr.², personal communication). Since many of the rice producing parishes in Louisiana also produce soybean and corn (NASSc 2009), the potential exists for off-target herbicide drift from one of these crops to rice.

In 2000, glufosinate drift was observed on rice from an application applied to commercially produced glufosinate-resistant rice in Louisiana (Steven D. Linscombe¹, personal communication). Averaged over the ten year period 1999 to 2008, the Louisiana Department of Agriculture and Forestry processed 76 Pesticide Investigation Reports per year listing ground or aerial applicators in Louisiana, and, on average, six reports per year involved rice (Lisa Gautreaux³, personal communication). However, it was reported that, in 2009, at least 50 rice fields were suspected of being affected by glyphosate drift and at least 25 rice fields were suspected of being affected by imazethapyr drift, many of which were not reported to the LDAF (Ronald J. Levy, Jr.², personal communication; John K. Saichuk⁴, personal communication). Therefore, the number of rice fields actually affected by a drift event each year may be underrepresented by the number of official complaints processed by the LDAF.

It has been reported that fine spray droplets less than 150 μm in size have a greater potential to drift off-target (Hanks 1995; SDTF 1997). The use of adjuvants and selection of proper spray nozzle type, size, and application pressure can be beneficial in reducing the amount of fine spray droplets in the spray cloud (Hanks 1995; Jones et al. 2007; Nuyttens et al.

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2007; VanGessel and Johnson 2005). This increase in droplet size can reduce the potential for off-target drift from droplets larger than 150 μm ; however, environmental conditions at the time of herbicide application can also impact the off-target drift of spray solutions (Bouse et al. 1976; Crabbe et al. 1994; Thistle 2004).

Wind speed and direction may be considered the two most important factors affecting spray droplets in the atmosphere, a stable atmosphere may be the third most important factor (Thistle 2004). A stable atmosphere, commonly referred to as an inversion, is an atmosphere that has a change of temperature with a change in elevation in the atmosphere. In a stable atmosphere, warm air overlies cool air. If air in a particular layer is displaced upward or downward it will be colder or warmer, respectively, than the immediately adjacent layer it enters and thus returns to the original layer. If a herbicide application is made during an inversion scenario, the fine droplets that do not succumb to gravity will remain in the layer in which they are deposited due to the lack of layers mixing. The droplets in this layer can be very concentrated and may horizontally move off-target, and in some cases, great distances.

Ultra-low-volume applications made during a stable atmosphere produced 35% more herbicide drift than applications made during a turbulent atmosphere with light wind speeds (Crabbe et al. 1994). The conditions in which the greatest drift occurred were in moderately stable conditions with wind at 3 m/s resulting in off-target drift 71% at 400 m and 50% at 2200 m from the application site and in slightly stable conditions with wind at 5 m/s resulting in off-target drift 77% at 400 m and 27% at 2200 m from the application site. It is recommended that herbicide applications should be avoided during the early morning and late evening because these times are most favorable for the development of inversion conditions (Crabbe et al. 1994; Thistle 2004).

Through the use of simulated herbicide drift studies, the potential effects of glufosinate drift to rice can be evaluated. In previous research, simulated drift studies varying the spray volume proportionally with reduced herbicide rates to simulate herbicide drift resulted in increased crop injury compared with the same herbicide rate applied in a constant spray volume (Banks and Schroeder 2002; Ellis et al. 2002; Ramsdale et al. 2003; Roider et al. 2008). Ellis et al. (2002) reported that glyphosate and glufosinate applied at 6.3 and 12.5% of their labeled use rates in a proportionally reduced carrier volume reduced corn yield four times and one-and-one-half times more, respectively, compared to the same herbicide doses applied with a constant carrier volume of 234 L/ha. The no-effect glyphosate rate for sweet corn was four times greater when using a spray volume proportional to the reduced glyphosate rate compared to when reduced glyphosate rates were applied in a constant spray volume (Banks and Schroeder 2002).

Glufosinate applied to rice at a simulated drift rate of 53 g/ha in a constant spray volume reduced rice yield 30% (Ellis et al. 2003). When glufosinate was applied to grain sorghum (*Sorghum bicolor* L.) at 1, 3, 10, and 33% of its labeled use rate only the 10 and 33% rates resulted in reduced grain sorghum yield (Al-Khatib et al. 2003).

Rapid rice seed germination and seedling growth can enhance plant stand establishment in commercial fields (Krishnasamy and Seshu 1989; Pollock and Roos 1972; Wright 1980). Rice seed germination was reduced by simulated glufosinate drift when evaluated at 16 C (Ellis et al. 2003). Bennett and Shaw (2000) found that applications of glufosinate applied preharvest in soybean reduced seed germination of sicklepod (*Senna obtusifolia* (L) Irwin and Barnaby) and pitted morningglory (*Ipomoea lacunosa* L.). Glufosinate applied for preharvest desiccation of grain sorghum (*Sorghum bicolor* (L.) Moench) did not effect grain sorghum seed germination (Bovey et al. 1999). Glyphosate applied to wheat at first node, boot, and early flowering growth

stages resulted in 16 to 36% reductions in seed weight (Roider et al. 2007). A negative correlation between 100-count seed weight and rice seed germination has been observed (Krishnasamy and Seshu 1989). A need exists to evaluate the possible effects of a glufosinate drift event on rice crop seed germination and seedling vigor.

Even though a published study evaluating the effects of simulated glufosinate drift on rice exists (Ellis et al. 2003), this study was not conducted using spray volumes proportional with reduced herbicide dosage. The objectives of this research were to evaluate the effects of simulated glufosinate drift applied to rice during the primary rice crop on the crop response and impact on the seed produced on treated rice in the primary and ratoon rice crops.

Materials and Methods

Simulated Glufosinate Drift Field Study. A study was conducted on rice grown in 2005 through 2007 at the LSU AgCenter Rice Research Station near Crowley, Louisiana on a Crowley silt loam (fine montmorillonitic, thermic Typic Albaqualf) with pH 5.5 and 1.2% organic matter. Field preparation consisted of a fall and spring disking and two passes in opposite directions with a two-way bed conditioner equipped with rolling baskets and S-tine harrows 15 cm deep. The long grain rice cultivar 'Cocodrie' was drill-seeded March 28 to April 17 in 2005 through 2007. Plots consisted of twelve-18 cm spaced rows 6 m long.

The experimental design was an augmented two-factor factorial arrangement of treatments in a randomized complete block with four replications. Factor A consisted of glufosinate applied at simulated drift rates of 6.3 and 12.5% of the labeled usage rate of 493 g ai/ha, or 31 and 62 g/ha, respectively. Factor B consisted of application timings at different growth stages: one-tiller, panicle differentiation (PD), boot, and physiological maturity. Each herbicide application was made with the spray

volume varying proportionally to herbicide dosage based on a constant spray volume of 234 L/ha. The 12.5% herbicide rate was applied at a spray volume of 29 L/ha and the 6.3% herbicide rate was applied at a spray volume of 15 L/ha. Each application was made with a tractor-mounted CO₂-pressurized sprayer calibrated to deliver a constant carrier volume with speed adjusted to vary application rate and equipped with Teejet^{®5} TX-2 Conejet[®] 800033 nozzles. A ratoon rice crop was not produced in 2006 due to unfavorable weather following primary crop harvest.

The study area was maintained weed-free using clomazone at 420 g ai/ha applied preemergence followed by propanil at 4483 g ai/ha plus halosulfuron at 53 g ai/ha applied postemergence. For the primary rice crop a preplant application of 280 kg/ha of 8-24-24 (N-P₂O₅-K₂O) fertilizer and a preflood application of 365 kg/ha 46-0-0 urea fertilizer were applied to the study area and for the ratoon rice crop a preflood application of 100 kg/ha 46-0-0 urea fertilizer was applied to the study area to maintain proper fertility and to maximize yields in the primary and ratoon crops. Standard agronomic and pest management practices were implemented throughout the growing season to maximize yield.

Rice plant height and rice injury in the primary rice crop were obtained 7 days after herbicide treatment (DAT) and continued weekly for 28 DAT. Rice plant height was obtained by measuring four plants per plot from the soil surface to the tip of the extended uppermost emerged leaf or extended rice panicle. Rice injury was evaluated based on chlorosis and necrosis of foliage and reduced plant height using a scale of 0 to 100% where 0 = no injury and 100 = plant death. Rice plant height at primary crop harvest and rough rice yield, 100-count seed weight, and stem and panicle counts for the primary and ratoon crop were also obtained. Whole plots were

⁵ Spraying Systems Co., P. O. Box 7900, Wheaton, IL 60187.

mechanically harvested and rough rice yield was adjusted to 12% moisture. Total stem and panicle counts were calculated by hand harvesting a 0.46 m section of row and determining the number of stems present at the mid-height of the plants and the number of panicles with bases emerged beyond the sheath of the flag leaf, or the last leaf to emerge prior to the panicle.

All data were subjected to the Mixed Procedure of SAS (SAS 2003). Year, replications (nested within year), and all interactions containing either of these effects were considered random effects. Application timing and rate were considered fixed effects. Considering year or combination of year as random effects permits inferences about treatments over a range of environments (Carmer et al. 1989; Leon et al. 2008). Type III statistics were used to test all possible effects of fixed factors (application timing and rate) and least square means were used for mean separation at the 5% probability level ($p \leq 0.05$).

Seed Germination Study. The germination potential of seed collected from grain harvested in the simulated glufosinate drift field study at primary crop harvest, 2005 through 2007, and at ratoon crop harvest, 2005 and 2007, was evaluated at multiple temperatures. Seed collected from each plot was air-dried and stored at 8 C. Germination temperatures evaluated were 13, 16, 19, 22, and 25 C. Temperature selection and germination testing procedure for this study were based on procedures previously described by Webster et al. (2003) and follow standard germination procedures recommended by Association of Official Seed Analysts (AOSA) (AOSA 2006). Temperature selection was based on 19 C being the historical mean 10-cm soil temperature in Crowley, LA on April, 1, which corresponds to 50% of the rice being planted across the state (Webster et al. 2003).

One hundred seeds from each field plot were prepared by soaking for 30 min in a 50:50 (v/v) solution of chlorine bleach and distilled water to decrease seedling diseases. After soaking, seeds were triple rinsed with

distilled water. After seed preparation, seeds were placed in a 10 cm plastic Petri dish between two 9 cm germination blotters⁶. Next, 10 ml of carboxin (5,6-dihydro-2-methyl-*N*-phenyl-1,4-oxathiin-3-carboxamide) plus thiram (tetramethylthiuram disulfide) plus distilled water solution (52 ml of a 10% carboxin and 10% thiram premix liquid fungicide combined with 948 ml distilled water) was applied in each Petri dish to reduce seedling diseases. Petri dishes were sealed with Parafilm M⁷ to prevent moisture loss and placed in a constant-temperature growth chamber in total darkness. Germination counts were taken 5, 9, and 14 d after initiation (DAI) of the study. A seed was considered germinated if the radical had reached a length of 1 mm.

Seed germination data were arranged as repeated measures and subjected to the Mixed Procedure of SAS (SAS 2003). Year, replications (nested within year), DAI (nested within replications), and all interactions containing either of these effects were considered random effects. Application timing and rate and germination temperature were considered fixed effects. Considering year or combination of year as random effects permits inferences about treatments over a range of environments (Carmer et al. 1989; Leon et al. 2008). Type III statistics were used to test all possible effects of fixed factors (application timing and rate and germination temperature) and least square means were used for mean separation at the 5% probability level ($p \leq 0.05$).

Seedling Vigor Study. Vigor of seedlings from grain collected at primary crop harvest in the simulated glufosinate drift field study in 2006 and 2007 was examined. Seedling vigor, as defined by AOSA (AOSA 2002), is "seedling vigor comprises those seed properties which determine the potential for rapid, uniform emergence, and development of normal seedlings under a wide

⁶ Anchor Steel Blue Seed Germination Blotter[®], SDB 3.5. Anchor Paper Company, 480 Broadway, St. Paul, MN 55101.

⁷ Parafilm M[®]. Pechiney Plastic Packaging, Menash, WI 54952.

range of field conditions" and it is used as a measure of seed quality by producers. Since there is no accepted standard vigor test for rice, vigor testing procedures for this study were based on procedures previously described by Webster et al. (2003).

Approximately 100 seeds from each field plot were prepared by soaking for 30 min in a 50:50 (v/v) solution of chlorine bleach and distilled water to decrease seedling diseases. After soaking, seeds were triple rinsed with distilled water. Following seed preparation, seeds were pre-germinated by soaking in distilled water for 24 h. Ten pre-germinated seeds from each field plot were placed on a single sheet of nontreated germination paper⁸ cut to fit a 12 by 23 by 0.3 cm acrylic sheet. Germination paper was moistened by submerging in distilled water for 5 seconds to facilitate adherence to the acrylic sheet and provide residual moisture to rice seeds. Seeds were placed along the center of germination paper oriented with the radical end of the seed toward the lower half of the sheet. A one-ply paper towel strip was placed over the seed, and 5 ml of a mancozeb [ethylene (bis)-dithiocarbamate] plus distilled water solution (dry formulation of mancozeb at 1640 mg ai/L distilled water) was applied on top of the strip to reduce seedling diseases. The plated seeds were then placed vertically in a rack and then placed in a 30 by 51 by 5 cm dish with 1,420 ml of distilled water to allow for evaporation. The dish and racks of plates were wrapped in plastic wrap to prevent desiccation. The glass dish was placed in a constant temperature growth chamber at 21 C for 12 d in total darkness. At the end of 12 d, shoot lengths were measured and an average of the 10 shoot lengths was obtained for data analysis.

Seed vigor data were subjected to the Mixed Procedure of SAS (SAS Institute 2003). Year, replications (nested within year), and all

⁸ Anchor Steel Blue Seed Germination Blotters®, SDB 1924. Anchor Paper Company, 480 Broadway, St. Paul, MN 55101.

interactions containing either of these effects were considered random effects. Application timing and rate were considered fixed effects. Considering year or combination of year as random effects permits inferences about treatments over a range of environments (Carmer et al. 1989; Leon et al. 2008). Type III statistics were used to test all possible effects of fixed factors (application timing and rate) and least square means were used for mean separation at the 5% probability level ($p \leq 0.05$).

Results and Discussion

Simulated Glufosinate Drift Field Study. A crop injury response was observed in the primary crop (Table 4.1). At 7 and 14 DAT, averaged across application rates, the greatest crop injury was 24 and 14% at the boot timing, respectively. Crop injury at 21 DAT was 6% with glufosinate applied at PD and boot and at 28 DAT crop injury was less than 5% for all timings evaluated. These data indicate a trend of increased crop injury when glufosinate is applied at later growth stages. Ellis et al. (2003) reported similar findings when evaluating glufosinate drift on rice.

Foliar symptoms observed on rice plants treated with glufosinate begin as small reddish-brown lesions within 2 DAT becoming irregularly shaped chlorotic lesions within 7 DAT on affected leaves (Figure 4.1, 4.2, 4.3). By 14 DAT, new leaf growth had initiated in plants treated at one-tiller and PD with chlorotic lesions increasing in size on the lower leaves resulting in necrosis of the leaf (Figure 4.4). By 28 DAT, visual symptoms were often undetectable, compared with nontreated plants.

A plant height response was observed in the primary rice crop when a simulated glufosinate drift application was applied to rice (Table 4.2). Rice plant height at 7 through 28 DAT was 94 to 96% of the nontreated when glufosinate was applied to rice at one-tiller. Regardless of timing, no rice plant height was less than 98% of the nontreated at harvest. These data

Table 4.1. Effects of simulated glufosinate drift application timing on primary rice crop injury 7, 14, 21, and 28 days after treatment (DAT), 2005 through 2007, Crowley, Louisiana.^a

Glufosinate timing	Injury			
	7 DAT	14 DAT	21 DAT	28 DAT
	%			
1-tiller	10 c	4 c	1 b	1 b
PD	15 b	9 b	6 a	1 b
Boot	24 a	14 a	6 a	3 a
Maturity	0 d	0 d	0 b	0 b
Nontreated	0 d	0 d	0 b	0 b

^a Means within a column followed by the same letter were not statistically different according to the t-test on difference of least square means at P = 0.05.

^b Data averaged across application rates of 31 and 62 g ai/ha glufosinate applied at spray volumes of 15 and 29 L/ha, respectively.



Figure 4.1. Symptoms observed with a PD application of 62 g ai/ha glufosinate.



Figure 4.2. Symptoms observed with a PD application of 31 g ai/ha glufosinate.



Figure 4.3. Symptoms observed with a PD application of 62 g ai/ha glufosinate.



Figure 4.4. Symptoms observed with a PD application of 31 g ai/ha glufosinate.

Table 4.2. Effects of simulated glufosinate drift application timing on primary crop rice plant height at 7, 14, 21, and 28 days after treatment (DAT) and at harvest, 2005 through 2007, as percent of the nontreated, Crowley, Louisiana.^a

Glufosinate timing	Rice plant height				
	7 DAT ^b	14 DAT ^c	21 DAT ^d	28 DAT ^e	Harvest ^f
	% of nontreated ^g				
1-tiller	94 b	96 b	96 c	96 c	99 ab
PD	98 a	99 a	99 ab	98 b	98 b
Boot	99 a	98 ab	98 b	99 ab	98 b
Maturity	99 a	100 a	100 a	99 ab	100 a
Nontreated	100 a	100 a	100 a	100 a	100 a

^a Means within a column followed by the same letter were not statistically different according to the t-test on difference of least square means at P = 0.05.

^b Actual heights of nontreated rice for the 1-tiller, PD, boot, and maturity timings were 41, 65, 93, and 51 cm, respectively.

^c Actual heights of nontreated rice for the 1-tiller, PD, boot, and maturity timings were 54, 71, 99, and 56 cm, respectively.

^d Actual heights of nontreated rice for the 1-tiller, PD, boot, and maturity timings were 65, 78, 98, and 60 cm, respectively.

^e Actual heights of nontreated rice for the 1-tiller, PD, boot, and maturity timings were 71, 88, 97, and 64 cm, respectively.

^f Actual height of nontreated rice at primary crop harvest was 96 cm.

^g Data averaged across application rates of 31 and 62 g ai/ha glufosinate applied at spray volumes of 15 and 29 L/ha, respectively.

indicate rice has the ability to recover from early injury caused by glufosinate drift with little or no impact on plant height. A similar trend of greater injury at earlier application timings was reported by Hoss et al. (2003) when evaluating efficacy of glufosinate on prairie cupgrass (*Eriochloa contracta* Hitchc.).

A crop yield response was observed (Table 4.3). Averaged across application rates, primary crop yield was reduced to 90% of the nontreated when simulated glufosinate drift was applied to rice at the boot growth stage. A reduction in ratoon crop yield was not observed; however, total crop yield was 93% of the nontreated when glufosinate was applied at the boot stage. These data indicate rice is susceptible to glufosinate drift occurring at the boot growth stage. These findings support the trend of increased crop injury with a glufosinate drift event at the boot stage. Ellis et al. (2003) reported a late reproductive growth stage application of glufosinate reduced rice yield. Though a yield reduction of this magnitude would reduce grower's profits, the yield reduction observed by Ellis et al. (2003) and Hensley et al. (2009) when evaluating glyphosate drift on rice resulted in a more significant decrease in primary crop yield. The rice plant stem and panicle counts in the primary and ratoon crop were not affected by glufosinate applications (data not shown). The carbohydrate source for developing rice grain is the three or four uppermost leaves of the rice plant (Dunand and Saichuk 2009). Since the visual crop injury observed with a glufosinate application at the boot stage would affect these uppermost leaves, this injury could account for the reduction in primary crop yield.

Seed Germination and Seedling Vigor Studies. Simulated glufosinate drift applications did not affect primary crop rice seed weight (Table 4.4) or ratoon crop rice seed weight (data not shown). It is expected that any unfilled or malformed grain on rice panicles on treated plants would be separated and expelled by the mechanical plot harvester. This separation is

Table 4.3. Effects of simulated glufosinate drift application timing on primary crop rice yield, 2005 through 2007, and ratoon and total crop rice yield, 2005 and 2007, as percent of the nontreated, Crowley, Louisiana.^a

Glufosinate timing	Yield		
	Primary crop	Ratoon crop	Total crop
	% of nontreated ^{b,c}		
1-tiller	100 a	98 a	98 a
PD	100 a	102 a	99 a
Boot	90 b	106 a	93 b
Maturity	103 a	93 a	99 a
Nontreated	100 a	100 a	100 a

^a Means within a column followed by the same letter were not statistically different according to the t-test on difference of least square means at P = 0.05.

^b Data averaged across application rates of 31 and 62 g ai/ha glufosinate applied at spray volumes of 15 and 29 L/ha, respectively.

^c Actual nontreated yield for the primary, ratoon and total crops were 6100, 1500, and 7600 kg/ha, respectively.

Table 4.4. Effects of simulated glufosinate drift application rate on primary crop rice seed weight, 2005 through 2007, and seedling vigor, 2006 and 2007, as percent of the nontreated, Crowley, Louisiana.^a

Glufosinate rate	100-seed weight	Seedling vigor
g ai/ha	————— % of nontreated ^{b,c} —————	
31	100 a	88 b
62	100 a	91 b
Nontreated	100 a	100 a

^a Means within a column followed by the same letter were not statistically different according to the t-test on difference of least square means at P = 0.05.

^b Data averaged across the one-tiller, PD, boot, and maturity application timings.

^c Actual nontreated 100-seed weight and seedling vigor shoot length was 2400 mg and 43 mm, respectively.

similar to a commercial harvesting operation so any affect on seed weight, germination, and seedling vigor of harvested grain observed in this study is reflective of the impact expected on commercial seed rice producers. Studies conducted using hand-harvesting of seeds, such as Walker and Oliver (2008), which bypass a separation process, may misrepresent the impact of herbicides on seed in mechanically harvested grain crops.

A primary crop rice seed germination response was observed (Table 4.5). Averaged across application timings, glufosinate applied at 31 and 62 g/ha resulted in rice seed germination 92 to 93% of the nontreated when evaluated at 19 C. Glufosinate applications had no affect on rice seed germination when evaluated at 13, 16, 22, and 25 C. Ellis et al. (2003) reported a simulated glufosinate drift application to rice resulted in a reduction in primary crop seed germination. Reductions in germination of this magnitude can lead to an increase in seed cost to rice producers. Ratoon crop rice seed germination was not reduced by simulated glufosinate drift applications, compared with the nontreated (Table 4.6).

A reduction in primary crop rice seedling vigor was observed (Table 4.4). Averaged across application timings, glufosinate applied at 31 and 62 g/ha resulted in primary crop rice seedling vigor 88 to 91% of the nontreated. These data indicate that even with no notable reduction in seed weight, rice seed germination and seedling vigor may be affected by glufosinate drift. If seed rice is affected by a glufosinate drift event, extra caution should be taken before that seed is sold to producers.

In conclusion, simulated glufosinate drift to rice at the one-tiller, PD, and boot growth stages resulted in visual crop injury and reduced rice plant height. Primary and total crop rice yield was reduced by glufosinate applied at the boot growth stage. Averaged across application timings, primary crop rice seed germination and rice seedling vigor were reduced by

Table 4.5. Effects of simulated glufosinate drift application rate on primary crop rice seed germination at various temperatures, 2005 through 2007, as percent of the nontreated, Crowley, Louisiana.^a

Glufosinate rate	Temperatures				
	13 C	16 C	19 C	22 C	25 C
g ai/ha	————— % of nontreated ^{b,c} —————				
31	125 a	93 a	92 b	103 a	102 a
62	113 a	89 a	93 b	100 a	100 a
Nontreated	100 a	100 a	100 a	100 a	100 a

^a Means within a column followed by the same letter were not statistically different according to the t-test on difference of least square means at P = 0.05.

^b Data averaged across the one-tiller, PD, boot, and maturity application timings.

^c Actual germination of the nontreated seed at 13, 16, 19, 22, and 25 C was 8, 45, 59, 76, and 68%, respectively.

Table 4.6. Effects of simulated glufosinate drift application timing on ratoon crop rice seed germination at various temperatures, 2005 and 2007, as percent of the nontreated, Crowley, Louisiana.^a

Glufosinate timing	Temperatures				
	13 C	16 C	19 C	22 C	25 C
	————— % of nontreated ^{b,c} —————				
1-tiller	100 a	127 a	113 a	117 a	120 a
PD	67 a	105 b	102 b	98 c	112 b
Boot	67 a	109 ab	107 ab	112 ab	120 a
Maturity	100 a	91 b	98 b	106 b	97 c
Nontreated	100 a	100 b	100 b	100 bc	100 c

^a Means within a column followed by the same letter were not statistically different according to the t-test on difference of least square means at P = 0.05.

^b Data averaged across application rates of 31 and 62 g ai/ha glufosinate applied at spray volumes of 15 and 29 L/ha, respectively.

^c Actual germination of the nontreated seed at 13, 16, 19, 22, and 25 C was 3, 22, 46, 52, and 59%, respectively.

glufosinate applications, regardless of rate. Glufosinate applications did not affect rice treated at primary crop maturity.

The ability to identify glufosinate drift on rice can be helpful to producers, Cooperative Extension Service personnel, crop consultants, and state regulatory agencies in distinguishing between herbicide drift and injury associated with soil fertility issues, diseases, and other disorders affecting rice. Misidentification of herbicide drift symptoms as injury associated with these factors can lead to loss in profitability if growers apply unnecessary applications of inputs to correct these factors when the symptoms present are actually a result of herbicide drift. The ability to correlate the symptoms observed to glufosinate drift also may assist state regulatory agencies in identifying the source of a herbicide drift event. If glufosinate can be identified by observation of plant symptoms this can reduce the cost associated with confirmation of a herbicide drift event through the use of diagnostic testing of foliar residue since most analytical facilities charge per evaluation and the diagnostic tests involved are often herbicide specific.

A glufosinate drift event occurring to a producer's field at the boot growth stage of rice can reduce yield. The negative effects of a glufosinate drift event occurring to a seed producer's field to rice in the boot growth stage has the potential to be two-fold. The reduction in profitability the year of the event from reduced yield and a reduction in seed germination and seedling vigor has the ability to reduce profitability in the subsequent year's crop due to an increase in seeding rate to offset the reduced seed germination and seedling vigor.

Caution should be taken when applying glufosinate near adjacent susceptible rice fields, especially when making applications near rice in the boot growth stage. The potential effect on grain yield and the seed

germination and seedling vigor potential of the harvested grain could be highly detrimental to rice producers.

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Chapter 5

Rice Crop and Seed Rice Response to Off-target Drift of Imazamox

Introduction

Imazamox is a selective imidazolinone herbicide used to control annual and perennial weeds in soybean [*Glycine max* (L.) Merr.], edible legumes, and imidazolinone-resistant crops (Senseman 2007). The mechanism of action for imazamox is inhibition of acetolactase synthase (ALS) (EC 4.1.3.18) also called acetoxyacid synthase (AHAS), a key enzyme in the biosynthesis of the branched-chain amino acids isoleucine, leucine, and valine (Muhitch et al. 1987; Senseman 2007; Shaner 1991; Shaner et al. 1984; Stidham 1991; Stidham and Singh 1991). Plant death results from events occurring in response to ALS inhibition, specifically the inhibition of isoleucine, leucine and valine, but the actual sequence of phytotoxic processes is unclear (Shaner 1991; Stidham and Singh 1991). Some secondary effects may include disruption of photosynthate translocation, hormone imbalance due to interruption of source/sink relationships, and interference in DNA synthesis and cell growth.

The symptoms expressed from this toxicity are growth is inhibited within a few hours of herbicide application, meristematic areas become chlorotic, followed by a slow general foliar chlorosis and necrosis (Shaner 1991). This injury to meristematic areas can be attributed to inhibition of branched-chain amino acids in the meristematic region. Even though plants have the ability to scavenge amino acids from preexisting proteins, the meristematic region lacks the protein reserve pools that are available in the mature regions of the plant. Injury symptoms usually appear within 7 to 14 d for susceptible species.

In 1993, imidazolinone-resistant rice (*Oryza sativa* L.) was developed and exhibited tolerance to the imidazolinone class of herbicides (Croughan

1994; Pellerin et al. 2004; Webster and Masson 2001). The first commercially available imidazolinone-resistant rice cultivars were 'CL 121' and 'CL 141' which were derived from the imidazolinone-resistant parent line 'IMI-tolerant 93AS-3510' (Carlson et al. 2002; Croughan 1994, 1998; Gealy et al. 2003; Tan et al. 2005; Webster and Masson 2001). Webster and Masson (2001) evaluated resistance of this parent line to several ALS-inhibiting herbicides and reported that the parent line exhibited decreased tolerance to the imidazolinone herbicides imazamox and imazapic, compared with imazethapyr, the herbicide targeted for use with commercial imidazolinone-resistant rice varieties.

Subsequently, the cultivar 'CL 161' and the hybrid 'CLXL 8' were released which were produced from a different parent line than those cultivars previously released (Gealy 2003; Tan 2005; Croughan 2008). Meins et al. (2004) found 'CL 161' and 'CLXL 8' to have increased tolerance to imazamox equivalent to that of imazethapyr. Imazamox¹ is currently labeled for use in imidazolinone-resistant rice cultivars and hybrids; however, imazamox can only be applied following at least two imazethapyr applications.

Imazamox can be useful following imazethapyr applications due to its effectiveness controlling escaped red rice (*Oryza sativa* L.) plants that survived previous imazethapyr applications and because of its limited soil activity (Meins et al. 2003; USDA 2004). The half life of imazamox in soil was reported to be 1.4 wk compared to 16 wk for imazethapyr (Aichele and Penner 2005). This would result in less potential for injury of subsequent crops from residual herbicide residue.

Rice is a major crop produced in the four state region of Arkansas, Louisiana, Mississippi, and Texas, with these states accounting for 76% of the 1.2 million total hectares of rice planted in the U.S. and 70% of the

¹ Beyond® herbicide label. BASF Corporation, Research Triangle Park, NC, 27709.

\$3.4 billion of total value of rice produced in the U.S. in 2008 (NASSa 2009; NASSb 2009). Louisiana planted approximately 184,160 hectares of rice in 2008 with approximately 59% planted in imidazolinone-resistant rice cultivars and hybrids (LSUA 2009). Since many of the rice producing parishes in Louisiana also produce imidazolinone-resistant rice, the potential exists for off-target drift of imazamox to conventional rice.

Averaged over the ten year period 1999 to 2008, the Louisiana Department of Agriculture and Forestry (LDAF) processed 76 Pesticide Investigation Reports per year listing ground or aerial applicators in Louisiana, and, on average, six reports per year involved rice (Lisa Gautreaux², personal communication). However, it was reported that, in 2009, at least 50 rice fields were suspected of being affected by glyphosate drift and at least 25 rice fields were suspected of being affected by imazethapyr drift, many of which were not reported to the LDAF (Ronald J. Levy, Jr.³, personal communication; John K. Saichuk⁴, personal communication). Therefore, the number of rice fields actually affected by a drift event each year may be underrepresented by the number of official complaints processed by the LDAF.

It has been reported that fine spray droplets less than 150 μm in size have a greater potential to drift off-target (Hanks 1995; SDTF 1997). The use of adjuvants and selection of proper spray nozzle type, size, and application pressure can be beneficial in reducing the amount of fine spray droplets in the spray cloud (Hanks 1995; Jones et al. 2007; Nuyttens et al. 2007; VanGessel and Johnson 2005). This increase in droplet size can reduce the potential for off-target drift from droplets larger than 150 μm ; however,

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environmental conditions at the time of herbicide application can also impact the off-target drift of spray solutions (Bouse et al. 1976; Crabbe et al. 1994; Thistle 2004).

Wind speed and direction may be considered the two most important factors affecting spray droplets in the atmosphere, a stable atmosphere may be the third most important factor (Thistle 2004). A stable atmosphere, commonly referred to as an inversion, is an atmosphere that has a change of temperature with a change in elevation in the atmosphere. In a stable atmosphere, warm air overlies cool air. If air in a particular layer is displaced upward or downward it will be colder or warmer, respectively, than the immediately adjacent layer it enters and thus return to its layer of origin. If a herbicide application is made during an inversion scenario, the fine droplets that do not succumb to gravity will remain in the air layer in which they are applied due to the lack of layers mixing. The droplets in this layer can be very concentrated and horizontally may move off-target great distances.

Ultra-low-volume applications made during a stable atmosphere produced 35% more herbicide drift than applications made during turbulent atmosphere with light wind speeds (Crabbe et al. 1994). The conditions in which the greatest drift occurred were in moderately stable conditions with wind at 3 m/s resulting in off-target drift 71% at 400 m and 50% at 2200 m from the application site and in slightly stable conditions with wind at 5 m/s resulting in off-target drift 77% at 400 m and 27% at 2200 m from the application site. It is recommended that herbicide applications should be avoided during the early morning and late evening as these times are most favorable for the development of inversion conditions (Crabbe et al. 1994; Thistle 2004).

Through the use of simulated herbicide drift studies, the potential effects of imazamox drift to rice can be evaluated. In previous research,

simulated drift studies varying the spray volume proportionally with reduced herbicide rates to simulate herbicide drift resulted in increased crop injury compared with the same herbicide rate applied in a constant spray volume (Banks and Schroeder 2002; Ellis et al. 2002; Ramsdale et al. 2003; Roider et al. 2008). Banks and Schroeder (2002) reported varying spray volume proportionally with herbicide dosage, thus maintaining constant herbicide concentration in the spray, would change the response of sweet corn (*Zea mays* L.) to glyphosate when compared with a constant spray volume where herbicide concentration would vary and be more dilute in the carrier. The no-effect glyphosate rate for sweet corn was 0.046 kg ae/ha when using a spray volume proportional to the reduced glyphosate rate; however, the no-effect glyphosate rate was four times greater when glyphosate was applied in a constant spray volume.

Deeds et al. (2006) reported imazamox applied to wheat (*Triticum aestivum* L.) at the flowering and jointing growth stages at 33% of the labeled use rate of 35 g ai/ha reduced wheat yield more than 90%. A simulated drift application of the commercial herbicide premix of imazethapyr plus imazapyr affected rice plant height and yield; however, simulated drift of the imazethapyr plus imazapyr premix did not affect yield when applied to corn (Bond et al. 2006).

Rapid rice seed germination and seedling growth can enhance plant stand establishment in commercial fields (Krishnasamy and Seshu 1989; Pollock and Roos 1972; Wright 1980). Simulated imazamox drift applications applied to wheat at the jointing and flowering growth stages had no effect on wheat seed germination (Deeds et al. 2006). Regardless of timing, applications of glyphosate and imazethapyr to red rice (*Oryza sativa* L.) in the two to three-tiller, boot, and bloom growth stages reduced red rice seed germination (Brommer et al. 1998). Glyphosate applied to wheat at first node, boot, and early flowering growth stages resulted in 16 to 36% reductions in seed weight

(Roider et al. 2007). A negative correlation between 100-count seed weight and rice seed germination has been observed (Krishnasamy and Seshu 1989). A need exists to evaluate the possible effects of an imazamox drift event on rice crop seed germination and seedling vigor.

The objectives of this research were to evaluate the effects of simulated imazamox drift applied to rice during the primary crop on the primary and ratoon rice crops and the impact on the seed produced on treated rice.

Materials and Methods

Simulated Imazamox Drift Field Study. A study was conducted on rice grown in 2005 through 2007 at the LSU AgCenter Rice Research Station near Crowley, Louisiana on a Crowley silt loam (fine montmorillonitic, thermic Typic Albaqualf) with pH 5.5 and 1.2% organic matter. Field preparation consisted of a fall and spring disking and two passes in opposite directions with a two-way bed conditioner equipped with rolling baskets and S-tine harrows 15 cm deep. The long grain rice cultivar 'Cocodrie' was drill-seeded March 28 to April 17 in 2005 through 2007. Plots consisted of twelve-18 cm spaced rows 6 m long.

The experimental design was an augmented two-factor factorial arrangement of treatments in a randomized complete block with four replications. Factor A consisted of imazamox applied at simulated drift rates of 6.3 and 12.5% of the labeled usage rate of 44 g ai/ha, or 2.7 and 5.5 g/ha, respectively. Factor B consisted of application timings at different growth stages: one-tiller, panicle differentiation (PD), boot, and physiological maturity. Each herbicide application was made with the spray volume varying proportionally to herbicide dosage based on a constant spray volume of 234 L/ha. The 12.5% herbicide rate was applied at a spray volume of 29 L/ha and the 6.3% herbicide rate was applied at a spray volume of 15 L/ha. Each application was made with a tractor-mounted CO₂-pressurized

sprayer calibrated to deliver a constant carrier volume with speed adjusted to vary application rate and equipped with Teejet^{®5} TX-2 Conejet[®] 800033 nozzles. A ratoon rice crop was not produced in 2006 due to unfavorable weather following primary crop harvest.

The study area was maintained weed-free using clomazone at 420 g ai/ha applied preemergence followed by propanil at 4483 g ai/ha plus halosulfuron at 53 g ai/ha applied postemergence. For the primary rice crop a preplant application of 280 kg/ha of 8-24-24 (N-P₂O₅-K₂O) fertilizer and a preflood application of 365 kg/ha 46-0-0 urea fertilizer were applied to the study area and for the ratoon rice crop a preflood application of 100 kg/ha 46-0-0 urea fertilizer was applied to the study area to maintain proper fertility and to maximize yields in the primary and ratoon crops. Standard agronomic and pest management practices were implemented throughout the growing season to maximize yield.

Rice plant height and rice injury in the primary rice crop were obtained 7 days after herbicide treatment (DAT) and continued weekly for 28 DAT. Rice plant height was obtained by measuring four plants per plot from the soil surface to the tip of the extended uppermost emerged leaf or extended rice panicle. Rice injury was evaluated based on chlorosis and necrosis of foliage and reduced plant height using a scale of 0 to 100% where 0 = no injury and 100 = plant death. Rice plant height at primary crop harvest and rough rice yield, 100-count seed weight, and stem and panicle counts for the primary and ratoon crop were also obtained. Whole plots were harvested using a mechanical plot harvester and rough rice yield was adjusted to 12% moisture. Total stem and panicle counts were calculated by hand harvesting a 0.46 m section of row and determining the number of stems present at the mid-height of the plants and the number of panicles with bases

⁵ Spraying Systems Co., P. O. Box 7900, Wheaton, IL 60187.

emerged beyond the sheath of the flag leaf, or the last leaf to emerge prior to the panicle.

All data were subjected to the Mixed Procedure of SAS (SAS 2003). Year, replications (nested within year), and all interactions containing either of these effects were considered random effects. Application timing and rate were considered fixed effects. Considering year or combination of year as random effects permits inferences about treatments over a range of environments (Carmer et al. 1989; Leon et al. 2008). Type III statistics were used to test all possible effects of fixed factors (application timing and rate) and least square means were used for mean separation at the 5% probability level ($p \leq 0.05$).

Seed Germination Study. The germination potential of seed collected from grain harvested in the simulated imazamox drift field study at primary crop harvest, 2005 through 2007, and at ratoon crop harvest, 2005 and 2007, was evaluated at multiple temperatures. Seed collected from each plot was air-dried and stored at 8 C. Germination temperatures evaluated were 13, 16, 19, 22, and 25 C. Temperature selection and germination testing procedure for this study were based on procedures previously described by Webster et al. (2003) and follow germination procedures recommended by Association of Official Seed Analysts (AOSA) (AOSA 2006). Temperature selection was based on 19 C being the historical mean 10-cm soil temperature in Crowley, LA on April, 1, which corresponds to 50% of the rice being planted across the state (Webster et al. 2003).

One hundred seeds from each field plot were prepared by soaking for 30 min in a 50:50 (v/v) solution of chlorine bleach and distilled water to decrease seedling diseases. After soaking, seeds were triple rinsed with distilled water. After seed preparation, seeds were placed in a 10 cm

plastic Petri dish between two 9 cm germination blotters⁶. Next, 10 ml of carboxin (5,6-dihydro-2-methyl-*N*-phenyl-1,4-oxathiin-3-carboxamide) plus thiram (tetramethylthiuram disulfide) plus distilled water solution (52 ml of a 10% carboxin and 10% thiram premix liquid fungicide combined with 948 ml distilled water) was applied in each Petri dish to reduce seedling diseases. Petri dishes were sealed with Parafilm M⁷ to prevent moisture loss and placed in a constant-temperature growth chamber in total darkness. Germination counts were taken 5, 9, and 14 d after initiation (DAI) of the study. A seed was considered germinated if the radical had reached a length of 1 mm.

Seed germination data were arranged as repeated measures and subjected to the Mixed Procedure of SAS (SAS 2003). Year, replications (nested within year), DAI (nested within replications), and all interactions containing either of these effects were considered random effects. Application timing and rate and germination temperature were considered fixed effects. Considering year or combination of year as random effects permits inferences about treatments over a range of environments (Carmer et al. 1989; Leon et al. 2008). Type III statistics were used to test all possible effects of fixed factors (application timing and rate and germination temperature) and least square means were used for mean separation at the 5% probability level ($p \leq 0.05$).

Seedling Vigor Study. Vigor of seedlings from grain collected at primary crop harvest in the simulated imazamox drift field study in 2006 and 2007 was examined. Seedling vigor, as defined by AOSA (AOSA 2002), is "seedling vigor comprises those seed properties which determine the potential for rapid, uniform emergence, and development of normal seedlings under a wide range of field conditions" and it is used as a measure of seed quality by producers.

⁶ Anchor Steel Blue Seed Germination Blotter®, SDB 3.5. Anchor Paper Company, 480 Broadway, St. Paul, MN 55101.

⁷ Parafilm M®. Pechiney Plastic Packaging, Menash, WI 54952.

Since there is no accepted standard vigor test for rice, vigor testing procedures for this study were based on procedures previously described by Webster et al. (2003).

Approximately 100 seeds from each field plot were prepared by soaking for 30 min in a 50:50 (v/v) solution of chlorine bleach and distilled water to decrease seedling diseases. After soaking, seeds were triple rinsed with distilled water. Following seed preparation, seeds were pre-germinated by soaking in distilled water for 24 h. Ten pre-germinated seeds from each field plot were placed on a single sheet of nontreated germination paper⁸ cut to fit a 12 by 23 by 0.3 cm acrylic sheet. Germination paper was moistened by submerging in distilled water for 5 seconds to facilitate adherence to the acrylic sheet and provide residual moisture to rice seeds. Seeds were placed along the center of germination paper oriented with the radical end of the seed toward the lower half of the sheet. A one-ply paper towel strip was placed over the seed, and 5 ml of a mancozeb [ethylene (bis)-dithiocarbamate] plus distilled water solution (dry formulation of mancozeb at 1640 mg ai/L distilled water) was applied on top of the strip to reduce seedling diseases. The plated seeds were then placed vertically in a rack and then placed in a 30 by 51 by 5 cm dish with 1,420 ml of distilled water to allow for evaporation. The dish and racks of plates were wrapped in plastic wrap to prevent desiccation. The glass dish was placed in a constant temperature growth chamber at 21 C for 12 d in total darkness. At the end of 12 d, shoot lengths were measured and an average of the 10 shoot lengths was obtained for data analysis.

Seedling vigor data were subjected to the Mixed Procedure of SAS (SAS 2003). Year, replications (nested within year), and all interactions containing either of these effects were considered random effects.

⁸ Anchor Steel Blue Seed Germination Blotters®, SDB 1924. Anchor Paper Company, 480 Broadway, St. Paul, MN 55101.

Application timing and rate were considered fixed effects. Considering year or combination of year as random effects permits inferences about treatments over a range of environments (Carmer et al. 1989; Leon et al. 2008). Type III statistics were used to test all possible effects of fixed factors (application timing and rate) and least square means were used for mean separation at the 5% probability level ($p \leq 0.05$).

Results and Discussion

Simulated Imazamox Drift Field Study. A crop injury response was observed in the primary crop (Table 5.1). Regardless of rate, simulated imazamox drift applications at one-tiller resulted in the greatest amount of injury at 7, 14, and 21 DAT, 25 to 36%. Imazamox at 5.5 g/ha applied at PD and boot resulted in 11 to 14% injury 21 DAT. At 28 DAT, imazamox at either rate at one-tiller and boot and 5.5 g/ha at PD resulted in 15 to 26% injury. An increase in injury from 21 to 28 DAT with rice treated with imazamox at boot was noted because necrosis of the flag leaf was observed at 28 DAT that was not present at 21 DAT. No response was observed on rice treated with imazamox at maturity. These data indicate that visual injury to rice is more severe when imazamox is applied during the early, vegetative growth stage of rice. As with actual drift events, identifying drift based on visual injury is more difficult as rice matures (Eric P. Webster⁹, personal communication). When evaluating the efficacy of imazethapyr on selected weed species, visual injury symptoms were more severe on plants treated at earlier timings (Hoss et al. 2003; Shaw et al. 1990).

This reduction in visual injury during reproductive growth stages may be due to the translocation of imazamox to meristematic tissue (Shaner et al. 1984). This tissue is located in the internal portions of the rice plant

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Table 5.1. Effects of simulated imazamox drift application rate and timing on primary rice crop injury 7, 14, 21, and 28 days after treatment (DAT), 2005 through 2007, Crowley, Louisiana.^a

Imazamox		Injury			
		7 DAT	14 DAT	21 DAT	28 DAT
rate ^b	Timing	%			
2.7	1-tiller	25 a	27 a	25 a	20 a
	PD	2 b	8 b	5 bc	5 bc
	Boot	7 b	8 b	9 bc	23 a
	Maturity	0 b	0 b	0 c	0 c
5.5	1-tiller	32 a	36 a	33 a	26 a
	PD	3 b	11 b	14 b	15 ab
	Boot	7 b	8 b	11 b	23 a
	Maturity	0 b	0 b	0 c	0 c
Nontreated		0 b	0 b	0 c	0 c

^a Means within a column followed by the same letter were not statistically different according to the t-test on difference of least square means at P = 0.05.

^b The 2.7 and 5.5 g/ha imazamox rates were applied at spray volumes of 15 and 29 L/ha, respectively.

during the reproductive stages of growth and would not be expressed on foliar tissue.

The injury symptoms observed in this study on plants treated at the one-tiller timing were an interveinal chlorosis in the uppermost leaves (Figure 5.1) to plant death. Leaves of treated plants often exhibited small, narrow reddish-brown leaf lesions similar to those associated with leaf blast disease of rice (Groth et al. 2009). Subsequent tillers on recovering treated plants often emerged along a single plane causing a flat, fan-shaped appearance in plants. Also, an overall stunting of plants was observed on plants treated at the one-tiller and PD timings (Table 5.2).

Visual symptomology observed on plants treated with imazamox at PD and boot, often beyond the rating dates evaluated in this study, were various forms of foliar and inflorescence malformations. Foliar symptoms were plants having multiple shoots arising from the secondary nodes of the main stem (Figure 5.2). The flag leaf on the main stem and secondary shoots would often appear malformed, wrinkled, contorted, or rolled. In some instances secondary shoots were stunted or both stunted and malformed. Panicles may partially exert beyond the flag leaf sheath or emerged from the side of the sheath (Figure 5.3). Often panicles failed to initiate emergence from the flag leaf sheath and decomposed in the leaf sheath causing necrosis of the flag leaf if the plants were treated at the boot stage (Figure 5.4). Some of the inflorescence malformations were due to a malformed panicle axis and partially emergence of the panicle due to fusing of the panicle with the flag leaf sheath. Individual florets malformations that were observed were florets with the tips of the lemma excessively curved toward the palea (Figure 5.3) causing an appearance often referred to as "parrot beaked" when observed in association with the straighthead physiological disorder of rice (Groth et al. 2009).

Table 5.2. Effects of simulated imazamox drift application rate and timing on primary crop rice plant height at 7, 14, 21, and 28 days after treatment (DAT) and at harvest, 2005 through 2007, as percent of the nontreated, Crowley, Louisiana.^a

		Rice plant height				
Imazamox						
rate ^b	Timing	7 DAT ^c	14 DAT ^d	21 DAT ^e	28 DAT ^f	Harvest ^g
g ai/ha		% of nontreated				
2.7	1-tiller	80 c	91 de	87 e	86 cd	93 b
	PD	99 a	96 cd	96 cd	97 ab	100 a
	Boot	98 ab	96 cd	96 cd	91 bc	92 b
	Maturity	95 ab	102 ab	108 a	101 a	100 a
5.5	1-tiller	66 d	82 f	85 e	83 d	90 b
	PD	93 b	89 e	88 e	88 cd	92 b
	Boot	97 ab	94 d	94 d	91 bc	91 b
	Maturity	98 ab	106 a	105 ab	103 a	100 a
Nontreated		100 a	100 bc	100 bc	100 a	100 a

^a Means within a column followed by the same letter were not statistically different according to the t-test on difference of least square means at P = 0.05.

^b The 2.7 and 5.5 g/ha imazamox rates were applied at spray volumes of 15 and 29 L/ha, respectively.

^c Actual heights of nontreated rice for the 1-tiller, PD, boot, and maturity timings were 40, 65, 93, and 40 cm, respectively.

^d Actual heights of nontreated rice for the 1-tiller, PD, boot, and maturity timings were 51, 72, 99, and 51 cm, respectively.

^e Actual heights of nontreated rice for the 1-tiller, PD, boot, and maturity timings were 64, 77, 99, and 57 cm, respectively.

^f Actual heights of nontreated rice for the 1-tiller, PD, boot, and maturity timings were 77, 86, 98, and 62 cm, respectively.

^g Actual height of nontreated rice at primary crop harvest was 95 cm.



Figure 5.1. Symptoms observed with a one-tiller application of 2.7 g ai/ha imazamox.



Figure 5.2. Symptoms observed with a boot application of 5.5 g ai/ha imazamox.



Figure 5.3. Symptoms observed with a boot application of 2.7 g ai/ha imazamox.



Figure 5.4. Symptoms observed with a boot application of 5.5 g ai/ha imazamox.

A plant height response was observed in the primary rice crop when imazamox was applied to rice (Table 5.2). With the exception of a boot application at 7 DAT, 5.5 g/ha imazamox applied at one-tiller, PD, and boot reduced rice plant height at 7, 14, 21, and 28 DAT. Imazamox applied at 2.7 g/ha resulted in reduced plant height at 7 through 28 DAT when applied at one-tiller. Imazamox applied at 5.5 g/ha resulted in the greatest reduction in height at 7 and 14 DAT when applied at one-tiller, 66 and 82% of the nontreated, respectively. At primary crop harvest, rice plant height was 90 to 100% of the nontreated, regardless of application rate or timing. Imazamox applied to mature rice had no affect on primary crop rice plant height. These findings support the trend of increased crop injury at earlier application timings. Similar findings were reported by Bond et al. (2006).

Stem and panicle counts in the primary and ratoon crops were affected by imazamox applications (Table 5.3). Imazamox applied at PD and boot increased secondary plant stems in the primary crop resulting in an increase in stem count compared with the nontreated. This increase was due to imazamox causing an excess of secondary stems to be produced on the upper plant nodes in the primary rice crop. However, this increase in stems did not translate into an increase in the number of panicles in the primary crop. In the ratoon crop, an increase in stem counts was observed when imazamox was applied to rice at one-tiller and boot; however, an increase in panicle counts was only observed in rice treated at the boot stage, compared with the nontreated (Table 5.3).

A primary crop rice yield response was observed (Table 5.4). Primary crop yield was 79 and 83% of the nontreated when imazamox was applied to rice at 2.7 g/ha at one-tiller and 5.5 g/ha at PD, respectively. Imazamox applied to rice at 5.5 g/ha at one-tiller and 2.7 g/ha at boot resulted in a primary crop yield 54 to 58% of the nontreated. The greatest reduction in primary

Table 5.3. Effects of simulated imazamox drift application timing on primary crop rice stem and panicle counts, 2005 through 2007, and ratoon crop rice stem and panicle counts, 2005 and 2007, as percent of the nontreated, Crowley, Louisiana.^a

Imazamox timing	Primary crop counts		Ratoon crop counts	
	Stem	Panicle	Stem	Panicle
	% of nontreated ^{b,c}			
1-tiller	92 c	86 b	123 b	103 b
PD	112 b	112 a	112 bc	99 b
Boot	141 a	90 b	156 a	188 a
Maturity	97 c	96 ab	110 bc	103 b
Nontreated	100 c	100 ab	100 c	100 b

^a Means within a column followed by the same letter were not statistically different according to the t-test on difference of least square means at P = 0.05.

^b Data averaged across application rates of 2.7 and 5.5 g ai/ha imazamox applied at spray volumes of 15 and 29 L/ha, respectively.

^c Actual nontreated primary crop stem and panicle counts were 38 and 35 per 0.46 m of row, respectively, and nontreated ratoon crop stem and panicle counts were 38 and 30 per 0.46 m of row, respectively.

Table 5.4. Effects of simulated imazamox drift application rate and timing on primary crop rice yield, 2005 through 2007, and ratoon and total crop rice yield, 2005 and 2007, as percent of the nontreated, Crowley, Louisiana.^a

Imazamox rate ^b	Timing	Yield		
		Primary crop	Ratoon crop	Total crop
g ai/ha		% of nontreated ^c		
2.7	1-tiller	79 b	96 b	72 b
	PD	100 a	100 b	101 a
	Boot	58 c	135 a	60 bc
	Maturity	99 a	99 b	101 a
5.5	1-tiller	54 c	103 b	62 bc
	PD	83 b	97 b	89 a
	Boot	34 d	156 a	47 c
	Maturity	102 a	104 b	101 a
Nontreated		100 a	100 b	100 a

^a Means within a column followed by the same letter were not statistically different according to the t-test on difference of least square means at P = 0.05.

^b The 2.7 and 5.5 g/ha imazamox rates were applied at spray volumes of 15 and 29 L/ha, respectively.

^c Actual nontreated yield for the primary, ratoon, and total crops were 5900, 1300, and 7200 kg/ha, respectively.

crop yield was observed on plants treated with 5.5 g/ha imazamox at boot, 34% of the nontreated. However, regardless of rate, imazamox applied at boot resulted in a ratoon crop rice yield 135 to 156% of the nontreated. This increase was due to imazamox causing an excess of secondary stems to be produced on the upper plant nodes in the primary rice crop (Table 5.3). This excess of secondary stems did not produce panicles in the primary crop but did produce panicles in the ratoon crop. This increase in ratoon crop panicles was not observed with rice treated at the other timings. However, when primary and ratoon crop yields were combined, the increase in ratoon crop yield did not compensate for the primary crop yield loss. Total crop yield was 47 to 72% of the nontreated when imazamox was applied to rice at one-tiller and boot. Imazamox had no effect on primary, ratoon, or total crop yield when applied to mature rice.

Though primary crop rice yield was reduced by simulated imazamox drift applications at the one-tiller, PD, and boot timings, it appears that rice is most susceptible to imazamox during the boot growth stage. Similar results were reported by Hensley et al. (2009) when evaluating simulated imazethapyr drift on rice. Rice producers in Louisiana may have the ability to recover some yield loss from an imazamox drift event occurring to rice during the boot growth stage by increasing ratoon crop yield; however, the reduction in total crop yield from an imazamox drift event at the boot growth stage of rice has the potential to be significant. These data also indicate an increased susceptibility to imazamox drift occurring at the one-tiller timing compared to the PD timing. This may be due to the reduced plant biomass at this growth stage compared to the later PD growth stage. Shaw et al. (1990) reported an increased susceptibility to imazethapyr in smaller plants when evaluating its effects on johnsongrass (*Sorghum halepense* L.) at 15, 30, and 60 cm plant heights. Though rice has the ability to recover from imazamox drift occurring at the vegetative one-tiller stage, if a combination of

herbicide drift and climatic conditions unsuitable for growth hinder recovery, yield losses may be significant (Eric P. Webster⁷, personal communication).

Seed Germination and Seedling Vigor Studies. Simulated imazamox drift applications did not affect primary crop rice seed weight (Table 5.5) or ratoon crop rice seed weight (data not shown). It is expected that any unfilled or malformed grain observed on rice panicles on treated plants was separated and expelled by the mechanical plot harvester. This separation is similar to a commercial harvesting operation so any affect on seed weight, germination, or seedling vigor of harvested grain observed in this study is reflective of the impact expected on commercial seed rice producers. Studies conducted using hand-harvesting of seeds, such as Walker and Oliver (2008), which bypass a separation process, may misrepresent the impact of herbicides on seed in mechanically harvested grain crops.

Primary crop rice seed germination was affected by simulated imazamox drift applications (Table 5.6). Imazamox applications had no affect on primary crop germination at 13 and 16 C; however, the greatest reduction in germination, at 19, 22, and 25 C were observed in plants treated at boot with 5.5 g/ha imazamox. Primary crop rice seed germination was reduced at 19 and 22 C when 5.5 g/ha imazamox was applied at PD and at 22 and 25 C when 2.7 g/ha imazamox was applied at boot. These data indicate an increased susceptibility to reduced rice seed germination from imazamox drift occurring to rice in the reproductive growth stages, especially the boot stage. Reductions in germination of this magnitude can lead to a significant increase in seed cost to rice producers. Compared with the nontreated at 19 C, a producer using seed affected by the 5.5 g/ha imazamox rate at boot would have to approximately double their seeding rate to achieve a rice plant density comparable to that of the nontreated seed. Imazamox applications

Table 5.5. Effects of simulated imazamox drift application timing on primary crop rice seed weight, 2005 through 2007, and seedling vigor, 2006 and 2007, as percent of the nontreated, Crowley, Louisiana.^a

Imazamox timing	100-seed weight	Seedling vigor
	————— % of nontreated ^{b,c} —————	
1-tiller	98 a	98 ab
PD	98 a	103 a
Boot	97 a	90 b
Maturity	99 a	103 a
Nontreated	100 a	100 a

^a Means within a column followed by the same letter were not statistically different according to the t-test on difference of least square means at P = 0.05.

^b Data averaged across application rates of 2.7 and 5.5 g ai/ha imazamox applied at spray volumes of 15 and 29 L/ha, respectively.

^c Actual nontreated 100-seed weight and seedling vigor shoot length was 2300 mg and 35 mm, respectively.

Table 5.6. Effects of simulated imazamox drift application rate and timing on primary crop rice seed germination at various temperatures, 2005 through 2007, as percent of the nontreated, Crowley, Louisiana.^a

Imazamox rate ^b		Temperatures				
		13 C	16 C	19 C	22 C	25 C
g ai/ha		% of nontreated ^c				
2.7	1-tiller	67 a	86 a	94 abc	93 de	100 a
	PD	111 a	98 a	100 ab	102 b	96 ab
	Boot	89 a	86 a	88 bc	89 ef	88 b
	Maturity	111 a	95 a	98 abc	101 b	100 a
5.5	1-tiller	44 a	86 a	98 abc	96 cd	100 a
	PD	89 a	95 a	86 c	87 f	97 a
	Boot	78 a	79 a	55 d	80 g	62 c
	Maturity	100 a	98 a	102 a	109 a	101 a
Nontreated		100 a	100 a	100 ab	100 bc	100 a

^a Means within a column followed by the same letter were not statistically different according to the t-test on difference of least square means at P = 0.05.

^b The 2.7 and 5.5 g/ha imazamox rates were applied at spray volumes of 15 and 29 L/ha, respectively.

^c Actual germination of the nontreated seed at 13, 16, 19, 22, and 25 C was 9, 43, 49, 70, and 65%, respectively.

did not reduce ratoon crop rice seed germination, compared with the nontreated (Table 5.7).

A primary crop seedling vigor response was observed (Table 5.5). Primary crop seedling vigor was reduced to 90% of the nontreated when imazamox was applied to rice at boot. These data indicate that even if there is no notable reduction in seed weight, the grain that remains after mechanical harvesting may have reduced seed germination and seedling vigor if an imazamox drift event occurs to rice, especially at the boot growth stage. If seed rice is affected by an imazamox drift event, extra caution should be taken before that seed is sold to producers.

In conclusion, simulated imazamox drift applications at the one-tiller, PD, and boot timings resulted in reduced plant height and primary crop yield losses. Total crop yield was reduced when imazamox was applied at one-tiller and boot with the greatest reduction in primary and total crop yield resulting from imazamox applied at the boot growth stage. Imazamox applications to mature rice had no effect on rice plant height or yield. Primary crop rice seed weight was not reduced by an imazamox application to rice; however, primary crop rice seed germination was reduced when imazamox was applied at one-tiller, PD, and boot, with increased susceptibility at the boot growth stage. Primary crop seedling vigor was reduced when imazamox was applied to rice at boot. Ratoon crop rice seed weight and seed germination was not reduced by imazamox applications.

The ability to identify imazamox drift on rice can be helpful to producers, Cooperative Extension Service personnel, crop consultants, and state regulatory agencies in distinguishing between herbicide drift and injury associated with soil fertility issues, diseases, and other disorders affecting rice. Misidentification of herbicide drift symptoms as injury associated with these factors can lead to loss in profitability if growers

Table 5.7. Effects of simulated imazamox drift application timing on ratoon crop rice seed germination at various temperatures, 2005 and 2007, as percent of the nontreated, Crowley, Louisiana.^a

Imazamox timing	Temperatures				
	13 C	16 C	19 C	22 C	25 C
	————— % of nontreated ^{b,c} —————				
1-tiller	133 a	139 a	116 a	109 a	113 a
PD	100 a	89 b	91 b	98 c	97 c
Boot	100 a	100 b	111 a	105 ab	103 bc
Maturity	100 a	89 b	100 b	100 bc	100 bc
Nontreated	100 a	100 b	100 b	100 bc	100 bc

^a Means within a column followed by the same letter were not statistically different according to the t-test on difference of least square means at P = 0.05.

^b Data averaged across application rates of 2.7 and 5.5 g ai/ha imazamox applied at spray volumes of 15 and 29 L/ha, respectively.

^c Actual germination of the nontreated seed at 13, 16, 19, 22, and 25 C was 3, 18, 45, 57, and 62%, respectively.

apply unnecessary applications of inputs to correct these factors when the symptoms present are actually a result of herbicide drift. The ability to correlate the symptoms observed to imazamox drift also may assist state regulatory agencies in identifying the source of a herbicide drift event. If imazamox can be identified by observation of plant symptoms this can reduce the cost associated with confirmation of a herbicide drift event through the use of diagnostic testing of foliar residue since most analytical facilities charge per evaluation and the diagnostic tests involved are often herbicide specific.

An imazamox drift event occurring to a producer's field at the one-tiller, PD, or boot growth stages of rice can reduce yield and germination of harvested seed; however, this study indicates that an imazamox drift event occurring to rice at the boot stage may be the most detrimental to yield and seed germination. Rice receiving a drift event in vegetative growth stages, one-leaf to one-tiller, can often recover if stand is maintained at recommended densities (Eric P. Webster⁹, personal communication). However, an imazamox drift event occurring to rice in the reproductive stage of growth may have little to no visual foliar injury and often symptoms may not appear until rice plants near crop maturity. This may lead to loss of yield and profitability due to continuing to supply crop inputs, such as increased fertilizer, insecticide, and fungicide applications, to a crop that has an already reduced yield potential. Unfortunately, due to the use of imazamox for late season red rice control following imazethapyr applications, the potential for off-target drift of imazamox is greater during the reproductive growth stages of rice. The negative effects of an imazamox drift event occurring to a seed producer's field to rice in the PD or boot growth stages has the potential to be two-fold. The reduction in profitability the year of the event from reduced yield in combination with the reduction in seed germination has the ability to reduce profitability in the subsequent year's

crop due to an increase in seeding rate to offset the reduced seed germination. If seed germination is too low seed may not be sold as seed rice which can decrease profitability more.

Caution should be taken when applying imazamox near adjacent susceptible rice fields, especially when making applications near rice in the reproductive growth stages. Though the effects of imazamox drift on rice may not be immediately apparent by visual observation, the potential affect on grain yield and the germination potential of the harvested grain could be highly detrimental to rice producers.

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Chapter 6

Summary and Conclusions

Simulated glyphosate drift applied to rice at the one-tiller, PD, and boot timings resulted in reduced plant height and primary and total crop yield losses, with the greatest reduction in primary crop yield resulting from glyphosate applied at the boot growth stage. Primary crop rice seed weight was not affected by glyphosate applications; however, primary crop rice seed germination was reduced when glyphosate was applied at one-tiller, PD, and boot, with increased susceptibility at the boot growth stage. Seeding vigor of primary crop rice seed was not affected by glyphosate applications. A reduction in ratoon crop rice seed weight by glyphosate applications at one-tiller was observed; however, ratoon crop rice seed germination was not reduced.

Simulated imazethapyr drift applied to rice at the one-tiller, PD, and boot timings resulted in reduced plant height and primary and total crop yield losses, with the greatest reduction in primary crop yield resulting from imazethapyr applied at the boot growth stage. Primary crop rice seed weight was reduced by an imazethapyr application to rice at boot. Primary crop rice seed germination was reduced when imazethapyr was applied at one-tiller, PD, and boot, with increased susceptibility at the boot growth stage. A reduction in ratoon crop rice seed weight was not observed; however, ratoon crop rice seed germination was reduced by an imazethapyr application at the PD growth stage.

Simulated glufosinate drift applied to rice at the one-tiller, PD, and boot growth stages resulted in reduced rice plant height. Primary and total crop rice yield was reduced by glufosinate applied at the boot growth stage. Averaged across application timings, primary crop rice seed germination and rice seedling vigor were reduced by glufosinate applications, regardless of rate.

Simulated imazamox drift applied to rice at the one-tiller, PD, and boot timings resulted in reduced plant height and primary crop yield losses. Total crop yield was reduced when imazamox was applied at one-tiller and boot with the greatest reduction in primary and total crop yield resulting from imazamox applied at the boot growth stage. Primary crop rice seed weight was not reduced by an imazamox application to rice; however, primary crop rice seed germination was reduced when imazamox was applied at one-tiller, PD, and boot, with increased susceptibility at the boot growth stage. Primary crop seedling vigor was reduced when imazamox was applied to rice at boot. Ratoon crop rice seed weight and seed germination was not reduced by imazamox applications.

Simulated glyphosate, imazethapyr, glufosinate, and imazamox drift applications did not affect rice when applied at maturity. The greatest reduction in primary crop yield was observed when each herbicide was applied to rice at the boot growth stage. Although each herbicide was evaluated separately, the yield reduction observed with simulated glyphosate, imazethapyr, and imazamox drift applied to rice at the boot growth stage was greater than that observed with glufosinate.

Simulated drift of glyphosate, imazethapyr, glufosinate, and imazamox reduced primary crop rice seed germination. Generally, glyphosate, imazethapyr, and imazamox applications at the boot timing resulted in the greatest reduction in seed germination. Glufosinate reduced primary crop seed germination regardless of timing.

The negative effects of a glyphosate, imazethapyr, glufosinate, or imazamox drift event occurring to a seed producer's field to rice in the boot growth stage has the potential to be two-fold. The reduction in profitability the year of the event from reduced yield and a reduction in seed germination has the ability to reduce profitability in the subsequent year's crop due to an increase in seeding rate to offset the reduced seed

germination. Caution should be taken when applying any of these herbicides near adjacent susceptible rice fields, especially when making applications near rice in the reproductive growth stages. Though the effects of glyphosate, imazethapyr and imazamox drift on rice may not be immediately apparent by visual observation, the potential effect on grain yield and the germination potential of the harvested grain could be highly detrimental to rice producers.

One important aspect of this research is the identification and documentation of the symptomology associated with glyphosate, imazethapyr, glufosinate, and imazamox drift to rice. While an understanding of the potential for yield loss is important, one must first be able to identify the causal agent of a suspected drift event before a potential detriment to yield can begin to be estimated.

The ability to identify drift of these herbicides can be helpful to producers, Cooperative Extension Service personnel, crop consultants, and state regulatory agencies in distinguishing between herbicide drift and injury associated with soil fertility issues, diseases, and other disorders affecting rice. Misidentification of herbicide drift symptoms as injury associated with these factors can lead to loss in profitability if growers apply unnecessary applications of inputs to correct these factors when the symptoms present are actually a result of herbicide drift. The ability to correlate the symptoms observed to a particular herbicide also may assist state regulatory agencies in identifying the source of a herbicide drift event. If the suspected herbicide can be identified by observation of plant symptoms this can reduce the cost associated with confirmation of a herbicide drift event through the use of diagnostic testing of foliar residue since most analytical facilities charge per evaluation and the diagnostic tests involved are often herbicide specific.

The landscape of agriculture in Louisiana is one of diversity which provides ample opportunity for off-target herbicide drift to affect adjacent crops. While the knowledge of the detrimental effects and symptoms associated with glyphosate, glufosinate, imazethapyr, and imazamox drift are valuable following a drift event of these herbicides, the gains involved in avoiding off-target drift to susceptible crops are much greater. Growers and commercial applicators must use caution when applying these herbicides near susceptible rice.

Vita

Justin Brian Hensley was born in December 1975 and was raised in Lonoke, Arkansas. He attended Lonoke High School, graduating in 1994. He began his college career at the Beebe branch of Arkansas State University but completed his Bachelor of Science at the University of Arkansas, Fayetteville, in 1999. Following graduation he worked as a County Extension Agent in St. Francis County Arkansas for the University of Arkansas Cooperative Extension Service with responsibility in row crop agriculture. During his time as a County Agent, he began pursuing a Master of Science degree which he completed in May 2004. In January 2005 he began pursuing the degree a Doctor of Philosophy in weed science under the direction of Dr. Eric Webster at Louisiana State University.