EVALUATION OF EFFICACY, SOIL BEHAVIOR AND DISSIPATION OF HERBICIDES IN AGRONOMIC CROPS

by

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(Under the Direction of Timothy Grey)

ABSTRACT

Field, greenhouse and laboratory experiments were conducted to investigate fomesafen soil behavior, degradation, dissipation and cotton tolerance. Fomesafen adsorption to soil was significantly affected by pH and clay content while desorption was correlated to sand, silt, clay fraction, pH and soil organic matter. Fomesafen degradation was minimum in Cecil sandy loam or Tifton loamy sand during a 90 day laboratory incubation. Under field conditions, fomesafen persistence varied significantly between Cecil sandy loam and Tifton loamy sand. The half-life in the respective soils was 47 and 6 d for 280 g ai ha^{-1}; and 34 and 4 day for 560 g ai ha^{-1}. Cotton was not damaged when fomesafen applied preemergence within the 280 to 420 g ai ha^{-1} registered use rates. However stand count, height and yield may be reduced by fomesafen rates exceeding 1120 g ai ha^{-1}. Herbicide tolerance and efficacy were initiated for *Miscanthus* × *giganteus* in laboratory, greenhouse and field, with the objective of screening potential herbicides to control weeds during *M. giganteus* establishment and eradicating *M. giganteus* for crop rotation and invasive control. Preemergence herbicide screening in greenhouse experiments indicated *M. giganteus* rhizomes were tolerant of atrazine, *S*-metolachlor, mesotrione, pendimethalin, acetochlor and metribuzin. However, experiments that screened preemergence

herbicides using *M. giganteus* fertile seeds indicated seed germination failed completely when treated with dinitroanilines, cellulose synthesis inhibitor, and protoporphyrinogen oxidase inhibitors; germination responses to very long chain fatty acid inhibitors varied from 46 to 94%. In postemergence herbicide screening experiments, nicosulfuron, trifloxysulfuron, sulfometuron, pyrithiobac, clodinafop and fluazifop reduced shoot dry weight of rhizome-established *M. giganteus* but only sulfometuron and fluazifop affected shoot regrowth from rhizomes. The glyphosate rate to reduce 50% growth compared to nontreated control for Miscanthus shoot dry weight, underground biomass and regrowth shoot dry weight were 702, 1174 and 1637 g ae ha^{-1} respectively. Single glyphosate application of 1.68 kg ae ha^{-1} reduced shoot height and dry weight, but did not affect underground biomass and shoot regrowth; two applications were required to eliminate regrowth. Postemergence glyphosate tank mixed with fluazifop, imazapyr, pyrithiobac or sulfometuron improved control efficacy compared to glyphosate alone.

INDEX WORDS: Fomesafen, soil behavior, degradation, soil dissipation, cotton tolerance, *Miscanthus* × *giganteus*, eradication, weed control, glyphosate, preemergence and postemergence herbicides, seed germination.

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BS, China Agricultural University, China, 2008

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A Dissertation Submitted to the Graduate Faculty of The University of Georgia in Partial Fulfillment of the Requirements for the Degree

DOCTOR OF PHILOSOPHY

ATHENS, GEORGIA

2014

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August 2014

ACKNOWLEDGEMENTS

I would like to thank my major advisor Dr. Timothy Grey and co-advisor Dr. William Vencill for their kind guidance, support and advice during my PhD work at University of Georgia. I also want to thank the rest of my committee, Dr. Theodore Webster, Dr. Donn Shilling and Dr. Mark Czarnota for their time and patience. I want to express my sincere appreciation to our technicians Sidney Cromer, Mike Mathis and fellow graduate students Brian Blanchett and Daryl Chastain for their assistance in this research. Finally, I would like to especially thank my parents Li Dawei, Sun Hongjuan and my fiancée Callie Yuan for their long spiritual and financial support for my education and research. I could never achieve these accomplishments without their love, understanding and encouragement.

TABLE OF CONTENTS

Page

ACKNOWLEDGEMENTS iv

LIST OF TABLES viii

LIST OF FIGURES x

CHAPTER

1 LITERATURE REVIEW 1

Fomesafen introduction, weed control and application in agronomic crops 1

Fomesafen soil behavior and dissipation in soil 5

Miscanthus × *giganteus*: a promising bioenergy crop in the US 9

Invasive potential and eradication of *M. giganteus* 13

Literature Cited 17

2 ADSORPTION, DESORPTION AND DEGRADATION OF FOMESAFEN IN SOIL 27

Abstract 28

Introduction 30

Materials and Methods 33

Results and Discussion 35

Literature Cited 40

3 FOMESAFEN SOIL DISSIPATION AND COTTON RESPONSE 50

Abstract 51

Introduction.....53
Materials and Methods.....57
Results and Discussion.....60
Literature Cited.....65
4 TOLERANCE EVALUATION OF VEGETATIVELY-ESTABLISHED MISCANTHUS × GIGANTEUS TO NUMEROUS HERBICIDES.....78
Abstract.....79
Introduction.....81
Materials and Methods.....83
Results and Discussion.....85
Literature Cited.....90
5 PREEMERGENCE HERBICIDE SCREENING AND TOLERANCE EVALUATION OF SEEDED-TYPE MISCANTHUS × GIGANTEUS.....97
Abstract.....98
Introduction.....100
Materials and Methods.....102
Results and Discussion.....104
Literature Cited.....110
6 GROWTH AND PHYSIOLOGICAL RESPONSES OF MISCANTHUS × GIGANTEUS TO POST HERBICIDES.....118
Abstract.....119
Introduction.....121

Materials and Methods.....123

Results and Discussion 126

Literature Cited.....131

CONCLUSIONS.....146

REFERENCES 151

LIST OF TABLES

Page

Table 1: Soil information of adsorption and desorption study 43

Table 2: Sorption coefficient estimates for fomesafen 44

Table 3: Percentage of fomesafen desorbed from soil.....45

Table 4: Correlations of soil parameters to K_f and desorption.....46

Table 5: Locations, planting and harvesting dates and soil information of field cotton trials.....69

Table 6: Parameter estimates of greenhouse cotton height.....70

Table 7: Parameter estimates of greenhouse cotton dry weight 71

Table 8: Field cotton stand count and height as affected by fomesafen 72

Table 9: Cotton yield as affected by fomesafen.....73

Table 10: Parameter estimates of fomesafen persistence in soils under field conditions.....74

Table 11: Responses of *M. giganteus* to 21 different PPI and PRE herbicides or herbicide combinations applied at two rates.....93

Table 12: *M. giganteus* shoot dry weight, height and injury affected by 27 POST herbicides 95

Table 13: *M. giganteus* seed germination and shoot emergence as affected by preemergence herbicides in petri dish assay 113

Table 14: Parameters of *M. giganteus* height and GR_{50} for the PRE herbicides used in the greenhouse bioassay.....114

Table 15: Parameters of *M. giganteus* shoot biomass and GR_{50} for the PRE herbicides used in the greenhouse bioassay.....115

Table 16: Parameter estimates of *M. giganteus* shoot dry weight, underground biomass and regenerated shoot dry weight as affected by various rates of glyphosate.....136

Table 17: Parameter estimates of *M. giganteus* PSII efficiency (Fv/Fm) 137

.....

Table 18: Parameter estimates of *M. giganteus* chlorophyll content.....138

Table 19: *M. giganteus* response to a single application of POST treatments 139

Table 20: *M. giganteus* response to a two applications of POST treatments 140

LIST OF FIGURES

Page

Figure 1: Fomesafen adsorption to Cecil sandy loam over 24 hr period 47

Figure 2: Fomesafen adsorption isotherms on 7 soils 48

Figure 3: Fomesafen degradation in Cecil sandy loam and Tifton loamy sand under laboratory environment 49

Figure 4: Effect of fomesafen rates and soil types on greenhouse cotton height and dry weight 75

Figure 5: Fomesafen persistence in Cecil sandy loam and Tifton loamy sand as grouped by rate 76

Figure 6: Fomesafen persistence in Cecil sandy loam and Tifton loamy sand as grouped by soil 77

Figure 7: *M. giganteus* height affected by various rates of metolachlor, acetochlor, mesotrione and atrazine in greenhouse 116

Figure 8: *M. giganteus* shoot biomass affected by various rates of metolachlor, acetochlor, mesotrione and atrazine in greenhouse 117

Figure 9: Response of *M. giganteus* shoot dry weight and underground biomass to various doses of glyphosate 141

Figure 10: Response of *M. giganteus* regrowth shoot dry weight to various doses of glyphosate 142

Figure 11: Response of *M. giganteus* chlorophyll content and PSII efficiency (Fv/Fm) to two rates of glyphosate 143

Figure 12: *M. giganteus* PSII efficiency (Fv/Fm) as affected by four POST combination treatments 144

Figure 13: *M. giganteus* chlorophyll content as affected by four POST combination treatments 145

CHAPTER 1

LITERATURE REVIEW

Fomesafen introduction, weed control and application in agronomic crops. Fomesafen, 5-(2-chloro-α, α, α-trifluoro-p-tolyloxy)-*N*-mesyl-2-nitrobenzamide, is a protoporphyrinogen oxidase (PPO) inhibitor used for weed control in cotton (*Gossypium hirsutum* L), soybeans (*Glycine max* L. Merr.), snap bean (*Phaseolus vulgaris* L,) pepper (*Capsicum* spp.), tomato (*Solanum lycopersicum* L.) and potato (*Solanum tuberosum* L.) (Campbell et al. 2012; Syngenta 2014). Fomesafen is an active ingredient in 27 products, five of which are registered by Syngenta Crop Protection (Flexstar, Flexstar GT, Flexstart GT 3.5, Prefix, Reflex) (Campbell et al. 2012). It is a weak acid (pK_a =2.7) with solubility of 50 mg L^{-1} at pH 7, and its solubility and bioavailability in soil are affected by pH (solubility <1 mg L^{-1} at pH 1) (Weber 1993). Fomesafen is rapidly absorbed by leaf tissue within 1 hr from a post-emergence (POST) application, and is primarily xylem mobile. Fomesafen injured plants typically produce symptoms such as chlorosis, necrosis, and leaf desiccation within 3 d; sub-lethal doses cause foliar bronzing on young leaves. Due to the rapid expansion of herbicide resistant weeds in the Southeast, especially glyphosate and acetolactate synthase (ALS) inhibitor resistant Palmer amaranth, fomesafen pre-emergence (PRE) applied has become an indispensable component of weed control in cotton and soybean (Culpepper 2009; Sosnoskie et al. 2009; Wise et al. 2009).

Fomesafen can provide control of many weed species in agronomic crops, including pigweed spp., (*Amaranthus* spp.), morningglory species (*Ipomoea* spp.), jimsonweed (*Datura stramonium* L.), wild mustard (*Sinapis arvensis* L. ssp. *Arvensis*), black nightshade (*Solanum nigrum* L.) and

ragweed (*Ambrosia* spp.) (Senseman, 2007). Culpepper (2009) reported in glufosinate-resistant cotton, fomesafen plus pendimethalin at 280 and 506 g ai ha^{-1} applied PRE, followed by glufosinate POST and diuron plus MSMA POST-directed (PD) provided the best late season Palmer amaranth control (95%) and greatest seed cotton yield (1341 kg ha^{-1}) over other herbicide systems evaluated. Fomesafen plus pendimethalin at 280 and 602 g ai ha^{-1} PRE, followed by glyphosate plus pyrithiobac POST and diuron plus MSMA applied PD provided 88% late season control of Palmer amaranth and 1300 kg ha^{-1} seed cotton yield in glyphosate-resistant cotton (Culpepper 2009). In another study, fomesafen applied pre-plant incorporated (PPI) at 280 g ai ha^{-1} was the least effective treatment to control Palmer amaranth (69%); however, increasing rate to 420 g ai ha^{-1} improved control to 81%; PRE treatments at 280 and 420 g ai ha^{-1} provided 72 and 81% control of Palmer amaranth, respectively. Fomesafen split applications provided the most effective control (> 91%) of Palmer amaranth among all treatments examined (Kichler and Culpepper 2012). Treatments containing fomesafen improved early-season common cocklebur (*Xanthium strumarium* L.) and *Ipomoea* spp. control when properly activated by irrigation or precipitation (Stephenson et al 2004). Another study suggested fomesafen plus pendimethalin (280 and 1120 g ai ha^{-1}, respectively) applied PRE followed by glufosinate mid-POST provided over 90% control of Palmer amaranth, common lambsquarter (*Chenopodium album* L.), common ragweed (*Ambrosia artemisiifolia* L.), large crabgrass (*Digitaria sanguinalis* L.) and goosegrass (*Eleusine indica* Elein) (Everman et al. 2009). Fomesafen tank mixed with flumeturon at 0.42 and 1.68 kg ai ha^{-1} rates provided higher weed control than either herbicide used alone; fomesafen applied 0.42 kg ai ha^{-1} in a tank mixed with MSMA at 2.24 kg ai ha^{-1} provided excellent control of yellow nutsedge (*Cyperus esculentus* L.), morningglory species and pigweed species when PD to cotton (Lunsford et al. 1998). However, fomesafen did not control purple

nutsedge (*Cyperus rotundus* L.) and sicklepod (*Senna obtusifolia* L.) (Murdock and Keeton, 1998).

Previous research indicates fomesafen half-life (DT_{50}) varies significantly under different environmental and soil conditions. Fomesafen field DT_{50} was reported from 28 to 66 d with an average of 50 d, after 0.18 kg ai ha^{-1} alone or 0.09 followed by 0.18 kg ai ha^{-1} applications in July and August in NY. Fomesafen residue was still detectable 350 d after treatment in this Madalin silty clay loam (Rauch et al. 2007). Fomesafen dissipation under anaerobic conditions was less than 3 wk but soil persistence varied significantly under aerobic field conditions with DT_{50} varying from 6 to 12 months (Senseman, 2007). Due to residual persistence, fomesafen may injure susceptible rotational crops, such as sugarbeets (*Beta vulgaris* L.), sunflowers (*Helianthus annuus* L.) and sorghum (*Sorghum bicolor* L.) up to one year after application (Senseman, 2007). The minimal rotational interval for small grains such as wheat (*Triticum aestivum* L.), barley (*Hordeum vulgare* L.) and rye (*Secale cereale* L.), is 4 months and for corn (*Zea mays* L.), peanut (*Arachis hypogaea* L.), peas (*Pisum sativum* L.) and rice (*Oryza sativa* L.) is 10 months (Syngenta 2014). In one study, all the bioassay crops, snap bean, sunflower, watermelon (*Citrullus Schrad*.), cucumber (*Cucumis sativus* L.) and mustard [*Brassica juncea* (L.) Czern], exhibited various level of injury (11-99%) when planted 2 wk after a 0.28 kg ai ha^{-1} fomesafen application. Injury on snap bean, sunflower, cucumber and mustard was 35, 42, 11 and 100% when planted back 11 wk after treatment (Johnson and Talbert, 1993). Dotray et al. (2010) reported fomesafen injury on peanut when applied PRE, AC (at cracking) and EPOST at two TX locations as unacceptable (> 46%) even though it generated good to excellent control of some broadleaf weeds. In this experiment, fomesafen applied PRE at 280 and 560 g ai ha^{-1} applications caused up to 46 and 59% peanut injury, respectively. Late season injury was

apparent and yield reduction was observed in most of the treatments (Dotray et al. 2010). Gilbert et al. (2009) reported fomesafen applied from 220 to 560 g ai ha^{-1} caused significant peanut injury and yield reduction was common regardless of application timing (PRE, AC and EPOST) in all four trial locations (Lamesa TX, Tifton GA, Citra FL and Lewiston-Woodville NC).

Fomesafen is mainly used in cotton PPI or PRE for weed control. The fomesafen cotton registration allows preplant surface application to medium or fine-textured soils for rates of 280 ai $ha^{-1}$and PRE application to coarse-textured soils for rates of 280 to 420 g ai ha^{-1} (Syngenta, 2014). The use of fomesafen in cotton production has rapidly increased over the past decade. Total usage of fomesafen in all cotton producing states increased by 4.9-fold from 2007 to 2010. A total of 66,636 ha in 2007 was treated with fomesafen in GA and this number increased to 220,742 ha in 2010. (USDA-NASS, 2010). However, cotton are concerned for fomesafen injury on cotton. Kichler and Culpepper (2012) reported greatest fomesafen cotton injury was observed 12 DAT. PRE treatment caused 8 and 15% injury when applied at 280 and 420 g ai ha^{-1}, while PPI treatments only produced 0 and 2% injury for the respective rates. Murdock and Keeton (1998) reported fomesafen cotton injury was generally greater when applied PRE than PPI; average injury was 5, 9, 14 and 23% respectively when fomesafen was applied PRE at 280, 426, 560 or 840 g ai ha^{-1}. When applied PPI, average injury was 1, 4, 5 and 15% for those respective rates. Schrage et al. (2012) concluded low seed vigor incurred 20% greater fomesafen injury on cotton and deep planting at 2.5 cm caused 15% more injury than at 0.6 cm.

Similar to fomesafen, other PPO inhibitors have been reported to cause injury to cotton and soybean. Flumioxazin was very effective against herbicide resistant Palmer amaranth but due to crop injury concerns, it can only be applied at least 14 d before cotton planting. If planted within 14 d of flumioxazin application, a strip-tillage is needed to safen cotton from flumioxazin injury,

but this may decrease weed control by 25 to 40% (Culpepper 2009; Kichler et al. 2007). Flumioxazin injury on cotton leaves occurred when heavy rainfall splashed treated soil onto leaf surface of 15 cm tall cotton (Wilcut et al. 2000). Therefore, flumioxazin application should be restricted to the cotton bark when PD applied and misapplication over the top or to small cotton with green stems can cause serious injury (Wilcut et al. 2000; Cranmer et al. 2000). Similar restriction has been specified in fomesafen label, which forbid POST application over cotton foliage. PD fomesafen applications in cotton need to be made with precision, hooded or shielded application equipment (Syngenta 2014). In soybean, sulfentrazone at 0.22 and 0.44 kg ai ha^{-1} caused greater soybean injury, reduced stand and yield in a soil with 1.1% organic content (OC) as compared to soils with 2.3% and 2.9% OC when applied 7 d before planting, at planting and at 50% hypocotyl emergence (Reiling et al. 2006). It has been noted that early season injury on cotton may delay plant development, fruiting and maturity, so plants were greener at harvest, causing more trash in the lint and lower quality (Hayes et al. 1981). Overall, little work has been done to systematically examine soil types and soil properties on fomesafen injury. Considering limited information is available regarding the impact of fomesafen injury on cotton yield, further research is needed to evaluate cotton tolerance to fomesafen and provide recommendations for growers.

Fomesafen soil behavior and dissipation in soil. Soil properties, adsorption, desorption, mobility and biological degradation are important factors that determine pesticide persistence and bioavailability. Fomesafen is a weak acid with pK_a of 2.7 (Senseman, 2007), therefore, increased sorption of fomesafen at low pH or acidic soil surfaces may reduce the water solubility, mobility and bioavailability of this herbicide in soil, due to the formation of

hydrophobic bonding between fomesafen molecule and lipophilic sites on the organic colloidal surfaces (Weber 1993a; Tanford 1973). Weber (1993b) suggested for weak acids like fomesafen, adsorption occurred by physical force near neutral pH and hydrophobic bonding or precipitation at low pH.

Freundlich isotherms is frequently used to describe pesticide adsorption and desorption processes (Stougaard et al. 1990; Weber 1993a; Grey et al. 1997)

$$Cs = K_f Ce^{1/n} \quad (1)$$

where *C*s (µmol kg^{-1}) is the amount of pesticide adsorbed at the equilibrium concentration *C*e (µmol L^{-1}); K_f and 1/*n* are constants that characterize the relative sorption capacity and the sorption intensity, respectively. K_f is the mathematical description of distribution of the pesticide between the solid and liquid phases, which represents the amount of adsorbed pesticide on the sorbent when equilibrium concentration is 1 µmol L^{-1}. K_{OC} (soil organic carbon adsorption coefficient) is usually calculated as:

$$K_{OC} = (K_f \div \text{OC \%}) \times 100 \text{ (OC= organic carbon)} \quad (2)$$

In one study, decreasing soil pH from 6.05 to 3.46 increased imazapyr (pK_a= 3.8) adsorption (K_f) to two soils by 10.9 and 2.6 fold respectively (Pusino et al. 1997), possibly due to the formation of hydrophobic bonding. Moreover, at agronomic soil pH ranges (5 to 8), adsorption to Fe and Al oxides could occur to many weak acids because they mainly appear in their anionic forms (Newby and White 1981; Pusino et al. 1997). No published literature is available regarding the effect of soil Fe and Al oxides on fomesafen adsorption. But in one study investigating adsorption and desorption of imazapyr, which is also a weak acid, correlation results indicated that imazapyr adsorption and desorption in soil were highly related to iron oxide content, CEC, and soil organic matter (OM). The adsorption coefficient (K_d) of imazapyr to iron

oxide was 32.7 at pH 4.8, which was higher than humic acid and Ca^{2+} saturated humate, but it drastically decreased to 1.7 at pH 7.1. These results may imply the complicity of weak acid herbicide adsorption and desorption since these processes could be affected by multiple soil components simultaneously.

Guo et al. (2003) reported Freundlich isotherms provided good description of fomesafen adsorption to soil. K_f varied from 1.38 to 3.02 on six Chinese soils. OM and pH were significantly correlated to fomesafen adsorption but soil pH was more important than organic matter content. Weber (1993a) investigated ionization and sorption of fomesafen by soil and soil constituents at suspension pH of 2 to 6.3. The sorbent used in this study included H^+ and Ca^{2+}-saturated soil organic matter, Ca^{2+}-saturated montmorillonite clay, Norfolk sandy loam and Drummer silt loam. Results suggested decreasing suspension pH to 2 increased fomesafen adsorption to all sorbents by 5.3 to 42.1 fold. K_f of Drummer silt loam and Norfolk sandy loam was 3.6 and 3.5, respectively but K_{oc} for the respective soils was 86 and 700, which varied 8.1 fold. This indicated there were soil constituents other than OM involved in fomesafen adsorption. Usually, a hydrophobic molecule should have relatively constant K_{oc} over different soil types since this molecule could partition from aqueous phase into soil and form strong hydrophobic bonds with OM in soil (Morillo et al. 2004). However, this may not be the case for fomesafen since it is an ionizable molecule with an affinity to OM which can also be affected by soil pH.

Fomesafen has been suggested to have moderate leaching potential (Newby and White 1981). In a field study, 60% of applied fomesafen was found 0 to 10 cm deep 63 d after a 0.3 kg ai ha^{-1} application with 660 mm of precipitation (Weissler and Poole 1982). Guo et al. (2003) concluded that fomesafen did not move in three of the five soils tested in the soil thin-layer

chromatography study; 89.92% of the applied ^{14}C-fomesafen remained in the top 5 cm when investigated with one soil under field conditions. Weber (1993b) reported different leaching potential of fomesafen in four soils. Fomesafen exhibited higher mobility in sandier Norfolk sandy loam than three other soils when irrigated 1.25 cm d^{-1} for 40 d or 50 cm water continuously. Liming the Norfolk sandy loam also resulted in greater fomesafen mobility. Correlation results indicated fomesafen mobility in soil was negatively related to CEC, OM, humic matter and pH but was not affected by clay content. The results of these studies suggested, fomesafen mobility might vary dramatically between soils since this process could be affected by multiple soil components and properties. Similarly, previous research on imazapyr (pKa = 3.8) suggested that imazapyr desorption from soils were highly related to OM, soil pH, CEC and Fe oxide.

Fomesafen persistence in soil varies significantly and half-life (DT_{50}) ranges from 6 to 12 month under aerobic conditions. However, fomesafen degradation under anaerobic conditions was less than 3 wk (Senseman, 2007). Rauch et al. (2007) reported fomesafen applied at 0.18 kg ai ha^{-1} or 0.09 followed by 0.18 kg ai ha^{-1} had field DT_{50} varied between 28 to 66 d, with an average of 50 d in a Madalin silty clay loam. Cobucci et al. (1997) reported fomesafen was detected in 0-5 cm, 5-10 cm and 10-20 cm of a Brazilian soil (61% clay, 26% sand, OM 3.92%, pH 6.1) 232 d after 0.25 and 0.5 kg ai ha^{-1} application and most of the fomesafen concentrated in the 0-10 cm. Oymada and Kuwatsuka (1988) investigated the persistence of three diphenylether herbicides in soil and found the DT_{50} varied greatly by soils and environmental conditions. The DT_{50} was 9 to 173 d for chlornitrofen, 3 to 87 d for nitrofen and 8 to 64 d for chlomethoxynil. Similar to fomesafen, these herbicides dissipated rapidly under anaerobic and low redox potential conditions. It has been noted in this research that DT_{50} was negatively related to soil

redox potentials and soil microorganisms may have been involved in the dissipation process since adding organic matter expedited fomesafen degradation, but no direct data supported this assumption. Since fomesafen is now widely used in cotton on a wide range of soils, it is imperative to know how soil properties affect fomesafen behavior, considering limited published data regarding its persistence and degradation in soil.

***Miscanthus* × *giganteus*: a promising bioenergy crop in the US**. *M. giganteus* has been grown in Europe as a cellulosic bioenergy crop for several decades and is currently under field evaluation at multiple locations in the US. The genus *Miscanthus* consists of 17 species and originated from East Asia (Greef and Deuter 1993). The specific genotype used in Europe and US for bioenergy production, *M. giganteus* was introduced to Denmark from Japan in the 1930's (Greef and Deuter 1993; Lewandowski et al. 2000). *M. giganteus* is a natural hybrid between *Miscanthus sinensis* and *Miscanthus sacchariflorus* with 57 somatic chromosomes. Due to triploidy, *M. giganteus* seeds are sterile and therefore, reproduction in natural habitat solely relies on vegetative propagation (Lewandowski et al. 2000; Linde-Laursen 1993). Previous tests have shown that *M. giganteus* biomass can be used as solid fuel, construction materials such as pressed particle-board, and as a source of cellulose. Key disadvantages include relatively high establishment costs, narrow genetic base and low cold tolerance in the first winter following establishment (Lewandowski et al. 2000).

M. giganteus has potential as a bioenergy crop because of its significant biomass production advantage compared to maize (*Zea mays* L.) for ethanol production and other bioenergy species, such as switchgrass (*Panicum virgatum* L.) (Heaton et al. 2008). Field trials have shown that at many locations in Europe, *M. giganteus* has yielded the greatest energy of all potential bioenergy

crops in terms of net MJ ha^{-1}. It also has the highest energy-use efficiency (EUE), in terms of the energy cost of production, due to relatively high yields and low inputs (Heaton et al. 2004). In Europe, experiments conducted from Denmark and Germany suggested yields without irrigation typically ranged from 10-25 t dry matter (DM) ha^{-1} (Lewandowski et al. 2000); irrigated trials generally produced yields in excess of 30 t ha^{-1}. Research data suggested *M. giganteus* produced an average yield of 30 t DM ha^{-1} and maximum yield of 61 t DM ha^{-1} in Illinois trials over 3 years and in the same study, regionally adapted switchgrass variety 'Cave-in-Rock' generated lower yields (10 t ha^{-1}**) (**Heaton et al. 2008). Another review paper analyzed the published yield data of Miscanthus and switchgrass from peer-reviewed articles (97 observations for Miscanthus, 77 for switchgrass) and the authors suggested Miscanthus can potentially produce an annual biomass of 22 t ha^{-1} compared to 10 t ha^{-1} of switchgrass (Heaton et al. 2004). In contrast to maize grain, *M. giganteus* also has an advantage in ethanol production cost since it requires lower management (i.e. tillage, nitrogen fertilizer, pesticide) and financial input (Lewandowski et al. 2000). The energy balance ratios (output energy/input energy) of maize and *M. giganteus* were 1.4-3.8 and 12-66, respectively (Venturi and Venturi, 2003). The net energy balance of ethanol (NEB) obtained from maize grain ranged from 10-80 GJ ha^{-1} yr^{-1} while NEB range of ethanol derived from *M. giganteus* cellulose biomass was 250-550 GJ ha^{-1} yr^{-1} (Yuan et al. 2008)**.**

M. giganteus is a C_4 grass with high water use efficacies and high biomass yield. Plants with C_4 photosynthesis may out yield C_3 plants because of higher radiation, water and nitrogen use efficacies, but they require a warmer climate to initiate growth in spring (Long 1983). Usually, *M. giganteus* rhizomes begin growth when soil temp reaches 10 to 12 C (Clifton-Brown 1997). The water use efficiency of pot and field-established *M. giganteus* ranged from 250 to 340 g g^{-1}

and 80 to 330 g g^{-1} respectively (mass of water per unit dry matter accumulated) (Lewandowski et al. 2000). Although water use efficiency is higher than most of C_3 plants, growth is often water limited (Beale and Long 1997). *M. giganteus* does not respond well to nitrogen fertilization, however, supplemental nitrogen may be necessary in areas where it is limiting (Lewandowski et al. 2000). There have been no reports of plant disease and insects which significantly reduced the yield of *M. giganteus* (Lewandowski et al. 2000).

Although an excellent bioenergy crop candidate, there are two major challenges that limit *M. giganteus* production, low tolerance to cold and high establishment cost (Lewandowski et al. 2000; Lewandowski 1998). *M. giganteus* rhizomes are killed when soil temperatures go below -3.5 C while its parent, *M. sinensis* rhizomes can tolerate cold stress to -6.5 C (Clifton-Brown and Lewandowski 2000). Therefore, in areas where soil temperatures fall below -3.5 C, more cold-tolerant genotypes or *M. sinensis* are recommended (Clifton-Brown et al. 2001). One study suggested rhizome size, planting depth, rhizome storage length and storage conditions have significant effect on the survival of *M. giganteus* within the first year of establishment, (Pyter et al. 2010). Clifton-Brown et al. (2011) studied the base temperatures below which the germination of at least 50% viable seeds ceased and reported that the base temperature for perennial ryegrass (*Lolium perenne* L.) and maize were 3.4 and 4.5 C, respectively. However, the base temperature of *Miscanthus* genotypes varied from 9.7 to 11.6 C, which was higher than maize and switchgrass (*Panicum virgatum* L.).

High establishment cost is another major obstacle in *M. giganteus* production. Because of seed sterility, *M. giganteus* stands are typically established with vegetative propagated rhizomes which are more expensive to produce and store, and difficult to plant as compared to seed. Also, some special planting equipment is needed to plant rhizomes and may not be available to

growers, therefore, the adoption of sterile *M. giganteus* has been slow (Heaton et al. 2010). Lewandowski et al. (2000) estimated stand establishment could cost $3906 to $7811 ha^{-1} with rhizomes and requires special planting equipment. However, cell culture techniques and micro-propagated plants from somatic cells or meristems may significantly reduce *M. giganteus* establishment cost to $456 ha^{-1}. Jones (2009) suggested propagation through either tissue culture or rhizomes could cost $2586 ha^{-1} and is largely supported by EU grants of some countries. Planting cost using *M. giganteus* fertile seeds would be $608 ha^{-1} (Clifton-Brown et al. 2011), which could significantly reduce establishment expenses as compared to rhizome propagation. Fertile varieties of *M. giganteus* are currently under development and may be commercially available in a near future (Smith and Barney 2014; Ross 2011) but these varieties have raised concerns over their invasive potential (Matlaga and Davis 2013; Quinn et al. 2011; Smith and Barney 2014).

Due to slow initial growth of *M. giganteus*, weed control in the first year is crucial to successful establishment and high biomass yield (Lewandowski et 2000; Anderson et al. 2011). Up to date, limited information is published concerning herbicide options available in *M. giganteus*. Some researchers suggested herbicides registered for corn (*Zea mays* L.) are generally safe on *M. giganteus* (Lewandowski et al. 2000); however, several exceptions have been identified. Corn herbicides EPTC applied at 4478 g ai ha^{-1}, nicosulfuron applied at 35 g ai ha^{-1} and trifloxysulfuron applied at 16 g ai ha^{-1} reduced *M. giganteus* shoot height and dry weight (Li et al. 2013). In another study, foramsulfuron applied at 37 g ai ha^{-1}, glyphosate applied at 840 g ai ha^{-1}, imazamox applied at 44 g ai ha^{-1} and nicosulfuron applied at 35 g ai ha^{-1} produced lower *M. giganteus* aboveground and belowground biomass than non-treated check (NTC) (Everman et al. 2011). Anderson et al. (2010) evaluated *M. giganteus* tolerance to 11 PRE and

16 POST treatments in greenhouse and 24 treatments in field. They reported 8980 g ai ha^{-1} atrazine, 284 g ai ha^{-1} imazethapyr, 316 g ai ha^{-1} isoxaflutole, 6400 g ai ha^{-1} pendimethalin, 420 g ai ha^{-1} isoxaflutole caused injury and reduced shoot dry weights in greenhouse trial. However, these herbicides, if applied at lower rates, did not produce any negative effect on *M. giganteus* growth. For POST treatments, clethodim, imazethapyr, imazapic, sethoxydim, tembotrione and topramezone produced various level of injury (17-58%) and dry weight reductions on greenhouse plants as compared to NTC. In field trials, treatments containing imazamox 44 to 176 g ai ha^{-1}generally decreased shoot dry weights.

Invasive potential and eradication of *M. giganteus*. The invasive potential of sterile and fertile *M. giganteus* has been evaluated in previous publications (Matlaga and Davis 2013; Quinn et al. 2011; Smith and Barney 2014). Sterile *M. giganteus* has been reported to possess less invasive potential than fertile varieties (Smith and Barney 2014). Sterile *M. giganteus* received a low score in the widely accepted Australian weed risk assessment (WRA) protocol and was considered 'minor risk' for invading natural areas in the US. Other bioenergy species received 'evaluated further' and 'reject' score in this evaluation, except for sterile genotypes of switchgrass in California (Barney and DiTomaso, 2008). Gordon et al. (2011) evaluated the invasive potential of 12 bioenergy species proposed in Florida and the US and sterile *M. giganteus* was given the lowest invasive score (-8 and -9, respectively for FL and the US) among all 12 species using WRA and was considered acceptable in FL and the US. Matlaga and Davis (2013) suggested the growth rate of sterile *M. giganteus* was slightly smaller than 1 (value less than 1 means growth of the population can not compensate the portion lost to senescence, physical and environmental damages, etc.), indicating the population is not self-sustainable and

would gradually decline over time without clonal recruitment. They also concluded that a sterile *M. giganteus* population may increases in number and space only if annual rhizome sprouting is greater than 20% and rhizome production is equal or greater than 1 per plant. Although no single case of escape has been reported in Europe for sterile *M. giganteus* after nearly three decades of research and production (Lewandowski et al. 2000), it has been suggested that sterile *M. giganteus* should be grown away from riparian areas, riverbanks and areas experience frequent soil disturbance (Matlaga and Davis 2013).

Compared to sterile variety, the new fertile varieties of *M. giganteus* can largely decrease planting cost, but also raised concerns over their invasiveness since these fertile varieties could produce large number of viable seeds in field (Smith and Barney 2014). It has been reported that a single *M. giganteus* plant can produce over 100 inflorescences after second year of growth, with each inflorescence can generate an average of 1,270 spikelets. These could total over 2.5 billion spikelet per ha per yr (Smith and Barney 2014). *Miscanthus* fruits (caryopses) are low in weight (0.8 to 1 mg per seed) and known to be dispersed by wind in native grassland because of the silky hairs on the caryopses (Ohtsuka et al. 1993; Quinn et al. 2011). Previous experiments indicated most Miscanthus caryopses (95% for *M. sinensis* and 77% for *M. giganteus*) were captured within 50 m of source, but a small portion (0.2% -3%) was found at 300 m and 400 m (Quinn et al. 2011). Caryopses could travel even further in high wind speeds and these fertile seeds will be nearly impossible to contain (Matlaga and Davis 2013; Quinn et al. 2011). Smith and Barney (2014) compared the invasive potential of a fertile variety of *M. giganteus* to five invasive and three noninvasive species at seven habitats in VA and GA. Their results suggested overall seed germination rate was low for all the species evaluated in all geographies and habitats. Final seedling mortality rate for fertile *M. giganteus* was 99.9% (one in 16,000 spikelet

survived and was 4 cm tall at the end of 6 mo study). Similar to *M. giganteus*, *M. sinensis* seeds yielded only 3% survival rate 12 wk after sowing (Christian et al. 2005). Although survival and germination rate were low, the invasive potential of fertile *M. giganteus* still warrant further investigation due to the massive amount of seeds fertile varieties can produce during one season (2.5 billion spikelet ha^{-1}) (Smith and Barney, 2014). Model estimates suggested sterile and fertile *M. giganteus* possess remarkably different invasive potential (Matlaga and Davis 2013). For the fertile varieties, rapid population expansion is possible even if the seed viability and survival rate is low (Matlaga and Davis 2013). Furthermore, some ideal traits of bioenergy crops (C_4 photosynthesis pathway, high water, nitrogen use efficiency and biomass accumulation ability, no or few pests and diseases, etc.) make them perfect invasive weeds (Raghu et al. 2006).

One parent of *M. giganteus*, *M. sinensis*, have long history of escaping cultivation in Eastern United States, particularly within the Appalachian region (Quinn et al. 2010). *M. sinensis* is a C_4 perennial grass native to eastern Asia and pacific islands. It was introduced to the US from Japan in 19^{th} century (Dougherty et al. 2014). It has become the most popular and recommended ornamental grass in the US (Maynard 2012) and sales of *M. sinensis* in NC amounts to nearly $40 million (Trueblood 2009). There have been new interests of developing *M. sinensis* as a bioenergy crop and breeding germplasm for novel lines of *M. giganteus* (Stewart et al. 2009). However, *M. sinensis* can produce viable seeds (Meyer and Tchida 1999) and can tolerate a number of stressful conditions, such as low fertility, cold temperatures, heavy metal contamination, low pH, shade and frequent burning (Stewart et al. 2009; Meyer 2003; Horton et al. 2010). It is considered to be more drought-tolerant than *M. giganteus* (Clifton-Brown et al. 2002). Dougherty et al. (2014) surveyed 18 naturalized *M. sinensis* population from NC to MA and they concluded that *M. sinensis* strongly favor highly disturbed and unmanaged habitats such

as roadsides and forest edges. Soil types and nutrient availability did not affected population size and plant morphology (tiller height, number and basal diameter) while low light availability did not have impact on plant size and vigor. These researchers suggested *M. sinensis* can tolerate broad range of climatic conditions and environments in the eastern US and the invasion beyond its current distribution is possible. Some researchers recommended that sterile varieties of *M. sinensis* should be developed due to its invasive potential (Quinn et al. 2010).

Eradication of Miscanthus with herbicides and tillage has been evaluated in previous experiments. Currently, control options heavily rely on glyphosate because of its efficacy against perennial grasses and mobility to underground rhizomes (Everman et al 2011; Anderson et al. 2011a, 2011b; Omielan et al. 2012; Cutts et al. 2011; Spencer et al. 2008, 2011). Everman et al. (2011) reported glyphosate applied at 0.84 kg ae ha^{-1} produced the lowest aboveground and underground biomass among 17 POST herbicides examined. Foramsulfuron applied at 0.037 kg ai ha^{-1}, imazamox at 0.044 kg ai ha^{-1}, nicosulfuron at 0.035 kg ai ha^{-1} also resulted decreased aboverground and underground biomass. Glyphosate, foramsulfuron and nicosulfuron produced the most injury among all treatments examined (54, 32 and 28% respectively). *M. sinensis* could be effectively controlled (> 90% control at 397 DAT) by glyphosate alone at 1.26 kg ae ha^{-1} and in combination with imazapyr 560 g ai ha^{-1} (Omielan et al. 2012). Anderson et al. (2011a) reported one application of 1.7 kg ae ha^{-1} glyphosate applied at either fall or spring did not reduce dry weight and summer shoot number of field established *M. giganteus*, both fall and spring applications were needed to decrease dry weight and shoot number. Tillage was effective to decrease shoot dry weight and number. Spring tillage with one or two application of 2.5 kg ae ha^{-1} glyphosate reduced aboveground biomass by 94 and 95% respectively, and reduced shoot number by 38 and 67% respectively in the same growing season. Although tillage and

glyphosate were effective options to eradicate *M. giganteus*, these researchers believed it would still take more than one year to completely remove established *M. giganteus* from field due to large amount of underground rhizome mat. In another study, rotating mature field *M. giganteus* to glyphosate resistant (GR) corn and soybean was evaluated (Anderson et al. 2011b). *M. giganteus* were harvested 10 cm to the ground in the previous fall, and then field was tilled prior to planting GR corn and soybean. Two applications of glyphosate at 1.26 kg ae ha^{-1} were made in corn during the season. For soybean, first glyphosate application was 1.74 kg ae ha^{-1} and second one was 0.79 kg ae ha^{-1}. Their results showed that two applications of glyphosate plus manual weeding (remove all the weeds except Miscanthus) resulted in highest crop yield, lowest *M. giganteus* shoot number and height among all treatments in both corn and soybean. *M. giganteus* was suppressed but not eradicated in the tested field during the growing season. Therefore, they concluded rotating glyphosate resistant crops after *M. giganteus* is feasible without yield loss but complete removal of *M. giganteus* would require more than one growing season. Considering the difficulty of removing *M. giganteus* and the increasing popularity of this crop for energy production, further study is needed to increase the control efficacy of *M. giganteus* with more effective herbicide options and agronomic practices.

Literature Cited

Anderson EK, Voigt TB, Bollero GA, Hager AG (2010) *Miscanthus* × *giganteus* response to preemergence and postemergence herbicides. Weed Technol 24:453-60

Anderson EK, Voigt TB, Bollero GA, Hager AG (2011a) *Miscanthus giganteus* response to tillage and glyphosate. Weed Technol 25:356-62

Anderson EK, Voigt TB, Bollero GA, Hager AG (2011b) Rotating a field of mature *Miscanthus* × *giganteus* to glyphosate-resistant crops. Agron J 103:1383-88

Barney JN, DiTimaso JM. (2008) Nonnative species and bioenergy: Are we cultivating the next invader? BioSci 58:64-70

Beale CV, Long SP (1997) The effects of nitrogen and irrigation on the productivity of C4 grasses *Miscanthus* × *giganteus* and *Spartina cynosuroides*. Aspects of Applied Biology 49:225-30

Campbell D, Overmyer J, Bang J, Perine J, Brain R (2012) Endangered species assessments conducted under registration review: fomesafen case study. Chapter 9 *in* Pesticide regulation and endangered species act. ACS symposium series, American chemical society: Washingtong , DC, 2012

Christian DG, Yates NE, Riche AB (2005) Establishing *Miscanthus sinensis* from seed using conventional sowing methods. Ind Crop Prod 21:109–11

Clifton-Brown JC (1997) The importance of temperature in controlling leaf growth of Miscanthus in temperate climates. PhD. thesis, Trinity College Dublin

Clifton-Brown JC, Lewandowski I (2000) Winter frost tolerance of juvenile Miscanthus plantations: studies on five genotypes at four European sites. New Phytol 148:287–94

Clifton-Brown JC, Lewandowski I, Andersson B, Basch B, Christian DG, Bonderup-Kjeldsen J, Jørgensen U, Mortensen J, Riche AB, Schwarz KU, Tayebi K, Teixeira F (2001) Performance of 15 Miscanthus Genotypes at Five Sites in Europe. Agronomy J 93:1013–19

Clifton-Brown JC, Lewandowski I, Bangerth F, Jones MB (2002) Comparative responses to

water stress in stay-green, rapidand slow senescing genotypes of the biomass crop, Miscanthus. New Phytologist 154:335–45

Cobucci T, Silva JB, Prates HT (1997) Carryover effect on fomesafen, applied on edible bean, on successional maize. Planta Daninha 5:180-89

Cranmer, JR, Altom JV, Braun JC, Pawlak JA (2000) Valor herbicide: a new herbicide for weed control in cotton, peanuts, soybeans, and sugarcane. Proc South Weed Sci Soc 53:158

Culpepper AS (2009) Impact of EPA's ecological risk assessment for fomesafen. Subject: re-registration of fomesafen (Docket # EPA-HQ-OPP-2006-0239) US-EPA. Washington, DC. 20460

Cutts GS, Webster TM, Grey TL, Vencill WK, Lee RD, Tubbs RS, Anderson WF (2011) Herbicide effect on napiergrass (*Pennisetum purpureum*) control. Weed Sci 59:255-62

Dotray PA, Grichar WJ, Gilbert LV (2010) Peanut tolerance and weed control following fomesafen applied at different rates and timing in Texas. Proc American Peanut Research and Education Soc 42:30-32

Dougherty RF, Quinn LD, Endres AB, Voigt TB, Barney JN (2014) Natural history survey of the ornamental grass Miscanthus sinensis in the introduced range. Invasive Plant Sci Manag 7:113-20

Everman WJ, Clewis SB, York AC, Wilcut JW (2009) Weed control and yield with flumioxazin, fomesafen and S-metolachlor systems for glufosinate-resistant cotton residual weed management. Weed Technol 23:391-97

Everman WJ, Lindsey AJ, Henry GM, Glaspie CF, Phillips K, McKenney C (2011) Response of

Miscanthus × *giganteus* and *Miscanthus sinensis* to postemergence herbicides. Weed Technol 25:398-03

Ferrell JA, Vencill WK, Xia K, Grey TL (2005) Sorption and desorption of flumioxazin to soil, clay minerals and ion-exchange resin. Pest Manag Sci 61:40-46

Gilbert LV, Dotray PA, Prostko EP, Grichar WJ, Ferrell JA, and Jordan DL (2009) Peanut response to fomesafen. Proc American Peanut Research and Education Soc 41:28-30

Gordon DR, Tancig KJ, Onderdonk DA, Gantz CA (2011) Assessing the invasive potential of biofuel species proposed for Florida and the United States using the Australian weed risk assessment. Biomass Bioenerg 35:74-79

Greef, JM, Deuter M (1993) Syntaxonomy of *Miscanthus* × *giganteus* GREEF et DEU. Angewandte. Botanik. 67: 87-90

Guo J, Zhu G, Shi J, Sun J (2003) Adsorption, desorption and mobility of fomesafen in Chinese soils. Water Air and Soil Pollut 148:77-85

Hayes RM, Hoskinson PE, Overton JR and Jeffery LS (1981) Effect of consecutive annual applications of fluometuron on cotton (*Gossypium hirsutum*). Weed Sci 29:120-23

Heaton E, Voigt T, Long SP (2004) A quantitative review comparing the yields for two candidate C4 perennial biomass crops in relation to nitrogen, temperature and water. Biomass Bioenerg 27: 21–20

Heaton EA, Dohleman FG, Long SP (2008) Meeting US biofuel goals with less land: the potential of Miscanthus. GCB bioenergy 14: 2000-14

Hodkinson TR, Renvoise S (2001) Nomenclature of *Miscanthus giganteus* (Poacae). Kew

Bulletin 56:759–60

Horton JL, Fortner R, Goklany M (2010) Photosynthetic characteristics of the C4 invasive exotic grass *Miscanthus sinensis* Anderss on growing along gradients of light intensity in the southeastern USA. Castanea 75:52–66

Jones JVH (2009) Testing the economic viability of energy crop production in competition with alternative land uses. pp. 402-423 *in* Proceedings of the 17th International Farm Management Congress. Bloomington/Normal, IL: International Farm Management Association

Johnson DH, Talbert RE (1993) Imazaquin, chlorimuron, and fomesafen may injure rotational vegetables and sunflower (*Helianthus annuus*). Weed Technol 7:573-77

Kichler JM, Culpepper AS, York AC (2007) Response of glyphosate-resistant Palmer amaranth to thirteen residual herbicides. pp. 1229 *in* Proc of the Beltwide Cotton Conference, New Orleans, LA. Natl. Cotton Counc. Am., Memphis, TN

Kichler J, Culpepper AS (2012) How tillage and application timing of reflex affects palmer amaranth control and cotton injury. pp.1543 *in* Proc of the Beltwide Cotton Conference, Orlando, FL. Natl. Cotton Counc. Am., Memphis, TN

Lewandowski I (1998) Propagation method as an important factor in the growth and development of *Miscanthus* × *giganteus*. Ind Crop Prod 8:229-45

Lewandowski I, Clifton-Brown JC, Scurlock JMO, Huisaman M (2000) Miscanthus: European experience with a novel energy crop. Biomass Bioenerg 19:209-27

Li X, Grey TL, Blanchett BH, Lee RD, Webster TM, Vencill WK (2013) Tolerance evaluation of vegetatively established *Miscanthus* × *giganteus* to Herbicides. Weed Technol 27:735-40

Linde-Laursen IB (1993) Cytogenetic analysis of Miscanthus `Giganteus', an interspecic hybrid. Hereditas. 119:297-00

Long SP (1983) C4 photosynthesis at low temperature. Plant Cell Environ 6:345-63

Lunsford JN, Harrison S, Smith JD (1998) Reflex use in cotton. Proc South Weed Sci Soc 51:12-13

Matlaga DP, Davis AS (2013) Minimizing invasive potential of *Miscanthus* × *giganteus* grown for bioenergy: identifying demographic thresholds for population growth and spread. J Appl Ecol 50:479–87

Maynard B (2012) Ornamental Grasses. http://www.uri.edu/ce/factsheets/sheets/orngrasses.html Accessed September 12, 2012

Meyer MH, Tchida CL (1999) *Miscanthus Anderss* produces viable seed in four USDA hardiness zones. J Environ Hort 17:137–40

Meyer MH (2003) Miscanthus: Ornamental and Invasive Grass: a Sabbatical in the Parks. Project Report. University of Minnesota, St Paul, MN.

Morillo E, Undabeytia T, Cabrera A, Villaverde J, Maqueda C (2004) Effect of soil type on adsorption-desorption, mobility and activity of the herbicide norflurazon. J Agric Food Chem 52:884-90

Murdock EC, Keeton A (1998) Where does fomesafen fit in South Carolina cotton weed management programs? Proc South Weed Sci Soc 51:12

Newby SE, White BG (1981) PP021: leaching on soil thick-layer chromatograms. London, ICI. pp.4 (Report RJ0156B)

Ohtsuka T., Sakura T, Ohsawa M (1993) Early herbaceous succession along a topographical gradient on forest clear-felling sites in mountainous terrain, central Japan. Ecol Res 8:329–40

Omielan J, Gumm D, Witt W (2012) Evaluation of management options for control of Chinese silvergrass (*Miscanthus sinensis* Anders.) Poster, Southeast exotic pest plant council annual meeting. May 8-10, 2012. Auburn AL. Available from: http://www2.ca.uky.edu/pss/weeds/ivm/pdf/Miscanthus%20Poster%20for%20AL.pdf

Oymada M, Kuwatsuka S (1988) Effects of soil properties and conditions on the degradation of three diphenylether herbicides in flooded soils. J Pestic Sci 13:99-105

Pyter RJ, Dohleman FG, Voigt TB (2010) Effects of rhizome size, depth of planting and cold storage on *Miscanthus* × *giganteus* establishment in the Midwestern USA. Biomass Bioenerg 34:1466-70

Pusino A, Petretto S, Gessa C (1997) Adsorption and desorption of Imazapyr by soil. J Agric Food Chem 45:1012-16

Quinn LD, Allen Dj, Stewart JR (2010) Invasive potential of Miscanthus sinensis: implications for bioenergy production in the United States. GCB Bioenergy 2:310-20

Quinn LD, Matlaga DP, Stewart JR, Davis AS (2011) Empirical evidence of long-distance dispersal in Miscanthus sinensis and *Miscanthus* × *giganteus*. Invasive Plant Sci Manage 4:142–50

Raghu S, Anderson RC, Daehler CC, Davis AS, Wiedenmann RN, Simberloff D, Mack RN (2006) Adding biofuels to the invasive species fire? Science 313:1742

Rauch BJ, Bellinder RR, Brainard DC, Lane M, Thies JE (2007) Dissipation of fomesafen in

New York state soil and potential to cause carryover injury to sweet corn. Weed Technol 21:206-12

Reiling, KR, Simmions FW, Reichers DE, Steckel LE (2006) Application timing and soil pH effects on sulfentrazone phytotoxicity to two soybean (*Glycine max* (L) Merr.) cultivars. J Crop Prot 25:230-34

Ross M (2011) New Miscanthus development possible biomass game changer? FarmWeek, May 16, 2011. pp. 9

Schrage BW, Norsworthy JK, Smith KL, Johnson DB, Bagavathiannan MV, Riar DS (2012) Factors contributing to cotton injury from soil-applied residual herbicides. Summary of Arkansas Cotton Research 2012:102-06

Senseman SA, ed (2007) Herbicide Handbook. 9th ed. Lawrence, KS: Weed Science Society of America. pp 207

Sims REH, Hastings A, Schlamadinger B, Taylor G, Smith P (2006) Energy crops: current status and future prospects. GCB Bioenergy 12: 2054–76

Smith LL, Barney JN (2014) The relative risk of invasion: evaluation of *Miscanthus* × *giganteus* seed establishment. Invasive Plant Sci Manage 7:93-106

Stephenson DO, Patterson MG, Faircloth WH and Lunsford JN (2004) Weed management with fomesafen preemergence in glyphosate resistant cotton. Weed Technol 18:680-86

Stewart JR, Toma Y, Fernandez FG, Nishiwaki A, Yamada T, Bollero G (2009) The ecology and agronomy of Miscanthus sinensis, a species important to bioenergy crop development, in its native range in Japan: a review. GCB Bioenergy (1) 126–53.

Sosnoskie LM, Kichler JM, Wallace RD, Culpepper AS (2011) Multiple resistance in Palmer Amaranth to glyphosate and pyrithiobac confirmed in Georgia. Weed Sci 59:321-25

Spencer DF, Tan W, Liow P, Ksander GG, Whitehan LC (2008) Evaluation of glyphosate for managing giant reed (*Arundo donax*). Invasive Plant Sci Manage 1:248-54

Spencer DF, Ksander GG, Tan W, Liow PS, Whitehand LC (2011) Influence of application timing on the impact of glyphosate on giant reed (*Arundo donax* L.). J Aquat Plant Manage 49:106-10

Syngenta Crop Protection (2014) Reflex label. Greensboro, NC. 27419. Available from http://www.cdms.net/LDat/ld6BJ002.pdf

Tanford C (1973) The hydrophobic effect. John Wiley and Sons, Inc,. New York.

Trueblood CE (2009) An Invasive Species Assessment System for the North Carolina Horticultural Industry. M.S. thesis. Raleigh, NC: North Carolina State University. pp 286

Venturi P. and G. Venturi. 2003. Analysis of energy comparison for crops in European agricultural systems. Biomass bioenerg. 25:235-55

Weber JB (1993a) Ionization and sorption of fomesafen and atrazine by soils and soil constituents. Pestic Sci 39:31-38

Weber JB (1993b) Mobility of fomesafen and atrazine in soil columns under saturated and unsaturated flow conditions. Pestic Sci 39:39-46

Weissler MS, Poole, NJ (1982) Mobility of fomesafen and degradation products in soil columns. London, ICI, 1982 pp 3. (Report RJ0241 B)

Wilcut, JW, Askew SD, Price AJ, Scott GH, Cranmer J. (2000) Valor: new weed management

option for cotton. Proc South Weed Sci Soc 53:159–60

Wise AM, Grey TL, Prostko EP, Vencill WK, Webster TM (2009) Establishing the geographic distribution level of acetolactate synthase resistance of Palmer amaranth (*Amaranthus palmeri*) accessions in Georgia. Weed Technol 23:214-20

USDA-NASS (2010) Agricultural chemical use program. Available from: http://www.nass.usda.gov/Surveys/Guide_to_NASS_Surveys/Chemical_Use/index.asp

Yuan JS, Tiller KH, Al-Ahmad H, Stewart NR, Stewart Jr CN (2008) Plants to power: bioenergy to fuel the future. Trends Plant Sci 13:421-29

CHAPTER 2

ADSORPTION, DESORPTION AND DEGRADATION OF FOMESAFEN IN SOIL[1]

[1] Xiao Li, Timothy L. Grey, and William K. Vencill. To be published in Pest Management Science.

Adsorption, desorption and degradation of fomesafen in soil

Xiao Li, Timothy L. Grey, and William K. Vencill [2]

Fomesafen provides excellent control of glyphosate resistant Palmer amaranth in cotton but limited information is available regarding its soil behavior and degradation in southern soils. Therefore, fomesafen adsorption and desorption were evaluated on three GA soils (Cecil sandy loam, Greenville sandy clay loam and Tifton loamy sand) and four soils from KY, CO, ID and TX (Sonora silt loam, Haxtun Sandy Loam, Minidoka silt loam and Tremona sand, respectively). The Freundlich distribution coefficient (K_f) was generally low for all soils (1.30 to 9.28). The desorption study indicated fours soils had a desorption rate varied from 11 to 29%, while Tremona sand, Haxtun Sandy Loam and Tifton loamy sand showed higher desorption rate (26 to 81%). There was a negative correlation between soil pH and K_f, while clay content positively correlated to K_f. Organic matter (OM), clay, and silt content were inversely related to fomesafen desorption, while pH and sand content were positively related to desorption. Soil pH had the largest impact on K_f, and OM showed greatest effect on fomesafen desorption. In fomesafen degradation study, a Cecil sandy loam and Tifton loamy sand treated with fomesafen was

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incubated at 0.28 and 0.56 kg ai ha^{-1} in conical flasks under 20 C and ambient soil moisture for 90 d. No significant reduction in fomesafen concentration was observed in any of the soil by the end of the study. Non-linear regression using exponential decay model indicated the slope parameter (b_1) failed to be significant for both soils. These study results indicated fomesafen soil behavior, mobility and bioavailability could be affected by multiple soil properties such as pH, sand clay and organic content, and fomesafen was not susceptible to biological degradation in soils during the incubation. Therefore, long fomesafen persistence in soil under adverse environmental conditions should be expected.

Nomenclature: Fomesafen; Palmer amaranth, *Amaranthus palmeri* S. Wats. AMAPA; cotton, *Gossypium hirsutum* L.

Key words: Fomesafen, soil behavior, adsorption, desorption, biological degradation.

Introduction

Fomesafen is registered in cotton and soybean (*Glycine Max* L.) for weed control at rates of 280 to 420 g ai ha^{-1} (Syngenta Crop Protection, 2014). Fomesafen is in the diphenylether herbicide family, the mechanism of action is inhibition of protoporphyrinogen oxidase (PPO). Fomesafen can be applied PRE or POST and controls many troublesome broadleaf weeds, including pigweed species, (*Amaranthus* spp.), morningglory species (*Ipomoea* spp.), jimsonweed (*Datura stramonium* L.), wild mustard (*Sinapis arvensis* L. ssp. *Arvensis*), black nightshade (*Solanum nigrum* L.) and ragweed species (*Ambrosia* spp.) (Senseman, 2007). Published research indicates that fomesafen soil persistence varied significantly and half-life (DT_{50}) ranges from 6 to 12 months under aerobic conditions in lab experiments. However, fomesafen degradation under anaerobic conditions was less than 3 wk (Senseman, 2007). Rauch et al. (2007) reported fomesafen field DT_{50} varied between 28 and 66 d, with an average of 50 d in a Madalin silty clay loam from NY. Oymada and Kuwatsuka (1988) investigated the persistence of three diphenylether herbicides in soil and noted the DT_{50} varied greatly by environmental conditions. The DT_{50} ranged from 9 to 173 d for chlornitrofen, 3 to 87 d for nitrofen and 8 to 64 d for chlomethoxynil. Similar to fomesafen, these herbicides dissipated rapidly in anaerobic conditions. Soil microorganisms may have been involved in the dissipation process, since adding organic matter expedited fomesafen degradation.

Soil properties, adsorption, desorption, mobility and biological degradation are important factors that determines fomesafen persistence under field conditions. Fomesafen is a weak acid with pK_a of 2.7. The solubility of fomesafen is 50 mg L^{-1} at pH 7 and decreases to less than 1 mg L^{-1} at pH 1 (Senseman, 2007). Therefore, increased sorption of fomesafen at low pH or at acidic soil surfaces may reduce the water solubility, mobility and bioavailability in soil because

of the formation of hydrophobic bonding between the fomesafen molecule and lipophilic sites on the organic colloidal surfaces (Weber 1993a; Tanford 1973). Weber (1993a) reported that decreasing the suspension pH from 6.3 to 2 greatly increased adsorption of fomesafen by all sorbents of this study (H^+ and Ca^{2+} saturated soil organic matter, Ca^{2+} saturated soil organic matter, Ca^{2+} saturated montmorillonite clay, Norfolk sandy loam and Drummer silt loam). Fomesafen sorption to Drummer silt loam and Norfolk sandy loam increased 4.1 and 19 fold respectively, when decreasing natural soil pH (6.3 for Drummer silt loam and 5.3 for Norfolk sandy loam) to 2. Guo et al. (2003) tested the adsorption, desorption and mobility of fomesafen in six soils from China and noted that soil pH was more important than soil OM for adsorption. In addition, 44 to 81% of the absorbed fomesafen was desorbed from these soils with one desorption process, therefore making it more readily available for herbicidal activity. At agronomic soil pH ranges (5 to 8), fomesafen adsorption to Fe and Al oxides could occur because many weak acids mainly appear in their anionic forms (Newby and White 1981; Pusino et al. 1997).

Similar to fomesafen, other weak acid herbicides, such as chlorsulfuron, perfluridone, and imidazolinones, have been reported to have less soil adsorption with moderate to high mobility under neutral or alkaline conditions (Ketchersid and Merkle 1975; Weber 1993a, 1993b). Mersie and Foy (1986) reported chlorsulfuron mobility (R_f) was positively correlated to soil pH (r=0.97) and negatively correlated to organic carbon (OC) (r=0.93) in a soil thin-layer chromatography study. Chlorsulfuron was 2.6 fold more mobile in Kenansville loamy sand (pH 6.9, OC=0.16%) than in Acedale silt loam (pH 4.6, OC=1.42%). Only 0.1% of the applied perfluridone was detected at 15.24 cm of a Sawyer loamy sand with pH 4, however, 72% of the applied perfluridone was found at the same depth when soil pH was elevated to 8.5 with lime.

Increasing pH from 4 to 8.5 also dramatically increased the mobility of 2, 4, 5-T and picloram in a Sawyer loam sand (Ketchersid and Merkle, 1974). Similarly, imidazolinones (imazamox, imazethapyr and imazaquin) exhibit greater adsorption to soil at pH 5 as compared to pH 7 and herbicide metabolism in soil was negatively related to soil adsorption (Aichele and Penner 2005). Imazapyr soil adsorption was strongly affected by the pH and the charge of the absorbing component; enhanced imazapyr adsorption to amorphous Fe oxide was observed, likely due to ligand exchange process (Pusino et al. 1997). In another study, imazethapyr persistence in Crosby silt loam and carryover injury of imazaquin on corn in Hoytville clay increased as pH decreased (Loux and Reese 1993). These results indicated weak acid herbicides exhibit greater adsorption and low mobility under agronomic low pH conditions.

Leaching could be another dissipation pathway for fomesafen from the soil surface. Weber (1993b) suggested fomesafen leaching occurred in tested soils when irrigated. Fomesafen exhibited higher mobility in sandier Norfolk sandy loam than other three soils when irrigated 1.25 cm d^{-1} for 40 d or 50 cm water continuously. Fomesafen mobility was negatively related to soil OM, humic matter, pH and CEC while soil liming increased fomesafen mobility in a Norfolk sandy loam. Although there has been published research regarding fomesafen adsorption, desorption and soil mobility, limited information is available regarding fomesafen behavior in southern US soils, and the effect of biological degradation on fomesafen persistence under aerobic condition. Therefore, the objective of this experiment was to: 1) evaluate fomesafen soil behavior as affected by various soil properties and 2) investigate fomesafen biological degradation in two GA soils.

Materials and Methods

Adsorption and desorption. Seven distinct soils were used to evaluate fomesafen adsorption and desorption (Table 1). From each soil, 10 g of air-dried and sieved soil with 20 ml of $CaCl_2$ solution containing 3, 6, 12, 24 and 48 µmol L^{-1} of fomesafen was added to 50 ml polypropylene centrifuge tubes and mixed. Fomesafen sodium salt (Reflex 2SL, Syngenta Crop Protection, Inc. Greensboro, NC 27419) was used to prepare the $CaCl_2$ solutions mixed with soil samples. Mixed samples were shaken for 24 h at 22 C to reach equilibrium. Then, slurry was centrifuged at 4000 RPM (Beckman Model TJ-6 centrifuge, Indianapolis, IN. 46268) for 5 min and 2 ml of supernatant was filtrated with 0.25 µm nylon syringe filter (Fisher Scientific, Pittsburgh, PA. 15275) for HPLC analysis. Fomesafen quantification was performed with Waters 2695 HPLC and Waters 2996 PDA detector. Separation was conducted by a Waters XTerra Shield RP18 column (4.6 mm × 250 mm, 5 µm. Waters Co. Milford, MA. 01757) at 60 C, using two mobile phases, 0.1% formic acid in water (A) and acetonitrile (B). Flow program ratio was set as 62% A/ 38% B initially and linearly decreased to 10% A/ 90% B in 7.5 min, then held isocratic for 2 min. Fomesafen was eluted at 6.95 min without interference. System flow rate was 0.75 ml min^{-1} and detection wavelength was 290 nm. Quantification limit of fomesafen in water was 0.05 ug ml^{-1}. The concentration difference between the initial and final equilibrium solutions was used to calculate fomesafen adsorption to soil. The study had three replications and was repeated twice.

Samples of each soil that mixed with 6, 24 and 48 µmol L^{-1} fomesafen solution were used to perform desorption study. The supernatant was decanted after initial equilibrium had been reached and 20 ml of blank $CaCl_2$ solution was added into each tube and shaken 24 hrs to achieve new equilibrium. Following preparation procedures were similar as the adsorption study

and final liquid samples were analyzed by HPLC. The desorption process was conducted only once on selected soil samples.

Adsorption data was fitted to the logarithmic form of the Freundlich isotherms (Stougaard et al. 1990; Weber 1993a; Grey et al. 1997):

$$\log Cs = \log K_f + 1/n \log Ce \quad [1]$$

where Cs (µmol kg^{-1}) is the amount of herbicide adsorbed at the equilibrium concentration Ce (µmol L^{-1}); K_f and $1/n$ are constants that characterize the relative sorption capacity and the sorption intensity, respectively. K_f is the mathematical description of distribution of the herbicide between the solid and solution phases. K_f value for each soil is reported and K_{OC} (soil organic carbon adsorption coefficient) is calculated as:

$$K_{OC} = (K_f / OC\%) \times 100 \quad [2]$$

$$OC\% = OM\% \times 0.58 \quad [3]$$

Fomesafen incubation. A fomesafen soil dissipation experiment was conducted in the laboratory using a Cecil sandy loam and Tifton loamy sand. To initiate the experiment, 50 g of dry soil was added to 250 ml conical flasks and brought to 12% soil moisture with 6 ml of fomesafen solution. Initial fomesafen soil concentration was set as 0.5 mg kg^{-1}, to simulate a 560 g ai ha^{-1} field PPI application. Incubation flasks were sealed with parafilm to prevent soil drying and then soil samples were incubated under lab condition at 22 C. Soils were sampled at 1 hr after treatment, 1, 2, 7, 14, 28, 56 and 90 d after treatment (DAT). Samples were kept in the dark storage at 0 C until extraction. Fomesafen residue was extracted by shaking soil sample in each bioassay flask with 100 ml 50:50 HPLC grade water and dichloromethane plus 0.5% acetic acid for 2 hr. Then slurry of each sample was poured into 50 ml polypropylene centrifuge tubes and centrifuged at 4000 RPM for 5 min. After centrifuge, supernatant was transferred to a separation

funnel, where lower phase (dichloromethane) was collected. Dichloromethane was evaporated to dryness, fomesafen residues was re-dissolved into 2ml of 70:30 water and acetonitrile solution by sonication, then filtrated through 2 μm nylon syringe filter for HPLC detection. Fomesafen recovery efficiency was proven to be over 90%. The experiment included 3 replications and was repeated twice.

Statistical analysis. The PROC CORR procedure in SAS (Version 9.3, SAS Institute Inc. Cary, NC. 27513) was used to conduct pairwise correlation in order to evaluate the effect of soil properties on fomesafen adsorption and desorption. Pearson correlation coefficients and corresponding P values were reported in Table 4. To calculate the fomesafen DT_{50}, non-linear regression was performed using Sigmaplot 12.0 (Systat Software, Inc. San Jose, CA 95110) using a two-parameter exponential decay function,

$$f(x) = b_0 e^{-b1(x)} \quad [4]$$

where y is the fomesafen concentration in soil samples; B_0 is the initial value of fomesafen concentration (y) when incubation time X is zero; B_1 is the rate of decline of concentration (slope) and X is incubation time.

Results and Discussion

Fomesafen adsorption kinetics. The adsorption kinetics of fomesafen to Cecil sandy loam is shown in Figure 1. Approximately 66% fomesafen in the solution was adsorbed to Cecil sandy loam at 30 min. Adsorption increased to 72% at 1 hr and final adsorption rate was 76% after 24 hr continuous shaking. This is consistent with Guo et al. (2003) who reported that fomesafen adsorption and desorption could reach equilibrium after shaking for 1 hr. Exposing additional sorption sites on soil particles as a result of prolonged shaking, could have caused the increased

herbicide sorption after the initial rapid phase (Savage and Wauchope 1974; Walker and Jurado-Exposito 1998; Ferrell et al 2005).

Adsorption and desorption. Three GA soils, one TX soil and one KY soil were included in the experiment as these soils occur in major cotton and soybean growing regions in the Southern US, where fomesafen is applied (Table 1). GA soils typically have higher sand fraction and lower pH as compared to other soils in this study. The Haxtun Sandy Loam from CO and Minidoka silt loam from ID were also chosen in order to evaluate fomesafen behavior in soils with high pH and low sand fraction. The coefficient K_f of the Freundlich adsorption isotherms were listed in Table 2 for soils used in this study and this constant represents the amount of pesticide adsorbed to the surface of an absorbent at an equilibrium concentration of 1 µmol L^{-1}. Therefore, it is a good description of pesticide adsorption to soil surface at low concentration. Adsorption non-linearity has occurred in this experiment since 1/n value ranged from 0.54 to 0.99 and Freundlich adsorption isotherms equation provided a good description of data with r^2 value $\geq$ 0.97 for all seven soils (Figure 2).

Fomesafen adsorption was generally low for soils used in this study (Table 2). The highest K_f was recorded with Cecil sandy loam (9.28) and lowest value was observed on Tremona sand (1.3), possibly due to a high sand fraction and low OM in this soil type. Similarly, the Tifton loamy sand had a low K_f, OM, and high sand fraction. Most of the soils examined had a K_f value lower than 3.0, which indicates that fomesafen may not be tightly bounded to surface in these soils and leaching is possible under certain environmental conditions. Fomesafen K_{oc} in this study varied significantly (11.7-fold) and this suggested that OM was not the major adsorptive fraction in the soil matrix and there should be other soil factors influence fomesafen adsorption process. Typically, a hydrophobic molecule should have higher possibility to enter

organic phase from aqueous phase in soil and form strong hydrophobic bonds with OM in soil. This process should be relatively independent to other soil factors and K_{oc} of this molecule should be relatively constant across a range of soils (Morillo et al. 2004). This prediction was consistent with the observed data (Ferrell et al. 2005) on flumioxazin soil adsorption, in which K_{oc} of flumioxazin only varied 1.6-fold over 6 soils, since this compound has low water solubility (1.78 mg L^{-1}) and is not ionizable (Harper 1994). However, this was not the case for fomesafen because it is a weak acid (pK_a of 2.7) with moderate water solubility (50 mg L^{-1}).

The fomesafen desorption rate varied dramatically between soils examined (Table 3). Fomesafen on the Cecil sandy loam, Sonora silt loam and Minidoka silt loam showed lower desorption rate than other soils examined. The highest fomesafen desorption values were recorded for the Tremona sand, Tifton loamy sand and Haxtun Sandy Loam. This was likely due to high sand fraction, high pH and low OM. Typically, fomesafen was more readily desorbed from soil surface at higher initial concentration than lower concentration, which suggested fomesafen molecules could be tightly adsorbed to soil surface at low concentrations and therefore, it is harder to desorb them. Similar findings have been reported by Morillo et al. (2004) on norflurazon desorption from 17 European soils and by Pusino et al. (1997) on imazapyr desorption from 6 Italian soils.

Pairwise correlation (Table 4) results suggested soil pH, clay and OM played significant role during adsorption. Pearson correlation coefficient for pH, clay and OM to K_f was -0.6832, -0.6444 and 0.4286 with corresponding p-value of 0.0006, 0.0016 and 0.0525, respectively. This indicated pH was more important for fomesafen adsorption than clay and OM, and it was inversely correlated to fomesafen adsorption. Previous studies have reported that soil pH was more important than OM during fomesafen adsorption to soils (Guo et al. 2003). Fomesafen is a

weak acid and its solubility, mobility and affinity to soil could be affected by soil pH (Weber, 1993a). Low pH decreases fomesafen water solubility and increases its affinity to soil OM due to the formation of hydrophobic bonds between fomesafen molecule and lipophilic sites on the organic colloidal surfaces (Tanford 1973). For desorption, all soil parameter listed in Table 4 were significant except for CEC. OM and sand fraction had the greatest impact on fomesafen desorption, followed by pH, silt and clay. Sand and soil pH were positively related desorption while silt, clay and OM were negatively related to desorption, which indicates fomesafen leaching potential may be escaladed in alkaline soils with high sand fraction and low OM content. Increased fomesafen leaching has been reported in a Norfolk sandy loam compared to other soils with higher OM and lower sand fraction (Weber 1993b). Moreover, liming this Norfolk soil increased fomesafen mobility. In another field bioassay study, cotton plants exhibited more stand and height reduction in Tifton loamy sand as compared to bioassays in Cecil sandy loam and Greenville sandy clay loam, possibly due to less fomesafen adsorption and more desorption from this sandy soil (Li et al. Unpublished data). These research data suggested fomesafen may possess stronger mobility and bioavailability to plants when soil properties favor less fomesafen adsorption and more desorption from soil surface. Similarly, Stougaard et al. (1990) reported mobility of imazaquin and imazethapyr increased when increasing soil pH from 5 to 7. More wheat height reduction was observed at pH 7 compared to pH 5 for both herbicides. Imazaquin and imazethapyr caused 15 to 20% height reduction at pH 5 and reduction increased to 40 to 60% at pH 7, as compared to non-treated check. This is possibly caused by more herbicide desorption from soil surface at high pH, thus increased their availability for plant uptake.

Fomesafen biological degradation. It has been reported that fomesafen degradation occurred rapidly in soil under anaerobic conditions (Senseman, 2007), however, little published data regarding fomesafen biological degradation under aerobic condition is available up to present. Lab incubation data suggested fomesafen was barely degraded by soil microorganisms in both Cecil sandy loam and Tifton loamy sand during the 90 d incubation, with ambient temperature and soil moisture (Figure 3). Approximately 79 and 99% of the applied fomesafen still remained in Cecil sandy loam and Tifton loamy sand 90 DAT. Although a decreasing trend was observed in Cecil sandy loam, non-linear regression with a two-parameter exponential decay model suggested slope failed to be significant for both soils (data not shown). The results of this study demonstrated that biological degradation may not be the major pathway for fomesafen dissipation under aerobic condition in field. Similar results have been reported that fomesafen had first order half-life of 90 wk in Frensham loamy sand, 75.3 wk in Gore silty clay loam and 29.7 wk in a Wisborough silty clay loam when incubated at 20 C and 40% soil water holding capacity (EPA 2006). Meanwhile, a fomesafen field dissipation study in GA determined that fomesafen DT_{50} in the top 7.5 cm layer was 34 and 4.5 d respectively, for Cecil sandy loam and Tifton loamy sand after 560 g ai ha^{-1} application. Fomesafen residue lasted over 120 d in Cecil sandy loam but was not detectable in Tifton loamy sand 28 DAT (Li et al. Unpublished data). Together, these findings demonstrated that biological degradation was not likely to be the major pathway for fomesafen dissipation in these soils; higher adsorption to Cecil sandy loam may account for the greater DT_{50} and longer fomesafen retention in this soil as compared to the Tifton loamy sand. Fomesafen may possess higher mobility and bioavailability to crops in soils with high sand fraction, high pH and lower OM.

Literature Cited

Aichele TM, Penner D (2005) Adsorption, desorption and degradation of imidazolinones in soil. Weed Technol 19:154-59

EPA (2006) Sodium salt of fomesafen. Docket ID: EPA-HQ-OPP-2006-0239-0010. Available from: http://www.regulations.gov/#!docketBrowser;rpp=25;po=0;s=EPA-HQ-OPP-2006-0239-0010;dct=PS%252BSR;D=EPA-HQ-OPP-2006-0239

Ferrell JA, Vencill WK, Xia K, Grey TL (2005) Sorption and desorption of flumioxazin to soil, clay minerals and ion-exchange resin. Pest Manag Sci 61:40-46

Grey TL, Walker RH, Hancock HG (1997) Sulfentrazone adsorption and mobility as affected by soil and pH. Weed Sci 45:733-38

Guo J, Zhu G, Shi J, Sun J (2003) Adsorption, desorption and mobility of fomesafen in Chinese soils. Water Air and Soil Pollut 148:77-85

Harper SS (1994) Sorption–desorption and herbicide behavior in soil. Rev Weed Sci 6:207–25

Ketchersid ML, Merkle MG (1975) Persistence and movement of perfluridone in soil. Weed Sci 23:344-48

Loux MM, Reese KD (1993) Effect of soil type and pH on persistence and carryover of imidazolinon herbicides. Weed Technol 7:452-58

Mersie W, Foy CL (1986) Adsorption, desorption and mobility of chlorsulfuron in soils. J Agric Food Chem 34:89-92

Morillo E, Undabeytia T, Cabrera A, Villaverde J, Maqueda C (2004) Effect of soil type on adsorption-desorption, mobility and activity of the herbicide norflurazon. J. Agric Food Chem 52:884-90

Newby SE, White BG (1981) PP021: leaching on soil thick-layer chromatograms. London, ICI. pp.4 (Report RJ0156B)

Oymada M, Kuwatsuka S (1988) Effects of soil properties and conditions on the degradation of three diphenylether herbicides in flooded soils. J Pestic Sci 13:99-105

Pusino A, Petretto S, Gessa C (1997) Adsorption and desorption of Imazapyr by soil. J Agric Food Chem 45:1012-16

Rauch BJ, Bellinder RR, Brainard DC, Lane M, Thies JE (2007) Dissipation of fomesafen in New York state soil and potential to cause carryover injury to sweet corn. Weed Technol 21:206-12

Savage KE, Wauchope RD (1974) Fluometuron adsorption–desorption in soil. Weed Sci 22:106-10

Senseman SA, ed (2007) Herbicide Handbook. 9th ed. Lawrence, KS: Weed Science Society of America. pp. 207

Stougaard RN, Shea PJ, Martin AR (1990) Effect of soil type and pH on adsorption, mobility and efficacy of imazaquin and imazethapyr. Weed Sci 38:67-73

Syngenta Crop Protection (2014) Reflex label. Greensboro, NC. 27419. Available from http://www.cdms.net/LDat/ld6BJ002.pdf

Tanford C (1973) The hydrophobic effect. John Wiley and Sons, Inc,. New York.

Walker A, Jurado-Exposito M (1998) Adsorption of isoproturon, diuron, and metsulfuron-methyl, in two soils at high soil:solution ratios. Weed Res 38:229-38

Weber JB (1993a) Ionizatin and sorption of fomesafen and atrazine by soils and soil constituents. Pestic Sci 39:31-38

Weber JB (1993b) Mobility of fomesafen and atrazine in soil columns under saturated and unsaturated flow conditions. Pestic Sci 39:39-46

Table 1. Soil information of adsorption and desorption study [a]

Location	Soil type	Taxonomy	pH	OM	Sand	Silt	Clay	CEC
				%	%	%	%	
Athens GA	Cecil sandy loam	Fine, kaolinitic, thermic Typic Kanhapludults	5.5	2.1	72	12	16	2.6
Plains GA	Greenville sandy clay loam	Fine, kaolinitic, thermic Rhodic Kandiudults	5.6	3.8	60	10	30	7.1
Tifton GA	Tifton loamy sand	Fine-loamy, kaolinitic, thermic Plinthic Kandiudults	5.6	1.0	90	6	4	2.5
Texas	Tremona sand	Loamy, fine sand, thermic Aquic Arenic Paleustalfs	7.9	0.4	92	2	6	4.2
Kentucky	Sonora silt loam	Fine-loamy, mixed, semiactive, mesic Typic Paleudalfs	6.9	3.5	38	46	16	14.0
Colorado	Haxtun sandy Loam	Fine-loamy, mixed, superactive, mesic Pachic Argiustolls	8.0	1.4	60	26	14	26.0
Idaho	Minidoka silt loam	Coarse-silty, mixed speractile, mesic Xeric Haplodorid	7.0	2.3	30	54	16	12.0

[a] Soil information was provided by University of Georgia Soil Testing Laboratory. Athens GA

Table 2. Sorption coefficient estimates for fomesafen

Soil type	K_f (± SEM)	K_{oc}	1/n (± SEM)	R^2
Cecil sandy loam	9.28 ± 0.68	810 ± 32	0.54 ± 0.02	0.97
Greenville sandy clay loam	7.76 ± 0.45	371 ± 12	0.67 ± 0.02	0.99
Tifton loamy sand	1.70 ± 0.15	323 ± 15	0.64 ± 0.02	0.98
Tremona sand	1.30 ± 0.12	578 ± 30	0.67 ± 0.02	0.98
Sonora silt loam	1.35 ± 0.13	69 ± 4	0.99 ± 0.02	0.98
Haxtun sandy Loam	2.05 ± 0.15	266 ± 11	0.64 ± 0.02	0.98
Minidoka silt loam	2.87 ± 0.17	231 ± 7	0.91 ± 0.01	0.99

Table 3. Percentage of fomesafen desorbed from soil

Soil type	% of desorption [a]		
	6 µMol/L	24 µMol/L	48 µMol/L
Cecil sandy loam	11	19	29
Greenville sandy clay loam	28	20	24
Tifton loamy sand	26	36	49
Tremona sand	40	49	81
Sonora silt loam	17	16	23
Haxtun sandy Loam	40	48	70
Minidoka silt loam	10	11	16

[a] Data presented was desorption rate after one 24 hr desorption process. Rate represented initial fomesafen solution concentration.

Table 4. Correlations of soil parameters to K_f and desorption

	K_f		Desorption	
Parameters	Correlation	P-value	Correlation	P-value
Sand	0.0101	0.9655	0.5812	**0.0057**
Silt	-0.2789	0.2209	-0.4883	**0.0247**
Clay	0.6444	**0.0016**	-0.4502	**0.0406**
pH	-0.6832	**0.0006**	0.4922	**0.0234**
CEC	-0.3648	0.1039	0.1664	0.4710
OM	0.4286	0.0525	-0.6328	**0.0021**

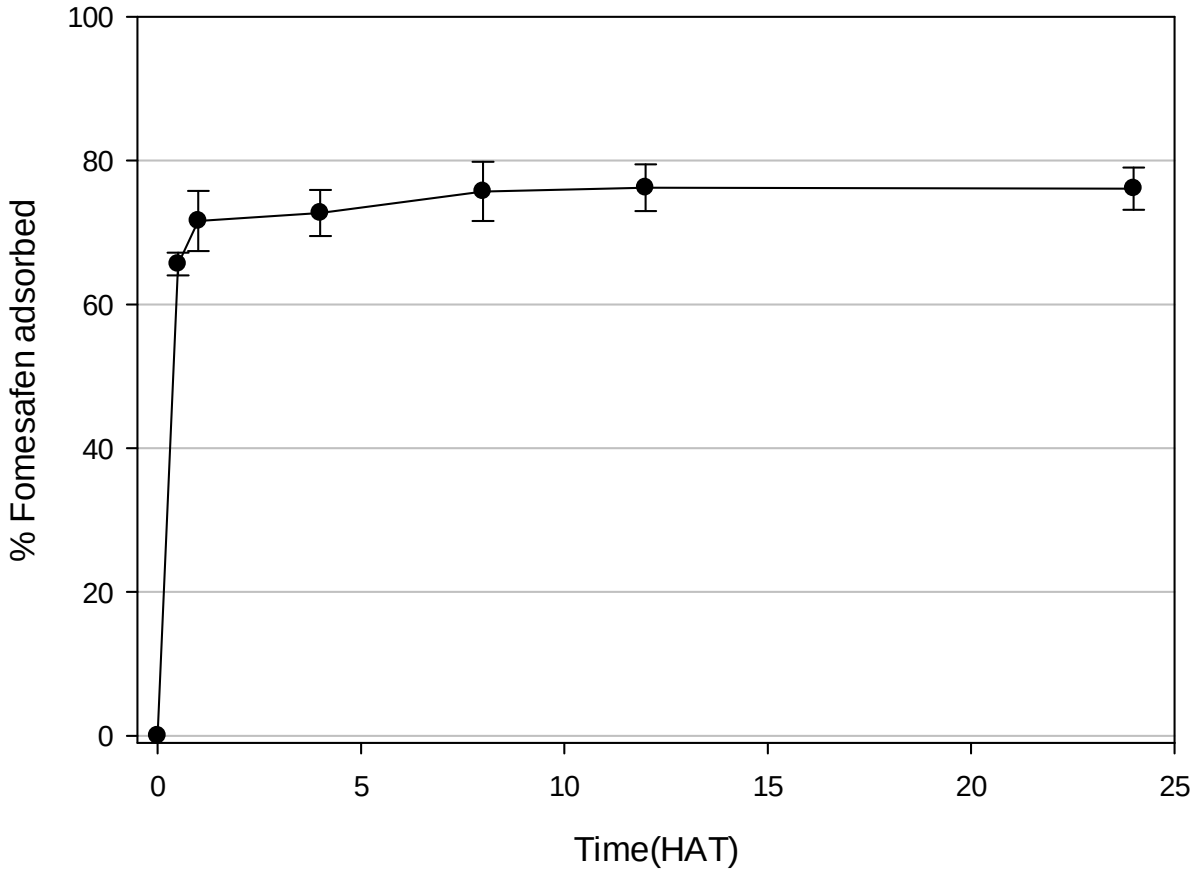

Figure 1. Fomesafen adsorption to Cecil sandy loam over 24 hr period. Error bars represent standard error of each mean

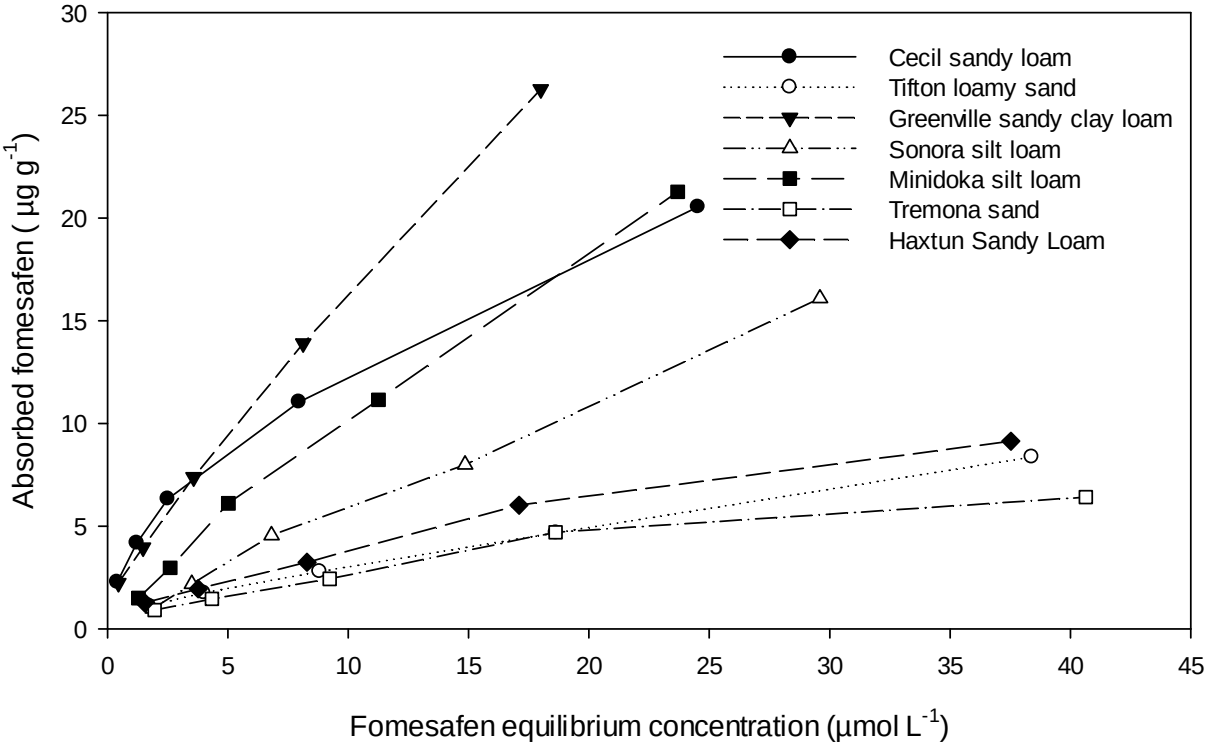

Figure 2. Fomesafen adsorption isotherms on 7 soils.

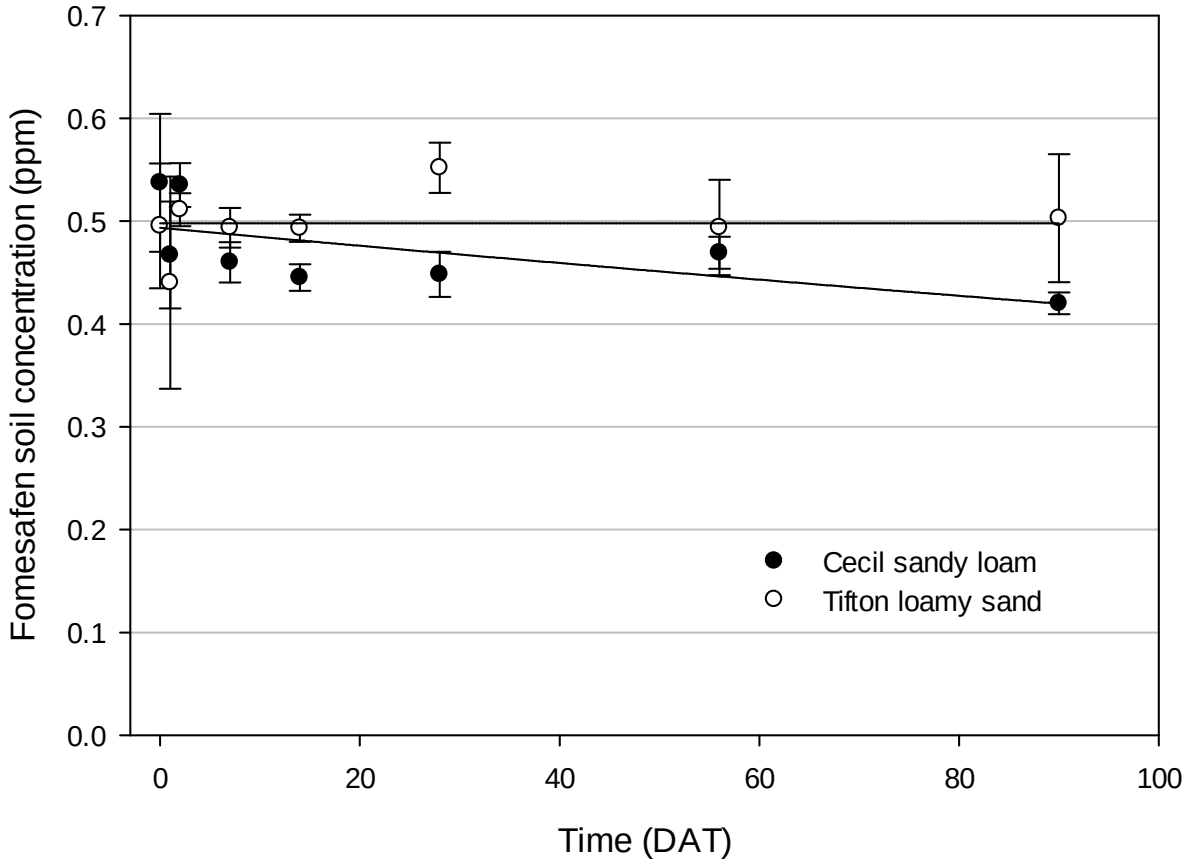

Figure 3. Fomesafen degradation in Cecil sandy loam and Tifton loamy sand under laboratory environment. Error bars represent standard error of each mean. Two-parameter exponential decay model was used to describe the data. F-test indicated that both models failed to be significant at 0.05 level.

CHAPTER 3

FOMESAFEN SOIL DISSIPATION AND COTTON RESPONSE [3]

[3] Xiao Li, Timothy L. Grey, Brian H. Blanchett, William K. Vencill. Theodore M. Webster. To be published in Journal of Cotton Science.

Fomesafen Soil Dissipation and Cotton Response [4]

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Fomesafen provides effective control of glyphosate resistant Palmer amaranth when applied PRE in cotton. However, cotton seedling injury is possible under adverse environmental conditions and coarse texture soil scenarios. Therefore, greenhouse and field experiments were conducted at three locations in Georgia (Athens, Plains and Ty Ty) to evaluate cotton growth and yield response to fomesafen applied PRE (0, 70, 140, 280, 560, 1120 and 2240 g ai ha^{-1}). Fomesafen dissipation under field conditions was also evaluated at Athens on a Cecil sandy loam and Ty Ty on Tifton loamy sand. Greenhouse cotton bioassay indicated fomesafen reduced cotton height and dry weight with increasing rate in the Cecil sandy loam and Tifton loamy sand but not in the Greenville sandy clay loam. In Athens, fomesafen did not negatively affect field cotton height as compared to NTC during the course of the season. At Plains, cotton exhibited

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height reduction when treated with the 2240 g ai ha^{-1} rate and at Ty Ty, cotton height was reduced by the two highest rates (1120 and 2240 g ai ha^{-1}) up to 71 d after treatment (DAT). Seed lint cotton yield was not affected by fomesafen at any location. Laboratory analysis indicated fomesafen field dissipation varied significantly between soils. Fomesafen persisted over 120 d for the Cecil sandy loam, but was not detectable past 28 DAT for the Tifton sandy loam. The half-life (DT_{50}) of fomesafen applied at 280 g ai ha^{-1} was 47 and 6 d for Cecil sandy loam and Tifton loamy sand, respectively. When applied at 560 g ai ha^{-1}, the DT_{50} was 34 and 4 d for Cecil sandy loam and Tifton loamy sand, respectively. These data indicated fomesafen persistence varied in different soils and cotton was not affected by fomesafen within 280-420 g ai ha^{-1} label rate.

Nomenclature: Fomesafen; Palmer amaranth, *Amaranthus palmeri* S. Wats. AMAPA; cotton, *Gossypium hirsutum* L.

Key words: Fomesafen, cotton tolerance, growth response, stand count, field persistence.

Introduction

Since the adoption of glyphosate resistant (GR) crops, herbicide-resistant Palmer amaranth has become common throughout the southeastern cotton-growing region (Sosnoskie et al. 2011; Wise et al. 2009). Due to multiple herbicide-resistance to glyphosate and acetolactate synthase (ALS) inhibitors, no effective topical option is available for growers to control GR and ALS-inhibitor resistant Palmer amaranth in glyphosate based cropping system (Culpepper, 2009; Sosnoskie et al. 2011). Herbicide resistant Palmer amaranth can be controlled by glufosinate, however, crop cultivars need to be glufosinate-resistant and applications have to be timely on small Palmer amaranth seedlings less than 10 cm tall (Culpepper et al. 2009; Marshall 2009). Therefore, it is recommended that growers use residual herbicides with different mechanisms of action in cotton to improve Palmer amaranth control and to minimize further herbicide resistance development. These residual herbicides are considered to be the key component in the current weed control programs for cotton.

Previous research has confirmed fomesafen was effective to control GR and ALS resistant Palmer amaranth in cotton (Culpepper 2009; Bond et al. 2006; Gardner et al. 2006; Troxler et al. 2002). Fomesafen is a diphenylether herbicide that inhibits protoporphyrinogen oxidase (PPO). Fomesafen cotton registration allows preplant surface application to medium or fine-textured soils for rates of 280 ai $ha^{-1}$and PRE application to coarse-textured soils for rates of 280 to 420 g ai ha^{-1} (Syngenta, 2014). Fomesafen controls annual broadleaf weeds including pigweed species, (*Amaranthus* spp.), morningglory species (*Ipomoea* spp.), jimsonweed (*Datura stramonium* L.), wild mustard (*Sinapis arvensis* L. ssp. *Arvensis*), black nightshade (*Solanum nigrum* L.) and

ragweed species (*Ambrosia* spp.) (Senseman, 2007). Treatments containing fomesafen improved early-season common cocklebur (*Xanthium strumarium* L.) and *Ipomoea* spp. control when properly activated by irrigation or precipitation (Stephenson et al 2004). Fomesafen plus pendimethalin (280 and 1120 g ai ha^{-1}, respectively) applied PRE followed by glufosinate mid-POST provided excellent control (> 90%) of Palmer amaranth, common lambsquarter (*Chenopodium album* L.), large crabgrass (*Digitaria sanguinalis* L.) and goosegrass (*Eleusine indica* Elein) (Everman et al. 2009). Fomesafen tank mixed with flumeturon at 0.42 and 1.68 kg ai ha^{-1} rates resulted in higher weed control than either herbicide used alone; fomesafen 0.42 kg ai ha^{-1} tank mixed with MSMA at 2.24 kg ai ha^{-1} effectively controlled yellow nutsedge, morningglory species and pigweed species when POST-directed to cotton (Lunsford et al. 1998). Another research indicated that average control of Palmer amaranth by fomesafen at 280 and 426 g ai ha^{-1} was 94% and yellow nutsedge (*Cyperus esculentus* L.) control ranged from 68 to 77% and 90 to 98% respectively at two SC locations, but fomesafen did not control purple nutsedge (*Cyperus rotundus* L.) and sicklepod (*Senna obtusifolia* L.) (Murdock and Keeton, 1998).

A major concern from cotton growers regarding fomesafen is potential injury to cotton seedlings, especially when applied PRE to moist soil (Kichler and Culpepper, 2012). Murdock and Keeton (1998) reported fomesafen cotton injury was generally greater when applied PRE than PPI; average injury was 5, 9, 14 and 23% respectively when fomesafen applied PRE at 280, 426, 560 and 840 g ai ha^{-1}. When applied PPI, average injury was 1, 4, 5 and 15% for those respective rates. Schrage et al. (2012) concluded cotton seeds with low vigor incurred 20% greater fomesafen injury, and deep

planting at 2.5 cm caused 15% more injury than planting at 0.6 cm. Similar to fomesafen, other PPO inhibitors have been reported to cause various levels of injury on cotton and soybean (*Glycine max* L.). One research has noted significant cotton injury occurred when flumioxazin applied at planting at 70 g ai ha^{-1}, but injury was not greater than 12% and cotton yield was not affected (Askew et al. 2001). Some researchers reported severe flumioxazin injury on cotton leaves occurred when heavy rainfall splashed treated soil onto leaf surface of 15 cm tall cotton (Wilcut et al. 2000). Therefore, flumioxazin application should be restricted to the cotton bark and a misapplication over the top or to small cotton with green stems could cause serious injury (Wilcut et al. 2000; Cranmer et al. 2000). In soybean, sulfentrazone at 0.22 and 0.44 kg ai ha^{-1} caused greater soybean injury, reduced stand and yield in a soil with 1.1% organic content (OC) as compared to soils with 2.3% and 2.9% OC when applied 7 d before planting, at planting and at 50% hypocotyl emergence (Reiling et al. 2006). Moreover, 15 soybean varieties exhibited different level of tolerance to flumioxazin and sulfentrazone; injury from sulfentrazone was 10% greater than flumioxazin over 3 rates evaluated and sulfentrazone at 224 g ai ha^{-1} reduced plant height 23 to 53% and caused 18 to 38% visual injury in greenhouse (Taylor-Lovell et al. 2001).

Fomesafen is a weak acid with pK_a of 2.7 (Senseman, 2007). Therefore, its solubility and bioavailability are expected to be affected pH (Weber 1993). Previous research indicated fomesafen half-lives (DT_{50}) varied dramatically under different environmental and soil conditions. Rauch et al. (2007) reported fomesafen field DT_{50} was 28 to 66 d with an average of 50 d. Fomesafen dissipation under anaerobic conditions was less than 3 wk, but persistence in soil varied significantly under field conditions with DT_{50} varying

from 6 to 12 months (Senseman, 2007). Oymada and Kuwatsuka (1988) investigated the persistence of three diphenylether herbicides in soil. The DT_{50} varied from 9 to 173 d for chlornitrofen, 3 to 87 d for nitrofen and 8 to 64 d for chlomethoxynil due to differences in chemical and soil properties. Similar to fomesafen, these herbicides degraded rapidly under anaerobic conditions. Due to long soil persistence, fomesafen residue may injure susceptible crops, such as sugar beets (*Beta vulgaris* L.), sunflowers (*Helianthus annuus* L.) and sorghum (*Sorghum bicolor* L.) up to one year after application (Senseman, 2007). The minimal rotational interval for small grains such as wheat (*Triticum aestivum* L.), barley (*Hordeum vulgare* L.) and rye (*Secale cereale* L.) is 4 months and for corn (*Zea mays* L.), peanut (*Arachis hypogaea* L.), peas (*Pisum sativum* L.) and rice (*Oryza sativa* L.) is 10 months (Syngenta 2014). Dotray et al. (2010) reported fomesafen injury on peanut when applied PRE, AC (at cracking) and EPOST at two TX locations was unacceptable even though it provided good to excellent control of some broadleaf weeds. In this experiment, 280 and 560 g ai ha^{-1} applications caused up to 59% mid-season injury in 2008 experiment and 46% injury in 2009 experiment. Late season injury was apparent and yield reduction was observed in most of the treatments (Dotray et al. 2010). Gilbert et al. (2009) reported fomesafen applied from 220 to 560 g ai ha^{-1} caused significant peanut injury. Yield reduction was common regardless application timing (PRE, AC and EPOST) in all four trial locations (Lamesa TX, Tifton GA, Citra FL and Lewiston-Woodville NC).

Fomesafen application in cotton production areas has dramatically increased in the past a few years. Total usage of fomesafen in all cotton producing states increased by 4.92-fold from 2007 to 2010. Total usage in GA was 18,573 kg on 66,636 ha in 2007 and

increased to 65,232 kg on 220,742 ha in 2010 (USDA-NASS, 2010). However, there is limited information regarding its soil dissipation, potential injury to cotton and carryover to susceptible crops in GA. Therefore, the objective of this research was to evaluate fomesafen field dissipation and study the cotton growth response to fomesafen when applied PRE in greenhouse and field experiments in GA.

Materials and Methods

Greenhouse cotton response. Cotton response to fomesafen was evaluated in University of Georgia greenhouse in Athens, GA from February to May 2012. The experiment was a complete randomized design with 5 reps, repeated twice. Plants were grown with a 30/20 C temperature setting and 16 hr photoperiod. The cotton variety was FM1845LLB2 (Fibermax®, Bayer Cropscience. RTP, NC. 27709) with seeds planted 1 cm deep in containers (25cm long, 6.5 cm diameter) filled with either Cecil sandy loam from Athens GA, Greenville sandy clay loam from Plains GA or Tifton loamy sand from Ty Ty GA. Fomesafen (Reflex®, 239 g ai L^{-1}, Syngenta Crop Protection, LLC. Greensboro, NC. 27419) was applied at 0, 70, 140, 280, 560, 1120 and 2240 g ai ha^{-1} with XR 9003VK (Teejet®, Spraying Systems Co. Wheaton, IL. 60187) flat-fan nozzle tips calibrated to deliver 183 L ha^{-1} at 187 kpa. Herbicide treatments were applied in a spray chamber on the day of planting, and containers were irrigated immediately after application (1.5 cm). After emergence, cotton was irrigated and fertilized biweekly with Miracle Gro® (The Scotts Company, LLC. Marysville, OH. 43041). Seedling height and visual injury were evaluated 1 month after treatment and aboveground biomass was harvested for dry weights.

Field cotton tolerance to fomesafen. Field trials were conducted at three locations in GA in 2013 (Table 5) using a randomized complete block design. Athens and Plains experiments had four replications while Ty Ty location had three. Athens, Plains and Ty Ty plots were 6, 10 and 7.5 m long and cotton was planted in 0.9 m-wide rows at each location. Athens plots had two rows while Plains and Ty Ty plots had four. The cotton variety was 'DP1137B2RF' (Deltapine®, Monsanto Co., St. Louis, MO. 63167). Soil texture, planting and harvesting dates were listed in Table 5. Fomesafen was applied at 0, 70, 140, 280, 560, 1,120, 2,240 g ai ha^{-1} and 280 g ai ha^{-1} with pendimethalin at 924 g ai ha^{-1} at the day of planting, using backpack sprayer with four nozzle tips (11003VK flat fan nozzles, Teejet®, Spraying Systems Co. Wheaton, IL. 60187) propelled by compressed CO_2. Spray volume was 187 L ha^{-1} at 207 kPa. Treated plots were irrigated immediately after fomesafen application to ensure soil activation, and then plots were irrigated as needed during growing season. Total rainfall and irrigation in season amounts to 84.5, 67.5 and 94.5 cm respective, for Athens, Plains and Ty Ty trials. Plots were maintained weed-free throughout the growing season with glyphosate (Roundup Weathermax®, 540 g ae L^{-1}, Monsanto Co., St. Louis, MO. 63167) and hand weeding. Cotton was defoliated one week prior to harvest with 1680 g ai ha^{-1} ethephon and 105 g ai ha^{-1} cyclanilide (Finish 6®, Bayer Cropscience. RTP, NC. 27709) plus 1.85 g ai ha^{-1} pyraflufen ethyl (ET®, 25 g ai L^{-1}, Nichino America, Wilmington, DE. 19808) plus 2.34 L ha^{-1} crop oil. Cotton stand was evaluated on 1 m stand from 35 to 42 DAT, and height data was recorded four times from 29 to 71 DAT at all locations. Stand count, height measure, and cotton seed-lint yield were averaged over the two center rows for statistical analysis.

Fomesafen residue persistence in field. Quantitative analysis of fomesafen was conducted in University of Georgia Tifton campus in 2013. Soil samples were taken from Athens, Plains and Ty Ty field plots that treated with 280 and 560 g ai ha^{-1} of fomesafen. Sampling dates were 1 hr after treatment, 1, 2, 7, 14, 42, 56, 84, 98 and 126 DAT. Soil samples were taken from surface to 7.5 cm deep, then wrapped in aluminum foil and kept frozen until extraction. Fomesafen residue was extracted by shaking 50 g soil of each sample with 100 ml 50:50 water: dichloromethane plus 0.5% acetic acid for 2 hr. Then supernatant of each sample was centrifuged (Beckman Model TJ-6 centrifuge, Indianapolis, IN. 46268) at 4000 RPM for 5 min and transferred to a separation funnel, where lower phase (dichloromethane) was collected. Dichloromethane was evaporated to dryness, re-dissolved with 2 ml of 70:30 water and acetonitrile solution, filtrated through 2 μm nylon syringe filter (Fisher Scientific, Pittsburgh, PA. 15275) for HPLC analysis. Fomesafen recovery efficiency was proven to be over 90%.

Residue quantification was performed with Waters 2695 HPLC and Waters 2996 PDA detector. Separation was conducted in a Waters XTerra Shield RP18 column (4.6 mm × 250 mm, 5 μm. Waters Co., Milford, MA. 01757) at 60 C, using two mobile phases, 0.1% formic acid in water (A) and acetonitrile (B). Flow program was set as 62% A/38% B initially and linearly decreased to 10% A/90% B in 7.5 min, then held isocratic for 2 min. Fomesafen peak eluted at 6.95 min without interference. System flow rate was 0.75 ml min^{-1} and detection wavelength was 290 nm. Quantification limit in soil was 0.002 ppmw or 2ug kg^{-1}.

Statistical analysis. All greenhouse and field data was converted to a percentage of the non-treated control (NTC) prior to statistical analysis. Non-linear regression did not

provide a good description of field data, therefore, these data was processed with PROC GLIMMIX procedure and means were separated with LSMEANS statement in SAS 9.3 (SAS Institute Inc. Cary, NC. 27513). Fomesafen rate was considered a fixed effect, while block and measuring dates were treated as a random effect. Since treatment-location interaction was significant constantly, results of field cotton height, stand count and yield were analyzed and presented by location. To describe greenhouse cotton and fomesafen field dissipation data, non-linear regression was performed with Sigmaplot 12 software (Systat Software, Inc. San Jose, CA 95110) using a two-parameter exponential decay function

$$f(x) = b_0 e^{-b1(x)} \quad (5)$$

, where y is the greenhouse cotton seedling height, dry weight or fomesafen concentration in field; b_0 is the initial value of the response variable (y) when rate X is zero; b_1 is the decline rate of the response variable (slope) and X is herbicide rate. Parameter estimates were given in table 3, 4 and 7, and b_1 was compared between treatments with LSD.

Results and discussions

Greenhouse cotton response. Cotton response to fomesafen varied significantly among three soil types (Figure 4). Fomesafen reduced cotton height and dry weight in Cecil sandy loam and Tifton loamy sand, but not in Greenville sandy clay loam. Slope (b_1 parameter) of non-linear regression revealed that the height of seedlings in Cecil sandy loam was most responsive to fomesafen, followed by Tifton loamy sand and Greenville sandy clay loam (Table 6). Similar to height, cotton dry weight showed greatest response to fomesafen in Cecil sandy loam, followed by Tifton loamy sand and was not affected

by fomesafen in Greenville sandy clay loam (Table 7). The GR_{50} in Cecil sandy loam was 1733 and 1155 g ai ha^{-1}, respectively, for cotton height and dry weight but could not be calculated with the rates examined in the Greenville sandy clay loam and Tifton loamy sand. Cotton received similar amount of injury in Cecil sandy loam as compared to Tifton loamy sand, with 20%, 35% and 70% injury for 560, 1120 and 2240 g ai ha^{-1} rates, respectively, when evaluated 2 WAT. Fomesafen injury evaluated 4 WAT on these two soils was 7.5%, 25% and 59% respectively, for 560, 1,120 and 2,240 g ai ha^{-1} rates. No significant injury was observed on cotton seedlings in Greenville sandy clay loam at any rate. High organic content and clay fraction in Greenville sandy clay loam may have reduced fomesafen injury on cotton by decreasing the amount of fomesafen available in soil solution for absorption. Similarly, Baumann et al. (1998) reported fomesafen applied from 560 and 840 g ai ha^{-1} treatment resulted up to 47% cotton injury in Amarillo sandy clay loam when applied PPI and PRE, but no injury was observed in Houston black clay when same treatments were applied. These results suggested soil texture is a critical factor determining fomesafen injury to cotton.

Field cotton tolerance to fomesafen. Cotton stand was evaluated between 35 to 42 DAT at each location (Table 8). In general, no effect of fomesafen on cotton stand was observed with any treatment except for the highest rate (2,240 g ai ha^{-1}), which reduced the stand by 24%, 39% and 52% in Athens, Plains and Ty Ty, respectively, as compared to the NTC. Cotton height was recorded four times between 29 to 71 DAT in all three locations and combined for data analysis. Overall, fomesafen did not adversely impact cotton height in Athens, but the highest rate reduced cotton height in Plains by 24%. Cotton was more responsive to fomesafen rates at Ty Ty than other locations: the highest

rate and 1,120 g ai ha^{-1} treatment reduced height by 41% and 27%, respectively, relative to the NTC. Although high rates of fomesafen may have the possibility to reduce cotton height and stand, cotton yield was not affected by fomesafen at any location (Table 9) since fixed effect fomesafen rates failed to be significant at 0.05 level for all locations. The only noticeable difference was the highest rate of fomesafen caused a 29% yield reduction in Ty Ty compared to NTC. These results suggested a good overall cotton tolerance to fomesafen but cotton could be responsive to very high rates of fomesafen in soils with large sand fraction and low OC. So growers should be cautious if high rate of fomesafen is accidentally applied in these soils due to miscalculation, spraying error, overlapping, etc.

Fomesafen plus pendimethalin was included in this study as a standard weed control practice in cotton growing area and this treatment did not incur any negative impact on cotton growth and yield at any location. Main et al. (2012) evaluated fomesafen applications on cotton from 0 to 840 g ai ha^{-1} in five Southern states. Fomesafen caused injury early to mid-season in three states and cotton yield was only reduced in North Carolina by 23 to 25% with 560 and 840 g ai ha^{-1} rates. Baumann et al. (1998) reported fomesafen applied PPI at 560 and 840 g ai ha^{-1} caused 22 and 47% early season injury in an Amarillo sandy clay loam, while mid-season injury decreased to 15 and 23%; PRE and POST-directed application in this soil resulted less than 10% injury and no yield reduction observed for any treatments applied to this soil. These results suggested cotton demonstrated good tolerance to fomesafen and yield was not affected when following label rates, but initial injury may be apparent under adverse environmental and soil conditions.

Fomesafen residue persistence in field. The field fomesafen dissipation experiment revealed significant differences among dissipation rate in Cecil sandy loam and Tifton loamy sand (Figure 5, 6). The DT_{50} of the 280 and 560 g ai ha^{-1} treatment was 47 and 34 d respectively for Cecil sandy loam. In Tifton loamy sand, the DT_{50} of fomesafen was 6 and 4 d, respectively, for the 280 and 560 g ai ha^{-1} treatment (Table 10). Fomesafen residues from the 280 and 560 g ai ha^{-1} treatments were detectable up to 126 DAT in Cecil sandy loam and 70 DAT in Greenville sandy clay loam (data not shown), but were not found in Tifton loamy sand 28 DAT. Non-linear regression indicated that dissipation rate (b_1) varied between soils and rates (Table 10). Fomesafen applied at 560 g ai ha^{-1} rate in Tifton loamy sand had the highest dissipation rate, followed by 280 g ai ha^{-1} in Tifton loamy sand and the 560 g ai ha^{-1} in Cecil sandy loam. Fomesafen applied at 280 g ai ha^{-1} in Cecil sandy loam had the slowest dissipation among all treatments. Dissipation of herbicides in soil and on plants was dependent on the physicochemical properties of the herbicides and environmental conditions (Ying and Williams 2000). Differences in soil dissipation have been reported for diphenylether herbicides. Fomesafen DT_{50} varied from 6 to 12 months under field conditions (Senseman 2007) and the DT_{50} three diphenylether herbicides (chlornitrofen, nitrofen and chlomethoxynil) varied from 9 to 173 d, 3 to 87 d and 8 to 64 d, respectively, on six Japanese soils. Previous research has confirmed that fomesafen has lower affinity to Tifton loamy sand, in term of K_d and K_f, and higher desorption rate as compared to Cecil sandy loam and microbial degradation was not the major dissipation pathway in these two soils (Li et al. Unpublished data). So it is reasonable to speculate that fomesafen may have leached out of the sampling zone

within 28 d in Tifton loamy sand under field conditions and more fomesafen remained in the sampling zone in Cecil sandy loam because of its high affinity to this soil.

Overall, cotton exhibited tolerance to fomesafen up to 2,240 g ai ha^{-1} in this study, although lower height and stand reduction may occur in field. Significant cotton visual injury (50 to 70%) was common early in the season following high rate applications (1,120 and 2,240 g ai ha^{-1}), but injured plants gradually recovered during the course of this experiment. Fomesafen applied to soils with high sand fraction and low organic matter may have more possibility to cause injury and reduce crop growth due to greater presence in soil solution than adsorbed to soil and OC surface (Li et al. Unpublished data). However, the benefits that fomesafen provides to cotton grower in controlling resistant Palmer amaranth could far exceed the injury potential of this herbicide (Main et al. 2012; Kichler et al. 2010). Meanwhile, fomesafen DT_{50} and field persistence varied significantly between Cecil sandy loam and Tifton loamy sand, which is likely due to the affinity difference of fomesafen to these soils. In areas where environmental and soil conditions are not favorable for fomesafen dissipation, growers need to be cautious about the potential carryover injury to susceptible crops and strictly follow the plant-back interval on fomesafen label.

Literature Cited

Askew SD, Wilcut JW, Cranmer JR (2002) Cotton (*Gossypium hirsutum*) and weed response to flumioxazin applied preplant and postemergence directed. Weed Technol 16:184-90

Baumann PA, Keeling JW, Morgan, GD, Smith JW (1998) Evaluation of fomesafen for weed control in Texas cotton. Proc South Weed Sci Soc 51:43-44

Bond JA, Oliver LR, Stephenson DO, IV (2006) Response of Palmer Amaranth (Amaranthus palmeri) accessions to glyphosate, fomesafen and pyrithiobac. Weed Technol 20:885-92

Cranmer, JR, Altom JV, Braun JC, Pawlak JA (2000) Valor herbicide: a new herbicide for weed control in cotton, peanuts, soybeans, and sugarcane. Proc South Weed Sci Soc 53:158

Culpepper AS, York AC, MacRae AW, Kichler J (2008) Glyphosate resistant Palmer amaranth response to weed management programs in Roundup Ready and Liberty Link Cotton. Pp. 1689 *in* Proc of the Beltwide Cotton Conferences. Natl. Cotton Counc. Am., Memphis, TN

Culpepper AS (2009) Impact of EPA's ecological risk assessment for fomesafen. Subject: re-registration of fomesafen (Docket # EPA-HQ-OPP-2006-0239) US-EPA. Washington, DC. 20460

Dotray PA, Grichar WJ, Gilbert LV (2010) Peanut tolerance and weed control following fomesafen applied at different rates and timing in Texas. Proc American Peanut Research and Education Soc 42:30-32

Everman WJ, Clewis SB, York AC, Wilcut JW (2009) Weed control and yield with flumioxazin, fomesafen and S-metolachlor systems for glufosinate-resistant cotton residual weed management. Weed Technol 23:391-97

Gardner AP, York AC, Jordan DL, Monks DW (2006) Management of annual grasses and Amaranthus spp. in glufosinate-resistant cotton. J Cotton Sci 10:328-38

Gilbert LV, Dotray PA, Prostko EP, Grichar WJ, Ferrell JA, and Jordan DL (2009) Peanut response to fomesafen. Proc American Peanut Research and Education Soc 41:28-30

Kichler JM, Culpepper AS, Chafin JE (2010) Glyphosate-resistant Palmer amaranth and cotton response to residual at-plant herbicides applied on the soil surface or preplant incorporated. Pp. 1654. *In* Proc of the Beltwide Cotton Conference, New Orleans, LA. Natl. Cotton Counc. Am., Memphis, TN

Kichler J, Culpepper AS (2012) How tillage and application timing of reflex affects palmer amaranth control and cotton injury. Pp.1543 *In* Proc of the Beltwide Cotton Conference, Orlando, FL. Natl. Cotton Counc. Am., Memphis, TN

Lunsford JN, Harrison S, Smith JD (1998) Reflex use in cotton. Proc South Weed Sci Soc 51:12-13

Main CL, Faircloth JC, Steckel LE, Culpepper AS, York AC (2012) Cotton tolerance to fomesafen applied preemergence. J Cotton Sci 16:80-87

Marshall MW (2009) Complementary herbicide programs for Palmer amaranth (*Amaranthus palmeri* S. Watts.) control in glyphosate-tolerant cotton and soybeans. Abstr Weed Sci Soc Am 49:99

Murdock EC, Keeton A (1998) Where does fomesafen fit in South Carolina cotton weed management programs? Proc South Weed Sci Soc 51:12

Oymada M, Kuwatsuka S (1988) Effects of soil properties and conditions on the degradation of three diphenylether herbicides in flooded soils. J Pestic Sci 13:99-105

Rauch BJ, Bellinder RR, Brainard DC, Lane M, Thies JE (2007) Dissipation of fomesafen in New York state soil and potential to cause carryover injury to sweet corn. Weed Technol 21:206-12

Reiling, KR, Simmions FW, Reichers DE, Steckel LE (2006) Application timing and soil pH effects on sulfentrazone phytotoxicity to two soybean (*Glycine max* (L) Merr.) cultivars. J Crop Prot 25:230-34

Schrage BW, Norsworthy JK, Smith KL, Johnson DB, Bagavathiannan MV, Riar DS (2012) Factors contributing to cotton injury from soil-applied residual herbicides. Summary of Arkansas Cotton Research 2012:102-06

Senseman SA, ed (2007) Herbicide Handbook. 9th ed. Lawrence, KS: Weed Science Society of America. pp 207

Sosnoskie LM, Kichler JM, Wallace RD, Culpepper AS (2011) Multiple resistance in Palmer amaranth to glyphosate and pyrithiobac confirmed in Georgia. Weed Sci 59:321-25

Stephenson DO, Patterson MG, Faircloth WH and Lunsford JN (2004) Weed management with fomesafen preemergence in glyphosate resistant cotton. Weed Technol 18:680-86

Syngenta Crop Protection (2014) Reflex label. Greensboro, NC. 27419. Available from http://www.cdms.net/LDat/ld6BJ002.pdf

Taylor-Lovell, S, Wax LM, Nelson R (2001) Phytotoxic response and yield of soybean (*Glycine max*) varieties treated with sulfentrazone or flumioxazin. Weed Technol 15:95-102

Troxler SC, Askew SD, Wilcut JW, Smith WD, Paulsgrove MD (2002) Clomazone, fomesafen and bromoxynil systems for bromoxynil-resistant cotton (*Gossypium hirsutum*). Weed Technol 16:838-44

USDA-NASS (2010) Agricultural chemical use program. Available from: http://www.nass.usda.gov/Surveys/Guide_to_NASS_Surveys/Chemical_Use/index.asp

Weber JB (1993) Ionization and sorption of fomesafen and atrazine by soils and soil constituents. Pestic Sci 39:31-38

Wilcut, JW, Askew SD, Price AJ, Scott GH, Cranmer J. (2000) Valor: new weed management option for cotton. Proc South Weed Sci Soc 53:159–60

Wise AM, Grey TL, Prostko EP, Vencill WK, Webster TM (2009) Establishing the geographic distribution level of acetolactate synthase resistance of Palmer amaranth (*Amaranthus palmeri*) accessions in Georgia. Weed Technol 23:214-20

Ying G, Williams B (2000). Dissipation of herbicides in soil and grapes in a South Australian vineyard. Agric Ecos Environm 78:283-89

Table 5. Locations, planting and harvesting dates and soil information of field cotton trials [a]

Location	Planting date	Harvesting date	Soil texture	pH	OM%	Sand	Silt	Clay
Athens	May 17th	Nov 22th	Cecil sandy loam [b]	5.49	2.08	71.9	12.0	16.1
Plains	May 20th	Nov 21st	Greenville sandy clay loam [c]	5.56	3.8	59.8	10.1	30.1
Ty Ty	May 1st	Oct 31st	Tifton loamy sand [d]	5.63	0.96	89.9	6.0	4.1

[a] Soil information was provided by University of Georgia Soil Testing Laboratory. Athens GA.

[b] Fine, kaolinitic, thermic Typic Kanhapludults.

[c] Fine, kaolinitic, thermic Rhodic Kandiudults.

[d] Fine-loamy, kaolinitic, thermic Plinthic Kandiudults.

Table 6. Parameter estimates of greenhouse cotton height [a]

Soil type	b_0 ± SEM	b_1 ± SEM [b]	GR_{50} [c] (g ai ha^{-1})	F value	P value
Cecil sandy loam	100.3 ± 5.0	4.00×10^{-4} ± 8.12×10^{-5} a	1733	31.00	0.0026
Greenville sandy clay loam	110.2 ± 3.5	3.29×10^{-6} ± 3.23×10^{-5}	NA	0.01	0.9227
Tifton loamy sand	94.6 ± 3.6	2.00×10^{-4}± 5.35×10^{-5} b	NA	27.97	0.0032

[a] Two-parameter exponential decay model $f(x) = b_0 e^{-b1(x)}$ was used for regression. SEM = standard error of the mean.

[b] Means followed by the same letter were not significant at 0.05 level using LSD separation.

[c]. GR_{50}: The herbicide rate causing 50% of growth reduction. In this study, 50% or greater height reduction was not observed in Greenville sandy clay loam and Tifton loamy sand at any rates evaluated.

Table 7. Parameter estimates of greenhouse cotton dry weight [a]

Soil type	$b_0 \pm$ SEM	$b_1 \pm$ SEM [b]	GR_{50} [c] (g ai ha^{-1})	F value	P value
Cecil sandy loam	95.6 ± 3.0	$6.00\times10^{-4} \pm 7.0\times10^{-5}$ a	1155	147.50	< 0.0001
Greenville sandy clay loam	107.1 ± 5.3	$4.26\times10^{-5} \pm 5.36\times10^{-5}$	NA	0.65	0.4573
Tifton loamy sand	89.3 ± 4.6	$3.00\times10^{-4} \pm 8.18\times10^{-5}$ b	NA	21.51	0.0056

[a] Two-parameter exponential decay model $f(x) = b_0 e^{-b1(x)}$ was used for regression. SEM = standard error of the mean

[b] Means followed by the same letter were not significant at 0.05 level using LSD separation.

[c]. GR_{50}: The herbicide rate causing 50% of growth reduction. In this study, 50% or greater height reduction was not observed in Greenville sandy clay loam and Tifton loamy sand at any rates evaluated.

Table 8. Field cotton stand count and height as affected by fomesafen [a]

Treatment	Stand Count [b]			Height [c]		
(g ai $ha^{-1)}$	Athens	Plains	Ty Ty	Athens	Plains	Ty Ty
	% of NTC					
0	100a	100a	100a	100cd	100c	100ab
70	88a	128a	110a	101cd	110ab	97abc
140	93a	128a	85a	119ab	115a	95bc
280	101a	130a	110a	115ab	116a	98ab
560	100a	100a	100a	110bc	105bc	106ab
1120	95a	106a	87a	96d	103bc	83c
2240	76b	61b	48b	97d	76d	59d
Fomesafen + Pendimethalin [b]	99a	130a	118a	125a	104bc	110a

[a] Means followed by same letter in the same column are not significant at $\alpha = 0.05$ level by Fisher's protected LSD. Data was expressed as percentage of non-treated control (0 g ai ha^{-1} treatment in the table).

[b] Stand data was recorded 35 DAT at Athens, 43 DAT at Plains and 42 DAT at Ty Ty.

[c] Height data was taken four times from 29 to 71 DAT at all three locations and combined for analysis.

[d] Fomesafen + Pendimethalin at 280 + 924 g ai ha^{-1}.

Table 9. Cotton yield as affected by fomesafen [a]

Treatment	Cotton Yield [b]		
(g ai $ha^{-1)}$	Athens	Plains	Ty Ty
	% of NTC		
0	100a	100a	100bc
70	103a	99a	107abc
140	103a	111a	130abc
280	89a	122a	118abc
560	132a	124a	170a
1120	92a	111a	132abc
2240	98a	102a	71c
Fomesafen + Pendimethalin [b]	136a	115a	143ab

[a] Means followed by same letter in the same column are not significant at $\alpha = 0.05$ level by Fisher's protected LSD. Data was expressed as percentage of non-treated control (0 g ai ha^{-1} treatment in the table). Fixed effect treatment failed to be significant at 0.05 level at all three locations.

Table 10. Parameter estimates of fomesafen persistence in soils under field conditions [a]

Soil type	Rate (g ai ha^{-1})	b_0 ± SEM	b_1 ± SEM [b]	DT_{50} [c]	F value	P value
Cecil sandy loam	280	0.1699 ± 0.0222	0.0147 ± 0.0055 d	47	13.82	0.0059
	560	0.3987 ± 0.0234	0.0206 ± 0.0035 c	34	101.68	< 0.0001
Tifton loamy sand	280	0.1829 ± 0.0215	0.1136 ± 0.0414 b	6	60.67	< 0.0001
	560	0.3358 ± 0.0120	0.1651 ± 0.0177 a	4	667.16	< 0.0001

[a] Two-parameter exponential decay model $f(x) = b_0 e^{-b1(x)}$ was used for regression. SEM = standard error of the mean.

[b] Means followed by the same letter are not significant at 0.05 level using LSD separation.

[c]. DT_{50}: Days required for 50% herbicide dissipation.

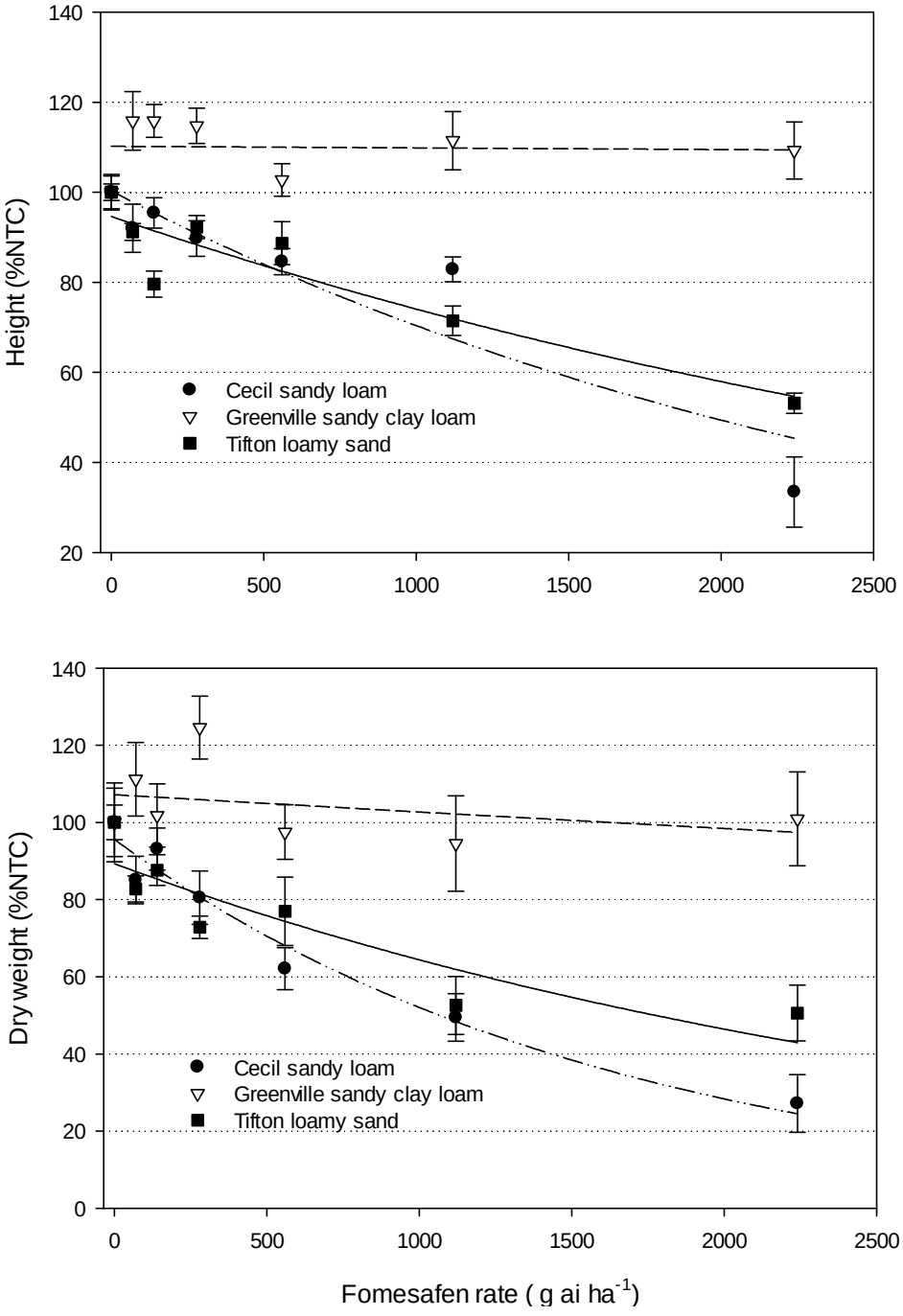

Figure 4. Effect of fomesafen rates and soil types on greenhouse cotton height and dry weight. Error bars represent standard error of each mean. Data described by two-parameter exponential decay model. Regression parameters and GR_{50} are listed in Table 2 and 3.

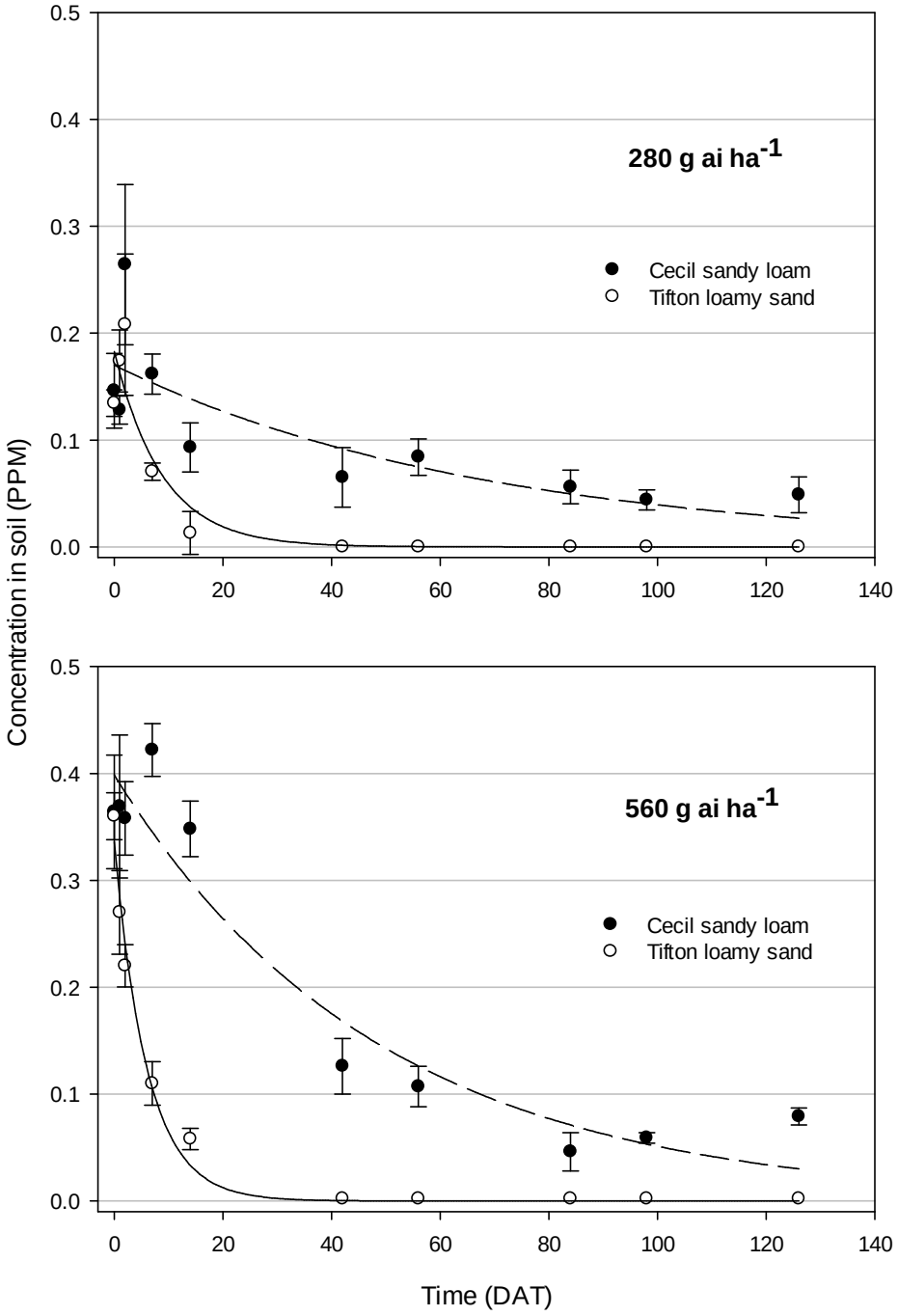

Figure 5. Fomesafen persistence in Cecil sandy loam and Tifton loamy sand as grouped by rate. Error bars represent standard error of each mean. Data described by two-parameter exponential decay model and parameter estimates are listed in table 7.

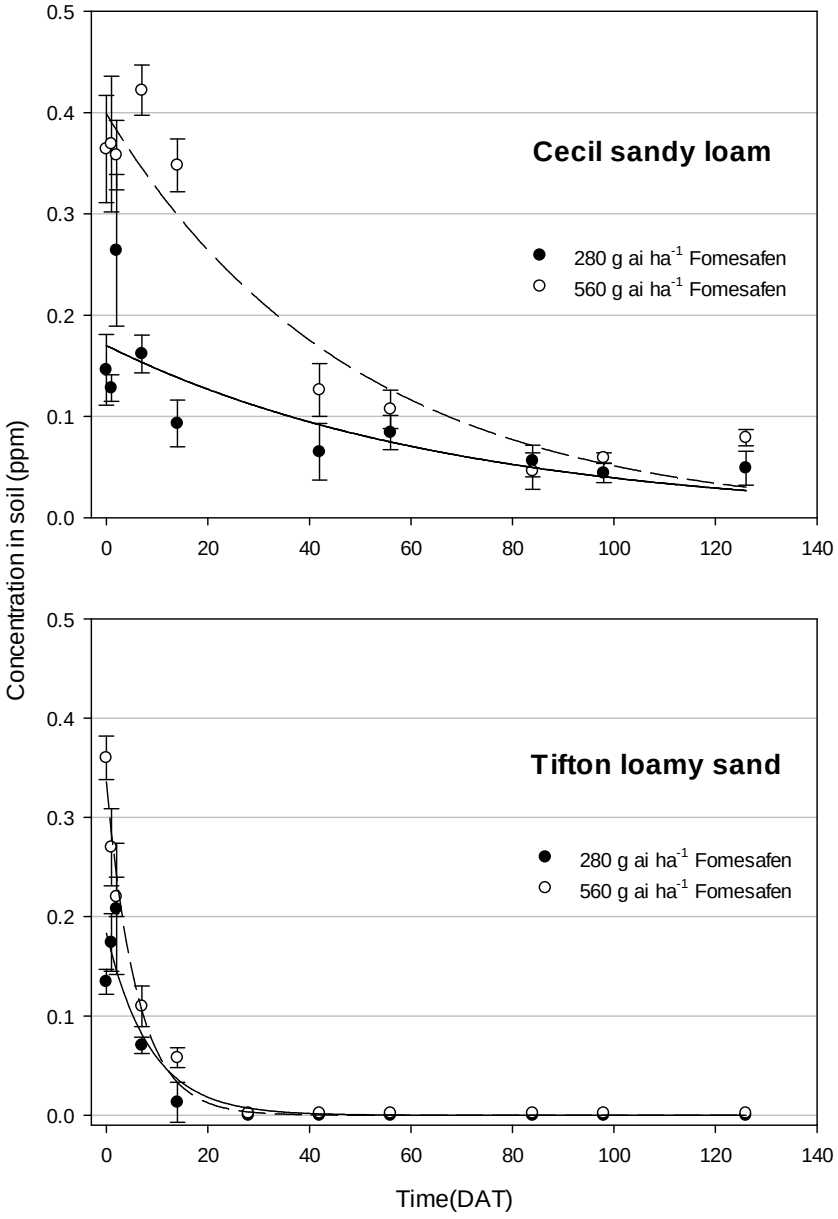

Figure 6. Fomesafen persistence in Cecil sandy loam and Tifton loamy sand as grouped by soil. Error bars represent standard error of each mean. Data described by two-parameter exponential decay model and parameter estimates are listed in table 7.

CHAPTER 4

TOLERANCE EVALUATION OF VEGETATIVELY-ESTABLISHED MISCANTHUS × GIGANTEUS TO NUMEROUS HERBICIDES [5]

[5] Xiao Li, Timothy L. Grey, Brian H. Blanchett, R. Dewey Lee, Theodore M. Webster and William K. Vencill. Published in Weed Technology 27:735-740. Reprinted here with permission of the publisher.

Tolerance evaluation of vegetatively-established *Miscanthus* × *giganteus* to numerous herbicides

Xiao Li, Timothy L. Grey, Brian H. Blanchett, R. Dewey Lee, Theodore M. Webster and William K. Vencill [6]

Miscanthus x giganteus is under consideration as a biofuel crop in the US, however there is little information on weed management for the establishment and survival of this crop. Therefore, greenhouse and field studies using ornamental pots were conducted in summer 2011 at Tifton, GA with the objective of screening potential PPI, PRE and POST emergence herbicides and herbicide combinations for *M. giganteus* when establishing from vegetative rhizomes. For the POST treatments, *M. giganteus* was established from rhizomes in 7.6 L containers in the field and treated with 27 POST herbicides to evaluate efficacy. Thifensulfuron, metsulfuron, tribenuron, chlorimuron, halosulfuron, rimsulfuron, cloransulam, pinoxaden, bentazon and metribuzin did not cause significant lower shoot height, reduced shoot dry weight and increased injury compared to non-treated control (NTC) when evaluated at 4 wk after treatment (WAT). Nicosulfuron, trifloxysulfuron, sulfometuron, clodinafop, fluazifop and pyrithiobac caused

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greatest injury, reduced plant height and dry weights as compared to the NTC. Sethoxydim, diclofop, flumioxazin, imazamox, imazapic and imazethapyr decreased plant heights or resulted in increased injury. PPI and PRE emergence treatments included 21 herbicides and herbicide combinations applied at two rates. Results indicated most treatments containing atrazine, metribuzin, pendimethalin, acetochlor, metolachlor and mesotrione did not cause significant injury and growth stunting; however, EPTC at 4.5 kg ai ha^{-1} significantly reduced height and dry weight and oxadiazon resulted in higher injury compared to NTC at both rates. These data indicated that PPI, PRE, and POST emergence herbicides can be utilized for establishment of *M. giganteus* from vegetative rhizomes. Further experiments are needed in field trials to evaluate establishment success and weed control spectrum utilizing these herbicides. Moreover, considering the invasive potential of *M. giganteus*, several POST herbicides evaluated in this study like fluazifop, pyrithiobac and sulfometuron may be viable options to control this specie if becomes invasive.

Nomenclature: Acetochlor, atrazine, bentazon, chlorimuron, clodinafop, cloransulam, diclofop, EPTC, fluazifop, flumioxazin, halosulfuron, imazamox, imazapic, imazethapyr, mesotrione, metribuzin, metolachlor, metsulfuron, nicosulfuron, oxadiazon, pendimethalin, pinoxaden, pyrithiobac, rimsulfuron, sethoxydim, sulfometuron, thifensulturon, tribenuron, trifloxysulfuron, *Miscanthus x giganteus*.

Key words: Carbon assimilation, cellulosic biofuel crop, growth reduction, herbicide injury, invasive species, weed control.

Introduction

Miscanthus is a genus of perennial rhizomatous grasses with C_4 photosynthesis, indigenous to East Asia but now found throughout a wide climate range due to its superior adaptability (Numata 1969, 1974; Greef and Deuter 1993). *Miscanthus giganteus* is a triploid with 57 somatic chromosomes, derived from a natural cross of *Miscanthus sacchariorus* (diploid) and *Miscanthus sinensis* (tetraploid). The triploidy resulted in sterility of this plant and it cannot produce viable seeds (Greef and Deuter 1993; Linde-Larson 1993).

M. giganteus has potential as a bioenergy crop due to its significant yield advantage compared to maize (*Zea mays* L.) in ethanol production and other bioenergy species, like switchgrass (*Panicum virgatum* L.) (Heaton et al. 2008). Heaton et al. (2008) reported *M. giganteus* achieved an average yield of 30 t ha^{-1} and maximum yield of 61 t ha^{-1} in a side-by-side trial with switchgrass in Illinois over 3 years, while switchgrass in this study averaged 10 t ha^{-1}. In contrast to maize grain, *M. giganteus* also has an advantage in ethanol production cost since it requires lower management (i.e. tillage, nitrogen fertilizer, pesticide) and financial input (Lewandowski et al. 2000). The energy balance ratios (output energy/input energy) of maize and *M. giaganteus* were 1.4-3.8 and 12-66, respectively (Venturi and Venturi, 2003). Net energy balance of ethanol (NEB) obtained from maize grain ranged from 10-80 GJ ha^{-1} yr^{-1} while NEB range of ethanol derived from *M. giganteus* cellulose biomass was 250-550 GJ ha^{-1} yr^{-1} (Yuan et al. 2008)

Despite many merits of *Miscanthus* spp., they have also been problematic weeds in East Asia and Japan for many years. For instance, *M. sinensis* has infested roadsides, rice (*Oryza sativa*), grassland and tree plantations at multiple areas in Japan (Sugimoto 2002, Hirata et al. 2007, Ito et al. 1982). As a foreign crop to the US, the invasive potential of *M. giganteus* needs to be

further evaluated, although the potential is relatively lower compared to its parent *M. sinensis* and some other bioenergy crop candidates such as giant reed (*Arundo donax* L.) and switchgrass due to its natural sterility and vegetative propagation (Barney and DiTomaso 2008; Quinn et al. 2010; Lewandowski et al. 2000). Previous research indicates that allopolyploidy does not guarantee continued sterility (Gray et al. 1991) and vegetative propagation is often associated with invasiveness (Daehler 1998; Kolar and Lodge 2001). Some of the ideal traits of bioenergy crops (i.e. C_4 photosynthesis pathway, high water and nitrogen use efficiency, high biomass accumulation ability, no or few pests and diseases, etc.) increased the risk of invasiveness (Raghu et al. 2006).

Grass weed control during crop establishment has been a major challenge in *M. giganteus* management as there are no herbicides registered for it as a crop in the US. It is postulated that herbicides registered for maize could be utilized for weed control in *M. giganteus* (Bullard et al. 1995; Lewandowski et al. 2000; Anderson et al. 2010). Anderson et al. (2010) tested various PRE and POST herbicide treatments on *M. giganteus* in greenhouse and field studies. They concluded that PRE and POST herbicides with mainly broadleaf activity did not injure *M. giganteus* or reduce its biomass as compared to herbicides with grass weed control activity, which caused injury rating up to 71% and reduced biomass up to 78%. Field experiments generally confirmed these results from greenhouse experiments. Their data support the previous recommendation that herbicides safe on maize are generally safe for *M. giganteus*, but with a few exceptions. Research studies compared responses of *M. giganteus* and *M. sinensis* to 18 and 10 POST herbicide treatments, respectively (Everman et al. 2011). The study results suggested that *M. giganteus* was injured by glyphosate at 840 g ae ha^{-1} (54% injury), foramsulfuron at 37 g ai ha^{-1} (32% injury), nicosulfuron at 35 g ai ha^{-1} (28% injury), and imazamox at 44 g ai ha^{-1} (10%

injury); these treatments also produced the lowest aboveground biomass values among all POST treatments. *M. sinensis* exhibited greater tolerance to POST herbicides evaluated in this study than *M. giganteus*.

Although progress has been made, growers still have limited options to selectively control grass weed species without causing excessive injury to *M. giganteus*. In Georgia, common bermudagrass (*Cynodon dactylon* (L.) Pers.), nutsedge (*Cyperus* spp.) and crabgrass (*Digitaria* spp.) were dominant weed species that caused issues during crop establishment in *M. giganteus* trials (Li, personal observation). These weeds were nearly non-controllable in the establishment year of *M. giganteus* in Southeast due to multiple factors: lack of available herbicide options, aggressive weed growth, prolonged growing season and slow canopy closure of *M. giganteus*. These weed species will likely compromise *M. giganteus* production in the Southeast. Therefore, the objectives of this research were to evaluate PPI, PRE and POST herbicides that primarily target monocot weeds and determining the most promising candidates to pursue in terms of crop safety in future large-scale field trials.

Materials and Methods

Tolerance to PPI and PRE herbicides. A greenhouse experiment was conducted at Tifton, GA from June to September 2011. There was no supplemental lighting and temperature in the greenhouse ranged from 25 to 40 C. The experiment design was completely randomized with 7 replicates and was repeated. *M. giganteus* rhizomes with excellent vigor (> 90% sprouting) were obtained locally (Lewis Taylor Farms Inc. Tifton, GA 31794). Only rhizomes exceeding 5 cm in length with at least one actively growing bud were used in the experiment. Selected rhizomes were planted horizontally 5cm deep in 1L pots filled with Tifton loamy sand (fine-loamy,

kaolinitic, thermic, Plinthic Kandiudult); 87%, 7%, and 6% sand, silt, and clay, respectively. PRE herbicide treatments included 21 different herbicides or herbicide combinations; each applied at two rates (Table 11) within 3 d of planting. All herbicide treatments were applied in a spray chamber calibrated to deliver 187 L/ha at 165 kpa using CO_2 as a propellant. The single nozzle system included a Teejet XR8002VS nozzle tip that was 45 cm above the pots. Following herbicide applications, pots received 2 cm overhead irrigation to provide soil activation of the herbicides. Due to potential EPTC volatility, it was soil incorporated by a soil tumbler immediately following application, and rhizomes were planted immediately after herbicide incorporation. Throughout the duration of the experiment, pots were maintained in trays that allowed for subsurface irrigation. Soil moisture and soil fertility (56 kg ha^{-1} N by 10-10-10 to each pot) were kept optimum for *M. giganteus* rhizome growth.

Crop shoot height and visual ratings of crop injury using a scale of 0% (no injury) to 100% (plant death) were recorded at 2 and 4 wk after treatment (WAT). Above ground plant biomass was sampled at 4 WAT by severing the shoots at the soil level and drying them prior to measuring biomass. Rhizomes were allowed to re-sprout shoots for another 4 wk and the second shoot harvest was conducted 8 WAT. Between harvests, treatments were maintained under the previously described growing conditions.

Tolerance to POST herbicides. This study was conducted in Tifton, GA between July and November 2011 in a nursery field site in pots. The experiment was arranged as a completely randomized design with 7 replicates and was repeated in time. *M. giganteus* plants were established in 7.6 L pots with pre-sprouted rhizomes that had approximately 15 cm of shoot growth. Potting soil used was amended with organic medium containing composted pine bark fines, perlite and reed sedge peat (Robin hood premium potting soil, Robin Hood Landscaping

Products, Inc. Adel, GA 31620). Pots were arranged in the field with a spacing of 90 cm by 90 cm and received daily drip irrigation throughout the experiment. There were 27 POST herbicide treatments (Table 12), applied when *M. giganteus* shoots had an average height of 40 cm. All treatments were sprayed in the field using a CO_2 backpack sprayer calibrated to deliver 187 L/ha at 4.8 km/h and 110 kpa with Teejet XR8002VS nozzle tips.

Data were collected as previously described in the PRE herbicide study, which included crop injury rating and shoot height measurements at 2 and 4 WAT; shoot dry biomass was evaluated at 4 WAT and reevaluated 8 WAT for the re-sprout shoots.

Statistical analysis. Data were subjected to ANOVA using PROC GLIMMIX (SAS Institute Inc, 2012). Dry shoot biomass, crop shoot height, and crop injury were analyzed in a mixed model containing fixed effects of herbicide treatment, herbicide rate, and their interaction, while random effects included trial repetitions and the associated interactions. All treatment means were compared to the non-treated control (NTC) using Dunnett's test at $\alpha = 0.05$ level.

Results and Discussion

Tolerance to PPI and PRE herbicides. Fixed effect of trial repetitions and interaction of trial repetitions by treatment were not significant, so data was combined for analysis and presentation. Relative to the NTC, crop injuries at 2 WAT were observed from both rates of oxadiazon (13 to 18%), the lower rate of pronamide (9%), higher rates of atrazine plus mesotrione (11%) and imazethapyr plus pendimethalin (9%). All other treatments did not injure *M. giganteus* at 2 WAT and there were no crop injury differences among any treatments at 4 WAT (Table 11). Derr (2002) determined two ornamental types of *M. sinensis* tolerated oxadiazon up to 9 kg ai ha^{-1} without causing significant shoot weight reduction. However, any rate of oxadiazon in this

experiment caused significant greater injury at 2 and 4 WAT compared to NTC, but dry weights were not significantly reduced. Another noticeable finding is EPTC, applied at 4.5 kg ai ha^{-1} with safener, caused significant reductions in *M. giganteus* shoot heights at 2 and 4 WAT as well as decreased shoot biomass at 4 WAT, relative to the NTC. EPTC is a widely used thiocarbamate herbicide registered in maize when applied 4.5 to 6.7 kg ha^{-1} PPI (Senseman 2007) and a common notion is herbicides used on maize are generally safe in *M. giganteus* production (Bullard et al. 1995; Lewandowski et al. 2000). For this study, PRE and POST experiments identified several herbicides that did not corroborate this conclusion, including EPTC, primisulfuron and nicosulfuron. Similar to EPTC, shoot heights at 4 WAT were reduced by the high rate of imazethapyr plus metolachlor but shoot heights and weights in all other treatments were not reduced relative to the NTC at 4 WAT; and there were no detectable differences in crop injury among any treatments at 4 WAT. Following the first harvest of shoot biomass at 4 WAT, new shoots emerged from rhizomes were allowed to grow for 4 more wk; at 8 WAT, there were no shoot biomass differences among any treatments to the NTC, which indicated no PRE treatment in this experiment severely injured *M. giganteus* rhizomes and prevented shoot regrowth (Data not shown). Anderson et al. (2010) suggested *M. giganteus* rhizomes grown in greenhouse demonstrated good tolerance to acetochlor, atrazine, pendimethalin and *S*-metolachlor up to 9.8, 4.5, 3.2 and 7.1 kg ai ha^{-1}, respectively. These PRE herbicides also provided similar shoot length, shoot number per plant and shoot dry weight compared to the weeded NTC under field conditions. Based on the results of this study, several PRE herbicides including atrazine, pendimethalin, acetochlor, metolachlor and mesotrione may have the potential to be applied in combinations during establishment of *M. giganteus* from rhizomes, to provide better and broader spectrum of weed control. The current recommendation

of PRE herbicide treatment on *M. giganteus* for growers in GA is atrazine plus pendimethalin, which is insufficient to control multiple aggressive weed species in late spring and early summer (Li, personal observation). Utilizing combinations including mesotrione, *S*-metolachlor and acetochlor will increase control efficacy of nutsedge and various grass species, which are the most troublesome weeds during *M. giganteus* establishment in GA. However, further field trials are still needed to evaluate the performances of these PRE combinations on weed control and crop injury.

Tolerance to POST herbicides. In the POST emergence study, there was no significant main effect of trial repetitions and associate interactions and data was therefore combined for analysis and presentation. Large differences of herbicide tolerance were observed among treatments, especially with the sulfonylureas (Table 12). The sulfonylurea herbicide treatments that were safe in terms of crop height, shoot dry weight and injury ratings at 2 and 4 WAT included thifensulfuron, tribenuron, chlorimuron, primisulfuron and halosulfuron. Sulfonylureas that caused initial injury at 2 WAT were metsulfuron and rimsulfuron, but plants grew out of the injury and stunting by 4 WAT and shoot dry weights were not affected. The most injurious sulfonylureas were nicosulfuron, trifloxysulfuron and sulfometuron, all of which reduced *M. giganteus* shoot height (≥22%), with ≥18 and ≥11% injury at 2 and 4 WAT and shoot biomass ≤57% relative to the NTC 4 WAT (100%). Nicosulfuron and trifloxysulfuron are registered in maize and sugarcane (*Saccharum officinarum* L.) as POST herbicides, respectively, but injured *M. giganteus* in this study. Four imidazolinone herbicides were generally injurious to *M. giganteus* and produced higher injury and reduced plant height compared to the NTC 2 WAT; these injuries were not reflected in the shoot biomass 4 WAT but plant height was decreased by imazamox, imazethapyr and imazapic at 4 WAT. Anderson et al. (2010) confirmed that

imazethapyr (142 g ai ha^{-1}), imazamox (22, 44, 88 g ai ha^{-1}) and imazapic (53, 106 g ai ha^{-1}) all reduced *M. giganteus* shoot dry weight under greenhouse conditions. Everman et al. (2011) concluded that rimsulfuron and halosulfuron were safe on *M. giganteus* but nicosulfuron caused injury that reduced biomass as compared to the NTC; imazethapyr and imazamox significantly reduced above-ground and below-ground biomass. These results confirmed the differential responses between sulfonylureas existed and imidazolinones could produce more injury to *M. giganteus*, therefore, caution is needed when applying imidazolinones on *M. giganteus*. For another two ALS-inhibitors in the POST study, cloransulam did not affect *M. giganteus* growth but pyrithiobac produced lowest plant height (51%) and dry weight (11%) compared to NTC (100%) among all treatments at 4 WAT; it also produced third highest injury 4 WAT (23%) among all treatments, only behind fluazifop (37%) and sulfometuron (25%).

Fluazifop caused the greatest injury at 2 and 4 WAT among all of the ACCase inhibitors; it reduced dry weight by 80% and plant height by 39% compared to the NTC (100%). Clodinafop reduced dry weight by 45% and plant height by 36% while sethoxydim, diclofop and pinoxaden treatments resulted in both higher injury and lower plant height compared to the NTC. Two protoporphyrinogen oxidase inhibitors (carfentrazone, flumioxazin) and two HPPD-inhibitors (tembotrione, topramezone) used in this study did not reduce plant height or dry weight but they caused visual injuries, especially with flumioxazin and topramezone which produced 29% and 23% injury 2 WAT as well as 17% and 14% at 4 WAT. Anderson et al. (2010) suggested tembotrione and topramezone caused significantly higher injury compared to the NTC but plant dry weight and leaf length were not affected, which correspond to the data of this study. PS II inhibitors bentazon and metribuzin were considered safe on *M. giganteus*; significant injuries

were reported at 2 WAT with both treatments but no visual injury, reduced height and dry weight were observed at 4 WAT.

A second-shoot harvest was conducted at 8 WAT to evaluate the effects of POST herbicide on shoot regrowth. Data suggested only sulfometuron and fluazifop reduced dry weight of new shoots as compared to the NTC (Table 12), indicating these two herbicides may have translocated to the underground rhizomes and affected shoot regrowth later on. This finding will be helpful for *M. giganteus* control if this specie becomes invasive in the future.

The PPI, PRE and POST emergence studies have identified various herbicides that have the potential to be utilized during and after *M. giganteus* establishment from vegetative rhizomes. Combinations of PRE herbicides plus multiple applications of POST herbicides will provide long-term weed control during the establishment, which will greatly improve the overall productivity of this crop due to its relatively slow establishment and canopy closure. However, further experiments are needed in field trials to evaluate establishment success and weed control efficacy utilizing these herbicides. Meanwhile, several POST herbicides in this experiment have demonstrated the potential to control *M. giganteus* and its shoot regrowth, but the control efficacy of these herbicides has not been compared with the standard glyphosate application. Therefore, research studies are currently underway to evaluate POST control efficacy of these herbicides compared to glyphosate and to identify best option for *M. giganteus* control.

Literature Cited

Anderson EK, Voigt TB, Bollero GA, Hager AG (2010) *Miscanthus* x *giganteus* response to preemergence and postemergence herbicides. Weed Technol 24:453-60

Barney JN, DiTimaso JM. (2008) Nonnative species and bioenergy: Are we cultivating the next invader? BioSci 58:64-70

Bullard MJ, Nixon PMI, Kilpatrick JB, Heath MC, Speller CS (1995) Principles of weed control in Miscanthus spp. under contrasting field conditions. pp 991–996 in Brighton Crop Protection Conference: Weeds. Proc. of an International Conference. Farnham, UK: British Crop Protection Council

Daehler C (1998) The taxonomic distribution of invasive angiosperm plants: ecological insights and comparison to agricultural weeds. Biol Conserv 84:167-80

Derr JF (2002) Tolerances of ornamental grasses to preemergence herbicides. J Environ Hort 20:161-65

Everman WJ, Lindsey AJ, Henry GM, Glaspie CF, Phillips K, McKenney C (2011) Response of *Miscanthus x giganteus* and *Miscanthus sinensis* to postemergence herbicides. Weed Technol 25:398-03

Gray AJ, Marshall DF, Raybouild AF (1991) A century of evolution in *Spartina anglica*. Adv Ecol Res 21:1-62

Greef JM, Deuter M (1993) Syntaxonomy of *Miscanthus x giganteus* GREEF et DEU. Angewandte. Botanik 67: 87-90

Heaton EA., Dohleman FG, Long SP (2008) Meeting US biofuel goals with less land: the potential of Miscanthus. GCB Bioenergy 14: 2000-14

Hirata M., Hasegawa N, Nogami K, Sonoda T (2007) Tuft, shoot and leaf dynamics in

Miscanthus sinensis in a young tree plantation under cattle grazing. Trop Grassl 41:113-28

Ito M, Ueki K, Sakamoto S (1982) Studies on the total vegetation control in railroad: 1. Major weeds and factors affecting their distribution. Weed Res Jpn 27:41-48

Kolar CS, Lodge DM (2001) Progress in invasion biology: predicting invaders. Trends Ecol Evol 16:199-04

Lewandowski, I, Clifton-Brown JC, Scurlock JMO, Huisaman W (2000) Miscanthus: European experience with a novel energy crop. Biomass Bioenerg 19:209-27

Linde-Laursen IB (1993) Cytogenetic analysis of Miscanthus `Giganteus', an interspecic hybrid. Hereditas 119:297-00

Numata M (1969) Progressive and retrogressive gradient of grassland vegetation measured by degree of succession-Ecological judgment of grassland condition and trend IV. Vegetatio 19:96-127

Numata M. ed. (1974) Grassland vegetation. The ora and vegetation of Japan. Tokyo: Elsevier: pp. 125-47

Raghu S, Anderson RC, Daehler CC, Davis AS, Wiedenmann RN, Simberloff D (2006) Adding biofuels to the invasive species fire? Science 313:1742

Quinn LD, Allen DJ, Stewart JR (2010) Invasiveness potential of *Miscanthus sinensis*: implications for bioenergy production in the United States. GCB Bioenergy 2:310-20

SAS Institute, Inc. 2012. SAS/STAT® 9.3 User's guide, Second Edition. http://support.sas.com/documentation/cdl/en/statug/63033/HTML/default/viewer.htm#statug_glimmix_a0000001405.htm

Senseman SA, ed (2007) Herbicide Handbook. 9th ed. Lawrence, KS: Weed Science Society of America. pp 308-09

Sugimoto Y (2002) Forest-pastoral systems – studies in Morotuka village, Miyazaki prefecture. Grassl Sci 47:644-51

Venturi P, Venturi G (2003) Analysis of energy comparison for crops in European agricultural systems. Biomass Bioenerg 25:235-55

Yuan JS, Tiller KH, Al-Ahmad H, Stewart NR, Stewart Jr CN (2008) Plants to power: bioenergy to fuel the future. Trends Plant Sci 13:421-29

Table 11. Responses of *M. giganteus* to 21 different PPI and PRE herbicides or herbicide combinations applied at two rates[a]

Treatment	Rate	Height	Injury	Height	Shoot dry
	g ai/ha	2 WAT[b]	2 WAT[c]	4 WAT	weight 4 WAT
		% of nontreated control			
Ethalfluralin	630	121	0	148	146
	1260	185	0	169	150
Oryzalin	2239	207	0	174	214
	4478	106	5	109	82
Trifluralin	560	116	0	94	79
	1120	112	0	92	64
EPTC[d]	2239	110	6	99	111
	4478	31*	4	31*	14*
EPTC + Atrazine	2239+2239	125	5	140	146
	4478+4478	59	2	98	79
Oxadiazon	2239	132	13*	120	114
	4478	133	18*	97	61
Imazethapyr	70	115	8	90	71
	140	119	7	118	96
Flufenacet + Metribuzin	123+31	184	0	172	139
	246+62	183	2	142	129
Metribuzin	280	122	0	96	71
	560	233	5	177	157
Pronamide	1119	175	9*	143	175
	2238	159	2	136	146
Atrazine + Pendimethalin	2239+831	154	3	155	157
	4478+1662	156	6	139	154
Atrazine + Acetochlor	2239+1343	123	7	133	161
	4478+2686	153	2	125	146
Atrazine + Metolachlor	2239+1422	116	3	106	100
	4478+2844	109	3	99	86
Atrazine + Mesotrione	2239+105	154	6	123	93
	4478+210	99	11*	104	114
Atrazine + Imazethapyr	2239+70	119	4	106	104
	4478+140	119	6	92	86

Mesotrione + Acetochlor	105+1343	180	3	172	157
	210+2686	133	0	133	132
Mesotrione + Pendimethalin	105+831	109	6	106	71
	210+1662	141	2	129	150
Mesotrione + Metolachlor	105+1422	102	4	90	64
	210+2844	143	3	164	182
Mesotrione + Imazethapyr	105+70	116	2	97	96
	210+140	93	4	74	54
Imazethapyr + Metolachlor	70+1422	88	2	64	89
	140+2844	65	7	60*	61
Imazethapyr + Pendimethalin	70+831	79	4	74	79
	140+1662	112	9*	109	93
NTC[e]		100	0	100	100

a. * indicates significant lower value compared to the non treated control (NTC). The value shown in the table is the ratio of treatment mean versus the corresponded mean of NTC. Injury at harvest data is not shown because no treatment was significant different compared to the NTC.

b. WAT= Weeks after treatment.

c. Injury rating varies from 0% (no injury) to 100% (complete death).

d. Due to volatility of EPTC, treatments that included it were mechanically incorporated into soil.

e. Original mean values for NTC were 8.1 cm, 0%, 15.9 cm and 0.28 g for height 2 WAT, injury 2 WAT, height 4 WAT and shoot dry weight 4 WAT, respectively.

Table 12. *M. giganteus* shoot dry weight, height and injury affected by 27 POST herbicides [a]

Treatment	Rate	Height	Injury	Height	Injury	Shoot dry	Shoot
	g ai/ha	2 WAT [b]	2 WAT [c]	4 WAT	4 WAT [c]	weight 4 WAT	regrowth 8 WAT
		——————% of nontreated control——————					
Thifensulfuron [d]	4	98	6	107	1	108	109
Metsulfuron [d]	4	87*	9*	106	2	123	116
Tribenuron [d]	18	98	4	107	0	113	113
Chlorimuron [d]	9	93	4	100	0	93	108
Nicosulfuron [d]	35	78*	18*	65*	11*	57*	70
Primisulfuron [d]	40	70	7	89	4	87	73
Halosulfuron [d]	35	107	2	115	0	139	108
Rimsulfuron [d]	35	74*	10*	89	4	85	76
Trifloxysulfuron [d]	16	72*	30*	59*	21*	33*	65
Sulfometuron [d]	105	72*	30*	57*	25*	19*	51*
Imazamox [d]	79	67*	10*	73*	5	75	73
Imazethapyr [d]	70	69*	8	79*	5	84	77
Imazaquin [d]	137	72*	11*	90	6	90	87
Imazapic [d]	67	72*	10	70*	3	77	72
Pyrithiobac [d]	107	67*	15*	51*	23*	11*	83
Cloransulam [e]	44	81*	8	95	4	95	86
Sethoxydim [f]	315	72*	9*	82*	7	73	83
Clodinafop [g]	70	70	34	64*	18	45*	61
Diclofop [f]	1119	80*	13*	88*	10*	75	84
Fluazifop [f]	210	76*	48*	61*	37*	20*	39*
Pinoxaden [h]	60	85*	9*	102	4	118	100
Carfentrazone [f]	18	96	13*	107	4	108	113
Flumioxazin [d]	107	87	29*	89	17*	70	76
Tembotrione [g]	92	100	15*	103	12*	84	118
Topramezone [i]	18	98	23*	98	14*	96	113
Bentazon [j]	1119	98	9*	100	6	98	98
Metribuzin [d]	390	94	10*	94	3	90	104
NTC [k]		100	0	100	0	100	100

a. * indicates significant lower value compared to the non treated control. The value shown in the table is the ratio of treatment mean versus the corresponded mean of NTC.

b. WAT = Weeks after treatment.

c. Injury rating varies from 0% (no injury) to 100% (complete death).

d. Sprayed with adjuvant NIS 0.25% v/v.

e. Sprayed with NIS 0.25% v/v + 2.24 kg ha^{-1} AMS.

f. Sprayed with COC 1% v/v.

g. Sprayed with MSO 0.25% v/v.

h. Built in adjuvant.

i. Sprayed with COC 1% + AMS 1.68 kg ha^{-1}.

j. Sprayed with COC 1% + AMS 2.8 kg ha^{-1}.

k. NTC = Non-treated control. Original mean values for NTC were 54 cm, 0%, 65.6 cm, 0%, 11.9 g and 9.3g for height 2 WAT, injury 2 WAT, height 4 WAT, injury 4 WAT, shoot dry weight 4 WAT and regrew shoot dry weight 8 WAT, respectively.

CHAPTER 5

PREEMERGENCE HERBICIDE SCREENING AND TOLERANCE EVALUATION FOR SEEDED-TYPE MSICANTHUS × GIGANTEUS[7]

[7] Xiao Li, Timothy L. Grey, R. Dewey Lee, William K. Vencill. To be published in Weed Technology.

Preemergence Herbicide Screening and Tolerance Evaluation for Seeded-type *Miscanthus* × *giganteus*[8]

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Miscanthus × *giganteus* has been considered as a potential bioenergy crop in the US for over a decade. However, very limited information concerning weed control during establishment from hybrid seed is available for this crop. Therefore, the objective of this research was to evaluate *M. giganteus* hybrid seed response to PRE herbicides and to assist in weed control during establishment. Herbicide screening using petri dish assay indicated that *M. giganteus* tolerated atrazine, flufenacet plus metribuzin, mesotrione, tembotrione, and acetochlor at concentrations equivalent to field use rates of 2239, 305 +76.3, 105, 92, 1343 g ai ha^{-1}, respectively; there were no reductions in seed germination as compared to the non-treated control (NTC). *S*-metolachlor, pyroxasulfone, trifluralin, ethalfluralin pendimethalin, sulfentrazone and indaziflam reduced or resulted in germination failure of *M. giganteus*. Additional studies on *M. giganteus* seed germination in response to four rates of mesotrione, acetochlor, *S*-metolachlor and atrazine were conducted in petri dishes and greenhouse. *M. giganteus* seed germination in petri dishes was not

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affected by acetochlor, atrazine and mesotrione up to 4480, 4480 and 224 g ai ha^{-1}, respectively. However, *S*- metolachlor at 1108 and 2216 g ai ha^{-1} significantly reduced *M. giganteus* germination relative to NTC. Greenhouse bioassays indicated that *M. giganteus* seed germination was mostly reduced by *S*- metolachlor, followed by mesotrione and acetochlor and was least susceptible to atrazine. Dose response bioassay in soil indicated herbicide rates causing 50% shoot dry weight reduction for *S*- metolachlor and acetochlor were 84 and 1386 g ai ha^{-1}, respectively; and rates causing 50% shoot height reduction were 291, 3209 g ai ha^{-1}, respectively, for *S*- metolachlor and acetochlor. However, those rates for atrazine and mesotrione were not achieved within the rate range evaluated in this bioassay. Results of this study indicated several PRE herbicides have the potential to be evaluated during seeded-type *M. giganteus* establishment for weed control in field trials.

Nomenclature: Acetochlor, atrazine, dinitroanilines, flufenacet, mesotrione, metolachlor, metribuzin, PPO inhibitors, tembotrione, *Miscanthus* × *giganteus*.

Key words: Herbicide tolerance, seeded-type *Miscanthus giganteus*, herbicide injury, PRE herbicides, dose-response, growth reduction.

Introduction

Miscanthus × *giganteus* has been grown in Europe as a cellulose bioenergy crop for several decades and currently under field evaluation at multiple locations in the US. Genus *Miscanthus* consists of 17 species and genetically originated from East Asia (Greef and Deuter 1993). The specific genotype used in Europe and US for bioenergy production, *Miscanthus* × *giganteus*, was introduced to Denmark from Japan in the 1930's (Greef and Deuter 1993; Lewandowski et al. 2003). *M. giganteus* is a natural cross between *Miscanthus sinensis* and *Miscanthus sacchariflorus* with 57 somatic chromosomes. Due to triploidy, *M. giganteus* seeds are sterile and therefore, reproduction in natural habitat solely relies on vegetative propagation (Lewandowski et al. 2003; Linde-Laursen 1993).

M. giganteus is a C_4 grass species with excellent nutrient use efficiency and high biomass yield. In Europe, experiments from Denmark and Germany noted yields without irrigation ranged from 10 to 25 t dry matter (DM) ha^{-1} (Lewandowski et al. 2000). Heaton et al. (2004) reported *M. giganteus* produced an average yield of 30 t DM ha^{-1} and maximum yield of 61 t DM ha^{-1} in Illinois trials over a 3-year study. Due to slow initial growth of *M. giganteus*, weed control in the first year is crucial to successful establishment and high biomass yield. Some researchers suggested herbicides registered for maize (*Zea mays* L.) are generally safe on *M. giganteus* (Lewandowski et al. 2000); however, several exceptions have been identified (Li et al. 2013). There have been no reports of plant disease or insects that significantly reduce the productivity of *M. giganteus* (Greef and Deuter 1993).

Although being an excellent bioenergy crop candidate, there are two major challenges that limit *M. giganteus* production in Europe and the US: low cold tolerance and high establishment cost due to vegetative establishment (Lewandowski et al. 2000; Lewandowski 1998). *M.*

giganteus rhizomes are killed when soil temperatures go below -3.5C, in constrast, *M. sinensis* rhizomes can tolerate soil temperature as low as -6.5C (Clifton-Brown and Lewandowski 2000). Therefore, in areas where soil temperatures can go below -3.5C, more cold-tolerant genotypes or *M. sinensis* is recommended (Clifton-Brown et al. 2001). One study suggested successful growth of *M. giganteus* within the first year of establishment, was affected by rhizome size, planting depth, rhizome storage length and storage conditions (Pyter et al. 2010). Clifton-Brown et al. (2011) studied the base temperatures below which the germination of at least 50% viable seeds ceased. They reported that the base temperature for perennial ryegrass (*Lolium perenne* L.) and maize were 3.4 and 4.5 C, respectively. However, the base temperature of ten *M. sinensis* genotypes varied from 9.7 to 11.6 C, which was higher than maize and switchgrass (*Panicum virgatum* L.).

High establishment cost is another major obstacle in *M. giganteus* production. Due to seed sterility, *M. giganteus* stands are typically established with vegetative propagated rhizomes, which are more expensive and difficult to plant as compared to seed. Lewandowski et al. (2000) estimated establishing stand cost \$3906 to \$7811 ha^{-1} with rhizomes. However, utilizing cell culture techniques and micro-propagated plants from somatic cells or meristems may significantly reduce establishment cost to \$456 ha^{-1}. Jones (2009) suggested propagation through either tissue culture or rhizomes could cost \$2586 ha^{-1} and is largely supported by EU grants of some countries. Planting cost if sowing *M. giganteus* seeds would be \$608 ha^{-1} (Clifton-Brown et al. 2011), which could reduce establishment expenses as compared to rhizome propagation. However, field establishment using seed is under development, but there are challenges given the small seed size, low nutrient reserves, and the high temperature and moisture requirements during germination. *M. giganteus* seed require optimum germination

conditions for successful field establishment, weed control is essential as this species competes poorly against weeds during the first year of growth (Greef and Deuter 1993; Anderson et al. 2010). Some herbicides have been evaluated and recommended for *M. giganteus* rhizomes but none have been selected for seeded-type *M. giganteu*s (Anderson et al. 2010; Li et al. 2013). Therefore, the objective of this study was to evaluate and identify PRE herbicides which have the potential to be safely used on *M. giganteus* when establishing from hybrid seeds using bioassay methods.

Material and Methods

PRE Herbicide screening. Studies were conducted in the herbicide plant physiology lab of University of Georgia from March to June, 2012. The experiment was a randomized complete design with 5 replications (petri dishes) for each herbicide treatment, and repeated twice. The effects of 13 PRE herbicides (Table 13) on *M. giganteus* seed germination were evaluated in plastic disposable petri dishes (100 ×15 mm, Fisherband) using similar setting as described by Voigt and Tischler (1996) and Emmerich and Hardegree (1991). Hybrid *M. giganteus* seeds (cultivar MX-45) were acquired from Mendel Biotechnology (Hayward, CA. 94545-3720) and were disinfected with 1% sodium hypochlorite solution and 70% isopropyl alcohol prior to experimentation. Seed vigor test indicated that greater than 90% of the disinfected seed germinated on filter paper with adequate moisture. Seed mold development from pathogens during the germination experiments was less than 5%.

To initiate experiments, two layers of filter paper were placed in each petri dish to retain moisture. Ten *M. giganteus* seeds were placed on filter paper in each petri dish and 3 ml of herbicide solution was added with a pipette. Herbicide solutions were made based on the typical

field use rate of each PRE herbicide and 3 ml of herbicide solution delivered equivalent amount of active ingredient (AI) as being applied on the area of one petri dish (78.5 cm^2) by a conventional sprayer in field. Herbicide solutions were made with commercially formulated products. After addition of herbicide solution, petri dishes were sealed with parafilm to prevent filter paper from drying, and were then stored at 20 C under room light with no supplementary light for 14 d before data collection. Number of germinated seed and seed with visible green shoot in each petri dish were recorded to evaluate root and shoot growth. Germination was defined as 5mm or longer radicle emergence.

Dose response bioassay. Responses of *M. giganteus* seed to four rates of acetochlor, metolachlor, atrazine and mesotrione were evaluated in a laboratory using petri dishes and soil filled pots in a greenhouse. Herbicides rates were 0.5, 1, 2 and 4X plus a non-treated control (NTC), where 1X rates for acetochlor, *S*-metolachlor, atrazine and mesotrione were 1120, 504, 1120 and 56 g ai ha^{-1}, respectively. Seed were germinated in petri dishes using the same procedures described for the PRE herbicide screening experiments.

For greenhouse dose response bioassay, ten *M. giganteus* seeds were planted at 1 cm depth in 0.26 L plastic cups filled with Cecil sandy loam (clayey, kaolinitic, thermic Typic Kanhapludult). Experiments were randomized complete designs with 5 replications (plastic cups), and repeated twice. Treatments were same as petri dish dose response bioassay and sprayed in a compressed air propelled spray chamber immediately after planting; nozzle tip was Teejet XR 8003VK and spray rate was 183 L ha^{-1} at 207 kpa pressure. After spray application, 1.25 cm equivalent of irrigation was applied to each cup to activate herbicides and pots were then kept in a greenhouse (30/20C day and night, 16 hr of photoperiod). Plants were irrigated properly and fertility optimally maintained with biweekly application of Miracle Grow (The

Scotts company, LLC. Marysville, OH. 43041). No pest damage or plant pathogens were observed throughout the experiments. Data included seedling height and shoot biomass collected 60 d after treatment.

Data analysis. Data for repeated experiments was combined for analysis since trial repetition was not significant. PRE herbicide screening and petri dish dose response data were subjected to SAS PROC GLIMMIX procedure. Mixed models used to analyze PRE herbicide screening data included fixed effect of herbicide treatment and random effect of trial repetition. All treatment means were compared to the non-treated control (NTC) with Dunnett's test at $\alpha = 0.05$ level.

Nonlinear regression model was applied to describe dose response data obtained in petri dishes and greenhouse; height and shoot dry weight were fitted into the two-parameter exponential decay equation,

$$f(x) = b_0 e^{-b1(x)} \quad (6)$$

where y is the seed germination, height or shoot biomass of *M. giganteus*; b_0 is the initial germination rate, height or shoot dry weight when rate X is zero (NTC); b_1 is the slope of regression and X is the herbicide rate (g ai ha^{-1}). Nonlinear regression analysis was performed with SAS nonlinear regression procedure (PROC NLIN); parameters and rate to cause 50% growth reduction (GR_{50}) were provided in table 3 and 4 based on SAS output. Slope of regression model was separated with LSD.

Results and Discussions

PRE Herbicide screening. Thirteen PRE herbicides with six mechanisms of action were evaluated in the herbicide screening experiment (Table 13). Two HPPD inhibitors mesotrione and tembotrione, mainly used in maize (*Zea mays* L.), did not adversely affect *M. giganteus* seed

germination and shoot growth as compared to the NTC; however the majority of shoots were chlorotic with white color attributed to both herbicides. *M. giganteus* seed germination with subsequent root and shoot growth were not affected by atrazine or flufenacet plus metribuzin. *M. giganteus* seeds exhibited different responses when treated with very long chain fatty acid inhibitory (VLCFA) herbicides: acetochlor did not affect seed germination, but seed germination and green shoot emergence were reduced by pyroxasulfone and *S*-metolachlor; *S*-metolachlor caused greater reductions in germination than other chloroacetamide tested in this study. Acetochlor, *S*-metolachlor and pyroxasulfone are herbicides that provide selective PRE control of grass weeds in maize and other small grains (Senseman 2007). Pyroxasulfone effectively controlled rigid ryegrass (*Lolium rigidum* L.) with little or no effect on wheat (*Triticum aestivum* L.) (Walsh et al. 2011). Doub et al. (1988) reported metolachlor controlled > 80% of large crabgrass (*Digitaria sanguinalis* L.) at the end of a 5 year study but incurred a weed shift to more tolerant fall panicum (*Panicum dichotomiflorum* Michx). In a New Zealand study, metolachlor was the most effective one among acetochlor, dimethenamid, alachlor and other PRE herbicide mixtures in controlling large crabgrass and bristly foxtail (*Setaria verticillata* L.); however all chloroacetamides were less effective against large seeded broom corn millet (*Panicum miliaceum* L.) than other grasses (James and Rahman 2009). Reports from the literature are consistent with the results of this research indicating that *M. giganteus* selectivity varied among VLCFA herbicides, with metolachlor caused greater injury.

The dinitroaniline herbicides trifluralin, ethalfluralin and pendimethalin caused germination failure with symptoms of short, round and swollen roots or shoots, possibly due to the fact that dinitroaniline herbicides inhibit spindle fiber production during cell mitosis, which causes cell division failure. Most seeds initiated germination process, and 38 to 56% had visible green

shoots, but growth ceased at very early stage with less than 5mm of roots emerged. At the end of germination study, most of the dinitroaniline treated seeds were molded or dead. It is well known that dinitroaniline herbicides primarily target at small-seed grasses and broadleaf weeds: tumble pigweed (*Amaranthus albus* L.) control in grain sorghum (*Sorghum bicolor* L.) was at least 99% with atrazine plus pendimethalin or trifluralin when applied early POST or late POST and Texas panicum [*Urochloa texana* (Buckley) R. Webster] control was at least 97% with the same treatments applied early POST (Grichar et al. 2005). Pendimethalin provided superior control of California brome (*Bromus carinatus* Hook. & Arn) and perennial ryegrass (*Lolium perenne* L.) (Mueller-Warrant 1999) while trifluralin was very effective against green foxtail [*Setaria viridis* (L.) P. Beauv.] (Kirkland 1996). Due to the fact that *M. giganteus* seeds are small and low in nutrient reserves and dinitroanilines mainly control small seed grass and broadleaf weeds (Vaughn and Lehnen 1991; Lewandowski et al. 2000), they may not have the potential to be used for weed control in seeded type of *M. giganteus*.

Cellulose biosynthesis inhibitor indaziflam and PROTOX inhibitors sulfentrazone and flumioxazin caused complete germination failure in all reps and no treated *M. giganteus* seed showed any sign of germination at the end of experiment. Indaziflam controlled annual bluegrass (*Poa annua* L.) in bermudagrass [*Cynodon dactylon* (L.) Pers.] turf and it also controlled smooth crabgrass [*Digitaria ischaemum* (Schreb.) Schreb. ex Muhl.] (Brosnan et al. 2012). Flumioxazin and sulfentrazone can offer various levels of activity on grass: flumioxazin control of Texas millet ranged from 36 to 76% when applied at 0.11 kg ai ha^{-1} (Grichar 2006) while sulfentrazone showed good control of green foxtail (Lyon and Wilson 2005). Results of this study suggested indaziflam, sulfentrazone and flumioxazin injured M. giganteus seedlings

and would not be weed control options during establishment. It is possible that they may have the potential to control seeds produced by other fertile *Miscanthus* species, like *M. sinensis*.

Dose response bioassay. Results of dose response bioassay in petri dishes indicated root growth and shoot emergence of *M. giganteus* seed were not negatively affected by acetochlor, mesotrione and atrazine; no significant differences were found between any rates compared to NTC (data not shown). However, as seen in the PRE herbicide screening study, *S*-metolachlor caused reduced germination with its two highest rates (Data not shown). At 1008 g ai ha^{-1}, germination decreased to 65% and further reduced to below 5% when treated with highest rate of 2016 g ai ha^{-1}; both root and shoot emergence were inhibited by *S*-metolachlor.

Dose response bioassay conducted in soil cups in greenhouse generated slightly different results, *M. giganteus* seeds were more sensitive to PRE herbicides in soil than in petri dishes. Acetochlor and mesotrione showed significant effect on seedling height, while *S*-metolachlor had most impact on seedling height among all herbicides (Figure 7). Atrazine did not have any effect on height within the rate range evaluated since the b_1 (the slope) was not significant different from 0 at α=0.05 level. GR_{50} for acetochlor and *S*-metolachlor on plant height were 3209 and 291 g ai ha^{-1}, respectively, and not available for atrazine and mesotrione in the rate range evaluated in this bioassay (Table 14).

Shoot dry weight followed the same trend as seedling height: *S*-metolachlor was the most injurious herbicide to *M. giganteus* seeds (Figure 8). Most of the seeds treated with 1008 and 2016 g ai ha^{-1} of *S*-metolachlor did not germinated by the end of 60 d experiment. Shoot dry weights were significantly reduced by any rate of *S*-metolachlor and GR_{50} was 84 g ai ha^{-1}. Moreover, acetochlor, atrazine and mesotrione all affected shoot biomass within the rates evaluated (Table 15). GR_{50} for acetochlor on shoot biomass was 1386 g ai $ha^{-1}$and was not

applicable for atrazine and mesotrione within the range studied. Slope comparisons revealed that *M. giganteus* seedling biomass were most responsive to *S*-metolachlor when rate increased; followed by mesotrione and acetochlor, with least response to atrazine.

Overall, propagating *M. giganteus* with seeds may significantly reduce production cost and results of this study suggested several PRE herbicides did not significantly inhibit seed germination and seedling establishment; they have the potential for evaluation in large scale field trials. Nevertheless, what requires special attention is the different response to PRE herbicides between *M. giganteus* seeds and rhizomes. Up to present, various PRE and POST herbicides have been screened for *M. giganteus* when propagating with rhizomes: Anderson et al. (2010) found *M. giganteus* rhizomes tolerated acetochlor and *S*-metolachlor up to 9,788 and 7,140 g ai ha^{-1}; rhizomes also showed good tolerance to high use rates of atrazine and pendimethalin when evaluated under greenhouse and field conditions. Li et al. (2013) studied the responses of *M. giganteus* rhizomes to 21 PRE herbicides and herbicide combinations with 8 mechanism of action: only EPTC at 4,478 g ai ha^{-1} significantly reduced shoot dry weight 1 month after application. These study results indicated better tolerance of rhizomes to PRE herbicides compared to seeds, possibly due to greater nutrient reserves and faster shoot and root growth. In the greenhouse dose response bioassay of this study, average shoot height of plants propagated by seeds was 17.5 cm 60 d after planting, but the shoots emerged from rhizomes which planted at the same time averaged 100 cm under the same greenhouse conditions. Differences in establishment speed may explain the response variance to herbicides between rhizome and seed. Similar to *M. giganteus*, johnsongrass (*Sorghum halepense* L.) is a perennial rhizomatous grass but it has the ability to produce viable seeds. Rosales-Robles et al. (1999) evaluated the influence of growth stage, herbicide rate and establishment method on POST johnsongrass

control. Johnsongrass plants established by rhizomes and seeds were sprayed with three rates of nicosulfuron, primisulfuron, fluazifop and clethodim at four growth stages. Their results suggested rhizome plants grew faster than seedling plants and for all four herbicides, rhizome plants required higher rate than seedling plants to reach ≥ 90% control at each growth stage, likely due to bigger size and more nutrient reserves.

Therefore, based on the results of this study and previous research, herbicide recommendations for rhizomes should not be inferred directly to seeds due to lower tolerance to PRE herbicides, otherwise severe injury or complete germination failure may occur.

Literature Cited

Anderson EK, Voigt TB, Bollero GA, Hager AG (2010) *Miscanthus* x *giganteus* response to preemergence and postemergence herbicides. Weed Technol 24:453-60

Brosnan JT, Breeden GK, McCullough PE, Henry GE (2012) PRE and POST control of annual bluegrass (*Poa annua*) with indaziflam. Weed Technol 26:48-53

Clifton-Brown JC, Lewandowski I (2000) Winter frost tolerance of juvenile *Miscanthus* plantations: studies on five genotypes at four European sites. New Phytol 148:287–94

Clifton-Brown JC, Lewandowski I, Andersson B, Basch B, Christian DG, Bonderup-Kjeldsen J, Jørgensen U, Mortensen J, Riche AB, Schwarz KU, Tayebi K, Teixeira F (2001) Performance of 15 *Miscanthus* Genotypes at Five Sites in Europe. Agronomy J 93:1013–19

Clifton-Brown J, Robson P, Sanderson R, Hastings A, Valentine J, Donnison I (2011) Thermal requirments for seed germination in Miscanthus compared with switchgrass (*Panicum virgatum*), reed canary grass (*Phalaris arundinaceae*), maize (*Zea mays*) and perennial ryegrass (*Lolium perenne*). GCB bioenergy 3:375-86

Doub JP, Wilson HP, Hines TE, Hatzios KK (1988) Consecutive annual applications of alachlor and metolachlor to continuous no-till corn (*Zea mays*). Weed Sci 36:340-44

Emmerich WE, Hardegree SP (1991) Seed germination in polyethylene glycol solution: effects of filter paper exclusion and water vapor loss. Crop Sci 31:454-58

Greef JM, Deuter M (1993) Syntaxonomy of *Miscanthus* × *giganteus* GREEF et DEU. Angewandte Botanik 67:87–90

Grichar WJ, Besler BA, Brewer KD (2005) Weed control and grain sorghum (*Sorghum bicolor*) response to postemergence applications of atrazine, pendimethalin and trifluralin. Weed Technol 19:999-1003

Grichar WJ (2006) Weed control and grain sorghum tolerance to flumioxazin. Crop Protect 25:174-77

Heaton E, Voigt T, Long SP (2004) A quantitative review comparing the yields for two candidate C4 perennial biomass crops in relation to nitrogen, temperature and water. Biomass Bioenerg 27:21-30

James TK, Rahman A (2009) Efficacy of Pre-emergence herbicides on three annual grass weeds in different soils. NZ Plant Protect 62:356-62

Jones JVH (2009) Testing the economic viability of energy crop production in competition with alternative land uses. Pages 402-423 *in* Proceedings of the 17th International Farm Management Congress. Bloomington/Normal, IL: International Farm Management Association

Kirkland KJ (1996) Use of incorporated and non-incorporated granular trifluralin for annual grass control in barley (*Hordeum vulgare*), wheat (*Triticum aestivum*), and canola (*Brassica napus*). Weed Technol 10:907-13

Lewandowski I (1998) Propagation method as an important factor in the growth and development of *Miscanthus* × *giganteus*. Ind. Crop Prod 8:229-45

Lewandowski I, Clifton-Brown JC, Scurlock JMO, Huisman W (2000) Miscanthus: European experience with a novel energy crop. Biomass Bioenerg 19:209–27

Lewandowski I, Scurlock JMO, Lindvall E, Christou M (2003) The development and current status of perennial rhizomatous grasses as energy crops in the US and Europe. Biomass Bioenerg 25:335-61

Li X, Grey TL, Blanchett BH, Lee RD, Webster TM, Vencill WK (2013) Tolerance evaluation of vegetatively-established *Miscanthus* × *giganteus* to numerous herbicides. Weed Technol

27:735-40

Linde-Laursen IB (1993) Cytogenetic analysis of Miscanthus ‘Giganteus’, an interspecific hybrid. Hereditas 119:297–300

Lyon DJ, Wilson RG (2005) Chemical weed control in dryland and irrigated chickpea. Weed Technol 19: 959-65

Mueller-Warrant GW (1999) Duration of control from preemergence herbicides for use in nonburned grass seed crops. Weed Technol 13:439-49

Pyter RJ, Rosales-Robels FE, Chander JM, Senseman SA, Prostko EP (1999) Influence of growth stage and herbicide rate on postemergence johnsongrass (*Sorghum halepense*) control. Weed Technol 13:525-29

Pyter RJ, Dohleman FG, Voigt TB (2010) Effects of rhizome size, depth of planting and cold storage on *Miscanthus* × *giganteus* establishment in the Midwestern USA. Biomass Bioenerg 34:1466-70

Senseman SA, ed (2007) Herbicide Handbook. 9th ed. Lawrence, KS: Weed Science Society of America.

Vaughn KC, Lehnen LP (1991) Mitotic disrupter herbicides. Weed Sci 39:450-57

Voigt PW, Tischler CR (1996) Effect of seed treatment on germination and emergence of 3 warm-season grasses. J. Range Manage 50:170-74

Walsh MJ, Fowler TM, Crowe B, Ambe T, Powles SB (2011) The potential for pyroxasulfone to selectivity control resistant and susceptible rigid ryegrass (*Lolium rigidum*) biotypes in Australian grain crop production systems. Weed Technol 25:30-37

Table 13. *M. giganteus* seed germination and shoot emergence as affected by preemergence herbicides in petri dish assay [a]

Treatment	Rate	Seed germination [b]	Shoot emergence [c]
	g ai ha^{-1}	%	
Mesotrione	105	95	93
Tembotrione	92	95	96
Atrazine	2239	90	90
Flufenacet plus Metribuzin	305 + 76	94	94
Acetochlor	1343	89	91
Metolachlor	1422	46*	33*
Pyroxasulfone	300	81*	73*
Trifluralin	560	0*	46*
Ethalfluralin	840	0*	38*
Pendimethalin	1064	0*	56*
Flumioxazin	89	0*	0*
Sulfentrazone	140	0*	0*
Indaziflam	63	0*	0*
NTC	NA	93	93

[a] Means followed by asterisk indicate significant differences compared to the NTC in the same column. Seeds were evaluated 2 weeks after herbicide treatment.

[b] Seed germination defined as 5 mm or longer radicle protrusion.

[c] Shoot emergence represented the percentage of seed in each treatment that produced visible green shoots.

Table 14. Parameters of *M. giganteus* height and GR_{50} for the PRE herbicides used in the greenhouse bioassay [a]

Herbicide	b_0	b_1 [b]	95% CI of b_1	R^2	GR_{50} [c] (g ai ha^{-1})
Acetochlor	18.7247	2.16×10^{-4} b	(1.10×10^{-4}, 3.23×10^{-4})	0.8604	3209
Atrazine [b]	16.4955	5.61×10^{-7}	(-6.00×10^{-5}, 6.40×10^{-5})	0.0003	NA
Mesotrione	17.3620	1.61×10^{-3} ab	(8.20×10^{-5}, 3.15×10^{-3})	0.9360	NA
Metolachlor	16.7519	2.38×10^{-3} a	(1.26×10^{-3}, 3.51×10^{-3})	0.9088	291

[a] Means followed with the same letter were not significant at 0.05 probability level. Data was described with a two parameter exponential decay model $f(x) = b_0 e^{-b1(x)}$.

[b] Means followed by the same letter are not significant at 0.05 level using LSD separation. Parameter B_1 (Slope) of atrazine failed to be significant at $\alpha = 0.05$ level.

[c] GR_{50}: The herbicide rate causing 50% of growth reduction. 50% or greater growth reduction was not obtained for atrazine and mesotrione at any rates evaluated in this study.

Table 15. Parameters of *M. giganteus* shoot biomass and GR_{50} for the PRE herbicides used in the greenhouse bioassay [a]

Herbicide	b_0	b_1 [b]	95% CI of b_1	R^2	GR_{50} [c] (g ai ha^{-1})
Acetochlor	0.2522	5.00×10^{-4} c	(3.78×10^{-4}, 6.21×10^{-4})	0.9747	1386
Atrazine	0.2637	1.19×10^{-4} d	(7.50×10^{-5}, 1.63×10^{-4})	0.9754	NA
Mesotrione	0.2678	2.96×10^{-3} b	(2.01×10^{-3}, 3.90×10^{-3})	0.9206	NA
Metolachlor	0.2589	8.26×10^{-3} a	(4.81×10^{-3}, 1.17×10^{-2})	0.9957	84

[a] Means followed by same letter in one column are not significant at 0.05 probability level. Data was described with a two parameter exponential decay model $f(x) = b_0 e^{-b1(x)}$.

[b] Means followed by the same letter are not significant at 0.05 level using LSD separation.

[c]. GR_{50}: The herbicide rate causing 50% of growth reduction. 50% or greater growth reduction was not obtained for atrazine and mesotrione at any rates evaluated in this study.

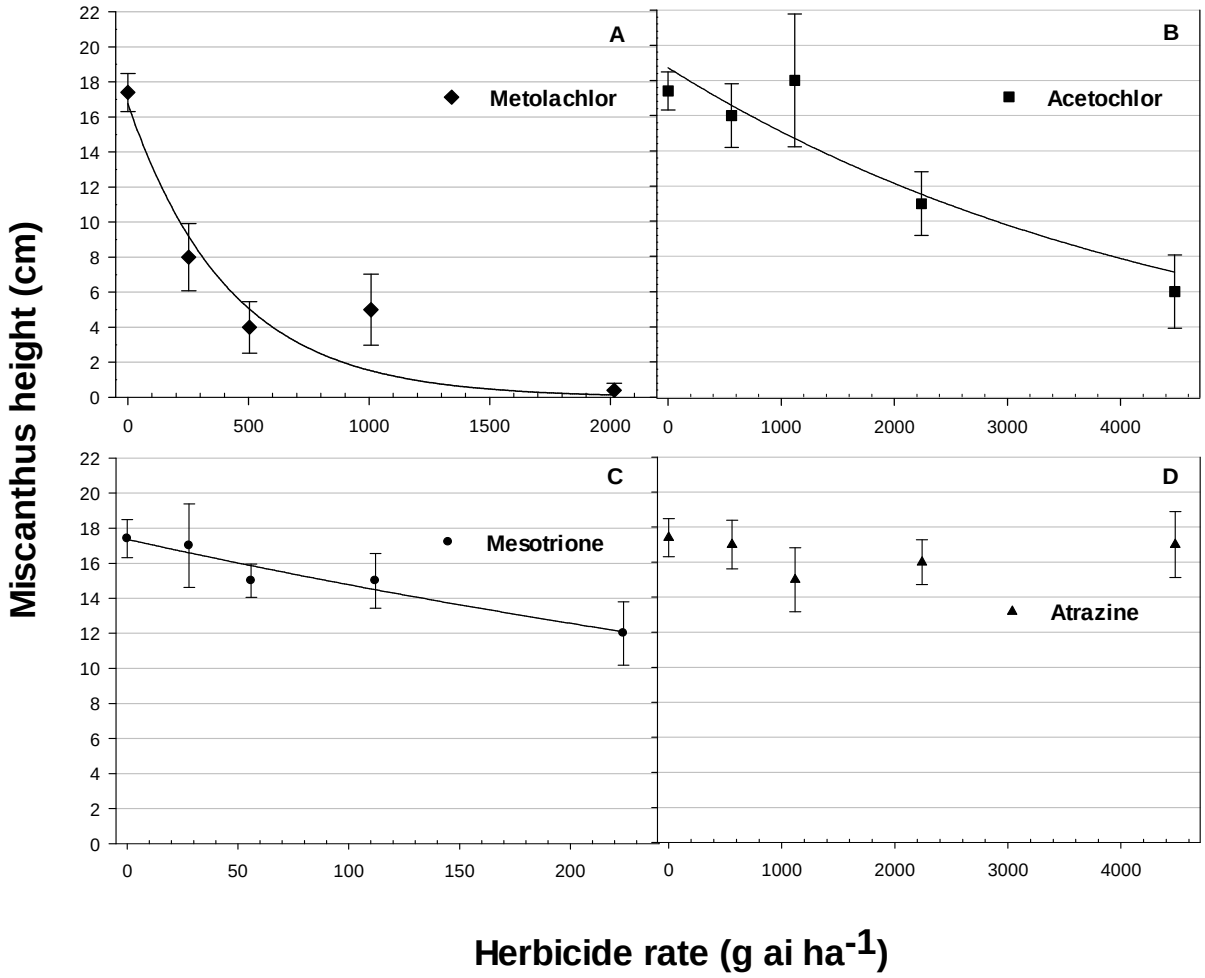

Figure 7. *M. giganteus* height affected by metolachlor (A), acetochlor (B), mesotrione (C) and atrazine (D) at various rates. Error bars represent standard error of each mean. Data were subjected to nonlinear regression and responses were described by the two-parameter exponential decay model. Regression parameters and GR_{50} are listed in Table 2.

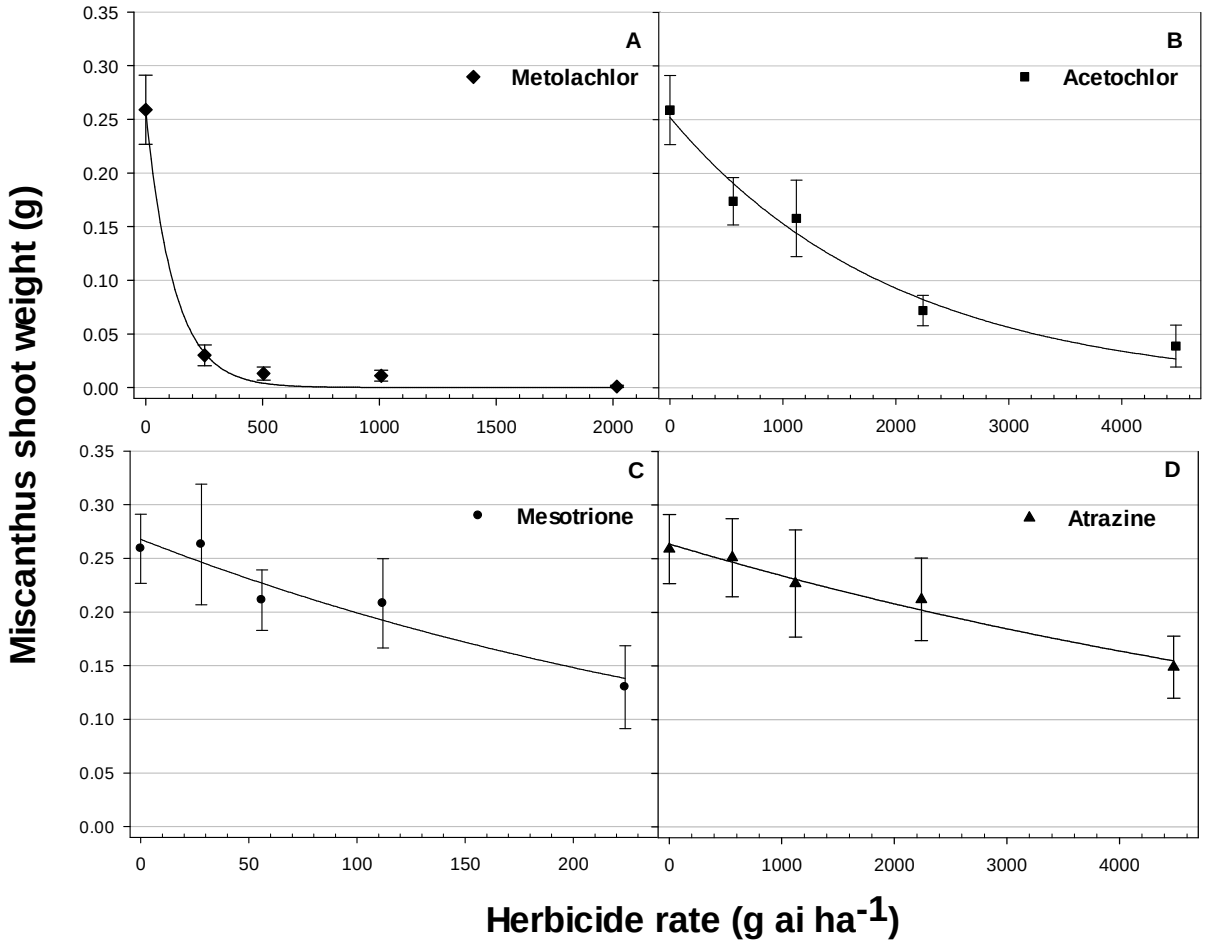

Figure 8. *M. giganteus* shoot biomass affected by metolachlor (A), acetochlor (B), mesotrione (C) and atrazine (D) at various rates. Error bars represent standard error of each mean. Data were subjected to nonlinear regression and responses were described by the two-parameter exponential decay model. Regression parameters and GR_{50} are listed in Table 3.

CHAPTER 6

GROWTH AND PHYSIOLOGICAL RESPONSES OF MISCANTHUS × GIGANTEUS TO POST HERBICIDES[9]

[9] Xiao Li, Timothy L. Grey, R. Dewey Lee and William K. Vencill. To be published in Invasive Plant Science and Management.

Growth and physiological responses of *Miscanthus* × *giganteus* to POST herbicides

Xiao Li, Timothy L. Grey, R. Dewey Lee, William K Vencill [10]

Miscanthus × *giganteus* established from rhizomes were evaluated for response to glyphosate applied alone or when used in combination with fluazifop, imazapyr, pyrithiobac and sulfometuron. In glyphosate dose-response research, rates to reduce 50% of growth (GR_{50}) were 702, 1,174 and 1,637 g ae ha^{-1} respectively, for shoot dry weight, underground biomass and regrowth of shoot dry weight (after clipping to the soil surface), respectively. Shoot regrowth was not eliminated with glyphosate rates of 4 kg ae ha^{-1} or less. Glyphosate at 10 kg ae ha^{-1} decreased 50 and 43% of *M. giganteus* chlorophyll content and photosynthesis system (PS) II efficiency (Fv/Fm) respectively, 10 d after treatment (DAT) as compared to non-treated control (NTC). Glyphosate at 2 kg ae ha^{-1} reduced chlorophyll content and Fv/Fm by 34 and 21% respectively, at 10 DAT. A single glyphosate of 1.68 kg ae ha^{-1} decreased *M. giganteus* shoot height and shoot dry weight by 8 and 17% respectively, as compared to the non-treated control (NTC) but it did not cause visual injury or reduce underground biomass and it did not prevent shoot regrowth. Two applications of glyphosate at 1.68 kg ae ha^{-1} increased control efficacy as

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compared to the single application; shoot dry weights and underground biomass were reduced by 59 and 69% compared to NTC and visual injury was 76%, and shoot regrowth was eliminated. Glyphosate 1.68 kg ae ha^{-1} plus 240 g ai ha^{-1} of fluazifop, 2240 g ai ha^{-1} imazapyr, 120 g ai ha^{-1} pyrithiobac or 120 g ai ha^{-1} sulfometuron in a single application caused greater visual injury (33 to 41%), shoot height, dry weight, and underground biomass reductions (24 to 25%, 33 to 41% and 43 to 56%, respectively) than glyphosate applied alone at 1.68 kg ae ha^{-1} and these combinations completely eliminated shoot regrowth. Two applications of combination treatments further reduced shoot dry weight and underground biomass. Reductions of chlorophyll content and Fv/Fm by glyphosate plus fluazifop, imazapyr, pyrithiobac or sulfometuron varied from 18 to 28% and 8 to 23%, respectively 10 DAT. These data suggested complete control of *M. giganteus* rhizomes and shoot regrowth would require high rates or multiple applications of glyphosate, and control efficacy could be improved by addition of POST herbicides that possessing activity against grasses.

Nomenclature: Glyphosate, fluazifop, imazapyr, pyrithiobac, sulfometuron, *Miscanthus* × *giganteus* J. M. Greef and Deuter ex Hodk. and Renvoize.

Key words: Invasive species, underground biomass, shoot regrowth, physiological response, glyphosate, POST herbicides.

Introduction

Miscanthus is a genus of perennial rhizomatous grasses with a C_4 photosynthetic pathway and the ability to yield greater biomass when compared to other bioenergy crops (Heaton et al. 2004, 2008; Lewandowski et al. 2000). *Miscanthus* originated from East Asia and has excellent adaptability to different environments, despite minimal genetic variation (Numata 1969, 1974; Greef and Deuter 1993; Barney and DiTomaso 2008). *M. giganteus* is a triploid with 57 somatic chromosomes from a natural cross of *Miscanthus sacchariorus* and *Miscanthus sinensis*. This species is considered sterile and cannot produce viable seeds (Greef and Deuter 1993; Linde-Larson 1993). Therefore, propagation of *M. giganteus* relies on vegetative propagules which increases establishment cost (Lewandowski et al. 2000; Jones 2009). Grower adoption of sterile *M. giganteus* has been slow since reproduction and storage of vegetative propagules are expensive and field planting requires specialized equipment (Heaton et al. 2010; Lewandowski et al. 2000; Smith and Barney 2014).

Although sterile varieties increase production cost, seed sterility also led to lower invasive risk to the introduced environment (Heaton et al. 2004). Sterile *M. giganteus* received a low score in the widely accepted Australian weed risk assessment (WRA) protocol and was considered “minor risk” for invading natural areas in the US (Barney and DiTomaso, 2008). Gordon et al. (2011) evaluated the invasive potential of 12 bioenergy species proposed in Florida and the US with WRA and sterile *M. giganteus* was given the lowest invasive score of -8 and -9 respectively and was considered acceptable in FL and the US (low WRA score represents low invasive risk); scores of other species ranged from 1 to 24. Matlaga and Davis (2013) suggested the growth rate of sterile *M. giganteus* population was slightly smaller than 1 (Value less than 1 means the population growth can not compensate the population lost to senescence, physical or

biological damages, etc.), which indicates its population would gradually decline over time without clonal recruitment. Moreover, no single case of escape has been reported in Europe for sterile *M. giganteus* after two decades of research and production (Lewandowski et al. 2000). It has been noted that fertile varieties of *M. giganteus* are under development and may be commercially available in the future (Smith and Barney 2014; Ross 2011). These new fertile varieties can largely decrease planting cost but also raised concerns over their invasiveness (Matlaga and Davis 2013; Quinn et al. 2011; Smith and Barney 2014)

Glyphosate has been used as a standard to eliminate bioenergy species such as napiergrass (*Pennisetum purpureum* Schum) (Cutts et al. 2011), giant reed (A*rundo donax* L.) (Spencer et al. 2008, 2011), *M. sinensis* (Omielan et al. 2012) and *M. giganteus* (Anderson et al. 2011a; Everman et al. 2011) for field crop rotation. Everman et al. (2011) noted that glyphosate applied at 0.84 kg ai ha^{-1} was the most effective treatment to reduce both aboveground and underground biomass of *M. giganteus* among 18 POST treatments evaluated. Glyphosate applied with POST herbicides that possess grass activity (i.e., imazapyr and fluazifop), can effectively control *Miscanthus* species (Speller 1993; Omielan et al. 2012). It has been suggested that other perennial grasses, such as quackgrass [*Agropyron repens* (L.) Beauv] and johnsongrass [*Sorghum halepense* (L.) Pers.] could also be controlled by glyphosate (Hamill and Zhang 1995; Parochetti et al. 1975). Spring and fall quackgrass coverage was reduced by over 80% with glyphosate at 0.56 and 0.84 kg ai ha^{-1} in soybean (*Glycine max* L.) and corn (*Zea mays* L.) (Hamill and Zhang 1995). Glenn et al. (1986) reported control of johnsongrass with fluazifop, sethoxydim and glyphosate in conventional tillage and no-tillage fields. Common reed [*Phragmites australis* (Cav.) Trin. ex Steud.] control was 80 to 100% with glyphosate plus imazapyr at 2.2 and 0.5 kg ai ha^{-1} (Rensburg 1996).

Glyphosate translocates in the symplast and accumulates in meristimatic and underground tissues (Senseman 2007). Research suggested limited amount of glyphosate applied, only 2 to 8%, was translocated to the underground rhizomes of johnsongrass (Lolas and Cobel 1980) and 7% was translocated to the rhizomes of alligatorweed [*Alternanthera philoxeroides* (Mart.) Griseb] (Bowmer et al. 1993). Moreover, Anderson et al. (2011a) reported single spring or fall application of glyphosate at 2.5 kg ae ha^{-1} did not reduce the number of shoot emerged in the following summer as compared to NTC and it did not control established *M. giganteus* in field. Multiple glyphosate applications plus tillage over several growing seasons may be needed to provide complete control. These researchers also suggested that glyphosate might not sufficiently translocate to the rhizome mass to control shoot regrowth.

Considering that insufficient information and limited research has been conducted on *M. giganteus* eradication, the objective of this experiment was to: 1) evaluate *M. giganteus* growth and physiological responses to various rates of glyphosate, and 2) to compare control efficacy of POST glyphosate applied alone and in combination with fluazifop, imazapyr, pyrithiobac and sulfometuron on *M. giganteus*.

Materials and Methods

Glyphosate dose-response bioassay. The experiment was conducted in a University of Georgia greenhouse in Athens from May to August 2012 and repeated twice. All experiments were completely randomized designs with four replications of each treatment. *M. giganteus* rhizomes were dug from field plots with shovels, cleaned and selected to ensure quality. Rhizomes were 10-15 cm long with 1 or 2 healthy buds, and rhizome viability was tested to be greater than 98%. Two rhizomes were planted 3 cm deep in Cecil sandy loam (Fine, kaolinitic, thermic Typic

Kanhapludults, pH 5.6, OM 2.6%) in 7.5 L pots. Plants were then allowed to establish in the greenhouse for 2 month, with irrigation and fertilizer applied as needed. Each pot typically had 2 to 3 actively growing shoots ranged from 130 to 150 cm tall. During establishment, temperatures were regulated to 35/25 (± 5) C diurnally with no supplemental lighting. No plant disease or insects infestations were observed during the experiment. Prior to trial initiation, *M. giganteus* plants were taken outside of greenhouse to harden for 1 wk and shoots were trimmed to 130 cm in height to ensure uniformity at treatment. Along with a NTC, glyphosate (Roundup Weathermax®, 540 g ae L^{-1}, Monsanto Co., St. Louis, MO. 63167) was applied at 0.5, 1, 2, 4, 6, 8 and 10 kg ae ha^{-1}. Treatments were sprayed in a compressed CO_2 propelled backpack sprayer with one nozzle tip (XR 8003VK flat-fan nozzles, Teejet®, Spraying Systems Co. Wheaton, IL. 60187) at 183 L ha^{-1} at 120 kpa pressure. Treated plants were kept in greenhouse for 1 month before shoots were harvested for dry weights. Then, rhizomes were given another month to regrow. Regrowth shoots and underground biomass (roots and rhizomes) in each pot were harvested 2 month after treatment for dry weights.

Physiological responses to herbicides. Leaf chlorophyll content and Fv/Fm were measured with a chlorophyll meter (SPAD-502plus. Konica Minolta, Ramsey, NJ. 07446) and OS5p modulated fluorometer (Opti-Sciences, Hudson, NH. 0.051). Previous research has demonstrated good correlation between SPAD meter readings and leaf chlorophyll content on multiple plant species (Ling et al. 2011; Uddling et al. 2007; Loh et al. 2002). Fv/Fm has been widely used as the maximum quantum yield and overall efficiency of PS II and the value of Fv/Fm varied from 0.78 to 0.84 on healthy plants (Bjorkman and Demmig 1987; Misra et al. 2012). Chlorophyll content and Fv/Fm measurements were taken on NTC and plants treated once with glyphosate alone at 2 and 10 kg ae ha^{-1}, glyphosate 1.68 kg ae ha^{-1} in combination with fluazifop at 0.22 kg ai ha^{-1},

imazapyr at 2.24 kg ai ha^{-1}, pyrithiobac at 0.11 kg ai ha^{-1} and sulfometuron at 0.11 kg ai ha^{-1}. Data were collected from 0 to 10 DAT on the first fully expanded leaf continuously. SPAD readings were measured at the midrib of leaf three times and averaged for each plant, readings of healthy non-treated plants generally varied from 0.3 to 0.4. Leaf was fully dark adapted before Fv/Fm was measured with fluorometer by wrapping a 5 cm leaf-section in aluminum foil 30 min. The initial Fv/Fm before treatments ranged from 0.75 to 0.81 when measured pre-dawn.

POST control efficacy evaluation. Experiments were conducted from July to October 2012 in University of Georgia green houses as a complete randomized design. *M. giganteus* plants were established and managed similar as used for the glyphosate dose-response bioassay. Each treatment was replicated four times and repeated twice. Glyphosate at 1.68 kg ae ha^{-1} was applied alone or in combination with fluazifop at 0.22 kg ai ha^{-1}, imazapyr at 2.24 kg ai ha^{-1}, pyrithiobac at 0.11 kg ai ha^{-1}, or sulfometuron at 0.11 kg ai ha^{-1} and an NTC was included. Nonionic surfactant at 0.25% v/v was added as needed. Treatments were applied using the same equipment and settings as the glyphosate dose-response bioassay. *M. giganteus* plants were trimmed to 130 cm in height, hardened 1 wk outside greenhouse and were randomly divided into two sets before treated. For the set treated only once, shoot height and visual injury was evaluated and aboveground shoots were harvested for dry weights 3 wk after treatment (WAT). Rhizomes were kept in greenhouse for another 3 wk and then regrowth shoot height, dry weights and underground biomass were collected 6 WAT. For the set that was treated twice, second treatment was applied 3 wk after initial treatment (WAIT), then shoot height, dry weights and visual injury were evaluated 6 WAIT. Rhizomes were allowed to grow another 3 wk to reproduce shoots and then regrowth shoot height, dry weights (if any) and underground biomass were collected 9 WAIT.

Statistical analysis. All data was converted to % of NTC prior to data analysis. Four-parameter log-logistic model was fitted to shoot dry weight, underground biomass and regenerated shoot dry weight data in glyphosate dose-response bioassay

$$f(x) = C + \frac{D-C}{1+\exp[b(\log(x)-\log(GR_{50}))]} \quad (7)$$

where C = lower limit, D = upper limit, b = reduction rate or slope and GR_{50} = the dose that produce 50% of response on the dependent variable. Leaf chlorophyll content and Fv/Fm data was described with a two-parameter exponential decay model,

$$f(x) = b_0 e^{-b1(x)} \quad (8)$$

in which b_0 is the initial value of the dependent variable when x is 0, b_1 is the slope or the decline rate of dependent variable and x is the time (DAT).

Data from POST control efficacy study was processed with PROC GLIMMIX procedure in SAS® (Version 9.3, SAS institute, Cary, NC. 27513). Trial repetition was not significant, therefore data from two repetitions were combined for analysis. Each dependent variable was analyzed with a mixed model containing fixed effect of treatment and random effect trial repetition. Means of treatments were separated by LSMEANS statement under PROC GLIMMIX procedure at $\alpha = 0.05$.

Results and Discussion

Glyphosate dose-response bioassay. *M. giganteus* shoot dry weight, underground biomass and regrowth shoot dry weight all decreased with increasing rate of glyphosate (Figure 9, 10) and parameter estimates of non-linear models were provided in Table 16. Glyphosate at 1 kg ae ha^{-1} reduced shoot dry weight and underground biomass by 40 and 26%, respectively. Glyphosate at 2 kg ae ha^{-1} decreased shoot dry weight and underground biomass by 42 and 43% respectively at

1 mo after treatment. When glyphosate rates were 4 kg ae ha^{-1} and greater, there were no differences in shoot dry weight and underground biomass reduction. Shoot regrowth did not occur with 4 kg ae ha^{-1} and higher rates. GR_{50} for shoot dry weight, underground biomass and regrowth shoot dry weight was 702, 1,174 and 1,637 g ae ha^{-1}. This indicated shoot growth was most sensitive to glyphosate application and increased rates are needed to control underground rhizomes and prevent shoot regrowth, possibly due to limited translocation to rhizomes. Anderson et al. (2011a) reported one application of glyphosate at 1.7 kg ae ha^{-1} did not control field *M. giganteus.* At that rate, glyphosate might not have adequately translocated to the entire rhizome mass to control new shoot growth effectively. Bowmer et al. (1993) studied glyphosate translocation in alligatorweed noting only 7% of the applied ^{14}C-glyphosate translocated to rhizomes and roots with up to 42% remaining in the treated leaf. Lolas and Coble (1980) reported that most of the applied ^{14}C-glyphosate remained in leaf surface (15 to 37%), treated area (6 to 10%) and rest of the treated leaf (18 to 47%) of johnsongrass, only 2 to 8% was translocated to the rhizomes. These results suggested most of the absorbed glyphosate may have remained in treated leaves and this could result in higher concentration in leaf tissue, thus a lower GR_{50} for leaf dry weight. This mechanism allowed perennial grasses to sacrifice dispensable shoots for the survival of reproductive organs (rhizomes and buds). So, controlling rhizomes and inhibiting new bud formation of perennial grasses might require increased rates or multiple glyphosate applications.

By comparing with previous studies on *M. giganteus* control, it has been noted that larger *M. giganteus* would require higher rates of glyphosate to achieve similar levels of control than on smaller seedlings. Everman et al. (2011) reported 0.84 kg ae ha^{-1} glyphosate caused 65% reduction of aboveground and belowground biomass on 40 cm *M. giganteus.* In another study,

0.4 kg ae ha^{-1} glyphosate application decreased shoot dry weight by 45% on *M. giganteus* plants varying in height from 40 to 100 cm (Anderson et al. 2011a). In this study, a 0.5 kg ae ha^{-1} glyphosate treatment resulted in 20 and 25% reduction of shoot dry weight and underground biomass respectively, likely due to the bioassay plants in this study was bigger (130 cm in height) and was established longer in greenhouse than those used in studies described above. Some researchers also suggested perennial grasses that possess large aboverground and underground biomass may 'dilute' absorbed glyphosate to a sub-lethal concentration in rhizomes, after a low dose application, thus allowing buds and rhizomes to survive glyphosate application (Hamill and Zhang, 1994). Therefore, higher rate of glyphosate or multiple glyphosate applications may be necessary to achieve lethal concentration in rhizomes and effectively control mature stand of *M. giganteus* with considerable amount of aboveground and underground biomass.

Physiological responses to herbicides. The Fv/Fm and chlorophyll content of *M. giganteus* exhibited a steady decline over time. Glyphosate at 10 kg ae ha^{-1} reduced Fv/Fm by 43% as compared to NTC (Figure 11). Glyphosate at 2 kg ae ha^{-1} reduced Fv/Fm by 21% at 10 DAT compared to NTC. Slope comparison suggested significant differences in reduction rate of Fv/Fm between 10 kg ae ha^{-1} and 2 kg ae ha^{-1} rate of glyphosate (Table 17). *M. giganteus* plants treated with one application of glyphosate plus imazapyr or pyrithiobac produced similar level of Fv/Fm reduction as 2 kg ae ha^{-1} glyphosate (Figure 12; Table 17), but not for glyphosate plus fluazifop or sulfometuron. Glyphosate at 10 kg ae ha^{-1} reduced chlorophyll content by 50% 10 DAT, as compared to 34% reduction by 2 kg ae ha^{-1} glyphosate (Figure 11). After comparing slopes of non-linear regression model, most of the combination treatment had similar reduction rate of chlorophyll content as 2 kg ae ha^{-1} glyphosate except for glyphosate plus fluazifop

(Figure 13, Table 18). Fv/Fm of *M. giganteus* showed significant decrease within 2 d by glyphosate treatment in this study, prior to any visible injury occurred. Chlorophyll fluorescence, particularly Fv/Fm, has been proved to be useful and convenient to detect physiological injury and environmental stresses prior to visible signs of injury (Percival and Fraser 2001, 2002; Percival 2004). In one study, researchers detected Fv/Fm reduction as soon as 24 hr after 0.086 and 0.86 kg ae ha^{-1} rate glyphosate applications on non-glyphosate resistant soybean (Huang et al. 2012). Meanwhile, the reduction of chlorophyll content in *M. giganteus* was in a good concurrence with the progress of leaf chlorosis and necrosis. Ketel et al. (1996) suggested low-dose application of 90, 180 and 360 g ai ha^{-1} of glyphosate resulted similar or higher chlorophyll content in common lambsquater (*Chenopodium album* L.) than the NTC and most of the treated plants survived low dose applications. They concluded that higher doses of glyphosate would cause chlorophyll breakdown, chlorosis, growth inhibition and eventually plant death. These findings agreed with the observations of this study.

POST control efficacy study. One application of glyphosate at 1.68 kg ae ha^{-1} reduced 8% shoot height and 17% dry weight compared to NTC, but did not affect underground biomass, while visual injury was not observed on glyphosate treated plants (Table 19). One application of four combination treatments produced more visual injury (59 to 64%), more reduction of shoot height (24 to 25%), shoot dry weight (48 to 51%) and underground biomass (43 to 56%) as compared to glyphosate applied alone. There were no significant differences among the control efficacy of combination treatments in any category. Shoot regrowth was also eliminated by these treatments but not glyphosate applied alone, no differences were found between glyphosate alone and NTC in shoot regrowth. When treated twice (Table 20), glyphosate decreased shoot height, dry weight and underground biomass by 34, 59 and 69% respectively, and reductions were similar to

the combination treatments except for shoot dry weight. Combination treatments constantly resulted in highest injury (83 to 94%) and lowest shoot dry weight (87 to 81% of reduction) when applied twice.

In this study, two applications of glyphosate markedly improved control efficacy as compared to one application and prevented shoot regrowth. Similar to this finding, previous reports suggested that two applications of glyphosate resulted in lower shoot numbers, height and dry weight than one application in a field *M. giganteus* control study (Anderson et al. 2011a, 2011b). Meanwhile, four POST combination treatments generated greater control as compared to glyphosate alone, which suggested glyphosate control efficacy might be increased with addition of POST herbicide with grass activity. It has been reported that imazapyr provided excellent control of common reed [*Phragmites australis* (Cav.) Trin. ex Steud.] in a container trial and 93% control in field experiments (Derr 2008). Fluazifop, sulfometuron and pyrithiobac applied at 0.21, 0.11 and 0.11 kg ai ha^{-1} produced severe injury and greatest dry weight reduction among all treatments on *M. giganteus* (Li et al. 2013). Thus glyphosate plus fluazifop, sulfometuron, pyrithiobac or imazapyr may have the potential to provide greater *M. giganteus* control than glyphosate alone.

Literature Cited

Anderson EK, Voigt TB, Bollero GA, Hager AG (2011a) *Miscanthus giganteus* response to tillage and glyphosate. Weed Technol 25:356-62

Anderson EK, Voigt TB, Bollero GA, Hager AG (2011b) Rotating a field of mature *Miscanthus* × *giganteus* to glyphosate-resistant crops. Agron J 103:1383-88

Anderson MA, Schnabel RR, Curran WS, Stout WL, Genito D, Tracy BF (2004) Switchgrass and big bluestem hay, biomass and seed yield response to fire and glyphosate treatment. Agron J 96:1688-92

Barney JN, DiTimaso JM. (2008) Nonnative species and bioenergy: Are we cultivating the next invader? BioSci 58:64-70

Bjorkman O, Demmig, B (1987) Photon yield of O2 evolution and chlorophyll fluorescence characteristics at 77 K among vascular plants of diverse origins. Planta 170:489-04

Bowmer KH, Eberbach PL, McCorkelle G (1993) Uptake and translocation of ^{14}C-glyphosate *Alternanthera philoxeroides* (Mart.) Griseb. (alligator weed) I. Rhizome concentrations required for inhibition. Weed Res 33:53-57

Cutts GS, Webster TM, Grey TL, Vencill WK, Lee RD, Tubbs RS, Anderson WF (2011) Herbicide effect on napiergrass (*Pennisetum purpureum*) control. Weed Sci 59:255-62

Derr JF (2008) Common reed (*Phragmites australis*) response to postemergence herbicides. Invasive Plant Sci Manag 1:153–57

Everman WJ, Lindsey AJ, Henry GM, Glaspie CF, Phillips K, McKenney C (2011) Response of *Miscanthus x giganteus* and *Miscanthus sinensis* to postemergence herbicides. Weed Technol

25:398-03

Glenn, S, Peregoy RS, Hook BJ, Heimer JB, Wiepke T. (1986) *Sorghum halepense* (L.) Pers. control with foliar-applied herbicides in conventional and no-tillage soybeans. Weed Res 26:245–50

Greef, JM, Deuter M (1993) Syntaxonomy of *Miscanthus x giganteus* GREEF et DEU. Angewandte Botanik 67:87-90

Gordon DR, Tancig KJ, Onderdonk DA, Gantz CA (2011) Assessing the invasive potential of biofuel species proposed for Florida and the United States using the Australian weed risk assessment. Biomass Bioenerg 35:74-79

Hamill AS, Zhang J (1995) Quackgrass control with glyphosate and SC-0224 in corn and soybean. Can J Plant Sci 75:293-99

Heaton EA, Voigt T, Long SP (2004) A quantitative review comparing the yields for two candidate C4 perennial biomass crops in relation to nitrogen, temperature and water. Biomass Bioenerg. 27:21-20

Heaton EA, Dohleman FG, Long SP (2008) Meeting US biofuel goals with less land: the potential of Miscanthus. Global Change Biol. 14:2000-14

Heaton EA, Dohleman FG, Miguez AF, Juvik JA, Lozovaya V, Widholm J, Zabotina OA, Mcisaac GF, David MB, Voigt TB, Boersma NN, Long SP (2010) Miscanthus: A Promising Biomass Crop. Adv Bot Res 56:75–137

Huang Y, Thomson SJ, Molin WT, Reddy KN, Yao H (2012) Early detection of soybean plant injury from glyphosate by measuring chlorophyll reflectance and fluorescence. J of Agric Sci 4:117-24

Jones JVH (2009) Testing the economic viability of energy crop production in competition with

alternative land uses. Proc of the 17th International Farm Management Congress. 1:402–23

Ketel DH (1996) Effect of low doses of metamitron and glyphosate on growth and chlorophyll content of common lambsquaters (*Chenopodium album*). Weed Sci 44:1-6

Lewandowski I, Clifton-Brown, Scurlock JM, Huisaman W (2000) Miscanthus: European experience with a novel energy crop. Biomass Bioenerg19:209-27

Li X, Grey TL, Blanchett BH, Lee RD, Webster TM, Vencill WK (2013) Tolerance evaluation of vegetatively established *Miscanthus* × *giganteus* to Herbicides. Weed Technol 27:735-40

Linde-Laursen IB. (1993). Cytogenetic analysis of *Miscanthus* `Giganteus', an interspecic hybrid. Hereditas 119:297-300

Ling Q, Huang W, Jarvis P (2011) Use of a SPAD-502 meter to measure leaf chlorophyll concentration in *Arabidopsis thaliana*. Photosynth Res 107:209-14

Loh FCW, Grabosky JC, Bassuk NL (2002) Using the SPAD 502 meter to assess chlorophyll and nitrogen content of Benjamin Fig and cottonwood leaves. Hort Tech 12:682-86

Lolas PC, Coble HD (1980) Translocation of 14C-glyphosate in johnsongrass (*Sorghum halepense* L. Pers.) as affected by growth stage and rhizome length. Weed Res 20:267-70

Matlaga DP, Davis AS (2013) Minimizing invasive potential of *Miscanthus* × *giganteus* grown for bioenergy: identifying demographic thresholds for population growth and spread. J Appl Ecol 50:479–87

Misra AN, Misra M, Singh R (2012) Chlorophyll fluorescence in plant biology. Pp.171 *in* Biophysics, Dr. Prof. Dr. A.N. Misra (Ed.), ISBN: 978-953-51-0376-9, InTech, Available from: http://www.intechopen.com/books/biophysics/chlorophyll-fluorescence-in-plant-biology

Numata M (1969) Progressive and retrogressive gradient of grassland vegetation measured by

degree of succession-Ecological judgment of grassland condition and trend IV. Vegetatio. 19:96-127

Numata, M. Editor. 1974. Grassland vegetation. The ora and vegetation of Japan. Tokyo: Elsevier: pp. 125-47

Omielan J, Gumm D, Witt W (2012) Evaluation of management options for control of Chinese silvergrass (*Miscanthus sinensis* Anders.) Poster, Southeast exotic pest plant council annual meeting. May 8-10, 2012. Auburn AL. Available from: http://www2.ca.uky.edu/pss/weeds/ivm/pdf/Miscanthus%20Poster%20for%20AL.pdf

Parochetti JV, Wilson HP, Burt GW (1975) Activity of glyphosate on johnsongrass. Weed Sci 23:395–400

Percival GC, Fraser GA (2001) Measurement of the salinity and freezing tolerance of Crataegus genotypes using chlorophyll fluorescence. J Arboric 27:233–45.

Percival GC, Sheriffs C (2002) Identification of drought-tolerant woody perennials using chlorophyll fluorescence. J. Arboric 28:215–24

Percival GC (2004) Evaluation of physiological tests as predictors of young tree establishment and growth. J. Arboric 30:80-91

Quinn LD, Matlaga DP, Stewart JR, Davis AS (2011) Empirical evidence of long-distance dispersal in Miscanthus sinensis and *Miscanthus* × *giganteus*. Invasive Plant Sci Manage 4:142–50

Raghu S, Anderson RC, Daehler CC, Davis AS, Wiedenmann RN, Simberloff D, Mack RN (2006) Adding biofuels to the invasive species fire? Science 313:1742

Rensburg, E. V. 1996. Reed eradication under transmission powerlines in South Africa using microlight aircraft. Pages 1095–1101. *in* Proc of the Second International Weed Control

Congress, Copenhagen, Denmark, 25–28 June 1996. Slagelse, Denmark: Department of Weed Control and Pesticide Ecology.

Rioux R, Bandeen JD, Anderson GW (1974) Effects of growth stage on translocation of glyphosate in quackgrass. Can J Plant Sci 54:397-401

Ross M (2011) New Miscanthus development possible biomass game changer? FarmWeek, May 16, 2011. pp. 9

Senseman SA, ed (2007) Herbicide Handbook. 9th ed. Lawrence, KS: Weed Science Society of America. pp 243

Speller CS (1993) Weed control in Miscanthus and other annually harvested biomass crops for energy or industrial use. pp. 671–676. *In* Brighton Crop Protection Conf., Weeds. Proc. of an Int. Conf., Brighton, UK. 22–25 Nov. 1993. British Crop Protection Council (BCPC), Farnham UK.

Spencer DF, Tan W, Liow P, Ksander GG, Whitehan LC (2008) Evaluation of glyphosate for managing giant reed (*Arundo donax*). Invasive Plant Sci Manage 1:248-54

Spencer DF, Ksander GG, Tan W, Liow PS, Whitehand LC (2011) Influence of application timing on the impact of glyphosate on giant reed (*Arundo donax* L.). J Auat plant manage 49:106-10

Uddling J, Gelang-Alfredsson J, Piikki K, Pleijel H (2007) Evaluating the relationship between leaf chlorophyll concentration and SPAD-502 chlorophyll meter readings. Phtotsynth Res 91:37-46

Table 16. Parameter estimates of *M. giganteus* shoot dry weight, underground biomass and regenerated shoot dry weight as affected by various rates of glyphosate [a]

Responsive variable	d ± SEM [b]	c ± SEM	b ± SEM	GR_{50} ± SEM [b]	F-value	P-value
Shoot dry weight	42.61 ± 3.59	100.24 ± 3.73	-3.25 ± 1.02	702 ± 130	69.29	0.0007
Underground biomass	34.63 ± 14.88	99.67 ± 4.27	-1.97 ± 0.95	1174 ± 755	49.01	0.0013
Regenerated shoot dry weight	-2.59 ± 2.23	98.31 ± 2.74	-6.46 ± 0.85	1637 ± 86	437.13	< 0.0001

[a] Four parameter log-logistic model was used to describe data. $f(x) = C + \frac{D-C}{1+\exp[b(\log(x)-\log(GR_{50}))]}$

[b] SEM = standard error of the mean.

[c] GR_{50} = Rate of glyphosate that cause 50% growth reduction.

Table 17. Parameter estimates of *M. giganteus* PSII efficiency (Fv/Fm) [a]

Treatment	Rate (g ai ha^{-1})	b_0 ± SEM [b]	b_1 ± SEM [c]	F value	P value
Glyphosate	2000	0.978 ± 0.015	0.026 ± 0.003 b	66.48	< 0.0001
Glyphosate	10000	0.976 ± 0.028	0.073 ± 0.007 a	116.19	< 0.0001
Glyphosate + Imazapyr	1680 + 2240	0.887 ± 0.028	0.028 ± 0.007 b	13.92	0.0029
Glyphosate + Fluazifop	1680 + 240	0.949 ± 0.027	0.015 ± 0.006 c	5.89	0.0320
Glyphosate + Pyrithiobac	1680 + 120	0.974 ± 0.018	0.030 ± 0.004 b	45.29	< 0.0001
Glyphosate + Sulfometuron	1680 + 120	0.969 ± 0.016	0.011 ± 0.004 c	9.24	0.0103

[a] Two-parameter exponential decay model was used to describe data. $f(x) = b_0 e^{-b1(x)}$

[b] SEM = standard error of the mean.

[c] Means followed by the same letter are not significant at 0.05 level using LSD separation.

Table 18. Parameter estimates of *M. giganteus* chlorophyll content [a]

Treatment	Rate (g ai ha^{-1})	b_0 ± SEM [b]	b_1 ± SEM [c]	F value	P value
Glyphosate	2000	0.952 ± 0.027	0.042 ± 0.006 b	44.37	< 0.0001
Glyphosate	10000	0.956 ± 0.027	0.082 ± 0.007 a	136.59	< 0.0001
Glyphosate + Imazapyr	1680 + 2240	1.003 ± 0.017	0.038 ± 0.004 b	90.49	< 0.0001
Glyphosate + Fluazifop	1680 + 240	0.893 ± 0.022	0.025 ± 0.006 c	19.48	0.0008
Glyphosate + Pyrithiobac	1680 + 120	1.004 ± 0.014	0.040 ± 0.003 b	150.55	< 0.0001
Glyphosate + Sulfometuron	1680 + 120	1.029 ± 0.017	0.038 ± 0.004 b	91.50	< 0.0001

[a] Two-parameter exponential decay model was used to describe data. $f(x) = b_0 e^{-b1(x)}$

[b] SEM = standard error of the mean

[c] Means followed by the same letter are not significant at 0.05 level using LSD separation.

Table 19. *M. giganteus* response to a single application of POST treatments [a]

Treatment	Rate (g ai ha^{-1})	Shoot height [b]	VI [c]	Shoot dw [b]	Underground biomass [d]	RSD [e]	RSH [f]
		% of NTC					
Glyphosate	1680	92b	2a	83b	88a	88a	98a
Glyphosate + Imazapyr	1680 + 2240	75c	59b	52c	57b	0b	0b
Glyphosate + Fluazifop	1680 + 240	76c	60b	51c	47b	0b	0b
Glyphosate + Pyrithiobac	1680 + 120	75c	67b	50c	44b	0b	0b
Glyphosate + Sulfometuron	1680 + 120	75c	64b	49c	44b	0b	0b
NTC	0	100a	0a	100a	100a	100a	100a

[a] Means followed by same letters in each column are not significant at 0.05 level. Results were presented as % of NTC. No injury was observed on regrowth shoots.

[b] Shoot height and dry weight was collected 3 WAT.

[c] Visual injury (VI) evaluated 3 WAT and rating varied from 0% (no injury) to 100% (complete death).

[d] Underground biomass reflected the dry weight of all underground biomass, including rhizomes and roots. Data was collected 6 WAT.

[e] RSD = Regrowth shoot dry weight. Data collected 6 WAT.

[f] RSH = Regrowth shoot height. Data collected 6 WAT.

Table 20. *M. giganteus* response to a two applications of POST treatments [a]

Treatment	Rate (g ai ha^{-1})	Shoot height [b]	VI [c]	Shoot dw [d]	Underground biomass [e]	RSD [f]	RSH [g]
		% of NTC					
Glyphosate	1680	66b	76c	41b	31b	0b	0b
Glyphosate + Imazapyr	1680 + 2240	60b	94a	15c	14b	0b	0b
Glyphosate + Fluazifop	1680 + 240	58b	83b	19c	15b	0b	0b
Glyphosate + Pyrithiobac	1680 + 120	55b	83b	18c	16b	0b	0b
Glyphosate + Sulfometuron	1680 + 120	57b	88ab	13c	14b	0b	0b
NTC	0	100a	0d	100a	100a	100a	100a

[a] Means followed by same letter in each column are not significant at 0.05 level. Results were presented as % of NTC. Second application was made 3 weeks after initial treatment (WAIT).

[b] Shoot height was measured 6 WAIT.

[c] Visual injury (VI) evaluated 6 WAIT and rating varied from 0% (no injury) to 100% (complete death).

[d] Shoot dry weight was measured 6 WAIT.

[e] Underground biomass reflected the dry weight of all underground biomass, including rhizomes and roots. Data was collected 9 WAIT.

[f] RSD = Regrowth shoot dry weight. Data collected 9 WAIT.

[g] RSH = Regrowth shoot height. Data collected 9 WAIT.

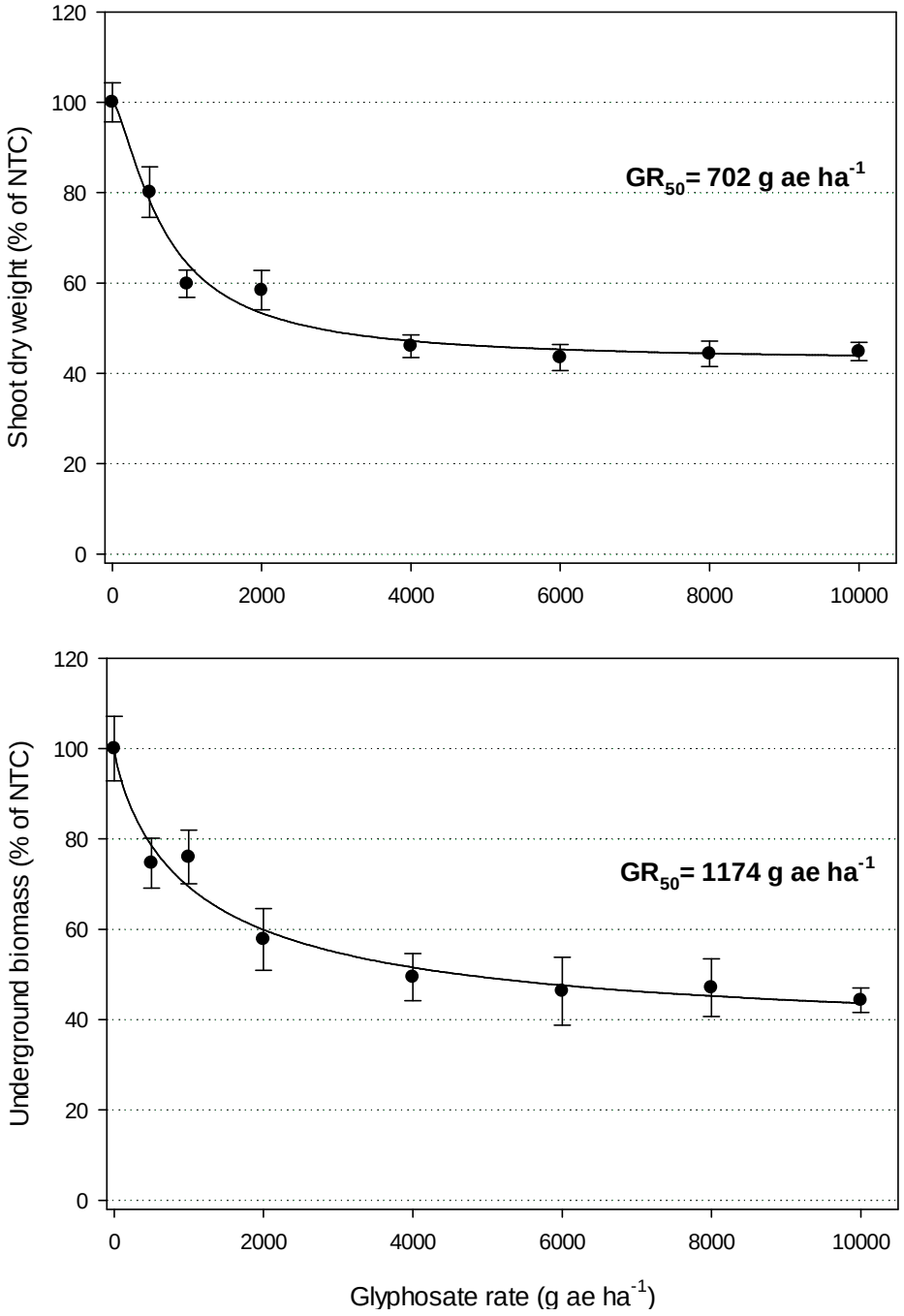

Figure 9. Response of *M. giganteus* shoot dry weight and underground biomass to various doses of glyphosate. Error bars represent standard error of each mean. Four-parameter log-logistic model was used to describe shoot and underground biomass dry weight data. Parameter estimates were given in Table 1.

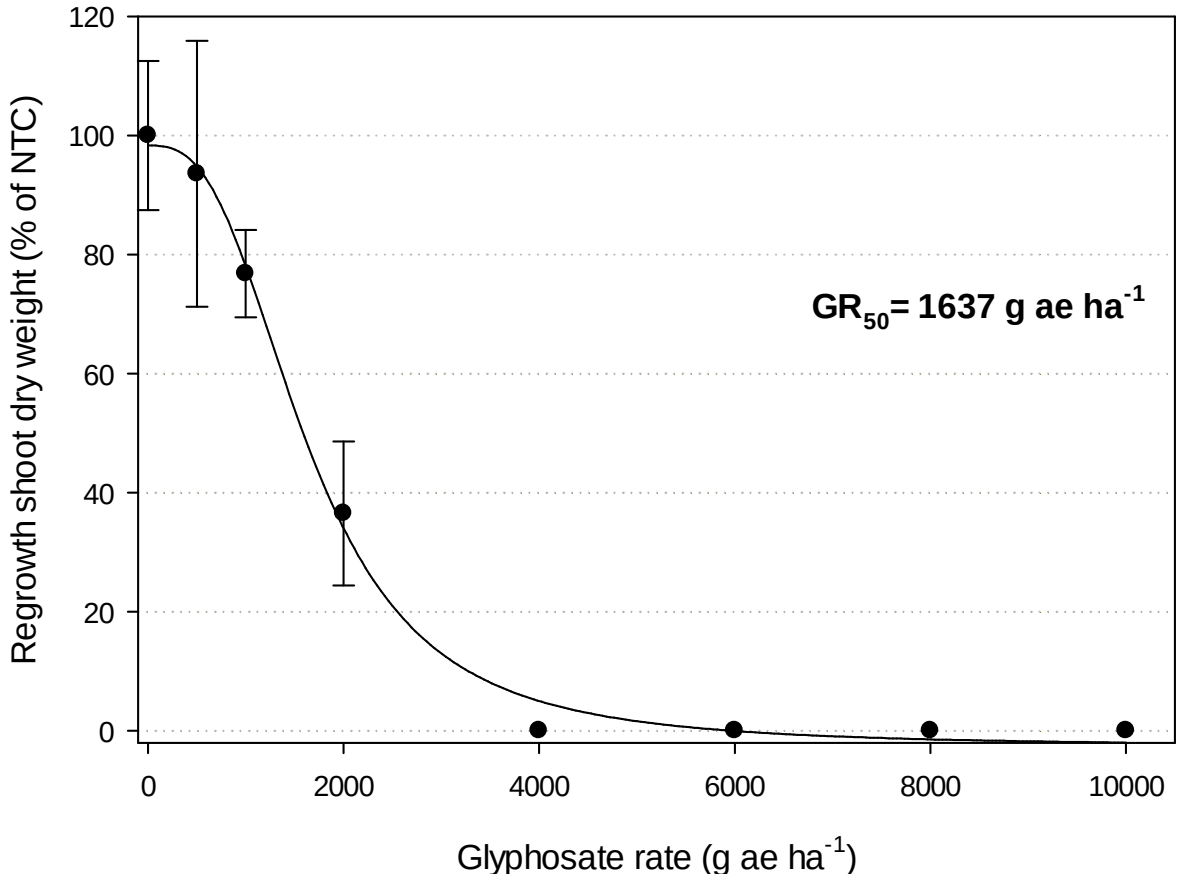

Figure 10. Response of *M. giganteus* regrowth shoot dry weight to various doses of glyphosate. Error bars represent standard error of each mean. Four parameter log-logistic model was used to describe regenerated shoot dry weight data. Parameter estimates were given in Table 1.

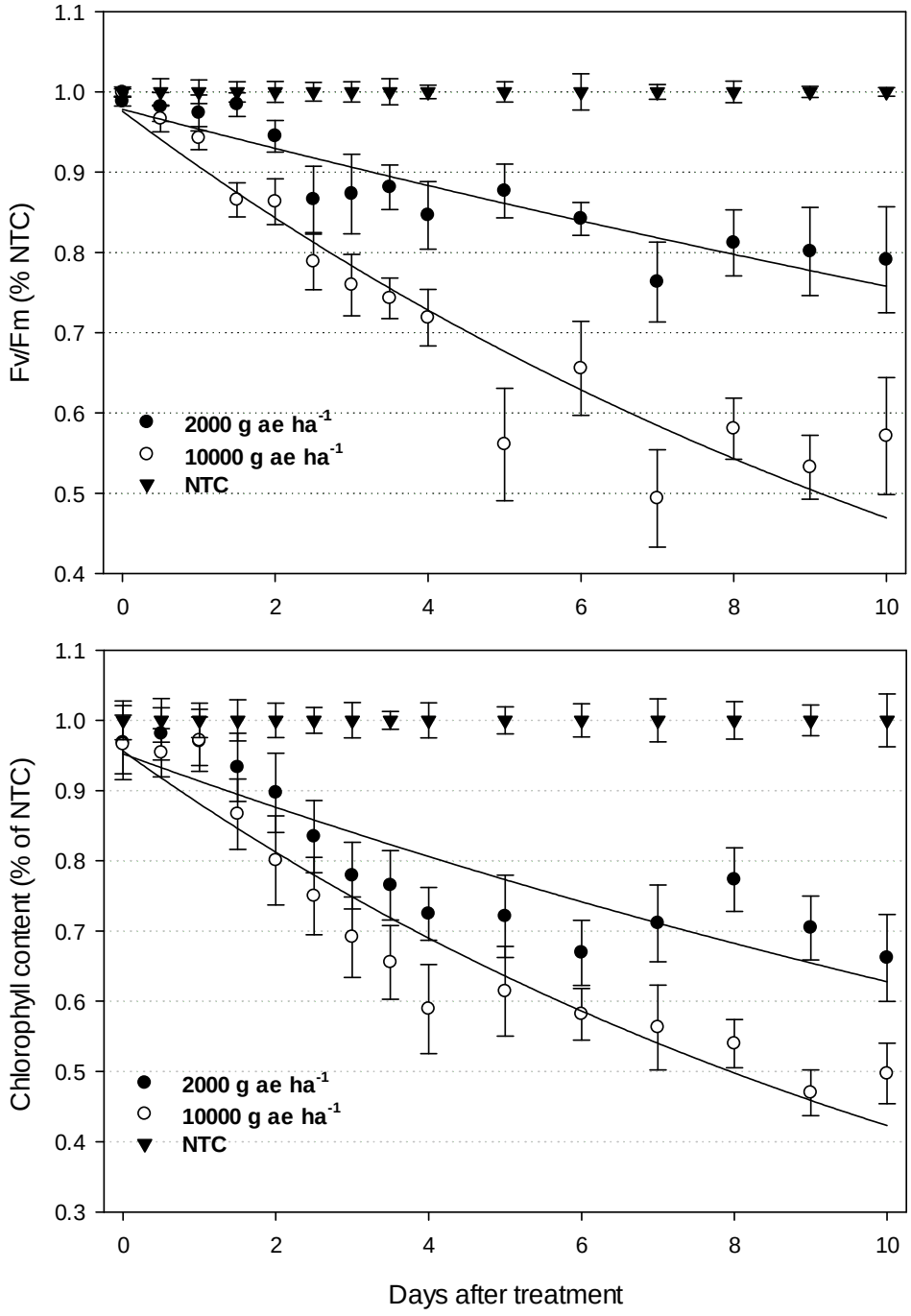

Figure 11. Response of *M. giganteus* chlorophyll content and PSII efficiency (Fv/Fm) to two rates of glyphosate. Error bars represent standard error of each mean. Data was described with two-parameter exponential decay model. Parameter estimates were given in Table 2 and 3.

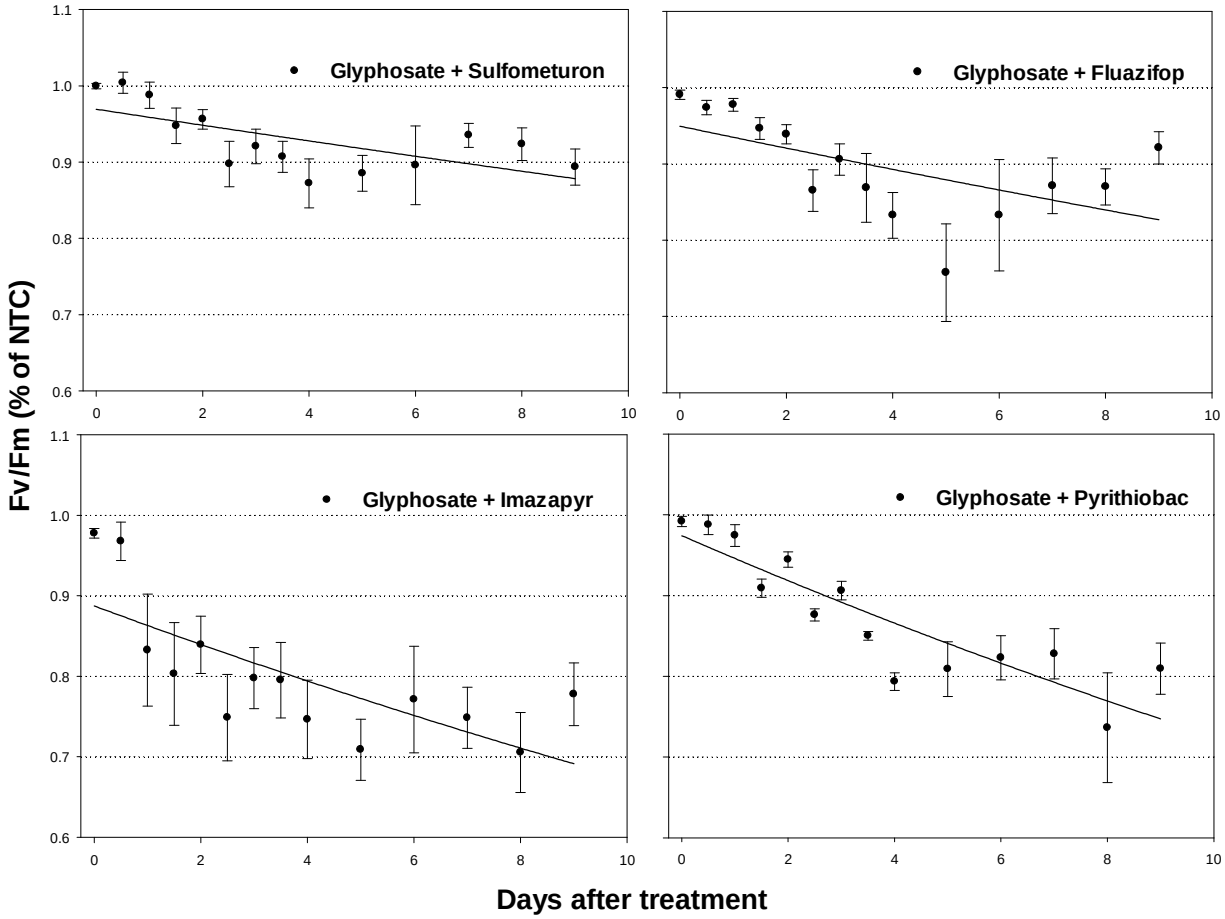

Figure 12. *M. giganteus* PSII efficiency (Fv/Fm) as affected by four POST combination treatments. Error bars represent standard error of each mean. Two-parameter exponential decay model was used to describe the data. Parameter estimates were given in Table 2.

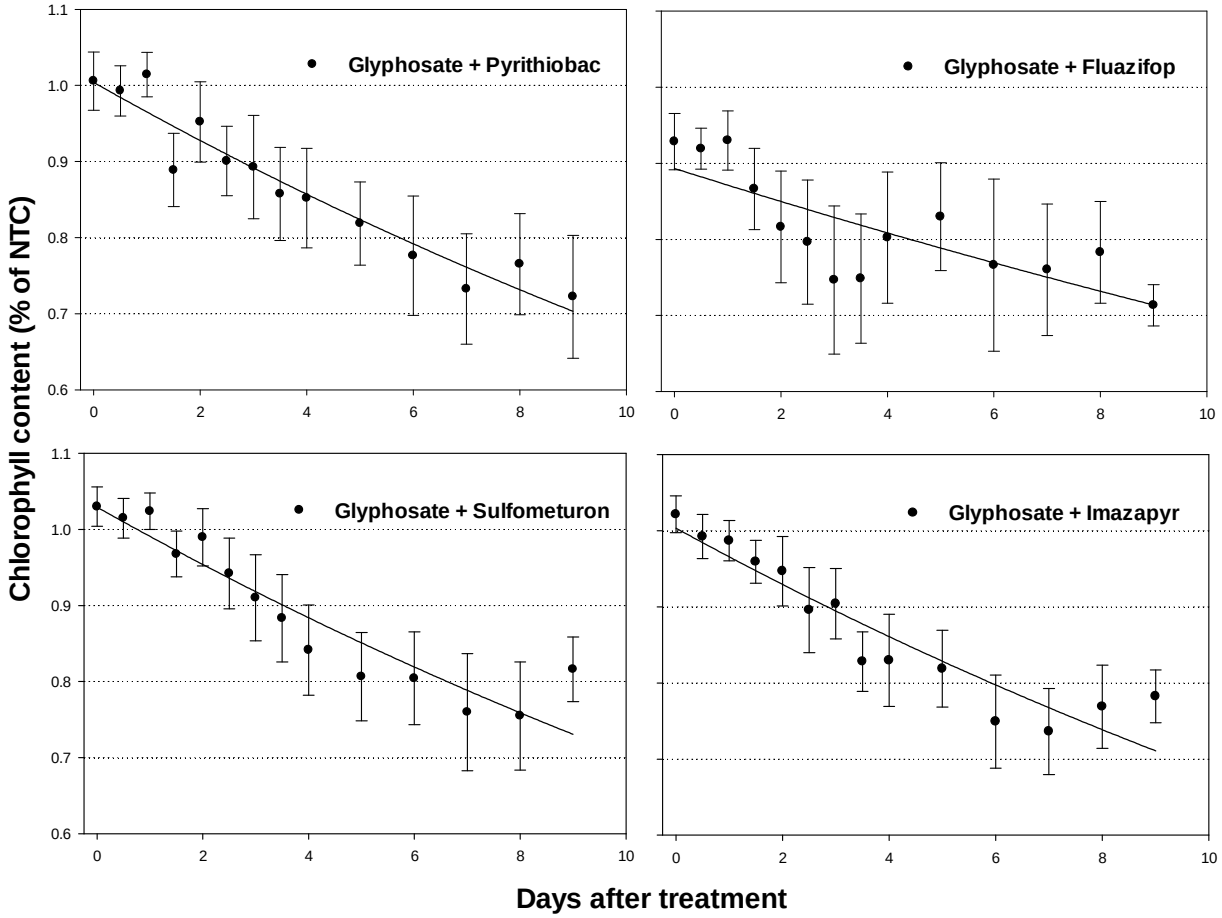

Figure 13. *M. giganteus* chlorophyll content as affected by four POST combination treatments. Error bars represent standard error of each mean. Two-parameter exponential decay model was used to describe the data. Parameter estimates were given in Table 3.

CONCLUSIONS

Experiment was conducted to evaluate fomesafen soil adsorption and desorption on 7 soils from GA, KY, CO, ID and TX (Cecil sandy loam, Greenville sandy clay loam, Tifton loamy sand, Sonora silt loam, Haxtun Sandy Loam, Minidoka silt loam and Tremona sand). Freundlich isotherms provided good description of fomesafen adsorption with R^2 greater than 0.97 for all soils used in this experiment. Adsorption equilibrium was reached after 1 hr shaking in a Cecil sandy loam. The Freundlich distribution coefficient (K_f) was generally low and K_f value varied from 1.30 to 9.28. Highest K_f was recorded with the Cecil sandy loam and lowest was found with the Tremona sand. K_{oc} ranged from 69 to 810, which suggests fomesafen adsorption was not primarily determined by soil organic matter (OM). Desorption rate after 24 hr shaking varied from 10 to 81%, with highest value observed with Tifton loamy sand, Tremona sand and Haxtun sandy loam. Fomesafen adsorption was negatively related to pH and positively related to clay content, while desorption was positively related to sand and pH and negatively related to silt, clay and OM. Lab incubation results indicated that fomesafen was barely degraded by soil microorganisms in Cecil sandy loam and Tifton loamy sand up to 90 d after treatment (DAT) under aerobic conditions. Non-linear regression using two-parameter exponential decay model suggested that model failed to be significant at 0.05 level in both soils. These results suggested fomesafen soil behavior largely depends on soil properties, microbial degradation may not be the major pathway for fomesafen dissipation in field.

Results of fomesafen greenhouse experiment suggested that the height and dry weight of cotton seedlings responded to increasing rate of fomesafen (0 to 2240 g ai ha^{-1}) in a Cecil sandy

loam from Athens and a Tifton loamy sand from Ty Ty, but not in a Greenville sandy clay loam from Plains. Field trials in Athens, Plains and Ty Ty showed that highest rate of fomesafen (2240 g ai ha^{-1}) reduced cotton stand count in all three locations as compared to the nontreated check (NTC). Cotton height was decreased by 2240 g ai ha^{-1} fomesafen in Plains and 1120, 2240 g ai ha^{-1} in Ty Ty as compared to NTC, but was not reduced in Athens at any rate evaluated in this study. Seed cotton yield was not affected by fomesafen since fixed effect rate failed to be significant at 0.05 level. The only significant yield reduction was observed with the highest rate in Ty Ty. Fomesafen persistence in Cecil sandy loam and Tifton loamy sand varied greatly. Fomesafen persisted over 120 d for the Cecil sandy loam, but was not detectable past 28 DAT for the Tifton sandy loam. The half-life (DT_{50}) of fomesafen applied at 280 g ai ha^{-1} was 47 and 6 d for Cecil sandy loam and Tifton loamy sand, respectively. When applied at 560 g ai ha^{-1}, the DT_{50} was 34 and 4 d for Cecil sandy loam and Tifton loamy sand, respectively. These data indicated fomesafen persistence varied in different soils and cotton was not affected by fomesafen within 280 to 420 g ai ha^{-1} label rate. However, initial injury and growth reduction might occur when high rate of fomesafen was accidentally sprayed to sandy soils due to miscalculation, overlapping and spraying errors, etc.

The growth response of rhizome-established *Miscanthus* × *giganteus* to various PRE and POST herbicides was evaluated in greenhouse and field. Most treatments containing atrazine, metribuzin, pendimethalin, acetochlor, metolachlor and mesotrione did not cause significant injury and growth stunting; however, EPTC at 4.5 kg ai ha^{-1} significantly reduced height and dry weight and oxadiazon resulted in higher injury compared to NTC at both rates. None of the PRE treatments affected shoot regrowth. In POST study, Thifensulfuron, metsulfuron, tribenuron, chlorimuron, halosulfuron, rimsulfuron, cloransulam, pinoxaden, bentazon and metribuzin did

not cause significant lower shoot height, reduced shoot dry weight and increased injury compared to NTC when evaluated at 4 wk after treatment. Nicosulfuron, trifloxysulfuron, sulfometuron, clodinafop, fluazifop and pyrithiobac caused greatest injury, reduced plant height and dry weights as compared to the NTC. Sethoxydim, diclofop, flumioxazin, imazamox, imazapic and imazethapyr decreased plant heights or resulted in increased injury. Within all the POST treatments studied, only fluazifop and sulfometuron injured *M. giganteus* rhizomes and reduced shoot regrowth. These results indicated that many PRE and POST herbicides in this study have the potential to control weeds in *M. giganteus* in future field trials.

The germination response of *M. giganteus* fertile seeds to various PRE herbicides were studied in petri dishes and greenhouse. Atrazine, flufenacet plus metribuzin, mesotrione, tembotrione, and acetochlor at concentrations equivalent to field use rates of 2239, 305 +76.3, 105, 92, 1343 g ai ha^{-1}, respectively, did not affected seed germination and shoot formation as compared to NTC. However, sulfentrazone, indaziflam, trifluralin, ethalfluralin and pendimethalin caused total germination failure at the end of this two-wk study while *S*-metolachlor, pyroxasulfone significant reduced germination. *M. giganteus* seed germination in petri dishes was not affected by acetochlor, atrazine and mesotrione up to 4x rates (4480, 4480 and 224 g ai ha^{-1}, respectively), however, *S*- metolachlor at 1108 and 2216 g ai ha^{-1} significantly reduced *M. giganteus* germination. In greenhouse dose-response study, *M. giganteus* seed height and dry weight were most responsive to *S*- metolachlor, followed by mesotrione, acetochlor and were least responsive to atrazine. Dose response bioassay in soil indicated herbicide rates causing 50% reduction (GR_{50}) of shoot dry weight for *S*- metolachlor and acetochlor were 84 and 1386 g ai ha^{-1}, respectively; and GR_{50} for shoot height were 291, 3209 g ai ha^{-1}, respectively, for *S*- metolachlor and acetochlor. However, those rates for atrazine and mesotrione were not

achieved within the rate range evaluated in this bioassay. Results of this study indicated several PRE herbicides have the potential to be evaluated during seeded-type *M. giganteus* establishment in large field trials. Moreover, what requires attention is several PRE herbicides proven safen on *M. giganteus* rhizomes, such as pendimethalin, trifluralin and *S*-metolachlor, may result in reduced germination or complete germination failure if used on fertile *M. giganteus* seeds due to tolerance differences.

Responses of rhizome-established *M. giganteus* to various rates of glyphosate and glyphosate in combination with fluazifop, imazapyr, pyrithiobac and sulfometuron were evaluated in greenhouse. The GR_{50} of glyphosate was 702, 1174 and 1637 g ae ha^{-1} respectively, for shoot dry weight, underground biomass and regrowth shoot dry weight, respectively. Shoot regrowth was eliminated with 4 kg ae ha^{-1} and higher rates of glyphosate. Glyphosate at 10 kg ae ha^{-1} decreased 50 and 43% of *M. giganteus* chlorophyll content and photosynthesis system (PS) II efficiency (Fv/Fm) respectively, 10 d after treatment (DAT) as compared to non-treated control (NTC). Glyphosate at 2 kg ae ha^{-1} reduced chlorophyll content and Fv/Fm by 34 and 21% respectively, at 10 DAT. One application of glyphosate at 1.68 kg ae ha^{-1} did not reduced underground biomass or cause significant visual injury, but it decreased shoot height and shoot dry weight by 8 and 17% respectively relative to NTC. Single application of glyphosate at 1.68 kg ae ha^{-1} did not have any effect on shoot regrowth. Two applications of glyphosate at 1.68 kg ae ha^{-1} dramatically improved control efficacy as compared to one application; shoot dry weights and underground biomass were reduced by 59 and 69% compared to NTC and visual injury was 76%. Shoot regrowth was eliminated with two application of glyphosate at 1.68 kg ae ha^{-1}. Glyphosate 1.68 kg ae ha^{-1} plus fluazifop, imazapyr, pyrithiobac and sulfometuron at 240, 2240, 120 and 120 g ai ha^{-1} respectively, in a single application caused more visual injury (33 to 41%),

shoot height, dry weight, and underground biomass reductions (24 to 25%, 33 to 41% and 43 to 56%, respectively) than glyphosate applied alone at 1.68 kg ae ha^{-1}, and these combinations completely eliminated shoot regrowth. Two applications of combination treatments reduced more shoot dry weight and underground biomass than one application. Reductions of chlorophyll content and Fv/Fm by glyphosate plus fluazifop, imazapyr, pyrithiobac or sulfometuron varied from 18 to 28% and 8 to 23%, respectively 10 DAT. These results suggested controlling *M. giganteus* rhizomes and shoot regrowth would require high rates or several application of glyphosate while control efficiency could be elevated by addition of POST herbicides that possessing activity against grasses.

REFERENCES

Aichele TM, Penner D (2005) Adsorption, desorption and degradation of imidazolinones in soil. Weed Technol 19:154-59

Anderson EK, Voigt TB, Bollero GA, Hager AG (2011a) *Miscanthus giganteus* response to tillage and glyphosate. Weed Technol 25:356-62

Anderson EK, Voigt TB, Bollero GA, Hager AG (2011b) Rotating a field of mature *Miscanthus* × *giganteus* to glyphosate-resistant crops. Agron J 103:1383-88

Anderson MA, Schnabel RR, Curran WS, Stout WL, Genito D, Tracy BF (2004) Switchgrass and big bluestem hay, biomass and seed yield response to fire and glyphosate treatment. Agron J 96:1688-92

Askew SD, Wilcut JW, Cranmer JR (2002) Cotton (*Gossypium hirsutum*) and weed response to flumioxazin applied preplant and postemergence directed. Weed Technol 16:184-90

Barney JN, DiTimaso JM. (2008) Nonnative species and bioenergy: Are we cultivating the next invader? BioSci 58:64-70

Baumann PA, Keeling JW, Morgan, GD, Smith JW (1998) Evaluation of fomesafen for weed control in Texas cotton. Proc South Weed Sci Soc 51:43-44

Bjorkman O, Demmig, B (1987) Photon yield of O2 evolution and chlorophyll fluorescence characteristics at 77 K among vascular plants of diverse origins. Planta 170:489-04

Bond JA, Oliver LR, Stephenson DO, IV (2006) Response of Palmer Amaranth (Amaranthus palmeri) accessions to glyphosate, fomesafen and pyrithiobac. Weed Technol 20:885-92

Bowmer KH, Eberbach PL, McCorkelle G (1993) Uptake and translocation of ^{14}C-glyphosate *Alternanthera philoxeroides* (Mart.) Griseb. (alligator weed) I. Rhizome concentrations required for inhibition. Weed Res 33:53-57

Brosnan JT, Breeden GK, McCullough PE, Henry GE (2012) PRE and POST control of annual bluegrass (*Poa annua*) with indaziflam. Weed Technol 26:48-53

Bullard MJ, Nixon PMI, Kilpatrick JB, Heath MC, Speller CS (1995) Principles of weed control in Miscanthus spp. under contrasting field conditions. pp 991–996 in Brighton Crop Protection Conference: Weeds. Proc. of an International Conference. Farnham, UK: British Crop Protection Council

Clifton-Brown JC, Lewandowski I (2000) Winter frost tolerance of juvenile *Miscanthus* plantations: studies on five genotypes at four European sites. New Phytol 148:287–94

Clifton-Brown JC, Lewandowski I, Andersson B, Basch B, Christian DG, Bonderup-Kjeldsen J, Jørgensen U, Mortensen J, Riche AB, Schwarz KU, Tayebi K, Teixeira F (2001) Performance of 15 *Miscanthus* Genotypes at Five Sites in Europe. Agronomy J 93:1013–19

Clifton-Brown J, Robson P, Sanderson R, Hastings A, Valentine J, Donnison I (2011) Thermal requirments for seed germination in Miscanthus compared with switchgrass (*Panicum virgatum*), reed canary grass (*Phalaris arundinaceae*), maize (*Zea mays*) and perennial ryegrass (*Lolium perenne*). GCB bioenerg 3:375-86

Cranmer, JR, Altom JV, Braun JC, Pawlak JA (2000) Valor herbicide: a new herbicide for weed control in cotton, peanuts, soybeans, and sugarcane. Proc South Weed Sci Soc 53:158

Culpepper AS., York AC, MacRae AW, Kichler J. (2008) Glyphosate resistant Palmer amaranth response to weed management programs in Roundup Ready and Liberty Link Cotton. Pp. 1689 *in* Proc of the Beltwide Cotton Conferences. Natl. Cotton Counc. Am., Memphis, TN

Culpepper AS (2009) Impact of EPA's ecological risk assessment for fomesafen. Subject: re-registration of fomesafen (Docket # EPA-HQ-OPP-2006-0239) US-EPA. Washington, DC. 20460

Cutts GS, Webster TM, Grey TL, Vencill WK, Lee RD, Tubbs RS, Anderson WF (2011) Herbicide effect on napiergrass (*Pennisetum purpureum*) control. Weed Sci 59:255-62

Daehler C (1998) The taxonomic distribution of invasive angiosperm plants: ecological insights and comparison to agricultural weeds. Biol Conserv 84:167-80

Derr JF (2002) Tolerances of ornamental grasses to preemergence herbicides. J Environ Hort 20:161-65

Derr JF (2008) Common reed (*Phragmites australis*) response to postemergence herbicides. Invasive Plant Sci Manag 1:153–57

Dotray PA, Grichar WJ, Gilbert LV (2010) Peanut tolerance and weed control following fomesafen applied at different rates and timing in Texas. Proc American Peanut Research and Education Soc 42:30-32

Doub JP, Wilson HP, Hines TE, Hatzios KK (1988) Consecutive annual applications of alachlor and metolachlor to continuous no-till corn (*Zea mays*). Weed Sci 36:340-344

EPA (2006) Sodium salt of fomesafen. Docket ID: EPA-HQ-OPP-2006-0239-0010. Available from: http://www.regulations.gov/#!docketBrowser;rpp=25;po=0;s=EPA-HQ-OPP-2006-0239-0010;dct=PS%252BSR;D=EPA-HQ-OPP-2006-0239

Emmerich WE, Hardegree SP (1991) Seed germination in polyethylene glycol solution: effects of filter paper exclusion and water vapor loss. Crop Sci 31:454-58

Everman WJ, Clewis SB, York AC, Wilcut JW (2009) Weed control and yield with flumioxazin, fomesafen and S-metolachlor systems for glufosinate-resistant cotton residual weed

management. Weed Technol 23:391-97

Everman WJ, Lindsey AJ, Henry GM, Glaspie CF, Phillips K, McKenney C (2011) Response of *Miscanthus x giganteus* and *Miscanthus sinensis* to postemergence herbicides. Weed Technol 25:398-03

Ferrell JA, Vencill WK, Xia K, Grey TL (2005) Sorption and desorption of flumioxazin to soil, clay minerals and ion-exchange resin. Pest Manag Sci 61:40-46

Gardner AP, York AC, Jordan DL, Monks DW (2006) Management of annual grasses and Amaranthus spp. in glufosinate-resistant cotton. J Cotton Sci 10:328-38

Gilbert LV, Dotray PA, Prostko EP, Grichar WJ, Ferrell JA, and Jordan DL (2009) Peanut response to fomesafen. Proc American Peanut Research and Education Soc 41:28-30

Glenn, S, Peregoy RS, Hook BJ, Heimer JB, Wiepke T. (1986) *Sorghum halepense* (L.) Pers. control with foliar-applied herbicides in conventional and no-tillage soybeans. Weed Res 26:245–50

Gordon DR, Tancig KJ, Onderdonk DA, Gantz CA (2011) Assessing the invasive potential of biofuel species proposed for Florida and the United States using the Australian weed risk assessment. Biomass Bioenerg 35:74-79

Gray AJ, Marshall DF, Raybouild AF (1991) A century of evolution in *Spartina anglica*. Adv Ecol Res 21:1-62

Greef, JM, Deuter M (1993) Syntaxonomy of *Miscanthus x giganteus* GREEF et DEU. Angewandte Botanik 67:87-90

Grey TL, Walker RH, Hancock HG (1997) Sulfentrazone adsorption and mobility as affected by soil and pH. Weed Sci 45:733-38

Grichar WJ, Besler BA, Brewer KD (2005) Weed control and grain sorghum (*Sorghum bicolor*)

response to postemergence applications of atrazine, pendimethalin and trifluralin. Weed Technol 19:999-1003

Grichar WJ (2006) Weed control and grain sorghum tolerance to flumioxazin. Crop Protect 25:174-77

Guo J, Zhu G, Shi J, Sun J (2003) Adsorption, desorption and mobility of fomesafen in Chinese soils. Water Air and Soil Pollut 148:77-85

Hamill AS, Zhang J (1995) Quackgrass control with glyphosate and SC-0224 in corn and soybean. Can J Plant Sci 75:293-99

Harper SS (1994) Sorption–desorption and herbicide behavior in soil. Rev Weed Sci 6:207–25

Heaton EA, Voigt T, Long SP (2004) A quantitative review comparing the yields for two candidate C4 perennial biomass crops in relation to nitrogen, temperature and water. Biomass Bioenerg. 27:21-20

Heaton EA, Dohleman FG, Long SP (2008) Meeting US biofuel goals with less land: the potential of Miscanthus. Global Change Biol. 14:2000-14

Heaton EA, Dohleman FG, Miguez AF, Juvik JA, Lozovaya V, Widholm J, Zabotina OA, Mcisaac GF, David MB, Voigt TB, Boersma NN, Long SP (2010) Miscanthus: A Promising Biomass Crop. Adv Bot Res 56:75–137

Hirata M., Hasegawa N, Nogami K, Sonoda T (2007) Tuft, shoot and leaf dynamics in *Miscanthus sinensis* in a young tree plantation under cattle grazing. Trop Grassl 41:113-28

Huang Y, Thomson SJ, Molin WT, Reddy KN, Yao H (2012) Early detection of soybean plant injury from glyphosate by measuring chlorophyll reflectance and fluorescence. J of Agric Sci 4:117-24

Ito M, Ueki K, Sakamoto S (1982) Studies on the total vegetation control in railroad: 1. Major

weeds and factors affecting their distribution. Weed Res Jpn 27:41-48

James TK, Rahman A (2009) Efficacy of Pre-emergence herbicides on three annual grass weeds in different soils. NZ Plant Protect 62:356-362

Jones JVH (2009) Testing the economic viability of energy crop production in competition with alternative land uses. Proc of the 17th International Farm Management Congress. 1:402–23

Ketchersid ML, Merkle MG (1975) Persistence and movement of perfluridone in soil. Weed Sci 23:344-48

Ketel DH (1996) Effect of low doses of metamitron and glyphosate on growth and chlorophyll content of common lambsquaters (*Chenopodium album*). Weed Sci 44:1-6

Kichler JM, Culpepper AS, Chafin JE (2010) Glyphosate-resistant Palmer amaranth and cotton response to residual at-plant herbicides applied on the soil surface or preplant incorporated. Pp. 1654. *In* Proc of the Beltwide Cotton Conference, New Orleans, LA. Natl. Cotton Counc. Am., Memphis, TN

Kichler J, Culpepper AS (2012) How tillage and application timing of reflex affects palmer amaranth control and cotton injury. Pp.1543 *In* Proc of the Beltwide Cotton Conference, Orlando, FL. Natl. Cotton Counc. Am., Memphis, TN

Kirkland KJ (1996) Use of incorporated and non-incorporated granular trifluralin for annual grass control in barley (*Hordeum vulgare*), wheat (*Triticum aestivum*), and canola (*Brassica napus*). Weed Technol 10:907-13

Kolar CS, Lodge DM (2001) Progress in invasion biology: predicting invaders. Trends Ecol Evol 16:199-04

Lewandowski I (1998) Propagation method as an important factor in the growth and development of *Miscanthus* × *giganteus*. Ind. Crop Prod 8:229-45

Lewandowski I, Clifton-Brown, Scurlock JM, Huisaman W (2000) Miscanthus: European experience with a novel energy crop. Biomass Bioenerg19:209-27

Lewandowski I, Scurlock JMO, Lindvall E, Christou M (2003) The development and current status of perennial rhizomatous grasses as energy crops in the US and Europe. Biomass Bioenerg 25:335-61

Li X, Grey TL, Blanchett BH, Lee RD, Webster TM, Vencill WK (2013) Tolerance evaluation of vegetatively established *Miscanthus* × *giganteus* to Herbicides. Weed Technol 27:735-40

Linde-Laursen IB. (1993). Cytogenetic analysis of *Miscanthus* `Giganteus', an interspecic hybrid. Hereditas 119:297-300

Linde-Laursen IB (1993) Cytogenetic analysis of Miscanthus ‘Giganteus’, an interspecific hybrid. Hereditas 119:297–300

Ling Q, Huang W, Jarvis P (2011) Use of a SPAD-502 meter to measure leaf chlorophyll concentration in *Arabidopsis thaliana*. Photosynth Res 107:209-14

Loh FCW, Grabosky JC, Bassuk NL (2002) Using the SPAD 502 meter to assess chlorophyll and nitrogen content of Benjamin Fig and cottonwood leaves. Hort Tech 12:682-86

Lolas PC, Coble HD (1980) Translocation of 14C-glyphosate in johnsongrass (*Sorghum halepense* L. Pers.) as affected by growth stage and rhizome length. Weed Res 20:267-70

Loux MM, Reese KD (1993) Effect of soil type and pH on persistence and carryover of imidazolinon herbicides. Weed Technol 7:452-58

Lunsford JN, Harrison S, Smith JD (1998) Reflex use in cotton. Proc South Weed Sci Soc 51:12-13

Lyon DJ, Wilson RG (2005) Chemical weed control in dryland and irrigated chickpea. Weed Technol 19: 959-65

Main CL, Faircloth JC, Steckel LE, Culpepper AS, York AC (2012) Cotton tolerance to fomesafen applied preemergence. J Cotton Sci 16:80-87

Marshall MW (2009) Complementary herbicide programs for Palmer amaranth (*Amaranthus palmeri* S. Wats.) control in glyphosate-tolerant cotton and soybeans. Abstr Weed Sci Soc Am 49:99

Matlaga DP, Davis AS (2013) Minimizing invasive potential of *Miscanthus* × *giganteus* grown for bioenergy: identifying demographic thresholds for population growth and spread. J Appl Ecol 50:479–87

Mersie W, Foy CL (1986) Adsorption, desorption and mobility of chlorsulfuron in soils. J Agric Food Chem 34:89-92

Misra AN, Misra M, Singh R (2012) Chlorophyll fluorescence in plant biology. Pp.171 *in* Biophysics, Dr. Prof. Dr. A.N. Misra (Ed.), ISBN: 978-953-51-0376-9, InTech, Available from: http://www.intechopen.com/books/biophysics/chlorophyll-fluorescence-in-plant-biology

Morillo E, Undabeytia T, Cabrera A, Villaverde J, Maqueda C (2004) Effect of soil type on adsorption-desorption, mobility and activity of the herbicide norflurazon. J. Agric Food Chem 52:884-90

Mueller-Warrant GW (1999) Duration of control from preemergence herbicides for use in nonburned grass seed crops. Weed Technol 13:439-49

Murdock EC, Keeton A (1998) Where does fomesafen fit in South Carolina cotton weed management programs? Proc South Weed Sci Soc 51:12

Newby SE, White BG (1981) PP021: leaching on soil thick-layer chromatograms. London, ICI. pp.4 (Report RJ0156B)

Numata M (1969) Progressive and retrogressive gradient of grassland vegetation measured by degree of succession-Ecological judgment of grassland condition and trend IV. Vegetatio. 19:96-127

Numata, M. Editor (1974) Grassland vegetation. The ora and vegetation of Japan. Tokyo: Elsevier: pp. 125-47

Omielan J, Gumm D, Witt W (2012) Evaluation of management options for control of Chinese silvergrass (*Miscanthus sinensis* Anders.) Poster, Southeast exotic pest plant council annual meeting. May 8-10, 2012. Auburn AL. Available from: http://www2.ca.uky.edu/pss/weeds/ivm/pdf/Miscanthus%20Poster%20for%20AL.pdf

Oymada M, Kuwatsuka S (1988) Effects of soil properties and conditions on the degradation of three diphenylether herbicides in flooded soils. J Pestic Sci 13:99-105

Parochetti JV, Wilson HP, Burt GW (1975) Activity of glyphosate on johnsongrass. Weed Sci 23:395–400

Percival GC, Fraser GA (2001) Measurement of the salinity and freezing tolerance of Crataegus genotypes using chlorophyll fluorescence. J Arboric 27:233–45.

Percival GC, Sheriffs C (2002) Identification of drought-tolerant woody perennials using chlorophyll fluorescence. J. Arboric 28:215–24

Percival GC (2004) Evaluation of physiological tests as predictors of young tree establishment and growth. J. Arboric 30:80-91

Pusino A, Petretto S, Gessa C (1997) Adsorption and desorption of Imazapyr by soil. J Agric Food Chem 45:1012-16

Pyter RJ, Rosales-Robels FE, Chander JM, Senseman SA, Prostko EP (1999) Influence of growth stage and herbicide rate on postemergence johnsongrass (*Sorghum halepense*)

control. Weed Technol 13:525-29

Pyter RJ, Dohleman FG, Voigt TB (2010) Effects of rhizome size, depth of planting and cold storage on *Miscanthus* × *giganteus* establishment in the Midwestern USA. Biomass Bioenerg 34:1466-70

Quinn LD, Allen DJ, Stewart JR (2010) Invasiveness potential of *Miscanthus sinensis*: implications for bioenergy production in the United States. GCB Bioenergy 2:310-20

Quinn LD, Matlaga DP, Stewart JR, Davis AS (2011) Empirical evidence of long-distance dispersal in Miscanthus sinensis and *Miscanthus* × *giganteus*. Invasive Plant Sci Manage 4:142–50

Reiling, KR, Simmions FW, Reichers DE, Steckel LE (2006) Application timing and soil pH effects on sulfentrazone phytotoxicity to two soybean (*Glycine max* (L) Merr.) cultivars. J Crop Prot 25:230-34

Rauch BJ, Bellinder RR, Brainard DC, Lane M, Thies JE (2007) Dissipation of fomesafen in New York state soil and potential to cause carryover injury to sweet corn. Weed Technol 21:206-12

Raghu S, Anderson RC, Daehler CC, Davis AS, Wiedenmann RN, Simberloff D, Mack RN (2006) Adding biofuels to the invasive species fire? Science 313:1742

Rensburg, E. V. 1996. Reed eradication under transmission powerlines in South Africa using microlight aircraft. Pages 1095–1101. *in* Proc of the Second International Weed Control Congress, Copenhagen, Denmark, 25–28 June 1996. Slagelse, Denmark: Department of Weed Control and Pesticide Ecology.

Rioux R, Bandeen JD, Anderson GW (1974) Effects of growth stage on translocation of glyphosate in quackgrass. Can J Plant Sci 54:397-01

Ross M (2011) New Miscanthus development possible biomass game changer? FarmWeek, May 16, 2011. pp. 9

SAS Institute, Inc. 2012. SAS/STAT® 9.2 User's guide, Second Edition. http://support.sas.com/documentation/cdl/en/statug/63033/HTML/default/viewer.htm#statug_glimmix_a0000001405.htm

Savage KE, Wauchope RD (1974) Fluometuron adsorption–desorption in soil. Weed Sci 22:106-10

Schrage BW, Norsworthy JK, Smith KL, Johnson DB, Bagavathiannan MV, Riar DS (2012) Factors contributing to cotton injury from soil-applied residual herbicides. Summary of Arkansas Cotton Research 2012:102-06

Senseman SA, ed (2007) Herbicide Handbook. 9th ed. Lawrence, KS: Weed Science Society of America. pp 207, 243, 308

Sosnoskie LM, Kichler JM, Wallace RD, Culpepper AS (2011) Multiple resistance in Palmer Amaranth to glyphosate and pyrithiobac confirmed in Georgia. Weed Sci 59:321-25

Speller CS (1993) Weed control in Miscanthus and other annually harvested biomass crops for energy or industrial use. pp. 671–676. *In* Brighton Crop Protection Conf., Weeds. Proc. of an Int. Conf., Brighton, UK. 22–25 Nov. 1993. British Crop Protection Council (BCPC), Farnham UK.

Spencer DF, Tan W, Liow P, Ksander GG, Whitehan LC (2008) Evaluation of glyphosate for managing giant reed (*Arundo donax*). Invasive Plant Sci Manage 1:248-54

Spencer DF, Ksander GG, Tan W, Liow PS, Whitehand LC (2011) Influence of application timing on the impact of glyphosate on giant reed (*Arundo donax* L.). J Auat plant manage 49:106-10

Stephenson DO, Patterson MG, Faircloth WH and Lunsford JN (2004) Weed management with fomesafen preemergence in glyphosate resistant cotton. Weed Technol 18:680-86

Stougaard RN, Shea PJ, Martin AR (1990) Effect of soil type and pH on adsorption, mobility and efficacy of imazaquin and imazethapyr. Weed Sci 38:67-73

Syngenta Crop Protection (2014) Reflex label. Greensboro, NC. 27419. Available from http://www.cdms.net/LDat/ld6BJ002.pdf

Sugimoto Y (2002) Forest-pastoral systems – studies in Morotuka village, Miyazaki prefecture. Grassl Sci 47:644-51

Tanford C (1973) The hydrophobic effect. John Wiley and Sons, Inc,. New York.

Taylor-Lovell, S, Wax LM, Nelson R (2001) Phytotoxic response and yield of soybean (*Glycine max*) varieties treated with sulfentrazone or flumioxazin. Weed Technol 15:95-102

Troxler SC, Askew SD, Wilcut JW, Smith WD, Paulsgrove MD (2002) Clomazone, fomesafen and bromoxynil systems for bromoxynil-resistant cotton (*Gossypium hirsutum*). Weed Technol 16:838-44

Uddling J, Gelang-Alfredsson J, Piikki K, Pleijel H (2007) Evaluating the relationship between leaf chlorophyll concentration and SPAD-502 chlorophyll meter readings. Phtotsynth Res 91:37-46

USDA-NASS (2010) Agricultural chemical use program. Available from: http://www.nass.usda.gov/Surveys/Guide_to_NASS_Surveys/Chemical_Use/index.asp

Vaughn KC, Lehnen LP (1991) Mitotic disrupter herbicides. Weed Sci 39:450-57

Venturi P, Venturi G (2003) Analysis of energy comparison for crops in European agricultural systems. Biomass Bioenerg 25:235-55

Voigt PW, Tischler CR (1996) Effect of seed treatment on germination and emergence of 3

warm-season grasses. J. Range Manage 50:170-74

Walker A, Jurado-Exposito M (1998) Adsorption of isoproturon, diuron, and metsulfuron-methyl, in two soils at high soil:solution ratios. Weed Res 38:229-38

Walsh MJ, Fowler TM, Crowe B, Ambe T, Powles SB (2011) The potential for pyroxasulfone to selectivity control resistant and susceptible rigid ryegrass (*Lolium rigidum*) biotypes in Australian grain crop production systems. Weed Technol 25:30-37

Weber JB (1993a) Ionization and sorption of fomesafen and atrazine by soils and soil constituents. Pestic Sci 39:31-38

Weber JB (1993b) Mobility of fomesafen and atrazine in soil columns under saturated and unsaturated flow conditions. Pestic Sci 39:39-46

Wilcut, JW, Askew SD, Price AJ, Scott GH, Cranmer J. (2000) Valor: new weed management option for cotton. Proc South Weed Sci Soc 53:159–60

Wise AM, Grey TL, Prostko EP, Vencill WK, Webster TM (2009) Establishing the geographic distribution level of acetolactate synthase resistance of Palmer amaranth (*Amaranthus palmeri*) accessions in Georgia. Weed Technol 23:214-20

Ying G, Williams B (2000). Dissipation of herbicides in soil and grapes in a South Australian vineyard. Agric Ecos Environm 78:283-89

Yuan JS, Tiller KH, Al-Ahmad H, Stewart NR, Stewart Jr CN (2008) Plants to power: bioenergy to fuel the future. Trends Plant Sci 13:421-29