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Evaluation of Foliar Fertilizer or Cytokinin Mixtures in Combination with Common Postemergence Soybean Herbicides

Huntington Tyler Hydrick

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Evaluation of foliar fertilizer or cytokinin mixtures in combination with common
postemergence soybean herbicides

By

Huntington Tyler Hydrick

A Thesis
Submitted to the Faculty of
Mississippi State University
in Partial Fulfillment of the Requirements
for the Degree of Master of Science
in Weed Science
in the Department of Plant and Soil Sciences

Mississippi State, Mississippi

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2017

Evaluation of foliar fertilizer or cytokinin mixtures in combination with common
postemergence soybean herbicides

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In an effort to reduce application costs and to integrate plant health management strategies in soybean, growers may combine POST herbicides with foliar fertilizers or cytokinin mixtures. Field experiments were conducted at the Delta Research and Extension Center in Stoneville, MS in 2015 and 2016 to evaluate soybean [*Glycine max* (L.) Merr.] injury, weed control, and agronomic performance when combining blended or single-nutrient foliar fertilizers with POST herbicide applications. Field experiments were also conducted at the Delta Research and Extension Center in Stoneville, MS in 2015 and 2016 to evaluate the influence of cytokinin mixtures on soybean injury and weed control when combined with common POST soybean herbicides.

DEDICATION

I would like to dedicate my work to my wife, Bobbi Hydrick, my father, mother, four brothers, and extended family for always encouraging me throughout this journey and letting me grow into the man I am today. I would also like to dedicate my research to the faculty in the biological sciences department at the University of the Ozarks for pushing me to achieve in school and in life. Lastly, I would like to dedicate this to my friends from Jonesboro, Arkansas and the University of the Ozarks for their undying support and encouragement throughout this moment of my life and many others.

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TABLE OF CONTENTS

DEDICATION	ii
ACKNOWLEDGEMENTS	iii
LIST OF TABLES	vi
CHAPTER	
I. AN EVALUATION ON THE INFLUENCE OF FOLIAR FERTILIZER IN COMBINATION WITH COMMON POSTEMERGENCE SOYBEAN HERBICIDES	1
Introduction	1
Literature Cited.....	15
II. AN EVALUATION ON THE INFLUENCE OF FOLIAR FERTILIZER IN COMBINATION WITH COMMON POSTEMERGENCE SOYBEAN HERBICIDES	23
Abstract.....	23
Introduction	24
Materials and Methods	30
Weed Control Study	30
Agronomic Study.....	32
Single-nutrient Foliar Fertilizer Study.....	34
Results and Discussion	35
Weed Control Study	35
Agronomic Study.....	38
Single-nutrient Foliar Fertilizer Study.....	38
Literature Cited.....	57
III. AN EVALUATION ON THE INFLUENCE OF CYTOKININ MIXTURES IN COMBINATION WITH COMMON POSTEMERGENCE SOYBEAN HERBICIDES	63

Abstract.....	63
Introduction	64
Materials and Methods	68
Results and Discussion.....	70
Literature Cited.....	76

LIST OF TABLES

2.1	Soybean injury 3, 7, and 14 d after treatment (DAT), Palmer amaranth and barnyardgrass control 28 DAT, height 14 DAT, and soybean yield following application of mixtures of POST soybean herbicides and a blended foliar fertilizer applied at the V3 growth stage in the Weed Control Study at Stoneville, MS, in 2015 and 2016a.	43
2.2	Antagonistic effects for Palmer amaranth control 7 d after treatment (DAT) with mixtures of POST soybean herbicides and a blended foliar fertilizer applied at the V3 growth stage in the Weed Control Study at Stoneville, MS, in 2015 and 2016.	44
2.3	Antagonistic effects for Palmer amaranth control 14 d after treatment (DAT) with mixtures of POST soybean herbicides and a blended foliar fertilizer applied at the V3 growth stage in the Weed Control Study at Stoneville, MS, in 2015 and 2016.	45
2.4	Antagonistic effects for Palmer amaranth control 21 d after treatment (DAT) with mixtures of POST soybean herbicides and a blended foliar fertilizer applied at the V3 growth stage in the Weed Control Study at Stoneville, MS, in 2015 and 2016.	46
2.5	Antagonistic effects for barnyardgrass control 7 d after treatment (DAT) with mixtures of POST soybean herbicides and a blended foliar fertilizer applied at the V3 growth stage in the Weed Control Study at Stoneville, MS, in 2015 and 2016.	47
2.6	Antagonistic effects for barnyardgrass control 14 d after treatment (DAT) with mixtures of POST soybean herbicides and a blended foliar fertilizer applied at the V3 growth stage in the Weed Control Study at Stoneville, MS, in 2015 and 2016.	48
2.7	Antagonistic effects for barnyardgrass control 21 d after treatment (DAT) with mixtures of POST soybean herbicides and a blended foliar fertilizer applied at the V3 growth stage in the Weed Control Study at Stoneville, MS, in 2015 and 2016.	49

2.8	Barnyardgrass control 28 d after treatment (DAT) with mixtures of POST soybean herbicides and a blended foliar fertilizer applied at the V3 growth stage in the Weed Control Study at Stoneville, MS, in 2015 and 2016a.	50
2.9	Soybean injury 3, 7, and 14 d after treatment (DAT), soybean dry weight 14 DAT, and soybean height 14 DAT following application of mixtures of POST soybean herbicides and a blended foliar fertilizer applied at the V3 growth stage in the Agronomic Study at Stoneville, MS, in 2015 and 2016a.	51
2.10	Soybean injury 3, 7, and 14 d after treatment (DAT) and Palmer amaranth control 14, 21, and 28 DAT with mixtures of POST soybean herbicides and single-nutrient foliar fertilizers applied at the V3 growth stage in the Single-nutrient Foliar Fertilizer Study at Stoneville, MS, in 2015 and 2016a,b.	52
2.11	Palmer amaranth and barnyardgrass control 7 d after treatment (DAT) with mixtures of POST soybean herbicides and single-nutrient foliar fertilizers applied at the V3 growth stage in the Single-Nutrient Foliar Fertilizer Study at Stoneville, MS, in 2015 and 2016a,b.	53
2.12	Barnyardgrass control 14 d after treatment (DAT) with mixtures of POST soybean herbicides and single-nutrient foliar fertilizers applied at the V3 growth stage in the Single-nutrient Foliar Fertilizer Study at Stoneville, MS, in 2015 and 2016a.	54
2.13	Barnyardgrass control 14, 21, and 28 d after treatment (DAT), soybean height 14 DAT, and mature soybean height following application of mixtures of soybean herbicides and single-nutrient foliar fertilizers applied at the V3 growth stage in the Single-nutrient Foliar Fertilizer Study at Stoneville, MS, in 2015 and 2016a.	55
2.14	Soybean yield following mixtures of POST soybean herbicides and single-nutrient foliar fertilizers applied at the V3 growth stage in the Single-nutrient Foliar Fertilizer Study at Stoneville, MS, in 2015 and 2016a,b.	56
3.1	Soybean injury 3, 7, and 14 d after treatment (DAT) and Palmer amaranth control 7, 14, 21, and 28 DAT with mixtures of POST soybean herbicides and cytokinin mixtures applied at the V3 growth stage in Stoneville, MS, in 2015 and 2016a.	73

3.2	Antagonistic responses for barnyardgrass control 14 d after treatment (DAT) with mixtures of POST soybean herbicides and cytokinin mixtures applied at the V3 growth stage in Stoneville, MS, in 2015 and 2016a,b.....	74
3.3	Barnyardgrass control 7, 21 and 28 d after treatment (DAT), height 14 DAT, mature height, and yield with mixtures of POST soybean herbicides and cytokinin mixtures applied at the V3 growth stage in Stoneville, MS, in 2015 and 2016a.....	75

CHAPTER I
AN EVALUATION ON THE INFLUENCE OF FOLIAR FERTILIZER IN
COMBINATION WITH COMMON POSTEMERGENCE
SOYBEAN HERBICIDES

Introduction

Soybean [*Glycine max* (L.) Merr.] dates back 5,000 yr when it was first domesticated in China (Hoeft et al. 2000). Soybean was first introduced to North America by Samuel Bowen in 1765 when it was planted on his farm near Savannah, GA (Hymowitz 2004). In 1954, the United States became the world's leading producer of soybean with more than 9.2 trillion kg of the world's 19.8 trillion kg production (Probst and Judd 1973). In 2016, 33.8 million ha of soybean were planted in the U.S. producing just over 117 trillion kg (Anonymous 2016). Mississippi accounted for 825,910 ha of soybean in the United States in 2015, which produced 2.1 trillion kg (Anonymous 2016).

Soybean is an annual plant reaching 75 to 125 cm in height with a diffuse root system and three types of leaves (Lersten and Carlson 2004). Leaf types include cotyledons, simple primary leaves, and trifoliolate leaves. During vegetative growth, soybean seed germinate and cotyledons emerge, followed by the simple primary leaves, then the trifoliolates (Lersten and Carlson 2004). Once axillary buds develop into flower clusters, the soybean has entered reproductive growth (Lersten and Carlson 2004).

Reproductive growth consists of the flowering, pod setting, seed formation, and mature stages (Hoeft et al. 2000).

Cultivar selection is the first major step to a successful soybean crop (Heatherly and Elmore 2004). Newer cultivars are released by public and private breeders, branded by their resistance to diseases, herbicides, insects, and nematodes, but no one cultivar is best for all production scenarios (Heatherly and Elmore 2004). Factors such as soil texture, irrigation, and field history (disease, nematodes, weeds, fertility requirements, etc.) influence the planting and growing environment (Heatherly 1999).

Cultivars are typically classified into one of 13 maturity groups (MG), or a group based on the length of time to reach maturity (Heatherly and Elmore 2004). Maturity groups typically utilized in Mississippi and other midsouthern U.S. states are III, IV, V, VI, and VII (Salmeron et al. 2014). Once the specifics of the planting situation are determined, variety or cultivar performance trials conducted by public or private entities can be used to choose the greatest yielding or most affordable cultivar (Heatherly and Elmore 2004; Heatherly 1999; Salmeron et al. 2014).

Planting date has been reported to be one of the most important factors affecting soybean yields (Salmeron et al. 2014). Salmeron et al. (2014) concluded that a May- to June-planted soybean produced 5 to 10% lower yield than March- to April-planted soybean. In a conventional soybean production system (CSPS), MG V or later cultivars are planted in May and June in the midsouthern U.S. (Heatherly 1999). These cultivars are often exposed to drought-like weather during their reproductive stages from mid-July to mid-September, which leads to stress and ultimately yield losses (Heatherly 1999). These yield losses can be correlated to high evaporation rates during the mid-summer

months (Heatherly 1999). In an early soybean production system (ESPS), soybean MG IV or earlier cultivars are planted in April with all tillage performed in the fall of the previous year and spring field preparation accomplished with preplant foliar applied herbicides (Heatherly 1999; Heatherly et al. 2002; Salmeron et al. 2014). Planting an earlier MG in April allows for adequate moisture and more favorable growing conditions during reproductive stages (Heatherly 1999; Heatherly and Spurlock 2001). Salmeron et al. (2014) reported MG IV cultivars were the most stable compared to other MG, producing greater or similar yields across sites and soil textures in Arkansas, Louisiana, Mississippi, Missouri, and Texas.

The critical period of weed control (CPWC) is the time where weeds that emerged with the crop must be controlled to avoid yield losses (Knezevic et al. 2003b). Weeds that germinate after the CPWC are not detrimental to crop yield (Knezevic et al. 2003b). Due to wide adoption of glyphosate-resistant (GR) soybean, CPWC has become an important part of integrated weed management (Knezevic et al. 2003b). Herbicide systems within a GR soybean crop can add a POST application of glyphosate, whereas a conventional crop may rely too heavily on PRE herbicides (Knezevic et al. 2003b). The critical time for weed removal (CTWR) is the time when weed control must be initiated to avoid yield losses (Knezevic et al. 2003b). The CTWR is best described as the initial stage of the CPWC. Critical times for weed removal can vary with different factors such as row spacing in soybean (Heatherly 1999; Knezevic et al. 2003b). By decreasing the row spacing, the CTWR is extended later in the growing season and ultimately, shortens the overall time for the CPWC (Knezevic et al. 2003b).

Burndown describes herbicide applications before and/or during planting (Owen et al. 2009). In an ESPS, one burndown herbicide application in February that includes a residual herbicide can control weeds until planting (Heatherly 1999). Canopy closure is extremely important in any soybean production system, and weeds are more competitive in systems utilizing wide compared with narrow row spacings due to light availability and more available soil nutrients (Knezevic et al. 2003a). If weeds are not removed during the CTWR, soybean yield losses can be 2% per leaf stage of delay (Knezevic et al. 2003a). Where new weeds emerge before soybean canopy closure, residual herbicides may need to be added to POST herbicide treatments (Knezevic et al. 2003a).

Amaranthus spp., known collectively as pigweeds, belong to the family *Amaranthaceae* and have ranked among the top ten most troublesome weeds in Mississippi soybean since the early 1970's (Anonymous 1972; Buchanan 1973, 1974). Palmer amaranth [*Amaranthus palmeri* (S.) Wats.] is native to northern Mexico, southern California, New Mexico, and Texas (Ehleringer 1983; Sauer 1957). The Navajo, Pima, Yuma, and Mohave peoples used this plant for food by grinding seeds into meal and cooking the leaves as greens (Moerman 1998).

Early in the 20th century, the range of Palmer amaranth in the United States increased, likely due to human activity transporting seed (Culpepper et al. 2010). By 1971, Palmer amaranth was present in Mississippi (Culpepper et al. 2010). Palmer amaranth increased in severity from 1995 to 2008, ascending from the number 10 to number one most troublesome weed in cotton [*Gossypium hirsutum* (L.)] and from 23 to number two in soybean (Webster and Nichols 2012). Nine of 10 southern U.S. states surveyed ranked Palmer amaranth as the most troublesome weed of cotton in 2012

(Webster and Nichols 2012). Among eight southern U.S. states surveyed in 2013, Palmer amaranth was ranked as the most troublesome weed of cotton and soybean in seven and three states, respectively (Webster 2013).

Palmer amaranth is characterized by oval-shaped leaves with petioles longer than the leaf blade and a green or reddish stem (Sauer 1955). Palmer amaranth is a tall (sometimes exceeding 2.5 m), frequently branching, summer annual (Bryson and DeFelice 2009). Because Palmer amaranth is a dioecious species, female (pistillate) and male (staminate) flowers appear on separate plants. Pistillate or staminate flowers cluster to form a single cylindrical inflorescence or spike up to 60 cm long on their respective plants. Inflorescences can be distinguished by touch with males having softer spikes and females having a rougher, prickly spike due to stiff bracts (Ward et al. 2013). Palmer amaranth seeds are smooth and round or disc shaped, and roughly 1 to 2 mm in diameter (Sauer 1955). When planted in June, Palmer amaranth growth rates were 0.21 and 0.18 cm per growing degree day across 2 yr (Horak and Loughin 2000).

Female Palmer amaranth plants produce copious amounts of seed even when growing in competitive environments or when they emerge late in the growing season (Ward et al. 2013). Female plants can produce over 200,000 seeds without competition if germination occurs between March and June (Keely et al. 1987). If season-long control of Palmer amaranth is not achieved, the soil seedbank is rapidly replenished due to Palmer amaranth's prolific seed production (Keely et al. 1987; Sellers et al. 2003). An advantage of Palmer amaranth growing in competitive environments is its ability to germinate within a day of favorable temperatures approximately 30 C, whereas some

other *Amaranthus* spp. require multiple days to germinate under favorable conditions (Steckel et al. 2004; Guo and Al-Khatib 2003).

Twelve weed species in Mississippi have been documented resistant to herbicides (Heap 2017). Georgia was the first state to report failure to control Palmer amaranth with glyphosate in 2004 (Culpepper et al. 2006). Arkansas reported a failure to control Palmer amaranth with glyphosate in 2005 (Norsworthy et al. 2008). In 2008, three accessions of Palmer amaranth from northeast Arkansas were evaluated for their response to various herbicides (Norsworthy et al. 2008). Glyphosate controlled the GR accession 73% 28 d after treatment (DAT), while other herbicides with different MOA resulted in $\geq 97\%$ control. Palmer amaranth in Mississippi is resistant to glyphosate and acetolactate synthase (ALS) inhibitors, such as pyriithiobac (Nandula et al. 2012). In Mississippi, the first glyphosate-resistant Palmer amaranth populations exhibited 14- to 17-fold resistance to glyphosate (Nandula et al. 2012). Nandula et al. (2012) reported 100% control with paraquat, glufosinate, and fomesafen across three biotypes. However, trifloxysulfuron, pyriithiobac, and chlorimuron all failed to control the resistant biotypes $>87\%$ (Nandula et al 2012). Yield reductions in cotton (Norsworthy et al. 2014), corn (*Zea mays* L.) (Massinga et al. 2001), and soybean (Klingaman and Oliver 1994) have been correlated with Palmer amaranth interference. Soybean yield reductions of 64 and 68% were reported with Palmer amaranth densities from 2 to 3.33 m⁻¹ of row (Klingaman and Oliver 1994).

Glyphosate-resistant Palmer amaranth can be managed in soybean with herbicides other than glyphosate and ALS inhibitors if timely applications are utilized (Whitaker et al. 2010). Protoporphyrinogen oxidase (PPO) inhibitors have become a staple group of

herbicides used for the PRE and POST control of problematic weeds in soybean. Fomesafen is a member of the diphenylether family of PPO inhibitors labeled in soybean (Stephenson et al. 2004; MSU-ES 2017). Fomesafen controls common cocklebur (*Xanthium strumarium* L.), prickly sida (*Sida spinosa* L.), and Palmer amaranth (Stephenson et al. 2004). Soybean yields were higher when fomesafen was applied POST to Palmer amaranth (Whitaker et al. 2010). Norsworthy et al. (2008) reported that glyphosate plus fomesafen controlled three accessions of Palmer amaranth 100%. The Mississippi State University Extension Service has published management strategies for GR Palmer amaranth (Bond et al. 2015; MSU-ES 2017). Fomesafen in combination with another MOA is recommended at 0.264 to 0.394 kg ai ha⁻¹ plus NIS at 0.25% (v/v) to control GR Palmer amaranth. (Bond et al. 2015; MSU-ES 2017). Bond et al. (2006) reported >96% Palmer amaranth control 21 DAT with fomesafen at 0.420 kg ha⁻¹. Palmer amaranth biomass reduction was >94% across accessions from Alabama, Arkansas, Kansas, Louisiana, Mississippi, Missouri, New Mexico, South Carolina, Tennessee, and Texas (Bond et al. 2006).

Barnyardgrass is one of the more problematic weeds in U.S. soybean production (Bagavathiannan et al. 2011; Holm et al. 1977). In 10 southern U.S. states surveyed in 1977, only Mississippi ranked barnyardgrass as one of the most troublesome weeds in soybean (McCormick 1977). By 2013, two of 10 states ranked barnyardgrass as one of the most troublesome weeds in soybean (Webster 2013). Mississippi and Arkansas ranked barnyardgrass as the fourth and eighth, respectively, most troublesome weed (Webster 2013).

Barnyardgrass is characterized as a summer annual reaching up to 2 m tall (Bryson and DeFelice 2009). Barnyardgrass sheath and leaves are glabrous and the plant has no ligule (Bryson and DeFelice 2009). The spikelets on mature plants should be crowded and rebranched (Bryson and DeFelice 2009). Seeds may be awned or awnless (Bryson and DeFelice 2009). Barnyardgrass has a range extending across the entire U.S. and as far north as central Canada (Bryson and DeFelice 2009).

Barnyardgrass is a troublesome weed around the world with resistance reported to 11 herbicide MOA (Heap 2017). Recently, Tennessee became the first state in the U.S. to confirm GR barnyardgrass (Steckel et al. 2017). This is particularly concerning for other midsouthern states that border Tennessee because a barnyardgrass population in Mississippi has developed resistance to several modes of action (Heap 2017; Wright et al. 2016). Possible GR barnyardgrass samples have been evaluated in Mississippi, but none have tested positive for resistance (Bond 2017). Overexposure to glyphosate can lead to resistance (Bagavathiannan et al. 2013). Bagavathiannan et al. (2013) reported glyphosate resistance evolution in barnyardgrass would occur by 2022 according to a model including five annual glyphosate applications in continuous GR cotton; however, by rotating to GR corn or glufosinate-resistant cotton cultivars, glyphosate resistance could be delayed up to six additional years.

Due to the wide adoption of GR crops, herbicide efficacy, and the prolonged emergence of barnyardgrass, glyphosate has been a principal herbicide for barnyardgrass control (Krausz et al. 2001; Sikkema et al. 2005; Riar et al. 2013). Sikkema et al. (2005) reported 82, 97, and 98% reduction in barnyardgrass density, dry weight, and seed production, respectively, 84 d after an application of glyphosate at 0.450 kg ae ha⁻¹ in GR

corn. When three consecutive applications of glyphosate at 1.06 kg ha⁻¹ were applied at the three-, seven-, and 14-leaf growth stages in GR-cotton, barnyardgrass control was 92% (Scroggs et al. 2007). In glyphosate-susceptible crops, acetyl CoA Carboxylase (ACCase), ALS, and photosystem II (PSII) inhibitors are common modes of action used to control barnyardgrass (MSU-ES 2017; Reddy 2003; Wilson et al. 2014).

Plant health management is the practice of understanding and overcoming several factors limiting plants from achieving their full genetic potential (Cook 2000). This concept can be applied to crops, trees, or any other plant (Cook 2000). The full genetic potential of a plant is a known or approximated capacity of a plant to grow, develop, and reproduce without limiting factors (Cook 2000). Breeding has increased the genetic potential of plants; however, plant health management focuses on improving upon the preexisting potential and not the modifications developed through breeding and genetic engineering (Cook 2000). Four major factors influencing plant health management include the use of high-quality seed, optimum fertility, irrigation, and proper pest management strategies (Cook 2000).

To sustain an acceptable level of soybean productivity each year, soil nutrients removed from the previous year's crop must be replenished (Varco 1999). In soybean, some nutrients are taken in during specific growth stages and others over the course of the growing season (Bender et al. 2015; Varco 1999). Nitrogen (N) is needed in the greatest quantity among all soil nutrients (Varco 1999). It is important to supply any field with the proper amount of N to prevent slowed growth and premature senescence where a field is deficient or delayed maturity where there is an excess (Varco 1999). Due to the symbiotic relationship between the bacterium *Bradyrhizobium japonica* and

soybean, soybean will fix N either within the soil or from the air through biological fixation (Varco 1999). Varco (1999) reported the relationship of applying fertilizer N to soybean yield is unpredictable, likely due to N fertilization repressing N fixation, variability in soil N-supplying capacity, soil water availability, and general environmental conditions.

Foliar fertilizers are routinely applied in a variety of crops to aid in plant health management (Clapp and Small 1968; Garcia and Hanway 1976; Mallarino et al. 2001; Poole et al. 1983). Research has shown inconsistent soybean and cotton responses to foliar fertilizers; possibly due to testing in the absence of a deficiency (Haq and Mallarino 2000; Mallarino et al. 2001; Yin et al. 2011). Soybean yield increases attributed to foliar fertilizer applications have been small and infrequent (Haq and Mallarino 2000). Garcia and Hanway (1976) reported a soybean yield increase with a 10-1-3-0.5 liquid nitrogen-phosphorous-potassium-sulfur (N-P-K-S) fertilizer applied at the R5 to R6 growth stages. Haq and Mallarino (1998) documented greater soybean yields compared with the nontreated following various rates of 3-8-15 (N-P-K) fertilizers applied at the V5 growth stage. Most research suggests no soybean yield increase with foliar S or micronutrients applied at reproductive stages (Clapp and Small 1968; Poole et al. 1983). The addition of micronutrients boron (B), iron (Fe), and zinc (Zn) to an N-P-K-S (10-4-8-1) fertilizer failed to improve soybean yields (Mallarino et al. 2001). Other research reported reduction in soybean yield following foliar fertilization with observed reduction attributed to leaf injury from the application (Haq and Mallarino 2000).

Plant growth regulators (PGRs) are used as plant health management tools in several crops, especially cotton (Ren et al. 2013). Mepiquat chloride is commonly

applied to cotton to control vegetative growth and prevent shading which causes fruit abscission and reduced yield (Guinn 1974). Some herbicides are also used as PGRs. Glyphosate applied at low rates has been used to suppress flowering and stimulate sucrose accumulation in sugarcane [*Saccharum spontaneum* (L.)] (Bennet and Montes 2003; Velini et al. 2010).

Cytokinin mixtures are available as PGRs for use in several crops. Cytokinins occur naturally in plants and are responsible for cell division and enlargement as well as the formation of flowers and fruits (Skoog and Armstrong 1970). Cytokinins have been reported to increase soybean cell proliferation in a tissue culture (Fosket and Short 1973). Kinetin, a specific cytokinin, has been reported to reverse the effect NaCl has on tobacco [*Nicotiana tabacum* (L.)] leaves when applied in solution to a tissue culture (Katz et al. 1978). Chemical manufacturers claim these mixtures improve vigor, promote root and shoot growth, reduce stress, and slow leaf aging (Anonymous 2017a; Anonymous 2017b). Data supporting the claimed benefits of applying cytokinin mixtures is limited. Most research detailing the effects of kinetin and other cytokinin mixtures have been contained to a tissue culture.

The option to mix different herbicide MOA provides the potential for increased weed control and a reduction in application costs (Hydrick and Shaw 1994). However, some components of herbicide mixtures can synergize or antagonize others. Synergism is the simultaneous action of two or more components in which the total response of the combination is greater than the sum of the individual components (Nash 1981). Antagonism is reported when the total response is less than the sum of the individual components (Nash 1981). Interactions between components (water, foliar fertilizers, and

other herbicides) of herbicide mixtures have been documented throughout the literature (Devkota and Johnson 2016b; Mahoney et al. 2014; Scroggs et al. 2009; Starke and Oliver 1998; Roskamp et al 2013; Vidrine et al. 1995).

Tests for synergistic, antagonistic, and additive responses have evolved over time. Eshel et al. (1976) reported synergistic effects on wild oat [*Avena fatua* (L.)] and wild mustard [*Sinapis arvensis* (L.)] control utilizing Colby's method (Colby 1967). The Blouin et al. (2004) nonlinear model was utilized by Webster et al. (2006) to evaluate a safening interaction on rice [*Oryza sativa* (L.)] treated with clomazone mixed with bensulfuron or halosulfuron. Blouin et al. (2010) expanded on the nonlinear model creating the augmented mixed-model methodology providing a more versatile model than Blouin et al. (2004). The augmented mixed-model methodology has been utilized by Fish et al. (2016) to determine synergistic and antagonistic effects on red rice [*Oryza sativa* (L.)] and barnyardgrass control when applying mixtures of propanil and imazamox.

Results of herbicide-by-herbicide interactions are abundant throughout the literature. Fish et al. (2016) reported synergism on red rice control with propanil and imazamox mixtures; however, the same mixtures antagonized barnyardgrass control. Minton et al. (1989) documented antagonism on barnyardgrass control when the ACCase inhibitors quizalofop or sethoxydim were combined with the PPO inhibitor lactofen. Starke and Oliver (1998) reported antagonism when fomesafen was combined with glyphosate on entireleaf morningglory [*Ipomoea hederacea* var. *integriuscula* (Gray)] but not on pitted morningglory [*Ipomoea lacunosa* (L.)].

Water is the primary carrier for herbicide applications (Roskamp et al. 2013). Because water is rarely found in pure form, other substances such as cations can be dissolved in water (Roskamp et al. 2013). The concentration of calcium (Ca) and magnesium (Mg) in water is referred to as the degree of water hardness (Roskamp et al. 2013). Herbicides such as dicamba, 2,4-D, and glyphosate are weak acids and can be affected by water hardness (Abouzienna et al. 2009; Buhler and Burnside 1983; Roskamp et al. 2013; Shilling and Haller 1989; Wills and McWhorter 1985). Glyphosate and many other aminopolyacids tend to form stable complexes with di- and trivalent cations such as Ca^{2+} and Mg^{2+} and Fe^{3+} (Glass 1984; Lundager Madsen et al. 1978; Thelen et al 1995). The cations in hard water can also be components of or utilized as foliar fertilizers. Herbicide efficacy in the presence of cations can also be affected by the targeted weed species (Mueller et al. 2006). Antagonism of glyphosate with manganese (Mn) and Zn foliar fertilizers has been documented across several weed species (Abouzienna et al. 2009; Bernards et al. 2005; Scroggs et al. 2009).

Fomesafen and lactofen are common treatments for Palmer amaranth control in soybean, but soybean injury is often observed following POST applications (Johnson et al. 2002; Mangialardi et al. 2016; MSU-ES 2017). In an effort to reduce application costs and decrease soybean injury, growers commonly add foliar fertilizers to POST herbicide applications (Bernards et al. 2005; Devkota et al. 2016a). Some cytokinin mixture labels do not mention tank mix partners beyond discussing the use of a surfactant based on experience or professional opinion (Anonymous 2017a; Anonymous 2017b). In order to reduce application costs by reducing trips through the field, growers may combine POST herbicides and cytokinin mixtures. Previous research is limited on detailing the

interaction between herbicides and foliar fertilizers or cytokinin mixtures. Therefore, research was conducted to determine the influence foliar fertilizers or cytokinin mixtures in combination with common POST soybean herbicides.

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CHAPTER II
AN EVALUATION ON THE INFLUENCE OF FOLIAR FERTILIZER IN
COMBINATION WITH COMMON POSTEMERGENCE
SOYBEAN HERBICIDES

Abstract

Field studies conducted in 2015 and 2016 in Stoneville, MS, evaluated the impact on soybean injury, weed control, and agronomic performance when combining blended or single-nutrient foliar fertilizers with POST herbicide applications. In the Weed Control Study, 14 antagonistic effects at various evaluations were detected on Palmer amaranth and barnyardgrass control when a blended foliar fertilizer (0.39 kg ai ha⁻¹ or 0.78 kg ha⁻¹) was mixed with glyphosate (1.37 kg ae ha⁻¹) alone or combined with *S*-metolachlor (1.42 kg ai ha⁻¹), fomesafen (0.395 kg ai ha⁻¹), or lactofen (0.128 kg ai ha⁻¹). Blended foliar fertilizer did not influence soybean injury. In the Agronomic Study, blended foliar fertilizer did not impact soybean injury, height, dry weight, nutrient concentration, or yield. In the Single-nutrient Foliar Fertilizer Study, Palmer amaranth and barnyardgrass control 7 DAT with glyphosate was reduced by zinc and manganese, respectively. Manganese reduced barnyardgrass control 14 DAT compared with treatments lacking a single-nutrient foliar fertilizer. These combinations of foliar fertilizer and POST herbicides should be avoided.

Nomenclature: Glyphosate; fomesafen; lactofen; S-metolachlor; Palmer amaranth *Amaranthus palmeri* S. Watts AMAPA; Barnyardgrass *Echinochloa crus-galli* L. Beauv. ECHCG; soybean, *Glycine max* L. Merr.

Keywords: Antagonism, foliar fertilizer rate, plant health management

Introduction

Amaranthus spp., known collectively as pigweeds, belong to the family *Amaranthaceae* and have ranked among the top ten most troublesome weeds in southern U.S. soybean since the early 1970's (Anonymous 1972; Buchanan 1973, 1974). Palmer amaranth increased in severity in the southern states from 1995 to 2008, ascending from the number 10 to the number one most troublesome weed in cotton [*Gossypium hirsutum* (L.)] and from 23 to number two in soybean (Webster and Nichols 2012). Nine of ten southern U.S. states surveyed ranked Palmer amaranth as the most troublesome weed of cotton in 2012 (Webster and Nichols 2012). Among eight southern U.S. states surveyed in 2013, Palmer amaranth was ranked as the most troublesome weed of cotton and soybean in seven and three states, respectively (Webster 2013).

Glyphosate-resistant (GR) Palmer amaranth is one of twelve weed species in Mississippi to be documented as herbicide-resistant (Heap 2017). Georgia was the first state to report glyphosate resistance in Palmer amaranth in 2004 (Culpepper et al. 2006), and this was followed with GR Palmer amaranth documentation in Arkansas (Norsworthy et al. 2008). Palmer amaranth in Mississippi was confirmed resistant to glyphosate and acetolactate synthase (ALS) inhibitors in 2008 (Heap 2017). In Mississippi, the first GR Palmer amaranth populations exhibited 14- to 17-fold resistance to glyphosate (Nandula et al. 2012).

Glyphosate-resistant Palmer amaranth can be managed in soybean with herbicides other than glyphosate and ALS inhibitors if timely applications are utilized (Whitaker et al. 2010). Protoporphyrinogen oxidase (PPO) inhibitors have become a staple group of herbicides used for PRE and POST control of problematic weeds in soybean. Fomesafen is a member of the diphenylether family of PPO inhibitors labeled in soybean, and it controls common cocklebur (*Xanthium strumarium* L.), prickly sida (*Sida spinosa* L.), and Palmer amaranth (Stephenson et al. 2004). Soybean yields were greater when fomesafen was applied POST to Palmer amaranth (Whitaker et al. 2010). Norsworthy et al. (2008) reported that glyphosate plus fomesafen controlled three accessions of GR Palmer amaranth 100%. Bond et al. (2006) reported >96% Palmer amaranth control 21 DAT with fomesafen at 0.420 kg ha⁻¹. Nandula et al. (2012) reported 100% control of three glyphosate- and ALS-resistant biotypes with paraquat, glufosinate, and fomesafen.

Barnyardgrass is one of the more problematic weeds in U.S. soybean production (Bagavathiannan et al. 2011; Holm et al. 1977). In ten southern U.S. states surveyed in 1977, only Mississippi ranked barnyardgrass as one of the most troublesome weeds in soybean (McCormick 1977). Mississippi and Arkansas ranked barnyardgrass as the fourth and eighth, respectively, most troublesome weed of soybean in 2013 (Webster 2013).

Barnyardgrass is a troublesome weed around the world with resistance reported to 11 herbicide MOA (Heap 2017). Recently, Tennessee became the first state in the U.S. to confirm GR barnyardgrass (Steckel et al. 2017). This is particularly concerning for other midsouthern states that border Tennessee because a barnyardgrass population in Mississippi has developed resistance to several modes of action (Heap 2017; Wright et al.

2016). Possible GR barnyardgrass samples have been evaluated in Mississippi, but none have tested positive for resistance (Bond 2017). Overexposure to glyphosate can lead to resistance (Bagavathiannan et al. 2013). Bagavathiannan et al. (2013) reported glyphosate resistance evolution in barnyardgrass would occur by 2022 according to a model including five annual glyphosate applications in continuous GR cotton; however, by rotating to GR corn (*Zea mays* L.) or glufosinate-resistant cotton cultivars, glyphosate resistance could be delayed up to six additional years.

Due to the wide adoption of GR crops, herbicide efficacy, and the prolonged emergence of barnyardgrass, glyphosate has been a principal herbicide for barnyardgrass control (Krausz et al. 2001, Sikkema et al. 2005; Riar et al. 2013). Sikkema et al. (2005) reported 82, 97, and 98% reduction in barnyardgrass density, dry weight, and seed production, respectively, 84 d after an application of glyphosate at 0.450 kg ae ha⁻¹ in GR corn. When three consecutive applications of glyphosate at 1.06 kg ha⁻¹ were applied at the three-, seven-, and 14-leaf growth stages in GR-cotton, barnyardgrass control was 92% (Scroggs et al. 2007). In glyphosate-susceptible crops, acetyl CoA Carboxylase (ACCase), ALS, and photosystem II (PSII) inhibitors are common modes of action used to control barnyardgrass (MSU-ES 2017; Reddy 2003; Wilson et al. 2014).

Plant health management is the practice of understanding and overcoming several factors limiting plants from achieving their full genetic potential (Cook 2000). This concept can be applied to crops, trees, or any other plant (Cook 2000). The full genetic potential of a plant is a known or approximated capacity of a plant to grow, develop, and reproduce without limiting factors (Cook 2000). Breeding has increased the genetic potential of plants; however, plant health management focuses on improving upon the

preexisting potential and not the modifications developed through breeding and genetic engineering (Cook 2000). Four major factors influencing plant health management include the use of high-quality seed, optimum fertility, irrigation, and proper pest management strategies (Cook 2000).

Foliar fertilizers are routinely applied in a variety of crops to aid in plant health management (Clapp and Small 1968; Garcia and Hanway 1976; Mallarino et al. 2001; Poole et al. 1983). Research has shown inconsistent soybean and cotton responses to foliar fertilizers (Haq and Mallarino 2000; Mallarino et al. 2001; Yin et al. 2011); however, deficiencies may have been absent. Soybean yield increases attributed to foliar fertilizer applications have been small and infrequent (Haq and Mallarino 2000). Garcia and Hanway (1976) reported a soybean yield increase with a 10-1-3-0.5 liquid nitrogen-phosphorous-potassium-sulfur (N-P-K-S) fertilizer applied at the R5 to R6 growth stages. Haq and Mallarino (1998) documented greater soybean yields compared with the nontreated following various rates of 3-8-15 (N-P-K) fertilizers applied at the V5 growth stage. Most research suggests no soybean yield increase with foliar S or micronutrients applied at reproductive stages (Clapp and Small 1968; Poole et al. 1983). The addition of micronutrients boron (B), iron (Fe), and zinc (Zn) to an N-P-K-S (10-4-8-1) fertilizer failed to improve soybean yields (Mallarino et al. 2001). Other research reported reduction in soybean yield following foliar fertilization with observed reduction attributed to leaf injury from the application (Haq and Mallarino 2000).

The option to mix different herbicide MOA provides the potential for increased weed control and a reduction in application costs (Hydrick and Shaw 1994). However, some components of herbicide mixtures can synergize or antagonize others. Synergism

is the simultaneous action of two or more components in which the total response of the combination is greater than the sum of the individual components (Nash 1981).

Antagonism is reported when the total response is less than the sum of the individual components (Nash 1981). Interactions between components (water, foliar fertilizers, and other herbicides) of herbicide mixtures have been documented throughout the literature (Devkota and Johnson 2016b; Mahoney et al. 2014; Scroggs et al. 2009; Starke and Oliver 1998; Roskamp et al 2013; Vidrine et al. 1995).

Tests for synergistic, antagonistic, and additive responses have evolved over time. Eshel et al. (1976) reported synergistic effects on wild oat [*Avena fatua* (L.)] and wild mustard [*Sinapis arvensis* (L.)] control utilizing Colby's method (Colby 1967). The Blouin et al. (2004) nonlinear model was utilized by Webster et al. (2006) to evaluate a safening interaction on rice [*Oryza sativa* (L.)] treated with clomazone mixed with bensulfuron or halosulfuron. Blouin et al. (2010) expanded on the nonlinear model creating the augmented mixed-model methodology providing a more versatile model than Blouin et al. (2004). The augmented mixed-model methodology has been utilized by Fish et al. (2016) to determine synergistic and antagonistic effects on red rice [*Oryza sativa* (L.)] and barnyardgrass control when applying mixtures of propanil and imazamox.

Results of herbicide-by-herbicide interactions are abundant throughout the literature. Fish et al. (2016) reported synergism on red rice control with propanil and imazamox mixtures; however, the same mixtures antagonized barnyardgrass control. Minton et al. (1989) documented antagonism on barnyardgrass control when the ACCase inhibitors quizalofop or sethoxydim were combined with the PPO inhibitor lactofen.

Starke and Oliver (1998) reported antagonism when fomesafen was combined with glyphosate on entireleaf morningglory [*Ipomoea hederacea* var. *integriscula* (Gray)] but not on pitted morningglory [*Ipomoea lacunosa* (L.)].

Water is the primary carrier for herbicide applications (Roskamp et al. 2013). Because water is rarely found in pure form, other substances such as cations can be dissolved in water (Roskamp et al. 2013). The amount of calcium (Ca) and magnesium (Mg) in water is referred to as the degree of water hardness (Roskamp et al. 2013). Herbicides such as dicamba, 2,4-D, and glyphosate are weak acids and can be affected by water hardness (Abouzienna et al. 2009; Buhler and Burnside 1983; Roskamp et al. 2013; Shilling and Haller 1989; Wills and McWhorter 1985). Glyphosate and many other aminopolyacids tend to form stable complexes with di- and trivalent cations such as Ca^{2+} and Mg^{2+} and Fe^{3+} (Glass 1984; Lundager Madsen et al. 1978; Thelen et al 1995). The cations in hard water can also be components of or utilized as foliar fertilizers. Herbicide efficacy in the presence of cations can also be affected by the targeted weed species (Mueller et al. 2006). Antagonism of glyphosate with manganese (Mn) and Zn foliar fertilizers has been documented across several weed species (Abouzienna et al. 2009; Bernards et al. 2005; Scroggs et al. 2009).

Fomesafen and lactofen are common treatments for Palmer amaranth control in soybean, but soybean injury is often observed following POST applications (Johnson et al. 2002; Mangialardi et al. 2016; MSU-ES 2017). In an effort to reduce application costs and decrease soybean injury, growers commonly add foliar fertilizers to POST herbicide applications (Bernards et al. 2005; Devkota et al. 2016a). Due to a limited amount of research on the interaction between herbicides and foliar fertilizers, three field studies

were conducted detailing the impact of adding a blended foliar fertilizer to POST soybean herbicide applications. The objectives of these studies were to (1) evaluate the influence of a blended foliar fertilizer on soybean injury and weed control with POST herbicides, (2) to characterize soybean agronomic performance following POST applications of mixtures of herbicides and a blended foliar fertilizer, and (3) identify a single-nutrient foliar fertilizer that antagonizes weed control when mixed with glyphosate plus fomesafen.

Materials and Methods

Weed Control Study

A field study was conducted at the Mississippi State University Delta Research and Extension Center in Stoneville, MS, in 2015 and 2016 to evaluate herbicide efficacy when a blended foliar fertilizer was added to POST herbicides in soybean. The study was performed at two sites in 2015 (33°26'29.18"N 90°54'41.92"W and 33°24'21.94"N 90°55'31.27"W) and 2016 (33°26'28.33"N 90°54'23.67"W and 33°24'21.94"N 90°55'31.27"W). In 2015, soil at one site was a Dundee very fine sandy loam (Fine-silty, mixed, active, thermic Typic Endoaqualfs) with a pH of 6.1 and 1.2% organic matter, and soil at the second site was a Newellton silty clay (Clayey over loamy, smectitic over mixed, superactive, nonacid, thermic Fluvaquentic Epiaquepts) with a pH of 6.9 and 1.6% organic matter. In 2016, soil at one site was a Commerce sandy clay loam (Fine-silty, mixed, superactive, nonacid, thermic Fluvaquentic Endoaquepts) with a pH of 6.8 and 1.6 % organic matter while that at the second was the Newelton silty clay loam utilized in 2015. The experimental sites were known to be heavily infested with barnyardgrass and Palmer amaranth. Each site was conventionally tilled prior to planting

to stimulate weed germination and ensure uniform weed emergence. ‘Asgrow 4632’ (Monsanto Company 800 N. Lindbergh Blvd. St. Louis, MO 63167) mid-maturity group IV soybean were utilized in all siteyears and sowed with a John Deere small-plot air planter (John Deere 1730, Deere and Company, One John Deere Place Moline, IL, 61265-8098).

The study was designed as a two-factor factorial within a randomized complete block with four replications. Factor A was herbicide treatment and included no herbicide, glyphosate (Monsanto Company, 800 N. Lindbergh Blvd., St. Louis, MO 63167) at 1.36 kg ha⁻¹ alone and in combination with *S*-metolachlor (Syngenta Crop Protection, LLC, P. O. Box 18300, Greensboro, NC 27419) at 1.42 kg ai ha⁻¹, fomesafen (Syngenta Crop Protection, LLC, P. O. Box 18300, Greensboro, NC 27419) at 0.375 kg ha⁻¹, and lactofen (Valent U.S.A. Corporation, P. O. Box 8025, Walnut Creek, CA 94596) at 0.128 kg ai ha⁻¹. Factor B was foliar fertilizer rate and consisted of a blended foliar fertilizer with a 4-0-0-3-3-3-0.25%, N-P-K-S-Mn-Zn-B guaranteed analysis (Brandt Consolidated, Inc., 2935 South Koke Mill Road, Springfield, IL 62711) applied at 0, 0.39, and 0.78 kg ha⁻¹. Treatments were applied with a tractor-mounted sprayer calibrated to deliver 140 L ha⁻¹ at 248 kPa, fitted with extended range flat-fan (XR10002 TeeJet® P.O. Box 7900 Wheaton, IL 60187) nozzles at the V3 soybean growth stage.

Visual estimates of soybean injury and weed control were recorded on a scale from 0 to 100% with 0 representing no injury or control and 100 representing soybean death or complete weed control. Soybean injury was evaluated 3, 7, 14, 21, and 28 d after treatment (DAT) and control of Palmer amaranth and barnyardgrass was evaluated 7, 14, 21, and 28 DAT. Heights of five soybean plants in each plot were measured from

the ground to the uppermost node 14 DAT and at maturity. Soybean were harvested using a small-plot combine (Kincaid Equipment, 210 West First St., P.O. Box 400; Haven, KS) on September 25 and October 5 in 2015 and September 16 and October 12 in 2016. Yield data were adjusted to 13% moisture content.

Square roots of visual injury and control estimates were arcsine transformed. The transformation did not improve the homogeneity of the variance based on visual inspection of the plotted residuals; therefore, nontransformed data were used in all analyses. Soybean injury and weed control data were analyzed utilizing the augmented mixed-model methodology described by Blouin et al. (2010). Data for soybean height and yield were subjected to ANOVA using the PROC MIXED procedure in SAS 9.4 (SAS Institute Inc. 100 SAS Campus Drive Cary, NC 27513-2414) with siteyear, replication (nested within siteyear), and treatment-by-rep interactions listed as the random effect parameters (Blouin et al. 2011). Type III Statistics were utilized to test the fixed effects of herbicide and foliar fertilizer. Least square means were calculated and mean separation ($p \leq 0.05$) was produced using PDMIX800 in SAS, which is a macro for converting mean separation output to letter groupings (Saxton 1998). When injury and weed control data did not return a significant synergistic or antagonistic effect (Blouin et al. 2010), data were analyzed as described for soybean height and yield.

Agronomic Study

A field study was conducted at the Mississippi State University Delta Research and Extension Center in Stoneville, MS, in 2015 and 2016 to evaluate soybean response when adding a foliar fertilizer to POST herbicides. The study was performed at two sites in 2015 (33°25'6.68"N 90°54'3.44"W and 33°24'54.02"N 90°54'3.44"W) and 2016

(33°25'6.68"N 90°54'3.44"W and 33°26'0.99"N 90°54'31.52"W). In 2015, soil at both sites was a Dundee very fine sandy loam with a pH of 6.1 and 1.2% organic matter. In 2016, one of the sites was the previously described Dundee very fine sandy loam and the second was a Commerce very fine sandy loam with a pH of 6.9 and 0.6% organic matter. Each site was conventionally tilled, then planted with a John Deere small-plot air planter. 'Pioneer 48T53' (Pioneer Hi-Bred P.O. Box 1000 Johnston, IA 50131-0184) and Asgrow 4632 mid-maturity group IV soybean were sowed in 2015 and 2016, respectively.

The treatment structure and experimental design for the Agronomic Study was the same as that for the Weed Control Study. However, the Agronomic Study was maintained weed-free each site year to prevent weed interference with soybean agronomic performance. Plots were hand-weeded or treated with POST and residual herbicides applied with a hooded sprayer (Willmar Fabrication 2205 Hall Ave. Benson, MN 56215) to prevent foliar soybean injury.

Visual estimates of soybean injury were recorded 3, 7, 14, 21, and 28 DAT on the previously described scale. Soybean heights were recorded 14 DAT and at maturity as previously described. Soybean biomass was collected from 1-m sections of rows 1 and 4 in each plot 14 DAT. Soybean biomass samples were dried at 60 C for one wk and weights converted to g m⁻². Ten trifoliolate leaves were collected from the second uppermost, fully expanded trifoliolate of plants in rows 2 and 3 14 DAT for tissue analysis. Tissue samples were air-dried in the greenhouse and sent to Waypoint Analytical (Waypoint Analytical Corporate Headquarters, 2790 Whitten Rd., Memphis, TN 38133) for analysis. Tissue samples were digested with concentrated nitric acid (HNO₃) and 30% hydrogen peroxide (H₂O₂) and analyzed by inductively coupled plasma atomic

emission spectroscopy (ICP-AES) for nutrient concentration (Jones and Case. Soybean were harvested using a small-plot combine on October 5, 2015, and September 27 and October 3, 2016. Yield data were adjusted to 13% moisture content. Data analyses were the same as in the Weed Control Study.

Single-nutrient Foliar Fertilizer Study

A field study was conducted at the Mississippi State University Delta Research and Extension Center in Stoneville, MS, in 2016 to evaluate the impact on herbicide efficacy when combining POST herbicides with single nutrients represented in the blended foliar fertilizer utilized in the previous studies. The study was repeated in space (33°26'28.33"N 90°54'23.67"W and 33°24'21.94"N 90°55'31.27"W). Soils, field preparation, and planting at each site were the same as described for 2016 sites in the Weed Control Study.

The study was designed as a two-factor factorial within a randomized complete block with four replications. Factor A was herbicide treatment and consisted of no herbicide, glyphosate at 1.36 kg ha⁻¹ alone and in combination with fomesafen at 0.375 kg ha⁻¹. Factor B was single-nutrient foliar fertilizer and consisted of no fertilizer, N at 0.235 kg N ha⁻¹, citric acid-chelated Zn at 0.175 kg Zn ha⁻¹, Mn derived from manganese sulfate at 0.175 kg Mn ha⁻¹, and B derived from boric acid at 0.015 kg B ha⁻¹. Treatment application, data collection, and data analyses were as previously described in the Weed Control Study.

Results and Discussion

Weed Control Study

No synergistic or antagonistic effects were detected for soybean injury at any evaluation interval. A main effect of herbicide treatment was detected for soybean injury at 3, 7, and 14 DAT (Table 2.1). Pooled across foliar fertilizer rates, glyphosate plus lactofen injured soybean more than other herbicide treatments 3, 7, and 14 DAT (Table 2.1). Glyphosate plus S-metolachlor injured soybean more than glyphosate alone, but not as severely as glyphosate plus fomesafen (Table 2.1). Bronzing and necrosis of plant tissue due to lactofen and fomesafen has been well-documented (Johnson et al. 2002; Mangialardi et al. 2016). By 21 and 28 DAT, no soybean injury was observed across all herbicide treatments (data not presented). The addition of foliar fertilizer to POST herbicide treatments did not influence soybean injury.

Palmer amaranth control with glyphosate alone was antagonized 7% at 7 DAT by the addition of foliar fertilizer at 0.39 kg ha⁻¹ and 11 and 13% at 14 and 21 DAT, respectively, by adding foliar fertilizer at 0.78 kg ha⁻¹ (Tables 2.2, 2.3, and 2.4). Control with glyphosate plus S-metolachlor was antagonized $\geq 11\%$ by adding foliar fertilizer at 0.39 or 0.78 kg ha⁻¹ 7 and 14 DAT (Tables 2.2 and 2.3). Bernards et al. (2005) documented antagonism on velvetleaf [*Abutilon theophrasti* (Medik)] control when glyphosate at 0.28 kg ha⁻¹ was combined with various formulations of Mn; however, some of the antagonistic effects were overcome by adding ammonium sulfate at 20 g L⁻¹. It should also be noted that the populations of Palmer amaranth in all siteyears were not completely resistant to glyphosate, because glyphosate alone provided 66% control 7 DAT. A herbicide main effect influenced Palmer amaranth control 28 DAT (Table 2.1).

Pooled over foliar fertilizer rates, Palmer amaranth control with glyphosate plus fomesafen was at least 7% greater than with all other herbicide treatments (Table 2.1). Palmer amaranth control with fomesafen has been well-documented; however, published research detailing control with glyphosate plus fomesafen is limited (Barkley et al. 2016; Everman et al. 2009; Miller and Norsworthy 2016; Whitaker et al. 2010). Miller and Norsworthy (2016) reported 93% Palmer amaranth control with glyphosate plus fomesafen and 2,4-D 14 DAT. Barkley et al. (2016) documented $\geq 90\%$ control of Palmer amaranth with varying rates of fomesafen alone 28 d after transplanting sweet potato [*Ipomoea batatas* (L.) Lam.]. Palmer amaranth control with glyphosate alone was 56% and less than that with all other herbicide treatments 28 DAT (Table 2.1). Since glyphosate is a POST herbicide lacking residual control, it should be expected that the residual activity from fomesafen and *S*-metolachlor would control Palmer amaranth better than glyphosate alone 28 DAT (Anonymous 2013; Anonymous 2015; MSU-ES 2017). Similar to glyphosate, there is no residual control with lactofen; however, glyphosate plus lactofen controlled Palmer amaranth 18 and 21% better than glyphosate alone 7 and 14 DAT, respectively (data not presented). Palmer amaranth control with glyphosate alone and glyphosate plus *S*-metolachlor was similar at all evaluations prior to 28 DAT (data not presented).

Barnyardgrass control was antagonized when foliar fertilizer at 0.78 kg ha⁻¹ was mixed with glyphosate alone 7 and 21 DAT, glyphosate plus fomesafen 14 and 21 DAT, and glyphosate plus *S*-metolachlor 14 DAT (Tables 2.5, 2.6, 2.7), and differences between the observed and expected levels of control ranged from 6 to 10%. Antagonism for barnyardgrass control was also detected when foliar fertilizer at 0.39 kg ha⁻¹ was

mixed with glyphosate plus fomesafen 14 DAT and glyphosate plus lactofen 21 DAT, and differences between the observed and expected levels of control were 9 and 6% for mixtures of glyphosate plus fomesafen or lactofen, respectively (Tables 2.6 and 2.7).

A main effect of herbicide treatment was also detected for barnyardgrass control 28 DAT (Table 2.1). Barnyardgrass control 28 DAT was greatest (82%) with glyphosate plus *S*-metolachlor (Table 2.1) due to its residual control of small-seeded broadleaves and grasses (Anonymous 2015). Scroggs et al. (2007) reported 88% residual control of barnyardgrass 56 DAT with glyphosate plus *S*-metolachlor in cotton. Residual control with fomesafen primarily targets broadleaf weeds (Anonymous 2013) and barnyardgrass control 28 DAT with glyphosate plus fomesafen in the current study was comparable to glyphosate alone or mixed with lactofen (Table 2.1). A main effect of foliar fertilizer was detected for barnyardgrass control 28 DAT (Table 2.8). The addition of foliar fertilizer at 0.39 or 0.78 kg ha⁻¹ reduced barnyardgrass control $\geq 4\%$ regardless of herbicide treatment (Table 2.8).

Pooled across foliar fertilizer rates, glyphosate plus lactofen reduced soybean height 14 DAT compared with the no herbicide and glyphosate alone treatments 5 and 4 cm, respectively (Table 2.1). This was similar to results reported by Mangialardi et al. (2016) where lactofen at 0.22 kg ha⁻¹ alone and mixed with COC reduced soybean height 4 and 5 cm, respectively, compared with the control or COC alone. Soybean height at maturity was not affected by foliar fertilizer rate or herbicide treatment. Pooled across foliar fertilizer rates, soybean yields in plots receiving herbicide were similar and greater than yield in the no herbicide treatment (Table 2.1).

Agronomic Study

A main effect of herbicide treatment was detected for soybean injury 3, 7, and 14 DAT, soybean dry weight 14 DAT, and soybean height 14 DAT (Table 2.9). Glyphosate plus lactofen caused the greatest soybean injury followed by glyphosate plus fomesafen. Soybean injury was less with glyphosate plus *S*-metolachlor compared with glyphosate plus fomesafen, but more than glyphosate alone at all evaluations. Treatments containing a PPO inhibitor reduced soybean dry weight $\geq 23 \text{ g m}^{-2}$ and soybean height $\geq 3 \text{ cm}$ 14 DAT compared with the no herbicide treatment. Soybean dry weights and heights 14 DAT with glyphosate plus *S*-metolachlor were comparable to no herbicide and glyphosate alone. Soybean dry weights were comparable with glyphosate plus *S*-metolachlor or fomesafen.

Foliar fertilizer rate did not affect any parameter measured. Some differences in nutrient concentration were detected; however, all values from the analysis were within the nutrient sufficiency range (data not presented; Mills and Jones 1996).

Single-nutrient Foliar Fertilizer Study

At 3, 7, and 14 DAT, a herbicide main effect was detected for soybean injury (Table 2.10). Treatments containing fomesafen injured soybean more than glyphosate alone and the no herbicide treatment at all evaluations (Table 2.10). No single-nutrient foliar fertilizer influenced soybean injury.

An interaction of herbicide treatment and single-nutrient foliar fertilizer was detected for Palmer amaranth control 7 DAT (Table 2.11). Glyphosate plus fomesafen plus all single-nutrient foliar fertilizers controlled more Palmer amaranth 7 DAT than glyphosate plus single-nutrient foliar fertilizers; however, control with glyphosate plus

fomesafen plus Zn and glyphosate plus Mn were comparable (Table 2.11). When comparing single-nutrient foliar fertilizers within each herbicide treatment, Palmer amaranth control 7 DAT was reduced $\geq 15\%$ with glyphosate plus Zn compared with other single-nutrient foliar fertilizer treatments (Table 2.11). Scroggs et al. (2009) documented reductions of 94 and 66% in Palmer amaranth and barnyardgrass control, respectively, when adding 10% zinc sulfate to a labeled glyphosate application in cotton. A herbicide main effect was detected 14, 21, and 28 DAT for Palmer amaranth control (Table 2.10). At each interval, glyphosate plus fomesafen controlled Palmer amaranth better than glyphosate alone (Table 2.10).

Barnyardgrass control 7 DAT was influenced by an interaction of herbicide treatment and single-nutrient foliar fertilizer was (Table 2.11). Glyphosate in combination with Mn reduced barnyardgrass control 11% compared with glyphosate alone (Table 2.11). Bernards et al. (2005) reported similar findings when combinations of glyphosate and Mn were applied to giant foxtail [*Setaria faberi* (Herrm)]. A main effect of single-nutrient foliar fertilizer was detected for barnyardgrass control 14 DAT (Table 2.12). Pooled across herbicide treatments, Mn reduced barnyardgrass control 5% 14 DAT compared with no single-nutrient foliar fertilizer (Table 2.12). Herbicide main effects were detected for barnyardgrass control 14, 21, and 28 (Table 2.13). Pooled across five single-nutrient foliar fertilizers, barnyardgrass control with glyphosate plus fomesafen was comparable with that from glyphosate alone at all evaluation intervals (Table 2.13).

A herbicide main effect was detected for soybean height 14 DAT and at maturity (Table 2.13). Pooled across five single-nutrient foliar fertilizers, treatments containing a

herbicide reduced plant heights 3 cm 14 DAT (Table 2.13); however, at maturity, treatments containing a herbicide were ≥ 3 cm taller (Table 2.13).

An interaction of herbicide treatment and single-nutrient foliar fertilizer was detected for soybean yield (Table 2.14). The addition of B to glyphosate improved soybean yield 870 kg ha^{-1} compared with glyphosate alone (Table 2.14). Sutradhar et al. (2017) reported B, Mn, and Zn fertilization did not improve soybean yield; however, these were not applied in a foliar application. There were no differences between single-nutrient foliar fertilizers within the no herbicide and glyphosate plus fomesafen treatments. When a herbicide was combined with Mn or N, yields were improved compared with the no herbicide plus Mn or N treatments. Soybean yield was inconsistent across single-nutrient foliar fertilizers and herbicide treatments likely due to different weed densities. To test for differences in soybean yield from single-nutrient foliar fertilization, a weed-free trial should be conducted.

The injury caused by POST soybean herbicide treatments evaluated in this research was not affected by the addition of a blended foliar fertilizer; therefore, injury from these POST soybean herbicides cannot be reduced with foliar fertilizers. The blended foliar fertilizer (4-0-0-3-3-3-0.25%; N-P-K-S-Mn-Zn-B) should not be combined with POST soybean herbicides with the intent to reduce injury.

Mixing a blended foliar fertilizer with POST soybean herbicides influenced herbicide efficacy. Palmer amaranth and barnyardgrass control was antagonized 7, 14, and 21 DAT by one or more of the herbicide treatments and blended foliar fertilizer rate combinations. Across species and evaluation intervals, 14 total antagonistic effects were detected. Antagonism of glyphosate from foliar fertilizer at 0.78 kg ha^{-1} was the most

common antagonistic effect regardless of weed species or evaluation interval with four detected effects. Antagonism of glyphosate plus *S*-metolachlor from foliar fertilizer at 0.78 kg ha⁻¹ was detected three times. Of the 14 total effects, nine were detected with foliar fertilizer at the higher rate of 0.78 kg ha⁻¹. Between the two weed species, seven antagonistic effects were detected for each regardless of herbicide treatment or foliar fertilizer rate. The only treatment combination in which an antagonistic effect was not detected was the glyphosate plus lactofen plus foliar fertilizer at 0.78 kg ha⁻¹. A grower with the intention of applying the blended foliar fertilizer (4-0-0-3-3-3-0.25%; N-P-K-S-Mn-Zn-B) at 0.78 kg ai ha⁻¹ with a POST soybean herbicide should expect antagonism, especially if glyphosate is included in the application.

In order to identify the cause of antagonism, the cations represented in the blended foliar fertilizer were applied as single-nutrient foliar fertilizers with glyphosate alone and in combination with fomesafen. Previous research documented cations interfering with herbicide efficacy (Bernards et al. 2005; Scroggs et al. 2009; Roskamp et al. 2013). Roskamp et al. (2013) reported reductions in horseweed [*Conyza canadensis* (L.) Cronq.] control 3 wk after treatment (WAT) when 2, 4-D at 0.266 kg ae ha⁻¹ was applied with deionized water plus Mn at 4.97 L ha⁻¹. Common lambsquarter [*Chenopodium album* (L.)] control 3 WAT was also reduced when 2, 4-D was applied with deionized water plus Ca at 590 mg L⁻¹ or Mn fertilizer at 4.97 L ha⁻¹ (Roskamp et al. 2013). Although antagonistic effects were not detected for weed control in the current research, interactions between the herbicide treatments and single-nutrient foliar fertilizers indicated reductions in control when adding some of the cations to the herbicide treatments. Palmer amaranth control was reduced when Zn was added to

glyphosate, and barnyardgrass control was reduced when Mn was added to glyphosate. Although antagonistic effects were not detected, the Single-nutrient Foliar Fertilizer Study should be repeated to improve the variability of the data.

Soybean agronomic performance was not improved by adding a blended foliar fertilizer to POST herbicide treatments. Since the blended foliar fertilizer (4-0-0-3-3-3-0.25%; N-P-K-S-Mn-Zn-B) did not affect soybean injury, height, dry weight, nutrient concentration, or yield, the addition of this blended foliar fertilizer would not be beneficial to soybean and would represent an added expense to the grower. Even when the blended foliar fertilizer was applied with no herbicide, the agronomic performance of soybean was not improved.

Foliar fertilizer in combination with POST soybean herbicides did not reduce soybean injury and produced inconsistent effects on herbicide efficacy across herbicide treatments and between weed species. Foliar fertilizers also did not improve agronomic performance of soybean. Zinc and Mn reduced Palmer amaranth and barnyardgrass control, respectively, when applied with glyphosate alone. Since this research evaluated only one blended foliar fertilizer and several individual nutrients represented in that foliar fertilizer, growers should be cautious of other foliar fertilizers applied with POST herbicides in soybean. If a soybean herbicide treatment includes glyphosate, no foliar fertilizer should be added, especially those containing Mn or Zn.

Table 2.1 Soybean injury 3, 7, and 14 d after treatment (DAT), Palmer amaranth and barnyardgrass control 28 DAT, height 14 DAT, and soybean yield following application of mixtures of POST soybean herbicides and a blended foliar fertilizer applied at the V3 growth stage in the Weed Control Study at Stoneville, MS, in 2015 and 2016a.

Herbicide treatment	Rate	Soybean injury			Palmer amaranth control	Barnyardgrass control	Height		Yield
		3 DAT	7 DAT	14 DAT	28 DAT	28 DAT	14 DAT	28 DAT	
		%			cm		kg ha ⁻¹		
No herbicide	-	0 d	0 d	0 d	0 d	0 d	38 b	2,659 c	
Glyphosate	1.37	1 d	1 d	0 d	56 c	77 bc	37 b	3,421 a	
Glyphosate plus fomesafen	1.37 + 0.395	12 b	10 b	4 b	78 a	74 c	36 ab	3,560 a	
Glyphosate plus lactofen	1.37 + 0.218	22 a	18 a	7 a	71 b	77 bc	33 a	3,377 ab	
Glyphosate plus S-metolachlor	1.37 + 1.42	6 c	5 c	2 c	67 b	82 a	36 ab	3,333 ab	

^a Data are pooled over four siteyears and three foliar fertilizer rates. Means followed by the same letter for each parameter and/or evaluation interval are not different at $p \leq 0.05$.

Table 2.2 Antagonistic effects for Palmer amaranth control 7 d after treatment (DAT) with mixtures of POST soybean herbicides and a blended foliar fertilizer applied at the V3 growth stage in the Weed Control Study at Stoneville, MS, in 2015 and 2016.

Herbicide treatment	Rate	Foliar fertilizer rate (kg ai ha ⁻¹)				p-value
		Expected ^a	Observed ^b	Expected	Observed	
	kg ae or ai ha ⁻¹	———— % ————	———— % ————	———— % ————	———— % ————	
		0.39			0.78	
Glyphosate	1.37	66	59*	66	61	0.1185
Glyphosate plus fomesafen	1.37 + 0.395	83	79	83	77	0.1025
Glyphosate plus lactofen	1.37 + 0.218	82	80	82	77	0.1831
Glyphosate plus S-metolachlor	1.37 + 1.42	73	62*	73	62*	0.0088

^a Expected values for each rate of foliar fertilizer are the same due to a lack of herbicidal activity from the foliar fertilizer; therefore, values are visual estimates of weed control for each herbicide treatment when foliar fertilizer rate was 0 kg ha⁻¹.

^b Asterisks denote antagonistic effects between herbicide treatment and foliar fertilizer rate when $p \leq 0.05$.

^c The p-value nested within each foliar fertilizer rate denotes significant differences between observed and expected values within the corresponding rate of foliar fertilizer.

Table 2.3 Antagonistic effects for Palmer amaranth control 14 d after treatment (DAT) with mixtures of POST soybean herbicides and a blended foliar fertilizer applied at the V3 growth stage in the Weed Control Study at Stoneville, MS, in 2015 and 2016.

Herbicide treatment	Rate	Foliar fertilizer rate (kg ai ha ⁻¹)				p-value
		Expected ^a	Observed ^b	Expected	Observed	
	kg ae or ai ha ⁻¹	———— % ————	———— % ————	———— % ————	———— % ————	
		0.39			0.78	
Glyphosate	1.37	66	57	66	55*	0.0294
Glyphosate plus fomesafen	1.37 + 0.395	81	77	81	74	0.1200
Glyphosate plus lactofen	1.37 + 0.218	83	80	83	78	0.2437
Glyphosate plus S-metolachlor	1.37 + 1.42	74	60*	74	60*	0.0200

^a Expected values for each rate of foliar fertilizer are the same due to a lack of herbicidal activity from the foliar fertilizer; therefore, values are visual estimates of weed control for each herbicide treatment when foliar fertilizer rate was 0 kg ha⁻¹.
^b Asterisks denote antagonistic effects between herbicide treatment and foliar fertilizer rate when $p \leq 0.05$.
^c The p-value nested within each foliar fertilizer rate denotes significant differences between observed and expected values within the corresponding rate of foliar fertilizer.

Table 2.4 Antagonistic effects for Palmer amaranth control 21 d after treatment (DAT) with mixtures of POST soybean herbicides and a blended foliar fertilizer applied at the V3 growth stage in the Weed Control Study at Stoneville, MS, in 2015 and 2016.

Herbicide treatment	Rate	Foliar fertilizer rate (kg ai ha ⁻¹)				p-value	p-value
		Expected ^a	Observed ^b	Expected	Observed		
	kg ae or ai ha ⁻¹	———— % ————	———— % ————	———— % ————	———— % ————		
		0.39				0.78	
Glyphosate	1.37	66	58	66	53*	0.0819	0.0099
Glyphosate plus fomesafen	1.37 + 0.395	79	78	79	72	0.6772	0.0886
Glyphosate plus lactofen	1.37 + 0.218	76	74	76	71	0.7159	0.2770
Glyphosate plus S-metolachlor	1.37 + 1.42	69	63	69	60	0.1451	0.0502

^a Expected values for each rate of foliar fertilizer are the same due to a lack of herbicidal activity from the foliar fertilizer; therefore, values are visual estimates of weed control for each herbicide treatment when foliar fertilizer rate was 0 kg ha⁻¹.

^b Asterisks denote antagonistic effects between herbicide treatment and foliar fertilizer rate when $p \leq 0.05$.

^c The p-value nested within each foliar fertilizer rate denotes significant differences between observed and expected values within the corresponding rate of foliar fertilizer.

Table 2.5 Antagonistic effects for barnyardgrass control 7 d after treatment (DAT) with mixtures of POST soybean herbicides and a blended foliar fertilizer applied at the V3 growth stage in the Weed Control Study at Stoneville, MS, in 2015 and 2016.

Herbicide treatment	Rate	Foliar fertilizer rate (kg ai ha ⁻¹)				p-value
		Expected ^a	Observed ^b	Expected	Observed	
	kg ae or ai ha ⁻¹	—— % ——	—— % ——	—— % ——	—— % ——	
		0.39			0.78	
Glyphosate	1.37	88	84	88	81*	0.0274
Glyphosate plus fomesafen	1.37 + 0.395	83	79	83	78	0.1082
Glyphosate plus lactofen	1.37 + 0.218	85	80	85	82	0.2646
Glyphosate plus S-metolachlor	1.37 + 1.42	82	82	82	80	0.6181

^a Expected values for each rate of foliar fertilizer are the same due to a lack of herbicidal activity from the foliar fertilizer; therefore, values are visual estimates of weed control for each herbicide treatment when foliar fertilizer rate was 0 kg ha⁻¹.

^b Asterisks denote antagonistic effects between herbicide treatment and foliar fertilizer rate when $p \leq 0.05$.

^c The p-value nested within each foliar fertilizer rate denotes significant differences between observed and expected values within the corresponding rate of foliar fertilizer.

Table 2.6 Antagonistic effects for barnyardgrass control 14 d after treatment (DAT) with mixtures of POST soybean herbicides and a blended foliar fertilizer applied at the V3 growth stage in the Weed Control Study at Stoneville, MS, in 2015 and 2016.

Herbicide treatment	Rate	Foliar fertilizer rate (kg ai ha ⁻¹)				p-value
		Expected ^a	Observed ^b	Expected	Observed	
	kg ae or ai ha ⁻¹	%		%		
		0.39			0.78	
Glyphosate	1.37	87	88	87	82	0.0818
Glyphosate plus fomesafen	1.37 + 0.395	84	75*	83	74*	0.0087
Glyphosate plus lactofen	1.37 + 0.218	85	81	85	81	0.2942
Glyphosate plus S-metolachlor	1.37 + 1.42	86	83	86	79*	0.0437

^a Expected values for each rate of foliar fertilizer are the same due to a lack of herbicidal activity from the foliar fertilizer; therefore, values are visual estimates of weed control for each herbicide treatment when foliar fertilizer rate was 0 kg ha⁻¹.

^b Asterisks denote antagonistic effects between herbicide treatment and foliar fertilizer rate when $p \leq 0.05$.

^c The p-value nested within each foliar fertilizer rate denotes significant differences between observed and expected values within the corresponding rate of foliar fertilizer.

Table 2.7 Antagonistic effects for barnyardgrass control 21 d after treatment (DAT) with mixtures of POST soybean herbicides and a blended foliar fertilizer applied at the V3 growth stage in the Weed Control Study at Stoneville, MS, in 2015 and 2016.

Herbicide treatment	Rate	Foliar fertilizer rate (kg ai ha ⁻¹)				p-value	p-value
		Expected ^a	Observed ^b	Expected	Observed		
	kg ae or ai ha ⁻¹	—— % ——	—— % ——	—— % ——	—— % ——		
		0.39				0.78	
Glyphosate	1.37	85	80	85	79*	0.0697	0.0397
Glyphosate plus fomesafen	1.37 + 0.395	82	79	82	72*	0.3155	0.0046
Glyphosate plus lactofen	1.37 + 0.218	83	77*	83	79	0.0465	0.1877
Glyphosate plus S-metolachlor	1.37 + 1.42	86	82	86	82	0.1971	0.1166

^a Expected values for each rate of foliar fertilizer are the same due to a lack of herbicidal activity from the foliar fertilizer; therefore, values are visual estimates of weed control for each herbicide treatment when foliar fertilizer rate was 0 kg ha⁻¹.

^b Asterisks denote antagonistic effects between herbicide treatment and foliar fertilizer rate when $p \leq 0.05$.

^c The p-value nested within each foliar fertilizer rate denotes significant differences between observed and expected values within the corresponding rate of foliar fertilizer.

Table 2.8 Barnyardgrass control 28 d after treatment (DAT) with mixtures of POST soybean herbicides and a blended foliar fertilizer applied at the V3 growth stage in the Weed Control Study at Stoneville, MS, in 2015 and 2016a.

Foliar fertilizer rate	Control
kg ai ha ⁻¹	%
0	65 a
0.39	61 b
0.78	60 b

^a Data are pooled over four siteyears and five herbicide treatments. Means followed by the same letter are not different at $p \leq 0.05$.

Table 2.9 Soybean injury 3, 7, and 14 d after treatment (DAT), soybean dry weight 14 DAT, and soybean height 14 DAT following application of mixtures of POST soybean herbicides and a blended foliar fertilizer applied at the V3 growth stage in the Agronomic Study at Stoneville, MS, in 2015 and 2016a.

Herbicide treatment	Rate kg ae or ai ha ⁻¹	Injury %			Dry weight g m ⁻²	Height cm
		3 DAT	7 DAT	14 DAT		
No herbicide	-	0 d	0 d	0 d	253 a	40 a
Glyphosate	1.37	1 d	1 d	0 d	254 a	39 a
Glyphosate plus fomesafen	1.37 + 0.395	17 b	12 b	4 b	230 b	37 b
Glyphosate plus lactofen	1.37 + 0.218	29 a	22 a	9 a	201 c	35 c
Glyphosate plus S-metolachlor	1.37 + 1.42	6 c	4 c	2 c	245 ab	39 a

^a Data are pooled over four siteyears and three foliar fertilizer rates. Means followed by the same letter for each parameter and/or evaluation interval are not different at $p \leq 0.05$.

Table 2.10 Soybean injury 3, 7, and 14 d after treatment (DAT) and Palmer amaranth control 14, 21, and 28 DAT with mixtures of POST soybean herbicides and single-nutrient foliar fertilizers applied at the V3 growth stage in the Single-nutrient Foliar Fertilizer Study at Stoneville, MS, in 2015 and 2016a,b.

Herbicide treatment	Rate	Injury			Palmer amaranth control		
		3 DAT	7 DAT	14 DAT	14 DAT	21 DAT	28 DAT
		kg ae or ai ha ⁻¹ _____ %					
No herbicide	-	0 b	0 b	0 b	0 c	0 c	0 c
Glyphosate	1.37	1 b	0 b	1 b	66 b	64 b	61 b
Glyphosate plus fomesafen	1.37 + 0.395	23 a	15 a	4 a	84 a	82 a	80 a

^a Data are pooled over two siteyears and five single-nutrient foliar fertilizers. Means followed by the same letter for each parameter and/or evaluation interval are not different at $p \leq 0.05$.

Table 2.11 Palmer amaranth and barnyardgrass control 7 d after treatment (DAT) with mixtures of POST soybean herbicides and single-nutrient foliar fertilizers applied at the V3 growth stage in the Single-Nutrient Foliar Fertilizer Study at Stoneville, MS, in 2015 and 2016a,b.

Single-nutrient foliar fertilizer	Rate kg ai ha ⁻¹	Palmer amaranth		Barnyardgrass		
		No herbicide	Glyphosate plus fomesafen	No herbicide	Glyphosate plus fomesafen	
%						
No single-nutrient foliar fertilizer	-	0 e	69 c	0 c	87 a	85 ab
Boron	0.015	0 e	69 c	0 c	84 ab	87 a
Manganese	0.175	0 e	74 bc	0 c	76 b	85 ab
Nitrogen	0.235	0 e	70 c	0 c	85 ab	88 a
Zinc	0.175	0 e	54 d	0 c	79 ab	85 ab

^a Data pooled over two siteyears. Means followed by the same letter for each weed species are not different at $p \leq 0.05$.

^b Herbicide treatments were no herbicide, glyphosate at 1.37 kg ae ha⁻¹, and glyphosate plus fomesafen at 1.37 kg ha⁻¹ plus 0.395 kg ai ha⁻¹.

Table 2.12 Barnyardgrass control 14 d after treatment (DAT) with mixtures of POST soybean herbicides and single-nutrient foliar fertilizers applied at the V3 growth stage in the Single-nutrient Foliar Fertilizer Study at Stoneville, MS, in 2015 and 2016a.

Single-nutrient foliar fertilizer	Rate	Control
	kg ai ha ⁻¹	%
No single-nutrient foliar fertilizer	-	57 a
Boron	1.37	54 ab
Manganese	0.395	52 b
Nitrogen	0.235	55 ab
Zinc	0.175	53 ab

^a Data are pooled over two siteyears and three herbicide treatments. Means followed by the same letter are not different at $p \leq 0.05$.

Table 2.13 Barnyardgrass control 14, 21, and 28 d after treatment (DAT), soybean height 14 DAT, and mature soybean height following application of mixtures of soybean herbicides and single-nutrient foliar fertilizers applied at the V3 growth stage in the Single-nutrient Foliar Fertilizer Study at Stoneville, MS, in 2015 and 2016a.

Herbicide treatment	Rate kg ae or ai ha ⁻¹	Barnyardgrass control			Height		
		14 DAT	21 DAT	28 DAT	14 DAT	14 DAT	Maturity
No herbicide	-	0 b	0 b	0 b	0	32 a	97 b
Glyphosate	1.37	82 a	75 a	68 a	29 b	29 b	101 a
Glyphosate plus fomesafen	1.37 + 0.395	81 a	73 a	70 a	29 b	29 b	100 a

^a Data are pooled over two siteyears and five single-nutrient foliar fertilizers. Means followed by the same letter for each parameter and/or evaluation interval are not different at $p \leq 0.05$.

Table 2.14 Soybean yield following mixtures of POST soybean herbicides and single-nutrient foliar fertilizers applied at the V3 growth stage in the Single-nutrient Foliar Fertilizer Study at Stoneville, MS, in 2015 and 2016a,b.

Single-nutrient foliar fertilizer	Rate kg ai ha ⁻¹	No herbicide	Glyphosate kg ha ⁻¹	Glyphosate plus fomesafen
No single nutrient foliar fertilizer	-	2,402 bc	2,356 bc	2,890 ab
Boron	0.015	2,317 bc	3,226 a	2,934 ab
Manganese	0.175	2,039 c	2,740 ab	2,947 ab
Nitrogen	0.235	2,015 c	2,777 ab	2,766 ab
Zinc	0.175	2,445 bc	2,859 ab	2,853 ab

^a Means followed by the same letter are not different at $p \leq 0.05$.

^b Herbicide treatments were no herbicide, glyphosate at 1.37 kg ae ha⁻¹, and glyphosate plus fomesafen at 1.37 kg ha⁻¹ plus 0.395 kg ai ha⁻¹

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CHAPTER III
AN EVALUATION ON THE INFLUENCE OF CYTOKININ MIXTURES IN
COMBINATION WITH COMMON POSTEMERGENCE
SOYBEAN HERBICIDES

Abstract

A field study was conducted in 2015 and 2016 in Stoneville, MS, to evaluate the influence of cytokinin mixtures on soybean injury and weed control when combined with common POST soybean herbicides. Cytokinin treatments included no cytokinin mixture and two formulated cytokinin mixtures (kinetin-1 and kinetin-2) applied at 0.000227 kg ai ha⁻¹. Herbicide treatments were no herbicide, glyphosate at 1.37 kg ae ha⁻¹ alone and in combination with *S*-metolachlor at 1.42 kg ai ha⁻¹ or fomesafen 0.395 kg ai ha⁻¹. Cytokinin mixtures had no impact on soybean injury, height, or yield. Glyphosate plus fomesafen provided the greatest level of Palmer amaranth control. Barnyardgrass control with glyphosate plus fomesafen was antagonized by one of two cytokinin mixtures. To prevent possible reductions in herbicide efficacy, cytokinin mixtures should not be applied to soybean in POST herbicide applications.

Nomenclature: Glyphosate; fomesafen; *S*-metolachlor; Palmer amaranth *Amaranthus palmeri* S. Watts AMAPA; Barnyardgrass *Echinochloa crus-galli* L. Beauv. ECHCG; soybean, *Glycine max* L. Merr.

Keywords: Cytokinins, kinetin mixtures, plant health management

Introduction

Palmer amaranth has been ranked as one of the most troublesome weeds in soybean in the southern U.S. since the 1970's (Anonymous 1972; Buchanan 1973, 1974). By 2013, Palmer amaranth was ranked as the most troublesome weed of soybean in three southern U.S. states (Webster 2013). Palmer amaranth has increased in severity, in part because of herbicide resistance. In 2004, Georgia reported the first glyphosate-resistant (GR) Palmer amaranth (Culpepper et al. 2006), followed by Arkansas in 2005 (Norsworthy et al. 2008). In Mississippi, GR Palmer amaranth was documented in 2008 (Heap 2017).

Various herbicides can be used for GR Palmer amaranth control in soybean (Whitaker et al. 2010). Protoporphyrinogen oxidase (PPO) inhibitors such as fomesafen are used for the PRE and POST control of Palmer amaranth (MSU-ES 2017). Stephenson et al. (2004) documented common cocklebur (*Xanthium strumarium* L.), prickly sida (*Sida spinosa* L.), and Palmer amaranth control with fomesafen. Bond et al. (2006) and Norsworthy et al. (2008) reported 96 and 100% GR Palmer amaranth control, respectively, with fomesafen at 0.420 kg ai ha⁻¹.

Barnyardgrass is also a problematic weed of U.S. soybean (Holm et al. 1977; Bagavathiannan et al. 2011). Tennessee was the first state to confirm GR barnyardgrass in the U.S. (Steckel et al 2017). Barnyardgrass in Mississippi has a history of resistance to multiple herbicide MOA (Heap 2017; Wright et al. 2016). With the state's close proximity to Tennessee, researchers in Mississippi have collected and tested barnyardgrass samples for possible glyphosate resistance (Bond 2017). In a model based on Arkansas' cotton [*Gossypium hirsutum* (L.)]-growing region, Bagavathiannan et al.

(2013) predicted GR barnyardgrass will develop by 2022 following five annual glyphosate applications in continuous GR cotton. By rotating to GR corn [*Zea mays* (L.)] or glufosinate-resistant cotton, resistance could be delayed 6 yr (Bagavathiannan et al. 2013).

Glyphosate-resistant barnyardgrass would be a major problem for growers because glyphosate is one of the principal herbicides used for barnyardgrass control (Krausz et al. 2001; Sikkema et al. 2005; Riar et al. 2013). Glyphosate at 0.450 kg ha⁻¹ reduced barnyardgrass density, dry weight, and seed production 84 d after application 82, 97, and 98%, respectively (Sikkema et al. 2005). Scroggs et al. (2007) reported 92% barnyardgrass control after three applications of glyphosate (1.06 kg ha⁻¹) at the three-, seven-, and 14-leaf growth stages.

Plant health management focuses on overcoming factors that limit plants from achieving their full genetic potential (Cook 2000). The full genetic potential is the capacity at which a plant can grow, develop, and reproduce without limiting factors (Cook 2000). Many techniques, such as breeding, have increased the genetic potential of plants; however, plant health management focuses on improving on the preexisting potential (Cook 2000). The use of high-quality seed, optimum fertility, irrigation, and integrated pest management strategies all influence plant health management (Cook 2000).

Plant growth regulators (PGRs) are used as plant health management tools in several crops, especially cotton (Ren et al. 2013). Mepiquat chloride is commonly applied to cotton to control vegetative growth and prevent shading which causes fruit abscission and reduced yield (Guinn 1974). Some herbicides are also used as PGRs.

Glyphosate applied at low rates has been used to suppress flowering and stimulate sucrose accumulation in sugarcane [*Saccharum spontaneum* (L.)] (Bennet and Montes 2003; Velini et al. 2010).

Cytokinins occur naturally in plants and are responsible for cell division and enlargement as well as the formation of flowers and fruits (Skoog and Armstrong 1970). Cytokinins have been reported to increase soybean cell proliferation in tissue culture (Fosket and Short 1973). Kinetin, a specific cytokinin, has been reported to reverse the effect of NaCl on tobacco [*Nicotiana tabacum* (L.)] leaves when applied in solution to a tissue culture (Katz et al. 1978). Cytokinin mixtures are available as PGRs for use in several crops, and labeling for formulated cytokinin mixtures claim these products improve vigor, promote root and shoot growth, reduce stress, and slow leaf aging (Anonymous 2017a, 2017b). Data supporting the supposed benefits of applying cytokinin mixtures is limited. Most research detailing the effects of kinetin and other cytokinin mixtures has been conducted with tissue culture.

Tank mixtures with multiple herbicide MOA offers the potential to increase weed control and reduce application costs (Hydrick and Shaw 1994). These combinations can produce synergistic, antagonistic, or additive effects (Nash 1981). Synergism occurs when the total response of the components is greater than the sum of the individuals (Nash 1981). Antagonism occurs when the sum is less than the response of the individual components (Nash 1981). The components could be herbicides, foliar fertilizers, water, or any other components (Devkota and Johnson 2016; Mahoney et al. 2014; Scroggs et al. 2009; Starke and Oliver 1998; Roskamp et al 2013; Vidrine et al. 1995).

Reports of herbicide-by-herbicide or -water interactions are abundant in the literature. Minton et al. (1989) reported barnyardgrass control was antagonized when quizalofop or sethoxydim were combined with lactofen. Starke and Oliver (1998) documented antagonism on entireleaf morningglory [*Ipomoea hederacea* var. *integriuscula* (Gray)] but not on pitted morningglory [*Ipomoea lacunose* (L.)] control when fomesafen and glyphosate were combined. Water may antagonize herbicides because of the cations present in hard water. Stable complexes are formed when glyphosate bonds with di- and trivalent cations, leading to glyphosate antagonism (Glass 1984; Lundager Madsen et al. 1978; Thelen et al 1995).

Various statistical techniques to test herbicide interactions in mixtures with other components have been outlined in the literature. Colby's method (Colby 1967) was utilized by Eshel et al. (1976) to report synergistic effects on wild oat [*Avena fatua* (L.)] and wild mustard [*Sinapis arvensis* (L.)] control. Colby's method has been one of the more popular tests for years and was used more recently to detail antagonism of volunteer GR corn control in dicamba-resistant soybean (Underwood et al. 2016). Blouin et al. (2004) developed the nonlinear model to test for interactions used by Webster et al. (2006) in evaluating a safening interaction on rice [*Oryza sativa* (L.)] treated with clomazone plus bensulfuron or halosulfuron. After expanding on the nonlinear model, Blouin et al. (2010) created the augmented mixed-model methodology utilized by Fish et al. (2016) to determine synergism and antagonism between propanil and imazamox on red rice [*Oryza sativa* (L.)] and barnyardgrass control.

Research detailing interactions between herbicides and cytokinin mixtures is limited. Also, labeling of formulated cytokinin mixtures does not mention mixtures with

other products beyond outlining the use of surfactants (Anonymous 2017a, 2017b). It has been hypothesized that cytokinins could reduce injury from flooding in corn (Rao et al. 2002). A patent also exists for a 1:1 mixture of glyphosate and kinetin to reduce glyphosate phytotoxicity (Ng and Wang 2012). In order to lower application costs by reducing the number of trips through the field, growers may combine POST herbicides and cytokinin mixtures. A field study was conducted to evaluate the influence on crop response and weed control of adding foliar cytokinin mixtures to POST soybean herbicide applications.

Materials and Methods

A field study was conducted at the Mississippi State University Delta Research and Extension Center in Stoneville, MS, in 2015 and 2016 to evaluate combinations of cytokinin mixtures and POST herbicides in soybean. The study was performed at two sites in 2015 (33°26'29.18"N 90°54'41.92"W and 33°24'21.94"N 90°55'31.27"W) and 2016 (33°26'28.33"N 90°54'23.67"W and 33°24'21.94"N 90°55'31.27"W). In 2015, soil at one site was a Dundee very fine sandy loam (Fine-silty, mixed, active, thermic Typic Endoaqualfs) with a pH of 6.1 and 1.2% organic matter, and soil at the other was a Newellton silty clay (Clayey over loamy, smectitic over mixed, superactive, nonacid, thermic Fluvaquentic Epiaquepts) with a pH of 6.9 and 1.6% organic matter. In 2016, soil at one site was a Commerce sandy clay loam (Fine-silty, mixed, superactive, nonacid, thermic Fluvaquentic Endoaquepts) with a pH of 6.8 and 1.6 % organic matter along with the Newellton silty clay loam utilized in 2015. The experimental sites were known to be heavily infested with barnyardgrass and Palmer amaranth. Each site was conventionally tilled prior to planting to stimulate weed germination and ensure uniform

emergence. ‘Asgrow 4632’ (Monsanto Company 800 N. Lindbergh Blvd. St. Louis, MO 63167) mid maturity group IV soybean were used in all siteyears and sowed with a John Deere small-plot air planter (John Deere 1730, Deere and Company, One John Deere Place Moline, IL, 61265-8098).

The study was designed as a two-factor factorial within a randomized complete block with four replications. Factor A was herbicide treatment and consisted of no herbicide, glyphosate (Monsanto Company, 800 N. Lindbergh Blvd., St. Louis, MO 63167) at 1.36 kg ha⁻¹ alone and in combination with *S*-metolachlor (Syngenta Crop Protection, LLC, P. O. Box 18300, Greensboro, NC 27419) at 1.42 kg ha⁻¹, and fomesafen (Syngenta Crop Protection, LLC, P. O. Box 18300, Greensboro, NC 27419) at 0.375 kg ha⁻¹. Factor B was cytokinin mixture and consisted of no cytokinin mixture, kinetin-1 (WinField Solutions, LLC, P. O. Box 64589, St. Paul, MN 55164) at 0.000227 kg ha⁻¹, and kinetin-2 (Loveland Products, Inc., P. O. Box 1286, Greeley, CO 80632) at 0.000227 kg ha⁻¹. Treatments were applied with a tractor-mounted sprayer calibrated to deliver 140 L ha⁻¹ at 248 kPa fitted with extended range flat-fan (XR10002 TeeJet® P.O. Box 7900 Wheaton, IL 60187) nozzles at the V3 soybean growth stage.

Visual estimates of soybean injury and weed control were recorded on a scale from 0 to 100% with 0 representing no injury or control and 100 representing soybean death or complete weed control. Soybean injury was evaluated 3, 7, 14, 21, and 28 d after treatment (DAT) and control of GR Palmer amaranth and barnyardgrass was evaluated 7, 14, 21, and 28 DAT. Heights of five soybean plants in each plot were measured from the ground to the uppermost node 14 DAT and at maturity. Soybean were harvested using a small-plot combine (Kincaid Equipment, 210 West First St., P.O. Box

400; Haven, KS) on September 25 and October 5 in 2015, and September 16 and October 12 in 2016. Yield were adjusted to 13% moisture content.

Square roots of visual injury and control estimates were arcsine transformed. The transformation did not improve the homogeneity of the variance based on visual inspection of the plotted residuals; therefore, nontransformed data were used in all analyses. Soybean injury and weed control data were analyzed utilizing the augmented mixed-model methodology detailed by Blouin et al. (2010). Data for soybean height and yield were subjected to ANOVA using the PROC MIXED procedure in SAS 9.4 (SAS Institute Inc. 100 SAS Campus Drive Cary, NC 27513-2414) with siteyear, replication (nested within siteyear), and treatment-by-rep interactions listed as the random effect parameters (Blouin et al. 2011). Type III Statistics were used to test the fixed effects of herbicide treatment and cytokinin mixture. Least square means were calculated and mean separation ($p \leq 0.05$) was produced using PDMIX800 in SAS, which is a macro for converting mean separation output to letter groupings (Saxton 1998). When injury and weed control data did not return a significant synergistic or antagonistic effect (Blouin et al. 2010), the data were analyzed as previously described for soybean height and yield.

Results and Discussion

No synergistic or antagonistic effects were detected for soybean injury at any evaluation interval. The main effect of cytokinin mixture did not influence soybean injury at any evaluation; however, a main effect of herbicide treatment was detected 3, 7, and 14 DAT (Table 3.1). Injury was at least 5% greater with glyphosate plus fomesafen compared with other treatments 3, 7 and 14 DAT (Table 3.1). Bronzing and necrosis of plant tissue from POST fomesafen applications has been well-documented (Johnson et al.

2002; Mangialardi et al. 2016). By 21 and 28 DAT, soybean injury was $\leq 1\%$ across all herbicide treatments (data not presented).

Data for Palmer amaranth control indicated no synergistic or antagonistic effects. Additionally, the main effect of cytokinin mixture was not significant for Palmer amaranth control. A main effect of herbicide treatment was detected for Palmer amaranth control at all evaluations (Table 3.1). Glyphosate plus fomesafen provided 84 and 67% control of Palmer amaranth 7 and 28 DAT, respectively (Table 3.1). Glyphosate alone or in combination with *S*-metolachlor did not control Palmer amaranth $> 68\%$ at any evaluation interval (Table 3.1). Across all evaluations, Palmer amaranth control was at least 6% greater with glyphosate plus fomesafen compared with other herbicide treatments (Table 3.1). Everman et al. (2009), Whitaker et al. (2010), Barkley et al. (2016), and Miller and Norsworthy (2016) all observed Palmer amaranth control after PRE or POST applications of fomesafen. Glyphosate alone controlled Palmer amaranth 58 to 65% across all evaluation intervals (Table 3.1), confirming the populations of Palmer amaranth contained GR individuals.

An antagonistic effect was detected on barnyardgrass control 14 DAT when kinetin-1 was combined with glyphosate plus fomesafen (Table 3.2). The addition of kinetin-1 to glyphosate plus fomesafen caused a 9% reduction in barnyardgrass control compared with glyphosate plus fomesafen or with no cytokinin mixture (Table 3.2). Across all other evaluation intervals, a main effect of herbicide treatment was detected for barnyardgrass control (Table 3.3). Glyphosate alone controlled more barnyardgrass than other herbicide treatments 7 DAT (Table 3.3). By 21 and 28 DAT, glyphosate plus *S*-metolachlor controlled barnyardgrass greatest (Table 3.3). Since glyphosate is a POST

herbicide lacking residual control, it should be expected that the residual control from *S*-metolachlor would control barnyardgrass better than glyphosate alone 28 DAT (Anonymous 2015; MSU-ES 2017). Glyphosate plus fomesafen provided 9 and 6% less barnyardgrass control 7 and 21 DAT, respectively, compared with glyphosate alone (Table 3.3). Barnyardgrass control 28 DAT with glyphosate plus fomesafen was comparable with glyphosate alone (Table 3.3).

Herbicide main effects were detected for soybean height 14 DAT, mature soybean height, and soybean yield (Table 3.3). Pooled across cytokinin mixtures, soybean heights 14 DAT and at maturity were greater for the no herbicide treatment compared with treatments that received a herbicide (Table 3.3). Height differences were attributed to a severe infestation of Palmer amaranth and barnyardgrass, increasing competition for sunlight necessary for photosynthesis during vegetative growth (Holt 1995). Pooled across cytokinin mixtures, treatments containing a herbicide produced greater soybean yields than the no herbicide treatment (Table 3.3).

Cytokinin mixtures did not reduce injury, improve soybean height, or increase soybean yield. Barnyardgrass control with glyphosate plus fomesafen was antagonized by the addition of kinetin-1. These cytokinin mixtures did not influence weed control when combined with glyphosate alone or in combination with *S*-metolachlor. Future studies should evaluate the possible agronomic benefit of using cytokinins as plant growth regulators in soybean to justify the application costs. Cytokinins should not be mixed with POST soybean herbicide applications because this research demonstrated cytokinin mixtures did not reduce soybean injury and could negatively influence control of certain weed species with some herbicide mixtures.

Table 3.1 Soybean injury 3, 7, and 14 d after treatment (DAT) and Palmer amaranth control 7, 14, 21, and 28 DAT with mixtures of POST soybean herbicides and cytokinin mixtures applied at the V3 growth stage in Stoneville, MS, in 2015 and 2016a.

Herbicide treatment	Rate	Injury								Palmer amaranth control			
		3 DAT	7 DAT	14 DAT	7 DAT	14 DAT	21 DAT	28 DAT	7 DAT	14 DAT	21 DAT	28 DAT	
None	-	0 c	0 c	0 b	0 c	0 c	0 c	0 c	0 c	0 c	0 c	0 c	
Glyphosate	1.37	1 c	0 c	1 b	65 b	63 b	62 b	58 b					
Glyphosate plus fomesafen	1.37 + 0.395	15 a	12 a	6 a	84 a	82 a	78 a	67 a					
Glyphosate plus S-metolachlor	1.37 + 1.42	6 b	6 b	1 b	64 b	68 b	63 b	61 b					

^a Data are pooled over four siteyears and three cytokinin mixtures. Means followed by the same letter for each parameter and/or evaluation are not different at $p \leq 0.05$.

Table 3.2 Antagonistic responses for barnyardgrass control 14 d after treatment (DAT) with mixtures of POST soybean herbicides and cytokinin mixtures applied at the V3 growth stage in Stoneville, MS, in 2015 and 2016a,b.

Herbicide	Rate kg ae or ai ha ⁻¹	Cytokinin mixture ^c					
		Kinetin-1			Kinetin-2		
		Expected	Observed	p-value	Expected	Observed	p-value
Glyphosate	1.37	89	87	0.5290	89	88	0.7514
Glyphosate plus fomesafen	1.37 + 0.395	82	73*	0.0047	82	81	0.8016
Glyphosate plus S-metolachlor	1.37 + 1.42	91	87	0.1781	91	91	0.9686

^a Expected values for each cytokinin mixture are the same due to a lack of herbicidal activity from the cytokinin mixtures; therefore, values are the percent weed control without a cytokinin mixture.

^b Asterisks denote antagonistic responses between herbicide treatment and cytokinin mixtures when $p \leq 0.05$.

^c Applications were made with kinetin-1 and 2 at 0.000227 kg ai ha⁻¹. The p-value nested within each cytokinin mixture denotes significant differences between observed and expected values within the corresponding cytokinin mixture.

Table 3.3 Barnyardgrass control 7, 21 and 28 d after treatment (DAT), height 14 DAT, mature height, and yield with mixtures of POST soybean herbicides and cytokinin mixtures applied at the V3 growth stage in Stoneville, MS, in 2015 and 2016a.

Herbicide treatment	Rate kg ae or ai ha ⁻¹	Barnyardgrass control			Height			Yield kg ha ⁻¹
		7 DAT	21 DAT	28 DAT	14 DAT	Maturity	Yield	
None	-	0 d	0 d	0 c	40 a	100 a	2,674 b	
Glyphosate	1.37	91 a	86 b	83 b	37 b	96 b	3,499 a	
Glyphosate plus fomesafen	1.37 + 0.395	82 c	80 c	79 b	36 b	97 b	3,640 a	
Glyphosate plus S-metolachlor	1.37 + 1.42	86 b	92 a	89 a	36 b	97 b	3,525 a	

^a Data are pooled over four siteyears and three cytokinin mixtures. Means followed by the same letter for each parameter and/or evaluation are not different at $p \leq 0.05$

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