Evaluation of kudzu bug as a pest in Mississippi soybean production systems

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The kudzu bug is an invasive species to the United States, and it has recently become a problem in the southern U.S. Experiments were conducted to examine the potential damage to vegetative stage soybean. Foliar insecticides and neonicotinoid seed treatments were studied to determine the efficacy of those treatments against kudzu bugs. Kudzu bug population densities of adults and nymphs along with egg masses were observed to determine peak activity in kudzu and soybean. This project was designed to develop a better understanding of the damage potential that kudzu bugs can cause as well as the best control methods available.

DEDICATION

I would like to dedicate this thesis to my wife, Callie. Thank you for supporting me in all that I do and pushing me to be the best man I can be. I would also like to dedicate this research to my parents, Bill and Meredith McRight. You have loved and supported me in everything I have done. Thank you for making me person I am today.

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CHAPTER I

INTRODUCTION

Soybean

Origin

Soybean, *Glycine max* (L.) Merr., is grown in nearly 50 countries worldwide (Wilcox 2004). Cultivation of the crop first began in China, which was the largest producer and exporter of soybean in the world until the middle of the 20^{th} century. The wild soybean was domesticated by ancient people using certain agricultural practices, proven by the number of chromosomes that the cultivated soybean shares with the wild soybean. Cultivation of soybean began in the United States as early as 1765 and was made popular by Benjamin Franklin in 1770 when he transported it from France to Philadelphia while working as an ambassador. Production rapidly increased in the United States by the 1950's, leading to the country becoming the world leader, a status continuing to present (Qiu and Chang 2010).

Soybean is one of the most valuable oilseed crops in the world (Singh and Shivakumar 2010). Although cultivation began for many different purposes, the identity of this crop was forever changed by the two World Wars because of its use for oil as well as the discovery of its potential as a vegetable oil. Production of soybean has remained popular in the United States, with over 33.2 million hectares harvested in 2016 (USDA-NASS 2016).

Cultivation systems vary in different regions of the world. Intercropping systems consist of mixing soybean with other seed at a desired ratio and planting the seed in a row or by broadcast. It has a low risk to the grower because at least one crop in the mix is likely to succeed (Qiu and Chang 2010). In contrast, a sequential cropping system is based on planting a single crop at a time to allow for maximum plant growth and reduced pest pressure leading to maximum yield. Soybean production in the United States today generally uses a sequential cropping system. Within this system, soybean production in the southern United States has undergone noticeable changes in recent years. This is especially true in Mississippi and other states in the Mid-South region of the country. Historically, soybean production used Maturity Group V, VI, and VII soybean varieties planted during May and June (Heatherly 1999). Drought and high temperatures are common in this area from July to September which is when these maturity groups are in their reproductive stages (Heatherly 1999). Planting earlier maturing varieties (Maturity groups IV & V) in April has been shown to be beneficial by allowing the soybean reproductive stages to occur earlier, when soil moisture is generally adequate. Planting early maturing soybean in April has not been shown to present any higher risk than using conventional tactics in the Mid-South region, and generally provides greater potential yield (Heatherly 1999).

Soybean row spacing and plant population varies among farm operations. Soybean is commonly grown on rows ranging from 18 cm in width to 102 cm (Heatherly et al. 1999). Although soybean grown in narrow row spacing has sometimes shown a higher yield than the same varieties planted in wider rows (Heatherly et al. 1999), yields do not vary much, so the most economical decision is planting soybean on a row width that is applicable for other crops grown in the operation. For instance, corn and cotton can be grown on 76 cm row spacing, therefore it would be economically beneficial to plant soybean on this row spacing so the same equipment can be used for all crops. The development of the early soybean planting system led to soybean becoming the most widely planted row crop in Mississippi (Bonnie 2008). In 2016, over 800 thousand hectares of soybean were harvested in Mississippi producing over 2.6 million metric tons (USDA-NASS 2016).

Biology

Soybean belongs to the family Leguminosae and subfamily Papilionoideae (Liu 1997). Depending on the environmental conditions and variety, soybean can reach heights ranging from 0.75m to 1.25m. The root system of soybean is comprised of a taproot and a large number of secondary roots that uphold several groups of smaller roots (Lersten 2004). A mutually beneficial relationship between *Bradyrhizobium japonicum*, a soil bacterium, and the soybean plant results in nodules forming on the root system (Kumudini 2010). This bacterium converts atmospheric nitrogen to ammonia, which is taken up by the plant. In return, the bacterium obtains carbon from the plant for energy.

When environmental conditions are favorable, the radicle will emerge from the seed in 1 or 2 days (Williams 1950). The quick maturation of the radicle results in the emergence of secondary roots in 4 to 5 days (Anderson 1961). The cotyledon will emerge shortly after this. This plant has 4 different types of leaves: cotyledons or seed leaves, unifoliate or primary leaves, trifoliolates and prophylls (Sun 1957). Cotyledons are oppositely arranged, emerging first on the plant (Kumudini 2010). Primary leaves develop at the first node above the cotyledon. The node references the section of the stem where the leaf is attached. Trifoliate, alternately arranged leaves follow on every node after the primary leaves (Lersten 2004). Prophylls appear as small pairs of simple leaves at the base of lateral branches (Hicks 1978). A system was developed by Fehr and Caviness (1977) to describe soybean development. Vegetative emergence (VE) is the earliest stage referring to when the cotyledon emerges above the soil surface. The next stage, vegetative cotyledon (VC), refers to the point when the cotyledons have emerged and are no longer touching (Fehr and Caviness 1977). All other vegetative stages after VC are derived from the number of nodes on the main stem (Fehr and Caviness 1977). Fehr and Caviness (1977) determined the beginning of the reproductive stage to be at the appearance of the first flower on the main stem, referring to this as R1. R1 and R2 refer to flower development on the plant. Development stages R3 and R4 refer the size of the pod, and R5 and R6 distinguish seed development. R7 and R8 are the last two stages, describing plant maturity.

Insect Pests of Soybean in Mississippi

There are numerous insect pests in soybean that can have a negative effect on overall production. Lepidopteran species that feed on the leaf tissue of the plant are common pests of soybean throughout the southern U.S. (Funderburk et al. 1999). Velvetbean caterpillar, *Anticarsia gemmatalis* Hübner, soybean looper, *Chrysodeixis includes* Walker, and green cloverworm, *Hypena scabra* Fabricius, are three species common to soybean fields. In the 2016 growing season, over 670,000 hectares of soybean were planted in Mississippi. Velvetbean caterpillar and green cloverworm each caused an estimated loss of 2,470 metric tons of soybean (Musser et al. 2017) while soybean looper damage resulted in a loss of 7,905 metric tons of soybean. Other lepidopteran caterpillars that can also be encountered include fall armyworm, *Spodoptera frugiperda* J.E. Smith, yellowstriped armyworm, *Spodoptera ornithogalli* Guenée, and beet armyworm, *Spodoptera exigua* Hübner,. These three pests can cause serious defoliation under certain environmental conditions. The corn earworm, *Helicoverpa zea* Boddie, is a lepidopteran species that can be a major pest on the pods and seed of soybean plants. Feeding habits of these pests can be described as green cloverworm feeding throughout the growing season; soybean loopers and corn earworms feeding mid to late season; armyworms feeding mid to late season; and velvetbean caterpillars feeding late season. The three most prevalent species of caterpillars can be distinguished from one another by the number of prolegs. The soybean looper has two pairs of prolegs, green cloverworm has three pairs of prolegs, and the velvetbean caterpillar has four pairs of prolegs.

The bean leaf beetle, *Ceratoma trifurcate* Förster, is another pest that is frequently found in Mississippi soybean fields. This pest is a native species that is commonly found in the eastern portion of the United States (Pedigo 1994). The adult bean leaf beetle is relatively small, only measuring about 5 mm in length. The coloration of the adult is variable, but the most common pattern is light yellow with four black spots and marginal, black lines. Other patterns include light yellow without spots and crimson with or without black spots. All adult bean leaf beetles are marked with a distinctive black triangle on the prothorax. Although both larvae and adult have chewing mouthparts, the larvae feed on the roots and nodules of the plant which is considered less important than the feeding damage caused by the adult which occurs as defoliation (Funderburk et al. 1999). The bean leaf beetle is one of the most damaging pests in soybean causing a loss of 23,716 metric tons of soybean in 2016 (Musser et al. 2017).

The stink bug complex (Hemiptera; Pentatomidae) is considered to be one of the most economically important pests to soybean in the southern United States (Musser et al. 2017). Both the nymph and adult stages of stink bugs can cause injury to soybean (McPherson et al. 1994). This pest uses its piercing and sucking mouthparts to penetrate plant tissue and withdraw fluids. They prefer to feed on developing seeds, but they will also obtain nutrients from the stems, foliage, and blooms of the soybean plant (McPherson et al. 1994). In 2016 the stink bug complex was responsible for the loss of 9,882 metric tons of soybean on 670,000 hectares, and the cost of insecticide treatment averaged $12.65/hectare (Musser et al. 2017). The kudzu bug, *Megacopta cribraria* Fabricius, is another hemipteran insect that has recently become a pest in Mississippi soybean. This pest was first noticed in the United States in Georgia in 2009 and it soon spread across southern parts of the country (Eger et al. 2010). In its native region of China, the kudzu bug is considered a pest of legumes (Zhixing et al. 1996). Although *M. cribraria* is known to feed on soybean, the full effect of its feeding in a Mid-South soybean production system is not fully understood. Contrary to the stink bug, the kudzu bug does not feed directly on the seed and pod, instead it uses piercing and sucking mouthparts to extract vascular fluid from the plant tissue, mainly stems (Ruberson et al. 2013a).

Insect Management, Threshold, and Economic Injury Level

There are several different methods used when it comes to managing insect pests. Biological control, the use of natural enemies to reduce a pest population, is one of the oldest methods of controlling insects (Pedigo and Rice 2009b). An example of this would be allowing a predatory insect to control a pest so that it does not cause economic damage (Pedigo and Rice 2009b). Another management tactic is cultural control, which is comprised of many agronomic practices starting before planting through the end of harvest (Summy and King 1992). Cultural practices such as soil tillage and removal of debris can expose pests to unfavorable weather conditions leading to death (Pedigo and Rice 2009b). Foliar insecticides and insecticidal seed treatments are two common forms of chemical control used in soybean pest management. The idea of using seed treated with chemicals to control pests of crops was first conceived by Junius Columella in A.D. 50 (David and Gardiner 1955).

The neonicotinoid class of insecticides is the most widely used insecticide class in the world (Sparks 2013), frequently being applied as seed treatments (Jeschke et al. 2011). Neonicotinoid seed treatments target many seedling pests of soybean in the Mid-South including bean leaf beetle, grape colaspis, *Colaspis brunnea* Fabricius, threecornered alfalfa hopper, *Spissistilus festinus* Say, and thrips (Thysanoptera) (Davis et al. 2009).

Economic decision levels play a major role in pest management decisions. These economic decision levels are measured as the number of insects per area or plant or animal unit as estimated by a sampling procedure (Pedigo and Rice 2009a). The economic injury level (EIL) concept was formally introduced by V.M. Stern and his colleagues in 1959. Economic damage is the amount of injury that justifies the use of control measures. Economic injury level is defined as the least number of insects that are capable of producing economic damage (Stern et al. 1959). Several formulas have been developed to calculate economic injury, and Pedigo et al. (1986) developed the one most commonly used today. This formula is expressed as C=VIDK where C = the price of management practices per production unit, V = the market value of each production unit, I = injury units per pest, D = amount of damage per injury unit, and K = the proportional reduction in pest pressure. The economic threshold (ET) is a commonly used terms in pest management (Pedigo and Rice 2009a). The ET, also known as action threshold, is the number of insects that indicate a need for management practices to prevent the population from exceeding the EIL. According to Pedigo and Rice (2009a), economic injury level and economic threshold work together so that action taken when a population of insects surpasses the economic threshold reduces the population before it causes economic loss by exceeding the economic injury level.

Kudzu Bug

Biology

Megacopta cribraria (Fabricius) (Hemiptera: Plataspidae) is a native insect to Asia and the Indian subcontinent (Srinivasaperumal et al. 1992). *Megacopta cribraria* is generally called the *M. cribraria* in the United States, but other names for this insect include the lablab bug, bean plataspid, and globular stink bug (Eger et al. 2010). The *M. cribraria* was first discovered in the United States in Georgia in 2009. In 2012 this pest was collected in Mississippi for the first time (Catchot 2012). *Megacopta cribraria* goes through two generations per year in the southeastern United States (Zhang et al. 2012). Studies in China determined that first generation adult *M. cribraria* lived between 1.5-3 months, while second generation adults lived 9-10 months as this is the overwintering generation (Zhang and Yu 2005). This insect has piercing and sucking mouth parts that it uses to feed on the sap from the stems and petioles of leguminous plants such as kudzu and soybean (Suiter et al. 2010). Male and female adults can be characterized by the shape of their terminal sternites, with the female identified by a distinct suture and V-shape while the male is rounded (Zhang et al. 2012). Adult *M. cribraria* congregate on the plant host to mate (Hibino 1985). Male *M. cribraria* actively attract the females, while the females are passive. Females prefer mating with larger males (Himuro et al. 2006). Eggs are deposited in clusters of two parallel rows, and the time of egg hatching is positively associated with temperature and occurs in the morning (Thippeswamy and Rajagopal 2005). Oviposition tends to take place in the upper canopy of leguminous crops on the petioles and underside of leaves (Tayutivutikul and Yano 1990). The eggs of the kudzu bug are oval shaped and white in color (Zhang et al. 2012). After emergence, nymphs will feed on a symbiotic gut capsule deposited at the base of the egg mass by the female (Jenkins 2010). Immature *M. cribraria* undergo five nymphal stages before molting into an adult. Adult kudzu bugs will appear white in color when they are newly molted then turn a shade of olive green (Waldvogel and Alder 2011). By feeding on the stems of the kudzu plant, *M. cribraria* most likely causes damage by physiologically stressing the plant (Ruberson et al. 2013). Kudzu bugs can reduce biomass of kudzu up to 32.5% after one year of infestation (Zhang et al. 2012), so in some contexts, it could be considered a beneficial insect.

Pest Status and Damage in Soybean

M. cribraria is considered a pest in areas of its native land (Zhixing et al. 1996). In the United States, the kudzu bug produced an average yield loss of 18% in studies done with untreated soybean fields in Georgia and South Carolina (Greene et al. 2012). When soybeans were exposed to adult *M. cribraria* populations exceeding 40 adults per plant, some delayed growth was noted as well as a decline in number of trifoliates, but this did not result in yield loss (Kikuchi and Kobayashi 2010). In cage trials consisting of soybean plots infested with 25 *M. cribraria* adults per soybean plant during early reproductive growth stages, densities increased to 184 nymphs and adults per plant later in the season, which resulted in a yield loss of 60% (Seiter et al. 2013b). Experiments conducted in Tifton, GA suggest that April and May planting dates receive more yield inhibiting pressure for *M. cribraria* than June or July planting dates (Blount et al. 2016). This could be due to the feeding habits of the insect coming out of overwintering. A study was performed in South Carolina to estimate the economic injury level from *Megacopta cribraria* on reproductive stage soybean using the formula C=VDIK. The study estimated economic injury levels ranging from 111 cumulative insect days per plant with control costs at $20.00/ha and commodity price being $0.6431/kg to 254 cumulative insect days per plant with control costs at $30.00/ha and commodity price being $0.4226/kg (Seiter 2014). They estimated that 0.2803 ± 0.0395 kg/ha of grain was lost for every insect day per plant.

Pest Status and Damage to Homeowners

The tendency to aggregate on white surfaces, as well as the overwintering habits combined with a foul odor exuded when disturbed, make *M. cribraria* a nuisance pest of urban structures (Suiter et al. 2010). The overwintering behavior that resulted in large aggregations on homes led to the discovery of *M. cribraria* in 2009. Along with being a pest based on the overwhelming numbers inhabiting urban areas, *M. cribraria* has been documented to produce a secretion that results in skin irritation and discoloration when crushed against the surface of the skin (Ruberson et al. 2013b).

Management Options

Biological Control

Megacopta cribraria is not known to have many natural enemies in its native area. However, a few have been documented in the United States. Egg masses on kudzu collected in Georgia, Alabama, and Mississippi showed parasitism by the wasp, *Paratelenomus saccharalis* Fabricius, (Gardner et al. 2013). Experiments were conducted in Auburn, Alabama, where approximately 300 adult *M. cribraria* were collected from soybean fields, to study female reproductive development (Golec et al. 2013). Upon dissection of the insects, researchers discovered individuals parasitized by the dipteran, *Strongygaster triangulifer* Loew. Two hundred fourteen specimens of adult *M. cribraria* were dissected, with the parasitism rate being 5%, regardless of the gender. This parasitoid generally parasitizes Coleoptera, not Hemiptera. Reports from India showed that *Beauvaria bassiana* (Balsamo) Vuillemin, a fungal pathogen, attacks *M. cribraria* (Borah and Dutta 2002). *M. cribraria* infected with *B. bassiana* have also been found in the southern United States (Ruberson et al. 2013a).

Beauveria bassiana, commonly known as the white-muscardine fungus, occurs worldwide (Tanada and Kaya 1993). This fungus commonly resides in Lepidoptera, Coleoptera, and Hemiptera (Tanada and Kaya 1993), but it has also been found in the nasal passages of humans as well as the lungs of rodents (McCoy et al. 1988). Infection of *B. bassiana* generally occurs through the integument. However, infection can occur in the digestive tract as proven by Broome et al. (1976). They fed *B. bassiana* conidia to fire ant, *Solenopsis richteri* Forel, larvae, which led to the conidia germinating in the digestive tract and penetrating the gut wall. *Beauveria bassiana* also attacks the larval digestive tract of the corn earworm, depleting the larvae of nutrients and causing starvation (Cheung and Grula 1982). Tests conducted in South Carolina in 2014 introduced this fungus, isolated from infected specimens, to be introduced to healthy adult *M. cribraria* (Seiter et al. 2014). White mycelia emerged from between the segments of host insects. Infected adults died 7 and 9 days after inoculation, while the control specimens remained uninfected. The use of *B. bassiana* as a biological control could be enhanced where populations of a host are high or under environmental conditions suitable for fungal growth (Seiter et al. 2014).

Chemical Control

Since the introduction of *M. cribraria* into the United States, the most common form of management in soybean has been synthetic insecticides (Seiter et al. 2015a). Studies were conducted in South Carolina and North Carolina to evaluate insecticide efficacy using a variety of chemicals. Bifenthrin was the most effective active ingredient tested (Seiter et al. 2015a). Presently, the recommended economic threshold level for *M. cribraria* in Mississippi is an average of 5 bugs per plant before the soybean blooms and an average of 1 nymph per sweep after bloom (Catchot et al. 2016) based on studies in the Southeast. A study conducted in the southeast United States showed that a single insecticide application at the R3 growth stage was adequate to protect against yield loss and reduce the total number of sprays (Seiter et al. 2015b). Additional experiments showed that soybean is most susceptible to *M. cribraria* feeding during pod and seed development (Seiter et al. 2016).

Residual efficacy of nine insecticides was tested on building materials commonly used in urban structures. This test showed pyrethroids and pyrethroid-neonicotinoid mixes were the most effective, providing 100% mortality 24 hours after application (Seiter et al. 2013).

There is no research published on the use of insecticidal seed treatments to control *M. cribraria.* However, based on the success of certain seed treatments in controlling other hemipteran insects (Willrich et al. 2003), it would be reasonable to think seed treatments would be effective against early season infestations of *M. cribraria*.

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CHAPTER II

REFINING THRESHOLD AND EVALUATING KUDZU BUG IMPACT ON VEGETATIVE STAGE SOYBEAN

Abstract

Kudzu bugs were first discovered on soybean in Mississippi in 2013. Although its primary host is kudzu, they have been shown to have damaging effects on soybean in its native land of Asia and in southeastern states in the U.S. Trials were conducted to evaluate the effect of kudzu bugs on vegetative stage soybean in Mid-South growing operations. At infestation levels up to 3 kudzu bugs/plant, there was no evidence of yield loss to soybean. Based on the low likelihood of having infestation levels greater than 3/plant on small plants, there is no reason to suggest that kudzu bugs will cause economic damage to vegetative stage soybean.

Introduction

The kudzu bug, *Megacopta cribraria* F. is a relatively new pest to soybeans in Mississippi, having only been found in this crop throughout the state since 2013. Both the adults and the nymphs of the kudzu bug commonly congregate at the plant nodes and feed on sap from the stems and petioles of the soybean (Tayutivutikul and Yano 1990). Kudzu bug damage resulted in an average yield loss of eighteen percent in Georgia and South Carolina in 2010 and 2011 (Ruberson et al. 2013a). This pest is capable of reducing soybean yield greater than fifty percent under high feeding pressure (Seiter et al. 2013b).

In recent years, Mid-South soybean operations have moved away from traditional soybean production systems that are still commonly practiced in the southeastern United States. These traditional systems consist of planting maturity group V, VI, and VII soybeans in late May and June (Heatherly 1999). In the Mid-South, planting maturity group IV and V soybeans during the months of April and May produce higher yields. These earlier planting dates and maturity groups are recommended to farmers in the Mid-South to prevent late season pest pressure as well as avoiding yield loss that can occur from drought that is common in late summer. The difference in production systems between the two regions left questions unanswered regarding the management of the kudzu bug during vegetative growth stages. To this point, soybean operations in the Mid-South have been using an adopted threshold from South-East production systems. Because *Megacopta cribraria* is capable of maturing completely from egg to adult on early vegetative stage soybean plants (Del Pozo- Valdivia and Reisig 2013), our objective was to identify the impact of *M. cribraria* on vegetative stage soybeans as well as test the validity of the current threshold within a Mid-South soybean production system.

Materials and Methods

Tests were conducted at the R. R. Foil Plant and Science Research Center in Starkville, MS during the 2016 and 2017 growing seasons to develop an economic threshold for kudzu bug in vegetative stages of soybean. Asgrow 5335 variety soybeans were planted on 9 May 2016 and 20 June 2016 on conventional tilled beds with 96.5 cm row spacing at a density of 26 plants per row meter. 1.8 m x 1.8 m x 1.8 m field cages covering two rows of soybean were erected prior to kudzu bug infestation. Infestation levels of 0, 1, 3, 5, or 10 bugs per plant were infested at plant growth stages of vegetative cotyledon (VC), V2, or V4. Each infestation was replicated three times. Kudzu bugs were collected from kudzu in surrounding areas on the day of infestation. Cages were infested on 16 May 2016 at the VC stage, 24 May 2014 at the V2 stage, and 14 June 2016 at the V4 stage for the first planting date. For the late planting date, cages were infested only at the V2 stage on 7 June 2016. *Megacopta cribraria* was kept in the cages to feed for three weeks or until the majority of the plants began flowering. At the end of the three weeks, the cages were removed and a broad spectrum insecticide (bifenthrin 467.6 ml/ha) was applied to the plants to rid the plots of any other soybean pests as well as infestations of *M. cribraria*. The plots were continually monitored and kept free of economically damaging insect populations for the rest of the growing season. Estimates of kudzu bugs/ plant as well as total number of infested plants were recorded weekly for three weeks to monitor the vigor of the insect on the soybean plant. A plant was considered infested if at least one kudzu bug was found on it. Upon maturity of the soybeans, the plants were harvested using a 2 row Kincaid (Massey Ferguson) 8XP plot combine. This combine had a 2 m platform header and data collection system that was used to measure yield, seed moisture and test weight of each plot. Using PROC GLIMMIX with a significance level of 0.05. Data were analyzed in SAS [SAS Institute, Version 9.4, Cary, NC].

Due to extremely low populations of *M. cribraria* in the 2017 growing season, it was not possible to collect enough kudzu bugs to conduct the trial. Therefore, there are no data available for the 2017 growing season.

Results and Discussion

For an unknown reason, nearly all kudzu bugs died in both V2 infestations. Therefore data are only shown for VC and V4 infestations. Within both growth stages, there were no differences in the plant growth measurements between uninfested and infested treatments. In the week between initial infestation and the first recorded observations, a large percentage of the infested *M. cribraria* died. From the VC infestation, no infestation level averaged more than 50% of the soybean plants infested with kudzu bugs (Table 2.1) and the % plants infested varied with infestation level ($F=42.98$, $df=4,36$, $P<0.0001$). Insects also clumped after the V4 infestation, so that the highest percentage of infested plants at any infestation level was 56% and the % plants infested varied with infestation level ($F=33.71$, $df=4,38$, $P<0.0001$). In both the VC infestation and the V4 infestation, there was no impact on soybean yield (VC: $F=0.68$ $df=4,10$ $P=0.6241$) (V4: $F=1.28$ $df=4,7.392$ $P=0.3579$) (Figure 2.1). Survival during the week after infestation averaged 21% and 37% in cages during VC and V4 stages, respectively. There was a significant difference in the yield between the stages of infestation ($F=7.37$ $df=1,19$ $P=0.0137$) (Figure 2.2), but this is likely due to the shade caused by the cage, not the insects. Although there are data showing the damaging effects of kudzu bug on reproductive stage soybean (Zhang et al. 2012), densities up to 3 kudzu bug/plant were not damaging. A greenhouse trial was conducted in Auburn, Alabama to test kudzu bug preference to soybean at different growth stages. Data showed the mean proportion of kudzu bug on R1, R3, R5, and V2 stage soybean was 0.40, 0.34, 0.15, and 0.11, respectively (Yang et al. 2017). Similarly, during a survey conducted in 2017, kudzu bugs seemed to prefer reproductive stage soybean to vegetative stages (See Chapter III). This preference in soybean maturity suggests that densities greater than 3 kudzu bugs/plant is not likely. Economic damage from kudzu bug on vegetative stage soybean is expected to be rare.

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Table 2.1 Mean cage observations taken 1, 2, and 3 weeks after infestation. Yield data corrected to 13% moisture.

GROWTH STAGE	# BUGS INFESTED/PLANT	# BUGS RECOVERED/PLANT	% PLANTS INFESTED
VC	0	0	0
VC	1	0.2	11%
VC	3	0.4	13%
VC	5	1.1	30%
VC	10	3	50%
V4	0	0	0%
V4	1	0.5	21%
V4	3	1	30%
V4	5	1.5	40%
V4	10	3.6	56%

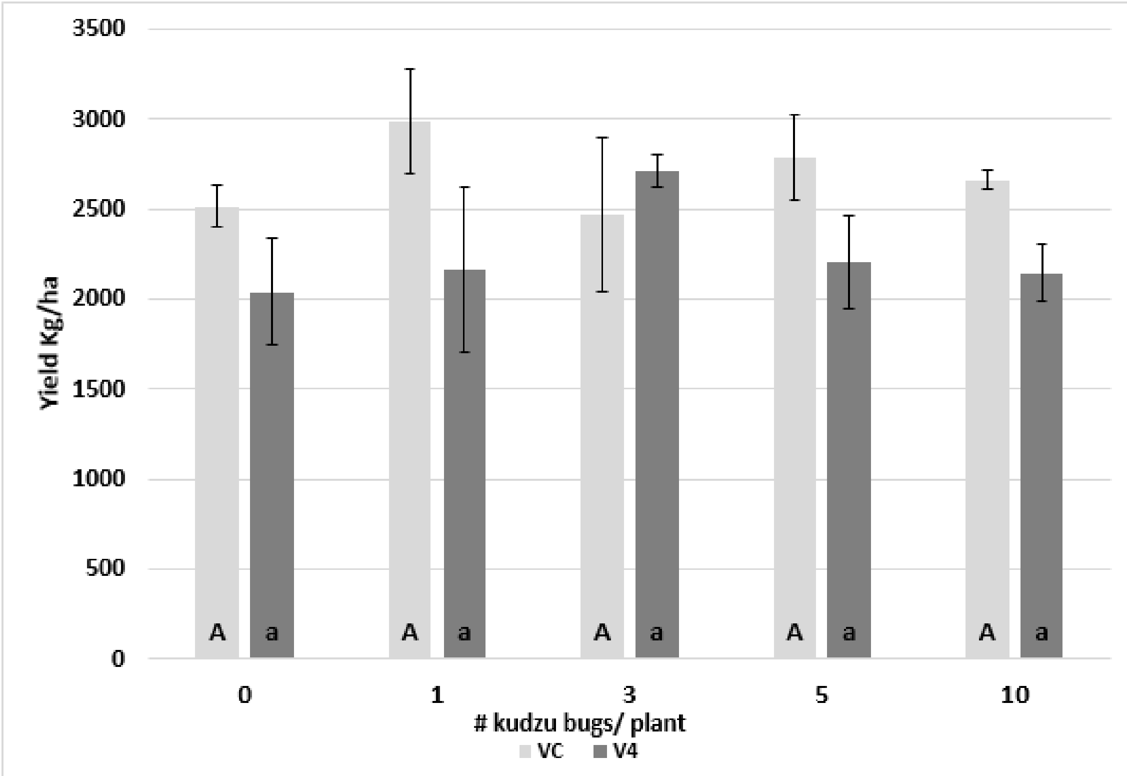

Figure 2.1 Soybean yield as impacted by kudzu bug infestations

Yield (+SEM) of soybean when infested with kudzu bugs for three weeks beginning at VC or V4 growth stages. Starkville, MS, 2016. Treatments containing the same letter are not significantly different (Fishers Protected LSD, α=0.05). VC and V4 infestations analyzed independently.

CHAPTER III

FOLIAR AND SEED TREATMENT INSECTICIDE EFFICACY ON KUDZU BUG

Abstract

The kudzu bug, *Megacopta cribraria* (F.), is a native species of Asia where it is commonly found on leguminous plants. First discovered in the U.S. in Georgia in 2009, this insect has quickly spread throughout the Mid-South and Southeastern United States. Although its primary host is kudzu, *Pueraria montana* (Lour.) Merr., *M. cribraria* can move into soybean, *Glycine max* (L.) Merr., where it is capable of causing economic damage. Chemical control is the most widely used management tactic for controlling the kudzu bug. Two trials were conducted to study the efficacy and residual of commonly used insecticides on adult and nymph *M. cribraria.* Overall, clothianidin and bifenthrin provided the highest mortality of adult kudzu bug up to 24 hrs after application. Greatest control of nymphs was with bifenthrin and lambda-cyhalothrin. Efficacy was low in all treatments by 7 days after application. No seed treatments provided mortality that consistently separated from the untreated control.

Introduction

Kudzu, *Puereria montana* (Lour.) Merr., and soybean, *Glycine max* (L.) Merr., are the primary hosts of kudzu bug, *Megacopta cribraria* F., that allow for development in the southeastern United States (Suiter et al. 2010). The kudzu bug feeds on the stem and petioles of soybean and can cause reduced photosynthesis, deformed pods, and yield loss (Xing et al. 2006). In Asia, *M. cribraria* has been found to reduce yield of field bean, *Lablab purpureus* Linnaeus, up to 44 percent (Thippeswamy and Rajagopal 1998). Thippeswamy and Rajagopal (1998) found that damage was more common with populations of kudzu bug during the vegetative stage of the crop rather than during the reproductive stage. If not for its potential to damage commercial, leguminous crops, *M. cribraria* could be considered a beneficial insect because of its ability to significantly reduce the biomass of kudzu (Zhang et al. 2012). However, under certain conditions, *M. cribraria* can be a significant pest on soybean. The offspring of the overwintering generation can abandon kudzu and finish their life cycle on early planted soybean (Del Pozo- Valdivia and Reisig 2013). Trials conducted in the Southeast U.S. in 2010 and 2011 showed yields in untreated soybean fields reduced from 0 to 47 percent compared to treated fields (Greene et al. 2012). Caged plot trials with 25 kudzu bugs/ plant infested at soybean growth stage of V12, had yields reduced up to 60 percent compared to uninfested cages (Seiter et al. 2013a). In that study, soybean plants in the infested plots had smaller pods than plants in the non-infested plots.

Since the establishment of *M. cribraria* in the United States in 2009, the most common form of management has been the use of synthetic insecticides (Seiter et al. 2015a). In one trial in North Carolina the pyrethroid, bifenthrin, proved to have the best efficacy. *M. cribraria* has proven to be a pest in urban areas as well (Seiter et al. 2013b), so the residual efficacy of nine insecticides was tested on building materials commonly used in urban structures. Pyrethroids, along with pyrethroid-neonicotinoid mixes, provided the highest efficacy of 100 percent mortality up to 24 hours after application. Insecticides are a critical component of kudzu bug management in soybean, but the duration of efficacy after an application has not been studied. For this reason, a trial was conducted to evaluate the longevity of potential insecticides for kudzu bug management.

The concept of placing pesticides on seed to aid in the control of crop pests is not a new idea. In fact, the practice was implemented in A.D. 50 by Junius Columella (David and Gardiner 1955). Modern seed treatments began with organic mercury used as a fungicide in the 1920's (Munkvold et al. 2006). The idea of treating a seed with insecticide that could translocate in the plant, and render the plant insecticidal was first noted in 1952 (Dowdy and Slessman 1952). While some early seed treatments provided good insect control, under certain conditions the use of these seed treatments resulted in poor germination, delay in maturity, and seedling phytotoxicity (Adkisson 1958). By the 1990's, advancements in insecticides and seed treatment had overcome earlier insecticidal seed treatment issues (Elbert et al. 2008). Neonicotinoids have proven to be one of the most effective insecticide classes for seed treatments because they translocate and control pests with sucking mouthparts. These insecticides work against the nicotinic acetylcholine receptor and have long residual activity (Maienfisch et al. 2001). Before the introduction of neonicotinoid seed treatments in 1990, the agrochemical market was dominated by organophosphates, pyrethroids, and carbamates. By 2005, neonicotinoids gained a 16 percent market share by replacing mainly organophosphates and carbamates. By 2011, 75 percent of soybean seed planted in Mississippi were treated with an insecticidal seed treatment, most of which were neonicotinoids (Musser et al. 2012). There is no research available showing the effects of neonicotinoid seed treatments on kudzu bug. However, because of the effectiveness neonicotinoids on insects with sucking mouthparts, it is logical to believe that they would be effective on kudzu bugs as they also have sucking mouthparts.

Therefore a trial was conducted to evaluate the impact of neonicotinoid seed treatments on kudzu bug mortality. Because the kudzu bug has piercing and sucking mouthparts, we hypothesized that seed treatments may work similarly on the kudzu bug as they do on other sucking insects such as thrips (Thysanoptera) and threecornered alfalfa hopper, *Spissistilus festinus* Say.

Materials and Methods

Foliar Insecticides

The efficacy of foliar insecticides against kudzu bug adults and nymphs was tested at the R.R. Foil Plant Science and Research Center in Starkville, MS during the 2016 and 2017 growing seasons. The insecticides tested were those that were labeled for use in soybean. In both years, experiments were designed as a randomized complete block with four replications with six insecticide treatments and an untreated control plot. The plots were four rows wide with a row spacing of 96.5 cm and 9.1 m long. Treatments are listed in Table 3.1 along with an untreated control. Insecticides were applied with a Mudmaster (Bowman Manufacturing, Newport, AR) sprayer with multiple four row spray booms. Insecticides were applied at 93.5 L / ha and 448 kPa (10 GPA and 65 P.S.I.) with TeeJet TX6 hollow cone spray tips. The soybean were at the R3 (Fehr and Caviness 1977) growth stage when the application was made. One 9 cm diameter piece of filter paper was placed into each 100 mm diameter X 15 mm petri dish and then dampened with water to insure the soybean tissue did not become dry. Leaf and petiole samples from each plot were collected at 1 hr, 1 day, 3 days, 5 days, 7 days, and 10 days after insecticide application. A single leaflet of a trifoliate and the base of the petiole, 2.5 cm long, randomly selected from the upper plant canopy of each plot was individually placed in ten petri dishes along with two adult or two nymph (3rd instar) kudzu bugs. Kudzu bugs used in the trial were collected from commercially grown soybean fields within 24 hrs of the start of the assay. The petri dishes were sealed with Parafilm M® All-Purpose Laboratory Film (Product No. 13-374-12, Fisher Scientific, Norcross, GA) to retain moisture and prevent escape and placed in a growth chamber at 26.6 ˚C with 70 % relative humidity and 16:8 hour light to dark photoperiod. Each infested petri dish was rated after 24 hrs and mortality was recorded. During the 2016 growing season, the assay was performed 1 hr and 1 day after spraying. Adult kudzu bug populations crashed after this, so kudzu bugs could not be collected at 3, 5, and 7 days after application. Each insect in a petri dish was recorded as either dead or alive. An insect was considered dead if no movement was detected. Data were corrected for control mortality using Abbott's (1925) formula (Abbott 1925). Data were analyzed in SAS (SAS Institute, Version 9.4, Cary, NC) using PROC GLIMMIX with a significance level of 0.05.

Insecticidal Seed Treatments

Similar to the efficacy test of foliar insecticides, during the 2016 growing season, experiments were conducted to test the efficacy of selected insecticidal seed treatments against kudzu bugs. Three different seed treatments along with an untreated control were planted in replicated small plots at the Delta Research and Extension Center in Stoneville, MS. The trial at the Delta Research and Extension Center took place twice during the 2016 growing season with the first one being planted on 30 June 2016 and the second on 15 July 2016. Each planting date consisted of three insecticide seed treatments (imidacloprid 0.78g a.i./kg of seed, clothianidin 0.5g a.i./kg of seed , thiamethoxam 0.5g a.i /kg of seed) along with non-treated soybean. Seeds were planted in each plot at a density of 29 seed/ row meter. Ten cages (20.32 cm diameter X 30.48 cm in height) each covering a soybean plant were infested with two adult kudzu bugs when the plants reached VC (vegetative cotyledon), V1 (first trifoliate), and V2 (second trifoliate) growth stages. The insects were left to feed on the plants for 48 hours. After this time, the cages were removed from the plants to evaluate mortality. Each treatment was replicated four times.

During the 2017 growing season, trials were conducted in Stoneville, Mississippi at the Delta Research and Extension and Center and in Starkville, Mississippi at the R.R. Foil Plant and Research Station. In Stoneville, the trial was conducted using a randomized complete block design, and in Starkville the trial was conducted using a completely randomized design. Each planting location consisted of three different insecticide seed treatments (imidacloprid 0.78g a.i./kg of seed, clothianidin 0.5g a.i./kg of seed , thiamethoxam 0.5g a.i /kg of seed) along with soybean not treated with insecticide. All seed was treated with ApronMax RFC fungicide (0.061g a.i./kg of seed). In Stoneville, Asgrow 4632 variety seed were planted on 5 July 2017 at a density of 29 seed/ row meter. In Starkville, Asgrow 5335 variety seed was planted on 26 June 2017 at the same density. Laboratory bioassays were utilized at both locations to test insecticide seed treatment efficacy. Soybean plants were allowed to grow in the field until the plants reached VC growth stage. Ten individual plants from each plot were then cut from the plot and a 2.5cm long stem along with both cotyledons was placed into a petri dish. Two adult kudzu bugs were placed into the petri dish and allowed to feed on the plant tissue for 24 hrs. After 24 hrs, kudzu bugs were rated for mortality. This test was repeated using stem samples at V1 and V2 growth stages in Stoneville and V1 growth stage in Starkville to determine duration of the efficacy of the seed treatments.

Results and Discussion

Foliar Insecticides

During the 2016 growing season, there was no difference in efficacy between 1hr and 1 day ($F=0.6$; $df=5,33$; $P=0.7005$), therefore the means over both ratings are presented. Mortality ranged from 61-100% ($F=12.9$; $df=5,39$; $P<0.01$). All 6 insecticide treatments provided some mortality (Figure 3.1). Clothianidin at 77g a.i./ha (1.1 oz./acre), bifenthrin at 56g a.i./ha (0.8 oz./acre), bifenthrin at 112g/ha (01.6 oz./acre), and acephate at 340g/ha (12 oz/acre) caused the highest mortality and were not significantly different from each other. During the 2017 growing season, mortality of adult kudzu bug ranged from 0 to 100% among treatments (Figure 3.2). All treatments in assays conducted 1 hr after application caused some mortality ($F=3.2$ $df=5,18$ $P=0.03$). Clothianidin, acephate, and the high rate of bifenthrin caused greater mortality than dimethoate and were not different from each other. Lambda-cyhalothrin and the low rate of bifenthrin were not significantly different from dimethoate or clothianidin, acephate, or the high rate of bifenthrin. At 1 day after application, all treatments still provided some mortality but were not different from each other (F= 0.77; df=5,15; P=0.58). Data collected at 3 days after application were removed from the analysis due to high control mortality. The high mortality is believed to be due to excessive moisture in the petri dishes from the wetting of the filter paper. Acephate provided the highest mortality at 6 days after application, and all treatments provided some mortality (F=14.66; df=65,18; *P*<0.01). Mortality from other treatments was not different from the untreated control. Clothianidin and acephate were the only two chemicals to provide at least 15 percent mortality 6 days after application, therefore, those chemicals were the only two that were tested against the untreated control 14 days after application. Mortality in the acephate, clothianidin and the treatments were not significantly different at 14 days after application. Throughout the trial, clothianidin and acephate provided as good or higher mortality than all other insecticides. The mortality provided by acephate on kudzu bug was expected as this efficacy has been previously reported (Brown et al. 2015).

Similar to the adult kudzu bug trial, a trial was conducted on kudzu bug nymphs during the 2017 growing season. In this trial, mortality ranged from 1-89% (Figure 3.3). Assays 1hr after application resulted in all insecticide treatments causing some mortality (F=2.8; df=5,15; P=0.05). At 1 day after application, all treatments caused some mortality and were not significantly different from each other (F=0.80; df=5,15; P=0.56). As with the adult trial, data for the nymph trial at 3 days after application was removed from analysis due to high control mortality. Between five and six days after insecticide application, a 10 cm rainfall event occurred. Seven days after application, only lambda-cyhalothrin and the low rate of bifenthrin provided mortality higher than 10 percent (F=4.1; df= 5,15; P=0.01). Due to low mortality among all treatments at 7 days after application, no treatments were tested at 14 days after application. No previous efficacy trials on kudzu bug nymphs have been reported. However, a trial at Louisiana State University was conducted to test the efficacy of southern green stink bug nymphs, *Nezara viridula* Linnaeus, using similar treatments. They found that lambda-cyhalothrin provided higher mortality than all other treatments (Willrich et al. 2003). This is consistent with the kudzu bug nymph trial, suggesting that lambda-cyhalothrin provides good control of immature hemipteran insects.

Insecticidal Seed Treatments

During the 2016 growing season, there were no significant interactions between planting date and seed treatment (F=0.44, df=1,69, P=0.7217) or plant growth stage and seed treatment on insect mortality (F=0.61; df=6,69; P=0.7205). Seed treatment was a significant factor on kudzu bug mortality when data were pooled across both planting dates and all rating dates (F=4.29; df=3,69; P=0.0078) (Figure 3.4). While thiamethoxam seed treatment resulted in higher mortality, average mortality was only 14% higher than the untreated check (48% vs. 34%). During the 2017 growing season, there was no interaction between locations and seed treatment (F=1.46, df=3,56, P=0.2359), so data were pooled across both locations. There was no interaction between seed treatment and soybean growth stage (F=1.00, df=6,63, P=0.4326), therefore data across all growth stages were combined. No seed treatment provided higher mortality than the untreated control (F=0.84, df=3,69.14, P=0.4741) (Figure 3.5).

There are no previous reports of seed treatment efficacy on kudzu bug, but there are reports on other Hempitera. Researchers in Louisiana conducted a trial using clothianidin, imidacloprid, and thiamethoxam seed treatments of corn, *Zea mays* L., to test the efficacy against brown stink bug *Euschistus servus* (Say) and southern green stink bug *Nezara viridula* L. (Willrich et al. 2003a). The stink bugs were caged on seedling corn at V2-V4 growth stages for 72 hours. Willrich et al. (2003) found that all seed treatments provided mortality of both species significantly higher than that found on untreated seed. In 2001 and 2002, researchers conducted trials to assess the efficacy of thiamethoxam seed treatment of snap bean *Phaseolus vulgaris* L. on potato leafhopper *Empoasca fabae* (Harris) (Nault et al. 2004). Nault et al. (2004) found that seed treatments of thiamethoxam at 30g (a.i) and 50g (a.i.)/100kg of seed provided control of leafhopper through bloom stage. Both rates of thiamethoxam provided equal control in contrast to almost 50 percent of the untreated plants expressing symptoms of leafhopper damage (Nault et al. 2004). In past experiments, seed treatments have proven to be beneficial in controlling pests of seedling crops. However, based on our findings, seed treatments are not an effective management option for controlling kudzu bug in vegetative stage soybean.

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Table 3.1 Foliar Insecticide Formulation Rates and Active Ingredient Rates Tested

INSECTICIDE	FORMULATION	FORMULATED RATE	A.I. RATE
Clothianidin	Belay	292ml/ha (4 fl oz./acre)	77g/ha (1.1 oz./acre)
Bifenthrin (low)	Brigade 2 EC	234ml/ha (3.2 fl oz./acre)	56g/ha (0.8 oz./acre)
Bifenthrin (high)	Brigade 2 EC	468ml/ha (6.4 fl oz./acre)	112g/ha (1.6 oz./acre)
Dimethoate	Dimethoate 4 EC	1169ml/ha (16 fl oz./acre)	561g/ha (8 oz./acre)
Lambda-cyhalothrin	Karate Z 2.08	141ml/ha (2 fl oz./acre)	35g/ha (0.5 oz./acre)
Acephate	Orthene 97	340g/ha (12 oz./acre)	330g/ha (11.6 oz./acre)

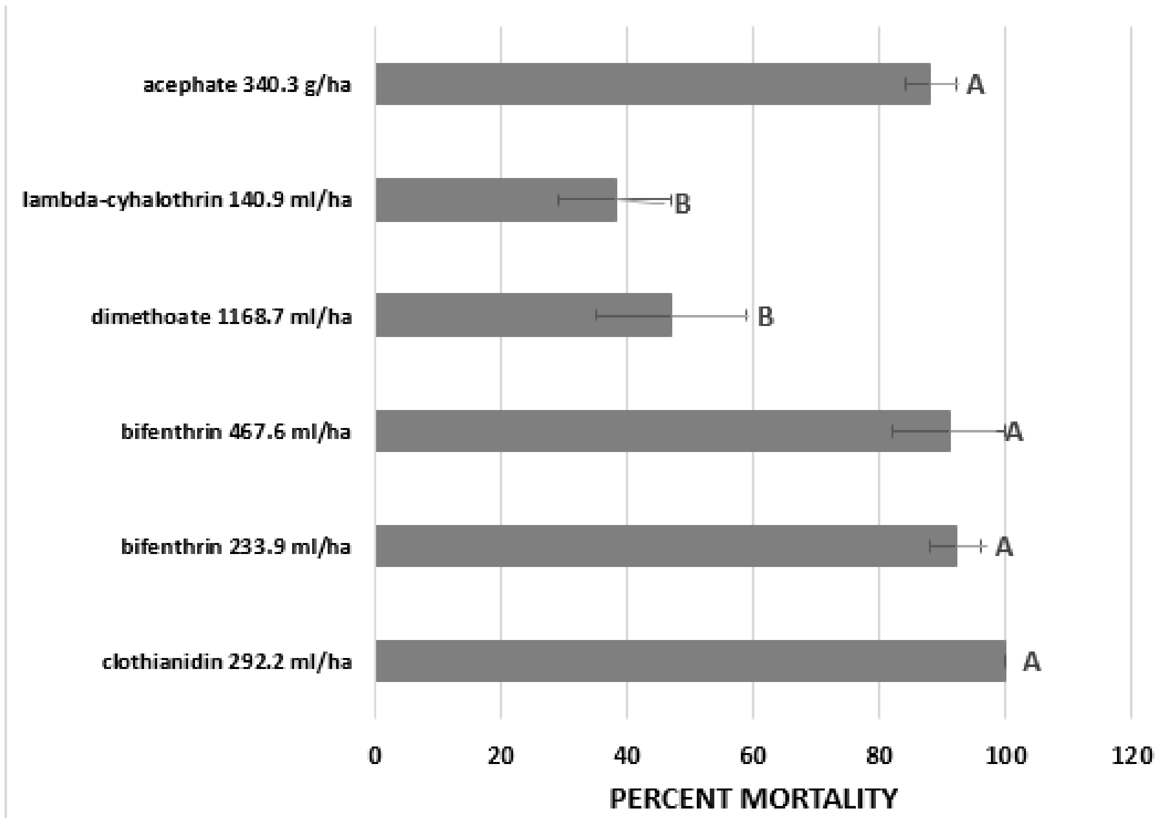

Figure 3.1 Foliar Insecticide Efficacy on Adults in 2016

Average mortality of adult kudzu bugs (+SEM) when fed treated soybean leaves and petioles 1 to 24 hrs after insecticide application. Starkville, MS, 2016. Treatments containing the same letter are not significantly different (Fishers Protected LSD, α=0.05).

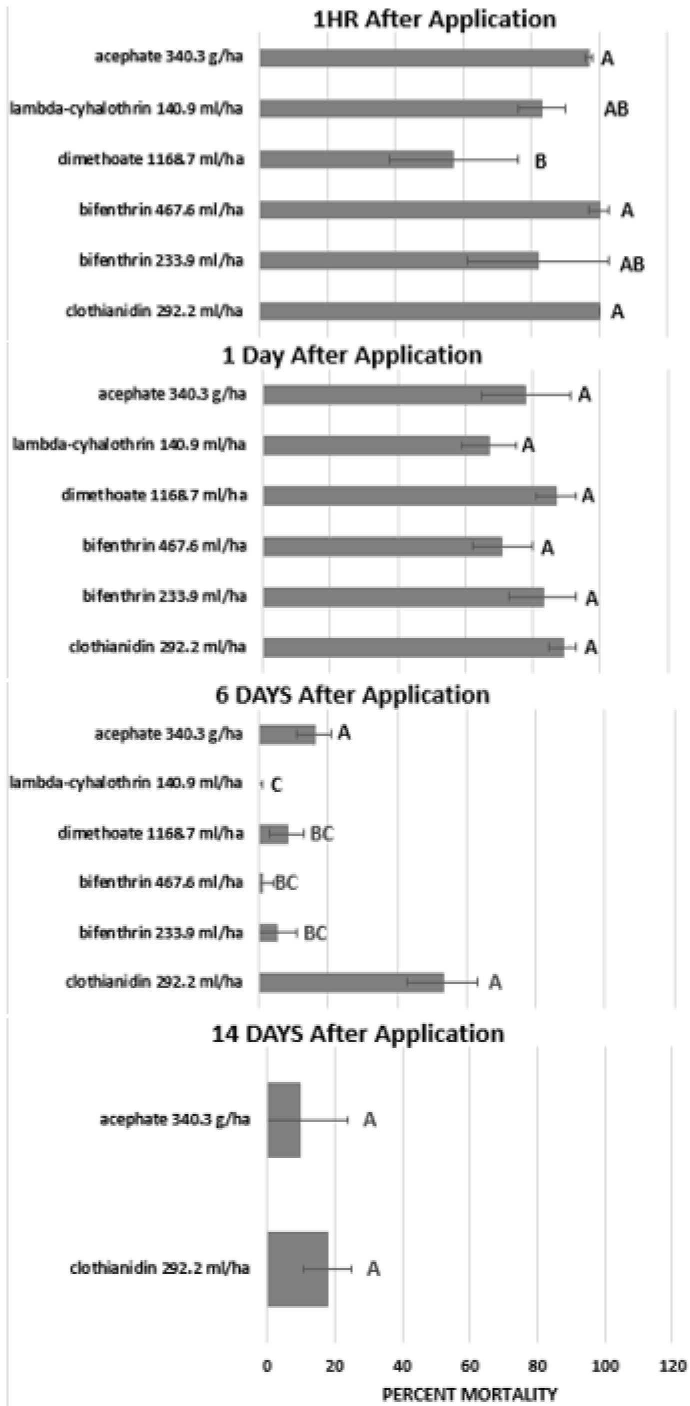

Figure 3.2 Foliar Insecticide Efficacy on Adults in 2017

Average mortality of adult kudzu bugs (+SEM) when fed treated soybean leaves and petioles 1hr to 14 days after insecticide application. Starkville, MS, 2017. Treatments containing the same letter within the same residual time are not significantly different (Fishers Protected LSD, α=0.05).

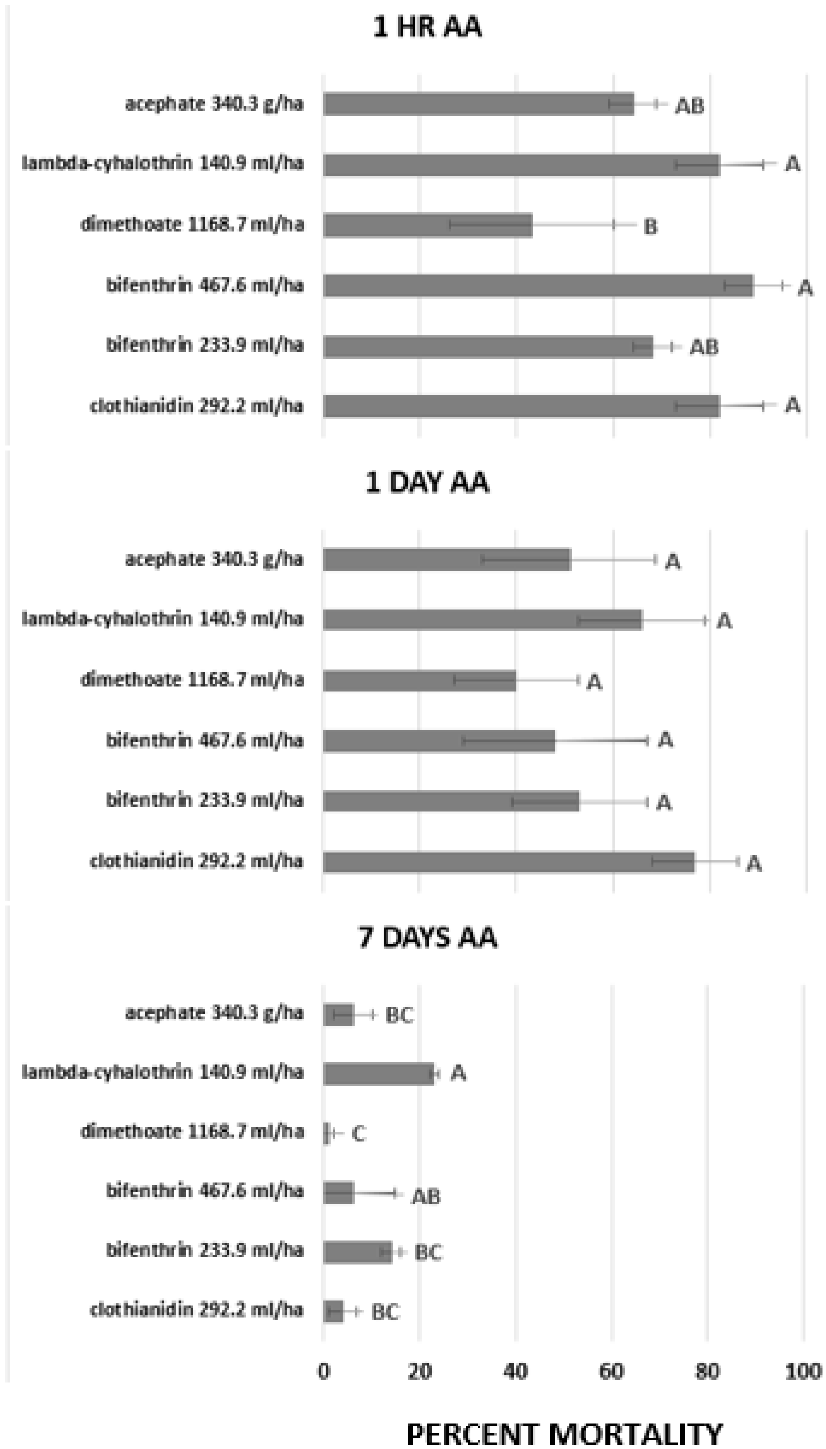

Figure 3.3 Foliar Insecticide Efficacy on Nymphs in 2017

Average mortality of nymph kudzu bugs (+SEM) when fed treated soybean leaves and petioles 1hr to 7 days after insecticide application. Starkville, MS, 2017. Treatments containing the same letter are not significantly different (Fishers Protected LSD, α=0.05).

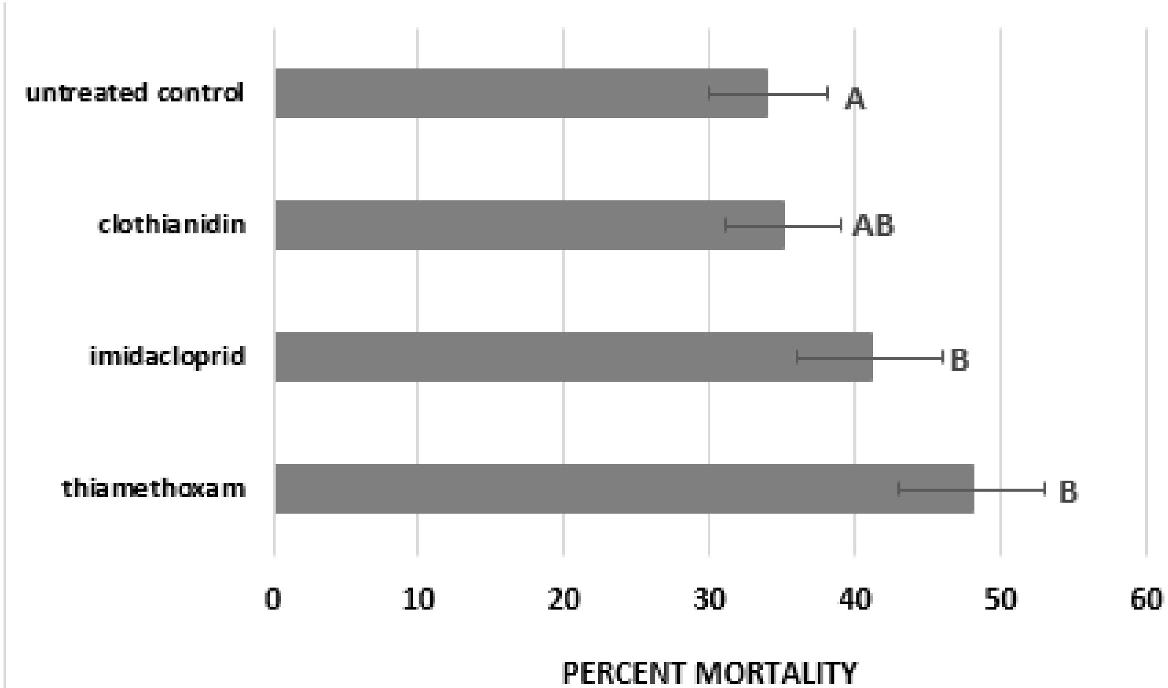

Figure 3.4 Percent Mortality for Each Seed Treatment in Stoneville During 2016

Average percent adult kudzu bug mortality (± SEM) as affected by seed treatments at VC, V1, and V2 soybean growth stages in 2016 at Stoneville, MS. Treatments containing the same letter are not significantly different (Fishers Protected LSD, α=0.05).

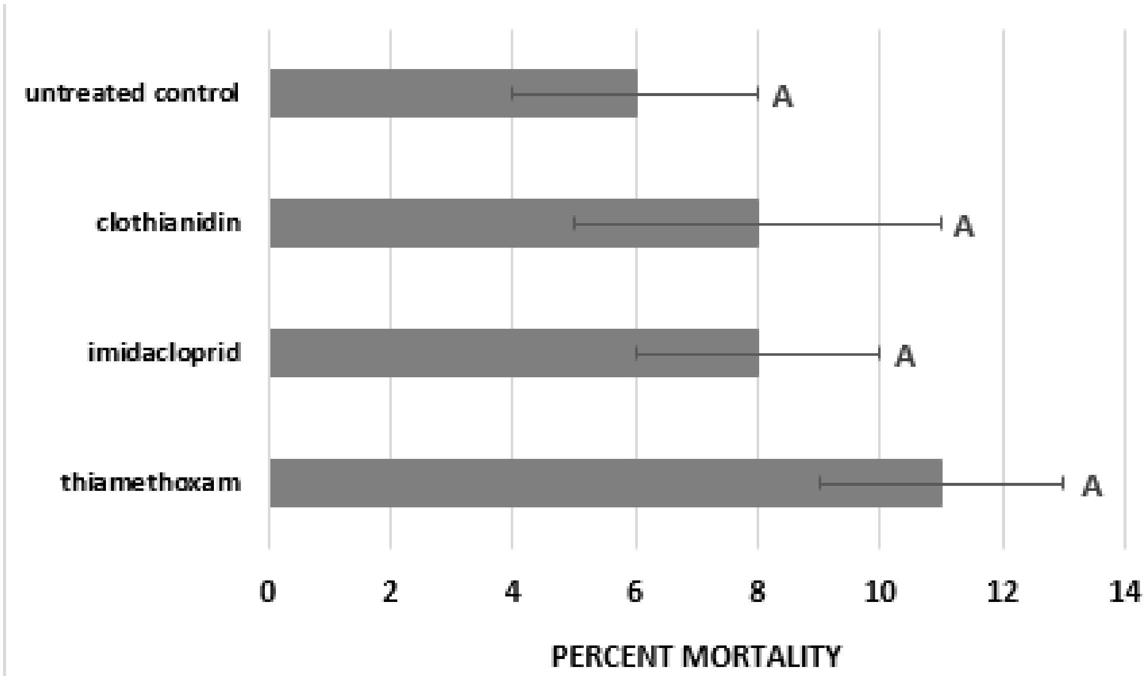

Figure 3.5 Percent Mortality for Each Seed Treatment for Combined Stoneville and Starkville Locations During 2017

Average percent adult kudzu bug mortality (± SEM) as affected by seed treatments at VC, V1, and V2 soybean growth stages in 2017 for combined Stoneville, MS and Starkville, MS locations. Treatments containing the same letter are not significantly different (Fishers Protected LSD, α=0.05).

CHAPTER IV

POPULATION DENSITY AND NATURAL ENEMIES OF *MEGACOPTA CRIBRARIA* IN KUDZU AND SOYBEAN IN MISSISSIPPI

Abstract

The primary hosts of *Megacopta cribraria* (F.) (Hemiptera: Plataspidae) are kudzu, *Pueraria montana* Lour. (Merr.) and soybean *Glycine max* (L.) Merr., so a survey was conducted to evaluate the population dynamics of this insect on these hosts. Kudzu (April - October) and soybean (late June - early October) were sampled every two weeks for adults, nymphs, and egg masses. One or more stages of *M. cribraria* were found in kudzu at all sampling dates. Adult densities increased beginning in early August and peaked in late September. The first nymphs were found in early May, but the number of nymphs found in kudzu was low until early July. Nymph densities peaked in late July and slowly decreased through late September. No nymphs were collected in kudzu after late September. A few egg masses were found in kudzu beginning in early April. The last egg masses found in kudzu were collected during early August. In soybean, adult *M. cribraria* densities did not exceed 10/ 40 sweeps until late September when they hit a peak of 21/ 40 sweeps. After early July, nymph populations slowly increased until early August at which point they peaked at 9/ 40 sweeps. Nymph densities decreased until late September when no nymphs were collected. Similar to that found in kudzu, no egg masses were found in soybean after early August. Based on populations that were

collected, we were not able to clearly determine that there is more than one generation of *M. cribraria* in both soybean and kudzu. However eggs were found from April until August, so two generations per year as previously reported is likely in Mississippi. *Beauveria bassiana* was first observed infecting kudzu bug in late August in both kudzu and soybean, and it was found at every sampling location where kudzu bugs were found for the remainder of the survey. No eggs collected in kudzu or soybean were parasitized.

Introduction

Kudzu bug, *Megacopta cribraria* (F.) (Hemiptera: Plataspidae) is a native species to Asia (Srinivasaperumal et al. 1992) and is called this because its primary host is kudzu, *Pueraria montana* Lour. (Merr.) (Eger et al. 2010). The kudzu bug goes through two generations per year in Asia and in the Southeastern United States (Zhang et al. 2012, Zhang and Yu 2005). First generation adult kudzu bugs live between 1.5-3 months, while the second generation adults live 9-10 months because they are the overwintering generation (Zhang and Yu 2005). Adults begin to oviposit within a week after emerging from overwintering (Del Pozo- Valdivia and Reisig 2013). Large numbers of kudzu bugs move into soybean from kudzu during summer months. The reason or cause for this dispersal is unknown. According to Zhang et al. (2012), kudzu bugs lay eggs on a number of hosts but are only able to develop into adults on kudzu and soybean. Trials conducted in North Carolina found that kudzu bug completed its life cycle in 45-50 days on soybean (Del Pozo- Valdivia and Reisig 2013). Del Pozo- Valdivia et al. (2013) were able to rear first generation kudzu bug with 90 percent surviving to develop into adults on potted soybean in a greenhouse. This is contradictory to the very low survival survival rate on soybean found by Zhang et al. (2012). Del Pozo- Valdivia et al. (2013) suggest that if first generation kudzu bugs can develop on soybean, then it could lead to even bigger second generation populations in soybean.

Kudzu bug is a new pest in the midsouthern United States, first reported in Mississippi in 2012 (Catchot 2012), so it is important to know when these generations occur in this geography. To document this, kudzu and soybean locations across the state of Mississippi were sampled throughout the growing season to get a better understanding of when kudzu bug development occurs in Mid-South environmental conditions. Specifically, our objectives were to determine when the overwintering generation starts to appear, when the overwintering generation begins to oviposit, how much kudzu bug generations overlap, when kudzu bug populations move into soybean and the level of oviposition and insect development in soybean and kudzu.

The capacity to which native natural enemies in North America can control kudzu bug has not been determined, but some native predators have been found to prey on adult and nymph kudzu bug (Ruberson et al. 2013a). Egg masses on kudzu collected in Georgia, Alabama, and Mississippi showed parasitism by the wasp, *Paratelenomus saccharalis* Dodd, (Gardner et al. 2013). *Beauvaria bassiana* (Balsamo) Vuillemin, a fungal pathogen, commonly known as the white-muscardine fungus, occurs worldwide (Tanada and Kaya 1993) and was found to attack kudzu bug in India (Borah and Dutta 2002) and in the Southern United States (Ruberson et al. 2013a). Infection by *B. bassiana* generally occurs through the integument. However, infection can occur in the digestive tract (Broome et al. 1976a) of some species. Seiter et al. (2014) introduced *B. bassiana* to healthy adult kudzu bug and observed white mycelia emerged from between the segments. Infected adults died 7 - 9 days after inoculation, while the control specimens remained uninfected. In the current survey, adult and nymph kudzu bugs were monitored for visual symptoms of infection by *B. bassiana,* while eggs were monitored for *Paratelenomus saccharalis* parasitism.

Materials and Methods

A survey was conducted in Webster, Carroll, Montgomery, Grenada, Yalobusha, Lafayette, Pontotoc, Noxubee, Kemper, and Winston counties in the North-Central and North-East regions of Mississippi from April through October 2017. A standard 38.1 cm (15 in.) sweep net was used to sample wild kudzu patches and commercial soybean fields for populations of *M. cribraria.* Also, leaves from both kudzu and soybean were sampled for egg masses. At each location, four sets of ten sweeps were taken and the number of kudzu bug adults and nymphs in each set of sweeps was recorded. Fifty leaves were sampled by observing the underside of each leaf at each location for egg masses. All collecting dates were separated into fifteen-day intervals representing the early and late part of each month. Approximately 15 locations of kudzu and 10 soybean locations were sampled during each fifteen-day interval. While the same general regions were sampled all year, specific locations changed as needed so potential hosts could be sampled. Each leaf containing an egg mass was collected and placed in a 100 mm diameter X 15 mm petri dish where the eggs were allowed to hatch inside a growth chamber maintained at 26.6 °C with 70 % relative humidity and 16:8 hour light to dark photoperiod. Throughout the survey, eggs were held in the growth chamber and observed daily for parasitoid emergence. All data were analyzed using PROC MEANS in SAS 9.4.

Results and Discussion

Population Dynamics in Kudzu

During the sampling periods from early April through early June, adult kudzu bugs averaged less than 3 insects/ 40 sweeps (Figure 4.1). In the four sampling periods from late June through early August, the populations in kudzu increased and adult kudzu bugs averaged between 5.5 and 15.2 insects/ 40 sweeps with the highest average recorded in early August. Zhang et al. (2012) first observed first-generation adults in late June, consistent with our increased densities. The largest increase in adult kudzu bugs was recorded between the early August sampling period and the late August sampling period. In this fifteen-day sampling interval, the number of adults increased from about 15/ 40 sweeps to almost 40/ 40 sweeps. The population of *M. cribraria* adults continued to increase through late September when the density of adults averaged over 50/ 40 sweeps. Adult kudzu bug numbers began to decline after late September.

No nymphs were found during April, however, some egg masses were collected during this time. The average number of nymphs slowly increased from early May through early July, but never exceeded 7/ 40 sweeps. Likewise, in 2010, Zhang et al. (2012) first observed nymphs in early May. *Megacopta cribraria* nymph densities dramatically increased during late July when they averaged 84/ 40 sweeps. During both sampling periods in August, nymphs averaged about 57/40 sweeps. Kudzu bug nymph denstities began to decrease during early September, and no nymphs were found by early October. From late August to late September, the declining nymph population aligned with the growing adult population as the older nymphs matured into adults.

Throughout the survey, egg mass collections were few, likely due to them being difficult to find in the dense foliage of the kudzu and the small size of the egg masses (Figure 4.2). However, egg masses were collected during each sampling period from early April through late May. Similarly, Zhang et al. (2012) found that oviposition began in the first week of April in Georgia. No egg masses were found in early June. According to Zhang et al. (2012), the second peak time for oviposition was in late June. In late June and early July, egg masses averaged 0.3/ and 0.2 / 50 leaves respectively. Our egg data appear to agree with Zhang et al. (2012), but numbers collected were too low to confidently draw conclusions of generations based on egg masses. No egg masses were collected during late July, and 0.5 egg masses/ 50 leaf sample were collected during early August. From late August through the end of the survey, no egg masses were collected.

Population Dynamics in Soybean

In soybean, the population densities of *M. cribraria* were more eratic than in kudzu (Figure 4.3). Because this was a survey in commercial fields, insecticide applications were made in some fields throughout the growing season, which likely affected the densities of kudzu bugs collected. The survey of soybean began in late June with adult populations averaging 3.3/ 40 sweeps. Beginning in late July and continuing into early September, average adult *M. cribraria* densities were between 5.7 and 6.8 / 40 sweeps. Late September saw an increase in density to 21 / 40 sweeps as nymphs molted into adults. Part of this increased density could also be a result of aggregation into unharvested fields as early-planted soybean fields were being harvested at this time. Kudzu bugs can readily move from harvested fields into the closest available unharvested soybean (Zhang et al. 2012). No nymphs were found in late June or early July soybean even though they were found in kudzu at this time. The first nymphs were found in soybean in late July and peaked in early August at 9 / 25 sweeps. In late August the density decreased and no nymphs were found in soybean after early September. Egg masses were found only during early June and early August (Figure 4.4). Because eggs, nymphs, and adult *M. cribraria* were found in soybean fields, one can assume that soybean is a viable host for the kudzu bug to complete its life cycle, consistent with Zhang et al. (2012) and Del Pozo-Valdivia et al. (2013). Although we began this survey in late June when at least 50 percent of the soybean crop was already in reproductive stages, late-planted fields with vegetative stage soybean seemed to have fewer kudzu bugs than soybean in reproductive stages. Yang et al. (2017) observed that adults that had already infested early-planted soybean would choose to remain on those plants rather than move to younger, later planted soybean. Research conducted in Auburn, Alabama suggests that when given the choice, adult kudzu bugs would prefer to infest soybean at early reproductive growth stage to other growth stages regardless of planting date (Yang et al. 2017). Although no data were collected, it appeared that when higher densities of kudzu bug were found in vegetative stage soybean, they were planted on narrower row spacing (38.1 cm or less). This was not an objective of the survey, so data were not collected to allow testing of this hypothesis. If this anecdotal observation is proven consistently, wide rows would be a cultural control method for kudzu bug. The narrower row spacing provides quicker canopy closure than soybean planted on wider rows, potentially attracting more kudzu bugs. While this behavior is not found among all insects, some other species have been documented to prefer narrower row spacing.

Scaphytopius acutus (Say) a leafhopper, was shown by Troxclair Jr. and Boehtel (1984) to have larger populations in soybean planted on narrower row spacing than on wider row spacing. Higher populations of velvetbean caterpillar, *Anticarsia gemmatalis* Hübner, and southern green stink bug, *Nezara viridula* (L.) were also observed in soybean planted on narrow row spacing (45 cm) than on wider row spacing (90 cm) (McPherson and Bondari 1991).

Natural Enemies

Beauveria bassiana was first observed infecting kudzu bug beginning in late August. From late August until the survey was concluded, *B. bassiana* was found infecting kudzu bug at every sampling location of both kudzu and soybean in which kudzu bugs were found. The abundance of mycelia appeared to vary among locations, but was never absent. Previous experiments showed that best germination of *B. bassiana* occurred at 100 % relative humidity, and relative humidity below 90 percent did not allow for germination (Walstad et al. 1969), which could explain the difference in severity of outbreaks between locations and sampling periods. We were unable to confirm the previous documentation of parasitism of kudzu bug eggs in Mississippi (Gardner et al. 2013) with this survey. No eggs collected during the survey were parasitized. No other natural enemies were observed during the survey in kudzu. Numerous generalist predators were found in soybean, but their impact of kudzu bug is unknown.

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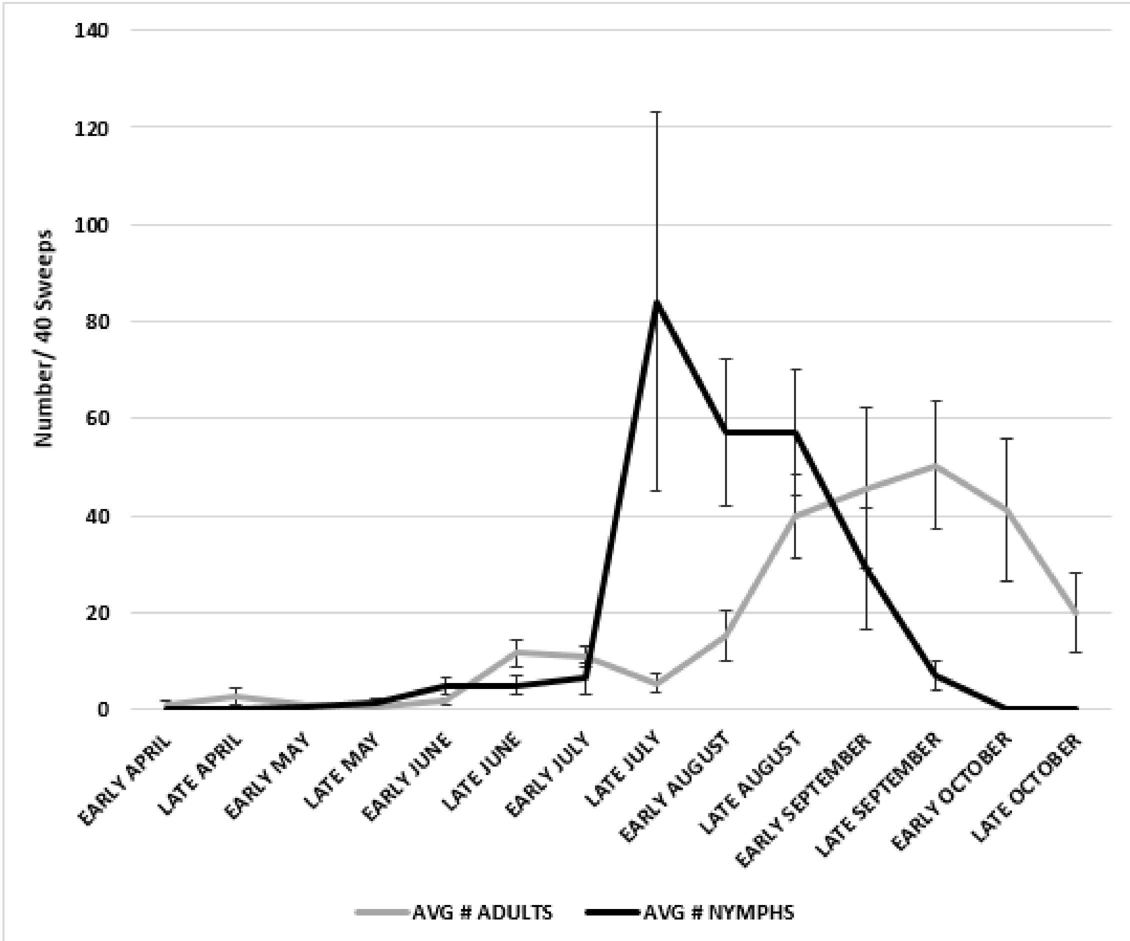

Figure 4.1 Kudzu Bug Population Densities in Kudzu

Average population density of kudzu bug adults and nymphs (+SEM) / 40 sweeps in kudzu from April through October 2017 in Northeastern Mississippi.

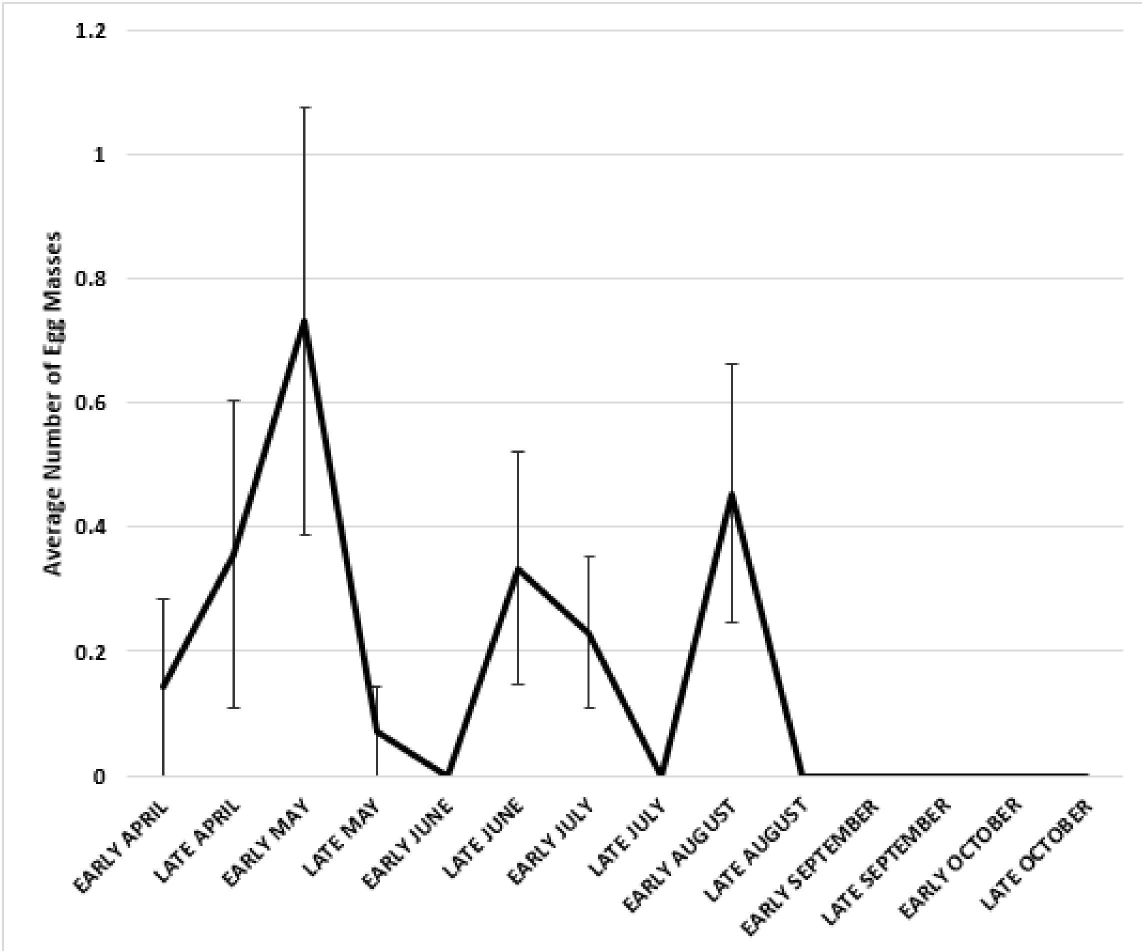

Figure 4.2 Number of Kudzu Bug Egg Masses in Kudzu

Average number of kudzu bug egg masses (+SEM)/50 kudzu leaves from April through October 2017 in Northeastern Mississippi.

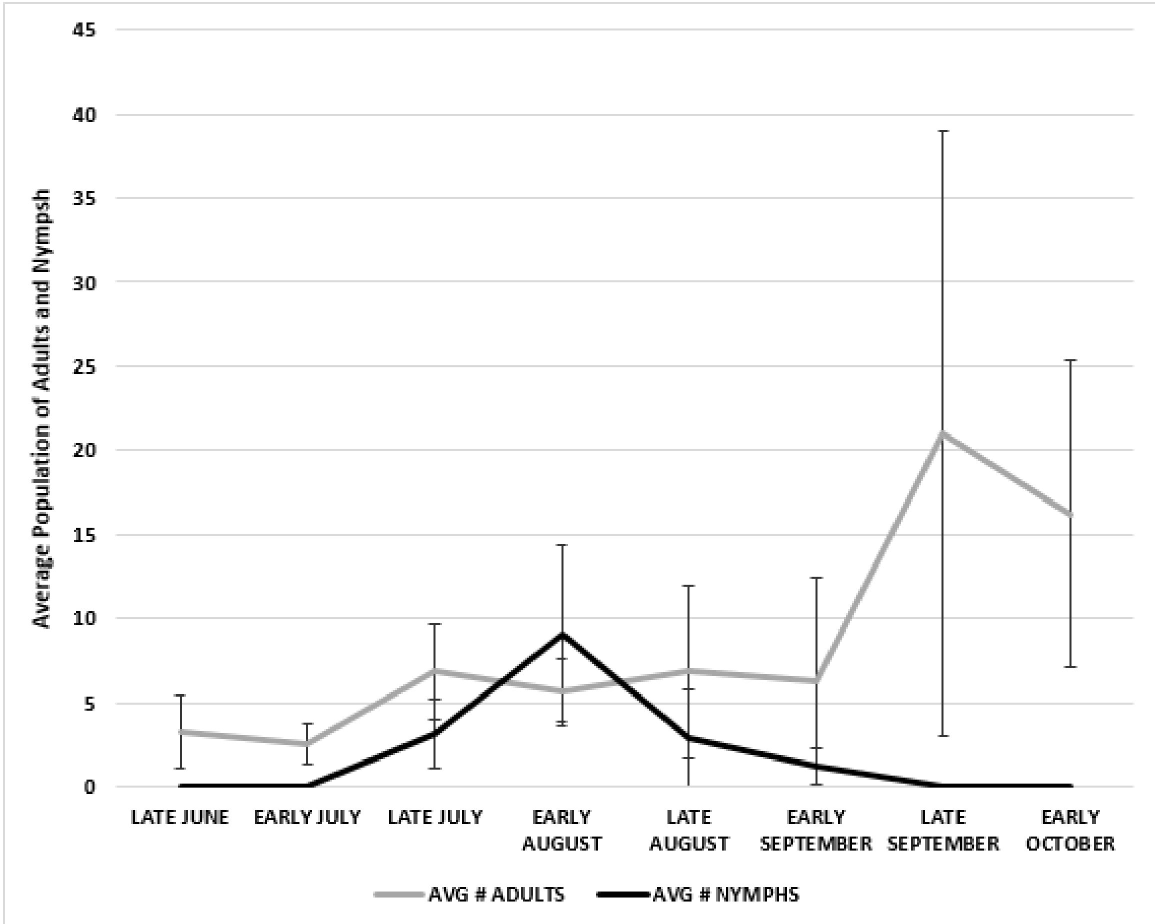

Figure 4.3 Kudzu Bug Population Densities in Soybean

Average population density of kudzu bug adults and nymphs (+SEM)/40 sweeps in soybean from late June through early October 2017 in Northeastern Mississippi.

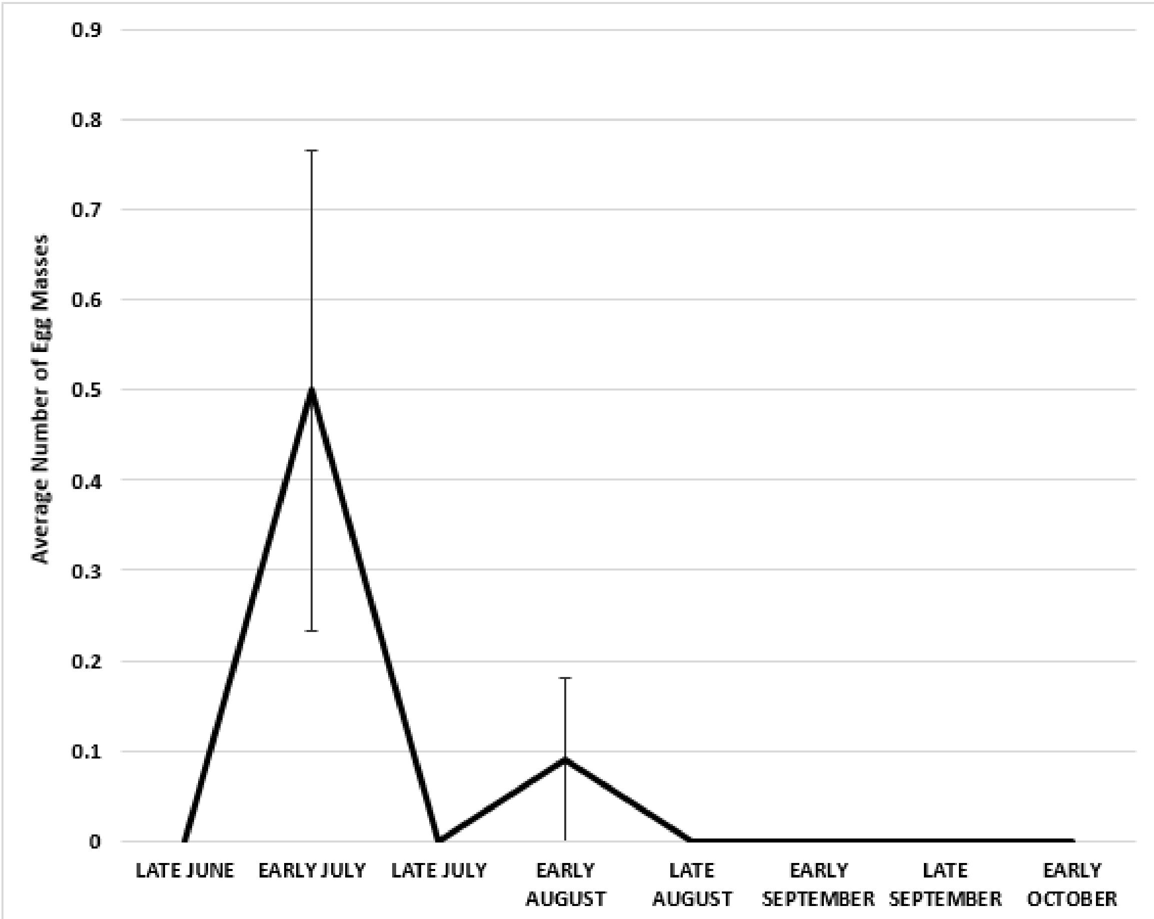

Figure 4.4 Number of Kudzu Bug Egg Masses in Soybean

Average number of kudzu bug eggs (+SEM)/50 soybean leaves from late June through early October 2017 in Northeastern Mississippi.

CHAPTER V

SUMMARY

The kudzu bug is a new pest of soybean in Mississippi. This insect is known to be able to reduce yield of reproductive stage soybean if left untreated. Little is known about the effects of kudzu bugs on soybean in Mid-South production systems. This gave rise for research to evaluate the effects of kudzu bug on vegetative stage soybean, evaluate control methods such as foliar insecticides and seed treatment, and evaluate population dynamics of kudzu bug in kudzu and soybean.

There was no impact on soybean yield from infestations of either VC or V4 stage soybean with densities up to 3 kudzu bugs/plant. Densities greater than this are expected to be rare events since kudzu bugs prefer reproductive stage soybean. All foliar insecticide applications provided adequate control of adult and nymph kudzu bugs for a short period. However, by six days after application no products provided much protection. Throughout the trial, clothianidin and acephate provided the highest mortality of all treatments on adult kudzu bug. Adult and nymph kudzu bugs can be easily controlled with common foliar insecticides. The neonicotinoid seed treatments currently used did not provide much control of adult kudzu bugs on VC, V1, or V2 soybean growth stage. Based on these findings, seed treatments are not recommended to control kudzu bugs in vegetative stage soybean. Based on a survey of kudzu bug adults, nymphs, and eggs, it is likely that there are two generations per year in Mississippi as previously reported in other regions. *Beauveria bassiana* was first observed infecting kudzu bugs in late August and was always present through the end of the survey. No eggs collected were parasitized, and no other natural enemies were observed.