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An evaluation of corn earworm damage and thresholds in soybean

By

Brian Patrick Adams

A Dissertation
Submitted to the Faculty of
Mississippi State University
in Partial Fulfillment of the Requirements
for the Degree of Doctor of Philosophy
in Life Sciences
in the Department of Biochemistry, Molecular Biology, Entomology and Plant Pathology

Mississippi State, Mississippi

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2015

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Interactions between corn earworm, *Helicoverpa zea* (Boddie), and soybean, *Glycine max* L. (Merrill), were investigated in the Mid-South to evaluate thresholds and damage levels. Field studies were conducted in both indeterminate and determinate modern cultivars to evaluate damage, critical injury levels, and soybean response to simulated corn earworm injury. Field studies were also conducted to evaluate the response of indeterminate cultivars to infestations of corn earworm. Field studies were also conducted to investigate the relationship between pyrethroid insecticide application and corn earworm oviposition in soybean.

Results of field studies involving simulated corn earworm damage indicated the need for a dynamic threshold that becomes more conservative as soybean phenology progressed through the reproductive growth stages. This suggested that soybean was more tolerant to fruit loss during the earlier reproductive stages and was able to compensate for fruit loss better during this time than at later growth stages.

Results of field studies involving infestations of corn earworm indicated that current thresholds are likely too liberal. This resulted in economic injury level tables

being constructed based upon a range of crop values and control costs, however, a general action threshold was also recommended for indeterminate soybean in the Mid-South.

Field study results investigating the relationship of pyrethroid application and corn earworm oviposition indicated that even in the presence of an insecticide, corn earworm prefers to oviposit in the upper portion of the canopy, as well as on the leaves as opposed to all other plant parts, consistent with all previous literature.

DEDICATION

I dedicate this research, the countless sleepless hours spent analyzing data and writing this dissertation to my wife, Julie, as well as to my son Hayes and our expected second child. This was for y'all. All of the time and effort spent to complete this degree was to help make a better life for my family, while allowing me to work in the agricultural industry, which I have long held dear to my heart.

I also dedicate this research to both of my grandmothers, Billie Spain and Emma Adams, as well as to the memory of my late grandfathers, Robert Spain and Bud Adams.

Lastly, I dedicate this research to my mother and father, Jerry and Lori Adams. Remember when I was little and told y'all I wanted to be a doctor? This may not be exactly what we all once thought, but guess what.....

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I'd like to take the opportunity to thank all of the major professors that I've worked under during my time here completing this degree, Angus Catchot and Don Cook. Their knowledge and guidance that imparted to me has been truly invaluable and I could not have completed this degree without their assistance. Additionally I'd like to thank Jeff Gore, currently a committee member on this dissertation, who previously served as the co-director of my thesis. His direction also proved to be invaluable while completing both degrees. Fred Musser served as a committee member for both degrees as well, and the assistance with data analysis and advice provided to me in our many office discussions will never be forgotten. Additionally, the wisdom on a variety of topics from my other two committee members, Bobby Golden and Trent Irby, have been very influential on many decisions made through the course of completing my degree. I'd also like to thank the Mississippi Soybean Promotion Board for their funding of this project. Lastly, but certainly not least I'd like to thank all of my fellow graduate students: Adam Whalen, John North, Taylor Dill, Jeff Ramsey, Nick Bateman, Scott Graham, and Andrew Adams as well as the many student workers who helped me to complete all of the labor that was required to complete this research.

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CHAPTER I

INTRODUCTION

Soybeans

Soybean, (*Glycine max*) [L.] Merrill, is one of the oldest cultivated crops in the world. The first record of domestication of soybean is by Chinese farmers around 1100 BC, and soybeans were grown in other countries by the first century AD (North Carolina Soybean Producers Association 2011). The first listed record of soybean cultivation in the United States was in 1765 by a colonist in Georgia (North Carolina Soybean Producers Association 2011). Soybeans were produced for a variety of purposes prior to the 20th century including as a forage for livestock as well as feeding of grain to livestock. At the beginning of the 20th century, George Washington Carver discovered the utility of soybeans as an oil crop and a good source of protein (North Carolina Soybean Producers Association 2011). Soybean production in China was halted during World War I and as a result the soybean production industry in the United States exploded during this period. The demand for oil products, lubricants and other products increased as a result of the war and the demand was met in large part through soybean production (North Carolina Soybean Producers Association 2011).

Soybean production in the U.S. has continued to rise since the 1940's as a result of the development of innovative uses of soybean oil and meal. During 2013, 31.2 million ha of soybean were planted in the U.S. (NASS 2014c). The U.S. produced 82.2

billion kg of soybean in 2012 (NASS 2014c). The Midwestern states of Iowa, Illinois, Indiana, and Minnesota are typically the largest producers of soybeans. However, production in the Midsouthern states is increasing (NASS 2014c).

Soybean is a photoperiod sensitive plant and flowering is triggered by declining day length. Soybeans are often divided into maturity groups from 00 to VIII (Hartwig 1973). These different maturity groups are separated by the amount of daylight required to begin flowering. Soybeans can possess either a determinate growth habit, where all vegetative growth ceases at the initiation of flowering or indeterminate growth habit where vegetative growth continues after the initiation of the flowering period (Kogan and Turnipseed 1980). The most commonly grown maturity groups in Mississippi are the Groups IV and V (Heatherly et al. 1999).

Soybean planting in Mississippi typically occurs from March to June (Heatherly et al. 1999). The March plantings occur on a limited basis and soybean planted at the end of May and into June are typical of a double crop system that is planted in to harvested wheat stubble. Soybean in Mississippi is grown under multiple agronomic practices (Heatherly et al. 1999). Row spacing varies from as narrow as 0.19 m rows up to as wide as 1.02 m row spacing with seeding populations generally ranging from 247,100-494,200 seed per hectare (Heatherly et al. 1999). Generally, as row spacing increases, seeding population decreases (Heatherly et al. 1999). In 2007, Mississippi yields averaged between 2,353-3,026 kg/ha (NASS 2014c). Those statistics can be misleading as yields in the Delta region for full season irrigated production often average greater than 4,700 kg/ha, and in places will exceed 6,000 kg/ha.

The method describing soybean growth stages by Fehr and Caviness (1977) was the first system for use with both determinate and indeterminate varieties. This method was slightly modified by Pedersen (2004) and is still the most commonly used system to describe soybean growth stages today. This system first defined the vegetative and reproductive stages separately. Soybean usually emerges 5-10 days after planting and is designated as VE, or emergence stage. During emergence the cotyledons begin to unroll and once unrolled the plant puts on unifoliate leaves at which the growth stage is termed VC or cotyledon stage (Pedersen 2004). From the unifoliate stage, soybean begins to put on trifoliate leaves and the subsequent remaining stages of vegetative growth are designated by the number of trifoliate leaves to the n^{th} degree, V^n . Fehr and Caviness (1977) define eight reproductive stages, R1-R8 and Pedersen (2004) made slight alterations to some stages. All reproductive stages, 1-8, will hereafter be referred to as R stages denoted by their corresponding number. Once the days have reached their proper length, growth becomes reproductive and soybean puts on an open flower designated as R1 (Pedersen 2004). Once soybean reaches R1, it continues to put on flowers at all nodes and once there is an open flower at the two uppermost nodes, it is defined as the R2 growth stage (Pedersen 2004). From R2, plants begin developing small 0.95 cm (3/16") pods where flowers were pollinated and once plants develop one 0.95 cm pod in the uppermost four nodes, it is termed R3 and is considered the beginning of pod setting (Pedersen 2004). At this stage pods continue to develop and grow in length and when one pod in the uppermost four nodes reaches 1.9 cm (3/4") in length it is designated as R4 and is considered the ending of pod setting (Pedersen 2004). Once pod setting is completed, pods begin to increase to their full size and seed fill begins. Once seed inside

Pods in the uppermost four nodes of the plant reach 0.3 cm (1/8") it is considered R5 (Pedersen 2004). From R5, seeds continue to grow in size within the pod cavities. R6 is the final stage of pod fill and defined by having a pod in the upper four nodes containing green seed that occupies the full pod cavity (Pedersen 2004). Once seed fill is completed the plants begin to reach physiological maturity. R7 is defined as having one mature brown pod present anywhere on the plant and is considered the beginning of maturity. The remainder of the pods begin to mature and reach their brown color beyond R7. Once 95% of all pods on the plant have reached their mature color, plants are considered to be at the R8 growth stage which is defined as full physiological maturity. As with most grain crops, moisture levels within the pods begin to drop as senescence progresses. Once R8 is reached, the soybeans are ready to be harvested. Most harvesting should take place when seed moisture is 13-15% unless drying aids will be used.

Corn Earworm

The corn earworm, *Helicoverpa zea* (Boddie), is a polyphagous insect with a broad host range that includes many food, fiber, oil, and horticultural crops as well as many uncultivated wild hosts (Fitt 1989). This insect species occurs in North and South America primarily between latitudes 40°N and 40°S. Corn earworm overwinters in the soil as a diapausing pupa. Winter survival is greatly reduced when temperatures decline below 0°C. Typical of other Noctuidae, once pupation from overwintering populations is complete, adults emerge and persist for approximately two weeks. During this time, females typically lay between 500-2000 eggs. Eggs hatch in approximately four days at 25°C. Corn earworm larvae feed primarily on fruiting structures. This habit makes it a direct pest of crops in which the fruiting structures are the marketable portion. The

preference for the fruiting structures of the plant is due to larval feeding preference for plant parts high in nitrogen (Hardwick 1965). In crops where corn earworm is a direct economic pest, economic thresholds tend to be low.

Fitt (1989) states that the polyphagy, high mobility, high fecundity, and facultative diapause of *Heliothis* spp. allow them to thrive in otherwise unsuitable ecosystems and exploit these crops. Corn earworm can be found feeding on many different plant families including but not limited to Asteraceae, Fabaceae, Leguminaceae, Malvaceae, Poaceae, and Solanaceae (Fitt 1989). Fitt (1989) states that the importance of polyphagy is threefold: that populations may develop at the same time on a variety of hosts in a given area, that populations can continuously develop under proper conditions on a variety of both cultivated and wild hosts through the year, and that populations possess the ability to persist at low levels in unsuitable areas due to the female's ability to locate a host that can sustain larval development. It is essential for corn earworm to take on long distance, local, and regional travel as part of its success as a pest. When dealing with the temporal and spatial diversity of cultivated and uncultivated hosts, especially those with a short period of suitability, the need for mobility both locally and regionally becomes critical in order to locate suitable hosts (Fitt 1989). Corn earworm and other Noctuids have two methods of dealing with the seasonality and senescence of their hosts, diapause through adverse climate conditions or migration to achieve spatial redistribution (Farrow and Daly 1987). Fitt (1989) states that unlike some Noctuids that are obligate migrators, *Heliothis* spp. are facultative migrators due to factors such as weather systems, mating, and uncondusive local conditions. He further states that the type of migration occurring depends largely upon the climate on the night of migration.

The ability of *Helicoverpa zea* to enter facultative diapause during the pupal stage is the third key trait for the success of the species. Fitt (1989) notes that winter is passed in diapause in subtropical and temperate regions of its range, but further notes that populations in these regions are often reestablished or supplemented by regular migration during warmer months. This diapause is critical to maintaining local populations when suitable hosts aren't available in the immediate area.

High fecundity is the final key element contributing to the success of corn earworm. This characteristic gives corn earworm the ability to quickly increase population size when coupled with a relatively short generation time (Fitt 1989). Quaintance and Brues (1905) reported in a laboratory study that a single female laid up to 3000 eggs in her lifetime. Female *H. zea* moths, however, have an average reproductive lifetime of 8-10 days and during this time may lay between 1000-1500 eggs per female singly as opposed to in masses (Fye and McAda 1972). Fecundity is most directly affected by climatic factors such as temperature and humidity, as well as nutrition during both larval and adult stages (Isely 1935). Prolonged high temperature appears to have the greatest impact on corn earworm. Exposure to temperatures above 35°C for prolonged periods significantly decrease not only fecundity, but fertility and survival of adults as well (Fye and McAda 1972).

Many pest problems in agriculture can be attributed to lack of host diversity in current agriculture systems. Large monocultures are a standard practice due to the efficiency and economics of planting and harvesting. In many cases it has been proven that increasing regional and in field host diversity could decrease pest populations. In the

case of corn earworm, studies have actually shown that corn earworm thrives in more diversified areas (Stinner et al. 1982).

In Mississippi, corn earworm typically undergoes 4-5 generations per year. There is a very evident host progression for each of these generations of corn earworm in Mississippi as well as other locations. In Mississippi, the first generation occurs on wild hosts such as crimson clover, *Trifolium incarnatum* L., in the Northeast portion of the state (Snow and Brazzel 1965). Stadelbacher (1981) reported crimson clover to be the primary host of first generation corn earworm larvae, but also noted that cut-leaved cranesbill, *Geranium dissectum* L. was an important host for first generation larvae in the Delta (Stadelbacher 1979). The second generation larvae occur primarily in corn, *Zea mays* L., but also in other wild hosts such as velvetleaf, *Abutilon theophrasti* Medik, in the Delta and Carolina geranium, *Geranium carolinianum* L., in the Northeast (Snow and Brazzel 1965, Stadelbacher 1981). The third and fourth generations occur primarily on cotton, *Gossypium hirsutum* L., and soybean after corn has naturally senesced with the fifth generation occurring primarily on wild hosts and volunteer corn (Hartstack et al. 1973).

In Mississippi, the three largest agronomic crops, corn, cotton, and soybean are all attacked routinely every year. Johnson et al. (1986) estimate that the corn earworm and tobacco budworm, *Heliothis virescens* [F] combine to cause over one billion dollars in losses annually on all infested crops in the United States. From 2000-2007 Mississippi planted between 445,000 and 650,000 hectares of cotton annually. Cotton production fell to 242,000 hectares in 2007 and all the way to 117,000 hectares in 2013 (NASS 2007). Mississippi producers planted between 450,000 and 690,000 hectares of soybeans

annually from 2000-2007, however from 2008-2013 soybean production ranged between 708,000 and 875,000 annually (NASS 2007). Reduced cotton production can be attributed to a combination of a decrease in commodity prices coupled with increases in input costs associated with production. During this time, corn and soybean commodity prices have generally increased, resulting in increased corn and soybean production.

Due to the shift in production, there's been an increase in soybean infested by and treated for corn earworm. This increase is multifaceted in the sense that not only is there a lack of cotton and an abundance of soybean for these middle and late generations, but also due to the increase in corn there are higher populations in the landscape. Prior to 2011, the stink bug complex and soybean looper, *Chrysodeixis includens* Walker, were managed as the primary pests of soybean. However with the continued decrease in cotton production, the corn earworm has become the primary pest managed in soybean in Mississippi in recent years (Musser and Catchot 2008, Musser et al. 2010, Musser et al. 2012, Musser et al. 2013). In 2007, cost of control plus loss for corn earworm only totaled \$637,000 compared to stink bugs the same year which totaled over \$28,000,000 (Musser and Catchot 2008). By 2009 there had been a major increase in cost plus loss for corn earworm to over \$17,600,000 while stink bug still totaled in excess of \$29,000,000 (Musser et al. 2010). By 2011 corn earworm cost plus loss totals had increased to a level exceeding that of stink bugs, greater than \$41,000,000 that year for corn earworm compared to just over \$5,000,000 for stink bug, making corn earworm the primary pest of soybean in Mississippi (Musser et al. 2012). Results were similar in 2012, corn earworm again was the most damaging pest of soybean in Mississippi with a total cost plus loss of

greater than \$34,000,000 compared to stink bugs just greater than \$10,000,000 (Musser et al. 2013).

Economic Injury Levels and Thresholds

Integrated pest management (IPM) is a strategy for managing pests that incorporates all types of control measures including physical, cultural, and chemical control. IPM strives to manage pest populations using the most economical and environmentally practical methods. Two key principles of IPM are economic injury levels and associated economic thresholds. An economic injury level is the lowest density of a pest that will cause economic damage (Stern et al. 1959). An economic threshold is the population density at which control measures should be taken to prevent an increasing pest population from reaching the economic injury level (Stern et al. 1959). Many mathematical models have been proposed to calculate economic injury levels, the one determined by Pedigo et al. (1986) is the one most commonly used today. In this model it is determined that $\text{economic injury level} = C/VIDK$. In his model, C= cost of the management tactic per unit of production, V= current market value per unit of the marketable crop, I= injury unit per insect per production unit, D= damage per unit injury, and K= proportionate reduction of the insect population by the management tactic.

One major goal of IPM is to practice control measures which would maintain pest populations below economic injury level to minimize the need for remedial control practices such as insecticide treatments (Eckel 1991). Knowledge of the particular ecosystem and pest biology are necessary for implementation of cultural and biological control practices in accordance with IPM goals. Eckel (1991) notes that though management of soybean pests have been reviewed by many different authors, caution

should be taken before trying to apply control strategies simply because they were successful in other regions due to differences in regional agroecosystems. He further states that results shouldn't be ignored from other regions as production systems still share some common practices.

Smith and Bass (1972b) reported variable impacts on soybean yield with various levels of corn earworm when infestations occurring at approximately R4 in cage studies. In one year, the highest infestation density (4 larvae/ 0.3 row-m) did not significantly impact yield. However, in the second year of the study, infestations of three or five larvae per row-m reduced yields and concluded the economic threshold fell between one and three corn earworm per row-m based on control costs and commodity prices at that time.

Studies using field cages conducted in Arkansas reported the effects of infestation level of corn earworm larvae on yield and damage to soybean at various growth stages (Mueller and Engroff 1980). Degree of damage depended upon the soybean growth stage at which the corn earworm infestation occurred. Also, smaller larvae, <3rd instar fed primarily on plant pubescence while third instar and older larvae fed on additional plant parts. All larvae exhibited a preference for blooms if present. However, no consistent yield reductions were observed at infestation levels up to 65 larvae per row meter at different growth stages. Consistent with other studies, these studies indicated that compensation was responsible for the lack of response in yield. As a result of these studies, the economic threshold for corn earworm in soybean was increased from 6.5 larvae per row meter to 10 larvae per row meter in Arkansas. The authors noted that this value was lower than it should be.

In Mississippi, egg infestation studies were conducted to evaluate the impact of tobacco budworm, *Heliothis virescens*, infestations on soybean yield (McWilliams 1984). Maturity group VII determinate 'Bragg' and maturity group VI 'Lee-74' soybeans were the varietal selections. Tobacco budworm eggs were applied as a spray to plots at rates of 0, 50, 100, and 200 eggs per 0.3 m of row at various stages of soybean growth. In 1977, egg infestations at the R2 stage significantly increased the number of damaged seed compared to the non-infested control and reduced the number of seed harvested per plot. However, 100 seed wt. was significantly higher in the infested plots and subsequently no differences in yield were observed. Similar to 1977 results, significant increases in seed weight were observed with increasing levels of damage in 1978. The only egg infestation timing that resulted in reduced yields was R2. Larvae resulting from egg infestations at this growth stage had developed to 3rd to 4th instars by R3 and 5th or 6th instar by R4. 5.8 larvae per 0.3 row meter caused a significant yield loss in 'Lee-74' while it took 11 larvae per 0.3 row meter to reduce yield in 'Bragg' when 5th or 6th instar larvae were present at R4.

Field plots were established in Virginia to determine the effects of corn earworm infestation level on soybean yield (McPherson and Moss 1989). In 1985, population densities of 4.8 corn earworm per 0.915 meter of row did not significantly reduce yields when compared to the non-infested control. In 1986, corn earworm populations peaked at 26.7 larvae per 0.915 meter of row. In both years of the study, significant yield losses were not observed for any infestation level due to an increase in weight of undamaged seed.

Due to the nature of the older cultivars and late maturity groups in these studies, evaluations with newer cultivars with shorter maturity levels need to be made. Results of all previous studies varied somewhat, however they all shared one commonality in that soybeans have a prolific ability to compensate given proper environmental conditions.

Simulated Corn Earworm Damage

Many studies of simulated corn earworm damage or pod removal have been conducted using various methods. Most studies utilized hand removal of whole pods (McAlister and Krober 1958, Hicks and Pendleton 1969, Smith and Bass 1972a, McPherson and Moss 1989) regardless of growth stage opposed to puncturing pods and seed removal (Kincade et al. 1971). Soybean, similar to other crops such as cotton produce a much larger number of flowers than mature fruit under normal conditions. Swen (1933) estimated between 30 and 85% of all floral buds or flowers produced on a soybean plant naturally abort and shed. Approximately 20% of total pod shedding occurs during the R1 growth stage, while 75% occurs during R2. Control of certain biotic factors, such as the natural infestations of many pest species in addition to the unreliability of natural corn earworm infestations, is the primary reason for using simulated damage techniques to evaluate corn earworm damage (McPherson and Moss 1989).

In a study by McAlister and Krober (1958), hand removal of 10% of pods did not significantly reduce numbers of mature pods present at harvest compared to the non-damaged control, while 20% pod removal did significantly reduce numbers of mature pods. The authors noted that the most probable cause for the apparent recovery in pods in the treatment is that the normal abortion of pods in the non-damaged plots was not

present in the treatments due to the mechanical removal of pods. McAlister and Krober also noted that there were no significant reductions in yield in that study until 80% of pods were removed. The study also found that seed size increased enough to compensate for fewer pods present at 30 and 40% pod removal levels.

Pods were damaged with a leaf cutter at levels of 0, 10, 20, 30, and 50% at late R4 to early R5 to simulate corn earworm feeding in a study in Mississippi performed by Kincade et al. (1971). The authors observed no significant difference in yield or weight per hundred seed in any of the injured plots compared to the untreated control during 1968. In 1969 however, yield was significantly reduced from the untreated control in the 30% and 50% damage plots though weights per hundred seed were not significantly different in any of the injured plots compared to the untreated control. It was further noted that abundant moisture was present in 1968 and was not during 1969, suggesting that late season moisture deficiency reduced yield and ultimately lowered the economic threshold for corn earworm in 1969 compared to 1968 and that the plants ability to compensate for damage is heavily dependent on proper environmental conditions.

Smith and Bass (1972a) performed a study simulating corn earworm damage by hand removing pods at levels of 10, 20, 30, 40, and 80% at various growth stages. No significant yield losses were observed until removal levels of 80% were performed prior to pod filling, however, once pod filling began, removal levels as low as 10% caused significant yield reductions.

In a study by McPherson and Moss (1989), 100% of pods and blooms if present were removed on August 26, 1986 for six different planting dates ranging from mid-May to early June in field plots in Virginia in 1986. The growth stage at the time of

depodding for the first planting date was late R4 and depodding caused a significant yield reduction from the untreated control. Both the second and third planting dates were mid R4 at the time of treatment and depodding caused a significant yield loss in both planting dates compared to the untreated control. The final three planting dates ranged from R2 to early R4 and there were no significant yield reductions due to depodding for any of the final planting dates. Depodding resulted in a significant increase in seed size for the first planting date compared to the untreated control as well as a significant increase in number of one bean pods present per plot and a significant decrease in numbers of two bean pods present per plot. The observed yield differences in the first three planting dates appeared to be a result of significantly less total seeds per plant in the depodded treatments that didn't appear in the final three planting dates. Six planting dates were used in Virginia in 1987 ranging from mid-May to late June. Depodding resulted in no significant yield differences in any planting date in 1987 in Virginia. In 1987, three planting dates in Georgia were also tested by removing all pods and blooms at R4. Significant yield reductions were observed at the last two planting dates in this study which were late R4 to R5. The authors also noted that in 1986, plants were under drought stress in Virginia before treatments were initiated through harvest, however there was adequate rainfall throughout the year in 1987, indicating again that the presence of moisture heavily influences soybean's ability to compensate for pod loss and that phenological growth stage of soybean at the time of injury also has major impacts on whether or not the plant has the ability to compensate.

Many factors have changed since the last time economic injury levels or economic thresholds for corn earworm in soybean were investigated. The effect of major

changes in production practices, such as shifting from later plantings and determinate varieties to earlier plantings and indeterminate soybeans could potentially have significant impact on previous findings. Economic injury levels and thresholds for corn earworm need to be investigated. Also, the amount of damage caused at various growth stages and the associated ability of the plants to compensate need to be investigated as well.

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CHAPTER II
THE IMPACT OF PYRETHROID APPLICATION ON CORN EARWORM
OVIPOSITION IN SOYBEAN

Introduction

The corn earworm, *Helicoverpa zea* (Boddie), is currently the most economically important insect pest of soybean, *Glycine max* [L.] Merrill, in Mississippi (Musser et al. 2013). In Mississippi, corn earworm typically undergoes five generations per year with the third and fourth generations occurring primarily in soybean and cotton, *Gossypium hirsutum* L. Previous research indicates that corn earworm has an ovipositional preference for soybean over cotton; however, corn, *Zea mays* L., was the most preferred host for oviposition (Johnson et al. 1975). In contrast, Hillhouse and Pitre (1976) found that corn earworm preferred cotton over soybean as an ovipositional host at most phenological stages of crop development. Corn earworm also has an ovipositional preference for soybean leaves compared to stems and pods (Hillhouse and Pitre 1976, Terry et al. 1987). Both studies used later maturing maturity group VI or VII varieties with determinate growth habits. Most cultivars used in current soybean production systems in Mississippi are maturity group IV and V and possess an indeterminate growth habit.

One common practice in current soybean production systems in Mississippi is the prophylactic application of a fungicide when soybeans reach approximately the R2 or R3

stages of growth as defined by Fehr and Caviness (1977) as a preventative measure against many common fungal diseases in Mississippi. In recent years, producers in Mississippi have often added a pyrethroid insecticide to accompany the fungicide application. This application of a pyrethroid often occurs even in the absence of threshold levels of any insect pest at the time of application and is not recommended. Due to this addition of broad spectrum pyrethroids, many beneficial insect species are eliminated from the field, which allows for corn earworm populations to establish very easily. One aspect of corn earworm oviposition in soybean that has not been investigated is what effect a pyrethroid insecticide may have on oviposition. Because pyrethroids can act as repellants, they may change the behavior of ovipositing corn earworm where more eggs are laid in the lower portions of the canopy to avoid insecticide residues in the upper portion of the canopy. The current experiment investigates if pyrethroid application has any effect on the ovipositional behavior of corn earworm in soybean.

Materials and Methods

Studies were conducted to examine the influence of pyrethroid insecticide applications on the ovipositional preference (plant structure and location within the canopy) of corn earworm. These studies were conducted at the Delta Research and Extension Center in Stoneville, MS and at the R.R. Foil Plant and Soil Sciences Research Farm in Starkville, MS during 2012-2013. The study was a randomized complete block design with four replications. At each location, a uniform planting of a maturity group IV indeterminate soybean variety (AG4605) was planted at a rate of 296,532 seed/ha into raised conventional till beds with a 0.96 m row spacing in Starkville and a 1.02 m row spacing in Stoneville and managed in accordance with Mississippi State University

Extension Service recommendations until the R1 stage of soybean development. Seed were treated with a commercial premix of imidacloprid, pyraclostrobin, metalaxyl, and fluxapyroxad (Acceleron®, Monsanto Company, St. Louis, MO) to minimize the impact of early season insect pests and seedling disease. Weeds and diseases were managed according to Mississippi State University Extension Service recommendations. At R1, eight 1.8 m x 1.8 m x 1.8 m field cages were erected at each location over the soybeans. Two to three days prior to infestation of moths at R2, one cage in each replication received an application of 1.12 kg ai/ha of methyl-parathion while the other cage received the same rate of methyl parathion plus a 0.018 kg ai/ha application of beta-cyfluthrin which served as our treatments in this study. Methyl parathion was applied to both cages to eliminate egg predators and parasitoids. At R2, approximately two to three days after application of insecticides, each cage was infested with ten pairs of newly emerged male and females moths reared in the laboratory. Insects used in this study were no more than two generations removed from a wild collected parent colony. Adults were allowed to mate and oviposit unhindered for a period of five days. Immediately after the five day ovipositional period, twenty randomly selected plants from each cage were removed and taken to the laboratory where they were examined for eggs. In Stoneville, cages were infested on August 11th, 2012 and July 24th, 2013. In Starkville, cages were infested on August 10th, 2012 and July 25th, 2013. Plants were partitioned vertically by mainstem node into three sections: top third, middle third, and bottom third of the plant. Each section was examined for eggs and the number of eggs were recorded by plant structure (leaves, stems, or blooms and small pods) within each section. Data were analyzed using mixed model analysis of variance in Proc Mixed in SAS (Littell et al. 2006). Subsamples

were averaged prior to analysis. In the model insecticide treatment, plant section, plant part (leaves, stems, pods) nested within plant section, and the corresponding interactions were designated as fixed effects. Year, location, and replications nested within years by locations were designated as random effects (Blouin et al. 2011) to allow inferences to be made over a wide range of environments (Carmer et al. 1989, Blouin et al. 2011). Degrees of freedom were calculated using the Kenward-Roger method. Differences were considered significant for $\alpha=0.05$.

Results and Discussion

Neither the interaction between treatment and plant part (part) nested within plant section (section) ($F= 0.57$; $df=6, 255$; $P=0.75$) nor the interaction between treatment and section ($F= 0.85$; $df=2, 255$; $P=0.43$) had a significant impact on the number of eggs laid by *H. zea* moths. Neither treatment ($F= 0.02$; $df=1, 255$; $P=0.9$) nor section ($F= 1.13$; $df=2, 255$; $P=0.07$) had a significant impact on the amount of eggs laid by corn earworm moths. Only part nested within section ($F= 13.68$; $df=6, 255$; $P\leq 0.01$) had a significant impact on eggs laid by corn earworm moths. Corn earworm moths preferred to oviposit on leaves as opposed to all other plant parts and they also preferred to oviposit in the upper portions of the canopy compared to lower portions. There were significantly more eggs laid on leaves within each section than all other part and section combinations, and there were significantly more eggs laid on the leaves in the upper portion of the canopy than on leaves in the lower portion of the canopy (Table 2.1).

One hypothesis of this study was that corn earworm moths would oviposit lower in the canopy in pyrethroid treated plots to avoid contact with pyrethroid residue. This was not the case. Regardless of treatment, results of this study were similar to Hillhouse

and Pitre (1976). Hillhouse and Pitre examined the ovipositional preferences of *H. zea* and *Heliothis virescens* between cotton and soybean of similar sizes and growth characteristics in 1.8 x 3.6 x 1.8m field cages at various stages of crop phenology and noted which portion of the canopy as well as plant structure on which eggs were laid. In their study, both *H. zea* and *H. virescens* laid significantly more eggs on leaves than on stems or floral/fruitlet structures of R2 soybeans. They also observed that eggs for both species were concentrated in the upper 2/3 of the plant canopy. In a separate component of the same study, Hillhouse and Pitre (1976) made the same observations on plants maintained in a 3.05 x 6.1 x 3.05m greenhouse. In contrast to the aforementioned component of the study, *H. zea* exhibited a preference for the fruitlet structure of soybean in the greenhouse study.

Our studies were consistent with that of Hillhouse and Pitre (1976), and demonstrated in the small field cages that corn earworm moths exhibited a preference to oviposit in the upper portions of the canopy and on leaves. The plants in our study ranged from 76-115 cm in height and had achieved canopy closure at all locations. One reason that corn earworm may have shown ovipositional preferences for upper portions of the canopy as well as leaves could be that the plant architecture was packed tightly and the two rows within our cages were pulled toward one another by the screens on the cage making penetration into the bottom of the canopy difficult. Also in this situation, petioles of these plants in our cages were standing more erectly than they would be without the cage, making the main stems and fruit/floral structures more difficult to reach than the leaves. Based upon this, caution should be used when extrapolating results of this study to production fields as there appears to be a spatial effect when comparing our results in

field cages as well as the results of Hillhouse and Pitre (1976) in field cages to their greenhouse experiments. To better extrapolate results of an experiment like this to field environments, the study needs to be repeated in larger cages that would give a more accurate representation of spatial distribution of plants within production fields. Additionally, in larger cages, the ability to design the test as a split sprayed/unsprayed study within the same cage could provide further insight into the effects of pyrethroid acting as a repellent with respect to *H. zea* oviposition.

Table 2.1 Mean number of eggs laid on each plant part in each canopy section per cage.

Part	Section	Mean	SEM
Leaves	Top	1.15 a	0.3
	Middle	0.92 ab	0.3
	Bottom	0.68 b	0.2
Stems	Top	0.07 c	0.1
	Middle	0.12 c	0.1
	Bottom	0.04 c	0.04
Blooms	Top	0.003 c	0.003
	Middle	0.02 c	0.02
	Bottom	0.0 c	0.0

Means within a column followed by the same letter are not significantly different according to Fisher's Protected LSD test ($\alpha=0.05$).

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CHAPTER III
IMPACT OF SIMULATED CORN EARWORM DAMAGE ON INDETERMINATE
SOYBEAN

Abstract

Field experiments were conducted in Starkville and Stoneville, Mississippi during 2012 and 2013 to evaluate fruit removal level and timing on soybean growth, crop maturity, and yield. Fruit removal treatments consisted of 0, 50, and 100% of all fruit removed at specified growth stages (R2, R3, R4, and R5.5). Plant heights were determined at least bi-weekly from the time damage was imposed until R7. The impact of fruit removal level and timing on crop maturity was determined by estimating the percentage of naturally abscised leaves at 137 DAP when control plots were approximately 10-15 days from harvest and the percentage of non-senesced main stems at 139 DAP. There was no significant impact of fruit removal timing or fruit removal level on plant height or canopy width. Significant delays in crop maturity were observed when fruit removal was imposed at the R5.5 growth stage. Significant reductions in yield and crop value were observed as early as R3 and R4 when 100% of fruit was removed. Both fruit removal levels at R5.5 resulted in a significant reduction in yield and crop value compared to the non-treated control. Indeterminate soybeans appear to have the ability to compensate for some fruit loss during the early to middle reproductive growth stages without delaying maturity. However, severe fruit loss causes increasingly more yield loss

as the plant approached maturity. Thresholds and economic injury levels therefore need to be adjusted accordingly to account for the dynamic nature of yield losses and crop maturity delays.

Introduction

Soybean, *Glycine max* (Merrill), production in Mississippi has increased from 469,435 ha during 2001 to 898,402 ha during 2014 (NASS 2014b). The area planted to cotton, *Gossypium hirsutum* (L.), has decreased from 687,965 ha in 2001 to 116,144 ha in 2014 (NASS 2014b). The reasons for this shift include more favorable commodity prices for soybean compared to cotton and other crops as well as lower production costs for soybean compared to cotton. Costs associated with insect management account for some of the differences between cotton and soybean input costs. Generally soybean requires less intensive insect management compared to cotton (Musser et al. 2013, Williams 2014). Transgenic insect control traits, such as those expressing *Bacillus thuringiensis* proteins, and the associated technology fees are not available in soybean in the U.S. at this time. While seed and associated technology costs are lower with soybean, other production costs such as weed control and seed treatments are comparable between the two crops. In recent years, lower input costs and higher commodity prices have made soybean production more profitable and less risky than an alternative crop such as cotton in the Mid-South.

During the last 20-25 years, Mississippi soybean yields have been greatly improved (NASS 2014b). Two reasons for this improvement include superior genetics as well as the adoption of the early soybean production system in Mississippi (Heatherly 1999). During this period, Mississippi has transitioned from later maturing determinate

varieties to earlier maturing indeterminate varieties (Heatherly 1999). Approximately 70% of the soybean production in Mississippi currently utilizes earlier maturing indeterminate varieties (Irby 2014).

Corn earworm, *Helicoverpa zea* (Boddie), has become the most important insect pest of soybean in Mississippi (Musser et al. 2013). As recently as 2007, the associated costs plus losses for corn earworm in soybean were slightly above \$1,500,000 across the entire statewide crop, while the cost for one insecticide application was only \$17.91 per hectare (Musser and Catchot 2008). These costs have risen dramatically and in 2012 Mississippi growers averaged approximately 2.97 applications per hectare targeting corn earworm, with a cost of \$37.07 per hectare (Musser et al. 2013). Cost of control plus the losses incurred from corn earworm infestations in 2012 were greater than \$34,000,000 across the entire soybean crop in Mississippi. The increase in control cost is a result of a shift from the use of less expensive pyrethroid insecticides for corn earworm control to the diamide insecticides due to less than satisfactory control with pyrethroids (Catchot et al. 2014). The reason for larger and more widespread corn earworm infestations in soybean may be related to an increase in corn production (NASS 2014c). Corn serves as the primary second generation host for corn earworm in the Mid-South, while soybean and cotton serve as primary hosts for the third and fourth generation larvae (Jackson et al. 2008). This increase in corn, the preferred host for corn earworm (Jackson et al. 2008), likely causes larger populations to develop in the landscape before infesting soybean and cotton.

Historically in Mississippi, corn earworm is only a pest of reproductive soybeans. Corn earworm commonly infests soybean during the flowering stage (R1-R2) where it

primarily feeds on floral buds. Leaf feeding does occur, but this injury is minor and not considered economically important. Oviposition by moths at this time can result in large larvae being present between R4 and R6. In recent years however, large quantities of production hectares have been experiencing levels of corn earworm in excess of published thresholds from R2-R6. As crop phenology progresses, corn earworm begins feeding on pods and seed within the pods. It is common for smaller, immature pods to completely abscise when fed upon. When corn earworms feed on larger, fully elongated pods, it is not as common for them to abscise, however, they are often plagued with seed rots from various causal agents and contribute minimally to yield. Current thresholds for corn earworm in Mississippi are static throughout the reproductive stages at nine corn earworm larvae per 25 sweeps with a standard 38cm sweep net or three larvae per 0.3m of row with only larvae 1.27cm in size or greater counted (Catchot et al. 2014).

The majority of research on the impact of corn earworm infestations and fruit loss on soybean has been conducted using later maturing determinate varieties (McAlister and Krober 1958, Hicks and Pendleton 1969, Kincade et al. 1971, Smith and Bass 1972a, McPherson and Moss 1989). The objectives of this study were to examine the impact of defined levels of fruit loss to mimic corn earworm feeding at different growth stages on maturity and yield of indeterminate soybean as well as identifying a defined range for a plant damage based economic injury level using manual removal of fruit.

Materials and Methods

To determine the effect of fruit loss, experiments were performed in Starkville, MS at the R.R. Foil Plant and Soil Sciences Farm and in Stoneville, MS at the Delta Research and Extension Center during 2012 and 2013. Soybeans were planted in

Stoneville, MS on April 13, 2012 and April 17, 2013. Soybeans were planted in Starkville, MS on April 27, 2012 and April 17, 2013. The experiments were conducted using an indeterminate maturity group (MG) IV variety (Asgrow 4605[®], Monsanto Company, St. Louis, Mo). Micro plots (2 rows by 1.5 m) were established in the center of larger two row by 6.1 m plots to eliminate edge effects. Soybeans were planted at 296,532 seeds/ha into raised conventional tilled beds with a 0.97 m row spacing in Starkville and a 1.02 m row spacing in Stoneville. Seed were treated with a commercial premix of imidacloprid, pyraclostrobin, metalaxyl, and fluxapyroxad (Acceleron[®], Monsanto Company, St. Louis, MO) to minimize the impact of early season insect pests and seedling disease. Weed and disease pests were managed according to Mississippi State University Extension Service recommendations. Insect pest populations were monitored by sampling with a 1.5 row-m drop cloth weekly and insecticides were applied when published thresholds were reached to minimize confounding insect damage.

The experimental design was a randomized complete block with a complete factorial arrangement of treatments with four replications. Factor A was soybean growth stage at the time of fruit removal (removal timing) and included the reproductive growth stages R2, R3, R4, and R5.5 as described in Fehr and Caviness (1977). Level of fruiting structure removal (0, 50, and 100%) served as factor B in the experiment. Corn earworm damage during R2-R4 stages is primarily consumption of the pods, which don't yet contain developed seeds. Corn earworm damage to soybean in phenological growth stages from R5 and beyond typically consists of entry into the pod and consumption of individual seed. For the purposes of this study, it was deemed that whole pod removal was a relevant method for simulating corn earworm damage at all these stages due to

pods abscising after corn earworm damage has occurred and seed rots that often infect larger pods that have been fed upon but do not abscise. Once soybean plots reached their designated growth stage, fruit removal treatments were imposed by removing the designated percentage of fruit by hand. For the purposes of this experiment, flowers were included in the removal treatments. The 50% damage level was achieved by removing all fruit and flowers from the top half of each plant in the designated micro plots. The top half of the plant was chosen as greater than 90% of eggs oviposited by corn earworm are laid in the top 1/3 of the soybean canopy (Adams et al. 2015, Dill et al. 2015). The 100% removal level was achieved by removing all fruit and flowers from each plant in the designated micro plots. Once treatments had been imposed, plant height and canopy width were determined at least bi-weekly by collecting three subsamples of both canopy width and plant height in each micro plot to determine the effects of fruit loss on plant height and canopy width. One impact of fruit loss is that plants do not completely senesce which may manifest as failure to naturally abscise and shed leaves and/or failure of stems to dry down properly. Impact of fruit loss on crop senescence was determined by visually estimating the percentage of abscised leaves in each micro plot at 137 days after planting when control plots (no fruit removal) were approximately 10-15 days from harvest in Stoneville in 2012 and both locations in 2013. Impact of fruit loss on crop senescence was also determined by estimating the percentage of mainstems that remained green in each micro plot at 139 days after planting when plots were approximately 10-15 days from harvest for the study in Starkville in 2012 and both locations in 2013. Once plots had reached maturity and a harvestable moisture, each microplot was machine

harvested with a Kincaid 8XP plot combine with weigh system and seed weights and moisture were determined. Seed yields were corrected to 13% moisture.

Plant height, canopy width, crop senescence, and yield data were analyzed using a generalized linear mixed model analysis of variance (PROC GLIMMIX SAS Institute Inc. 2011). Because the control plots for each growth stage were identical (no fruiting structure removal), data for the non-damaged plots were pooled by replication and termed as a pooled fixed effect in an analysis for a randomized complete block design with incomplete factorial treatment arrangement. The first replication and three plots of the second replication at Starkville in 2013 were removed from yield analysis due to observed premature senescence from an unknown cause. Yield data were examined for outliers. Outliers were determined by plotting residuals by predicted values. Based on a normal distribution, an additional two plots with residual values more than three standard deviations from the predicted value were removed from analysis. Fruit removal timing, fruit removal level, and the interaction between removal timing and level of fruit removal all nested within the pooled fixed effect to combine all controls as a single treatment and the pooled fixed effect were designated as fixed effects. Year, location, year by location and replication nested within year by location were designated as random effects to allow for inferences to be made over a range of environments (Carmer et al. 1989, Blouin et al. 2011). Error degrees of freedom were calculated using the Kenward-Roger method. All means were separated using Fisher's protected LSD test. Differences were considered significant for $\alpha=0.05$.

Results

Neither fruit removal level nor fruit removal timing had a significant impact on plant height or canopy width at any date; therefore, results are only presented for final measurements. There was no significant impact of fruit removal timing ($F= 2.34$; $df=3$, 168 ; $P=0.07$), removal level ($F= 0.56$; $df=1$, 168 ; $P=0.45$), or the corresponding interaction between timing and level ($F= 2.27$; $df=3$, 168 ; $P=0.08$) observed for final plant heights measured at the R7 growth stage (Table 3.1) defined by Fehr and Caviness (1977). Also, there was no significant impact of fruit removal timing ($F= 2.15$; $df=3$, 168 ; $P=0.1$), removal level ($F= 0.77$; $df=1$, 168 ; $P=0.38$), or the interaction between fruit removal timing and removal level ($F= 0.38$; $df=3$, 168 ; $P=0.77$) observed for final canopy width (Table 3.1).

No significant interaction between fruit removal timing and fruit removal level was observed for delay in natural senescence as measured by abscission of leaves at 137 days after planting ($F= 1.13$; $df=3$, 133 ; $P=0.34$). Removal level had no significant impact on normal leaf abscission at 137 days after planting ($F= 3.4$; $df=1$, 133 ; $P=0.07$). Fruit removal timing did significantly impact natural leaf senescence and abscission at 137 days after planting ($F= 116.25$; $df=3$, 133 ; $P\leq 0.01$). There were no significant differences in natural leaf senescence and abscission between the non-treated control plots and plots that received fruit removal treatments at the R2 growth stage (Table 3.2). Each successive removal timing beyond R2 resulted in a significant delay in crop senescence as measured by naturally senesced leaves at 137 days after planting from the previous removal timing. Plots that received fruit removal treatments at R3 retained significantly more leaves compared to plots that received fruit removal treatments at R2

and the control plots. Plots that received fruit removal treatments at R4 retained significantly more leaves than plots that incurred fruit removal treatments at R2, R3, or the control plots. Plots that received fruit removal treatments at R5.5 retained significantly more leaves (16.25% abscised leaves) than plots receiving any other treatment or the control.

Results similar to those for abscission of leaves were observed for main stem senescence. No significant interaction between fruit removal timing and fruit removal level was observed for main stem senescence at 139 DAP ($F= 0.44$; $df=3, 124$; $P=0.72$). Also, fruit removal level did not significantly impact main stem senescence ($F= 2.12$; $df=1, 124$; $P=0.15$) at 139 days after planting. Fruit removal timing significantly impacted main stem senescence ($F= 845.31$; $df=3, 124$; $P\leq 0.01$) 139 days after planting. Plots that received fruiting structure removal treatments at R2, R3, and R4 had similar levels of non-senesced main stems compared to the non-treated control plots (Table 3.3). Plots that received fruit removal treatments at R2 had a significantly lower percentage of non-senesced main stems compared to plots that received fruit removal treatments at R3 or R4. Plots that received fruit removal at R5.5 had a significantly higher percentage of non-senesced main stems compared to plots that received fruit removal treatments at R2, R3, R4, or the non-treated control plots. Mean percent non-senesced main stems in plots that received fruit removal treatments at R5.5 was 93.3% (± 1.9), while percent non-senesced main stems for plots that received fruit removal treatments at R2, R3, R4, and the non-treated plots ranged from 2.7% (± 0.8) to 8.1% (± 2.7).

A significant interaction between fruit removal timing and fruit removal level was observed for soybean yields ($F= 3.14$; $df=3, 152.2$; $P=0.02$). Mean soybean yields

ranged from 1886.4 (± 193.9) to 3943.6 (± 212.5) kg/ha. Fruit removal up to 100% at the R2 growth stage did not significantly impact yield compared to the control plots (Table 3.4). Removal of 50% of fruiting structures at R3 also did not significantly reduce yield compared to the non-treated control plots while 100% fruit removal at R3 did significantly reduce yields from the non-treated control plots, however the removal levels at R3 were not significantly different from each other. The 50% fruiting structure removal at R4 did not significantly reduce soybean yield compared to the non-treated control. However, removal of 100% of fruiting structures at R4 significantly reduced soybean yield compared to the non-treated control. At R5.5, both levels of fruiting structure removal resulted in significantly lower soybean yields compared to the non-treated control. Also, the 100% fruit removal treatment at R5.5 resulted in significantly greater yield reduction compared to all other fruit removal level and timing treatments.

Discussion

An increase in vegetative growth as measured by plant height and canopy width was not observed for any fruit removal level or timing in the current experiment. In contrast, increased vegetative growth has been observed in cotton as a response to fruit loss (Kennedy et al. 1986). One possible reason for the lack of a vegetative growth response in soybean following fruit loss is that soybean is an annual plant whereas cotton is a perennial. Our findings are consistent with McAlister and Krober (1958) who also did not observe a vegetative growth response in determinate 'Hawkeye' and 'Lincoln' soybean varieties following fruit removal when plants had a floral bud present in the terminal inflorescences.

Senescence of plant parts (stems, leaves, and pods) and abscission of leaves are normal processes that occur as a soybean plant matures. Failure of plant parts to senesce normally and retention of non-senesced leaves is a common response of indeterminate MG IV soybean varieties in the Mid-Southern U.S. to late season stress (Egli and Bruening 2006). In the current study, fruit loss that occurred at the R3 growth stage or later resulted in increased leaf retention, while fruit loss at the R5 stage resulted in a higher occurrence of main stems remaining green. This result is similar to that observed by Hicks and Pendleton (1969) in which the vegetative parts of soybean plants that incurred floral bud removal failed to senesce when 0-60 floral buds were removed at random timings beginning when the first flower appeared until the specified removal rate was obtained in determinate 'Wayne' soybeans. McPherson and Moss (1989) observed in determinate 'Essex' soybeans that were completely depodded at times ranging from R2 to R4, that some plants depodded at late R4 remained green through harvest while other plants with the same treatment as well as those depodded earlier resulted in a maturity delay of only two to five days and senesced normally. McAlister and Krober (1958) also noted plants depodded at 40% and 80% had less maturation of pods on entire plants than the untreated control in determinate 'Hawkeye' and 'Lincoln' soybeans. These responses are most likely the plant's attempt to compensate for fruit loss. This response can be prolonged if less than optimal environmental conditions, such as low temperatures, for plant growth are encountered (McPherson and Moss 1989). The presence of non-senesced leaves and main stems when grain has reached a harvestable moisture content is problematic for producers. This green tissue greatly decreases harvest efficiency and can also influence moisture content of the harvested grain as well as foreign matter content.

Harvest aids to desiccate non-senesced leaves and main stems can be applied to improve harvest efficiency and grain quality. However, these are additional expenses and generally are only moderately efficacious in desiccating main stems.

In the early reproductive growth stages (R2) no significant yield loss was observed, while during the middle reproductive stages (R3 and R4) only 100% fruit removal significantly reduced yield. Because a soybean plant normally aborts 30 to 85% of the flower buds it produces (Swen 1933, van Schaik and Probst 1958), compensation for fruit loss during the early to middle reproductive period should occur in most instances without significant delays in maturity. Fruit removal of 100% at R3 and R4 and both removal levels at R5.5 significantly reduced yield. McPherson and Moss (1989) also observed that 100% pod removal at R4 or later significantly reduced yield of determinate 'Essex' soybeans. McAlister and Krober (1958) observed significant yield loss when 80% of pods were removed when soybean plants had the first floral bud present in the terminal inflorescence in determinate 'Hawkeye' and 'Lincoln' soybeans. Kincade et al. (1971) observed significant yield loss when 30% and 50% of pods were damaged with a leaf cutter to simulate corn earworm injury when pods were fully elongated but not filled in determinate 'Lee' soybeans. Smith and Bass (1972a) performed a study evaluating pod removal levels of 0, 10, 20, 30, 40, and 80% in determinate 'Bragg' soybeans at multiple locations. Treatments in their study were performed at late R3 to early R4, R5, R6, and R7. They observed that 80% pod removal at R5 significantly reduced yields from the non-treated control plots. They also observed that all treatments at R6 and R7 significantly reduced yields from the non-treated controls. In contrast to our results, Hicks and Pendleton (1969) observed that removing

all floral buds from the stem or leaf axils caused a significant increase in yield compared to non-treated plots, which was likely a result of overcompensation. During the later reproductive growth stages most of the natural fruit shed has probably already occurred (Swen 1933). Also, the plant has committed a significant energy investment in developing and maturing the remaining fruiting structures. Compensation for fruit loss that occurred during the latter stages of reproduction should require more time compared to fruit loss occurring during the early reproductive stages as illustrated by the crop maturity data reported in this study. In the Mid-Southern U.S., corn earworm infestations in soybean generally peak during the middle to latter portion of the growing season when soybeans are in the reproductive growth stages. Typically larger larvae, which are capable of consuming large quantities of reproductive structures, are present during the later reproductive growth stages. With the loss of a significant energy investment (ex. R5 growth stage pods) and an increasing probability of environmental conditions less than favorable for plant growth occurring, indeterminate soybeans do not appear to have the ability to compensate for moderate to high levels of fruit loss occurring during the later reproductive growth stages.

This research illustrates an important preliminary step in determining economic injury levels for pod and seed consuming insect pests such as corn earworm based on plant damage. Based on these data, the early reproductive stages of soybean are not as susceptible to yield loss from depodding, especially at low to moderate levels compared to the later stages. While admittedly, simulated pod injury may not totally mimic natural insect feeding, it has been used many times in past literature that has been mentioned in this study. Certainly insect feeding would occur over a more progressive timing, as

opposed to one discrete pod removal as performed in this study. The purpose of the authors in this study was not simply to mimic corn earworm feeding, but to determine the relationship of fruit loss at given growth stages. Parallels may still be drawn to fruit loss through corn earworm feeding to some degree, however it should be known that with natural insect feeding, natural plant responses may be elicited that were not present in our study. Additionally, other varieties may respond differently to the fruit loss treatments imposed in this study, however information on other maturity group IV soybeans and their responses are not known.

These data also reflect the potential need for a dynamic economic injury level for insect numbers and damage throughout the reproductive period of soybean growth that would become lower as crop phenology progresses through the later reproductive stages. For example, assuming a soybean market price of \$0.37/kg (\$10/bu) and an insecticide application cost of \$54.32/ha (\$22/acre) and assuming 100% control from the applied control tactic, it would only require protection of 146.8 kg (2.2 bu/ac) of yield per hectare to equal the cost of control. Based on these assumptions, approximate plant based economic injury levels can be derived within a more defined range for each growth stage (Figure 3.1). The plant damage based economic injury level for R2 and R3 are similar and would be between 30 and 50% pod or floral bud loss. The plant damage based economic injury level during R4 would be between 10 and 25% pod loss, while the R5.5 stage would have the lowest economic injury level, found between 0 and 15% pod loss. Like Thomas et al. (1974), this study shows that soybeans are less sensitive to depodding during the early reproductive stages. However, while Thomas et al. (1974) found that soybeans depodded at the R3 growth stage never reached an economic injury level, we

determined there was an economic injury level between 30 and 50% pod loss. Estimated economic injury levels from this study during R4 and R5 are similar to those of Thomas et al. (1974). More research needs to be conducted to further specify economic injury levels at each growth stage based on plant damage, however these data indicate starting points for future research. Once these specific economic injury levels are defined for each growth stage, these plant damage based economic injury levels and their associated economic thresholds could be used in conjunction with insect feeding rates to develop plant and insect-based corn earworm thresholds as is already used for other insects such as soybean defoliators (Catchot et al. 2014).

Table 3.1 Effect of fruit removal timing and fruit removal level on final mean plant height and canopy width measured at R7.

Growth Stage	Removal Level	Mean Plant Height (cm)(SEM)	Mean Canopy Width (cm)(SEM)
Non-Treated Control		67.8 (1)	66.0 (0.8)
R2	50	63.8 (2.8)	64.0 (2.3)
	100	66.0 (2.8)	63.0 (1.5)
R3	50	68.3 (2.8)	65.5 (2.3)
	100	66.3 (2.3)	65.3 (1.3)
R4	50	68.6 (1.8)	67.6 (1.5)
	100	63.2 (2.8)	65.0 (1.5)
R5.5	50	68.3 (2.3)	63.0 (1.3)
	100	69.9 (2.3)	63.0 (1.3)

None of the differences within a column are significant according to Fisher's Protected LSD test ($\alpha=0.05$).

Table 3.2 Effect of fruit removal timing on mean percent of normally abscised leaves at 137 DAP.

Growth Stage	Mean Percent Abscised Leaves (SEM)
Non-Treated Control	80.6 (1.6) a
R2	84.3 (1.3) a
R3	72.2 (3.8) b
R4	63.9 (4.7) c
R5.5	16.2 (1.8) d

Means within a column followed by the same letter are not significantly different according to Fisher's Protected LSD test ($\alpha=0.05$).

Table 3.3 Effect of fruit removal timing on percent of non-senesced main stems at 139 DAP.

Growth Stage	Mean Percent Non-Senesced Main Stems (SEM)
Non-Treated Control	4.5 (0.9) bc
R2	2.7 (0.8) c
R3	7.2 (1.1) b
R4	8.1 (2.7) b
R5.5	93.3 (1.9) a

Means within a column followed by the same letter are not significantly different according to Fisher's Protected LSD test ($\alpha=0.05$).

Table 3.4 Effect of the interaction of fruit removal timing and fruit removal level on yield of soybeans in kg/ha.

Growth Stage	Removal Level	Mean Yield in kg/ha (SEM)
Non-Treated Control		3943 a (212)
R2	50%	3708 ab (361)
	100%	3617 ab (349)
R3	50%	3889 ab (409)
	100%	3443 bc (284)
R4	50%	3806 ab (422)
	100%	2961 c (287)
R5.5	50%	2995 c (260)
	100%	1884 d (193)

Means within a column followed by the same letter are not significantly different according to Fisher's Protected LSD test ($\alpha=0.05$).

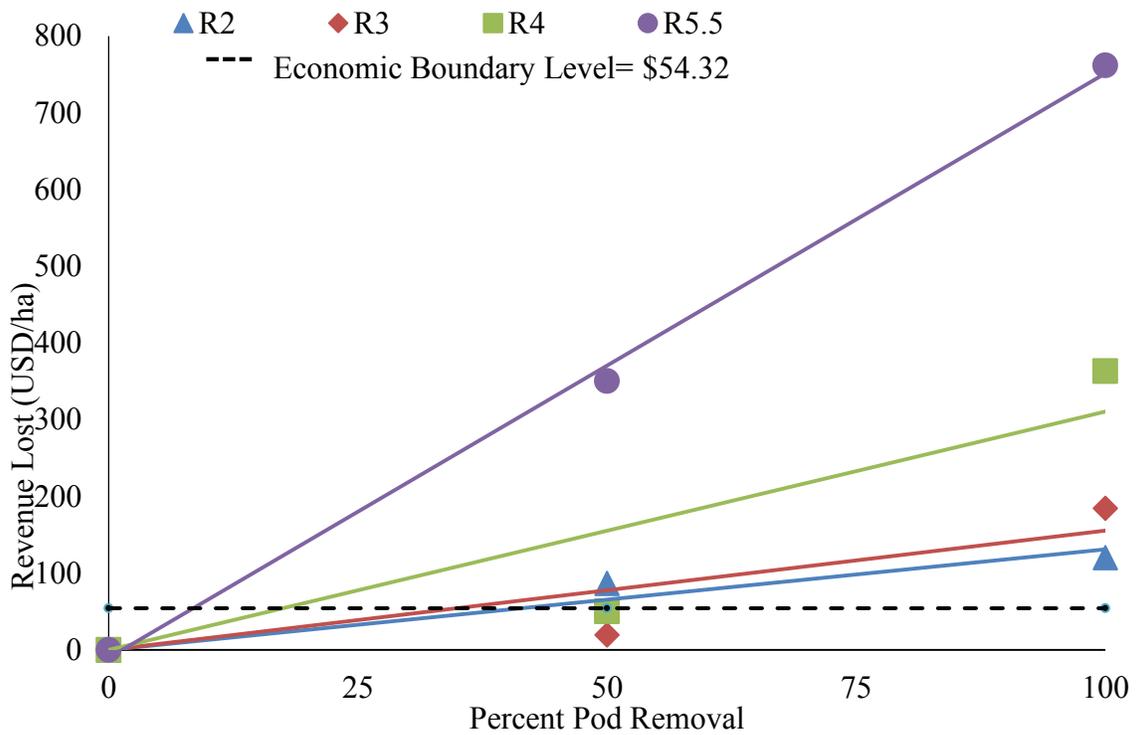


Figure 3.1 Plant based injury level for soybean reproductive growth stages expressed in mean revenue (USD/ha) lost from pod loss compared to non-damaged plants.

Trend lines are best linear fit of data for each growth stage with an intercept of 0. Economic injury level for each growth stage is where each line crosses the economic boundary line.

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CHAPTER IV
IMPACT OF SIMULATED CORN EARWORM DAMAGE ON DETERMINATE
SOYBEAN

Abstract

Field experiments were conducted in Starkville and Stoneville, MS during 2012 and 2013 to assess fruit removal level and timing on soybean growth, crop maturity, and yield. Fruit removal treatments consisted of 0, 50, and 100% of all fruit removed at specified growth stages (R2, R3, R4, and R5.5). Plant heights were determined at least bi-weekly from the time treatments were imposed until R7. The impact of fruit removal level and timing on crop maturity was determined by estimating the percentage of naturally abscised leaves at 139 DAP when control plots were approximately two weeks from harvest and the percentage of non-senesced main stems at 143 DAP. There was no significant impact of fruit removal level or timing on plant heights, however, 100% fruit removal at R5.5 resulted in significantly narrower canopy widths than all other treatments. Significant delays in crop maturity were observed when 100% of fruit was removed at R3 and R4, as well as both removal levels at R5.5. Significant yield reductions were observed at the 100% fruit removal level at R3, R4, and R5.5 compared to the non-treated control. Determinate soybeans appear to have the ability to compensate for some fruit loss between R2 and R5.5, however fruit loss at R5.5 could result in significant maturity delays. The highest fruit loss levels caused more yield loss

than the 50% removal treatments. Economic injury levels and associated thresholds need to be adjusted accordingly to account for the dynamic nature of yield loss and maturity delays.

Introduction

Soybean, *Glycine max* (Merrill), production in Mississippi has increased from 469,435 ha to 898,402 ha between 2001 and 2014 (NASS 2014c). During this same timeframe, the area planted to cotton, *Gossypium hirsutum* (L.), has decreased from 687,965 ha to 116,144 ha (NASS 2014b). The two primary reasons for this shift include favorable commodity prices for soybean compared to cotton, as well as lower production costs for soybean compared to cotton. Some of the differences in input costs between soybean and cotton can be attributed to insect management costs. Soybean insect management is generally less intensive compared to cotton (Musser et al. 2013, Williams 2014). Both cotton and soybean seed potentially incur technology fees associated with transgenic weed control traits, however cotton also incurs technology fees for transgenic insect control traits such as those expressing *Bacillus thuringiensis*, which is not commercially available in soybean in the U.S. While technology fees and seed cost for soybean are lower compared to cotton, production costs such as weed control and seed treatments are similar between the two crops. These lower production costs for soybean make it a more profitable and less risky crop for Mid-South producers.

Over the course of the last 25 years, soybean production in Mississippi has transitioned from utilizing late maturing maturity group (MG) VI and VII varieties that exhibit a determinate growth habit to earlier maturing MG IV and MG V varieties in adoption of the early soybean production system (Heatherly 1999). This shift allowed for

most of the soybean production in the Mid-South to reach the pod set and seed fill growth stages prior to the hottest and drought prone portions of the growing season. Also, earlier maturing varieties allow producers to more efficiently manage harvest in conjunction with other crops (Heatherly 1999). While most of the newer MG IV varieties and some of the MG V varieties possess an indeterminate growth habit, many of the MG V varieties in use still possess a determinate growth habit. Approximately 269,520 (30%) hectares of Mississippi's 898,402 hectares of soybean were planted to a variety with a determinate growth habit in 2013 (Irby 2014).

The corn earworm, *Helicoverpa zea* (Boddie), is the most important insect pest of soybean in Mississippi (Musser et al. 2013). In 2006, stink bugs were considered the primary pest of soybean with a revenue loss from stink bug injury plus cost of control that was just over \$19,000,000, while the revenue lost from corn earworm injury plus the cost of control totaled only \$86,000 (Musser and Catchot 2008). The average cost of one insecticide application for corn earworm at this time was only \$17.90 per hectare (Musser and Catchot 2008). By 2012, revenue lost from corn earworm damage plus the costs for control increased to over \$34,000,000 and the cost of one application of insecticide rose to \$41.98 per hectare (Musser et al. 2013). One reason could be attributed to the increase in corn production in recent years. As recently as 2006 there were less than 200,000 hectares planted to corn in Mississippi (NASS 2009). By 2012, the area planted to corn increased to 339,936 hectares (NASS 2014a). Corn serves as the primary second generation host for corn earworm in the Mid-South, while soybeans and cotton serve as primary hosts for the third and fourth generation larvae (Jackson et al. 2008). This

increase in corn, the preferred host of corn earworm, likely causes larger populations to develop on the landscape level before infesting soybean and cotton.

In Mississippi, corn earworm is typically only a pest of soybeans during the reproductive growth stages (Catchot 2014). Corn earworm usually infests soybean early in the reproductive period around R1 or R2 (Catchot 2014). At this time, larvae typically feed on the floral structures present. However, as crop phenology progresses, larvae feed more commonly on pods and seeds. Larval feeding upon leaves does occur, though less commonly than feeding on reproductive structures. Leaf feeding by corn earworm rarely causes economic damage. The threshold for corn earworm in Mississippi is currently static through the reproductive period at nine larvae per 25 sweeps with a 38cm sweep net (Catchot et al. 2014).

Previous research evaluating corn earworm infestations and fruit loss on soybean was conducted using later maturing varieties exhibiting a determinate growth habit (McAlister and Krober 1958, Hicks and Pendleton 1969, Kincade et al. 1971, Smith and Bass 1972a, Thomas et al. 1974, McPherson and Moss 1989). While research has previously been conducted using determinate varieties, those studies utilized MG VI through MG VIII varieties which aren't common to current production systems in the Mid-South. Currently, there is little information on earlier maturing determinate soybeans commonly grown in the Mid-South. The objectives of this study were to examine the impact of defined fruit removal levels at specific growth stages on maturity and yield of earlier maturing determinate soybean, and to identify a defined range for a plant damage based economic injury level using manual fruit removal.

Materials and Methods

To determine the effect of fruit loss, experiments were performed in both Starkville, MS at the R.R. Foil Plant and Soil Sciences Farm and in Stoneville, MS at the Delta Research and Extension Center during 2012 and 2013. Soybeans were planted in Stoneville, MS on May 7, 2012 and May 15, 2013. Soybeans were planted in Starkville, MS on May 21, 2012 and May 13, 2013. The experiments were conducted using an determinate maturity group (MG) V variety (Asgrow 5605[®], Monsanto Company, St. Louis, Mo). Methodology from this point forward was similar to Adams et al. (2015b). Micro plots (2 rows by 1.5 m) were established in the center of a larger two row by 6.1 m plot to eliminate edge effects. Soybeans were planted at 296,532 seeds/ha into raised conventional tilled beds with a 0.97 m row spacing in Starkville and a 1.02 m row spacing in Stoneville. Seed were treated with a commercial premix of imidacloprid, pyraclostrobin, metalaxyl, and fluxapyroxad (Acceleron[®], Monsanto Company, St. Louis, MO) to minimize the impact of early season insect pests and seedling disease. Weed and disease pests were managed according to Mississippi State University Extension Service recommendations. Insect pest populations were monitored by sampling with a 1.5 row-m drop cloth weekly and insecticides were applied when published thresholds were reached to minimize confounding insect damage.

The experimental design was a randomized complete block with a complete factorial arrangement of treatments with four replications. Factor A was soybean growth stage at the time of fruit removal (removal timing) and included the reproductive growth stages R2, R3, R4, and R5.5 as described in Fehr and Caviness (1977). Level of fruiting structure removal (0, 50, and 100%) served as factor B in the experiment. Corn earworm

damage during R2-R4 stages typically consists of consumption of pods which don't yet contain developed seeds. Corn earworm damage to soybean at the phenological growth stages of R5 and beyond typically consists of entry into the pod and consumption of individual seed. For the purposes of this study, it was deemed that whole pod removal was a relevant method for simulating corn earworm injury at this stage. At R5.5, pods abscise after corn earworm damage has occurred and seed rots often infect the larger pods that have been fed upon but do not abscise. Pods infected with seed rots typically do not contribute beyond a minimal level to overall soybean yield. Once soybean plots reached their designated growth stage, fruit removal treatments were imposed by removing the designated percentage of fruit by hand. For the purposes of this experiment, flowers were included in the removal treatments. The 50% damage level was achieved by removing all fruit and flowers from the top half of each plant in the designated micro plots. The top half of the plant was chosen as greater than 90% of eggs oviposited by corn earworm were laid in the top 1/3 of the soybean canopy (Adams et al. 2012, Dill et al. 2012). The 100% removal level was achieved by removing all fruit and flowers from each plant in the designated micro plots. Once treatments had been imposed, plant height and canopy width were determined at least bi-weekly by collecting three subsamples of both canopy width and plant height in each micro plot to determine the effects of fruit loss on plant height and canopy width. Plant height and canopy widths were not taken in the Stoneville location in either year as a result of substantial early lodging. One impact of fruit loss is that plants do not completely senesce which may manifest as failure to naturally abscise leaves and/or failure of stems to dry down properly. Impact of fruit loss on crop senescence was determined by estimating the percentage of abscised leaves in

each micro plot at 139 days after planting when control plots were approximately 10-15 days from harvest at both locations in both years. Impact of fruit loss on crop senescence was also determined by estimating the percentage of mainstems that remained green (green stems) in each micro plot at 143 days after planting when plots were approximately 10-15 days from harvest at both locations in 2013. Once plots had reached maturity and a harvestable moisture, each microplot was machine harvested and seed weights and moisture were determined. Seed yields were corrected for 13% moisture and converted to kg/ha.

Plant height, canopy width, crop senescence, and yield data were analyzed with general linear mixed model analysis of variance (PROC GLIMMIX SAS Institute Inc. 2011). Because the control plots for each growth stage were identical (no fruiting structure removal), data for the non-damaged plots were pooled by replication and termed as a pooled fixed effect to combine all controls as a single treatment in an analysis for a randomized complete block design with an incomplete factorial treatment arrangement. Fruit removal timing, fruit removal level, and the interaction between timing and level all nested within the pooled fixed effect and the pooled fixed effect were designated as fixed effects. Year, location, and replication nested within year by location were designated as random effects (Blouin et al. 2011). Years and locations were considered environmental or random effects; this allowed inferences to be made over a range of environments (Carmer et al. 1989, Blouin et al. 2011). Degrees of freedom were calculated using the Kenward-Roger method. All means were separated using Fisher's protected LSD test. Differences were considered significant for $\alpha=0.05$.

Results

Neither fruit removal level nor fruit removal timing had a significant impact on plant height at any date, therefore results are only presented for final measurements. There was no significant impact of fruit removal timing ($F= 1.57$; $df=3, 80$; $P=0.2$), removal level ($F= 0.58$; $df=1, 80$; $P=0.45$), or the corresponding interactions between growth stage and removal level ($F= 1.55$; $df=3, 80$; $P=0.21$) observed for final plant heights measured at the R7 growth stage. Mean final plant heights at R7 ranged from 71.3 (± 2.4) to 84.9 (± 1.6) cm (Table 4.1). The interaction of fruit removal timing and fruit removal level ($F= 3.63$; $df=3, 80$; $P=0.02$) had a significant impact on final canopy widths measured at the R7 growth stage. No treatment caused a significant change in canopy width compared to the non-treated control (Table 4.1). The 50 and 100% fruit removal treatments for R2, R3, and R4 were not significantly different from each other. At the R5.5 growth stage, 100% fruit removal resulted in significantly narrower canopy widths ($50.3\text{cm} \pm 2.4\text{cm}$) compared to the 50% fruit removal treatment ($61.7 \pm 1.9\text{cm}$).

There was a significant interaction observed between fruit removal timing and fruit removal level for delay in natural senescence ($F= 12.05$; $df=3, 180$; $P \leq 0.01$). Mean (SEM) soybean leaf abscission levels ranged from 27.5 (± 3.7) to 94.4 (± 1.1) %. The 50% removal level at R2, R3, and R4, as well as the 100% removal level at R2 did not result in a significant delay in crop senescence (Table 4.2). The 100% removal treatment at R3 resulted in significantly fewer naturally abscised leaves compared to the non-treated control, however it was not significantly different from the 50% removal treatment at R3. Plots that received 100% fruit removal at R4 contained significantly fewer abscised leaves than the non-treated control and significantly fewer abscised leaves

than plots that received the 50% removal treatment at R4. Both removal levels at R5.5 resulted in significantly fewer naturally abscised leaves compared to the non-treated control. The 100% removal treatment resulted in significantly fewer abscised leaves than the 50% removal treatment at R5.5 and all other level by timing treatments.

Results similar to those for naturally senesced leaves were observed for percentage of non-senesced main stems present 143 days after planting. There was a significant interaction observed between fruit removal timing and fruit removal level for delay in natural senescence of main stems ($F= 5.95$; $df=3, 87$; $P\leq 0.01$). Mean percent non-senesced main stems present ranged from 1.9 (± 0.7) to 87.5 (± 12.5) %. Plots that received the 50% fruit removal treatments at R2, R3, and R4 as well as the 100% removal treatment at R2 had similar levels of non-senesced main stems compared to the non-treated control plots (Table 4.2). Plots that received the 100% removal treatments at R3 had significantly more non-senesced main stems present than the non-treated control and also had significantly more non-senesced main stems present than plots that received the 50% removal treatment at R3. Similar to results at R3, the 100% removal treatment at R4 resulted in significantly more non-senesced main stems present than the non-treated control and the 50% removal treatment at R4. Both pod removal treatments at R5.5 resulted in a significant increase in percentage of non-senesced main stems present compared to the non-treated control. Also at R5.5, the 100% removal treatment resulted in significantly more non-senesced main stems than the 50% removal treatment and any of the other growth stage by fruit removal treatments.

A significant interaction between fruit removal timing and fruit removal level was observed for soybean yield ($F= 3.14$; $df=3, 152.2$; $P=0.02$). Mean yields ranged from

2249.8 (± 229.2) to 4140.3 (± 198.3) kg/ha. Fruit removal at R2 did not cause a significant reduction in soybean yields compared to the non-treated control (Table 4.3). The 50% removal treatment at R3 did not cause a significant reduction in yield compared to the non-treated control. However, the 100% removal treatment caused a significant reduction in yield from non-treated plots and it resulted in significantly lower yields compared to the 50% removal treatment at R3. Similar to results observed at R3, plots that received the 50% removal treatment at R4 had similar yields compared to the non-treated control. However, the 100% removal treatment again had significantly lower yields compared to control plots and the plots that received the 50% removal treatment at R4. The 50% removal treatment at R5.5 did not cause a significant reduction in yield compared to the non-treated control. The 100% removal treatment at R5.5 resulted in significantly lower yields compared to the non-treated control and all other treatments.

Discussion

An increase in vegetative growth as measured by plant height and canopy width was not observed for any fruit removal timing or level in this experiment compared to the untreated control. Increased vegetative growth is often observed in cotton as a response to fruit loss (Kennedy et al. 1986). One possible reason for the lack of vegetative growth in response to fruit loss is that the soybean cultivar used had a determinate growth habit compared to a crop such as cotton that has an indeterminate growth habit. Vegetative growth in this cultivar should have terminally ceased once reproductive growth began. Another reason for the lack of observed vegetative growth could be that soybean is an annual plant whereas cotton is a perennial. Findings in this study are consistent with those of McAlister and Krober (1958) who did not observe a vegetative growth response

in determinate ‘Lincoln’ and ‘Hawkeye’ cultivars following fruit removal once a floral bud was present in the terminal inflorescences.

Senescence of leaves, stems, and pods, as well as abscission of leaves are normal processes that occur as a soybean plant matures. It is common for soybean in the Mid-South to fail to senesce normally and retain leaves in response to late season stress (Egli and Bruening 2006). In this study, fruit loss of 100% at R3 and R4 and both fruit removal levels at R5.5 resulted in increased leaf retention, as well as an increase in the percentage of non-senesced main stems present. This result is similar to the observations of McPherson and Moss (1989) where they noted stems of some soybean plants that were depodded at late R4 remained green through harvest in the determinate cultivar ‘Essex’ while others plants of the same treatment only experienced a maturity delay of 2-5 days and senesced normally. Hicks and Pendleton (1969) observed vegetative parts of the determinate cultivar ‘Wayne’ failed to senesce when 0-60 floral buds were removed at random timings beginning when the first flower appeared on the plant. It was noted by McAlister and Krober (1958) that soybean plants of the varieties ‘Hawkeye’ and ‘Lincoln’ depodded at 40 and 80% during R4 had less maturation of pods on plants compared to the untreated control. As with our studies, these responses were attributed to the plant’s effort to compensate for fruit loss. This response can be exacerbated when less than optimal plant growth conditions, such as low temperatures, are encountered. The failure of plants to fully abscise leaves or senesce their main stems can be problematic for producers once grain has reached a harvestable moisture content. The green plant tissue can influence moisture content of the harvested seed and increase contaminating foreign matter in addition to decreasing harvest efficiency. These harvest

issues could create the need for harvest aids to desiccate non-senesced main stems and leaves to increase harvest efficiency and grain quality. This creates additional expenses for producers and are only moderately efficacious in desiccating main stems.

At R2, no removal level caused significant yield loss, while only the 100% removal levels at R3, R4, and R5.5 caused a significant reduction in yield. Compensation for fruit loss at low to moderate levels during the early reproductive period of soybeans should occur in most cases without significant maturity delays due to soybean plants normally aborting 30 to 85% of the flowers it produces (Swen 1933, van Schaik and Probst 1958). This study found that significant yield loss only occurred at 100% fruit removal levels at R3, R4, and R5.5 which is similar the findings of McPherson and Moss (1989) who observed significant yield reductions when 100% of pods were removed at R4 or later in 'Essex' soybeans. Kincade et al. (1971) found that pods damaged with a leaf cutter at levels as low as 30 and 50% when pods were fully elongated but not filled (approximately R4) resulted in significant yield reductions in 'Lee' soybeans. McAlister and Krober (1958) observed a significant yield loss in the cultivars 'Hawkeye' and 'Lincoln' when 80% of pods were removed from plants at the time of the appearance of the first floral bud in the terminal inflorescence. Smith and Bass (1972a) performed a study at late R3 to early R4, R5, R6, and R7 evaluating the impacts of 0, 10, 20, 30, 40, and 80% pod removal in the soybean cultivar 'Bragg'. They observed significant yield loss occurred at R5 when 80% of pods were removed and with all treatments imposed at R6 and R7. In contrast to the results of this study, Hicks and Pendleton (1969) observed a significant increase in yield when all floral buds were removed from the stem and leaf axils, which was likely a result of overcompensation. Compensation for early fruit loss in

this study was likely unnoticed due to the natural shedding of a large proportion of flowers during those times and the plant had time to extend the period in which it was producing floral buds to compensate for any excess loss incurred through fruit removal. In contrast, most of the natural shedding of floral buds and small pods had likely already occurred before the latter reproductive growth stages. Compensation for high levels (100%) of fruit loss during these later growth stages should require more time compared to fruit loss that occurred early in the reproductive period as evidenced by the crop maturity data reported in this study. Compensation did occur for the 50% fruit removal treatments at the latter growth stages (R3-R5.5). Commonly in the Midsouth, corn earworm infestations in soybean generally reach their highest levels in the middle to latter part of the growing season when soybeans are in the reproductive stages. Typically the larger, most damaging larvae are present during the later reproductive growth stages (R4-R5.5). With the increasing probability of environmental conditions less than favorable for plant growth occurring this late in the season coupled with the loss of significant energy investments of the plant (filled or partially filled pods), determinate soybeans do not appear to possess the ability to compensate for the higher levels (100%) of fruit loss during the middle to latter reproductive growth stages (R3-R5.5).

The data reported from this study indicate that the early reproductive stages (R2) of soybean at levels up to 100%, as well as the later reproductive stages from R3-R5.5 are not as susceptible to yield loss at fruit loss levels up to 50% compared to the higher levels of pod removal during the later reproductive period. Based on these data, there appears to be a potential need for a dynamic economic injury level for insect numbers and damaged pods throughout the reproductive period that decreases as crop phenology

progresses beyond R2, and the findings of this study represent an important preliminary step in determining a plant damage based economic injury level for pod/seed consuming insects such as corn earworm.

Table 4.1 Effect of fruit removal timing and fruit removal level on final mean plant height and canopy width measured at R7.

Growth Stage	Removal Level	Mean Plant Height (cm) (SEM)	Mean Canopy Width (cm) (SEM)
Non-Treated Control		82.4 (0.9)	55.7 (0.8) abc
R2	50	77.7 (2.1)	56.4 (1.2) abc
	100	84.9 (1.6)	58.4 (1.0) ab
R3	50	81.2 (1.9)	57.2 (1.3) abc
	100	76.3 (1.9)	52.6 (1.4) bc
R4	50	71.3 (2.4)	54.3 (1.5) abc
	100	78 (2.6)	60.7 (1.6) ab
R5.5	50	81.8 (2.6)	61.7 (1.9) a
	100	80.1 (2.7)	50.3 (2.4) c

Means within a column followed by the same letter are not significantly different according to Fisher's Protected LSD test ($\alpha=0.05$). Columns without letter groupings were not significantly different according to Fisher's Protected LSD test ($\alpha=0.05$).

Table 4.2 Effect of the interaction between fruit removal timing and fruit removal level on mean percent of normally abscised leaves at 139 days after planting and percent of non-senesced main stems at 143 days after planting.

Growth Stage	Removal Level	Mean Percent Abscised Leaves (SEM)	Mean Percent Non-Senesced Mainstems (SEM)
Non-Treated Control		94.4 (1.1) a	1.9 (0.7) a
R2	50	90.6 (4.4) ab	16.25 (12.1) ab
	100	91.9 (2.6) ab	2.5 (1.6) a
R3	50	90.9 (2.2) ab	5 (1.9) a
	100	85.3 (3.7) b	27.5 (8.6) b
R4	50	89.4 (2.5) ab	15 (5.3) ab
	100	63.1 (4.7) c	65 (11.2) c
R5.5	50	60.9 (5.9) c	63.8 (13.1) c
	100	27.5 (3.7) d	87.5 (12.5) d

Means within a column followed by the same letter are not significantly different according to Fisher's Protected LSD test ($\alpha=0.05$).

Table 4.3 Effect of the interaction of fruit removal timing and fruit removal level on yield of soybeans in kg/ha.

Growth Stage	Removal Level	Mean Yield in Kg/Ha (SEM)
Non-Treated Control		3954 (111) a
R2	50	3752 (238) ab
	100	3945 (198) ab
R3	50	4013 (216) a
	100	3487 (213) b
R4	50	4140 (198) a
	100	3495 (209) b
R5.5	50	3755 (241) ab
	100	2249 (229) c

Means within a column followed by the same letter are not significantly different according to Fisher's Protected LSD test ($\alpha=0.05$).

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CHAPTER V
COMPENSATION OF INDETERMINATE AND DETERMINATE SOYBEAN TO
MANUAL FRUIT REMOVAL

Abstract

Field experiments were conducted in Starkville and Stoneville, MS during 2012 and 2013 to evaluate the compensatory growth of fruiting structures of both determinate and indeterminate soybean subjected to manual fruit removal treatments. Removal treatments of 0, 50, and 100% were imposed to soybean at specified growth stages (R2, R3, R4, and R5.5). Soybean fruiting was portioned from the bottom of the plant to the top into four fruiting zones and the number of pods with one, two, three, and four seed as well as individual seed weights were determined for each zone. Both determinate and indeterminate soybean exhibited compensation of various levels. When fruit was removed after R2, soybean was not able to fully compensate through increasing pod loads to levels equivalent of the control regardless of growth habit. Fruit removal from R2 through R4 typically resulted in increased seed weight compared to the control. In contrast, fruit removal at R5.5 reduced seed weights. Overall, fruit removal after R2 does not appear to be able to fully compensate for fruit loss. Producers experiencing heavy infestation of pod consuming pests, such as corn earworm, *Helicoverpa zea* (Boddie), should monitor populations closely in order to mitigate yield losses when economical.

Introduction

Corn earworm, *Helicoverpa zea* (Boddie), has recently become the primary insect pest of soybean in Mississippi (Musser et al. 2010, Musser et al. 2011, Musser et al. 2012, Musser et al. 2013). Prior to 2009, the corn earworm ranked as a relatively minor pest when compared to the stink bug complex, the soybean looper, *Chrysodeixis includens* (Walker), and bean leaf beetle, *Cerotoma trifurcata* Forster, among others. In 2009, the pest status of corn earworm rose from relatively minor to the second most economic pest of Mississippi soybean causing over \$17,000,000 in crop loss plus control costs, only behind the stink bug complex (Musser et al. 2010). By 2010, corn earworm had become the most important economic pest of soybean in Mississippi causing nearly \$25,000,000 in crop loss and control costs (Musser et al. 2011). One proposed reason for the increase in infestation levels of corn earworm in soybean is the recent increase in area planted to soybean coupled with the decline in cotton production (NASS 2014b, c). Both crops are known to serve as major hosts for third and fourth generation corn earworm larvae in Mississippi (Jackson et al. 2008). In Mississippi soybean, corn earworm infestations usually occur during the beginning of the reproductive stages of the crop and can last through pod fill (Catchot 2014).

Over the course of the last 20 years, soybean production in Mississippi has experienced significant changes with respect to production practices. Prior to this time, most soybean planting began in May and ran through the end of June and utilized later maturity group (MG) V and VI varieties that exhibited a determinate growth habit. During this production shift, growers have adopted an early soybean production system, shifting to planting beans as early as March through the end of June (Heatherly 1999).

This system primarily employs the use of earlier maturing MG IV and V varieties. Most of the earlier varieties possess an indeterminate growth habit, however some still exhibit the determinate growth habit. Approximately 70% of the area planted to soybeans in Mississippi are planted to an indeterminate variety while determinate varieties make up the balance of the area (Irby 2014).

Aside from work done by Eckel (1991), little research exists on soybean compensation response mechanisms to pod/seed consuming pests such as corn earworm. Eckel (1991) utilized MG V, MG VI and MG VII soybean cultivars, all of which likely possessed a determinate growth habit and the latter two of which are not common to production in the Mid-Southern states. Since the time of that study, varietal characteristics have changed as well as experiencing increased yield potential. With the widespread adoption of varieties exhibiting an indeterminate growth habit, investigations need to be made into the potential of these cultivars to compensate for fruit loss. The purpose of this study is to investigate whether indeterminate and determinate varieties have the ability to compensate for fruit loss, and if so, by what mechanism(s) it is occurring.

Materials and Methods

To determine the compensation ability of indeterminate and determinate soybean cultivars incurring fruit loss in defined reproductive stages, experiments were conducted in Starkville, MS at the R.R. Foil Plant and Soil Sciences Farm and in Stoneville, MS at the Delta Research and Extension Center during 2012 and 2013. Due to differences in maturity and growth habits, separate studies were conducted using an indeterminate and determinate variety. One experiment was conducted using an indeterminate MG IV

variety (Asgrow 4605[®], Monsanto Company, St. Louis, Mo). The second study was conducted using a determinate MG V variety (Asgrow 5605[®], Monsanto Company, St. Louis, Mo). The methodology for each experiment was identical. Micro plots (2 rows by 1.5 m) were established in the center of a larger two row by 6.1 m plot to eliminate edge effects. Soybeans were planted at 296,532 seeds/ha into raised conventional tilled beds with a 0.97 m row spacing in Starkville and a 1.02 m row spacing in Stoneville. Seed were treated with a commercial premix of imidacloprid, pyraclostrobin, metalaxyl, and fluxapyroxad (Acceleron[®], Monsanto Company, St. Louis, MO) to minimize the impact of early season insect pests and seedling disease. Weed and disease pests were managed according to Mississippi State University Extension Service recommendations. Insect pest populations were monitored weekly and insecticides were applied when published thresholds were reached to minimize confounding insect damage. The experimental design was a randomized complete block with a complete factorial arrangement of treatments with four replications. Factor A was soybean growth stage at the time of fruit removal (removal timing) and included the reproductive growth stages R2, R3, R4, and R5.5 as described in Fehr and Caviness (1977). Level of fruiting structure removal (0, 50, and 100%) served as factor B in the experiment. Corn earworm damage to soybean at phenological growth stages from R5 and beyond typically consists of entry into the pod and consumption of individual seed. For the purposes of this study, it was deemed that whole pod removal was a relevant method for simulating corn earworm damage at this stage due to pods abscising after corn earworm damage has occurred. Once soybean plots reached their designated growth stage, fruit removal treatments were imposed by removing the designated percentage of fruit by hand. For the purposes of this

experiment, flowers were included in the removal treatments. The 50% damage level was achieved by removing all fruit and flowers from the top half of each plant in the designated micro plots. The 100% removal level was achieved by removing all fruit and flowers from each plant in the designated micro plots. When seed had reached maturity and a harvestable moisture content, five plants per micro plot were randomly collected for yield component and compensation analysis. In 2012, yield was partitioned by grouping mainstem nodes beginning at the base of the plant into fruiting zones. Zone 1 consisted of nodes 1-4, zone 2 consisted of nodes 5-8, zone 3 consisted of nodes 9-12, while zone 4 consisted of nodes 13 and above. Pods in each zone were further partitioned by the number of seed per pod and all seed were summed to obtain total number of bean seed per zone. After analysis of yield components in 2012, it was discovered that seed size was likely playing an important role in compensation, therefore, in 2013 seed weight was determined for each zone. The remaining plants in each plot were then machine harvested to determine yields as a comparison to seed and pod counts, as well as seed weights. Machine harvested weights are reported in this study but are also found in Adams et al. (2015a,b).

Number of one, two, three and four bean pods per zone, total bean seed per zone, and individual seed weight per zone were measured on the five plant subsamples per plot and averaged then analyzed with mixed model analysis of variance (Littell et al. 2006). Due to overall scarcity of four bean pods in both studies, individual analysis of four bean pods was excluded and was only accounted for in the measure of total bean seeds per zone. Because the control plots for each growth stage were identical (no fruiting structure removal), data for the non-damaged plots were pooled by replication in an

analysis for a randomized complete block design with incomplete factorial treatment arrangement. Fruit removal timing, fruit removal level, fruiting zone (zone) and all interactions between removal timing, level of fruit removal, and zone were designated as fixed effects. Year, location, and replication nested within year by location were designated as random effects to allow inferences to be made over a range of environments (Carmer et al. 1989, Blouin et al. 2011). Degrees of freedom were calculated using the Kenward-Roger method. All means were separated using Fisher's protected LSD test. Differences were considered significant for $\alpha=0.05$.

Results

Indeterminate Study Results

There was a significant interaction of fruit removal timing and fruit removal level observed for soybean yields ($F= 3.14$; $df=3, 152.2$; $P=0.02$). Fruit removal at R2, regardless of level, did not significantly reduce yields compared to the control (Figure 5.1). At R3 and R4, only 100% fruit removal significantly reduced yields compared to the control. At R5.5, both fruit removal levels significantly reduced yields below the non-treated control, and the 100% fruit removal treatment resulted in significantly lower yields than all other treatments.

There was no significant 3-way interaction between fruit removal level, fruit removal timing, and zone observed for total seed number ($F= 1.44$; $df=9, 525$; $P=0.17$) nor was there a significant interaction between fruit removal timing and level for total beans ($F= 1.56$; $df=3, 525$; $P=0.2$). A significant interaction between fruiting zone and fruit removal timing was observed for total seed number ($F= 7.17$; $df=9, 525$; $P\leq 0.01$). Findings from this study indicate that soybean plants not incurring fruit loss during the

reproductive stages had a seed distribution that was relatively uniform from the bottom of the plant to the top. Removal of fruit at R2 and R3 had minimal effect on soybean seed distribution throughout the plant with no zone at the R2 or R3 fruit removal timings having significantly less soybean seed for the same zone in control plots (Figure 5.2). When fruit was removed at R4, there was a significant change in total seed distribution, with more seed in zone 1 and fewer seed in zone 3 compared to the non-treated control. When fruit removal occurred at R5.5, all zones except zone 1 experienced a significant decrease in total seed present compared to the control.

There was also a significant interaction between removal level and zone for total seeds present ($F= 4.84$; $df=3, 525$; $P\leq 0.01$). When 50% of fruit was removed, zones 1 and 2 did not experience a significant decrease in total seeds present compared to the control, while zones 3 and 4 did experience a significant decrease in total seed compared to the control (Figure 5.2). This result was expected given that the 50% damage treatment was imposed to the top half of the plant. When 100% of fruit was removed, zone 1 and 4 did not incur a significant reduction in total seed present while zones 2 and 3 did experience a significant reduction in total seed present compared to the control plots.

Similar to the results of total seed analysis, there was no significant 3-way interaction between fruit removal timing, fruit removal level, and fruiting zone for pods with three seeds ($F= 1.18$; $df=9, 525$; $P=0.3$) nor was there a significant interaction between fruit removal timing and level for pods with three seeds ($F= 1.15$; $df=3, 525$; $P=0.32$). There was a significant interaction between fruit removal timing and zone for pods with three seeds ($F= 5.70$; $df=9, 525$; $P\leq 0.01$). In the control plots, pods with three

seeds were found to be distributed uniformly across all zones with means ranging from 6.2-7.1 per zone. Fruit loss at R2 and R3 had no significant effect on distribution of pods with three seeds compared to the control (Figure 5.2). When fruit was removed at R4 and R5.5, there was a significant change in distribution of pods with three seeds. Plants receiving fruit removal treatments at R4 had significantly fewer pods with three seeds in zone 3 than the control plants. When fruit loss occurred at R5.5, all zones had significantly fewer pods with three seeds than those for the control.

There was also a significant interaction between fruit removal level and zone for pods with three seeds ($F= 5.0$; $df=3, 525$; $P\leq 0.01$). Similar to results for total seed, when 50% of fruit was removed, there was no significant reduction in pods with three seeds in zones 1 or 2 compared to those zones for the control (Figure 5.2). Plants that received 50% fruit removal had significantly fewer pods with three seeds in zones 3 and 4 compared to the same zones for control plants. In plots that received 100% fruit removal, no significant reduction in pods with three seeds was observed for zones 1 or 4 while a significant reduction in pods with three seed was observed for zones 2 and 3 compared to the control.

There was no significant 3-way interaction between fruit removal timing, fruit removal level, and fruiting zone observed for pods with two seed ($F= 1.70$; $df=9, 525$; $P=0.08$). There was a significant interaction between fruit removal timing and fruit removal level for mean pods with two seeds present per zone ($F= 3.22$; $df=9, 525$; $P=0.02$). Control plots averaged 4.6 pods with two seeds per zone. Fruit removal of 50 or 100% at the R2 or R3 growth stages did not significantly impact the number of pods with 2 seeds present per zone (Figure 5.2). Fruit removal of 50% at R4 did not

significantly reduce the number of pods with 2 seeds. However, the plots that received the 100% fruit removal had significantly more pods with 2 seed/zone compared to plots that received the 50% removal treatment. When fruit was removed at R5.5, both the 50% and 100% removal treatments resulted in significantly fewer pods with two seeds per zone compared to the control plots.

There was also an interaction of fruit removal timing and zone for pods with two seeds ($F= 6.52$; $df=9, 525$; $P\leq 0.01$). In control plots, pods with two seeds were distributed somewhat uniformly among zones, with significantly fewer pods with two seeds in zone 1 compared to zone 2. Similar to results for pods with two seeds, fruit removal at R2 and R3 did not significantly change the distribution of pods with three seeds (Figure 5.2). For plots that received fruit removal treatments at R4, fruiting zone 1 contained significantly more pods with two seeds than the control plots, while fruiting zone 3 contained significantly fewer pods with two seeds than the control. When fruit was removed at R5.5, there were significantly fewer pods with two seeds in fruiting zones 3 and 4 compared to the control.

There was also an interaction between fruit removal level and zone for pods with two seeds ($F= 4.1$; $df=3, 525$; $P\leq 0.01$). Similar to results of pods with three seeds, when 50% of fruit was removed, there were significantly fewer pods with two seeds in zones 3 and 4 compared to the same zones in the non-treated control (Figure 5.2). When 100% of fruit was removed, no zone experienced significant reductions in number of pods with two seeds compared to the non-treated control.

There was no significant 3-way interaction of fruit removal timing, level and zone for pods with one seed ($F= 1.73$; $df=9, 525$; $P=0.08$). There was a significant interaction

of fruit removal timing and level for mean pods with one seed ($F= 5.16$; $df=3, 525$; $P\leq 0.01$). Control plots averaged 0.94 pods with one seed per zone. Fruit removal of 50% or 100% at the R2 and R3 growth stages did not significantly impact the number of pods with one seed present per zone compared to the control (Figure 5.2). When 50% of fruit was removed at R4 or R5.5, there was no significant impact on the number of one bean pods present per zone. However, 100% fruit removal at R4 or R5.5, resulted in a significant increase in the number of pods with one seed per zone compared to the control.

There was also an interaction of fruit removal timing and zone for pods with one seed present ($F= 6.52$; $df=9, 525$; $P\leq 0.01$). Pods with one seed were found to be distributed uniformly throughout all four zones in the non-treated control. Removal of fruit at R2 and R3 did not significantly impact the distribution of pods with one seed compared to the control (Figure 5.2). When fruit removal occurred at R4 and R5.5, there was a significant increase in the amount of pods with one seed present in zone 1 compared to the control.

There was also an interaction between fruit removal level and zone for pods with one seed ($F= 6.88$; $df=3, 525$; $P\leq 0.01$). When 50% of fruit was removed, no zones were significantly different from the same zone in the control plots with respect to pods with one seed (Figure 5.2). When 100% of fruit was removed, zone 1 experienced a significant increase in the amount of pods with one seed present compared to the non-treated control.

There was a significant 3-way interaction between fruit removal timing, fruit removal level, and zone for mean individual seed weight ($F= 2.3$; $df=9, 240$; $P\leq 0.01$). In

control plots, the upper three zones (2, 3, and 4) of the canopy produced significantly heavier seeds than the bottom zone. The same trend continues across both the 50% and 100% fruit removal levels at R2, R3, and R4 as the upper three zones for each removal timing and level combination produced significantly heavier seed than the bottom zone (Figure 5.3). Also, none of the four zones in any of the previously mentioned fruit removal timing by level combinations were significantly different from the same zone in the control plots. Zones 1 and 2 of the R5.5-50% removal plots produced significantly heavier seed than the same zones in the control plots, while zone 4 produced significantly lighter seed than zone 4 of the control. Zone 1 of the R5.5-100% removal plots also produced significantly heavier seed than zone 1 of the control plots, while zones 2-4 produced significantly lighter seeds than the same zones of the control.

Determinate Study Results

A significant interaction between fruit removal timing and fruit removal level was observed for soybean yield ($F= 3.14$; $df=3, 152.2$; $P=0.02$). Fruit removal at any level at R2 did not cause a significant yield loss (Figure 5.4). At R3, R4, and R5, only 100% fruit removal caused a significant reduction in yields compared to the control plots, and the 100% removal plots also yielded significantly less than the 50% removal plots. The 100% removal treatment at R5.5 resulted in significantly lower yields than all other treatments.

There was no significant 3-way interaction between fruit removal level, fruit removal timing, and zone for total seed ($F= 1.64$; $df=9, 525$; $P=0.1$). Also, there was no significant interaction between fruit removal timing and level for total seed ($F= 1.89$; $df=3, 525$; $P=0.13$) or between zone and fruit removal timing for total seed ($F= 0.85$;

df=9, 525; $P=0.6$). There was a significant interaction between removal level and zone for total seed present ($F= 6.02$; df=3, 525; $P\leq 0.01$). For the control plots, zone 2 had significantly more seed than all other zones, while zone 4 had significantly fewer seed than all other zones. When 50% of fruit was removed, seed distribution in zones 1 and 2 was unchanged compared to control plots. However, zones 3 and 4 had significantly fewer seed than zones 3 and 4 in the control plots (Figure 5.5). When 100% of fruit was removed, only zone 1 did not experience a significant reduction in seed compared to the same zone in the control plots.

Similar to the results for total seed analysis, there was no significant 3-way interaction between fruit removal timing, fruit removal level, and fruiting zone for pods with three seed ($F= 0.81$; df=9, 525; $P=0.6$). There was a significant interaction between fruit removal timing and level for pods with three seed ($F= 2.87$; df=3, 525; $P=0.04$). Both fruit removal levels at R2 resulted in similar numbers of pods with three seed per zone as the control (Figure 5.5). Both fruit removal levels at R3 resulted in significantly fewer pods with three seed per zone than the non-treated control, however, there was no significant differences between the 50% and 100% fruit removal levels. At R4, only the 100% removal level resulted in significantly fewer pods with three seed per zone than the control. At R5.5, both removal levels resulted in significantly fewer pods with three seed than the control.

There was no significant interaction between fruit removal timing and zone for pods with three seed ($F= 0.43$; df=9, 525; $P=0.9$). There was a significant interaction between fruit removal level and fruiting zone for pods with three seed present ($F= 4.97$; df=3, 525; $P\leq 0.01$). In control plots, zone 2 had significantly more pods with three seed

than zones 1 and 4, however, it was not significantly different from zone 3. Zone 3 had significantly more pods with three seed than zone 4 but was not significantly different from zone 1. When 50% of fruit was removed, only zone 3 had a significant reduction in pods with three seed compared to the same zone in the non-treated control (Figure 5.5). When 100% of fruit was removed, only zones 2 and 3 experienced a significant reduction in pods with three seed compared to the same fruiting zones in the control.

There was a significant 3-way interaction between fruit removal timing, fruit removal level, and zone for mean pods with two seed ($F= 1.87$; $df=9, 525$; $P=0.05$). In control plots, there were significantly more pods with two seed in zone 2 than in all other zones. Zone 3 had significantly more pods than zone 4, while zones 1 and 4 were not significantly different from each other. When fruit was removed at R2 regardless of level, distributions of pods with two seed were not significantly different from that observed in the control (Figure 5.5). When 50% of fruit was removed at R3, only zone 4 had significantly less pods with two seed than the same zone for the non-treated control. When 100% of fruit was removed at R3, zones 2 and 3 had significantly fewer pods with two seed present compared to the same zones in the control plots. When 50% of fruit was removed at R4, there was a significant increase in pods with two seed in zone 1, while there was a significant reduction of pods with two seed in zones 3 and 4 compared to the same zones in the control. When 100% of fruit was removed at R4, there were no significant differences for any zones compared to the control. When 50% of fruit was removed at R5.5, only zones 3 and 4 had a significant reduction in pods with two seed compared to the same zones for the control plots. When 100% of fruit was removed at

R5.5, zones 2, 3, and 4 all experienced a significant reduction in two bean pods compared to respective fruiting zones for the control.

There was no significant 3-way interaction between fruit removal timing, fruit removal level, and zone for pods with one seed ($F= 0.3$; $df=9, 525$; $P=0.9$). There were also no significant interactions between fruit removal timing and zone ($F= 1.01$; $df=9, 525$; $P=0.4$) or fruit removal level and zone ($F= 0.59$; $df=3, 525$; $P=0.6$). There was a significant interaction between fruit removal timing and level for pods with one seed ($F= 2.62$; $df=3, 525$; $P=0.05$). Fruit removal at R2, R3, and R4 regardless of level did not cause a significant change in pods with one seed per zone compared to the control (Figure 5.5). At R5.5, only the 50% removal level caused a significant reduction in number of pods with one seed per zone compared to the control.

There was no significant 3-way interaction between fruit removal timing, fruit removal level, and zone for mean individual seed weight ($F= 1.05$; $df=9, 239$; $P=0.4$). There was no significant interaction between removal level and zone for individual seed weight ($F= 0.78$; $df=3, 239$; $P=0.5$). There was a significant interaction of fruit removal timing and fruit removal level for mean seed weight across all zones ($F= 4.06$; $df=3, 239$; $P\leq 0.01$). In control plots, mean individual seed weight was 0.38 g. Neither fruit removal level at R2 caused a significant change in mean individual seed weight compared to the control (Figure 5.6). When fruit was removed at R3 and R4, 50% fruit removal did not significantly impact mean individual seed weight, while 100% fruit removal caused a significant increase in mean seed weight compared to the control. When fruit was removed at R5.5, both removal levels resulted in a significant decrease in mean individual seed weight.

There was also a significant interaction of fruit removal timing and zone for mean individual seed weight ($F= 3.37$; $df=9, 239$; $P\leq.01$). In the non-treated control plots, significantly heavier seed were present in zones 2 and 3 than zones 1 and 4. Also, in the control plots, seed weights for zones 2 and 3 were not significantly different from each other, and seed weights for zones 1 and 4 were also not significantly different from each other. Fruit removal at R2 and R4 did not significantly affect mean individual seed weights for any zone compared to the control plots (Figure 5.7). When fruit was removed at R3, significantly heavier seed were produced in zones 1 and 2 compared to the same zones in the control plots. When fruit was removed at R5.5, a significant increase in mean individual seed weight was observed in zone 1 compared to fruiting zone 1 for the control, while there was a significant reduction in seed weight for zones 3 and 4 compared to the same zones for the control plots.

Discussion

Significant yield reductions were not observed for the indeterminate cultivar until 100% of fruit was removed at R3 and R4, and when fruit was removed at both levels at R5.5 compared to the non-treated control. Generally, as fruit removal occurred beyond R2, there was an overall reduction of both pods with two and three seeds compared to the non-treated control across the entire plant. Similarly, as the injury level increased to 50 and 100% fruit removal, there were also reductions of pods with two and three seeds. In contrast, there was a general increase of pods with one seed as injury timing progressed through the reproductive stages of soybean, as well as when injury level increased after the R2 growth stage. At the 50% injury level, seed weights remained similar or increased compared to the control through the R4 stage. When 100% of fruit was removed, overall

seed weights began declining at R3. The ability of soybean to maintain or increase seed weight likely allowed for the yields of the 50% removal treatments at R3 and R4 to not be reduced below the non-treated control. The inability to maintain seed weight as well as the inability to produce similar levels of pods with two or three seed is what ultimately reduced yields in the 50% removal treatment at R5.5 as well as the 100% removal treatments at R3-R5.5. These data indicate that current indeterminate soybean cultivars may not possess the ability to compensate for late season fruit loss at high levels through increased pod production or seed weight. Data for pods with three seeds for indeterminate soybean suggest that although indeterminate soybean might be capable of resetting some pods to compensate for fruit loss at later reproductive stages, plants do not have the ability to produce enough additional pods with three seed to offset the original fruit loss. This further indicates that indeterminate soybeans are not capable of fully compensating for late season fruit losses, particularly at high levels solely through setting additional pods.

Significant yield reductions in the determinate cultivar were only observed at the 100% removal level at R3, R4, and R5.5. Generally, as fruit removal occurred beyond R2, there was a reduction in pods with three seeds with the exception of the 50% removal treatment at R4 compared to the non-treated control. Also, as injury level increased to 50 and 100%, generally there was a reduction of pods with three seeds compared to the non-treated control. The 50% removal treatment at all growth stages produced similar levels of pods with two seeds, while the 100% level experienced reductions in levels of pods with two seeds once removal occurred beyond the R2 timing. Only fruit removal treatments at R5.5 were not able to maintain similar levels of pods with one seed

compared to the non-treated control. Both fruit removal levels at R2, R3, and R4 either maintained or increased seed weight compared to the non-treated control while both removal levels at R5.5 resulted in reduced seed weights. All of the 50% fruit removal levels were able to compensate for fruit loss compared to the non-treated control through a combination of resetting pods with one and two seeds as well as maintaining seed weight, the latter of which likely being the primary contributor to compensation. The reduction of pods in the 100% removal levels at R3-R5.5 were too great to be overcome by resetting pods and increased seed weights.

Data regarding indeterminate and determinate soybean total seed produced suggest that producing additional pods after a fruit loss event during the later reproductive stages occurs more strongly at the bottom of the plant, and fruiting positions higher in the canopy are not capable of producing as many seed as those lower on the plant. It appears that plants are not capable of producing enough additional pods to offset those larger losses after fruit set has occurred regardless of growth habit, particularly once plants have reached R3 or later. In a study utilizing multiple cultivars, Eckel (1991) found that when corn earworm larvae were present prior to peak flowering, there was often a significant reduction of flowers. With some treatments, there were also reduced numbers of total seeds/pods. This was attributed to infestations that occurred prior to peak flowering and the reduction in flowers prior to pod formation.

It appears current indeterminate and determinate cultivars adapted for use in the Mid-South are not able to compensate fully through additional pod production or increased seed weights under the highest levels of fruit loss in the middle to latter reproductive stages. At this point, it is likely that soybean has contributed a significant

energy investment in the form of nutrients to setting and attempting to fill a full pod load, and is unable to completely restart the process. During the earlier reproductive growth stages (R2-R3), both indeterminate and determinate soybean appear to have a substantial ability to compensate for moderate to high levels of fruit loss. Also, these data would suggest that because the 50% removal level had less impact on yields of the determinate cultivar, that overall the later maturing determinate cultivar has more ability to compensate for moderate levels of fruit loss. This however could be due in part because of the extended relative maturity of this particular cultivar.

Overall, both indeterminate and determinate cultivars in our studies exhibited some degree of compensation, primarily through seed weights and to a lesser degree, additional pod production. However these responses did not always prove to be enough to offset yield losses incurred through fruit removal. Under severe infestation and the expectancy of heavy fruit damage, growers should monitor corn earworm populations closely and be prepared to take appropriate action and management steps to mitigate potential damage, as soybean doesn't appear to have the ability to compensate for the most severe levels of fruit loss.

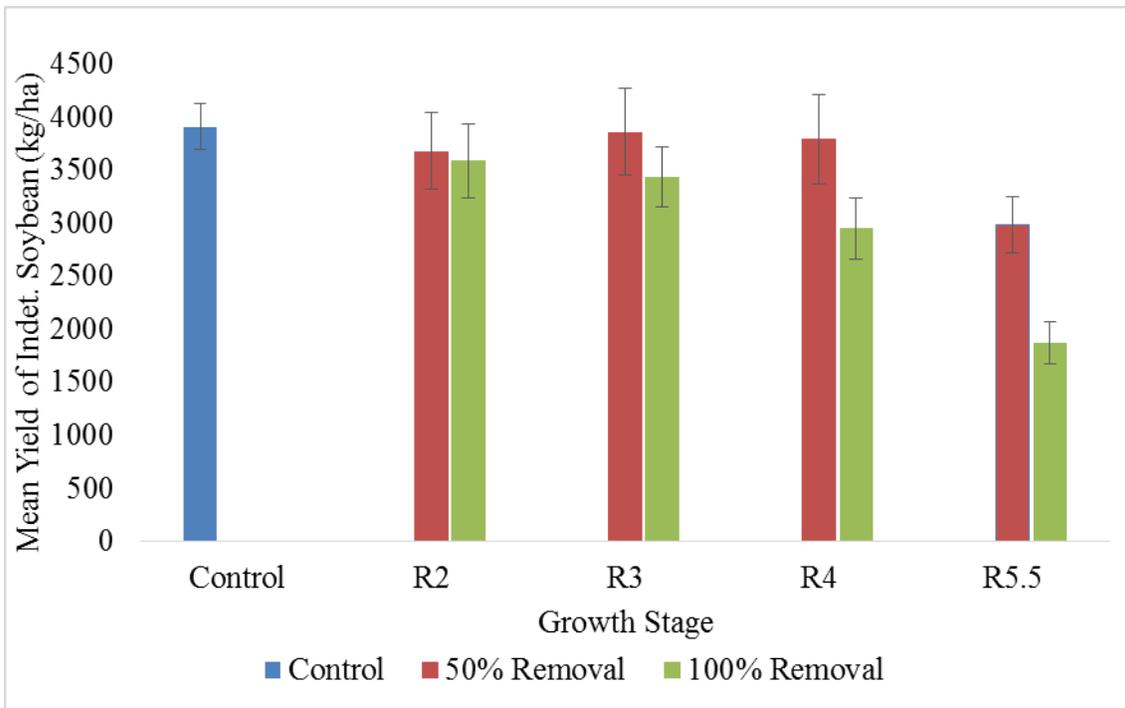


Figure 5.1 Interaction effect of fruit removal timing and fruit removal level on yield of indeterminate soybean in field plots in Mississippi in 2012-2013.

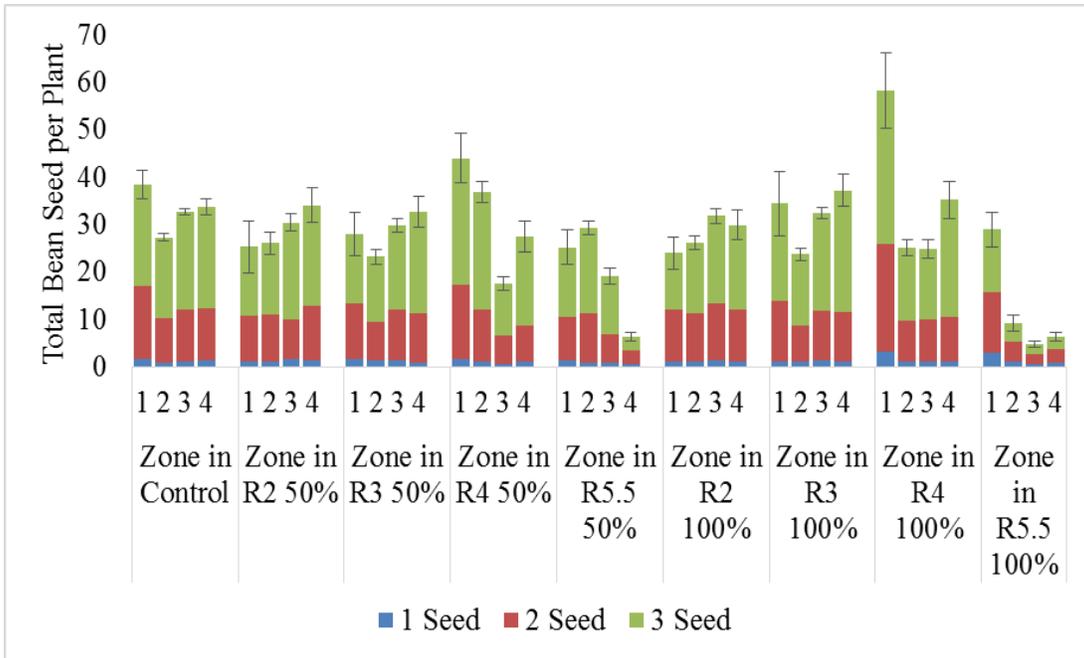


Figure 5.2 Contribution of pods with 1, 2, and 3 seed to total beans present in each zone of each growth stage by removal level treatment in indeterminate soybean from field plots in Mississippi in 2012-2013.

Standard errors presented are the standard errors for total beans for each zone.

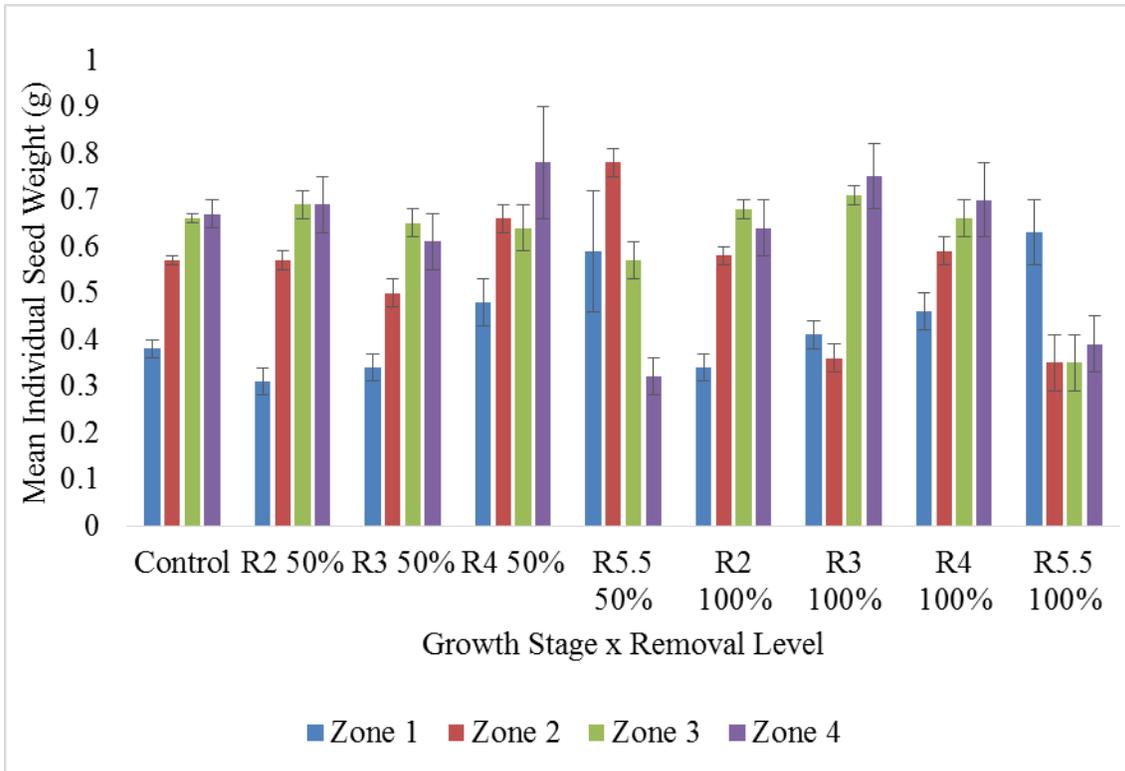


Figure 5.3 Interaction of fruit removal timing, level, and zone on mean individual seed weight of indeterminate soybean from field plots in Mississippi in 2012-2013.

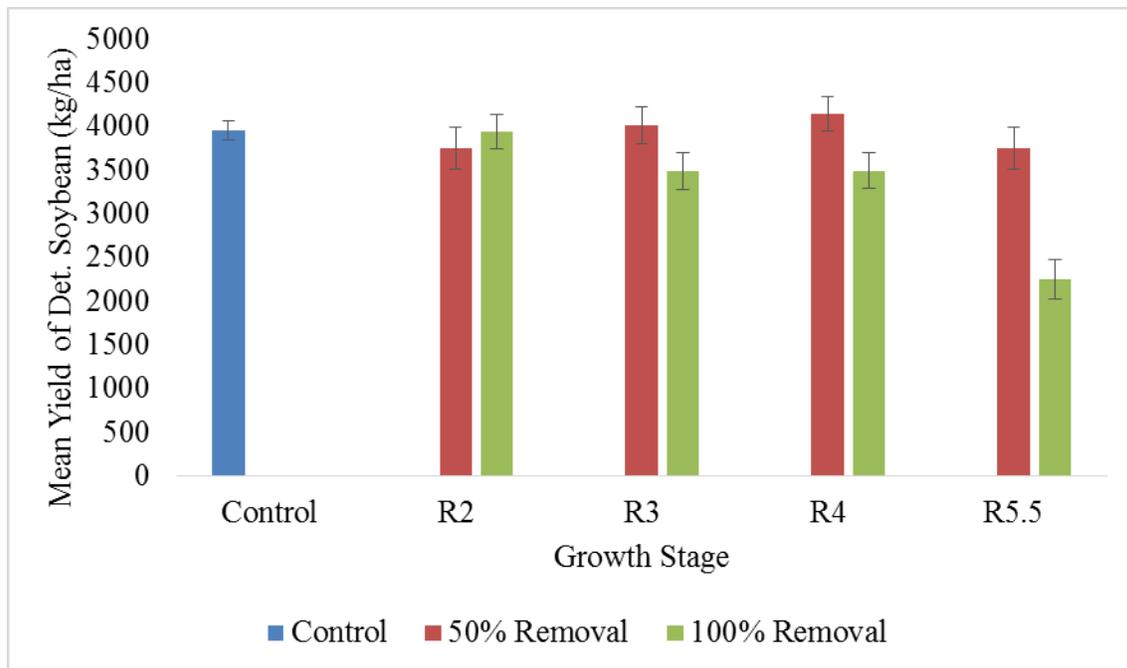


Figure 5.4 Interaction effect of fruit removal timing and fruit removal level on yield of determinate soybean in field plots in Mississippi in 2012-2013.

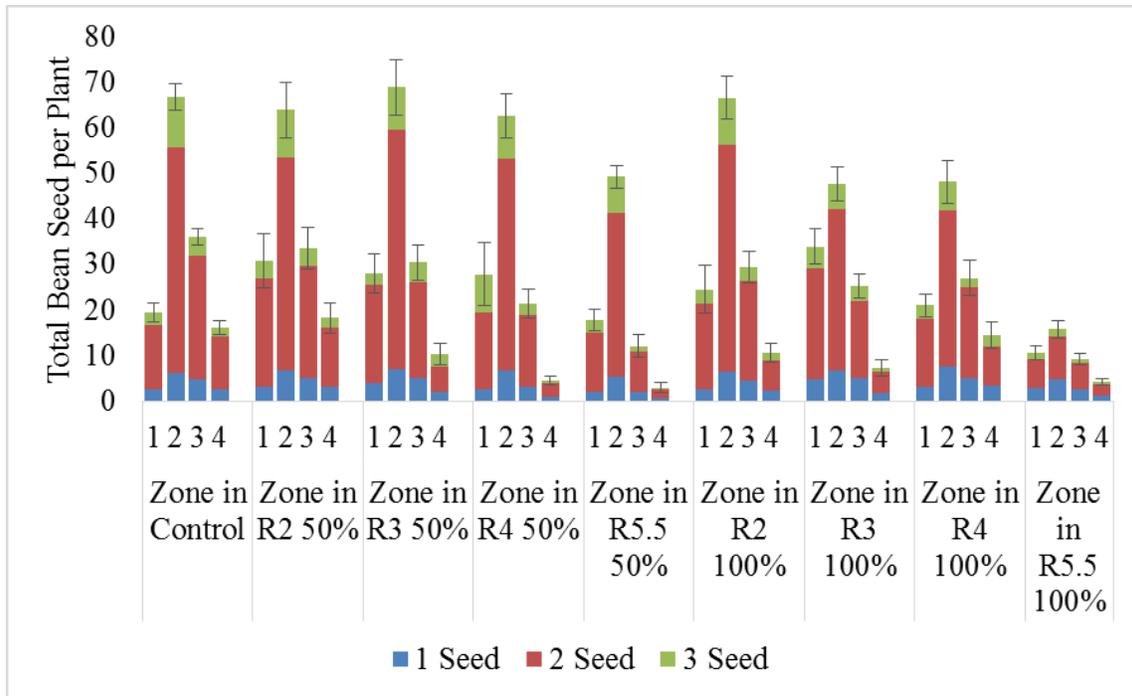


Figure 5.5 Contribution of pods with 1, 2, and 3 seed to total beans present in each zone of each growth stage by removal level treatment in determinate soybean from field plots in Mississippi in 2012-2013.

Standard errors presented are the standard errors for total beans for each zone.

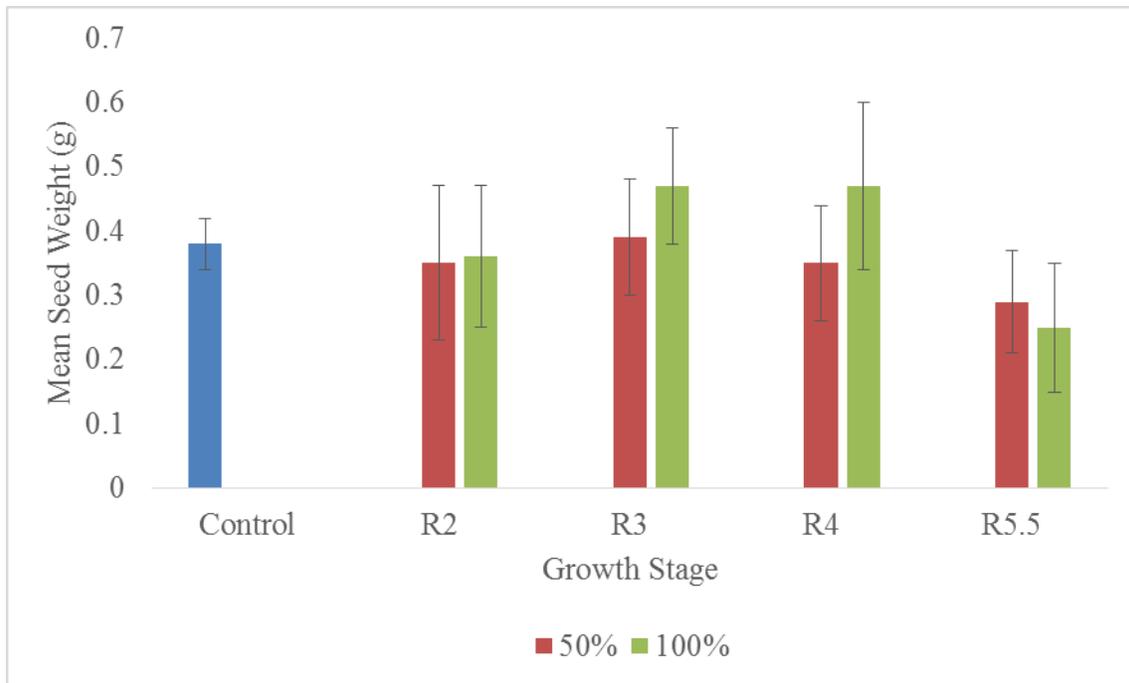


Figure 5.6 Interaction effect of fruit removal timing and level on mean seed weight across all zones on mean individual seed weight of determinate soybean from field plots in Mississippi in 2012-2013.

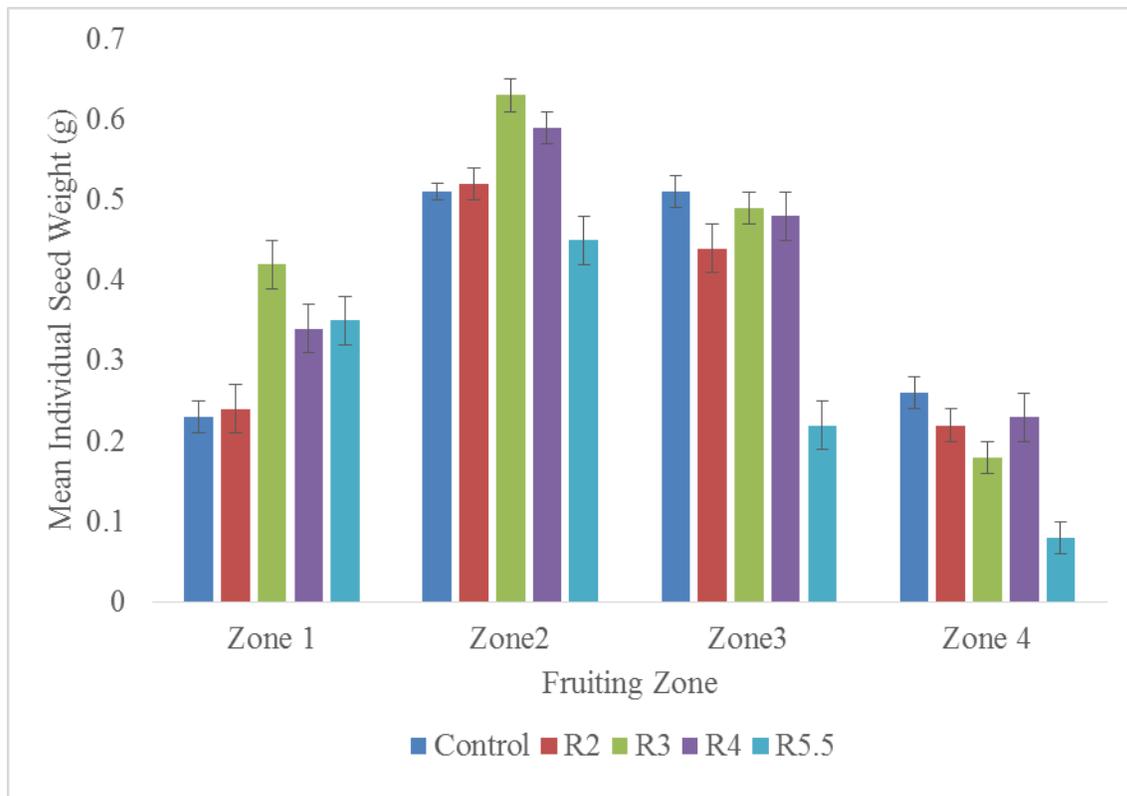


Figure 5.7 Interaction effect of fruit removal timing and zone on mean individual seed weight of determinate soybean from field plots in Mississippi in 2012-2013.

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CHAPTER VI

EVALUATION OF CORN EARWORM, *HELICOVERPA ZEA*, ECONOMIC INJURY LEVELS IN MID-SOUTH REPRODUCTIVE STAGE SOYBEAN

Abstract

Field experiments were conducted in Starkville and Stoneville, MS, Marianna, AR, Winnsboro, LA, and Jackson, TN during 2012 and 2014 to evaluate the relationship of corn earworm, *Helicoverpa zea* (Boddie), larval density and yield and the relationship between the percentage of damaged pods and yield in Midsouth soybean systems. Corn earworm moths were infested into field cages at R2 for 5-11 days to achieve a range of larvae within each plot. Larval density was estimated at 14 days after infestation. Total pods and damaged pods were determined at 19 days after infestation to obtain the percentage of damaged pods. Plots were harvested at the end of each growing season and yield recorded. Data were subjected to regression analysis and the relationship between larval density and yield and the relationship between the percentage of damaged pods and yield both resulted in a linear relationship. Each increase of one larvae per row-m resulted in a yield loss of 45.4 kg/ha. Similarly, each increase of 1% damaged pods resulted in a yield loss of 29.4 kg/ha. From these data, economic injury levels were developed for a range of crop values and control costs. These data suggest corn earworm thresholds for Mid-South soybean need to be reduced.

Introduction

Corn earworm, *Helicoverpa zea* (Boddie), is often the most economically important insect pest of soybean, *Glycine max* (Merrill), in Mississippi (Musser et al. 2013). Corn earworm in Mississippi usually undergoes 5 generations per year. The first generation typically occurs on wild hosts such as crimson clover, *Trifolium incarnatum* L., with the subsequent generation moving into corn, *Zea mays mays* L. Host preference of corn earworm is positively correlated to plant maturity and corn earworm strongly prefers plants in the flowering stage with corn being the most suitable of all hosts (Johnson et al. 1975). Once corn begins to senesce, it becomes unattractive to corn earworm adults as an ovipositional host. The third and fourth generations generally occur in other agronomic host crops such as soybean, cotton, *Gossypium hirsutum* L., and grain sorghum, *Sorghum bicolor* [L.] Moench, with the fifth generation occurring primarily on volunteer crop plants after harvest and other non-crop wild hosts (Hartstack et al. 1973).

In 2011, corn was worth an average price of \$0.05/kg (\$6.00/bu). With this recent increase in commodity prices, corn production in Mississippi has increased over the past several years (NASS 2014b). This increase in production of the preferred host of corn earworm may contribute to larger populations of the subsequent generations of corn earworm leading to more severe infestations in soybean and cotton. Historically, cotton has been a major sink for corn earworm infestations following the generation in field corn. There has also been a major decrease in cotton production and subsequent increase in soybean production (NASS 2014b). This production shift probably contributed to the higher infestation levels of corn earworm in soybean experienced during 2010 to 2013. Corn earworm became the most costly insect pest of soybean in Mississippi in 2010,

surpassing the soybean looper, *Chrysodeixis includens* (Walker), and the stink bug complex and has remained as such through the present time (Musser et al. 2011, Musser et al. 2013).

Early maturing soybean varieties (maturity group IV) have been a standard recommendation when planting during late March through late April for many years (Heatherly et al. 1999). Current maturity group IV and several maturity group V soybean varieties exhibit an indeterminate growth characteristic compared to the determinate characteristic exhibited by later maturity groups including some maturity group V varieties, as well as, maturity groups VI, VII, and VIII varieties. Early planting of soybean has been recommended as an IPM strategy to allow soybean to avoid damaging infestations of corn earworm (Jackson et al. 2008). With the increase in soybean production, planting now ranges from late March through mid-May for early planted full season soybean and late May through the middle of June for soybean double cropped following wheat, as well as planting soybean later to manage harvest of soybean and other crops (Heatherly et al. 1999). Changes in production systems, such as extending the planting window, may result in a higher proportion of the soybean crop at risk to significant infestations of late season insect pests such as corn earworm.

Most of the research on *Heliothine* economic injury levels and economic thresholds in soybean has been performed on later maturing cultivars (maturity group VI-VII) displaying a determinate growth habit. (Smith and Bass 1972b, Mueller and Engroff 1980, McWilliams 1984, McPherson and Moss 1989). There is currently little information on the impact of corn earworm infestations on the most commonly used earlier maturing varieties or cultivars with an indeterminate growth habit. This study

investigates the relationship between corn earworm larval density, damage, and yield in current Mid-Southern soybean production systems. The goal of this study was to validate or revise the current economic injury levels and their associated action thresholds for corn earworm larval densities in Mid-Southern soybean. The current action threshold for corn earworm in soybean is 10 corn earworm larvae per row-m (3/ row-ft) in Mississippi, Louisiana, and Tennessee (Beuzelin et al. 2014, Catchot et al. 2014, Stewart and McClure 2014). The action threshold for corn earworm in Arkansas is lower, recommending control measures be taken when 3.3 larvae at least 1.27 cm (0.5 in) in size are found per row-m (1/ row-ft) (Davis et al. 2014). In addition to revising or validating action thresholds based on larval numbers, this study attempts to propose an economic injury level and action threshold based upon pod damage from corn earworm expressed as percent of total pods damaged.

Materials and Methods

To determine the relationship between corn earworm larval density and yield, and between damaged pods and yield in current soybean production systems, experiments were performed in Starkville, MS at the R.R. Foil Plant and Soil Sciences Farm; in Stoneville, MS at the Delta Research and Extension Center; in Jackson, TN at the West Tennessee Research and Education Center; in Winnsboro, LA at the Macon Ridge Research Station; and in Marianna, AR at Lon Mann Cotton Research Station during 2012, 2013, and 2014.

The experimental design was a randomized complete block at all locations. The experiment included four replications at both Mississippi locations, as well as, the Louisiana location. The Arkansas and Tennessee locations each had three replications

for each year. Soybeans were planted at 296,532 seed/ha into raised conventional till beds with a 0.76 m row spacing in Jackson, TN, a 0.97 m row spacing in Starkville, MS, Marianna, AR, and Winnsboro, LA, and a 1.02 m row spacing in Stoneville, MS. Seed were treated with a commercial premix of imidacloprid, pyraclostrobin, metalaxyl, and fluxapyroxad (Acceleron®, Monsanto Company, St. Louis, MO) to minimize the impact of early season insect pests and seedling disease. Plots were maintained weed free in accordance with each state's extension service recommendations. An indeterminate maturity group (MG) IV soybean (Asgrow 4605®, Monsanto Company, St. Louis, Mo) cultivar was used in all plots in 2012 and 2013. Due to this cultivar no longer being commercially available, during 2014 a similar MG IV cultivar (Asgrow 4632®, Monsanto Company, St. Louis, Mo) was used.

Plots (2 rows x 1.8 m) were established and enclosed within field cages (1.8 m x 1.8 m x 1.8 m) when soybeans reached the R1 growth stage (Fehr and Caviness 1977). When plants within the cages reached R2, ten to twenty newly emerged corn earworm adults of each sex were infested into field cages containing the soybean cultivar. This infestation timing coincides with the period of physiological growth when the majority of corn earworm oviposition typically occurs under normal conditions in production fields in the Mid-South. Insects used in this study were no more than two generations removed from a wild collected parent colony. The study consisted of five infestation durations in order to achieve a range of larval densities. To achieve this larval density range, defined periods of adult oviposition were assigned to infested plots. These periods included 5, 7, 9, and 11 days. A non-infested control was also included.

Three days prior to adult infestation, all cages were treated with 0.56 kg-ai/ha of acephate to eliminate predators and other pest insects. Once soybeans reached the R2 growth stage, corn earworm adults were infested in designated cages. Moths were removed by hand at the designated removal time for each plot. One row of each cage was designated for sampling purposes while both rows were harvested for yield measurements. At 3 days after the final adult removal (14 days after infestation), larval density was estimated by sampling a portion (0.76 row-m) of one row with a 0.76 row-m drop cloth. On the 19th day after infestation, the total number of pods and total number of damaged pods were determined by sampling a portion (0.97 m) of the row that was previously sampled for larvae. The plants that were sampled for number of pods and damaged pods were not in the part of the row that was previously sampled for larvae. The larval sample represents an estimate of larval density when larvae would typically be first detected and management decisions made by consultants or producers. This left the remaining row undisturbed. Cages remained on all plots until the last sample was collected. Once plots were sampled for damaged pods, plots were maintained insect free with insecticides in order to eliminate confounding insect damage from other species or further infestations of corn earworm. At the end of the season, both rows were harvested with a Kincaid 8-XP plot combine and weights and moisture were recorded. All plot weights were corrected to 13% moisture.

The experiment at Winnsboro, LA during 2012 was lost to Hurricane Isaac. Experiments were attempted at all sites during 2013, however attempts to establish larval infestations failed at all locations. Additionally, 2/3 of the plots were lost at the Tennessee location during 2014 due to a high wind event, so data from the remaining

plots were not included in the analysis. Also larval infestations failed to establish at the Starkville, MS location during 2014, leaving us with 7 successful experiments out of 15 attempts during 2012 to 2014. Prior to analysis, the number of damaged pods in each plot was divided by the number of total pods for the same plot in order to obtain a total percentage of pods damaged by corn earworm. Data were analyzed with regression analysis in Proc Glimmix in SAS (PROC GLIMMIX SAS Institute Inc. 2011) to determine the relationship between larval density and yield and the relationship between the percentage of damaged fruit and yield. All data were examined for outliers. Outliers were determined by plotting residual and predicted values. Based on a normal distribution, any plots with residual values more than three standard deviations from the predicted value were removed from analysis. Initial analysis revealed multiple extreme outliers from the Arkansas location during 2012, possibly due to harvesting errors. Therefore, data from this location during 2012 was excluded from further analysis leaving a total of six site years (combination of year and location) for the final analysis. Additionally, nine plots at the Stoneville, MS location during 2014 had larval densities in excess of 125 larvae per 0.76 row-m. Initial analysis revealed that once larval numbers increased beyond 100 per 0.76 row-m, no further decrease in yield was observed. All nine plots incurred 100% pod damage. At this point, the ecological system was likely overwhelmed and no further damage/loss could be incurred from additional larvae. Because densities in excess of 100 larvae per 0.76 row-m were much greater than what is needed to cause economic damage, these plots were excluded from final analysis.

Preliminary analysis of the interactions between larval densities with site year were not significant ($F= 2.04$; $df=5, 69.29$; $P=0.09$). Preliminary analysis also did not

result in a significant interaction between percent damaged pods and site year ($F= 2.16$; $df=5, 73.86$; $P=0.08$). Therefore, site year and replication nested within site year were designated as random effects for all analyses allowing for inferences to be made over a range of environments (Carmer et al. 1989, Blouin et al. 2011).

Data from this research were used in the determination of economic injury levels for indeterminate soybean based on larval densities and percent corn earworm damaged pods. The yield loss equations for larval density and for the percentage of damaged pods were used to calculate the number of larvae present and percent of damaged pods needed to equal the cost of controlling the pest in two separate EIL equations which are represented by C in the formula $EIL = C/VIDK$ (Pedigo et al. 1986) where V is the crop value expressed in U.S. dollars per hectare, ID is based on the amount of yield loss from the percentage of pods damaged by corn earworm or for given larval densities, and K is the expected percent of control from the application of a control tactic. For the purposes of developing this economic injury level, control was assumed to be 100% as it is not specific to a particular control tactic (Owen et al. 2013). Scenarios given for EILs are based on a range of crop values and yields as this research was conducted in varying environments with respect to yield potential. Costs of control are based on total cost including the current value of an insecticide plus application costs.

Results and Discussion

Mean yield of non-infested plots was 3947 kg/ha ranging from 967 to 7498 kg/ha. Yields of infested plots ranged from 215 to 6513 kg/ha. Larval densities in infested plots ranged from 0 to 112 corn earworm larvae per row-m. Damaged pods in infested plots ranged from 0 to 1099 per row-m. Total pods ranged from 11 to 1825 per row-m among

all plots. Percent damaged pods in infested plots ranged from 0 to 99%. When data were analyzed across site years, there was a significant negative linear relationship between percentage of corn earworm damaged pods and yield ($F= 52.11$; $df=1, 78.16$; $P\leq 0.01$). There was a yield loss of 24.9 ± 3.4 kg/ha for each increase of one percent damaged pods (Fig. 6.1). Similarly, analysis of larval density data revealed a significant negative linear relationship with yield ($F= 52.6$; $df=1, 78.72$; $P\leq 0.01$). There was a yield loss of 26.3 ± 3.6 kg/ha for each increase of one corn earworm larvae per row-m (Fig. 6.2).

Results from previous studies using determinate later maturing cultivars have been variable. Smith and Bass (1972) estimated that the economic threshold for corn earworm infesting soybean during the R3 to R4 growth stages was 3.3 to 10 larvae per row-m based on the commodity and control tactic prices of the time. Mueller and Engroff (1980) observed no yield reductions from corn earworm infestations up to 65 larvae per row-m during the reproductive period regardless of growth stage. McWilliams (1984) infested tobacco budworm, *Heliothis virescens* (F.), eggs at R2 which resulted in fifth instar larvae at R5 during 1977. No yield reductions were observed at larval densities of 5 per row-m (lowest level) or greater. During 1978, infestations of fifth instar larvae at R4 did not result in yield reductions with one variety at densities of 21 larvae per row-m or less. A higher infestation level (36.7 per row-m) significantly reduced yield. With another variety used in the study, larval densities of 19.3 per row-m (lowest infestation level) or greater significantly reduced yield. Though *H. virescens* and *H. zea* are different species, the biology of the two species and the manner in which they infest fields and subsequently consume fruit is similar. McPherson and Moss (1989) observed no yield reductions at infestation levels up to 5.2 larvae per row-m at R5. Eckel

et al. (1992) observed no yield reductions at infestation densities of 4.7 large larvae (fourth to sixth instar) per row-m or lower at one location during 1983. However, during 1984 infestation levels as low as 2 large larvae per row-m significantly reduced yield. At another location, densities of large larvae ≥ 31.7 large larvae per row-m significantly reduced yield during 1983 and 1984. The next lowest infestation level was 0.3 and 0 larvae per row-m, respectively, and these levels had no significant impact on yield. Results from our study are similar to the previous studies in some cases and very different in others. Corn earworm typically infest indeterminate soybean in the Mid-South during the R1 to R2 growth stages which results in large larvae during the R4 to R5 growth stages. Infestations infrequently occur during other growth stages. One possible reason for differences in the results between the current study and previous ones is that earlier maturing varieties may respond differently than later maturing varieties to corn earworm injury. In manual simulated damage studies (Adams et al. 2015b, Adams et al. 2015a) yield responses to fruiting structure removal was similar between an indeterminate and a determinate cultivar at R4. However, responses between the cultivars to the lower level of damage imposed (50% pod removal) at R5.5 were different, indicating that determinate cultivars may be more tolerant to fruit loss during the later reproductive period than indeterminate cultivars.

Based on the relationships between larval density and yield, economic injury levels were calculated for a range of crop values and control costs (Table 6.1). Currently, the cost of a diamide insecticide, which is the most common chemistry used in the Mid-South to control lepidopteran pests, plus the application cost, is approximately \$50 per hectare (\approx \$20/ac). Assuming a soybean market price of \$0.40/kg (\approx \$11/bu) and an

insecticide application cost of \$50/ha, it would only require protection of 125 kg (1.9 bu/ac) of yield per hectare to equal the cost of control. The resulting EIL is 4.7 larvae per row-m. Based upon these findings, and given the nature of infestations intensifying quickly, the economic threshold for larval densities should be set between 25-50% lower than the given EIL depending on but not limited to scouting intervals, logistical considerations for control measures, etc. Similarly EILs were calculated based on percent pod damage (Table 6.2). Based on the same prices of \$0.40/kg for soybean and \$50/ha for insecticide, the resulting economic injury level would be 5% pod damage. Similar to the results of this study, in a study examining the impacts of simulated corn earworm damage, Adams et al. (2015b) found that the preliminary range for an EIL based upon plant damage at the R4 and R5.5 growth ranged from 5-20% (the time at which large larvae were present in the current study) which would further validate the findings of this study using actual corn earworm larvae.

Most Mid-South states currently have an action threshold published for corn earworm larval densities on a per row-m basis for a drop cloth. However, across the Mid-South the preferred method for agricultural consultants who work in soybean to monitor insect populations is using a sweep net. Due to the nature of our study being conducted in small field cages, using the sweep net was not a valid sampling method. Previous comparisons of drop cloth and sweep net sampling methods have shown that 9.84 larvae per row-m on a drop cloth is equivalent to 12-24 larvae collected in 25 sweeps with a sweep net (38cm diameter) (Hillhouse and Pitre 1974, Rudd and Jensen 1977). More recent data (Stewart 2013, unpublished data) suggests that 3.8 larvae per row-m on a drop cloth is equivalent to catching 9 larvae in 25 sweeps. Using the most

conservative sweep net estimate (Stewart 2013 conversion), this would mean that our previously mentioned EIL of 4.7 larvae per row-m is equivalent to 11.1 larvae per 25 sweeps. This EIL is only slightly above currently recommended thresholds.

Currently in Mississippi, action thresholds in reproductive stage soybean for defoliating insects such as soybean looper, *Chrysodeixis includens* Walker, utilize a combination of 20% defoliation and 19 loopers per 25 sweeps with a sweep net. The possibility exists moving forward to adopt a similar system for corn earworm action thresholds that utilizes the EIL and associated action threshold for the percentage of damaged pods in conjunction with corn earworm larval density EIL and action thresholds set forth in this study at a given commodity price and control cost. This system would assume that damage has reached the specified threshold and there is an active infestation of corn earworm larvae that would maintain at levels at or above threshold or thought to be increasing.

This study was conducted over a range of environments as evidenced by the large variation in yields among the non-infested plots. Sites were irrigated as needed, with the exception of the Tennessee location. Weather conditions during 2012 were typical of Mid-Southern sub-tropical summers with warm weather and average rainfall experienced across the entire region. During 2014, the growing season was much more temperate in nature with high temperatures being below the 10 year average for most of the growing season and experiencing above average rainfall amounts for the period. It is possible that if this study was conducted in different environments such, the response may be different than from what was observed in our study.

Indeterminate maturity group IV cultivars represent the majority of soybean production in the Mid-South. Over the course of our study, we attempted to choose two of the most popularly utilized cultivars for our study to be as representative as possible of the entire area. While we acknowledge that the use of more varieties would have been better, the logistics and costs of cage materials in addition to the repeated failures to establish larvae did not allow for using more than one cultivar in a given year. We acknowledge that there could potentially be differences in responses among cultivars of the same maturity and growth habit, however that information is not known for indeterminate maturity group IV soybeans at this time.

As previously mentioned, current action thresholds in Mississippi, Tennessee, and Louisiana are 10 corn earworm larvae per row-m while Arkansas's current action threshold is 3.3 corn earworm larvae per row-m. The findings of this study list EIL's based on a range of crop values and control costs and the associated thresholds should be set below the EIL (25-50%) for the given situation. The authors believe this method would be the most effective method of determining when to take action against corn earworm in indeterminate soybean in order to maximize a producer's return on investment when making an application of an insecticide to control corn earworm. In the event producers would not have this scale of EIL's in front of them, the need for a general action threshold may be necessary. Based upon the findings of this study, the authors recommend an action threshold of 3.5 corn earworm larvae per row-m in indeterminate soybean in the Mid-South in the event producers or consultants choose not to use the scale provided in Table 1 when making pest management decisions.

Table 6.1 Economic injury levels for corn earworm larval densities in indeterminate soybean at various control costs and crop values.

Crop Value (\$/kg)	Cost of Control (\$/ha)				
	\$15	\$30	\$45	\$60	\$75
	Economic Injury Level (Larvae/row-m)				
\$0.60	0.9	1.8	2.9	3.8	4.7
\$0.50	1.2	2.2	3.4	4.6	5.7
\$0.40	1.4	2.9	4.3	5.7	7.1
\$0.30	1.8	3.8	5.7	7.6	9.5
\$0.20	2.9	5.7	8.5	11.4	14.3

Table 6.2 Economic injury levels for percent of damaged pods in indeterminate soybean at various control costs and crop values.

Crop Value (\$/kg)	Cost of Control (\$/ha)				
	\$15	\$30	\$45	\$60	\$75
	Economic Injury Level (Percent Damaged Pods)				
\$0.60	1	2	3	4	5
\$0.50	1.2	2.4	3.6	4.8	6
\$0.40	1.5	3	4.5	6	7.5
\$0.30	2	4	6	8	10
\$0.20	3	6	9	12	15.1

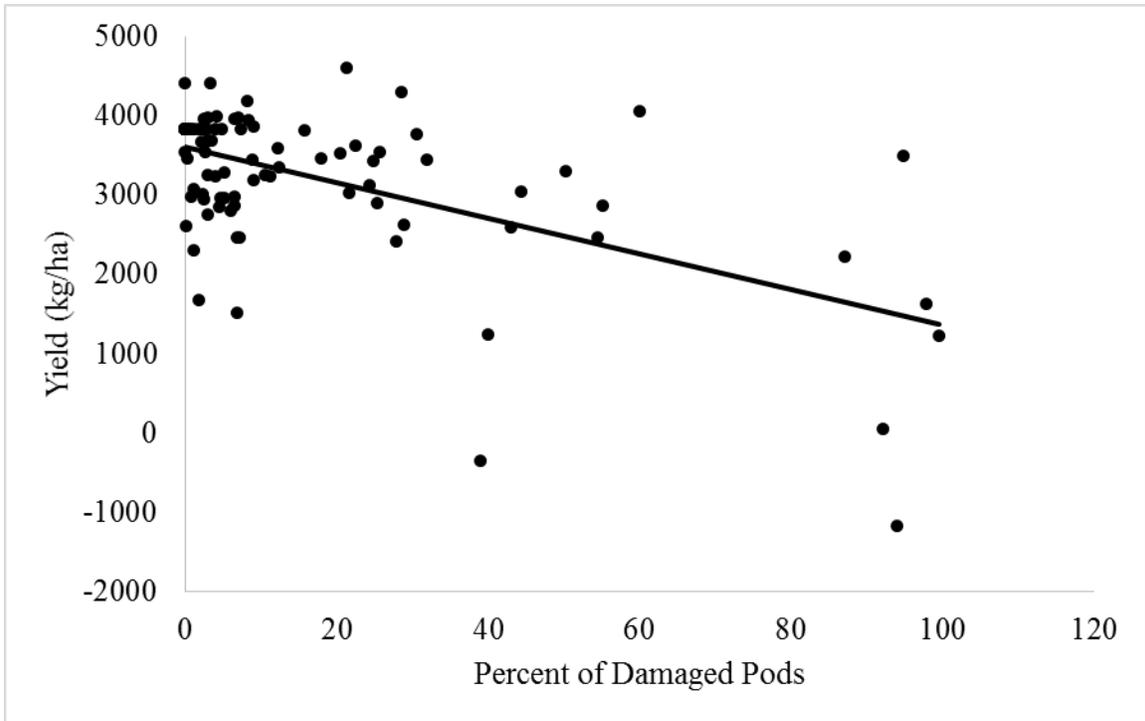


Figure 6.1 The relationship of yield and percentage of damaged pods from corn earworm infested field cage trials in the Mid-South during 2012-2014.

Yield is expressed in $\text{kg/ha} = 3779 - 24.9x$ where x is equal to percent of damaged pods. For the graph, yield of all plots in each replication for each site year was adjusted so that the non-infested control yield was equal to the overall mean yield of non-infested controls

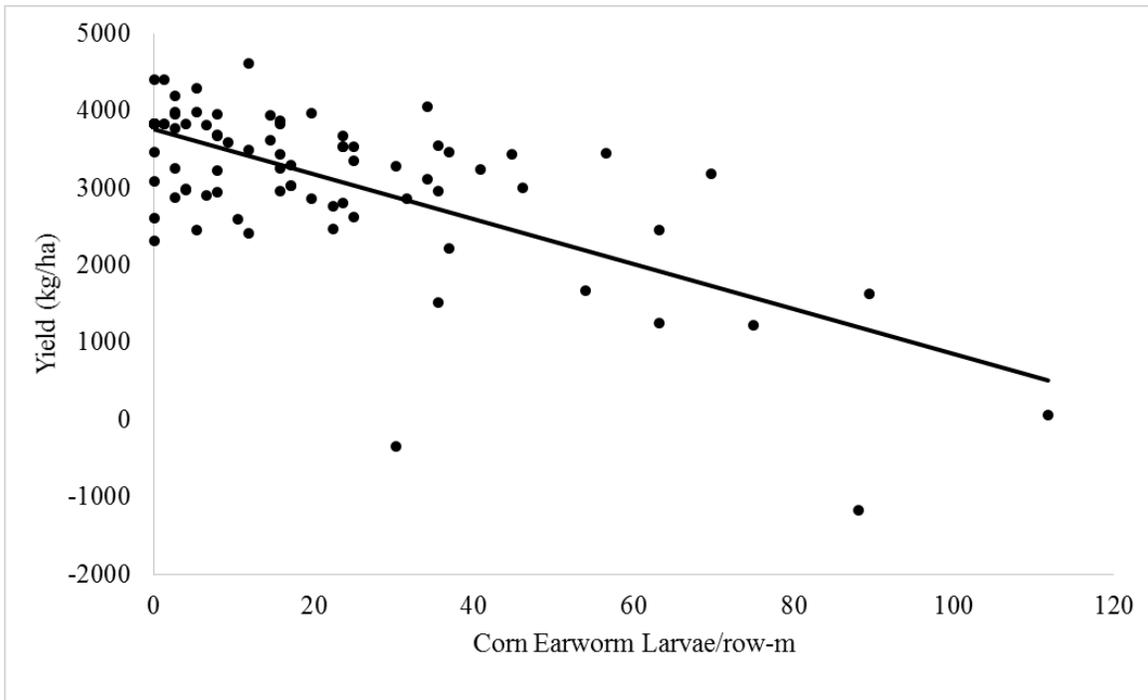


Figure 6.2 The relationship of yield and corn earworm larval density from infested field cage trials in the Mid-South during 2012-2014.

Yield is expressed in $\text{kg/ha} = 3830 - 26.3x$ where x is equal to corn earworm larvae/row-m. For the graph, yield of all plots in each replication for each site year was adjusted so that the non-infested control yield was equal to the overall mean yield of non-infested controls

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