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# Effects of Cultural Practices on Soybean Nectar Production

Tyler Miles Smith

Effects of cultural practices on soybean nectar production

By

Tyler Miles Smith

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By

Tyler Miles Smith

Approved:

Angus L. Catchot Jr.
(Co-Major Professor)

Jeffrey W. Harris
(Co-Major Professor)

Jeffrey Gore
(Committee Member)

Donald R. Cook
(Committee Member)

Natraj Krishnan
(Committee Member)

Kenneth O. Willeford
(Graduate Coordinator)

George M. Hopper
Dean
College of Agriculture and Life Sciences

Name: Tyler Miles Smith

Date of Degree: August 10, 2018

Institution: Mississippi State University

Major Field: Agricultural Life Science

Major Professors: Dr. Angus Catchot and Dr. Jeff Harris

Title of Study: Effects of cultural practices on soybean nectar production

Pages in Study 58

Candidate for Degree of Master of Science

Experiments were conducted to determine the effects of cultural practices such as irrigation, planting date, and maturity group on soybean nectar production. Multiple interactions between different factors influences the amount of total sugar present in soybean flowers. Pod production was increased in later planted soybean, but later planting resulted in reduction in yield. Another study was conducted to explore the effects that variety selection had on soybean nectar production. Soybean nectar production was highly variable among varieties and was influenced further depending on their growing location. Later maturing varieties produced significantly more soybean pods. This increase in pod production was not reflected in yield, as the later maturing varieties usually yielded less.

# DEDICATION

I would like to dedicate this research to my family. First, my parents Dale and Kim Smith. Words cannot express how much your support and guidance have helped me throughout all of my life. Your steady support in my life and work is more than I can ever repay. Next my sister Kristina, her husband Justin, and my niece Olivia Christian. Thank you for always allowing me in your home and for being by my side when things were tough. Next to my brother Jacob, thanks for always being someone that I can talk to about the struggles that we come across every day and for allowing me to hopefully be a big influence in your life. Lastly, I would like to dedicate this research to my grandparents Tawanda and the late Buddy Smith and Juanita and the late Richard Bussey. Thank you for your unwavering support as I complete the next chapter in my life.

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# CHAPTER I

# INTRODUCTION

## Soybean in Mississippi

Soybean, *Glycine max* (L.) Merr., production in Mississippi has changed substantially over the years. In the past, primary production systems involved planting maturity groups VI and VII. Early soybean production systems have been adopted in order to guard against drought stresses and insect infestations commonly experienced during July and August (Baur et al. 2000). Early maturing soybean cultivars (maturity group IV and V) are planted in early- to mid-April. Management decisions vary depending on factors such as location or issues that may occur during the growing year, such as weather, soil type, and insect infestations. Soybean is number three behind poultry and forestry in terms of value of agricultural commodities produced in Mississippi (MSUCares 2017). Today, the majority of the soybean acreage in Mississippi is planted in maturity groups IV and V (MSUCares 2017). In the 1980s and 1990s, state average yields were ca. 1345 to 1681 kg per hectare. During the 2014 and 2015 growing season, record yields of 3497 and 3093 kg per hectare were achieved. The total soybean production area for Mississippi in 2016 was 825,558 hectares averaging 3,195 kg/ha (NASS 2017).

## Soybean Biology

Soybean is an annual legume and is considered a short day plant. Most soybean cultivars initiate flowering during long nights (Borthwick and Parker 1938). Soybean cultivars are classified by thirteen maturity groups with maturity group 000 being the earliest cultivars and requiring short nights for development and maturity group X cultivars needing long nights for development. Maturity Group zones represent defined areas where varieties of certain maturities are best adapted; however, this does not imply that a certain variety from the Maturity Group can only be grown in that designated area (Heatherly and Elmore 2004). The early soybean production system used in the Mid-South is an example of using cultivars outside their assigned Maturity Group region.

Soybean varieties are also grouped into categories based on growth habit: determinate and indeterminate. Determinate varieties initiate flowering at all positions on the plant simultaneously. Soon after flowering begins, vegetative growth will cease. Indeterminate varieties begin flowering at the lowest node continue vegetative growth after flowering has initiated (Hodges and French 1985).

Karlton, Weber, and Eldredge (1949) created a development system for classifying growth stages of indeterminate soybean cultivars in order to assess hail damage. Although this system was adequate for indeterminate varieties, it was not satisfactory for determinate cultivars. Fehr et al. (1971) developed descriptions for stage of development for all genotypes of soybean. Once soybean seed are planted, emergence or VE, occurs in approximately 5 to 10 days. During emergence, cotyledons fully unroll, and unifoliate leaves develop, which is referred as cotyledon stage or VC. After the unifoliate leaves develop, all subsequent leaves are trifoliate. The remaining stages of vegetative growth are identified by the number of trifoliate leaves to the $n^{th}$ node, Vn (Fehr et al. 1971). There are eight reproductive growth stages of soybean, described as R1 to R8 (Fehr et al. 1971). R1, or beginning bloom, is identified when one open flower can be seen at any node on the soybean plant. The soybean plant reaches R2 when there are open flowers at one of the two uppermost nodes. The initiation of one 0.95 cm (3/16") pod in one of the uppermost four nodes is termed R3 or beginning pod. Pod development continues in the uppermost four nodes, and when one pod reaches 1.9 cm (3/4") in length it is designated at R4. This is the end of pod formation and the beginning of seed fill (Pedersen 2004). R5 is identified when a 0.3 cm (1/8") seed is present in a pod at one of the four uppermost nodes. Seed continue to grow from R5 and fill the inside of pods. R6 is defined as full seed, and a green seed fills the pod cavity in one of the four upper most nodes (Pedersen 2004). After pod fill, the soybean plants start to reach physiological maturity. When one mature pod is present anywhere on the plant it is termed R7. Soybeans have reached R8 growth stage when 95% of all pods on the plant have reached a mature color and have reached full physiological maturity (Pedersen 2004). Soybean plants are ready for harvest once R8 growth stage has been reached.

### Soybean Flower

Soybean flowers are variable in size with some being long and narrow, while others are short and broad. It is made up of five petals and can be purple to white in color. A definite nectar guide consisting of a series of lines converging at the entrance to the tongue channel is visible in soybean varieties that have purple flowers. That same guide is not present in varieties with white-colored flowers (Erickson 1975c). Two small, tightly clasped ventral keel petals partially enclose the sexual column. The stamens are diadelphous, with nine stamens fused in a common sheath and a single tenth stamen attached dorsally at the base of the standard petal. The soybean floral nectary surrounds the carpel base and is in turn surrounded by the staminal sheath. Soybean nectaries are discoidal, and similar to others in Fabaceae (Waddle and Lerstein 1973). Although soybean nectariferious tissue has a high proportion of phloem in the nectaries, the nectar that is produced has a high sugar content, upwards of 43 ± 15% (Erickson 1975a, 1975b, 1975c).

Flowering tendencies can be highly variable with anywhere from eight to thirty-five blossoms occurring in each cluster depending on the variety (Erickson 1975b). Soybean flowers first open at the base of the raceme then progressively upwards. Severson (1983) observed that each soybean flower is only open for a day, but from one to thirteen different flowers may be open simultaneously on a raceme depending on the cultivar (Erickson 1975a). If the leaf canopy is moved aside and several flowers are open simultaneously on some cultivars, they can appear quite showy. The number of flowers produced per hectare is highly variable among cultivars with anywhere from 1.3 to 4.1 million flowers per hectare per day (Erickson 1984a). Soybean does not compete well with other common legumes for the attention of bees. Other legumes, such as *Trifolium repens* and *Medicago sativa*, can produce upwards of ten times more flowers per hectare and greater number of flowers per cluster (Erickson 1984a).

Soybean flowers are reported to have functional nectaries, meaning that pollinators can visit the flowers and obtain the food reward (Erickson and Garment 1979). Sugar concentrations in soybean nectar are 5-10% higher than those of other legumes, such as alfalfa, when growing conditions are favorable (Erickson 1975c; Severson 1983). Soybean nectar production and honey bee visitation occur between 0900 and 1500 hr. The peak nectar flow can be variable depending on the variety and local weather condition. Soybean nectar volume per flower varies significantly among varieties, but is greatest in warmer conditions (Erickson 1975c). Erickson (1984a) observed that plants required up to three days to recover the ability to produce nectar, following a period of cold weather. Many have reported mean nectar sugar contents ranging from 37% to 45% depending on the variety sampled (Erickson 1975bc; Jaycox 1970). Floral sugar concentrations increased as nectar volume decreased with time of day and temperature due to evaporation. Sugar ratios (glucose:fructose:sucrose) differ between varieties, as well as with time of day within a cultivar. Severson (1983) reported that while glucose and fructose fluctuate mildly during the day and between cultivars, sucrose can be anywhere from the smallest available sugar to the most predominant sugar available. According to work done by Severson (1983), flower color does not impact attractiveness and sugar content among different varieties.

## Honey Bee

The honey bee ,*Apis mellifera* (L.), is a member of the family Apidae and order Hymenoptera. Although originally native to Europe, Asia, and Africa, honey bees are distributed all around the world due to the movement of the bees by beekeepers (Winston 1991). In many parts of the world, especially Europe, modern beekeeping is only a few centuries old with man taking sections of trees with established colonies and moving them closer to human swellings (Seeley 1985). Prior to these advances, man was more of a honey hunter that harvested honey from honey bees in natural cavities. In 1851, Lorenzo Langstroth was credited for developing the first hive with combs that can be removed, and this advancement is the foundation for modern beekeeping (Naile 1976). Although Langstroth is credited with this achievement, Petro Prokopovych was the first to create removable comb hives back in 1814.

At the center of every honey bee colony is a long-lived female, the queen, who is the mother to roughly thirty thousand offspring in a colony. Approximately 95 percent of a queen's offspring are workers, or females that never mate and carry out the day to day operations in a colony. The other 5 percent are sexual males, or drones, and other female queens (Jay 1968). Queens can control the sex of the offspring by a very elementary method: males arise from unfertilized eggs; whereas, females from fertilized eggs (Kerr 1969, Crozier 1977).

Whether a fertilized egg develops into a worker or queen is dependent on the composition of its food during the first three days of development of the larvae. The difference in concentrations of sugar in royal jelly and worker jelly, 35 percent and 10 percent, respectively, has a profound effect in the development of the larval bee. Beetsma (1979, 1982) concluded that worker jelly that is experimentally fortified with amounts of glucose and fructose caused queens to be developed. He notes that the sweetness difference triggers different larval feeding rates, different levels of juvenile hormone, and ultimately different developmental programs for the two types of female bees.

The division of workers into labor categories is central to the social organization of honey bees. Separating bees into labor specialists, combined with a high level of communication, gives a colony the ability to work much more efficiently than if it was comprised of a loose association of identical individuals. The basis for this principle in honey bees is age. During the six weeks of a summer bee's life, day to day duties within the colony vary with age, and is driven by hormones, such as juvenile hormone. These tasks start with cell cleaning, progressing into brood care and food storage, and then ending with foraging (Seeley 1985). The first three weeks are inside the hive; the last three weeks are spent foraging outside the hive. This typical time-course of worker tasks is plastic; individuals can either develop faster than normal or develop in retrograde to previous age-related activities if the colony needs them to do so.

Foraging by honey bees is a collective effort in which foragers from the colony work closely together to identify and exploit rich sources of nectar and pollen. The key to success with honey bees is that foraging is designed to achieve a high efficiency for the colony as whole and not just for the individual. Most foraging bees follow recruitment dances to know which food source to exploit next. A small minority of bees known as "scout bees", will fly out independently in order to search for new food source (Lindauer 1952, Seeley 1983). These bees tend to be older and more experienced foragers. This is an example of a honey bee decreasing their individual foraging success to boost the success of the colony.

There are four major resources that are needed for a honey bee colony to be successful: nectar, pollen, propolis, and water (Seeley 1985). Sugar contained in nectar is the primary energy fuel for bees (after conversion to honey), while pollen is the primary source of amino acids, sterols, minerals, vitamins, campesterol and some minor carbohydrates. Water is collected and used to cool the broodnest through evapoartion during the hot summer months. Water is also used to dilute stored honey for larval feeding. Propolis is used to seal the hive cavity to discourage outside invasions (Seeley 1985). The propolis envelope around the hive also has a role in “social immunity” in the hive, meaning it kills some fungal and bacterial pathogens (Simone-Finstrom and Spivak 2010).

## Soybean and Honey Production

Although soybeans are classified as self-fertilizing and self-pollinating, they can benefit from visitation by pollinators, especially honey bees and some leaf cutting bees (Erickson 1975c). Many species of bees, including honey bees, forage soybeans for nectar and pollen. Erickson (1975c) noted that the density of bees can exceed 1 bee per meter of row during peak foraging times. Many studies have shown that the presence of honey bees may increase yields by 20 percent or more. Erickson (1975a) demonstrated a yield increase of 13.9 percent for the variety ‘Corsoy’ in 1971 and increases of 5.2 percent and 16.9 percent in the variety ‘Hark’ during 1972 and 1973, respectively. In the Mississippi River Delta, soybean yields increased by 21.6 percent for the variety ‘Pickett’ at locations in Arkansas and Missouri. Significant differences in the number of filled pods versus empty pods were also noted. Plots where honey bees were present produced significantly more filled pods. Erickson (1978) attributed these results to increased pod set, since seeds per pod and weight per seed did not vary. Kettle and Taylor (1979) obtained soybean yield increases with honey bees of 5.1 percent, 19.9 percent and 36.0 percent for the variety ‘Forrest’ at three different locations in Kansas. The cultivar ‘Woodworth’, grown at yet another site’ had a yield differential of 10 percent.

Beekeepers around the southern and central United States have obtained excellent yields of honey from soybeans for decades (Erickson 1975a). Over this time, they have identified high yielding agricultural areas that are ripe for honey production. In return, they have also identified areas that are poor and unreliable for honey production. There is also little doubt that some beekeepers may unwittingly harvest larger quantities of soybean honey (Erickson and Robins 1979). Mississippi beekeepers consider soybeans to be an important source for honey production, and a typical yield is 90-130 pounds per colony (J. Harris, personal communication, 2018). Erickson (1975a,b,c) noted that soybeans are not exploited by beekeepers for honey production in other regions, and these could be untapped resources in some areas. Soybean honey has a distinctive aroma and flavor that can be easily identified with training and experience.

Plant husbandry plays a significant role in the production of soybean nectar, and variations among several factors influences yield and quality of honey. The most basic components of nectar are sugars, which are products of photosynthesis. The healthiest plants receiving the best growing conditions, such as, ample light, water, and being grown in the best soil should produce flowers with the highest quality of nectar and aroma, and these are the most attractive to foraging bees (Erickson and Robins 1979; Robacker et al. 1983). Soybean seed yields can be quite sensitive to soil nutrients, pH and moisture as well. Erickson (1984a) noted that soybean yields can be significantly reduced with poor soil pH (6.0-6.5 is considered optimum) and soil nutrient availability, such as nitrogen, potassium, and phosphorus. Moisture stress can also reduce photosynthesis, as well as, flowering and pod filling.

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CHAPTER II

# INFLUENCE OF IRRIGATION, MATURITY GROUP, AND PLANTING DATE ON TOTAL SUGAR PRODUCTION IN SOYBEAN

## Abstract

Field experiments were conducted during 2016 and 2017 at the R. R. Foil Plant Science Research Center in Starkville, MS and the Delta Research and Extension Center in Stoneville, MS to determine the effects of irrigation, maturity group, and planting date on the quantity of total sugar in soybean nectar. Soybean production has shifted to an early production system over the last few decades, and farmers favor earlier planting because soybean growth and yield is vastly improved. Soybean is also a primary nectar source for honey production by commercial beekeepers in the Delta region. Although the effects of cultural shifts in production of soybeans on yield have been well documented, there has been little or no research into how the same practices might affect nectar production. This study examined how common farming practices in different regions of the state affected nectar quality in soybeans over two years. A multi-factorial experimental design tested the effects of region (Delta vs. Hills), soybean maturity group (IV vs. V), planting date (early vs. late), irrigation (with vs. without) and separate years (2016 vs. 2017) on soybean nectar production. Total sugar content was determined from individual flowers that were collected from plants randomly sampled from plots grown

under different treatment combinations. A colorimetric assay based on an anthrone-sulfuric acid reagent was used to determine the amount of total sugar in each flower. Unwieldy variation in response between the two years and the two regions forced separate statistical analyses of the four region by year combinations. There were no significant differences in mean total sugar content per mg of flower weight between the Delta and Hills regions. During 2016 in the Hills, early planted Group IV soybeans produced significantly more mean total sugar. During 2017 in the Hills, early planted plots that were irrigated produced the highest mean total sugar. In the Delta region in 2016, late planted Group IV soybeans that were irrigated produced significantly more mean total sugar. During 2017 in the Delta region, early planted Group IV soybeans produced the highest mean total sugar.

## Introduction

Soybean production occurs across all areas of Mississippi. There are two distinct regions than comprise most of the production in the state, the Hills and the Delta. The Hills region is mostly comprised of smaller cultivatable fields interspersed across the landscape. In 2016, there were 1,143,610 hectares of soybeans planted. Overall, land dedicated to row crop agriculture makes up a small percentage of the overall land area (NASS 2017). In contrast, cultivatable land accounts for a larger percentage of the land area in the Delta region. In 2016, there were 3,544,450 hectares of soybeans planted. Fields dedicated to row crop agriculture are much larger and more contiguous in the Delta region than in the Hills region (NASS 2017).

Plant development, from germination through the onset of flowering and to maturity, is controlled by photoperiod and temperature (Major et al., 1975). How soybean cultivars respond to these abiotic factors determines to which Maturity Group they are categorized. Historical soybean production systems involve the planting of Maturity Group V, VI, and VII varieties in May and June in the midsouthern U.S. This resulted in poor and static yields in the 1970s and 1980s (Heatherly 1999). Drought stress and late season insect infestations drove the poor performance that was commonly seen in this production system.

Mitigation of these problems involved the introduction and implementation of the early soybean production system (ESPS). Soybean can tolerate a relatively wide range of planting dates in both the northern and southern states. The rationale of the early soybean production system is to plant earlier maturing varieties in April and early May to avoid drought stress and late season insect infestations (Heatherly 1999). Planting date affects the size of the plant before floral induction. In most cases, yield will decline rapidly with June and later planting in both the north and south U.S. (Heatherly and Elmore 2004).

When using the ESPS, optimum planting dates for the Southern U.S. are from late March to late April using implementation of the ESPS. Beuerlein (1988) observed a yield reduction in determinate and indeterminate cultivars of 22 kg/ha/day when planting occured after the first of May.

Although little research has been attempted on determining the effects of planting date on nectar secretion, the consensus that healthier plants produce more nectar is a well-known idea (Erickson 1984a). The basic components of nectar are sugars which are products by photosynthesis. The healthiest plants receiving the maximum amount of light and grown under the best possible conditions are likely to be the greatest producer of flowers with quality nectar and aroma, and should also be the most attractive to foraging bees (Erickson and Robbins 1979, Robacker et al. 1983).

Irrigation that is properly managed or applied is important for soybean production in several areas of the U.S. The Delta region of Mississippi, as well as eastern Nebraska, are areas of high concentrations of irrigated soybean. Soybean yields in Nebraska have increased 40% faster in irrigated versus traditional rain-fed soybean systems (Specht et al. 1999). In the midsouthern U.S., irrigation is required to consistently make a profitable soybean crop (Heatherly 1999). In the midsouthern U.S., moisture deficits increase in severity from April through August. This leads to serious drought stress during reproductive development of soybean nearly every growing season. Because pod and seed growth, which are quite sensitive to plant water deficits, occur later in the season when soil moisture and rainfall are at the lowest seasonal levels, the potential for significant reductions in their growth and development and subsequent yield is great (Heatherly and Elmore 2004).

This study was conducted to investigate the effects of certain cultural practices have on soybean nectar production. Erickson (1984b) reported that overall, the healthiest and the most well managed soybeans will produce the highest amount of quality nectar. Heatherly and Elmore (2004) conducted extensive testing to evaluate the early soybean production system and the effects on soybean growth and yield. There has been limited research into the effects that planting dates, irrigation practices, and maturity group selection has on nectar production. We hypothesize that factors producing faster growth and maturity will produce nectar with higher sugar content. We measured the total sugar content of single flowers from plants that varied based on planting dates, maturity group selections, and irrigation cycles. We adjusted the total sugar content to a per weight basis by weighing the flowers.

## Materials and Methods

This experiment was conducted in 2016 and 2017 in two separate locations: the Delta Research and Extension center in Stoneville, MS and the R.R. Foil Experiment Station in Starkville, MS to evaluate the influence of irrigation, maturity group, and planting date on nectar production in soybeans. Soybean was planted on 96 cm rows in Starkville and 102 cm rows in Stoneville with a plant density of 26 plants per row meter as recommended by Mississippi State University. Plot sizes were four rows by 7.6 to 12.2 meters long with four replications per test site. This experiment conducted was a randomized complete block design with a split-split plot arrangement of treatments. The major treatments factors were planting date, maturity group and irrigation. Levels of planting date included an early planting date during late April to early May and a late planting date during late May to early June. During 2016, plots were planted in Stoneville on 4 May and again on 24 May. Plots in Starkville were planted on 28 April and 2 June. During 2017, plots were planted in Stoneville on 28 April and again on 28 May. Plots in Starkville were planted on May 4$^{th}$ and again on May 30$^{th}$. AG4835 (Asgrow® 4835, Monsanto Company, St. Louis, MO) and AG5633 (Asgrow® 5633, Monsanto Company, St. Louis, MO) were used as levels of the maturity group factor. With the irrigation factor, plots were irrigated as needed or plants were grown without irrigation. Irrigation did not take place in the Hills region in Mississippi during 2016 due to timely rainfall events (Figure 2.1).

Nectar samples were collected at R1 or R2 growth stages (Fehr, et al. 1971) between the times of 10:00 AM and 2:00 PM (Erickson 1984a). Five flowers from the upper most fruiting position of 5 randomly selected plants were selected. Flowers were then removed and placed peduncle first into 200 μL of deionized water in a 1.5 mL Eppendorf tube. Samples were placed in a plastic cooler lined with frozen packs and taken to the lab. In the lab, flowers were weighed individually after removing the peduncle and calyx with a Denver scale (Instrument model P-114). Flowers were then placed upside down in a new 1.5 mL Eppendorf tube filled with 200μL of deionized water. Samples were centrifuged in a Sorvall RT6000 refrigerated centrifuge at 3000 RPM for 15 min at room temperature to aid with extraction of the nectar. Following centrifugation, the samples were frozen at -20°C until analysis.

The procedure used to process the nectar samples for total sugar analysis was an anthrone assay (Hansen and Moller, 1975). Anthrone is a spectrophotometric assay that measures furfural – anthrone complex by monitoring a wavelength of $\lambda$=630nm (Dreywood 1946). In the presence of sugars, the complex takes on a blue – green color. The anthrone reagent is made by dissolving anthrone in concentrated sulfuric acid to make a 0.2% solution. A 500 μL mixture of a chloroform-methanol (1:2) was added to the nectar sample and vortexed rapidly. The samples were then centrifuged at 8000 RPM for 10 minutes at 4°C. 150 μL of the supernatant were mixed with 100 μL of deionized water in a new 1.5 mL Eppendorf tube and then vortexed rapidly. 500 μL of anthrone reagent was then added to the mixture. The samples were then heated for 10 minutes at 90°C. 200 μL of the solution was then pipetted in triplicate in wells of a Fisher brand microtiter ELISA plate. The samples were then placed into a BioTek

Synergy H1M plate reader, and absorbance was measured at λ=630 nm. Total sugar content for each flower was estimated from a standard curve that was based on varying and known glucose concentrations, which was plotted in the GraphPad Prism 5 (GraphPad, CA) software program. The standard curve for glucose was made at a range of 0, 12.5, 25, 50, 75, 100, 150, and 200 µg/mL in deionized water, and all standards were processed the same as the nectar samples. A fresh standard curve was generated for each batch of flowers that were processed.

At the end of the season, soybean plots were mechanically harvested with a plot combine. Plot weights and moisture were recorded for soybeans from each plot to determine yield. Total sugar concentrations across all three factors were analyzed using analysis of variance (PROC GLIMMIX of SAS 9.4, SAS Institute, Cary, NC). Year, location, irrigation, planting date, and maturity group were considered fixed effects. Means and standard errors were calculated using the MEANS procedure. Fisher's Protected LSD was used to calculate *P*-Value at $\alpha=0.05$. Differences were considered significant at $\alpha=0.05$. Degrees of freedom were calculated using the Kenwood-Roger Method.

## Results and Discussion

### Total Sugar Content per Weight (µg sugar/mg of flower weight) of Flower

Mean total sugar content per weight (µg sugar/mg of flower weight) of soybean flower did not significantly differ between the Delta and Hills regions ($F=0.35$; $df=1,1204$; $P=0.56$). Additionally, the total sugar observed during the 2017 growing season was nearly twice the value observed in the 2016 growing season ($F=98.46$; $df=1,1204$; $P<0.01$) (Table 2.1). Severson et al. (1987) observed environmental effects when comparing two soybean varieties, Coker 237 and Centennial, from a similar two-year study in 1980 and 1981. During 1980 concentrations of fructose and glucose (reducing sugars) were three to four times higher than in 1981. Sucrose was omitted from their results despite it being measured because sucrose concentration was highly variable and dependent upon time of day of sampling (while the reducing sugars were more consistent through time of day).

The mean sugar content per weight (µg sugar/mg of flower weight) of soybean flower reported from the current study must be viewed with caution due to high variability between regions and years of the study. Total sugar content of soybean flowers was evaluated using an ANOVA for a multifactorial design with all treatment factors (irrigation, maturity group, planting date, year, and region) and all interaction terms among these modeled as fixed effects (Proc Glimmix, SAS). Each treatment factor consisted of two levels; therefore, any significant interactions could be considered disorderly and likely to profoundly limit any independent interpretation of the major factors. The five-way interaction (F=7.23; df=1, 1174, *P*=0.01) and four out of six of the three-way interactions were significant ($\alpha$=0.05) in the full model.

These results forced analyses of the four treatment combinations of region (Hills and Delta) by year (2016 and 2017). The analyses for each combination of region by year resulted in a reduced statistical model for comparison of total sugar content per flower (µg sugar/mg of flower weight). The ANOVA included the three treatment factors (irrigation, maturity group, planting date) and all interactions of these terms as fixed effects within each region and year.

### *Hills, 2016*

There was a significant interaction between maturity group and planting date ($F=50.11$; $df=1,391$; $P<0.01$). Maturity Group IV soybean plots that were planted early produced significantly more mean total sugar per weight of flower (µg sugar/mg of flower weight) than did any of the other three combinations (Table 2.2). The lowest sugar content per weight of flower (µg sugar/mg of flower weight) came from Maturity Group V soybeans planted at the early date (Table 2.2). There was a significant difference between maturity groups with respect to total sugar content per weight of flower in the early planting date. This difference is not observed in the later planting date. Erickson (1984) concluded that soybeans grown under the most optimal conditions produce higher quality and quantity of nectar as compared to poorer growing conditions. According to Heatherly and Elmore (2004), this is consistent with the early soybean production system as optimal times for soybean planting and Maturity Group IV variety selection. With the ESPS, early maturing Maturity Group V soybeans can also be planted at the same time. This discrepancy could be the result of the different genetics within the two varieties.

### *Hills, 2017*

There was a significant interaction between irrigation and planting date for mean total sugar ($F=5.63$; $df=1,152$; $P=0.02$). Early planted soybean that were irrigated produced significantly more mean total sugar per weight of flower (µg sugar/mg of flower weight) than other combinations (Table 2.3). There was a significant difference in total sugar content per weight of flower in the early planting date, but this trend is not observed in the later planting date. This is consistent with Severson et al. (1987)

findings in that lower nectar volumes and shifts in mean total sugar can be affected by water stress. Water stress has been identified as a major determinant of daily secretion rates of nectar in *Impatiens capensis* (Marden 1984). Both irrigated and non-irrigated plots that were planted early produced significantly more mean total sugar than later planted plots. This is consistent with findings from our 2016 plots suggesting that earlier planting dates can increase the amount of mean total sugar.

### *Delta, 2016*

There was a significant three-way interaction between irrigation, planting date, and maturity group selection (F=65.70; df=1,348.2; $P<0.01$). Late planted Maturity Group IV soybean plots that were irrigated produced significantly more mean total sugar than any other combination (Table 2.4). No differences in mean total sugar content per weight of flower were observed among maturity group or irrigation at the early planting date. In the later planting date, there were significant differences between the different combinations of factors. Comparable to Hills 2017 site year, avoiding water stress during the reproductive growth stages will keep nectar secretion higher. All late planted soybeans in this site year produced significantly more mean total sugar than the early planted soybeans as well. Many Mississippi beekeepers produce more honey on soybean and cotton during the hot months of June and July. Erickson (1984) noted that to insure nectar secretion, temperatures need to be above 22-24°C (70-75°F). In the later summer months of Mississippi, some nighttime temperatures never dip below this threshold. This could explain the increase in mean total sugar for later planted soybeans

### *Delta, 2017*

There was a significant interaction between maturity group and planting date (F=18.47; df=1,281.2; *P*<0.01). Early planted Group V soybeans produced significantly more mean total sugar per weight of flower (µg sugar/mg of flower weight) than other combinations (Table 2.5). There was a significant difference in mean total sugar per weight of flower in the early planted plots due to maturity group. This same trend is not observed in the later planted date. These results were the exact opposite of the trends seen in the Hills 2016. Explanation for these opposing results are most likely related to differences in the growing conditions between the two regions. With the early soybean production system, Group V varieties grown can compete with Group IV varieties when in optimal conditions (Heatherly 1999).

## Total Pods

Total pods produced on soybean plants was affected by planting date (Figure 2.1). For the 2017 growing season, later planted soybean plots produced significantly more total pods than early planted soybean plots for both the Hills region (F=11.17; df=1,91; *P*=0.01) and the Delta region (F=8.57; df=1,88; *P*=0.01). In response to decreasing day length, reproductive growth is initiated in soybeans (Bothwick and Parker 1938). Many pods are produced, but they do not have the proper nutrients to fill these pods with soybean seed. Even though these plots produced significantly more total pods, many pods were left unfilled.

There was no significant differences in total pods between soybeans that were irrigated and those grown without irrigation for both the Hills region (F=0.01; df=1,91; *P*=0.91) and the Delta region (F=0.15; df=1,88; *P*=0.70). Maturity group selection had no significant impact on total pod production in 2017. There were no significant differences observed with respect to total pod count for the Hills region (F=2.33; df=1,91; *P*=0.13) and the Delta region (F=3.57; df=1,88; *P*=0.06).

**Yield**

Planting date significantly affected yield for the 2016 Hills location (F=96.69; d=1,28; *P*<0.01). Early planted soybeans yielded significantly higher than later planted soybeans (Figure 2.2). This trend was also seen in the Hills region in 2017. Planting date significantly affected yield in the Hills region in 2017 (F=20.84; df=1,23; *P*=0.01). Early planted soybeans yielded significantly higher than those that were planted later (Figure 2.3). This result is congruent with past research with the early soybean production system that was adopted in the recent years (Heatherly 1999). Optimal planted soybeans can significantly out yield within the same varieties over their later planted counterparts (Heatherly and Elmore 2004).

There were no significant interactions between irrigation, maturity group selection, and planting date with respect to yield during the 2016 growing season in the Delta region (F=0.7061; df=1,24; *P*=0.71). None of the major factors significantly affected yield. Yield ranged from 1700-2744 kg $ha^{-1}$. There was a significant interaction between irrigation, maturity group selection, and planting date with respect to yield for the 2017 Delta region (F=42.90; df=1,24; *P*<0.01). Early planted Maturity Group V soybean plots that were both irrigated and not irrigated yielded significantly more than any other combination of factors (Table 2.6). Maturity Group IV and Maturity Group V soybean varieties are both used as part of the early soybean production system, so it is not uncommon to see some determinate varieties out yield indeterminate varieties when grown in optimal growing conditions.

Variation in mean total sugar content of nectar was high across all the site years that were tested. Mean total sugar per mg of flower weight did not differ between the locations. Due to major differences in the growing seasons between 2016 and 2017 (Figures 2.5-2.7), more mean total sugar was present in 2017. Differences in weather conditions during reproductive soybeans can influence the amount of mean total sugar each year. It is difficult to determine which cultural practices can increase total sugar production due to a multitude of different factors such as irrigation practices, location of planting, maturity group, and planting date. Some practices seem to benefit sugar production in one instance but not in another. Sugar production seems to highly variable and influenced by many of these factors.

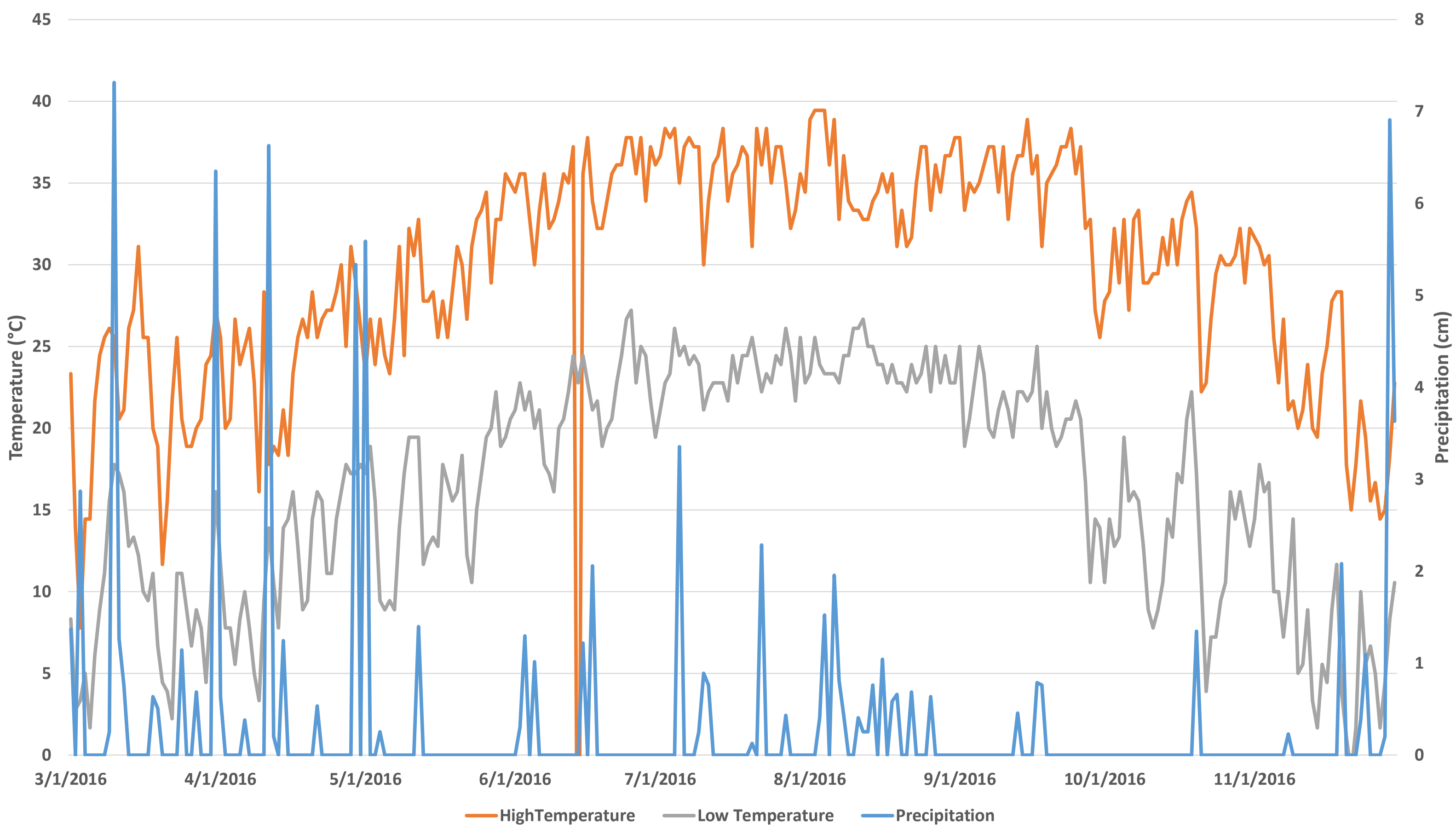


Figure 2.1 Weather data (precipitation, high temperature, and low temperature) for the Hills region of Mississippi in 2016 from 1 March 2016 to 30 November 2016.

Table 2.2 Differences in mean total sugar of soybean flowers between 2016 and 2017 across the Hills and Delta region of Mississippi.

| Year | Mean Total Sugar µg sugar/ mg flower weight | Standard error mean |
|---|---|---|
| 2016 | 24.20b | 0.71 |
| 2017 | 43.04a | 2.15 |

Means followed by the same letter are not significantly different (α=0.05, Fisher's PLSD).

Table 2.3 Effect of maturity group and planting date on mean total sugar concentration of soybean flowers for the 2016 Hills location in Mississippi.

| Planting Date | Maturity Group | Mean Total Sugar µg sugar/ mg flower weight | Standard error mean |
|---|---|---|---|
| Early | IV | 41.51a | 2.30 |
| | V | 17.27c | 0.95 |
| Late | IV | 23.86b | 1.01 |
| | V | 25.60b | 1.06 |

Means followed by the same letter are not significantly different (α=0.05, Fisher's PLSD).

Table 2.4 Effect of irrigation and planting date on mean total sugar concentration of soybean flowers for the 2017 Hills location in Mississippi.

| Planting Date | Irrigation | Mean Total Sugar µg sugar/ mg flower weight | Standard error mean |
|---|---|---|---|
| Early | Irrigated | 57.35a | 7.21 |
| | Non-irrigated | 41.33b | 3.84 |
| Late | Irrigated | 19.78c | 1.91 |
| | Non-irrigated | 25.36c | 3.22 |

Means followed by the same letter are not significantly different (α=0.05, Fisher's PLSD).

Table 2.5 Effect of irrigation, maturity group, and planting date on mean total sugar concentration of soybean flowers for the 2016 Delta location in Mississippi.

| Planting Date | Maturity Group | Irrigation | Mean Total Sugar µg sugar/ mg flower weight | Standard error mean |
|---|---|---|---|---|
| Early | IV | Irrigated | 9.52d | 0.75 |
| | | Non-irrigated | 8.00d | 0.59 |
| | V | Irrigated | 9.28d | 0.91 |
| | | Non-irrigated | 11.15d | 1.27 |
| Late | IV | Irrigated | 48.47a | 2.53 |
| | | Non-irrigated | 22.18c | 2.25 |
| | V | Irrigated | 23.64c | 1.66 |
| | | Non-irrigated | 39.80b | 4.09 |

Means followed by the same letter are not significantly different (α=0.05, Fisher's PLSD).

Table 2.6 Effect of maturity group and planting date on mean total sugar concentration of soybean flowers for the 2017 Delta location in Mississippi.

| Planting Date | Maturity Group | Mean Total Sugar μg sugar/ mg flower weight | Standard error mean |
|---|---|---|---|
| Early | IV | 33.96b | 2.89 |
| | V | 71.78a | 9.48 |
| Late | IV | 46.01b | 4.62 |
| | V | 31.64b | 3.37 |

Means followed by the same letter are not significantly different (α=0.05, Fisher's PLSD).

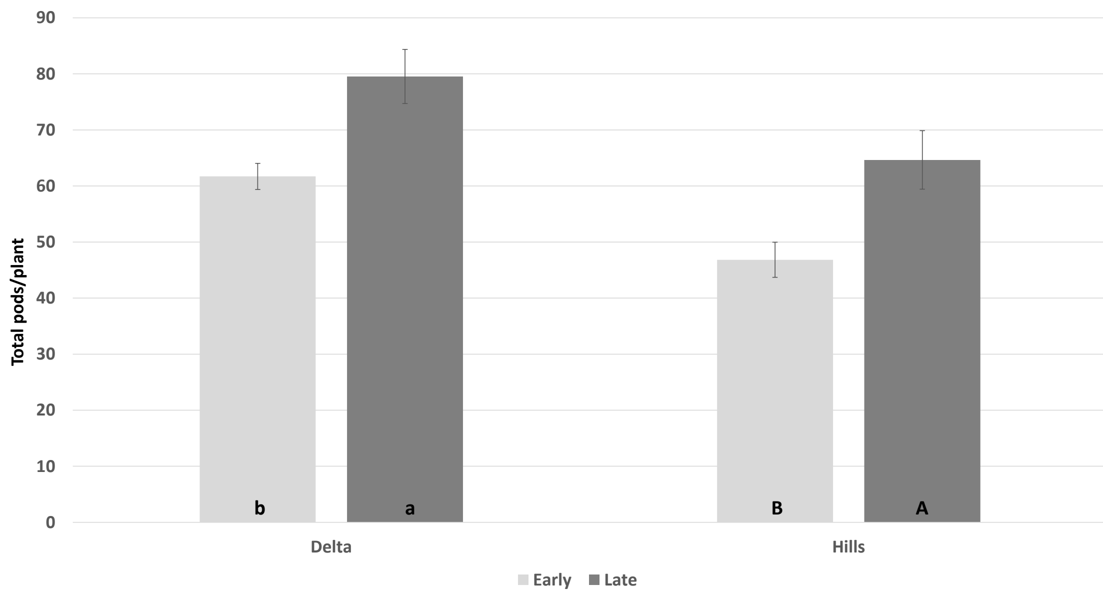


Figure 2.2 Total number of soybean pods for the Delta and Hills region of Mississippi for early and late planting dates in 2017.

Means followed by the same letter within a region are not significantly different (α=0.05, Fisher's PLSD).

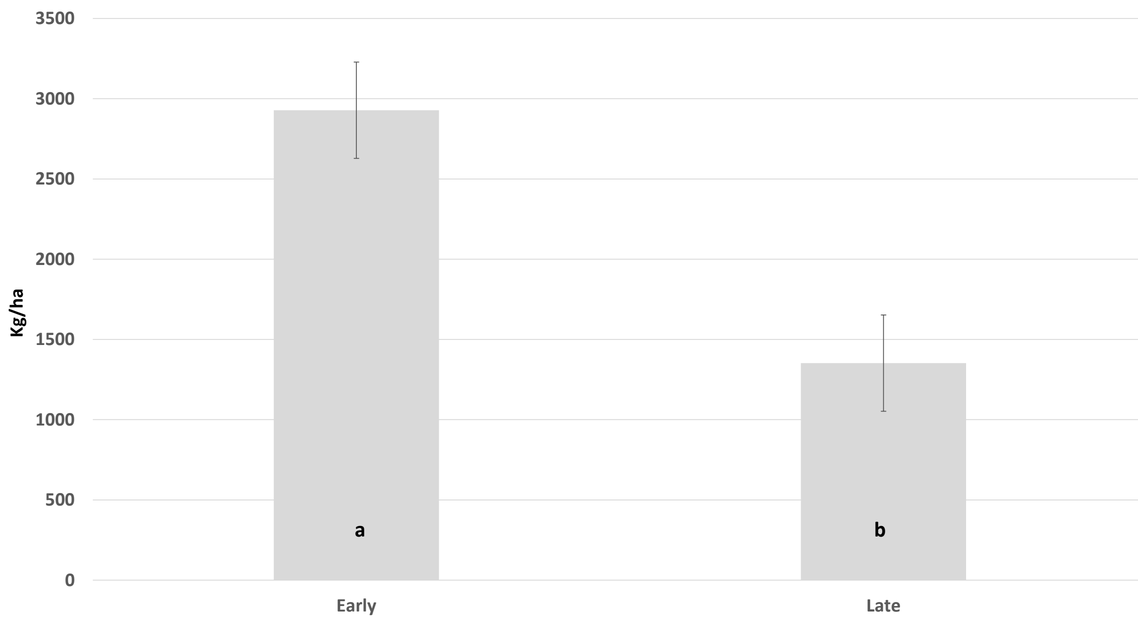


Figure 2.3 Yield (kg $ha^{-1}$) of soybeans for 2016 Hills region in Mississippi for early and late planting dates.

Means followed by the same letter are not significantly different (α=0.05, Fisher's PLSD).

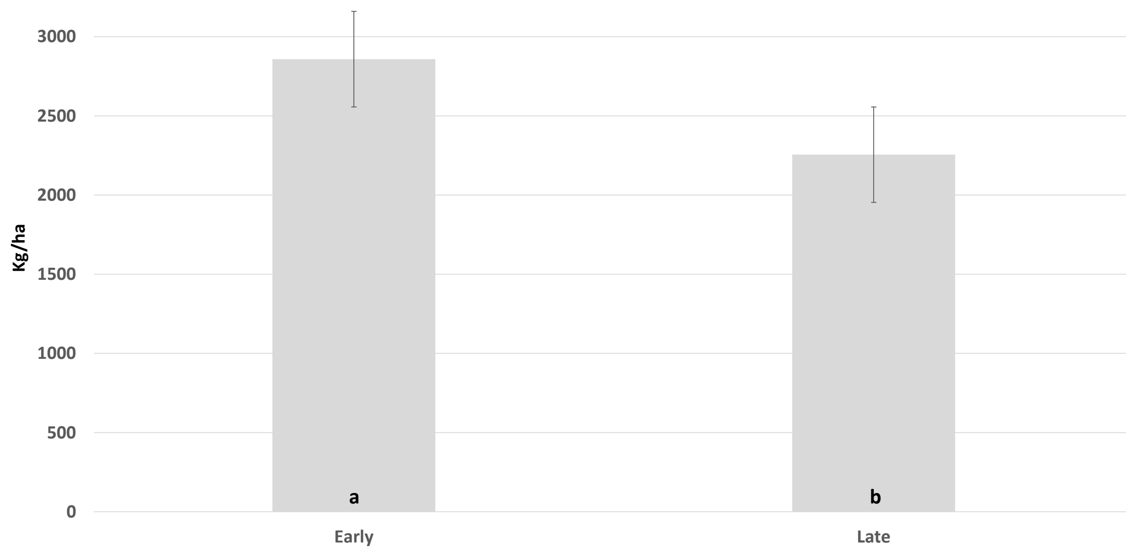


Figure 2.4 Yield (kg $ha^{-1}$) of soybeans for 2017 Hills region in Mississippi for early and late planting dates.

Means followed by the same letter are not significantly different (α=0.05, Fisher's PLSD).

Table 2.7 Effect of irrigation, maturity group, and planting date on yield ($kg\ ha^{-1}$) of soybeans for the 2017 Delta location in Mississippi.

| Planting Date | Maturity Group | Irrigation | Yield $kg\ ha^{-1}$ | Standard error mean |
|---|---|---|---|---|
| Early | Group IV | Irrigated | 2672.85cd | 64.12 |
| | | Non-irrigated | 2662.67cd | 143.32 |
| | Group V | Irrigated | 3512.83a | 73.96 |
| | | Non-irrigated | 3322.94ab | 61.35 |
| Late | Group IV | Irrigated | 3232.21b | 48.30 |
| | | Non-irrigated | 2505.31de | 60.09 |
| | Group V | Irrigated | 2407.22e | 22.81 |
| | | Non-irrigated | 2875.85c | 61.03 |

Means followed by the same letter are not significantly different (α=0.05, Fisher's PLSD).

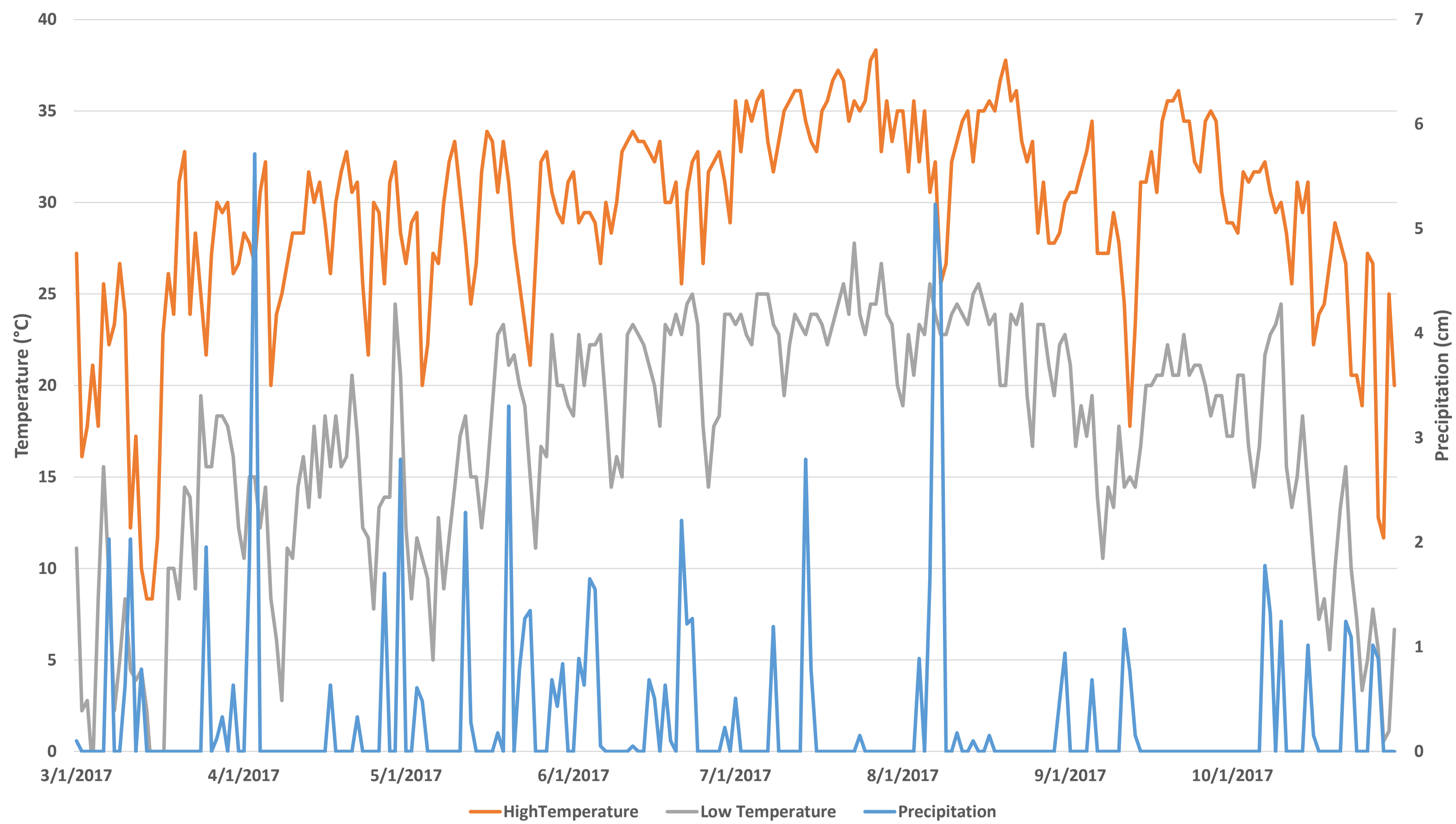


Figure 2.5 Weather data (precipitation, high temperature, and low temperature) for the Hills region of Mississippi in 2017 from 1 March 2017 to 30 November 2017.

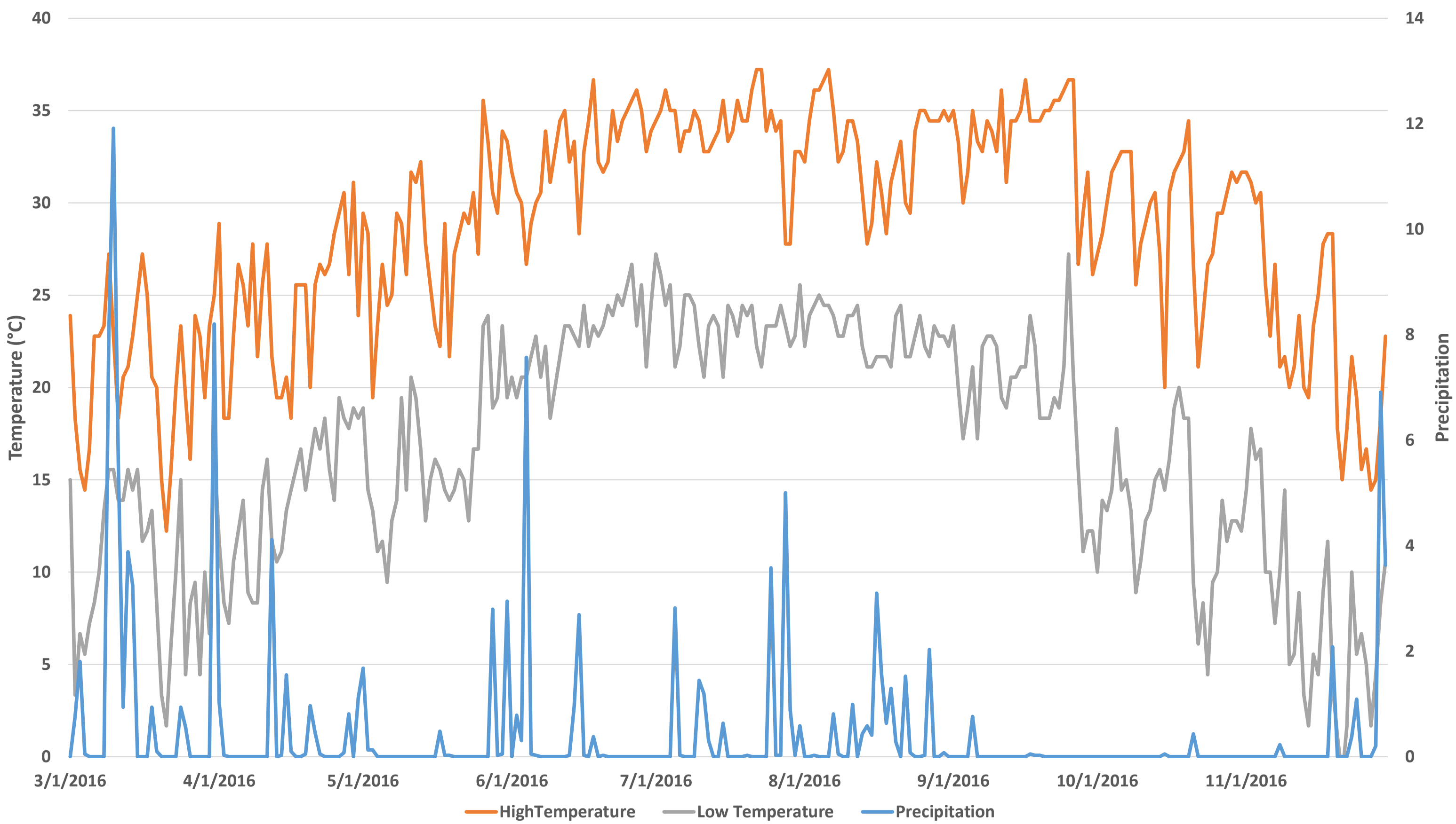


Figure 2.6 Weather data (precipitation, high temperature, and low temperature) for the Delta region of Mississippi in 2016 from 1 March 2016 to 30 November 2016.

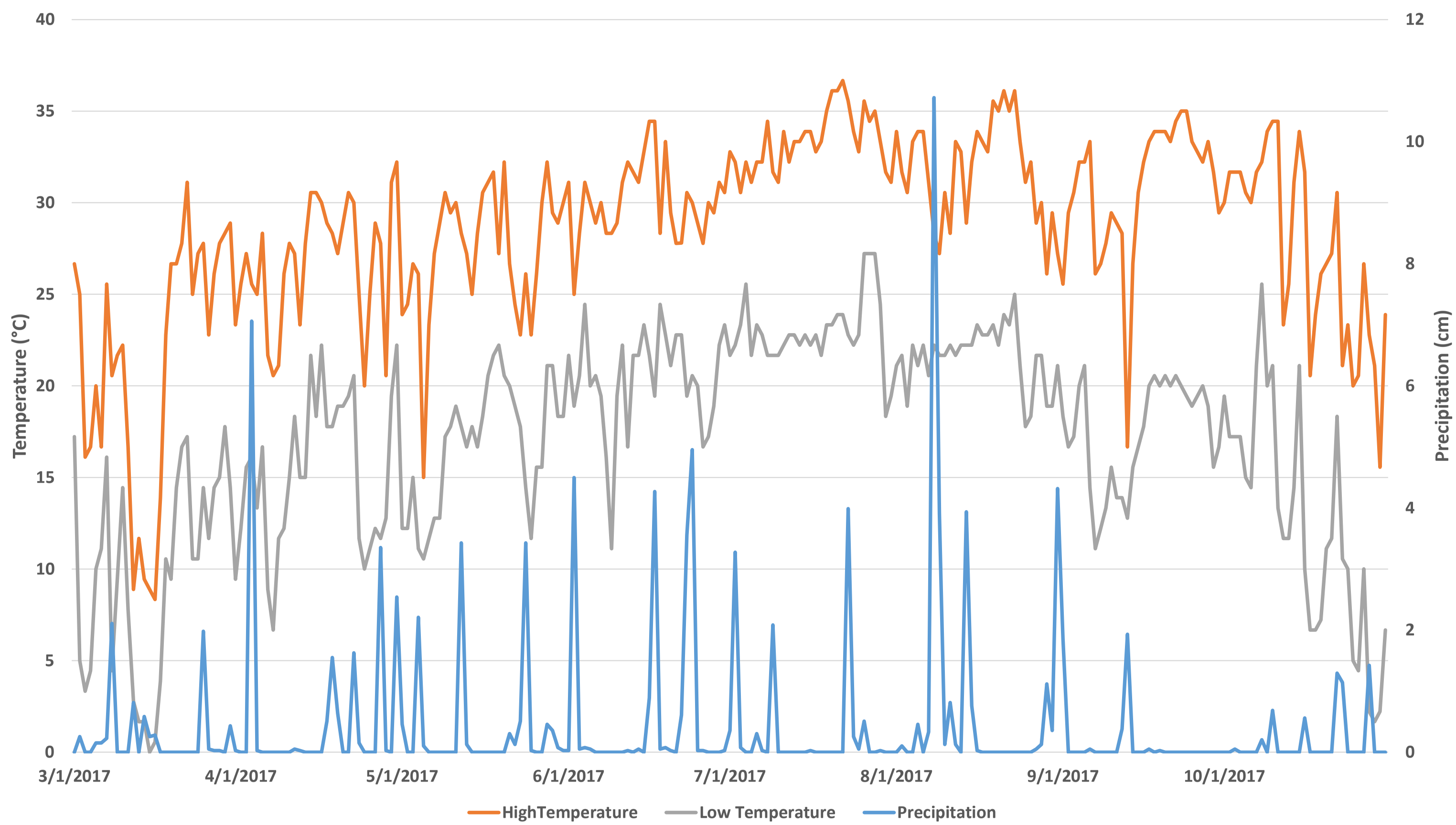


Figure 2.7 Weather data (precipitation, high temperature, and low temperature) for the Delta region of Mississippi in 2017 from 1 March 2017 to 30 November 2017.

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# CHAPTER III

# EVALUATING VARIETAL EFFECTS ON TOTAL SUGAR PRODUCTION IN SOYBEANS

## Abstract

Field experiments were conducted in 2016 and 2017 at the R. R. Foil Plant Science Research Center in Starkville, MS and the Delta Research and Extension Center in Stoneville, MS to determine the effects of variety selection on the total sugar content in soybean flowers. Sugar content was measured using a standard anthrone colorimetric assay. Soybean is a highly viable nectar source for visiting pollinators such as honey bees. With the steady increase in soybean production across Mississippi and the rest of the Mid-South, higher yielding varieties are being planted. As the landscape of soybean production changes, different varieties with different genetics are increasingly being introduced to pollinators. A location by variety interaction was observed for both years. There was considerable variability in sugar production between cultivars. Factors such as sampling time, temperature, and other varietal effects can impact nectar production. Later maturing soybean varieties tended to produce more total pods when compared to early maturing varieties. This result was the same for both the Hills and Delta regions. In the Hills region, yield was highly variable between all varieties tested. In the Delta

region, yields were not significantly different across all thirteen varieties. Total pod counts show a weak correlation to yield during the 2017 growing season.

## Introduction

There has been a proliferation of soybean varieties over the years, and many different varieties are commonly available that fit many different production systems. Because of this, no one variety is well suited for every soybean production system. Many varieties are chosen for specific sites depending on soil type, planting dates and other important factors identified by comparisons in state variety trials for a specific region (Heatherly 1999). State variety trials are conducted every year to compare and contrast the top varieties on the market. Growers consult these variety trials when making decisions about what varieties should be grown in their respective area.

Soybean productions occurs across all areas of Mississippi. There are two distinct regions than comprise most of the production in the state, the Hills and the Delta. The Hills region is mostly comprised of smaller cultivatable fields interspersed across the landscape. Overall, land dedicated to row crop agriculture makes up a small percentage of the overall land area (NASS 2017). In contrast, cultivatable land accounts for a larger percentage of the land area in the Delta region. Fields dedicated to row crop agriculture are much larger and more contiguous in the Delta region than in the Hills region (NASS 2017).

Over the years, there has been a wide shift into an early soybean production system in Mississippi. This system uses Maturity Group IV and Maturity Group V soybean varieties planted in Mid-April to May in order to avoid late season pests and drought conditions (Heatherly 1999). Bowers (1995) conducted 3 years of testing in the

1980s that helped establish this system. He concluded that early maturing varieties yielded more than conventional varieties when planted in Mid-April, and earlier maturing varieties also yielded more or as much as conventional varieties when planted in May.

**Soybean Nectar Production**

Variety and maturity group can also have an impact on the production of nectar. Nectar production is extremely dynamic and subject to genetic and environmentally induced variability (Severson et al. 1987). Severson and Erickson (1984) observed a significant difference in the amount of nectar that is produced among 18 cultivars. The amount of nectar that was produced ranged from 0.022 µl per flower to 0.127 µl per flower, and total carbohydrates ranged from 16 µg to 134 µg per flower. They also observed that sucrose is the most predominant sugar with the highest ratio being 1.2:1.0:6.7 (fructose:glucose:sucrose). Severson et al. (1987) tested the effects of sampling time and temperature on nectar production of two cultivars in Arkansas, and observed mean nectar to be highly variable between cultivars and between years. The same researchers also observed that the sugar ratio in 1980 was 1:1:1, but the ratio was sucrose dominant in 1981.

The current study was conducted to determine if the interactions of variety and location had an impact on the production of sugar in the nectar of different soybean varieties. It is well known that concentration and abundance of nectar in flowers affect honey bee foraging activity (Butler 1945; Kauffeld and Sorensen 1971). Previous research suggests that the variation in nectar production is a function of both extravarietal and intervarietal sources (Severson and Erickson 1984). However, published data to support this variation is minimal. We hypothesize that variety will have an impact on the production of soybean nectar during the reproduction growth phase. Our hope was to identify any varieties that had significantly higher mean total sugar in nectar of soybean flowers. To test this hypothesis, we looked at the amount of total sugar that was produced among different varieties in two uniquely different growing regions in Mississippi.

## Materials and Methods

This experiment was conducted at two locations: the R.R. Foil Experiment Station in Starkville, MS and the Delta Research and Extension Center in Stoneville, MS in the summers of 2016 and 2017. This test comprised thirteen different varieties from 3 maturity groups (Table 3.1) (Asgrow®, Monsanto Company, St. Louis, MO) arranged in a randomized complete block design with four replications. Soybean were planted on 96 cm rows in Starkville and 102 cm rows in Stoneville with a plant density of 26 plants per row meter as recommended by Mississippi State University. Plot sizes were four rows by 7.6 to 12.2 m long. In 2016, tests were planted in Stoneville on 5 May, 2016 and in Starkville on 11 May, 2016. In 2017, tests were planted in Stoneville on 23 June, 2017, and in Starkville on 26 June, 2017. The major factors were region, year, and variety.

Samples were collected at R1 or R2 growth stages between the times of 10:00 AM and 2:00 PM (Erickson 1984). Five flowers from the upper most fruiting position of 5 randomly selected plants were taken. Flowers were then cut and placed peduncle first into 200 µL of deionized water in a 1.5 mL Eppendorf tube. Samples were placed in a plastic cooler lined with frozen packs and taken to the lab. In the lab, flowers were weighed individually after removing the peduncle and calyx with a Denver scale

(Instrument model P-114). Flowers were then placed upside down in a new 1.5 mL Eppendorf tube filled with 200µL of deionized water. Samples were then centrifuged in a Sorvall RT6000 refrigerated centrifuge at 3000g for 15 min at room temperature to aid with extraction of the nectar. Following centrifugation, the samples were frozen at -20°C until analysis.

Total sugar was measured in nectar using an anthrone assay (Hansen and Moller, 1975). Anthrone is a spectrophotometric assay that measures absorbance by a furfural – anthrone complex by monitoring a wavelength of λ=630nm (Dreywood 1946). In the presence of sugars, formation of the complex takes on a blue – green color. The anthrone reagent is made by dissolving anthrone in concentrated sulfuric acid to make a 0.2% solution. Sugars were extracted using 500 µL of a chloroform-methanol (1:2) mixture that was added to the nectar sample and vortexed rapidly. The samples were then centrifuged at 8000g for 10 minutes at 4°C. 150 µL of the supernatant were be mixed with 100 µL of deionized water in a new 1.5 mL Eppendorf tube and then vortexed rapidly. 500 µL of anthrone reagent was then added to the mixture. The samples were then placed in a dry bath for 10 minutes at 90°C. 200 µL of the solution was then pipetted in triplicate in wells of a Fisher brand microtiter ELISA plate. The samples were then placed into a BioTek Synergy H1M plate reader and absorbance read at λ=630nm. A standard curve derived from known concentrations of glucose was used to estimate the total sugar content for each sample using the GraphPad Prism 5 (GraphPad, CA) software program. Standards for glucose were made at a range of 0, 12.5, 25, 50, 75, 100, 150, and 200 µg/mL in deionized water and processed with the anthrone reagent before measuring absorbance.

In October at the end of the growing season, plots were mechanically harvested with a plot combine. Plot weights and moisture were recorded for each plot. Total sugar, number of pods per 1 plant, and yield were analyzed using PROC GLIMMIX of SAS 9.4 (Version 9.4, SAS Institute, Cary, NC). Pearson's correlation was used to determine the relationship between flower weight and mean total sugar. Year, location, and replication were considered random effects, and variety was considered a fixed effect. Differences were considered significant at $P<0.05$. Fisher's Protected LSD was used to calculate *P*-value at $\alpha$. Degrees of freedom were calculated using the Kenwood-Roger Method.

Table 3.1 Soybean varieties used by maturity group.

| **Group III** | **Group IV** | **Group V** |
|---|---|---|
| AG3533 | AG4135 | AG5335 |
| AG3536 | AG4232 | AG5533 |
| AG3832 | AG4632 | AG5535 |
| AG3936 | AG4633 | AG5935 |
| | AG4835 | |

## Results and Discussion

There was a significant difference in mean total sugar content per weight of flower (µg total sugar/ mg of flower weight) between the Hills and Delta region ($F=19.14$; $df=1,1309$; $P<0.0001$). The soybeans grown in the Hills region produced significantly more mean total sugar than soybeans grown in the Delta region. The mean concentration of total sugar across all varieties in the Hills region was highly variable ($F=4.69$; $df=12,570.1$; $P<.01$). Varieties AG3533, AG4135, AG5935, AG3832, AG4835, AG4633, and AG4632 had significantly higher mean concentrations of total sugar compared to the other tested varieties (Figure 3.1).

The mean concentration of total sugar for varieties in the Delta region was also extremely variable (F=4.37; df=12,709.7; *P*<.01) (Figure 3.3). The varieties AG3533, AG4135, AG5935, AG4835, AG4632, AG4232, AG3936, and AG3536 had significantly higher mean concentrations of total sugar compared to other varieties (Figure 3.2). The correlation between flower weight and total sugar was significant. The Pearson's correlation (r=0.20) indicated that there was a positive relationship between flower weight and total sugar, but it was a weak to moderate correlation.

Nectar production in soybeans appears to be extremely dynamic, subject to genetic and environmentally induced variability. Consistent differences in nectar characteristics between the different varieties provide evidence for a genetic component. Environmental effects became evident after comparing the results from 2016 to 2017. The total sugar concentrations for 2016 were three to four times higher than those observed in 2017. In 2017, plots were planted much later than in 2016 due to seed availability. The trend in the data, that late planted soybeans produced significantly more total sugar, is consistent with findings from Severson et al. (1987). The range of sugar content in the 18 varieties tested was highly variable. The range of total carbohydrates from Severson et al. (1987) test ranged from 16 µg to 143 µg. The range of total carbohydrates, 82 µg to 143 µg, from the current study was consistent with this past research. From the results, we can conclude that there is a wide range of mean total sugar between all the varieties tested. This can be attributed to the genetic variability and the difference in the growing conditions between 2016 and 2017. This is consistent with findings from Severson and Erickson (1984). After testing two varieties in Missouri, Severson and Erickson (1984), concluded the high variability in nectar production due to genetic differences in the varieties and the subsequent environmental conditions.

For the 2017 growing season, later maturing varieties seemed to produce more total pods compared to earlier maturing varieties. This is consistent for both the Hills location ($F=3.52; df=11,129; P=0.01$) and Delta location ($F=5.92; df=11,114; P<.01$). AG4835, AG5335, AG5533, AG5535, AG5935 produced significantly more total pods compared to other varieties tested in the Hills location (Figure 3.3). AG5533, AG5535, AG5935 produced significantly more total pods in the Delta location compared to other varieties tested (Figure 3.4). Although these varieties produced more total pods, this was not reflected in yields. There was no significant correlation between number of pods per 1 plant and yield ($P=0.10$). Variety (genetics) and management strategies can influence the number of pods per 1 plant (Pederson and Lauer 2004). Different management strategies, such as irrigation, weed management, and insect pest management were used at the different locations in the current study.

Similar to total sugar concentrations, there was widespread variability in yield in the Hills location ($F=3.53$; $df=11,81.04$; $P=.01$). The Delta location showed no significant differences in yield when comparing all thirteen varieties tested ($F=0.44; df=10,77; P=0.9215$). AG3533, AG3832, AG3936, AG4135, and AG4232 yielded significantly more than other varieties tested in the Hills region (Figure 3.5). In the Delta region, yield ranged from 2951-4124 kg $ha^{-1}$. Differences in management systems among location can influence yield with different varieties (Pedersen and Lauer 2004).

Mean total sugar was highly variable for both tested locations. Where some varieties produced significantly more mean total sugar in one location, they sometimes produced significantly less in the other location. Mean total sugar ranged from 82 μg to 143 μg. Later maturing varieties produced significantly more total pods than early maturing varieties. This included a range of indeterminate and determinate varieties. Yield and total number of pods per 1 plant counts were not significantly correlated.

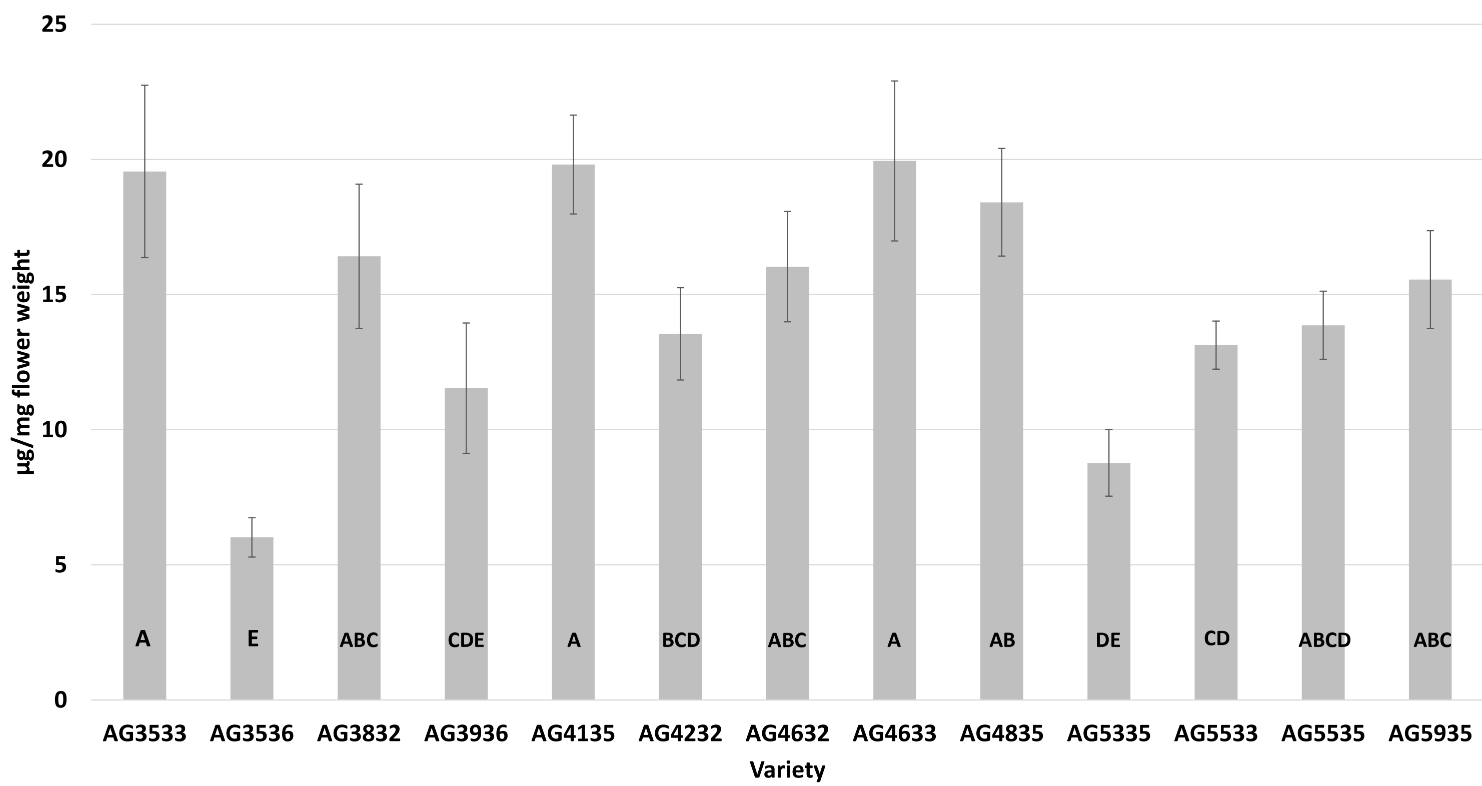


Figure 3.1 Total sugar concentration (µg sugar/mg flower weight) of soybean flowers by variety for the Hills region in Mississippi for 2016 and 2017.

Means followed by the same letter are not significantly different (α=0.05, Fisher's PLSD).

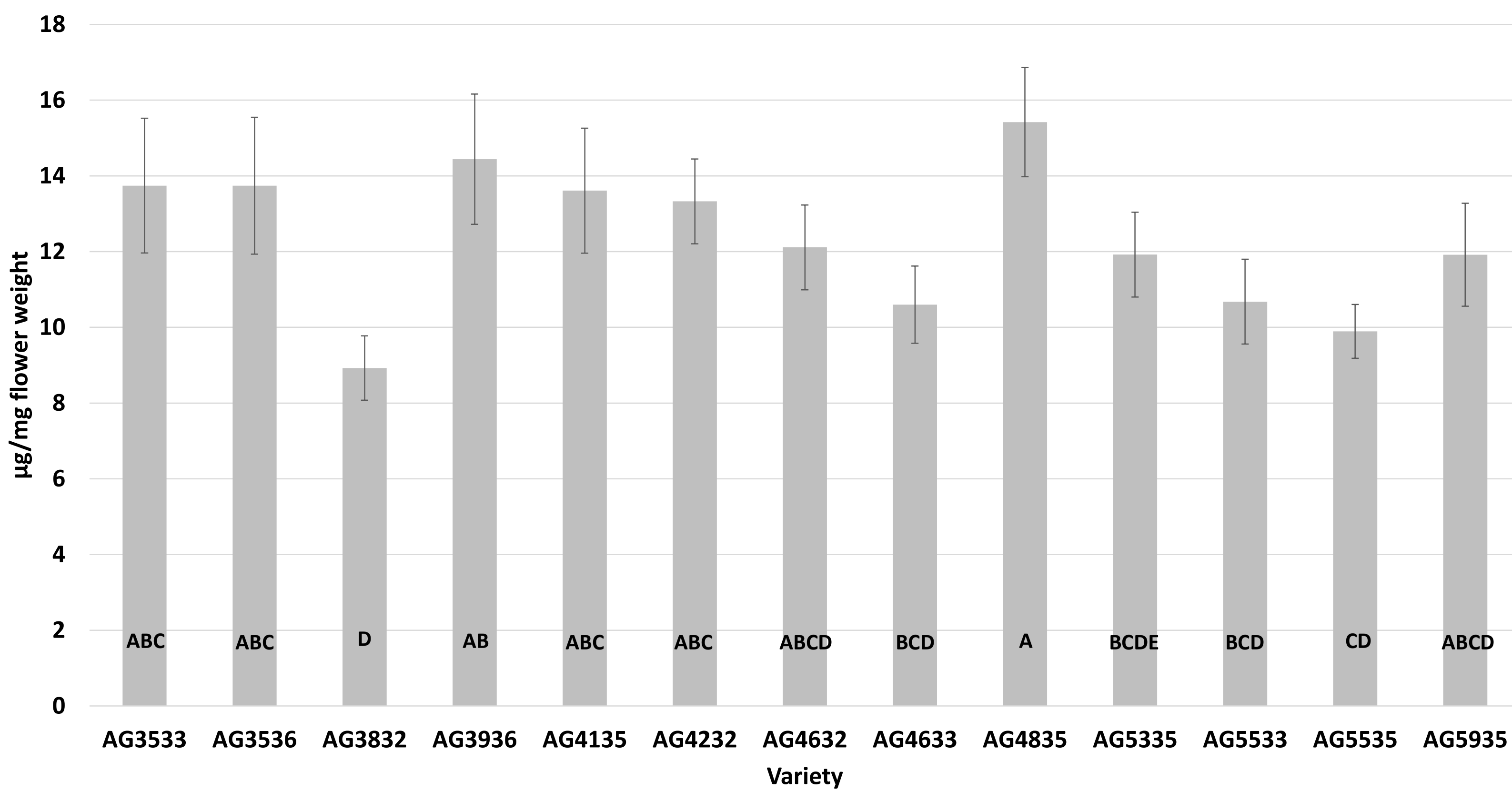


Figure 3.2 Total sugar concentration (µg sugar/mg flower weight) of soybean flowers by variety for the Delta region of Mississippi in 2016 and 2017.

Means followed by the same letter are not significantly different ($\alpha$=0.05, Fisher's PLSD).

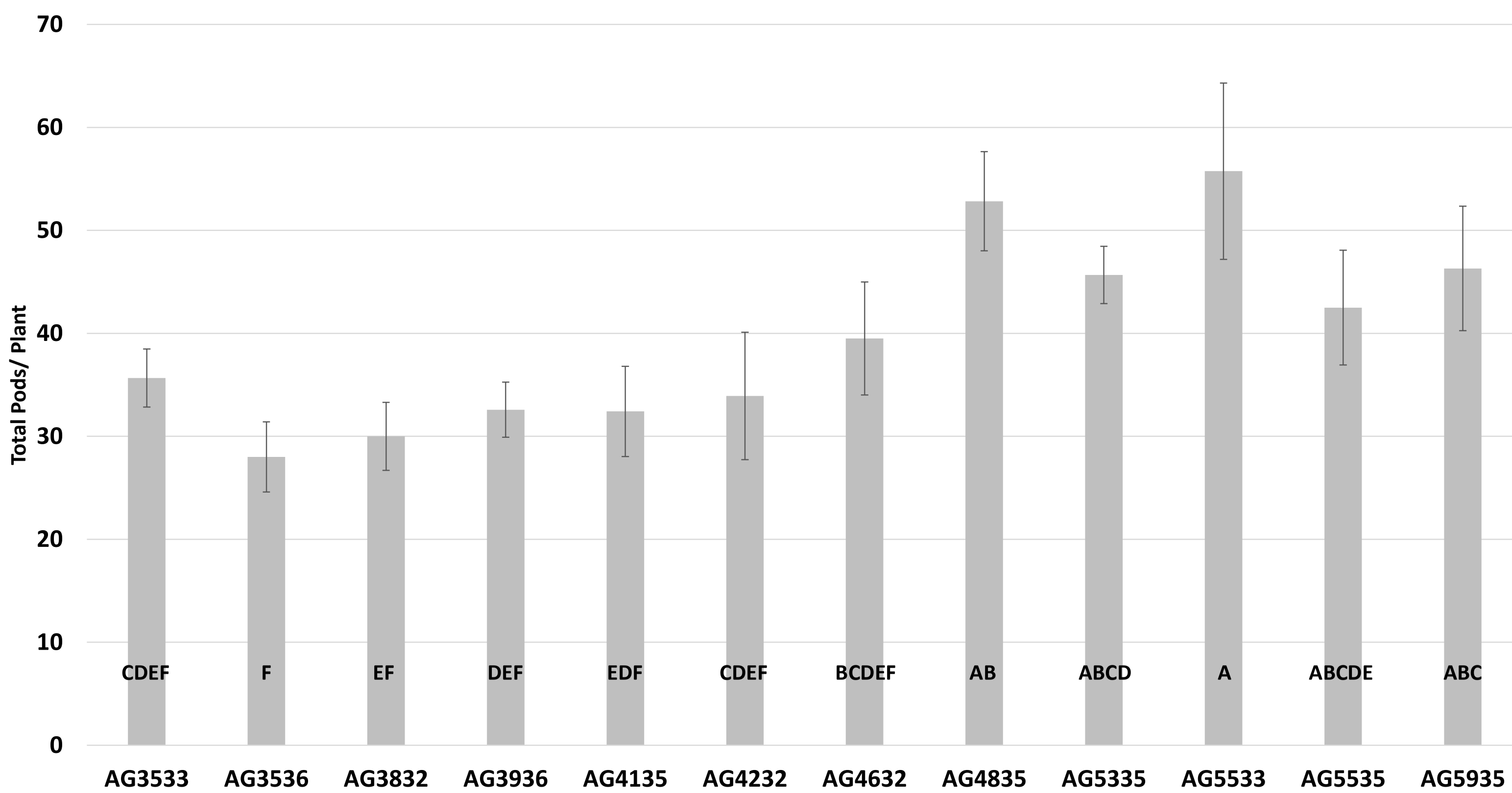


Figure 3.3 Total number of soybean pods by variety for the Hills region of Mississippi in 2017.

Means followed by the same letter are not significantly different (α=0.05, Fisher’s PLSD).

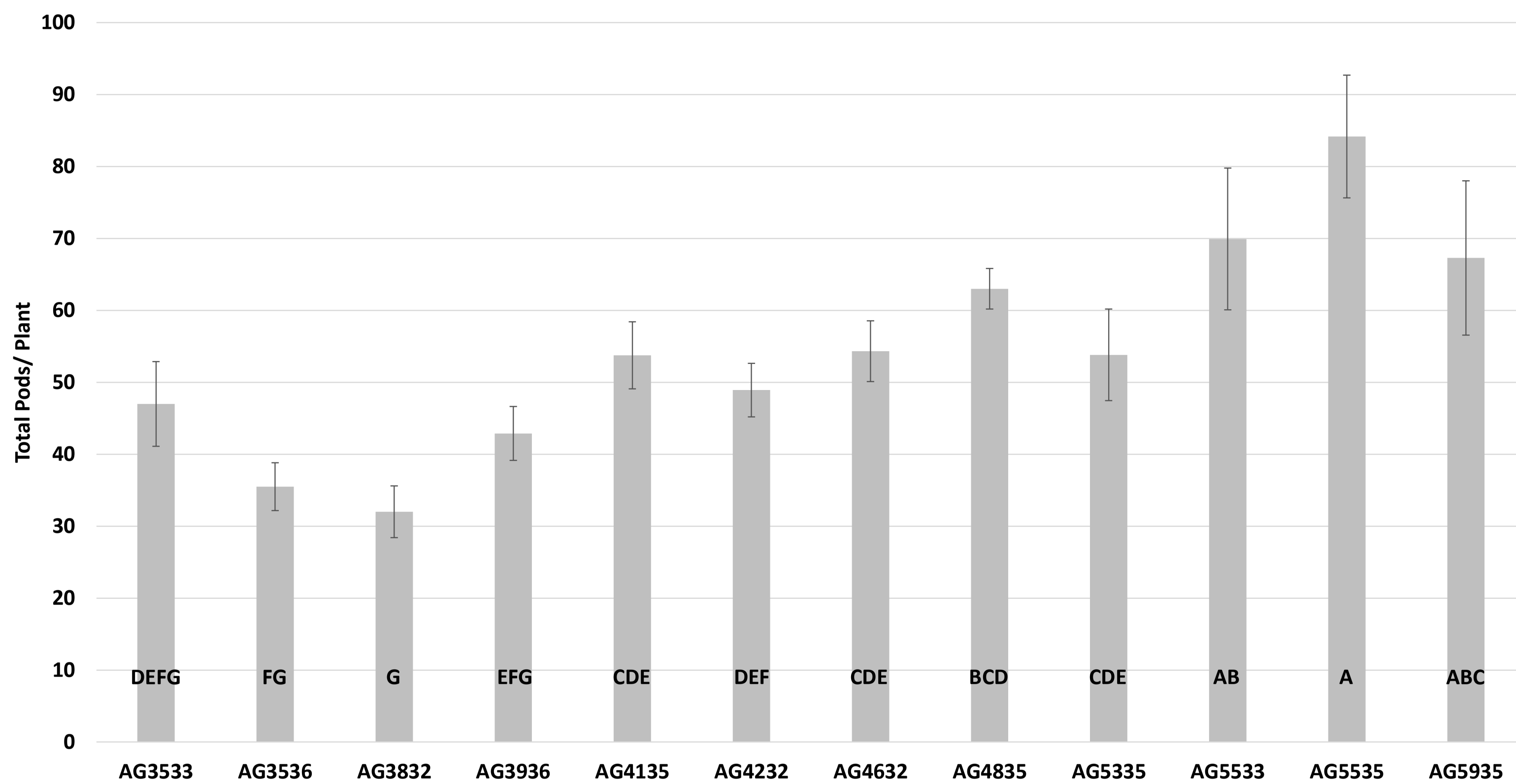


Figure 3.4 Total number of soybean pods by variety for the Delta region of Mississippi in 2017.

Means followed by the same letter are not significantly different (α=0.05, Fisher's PLSD).

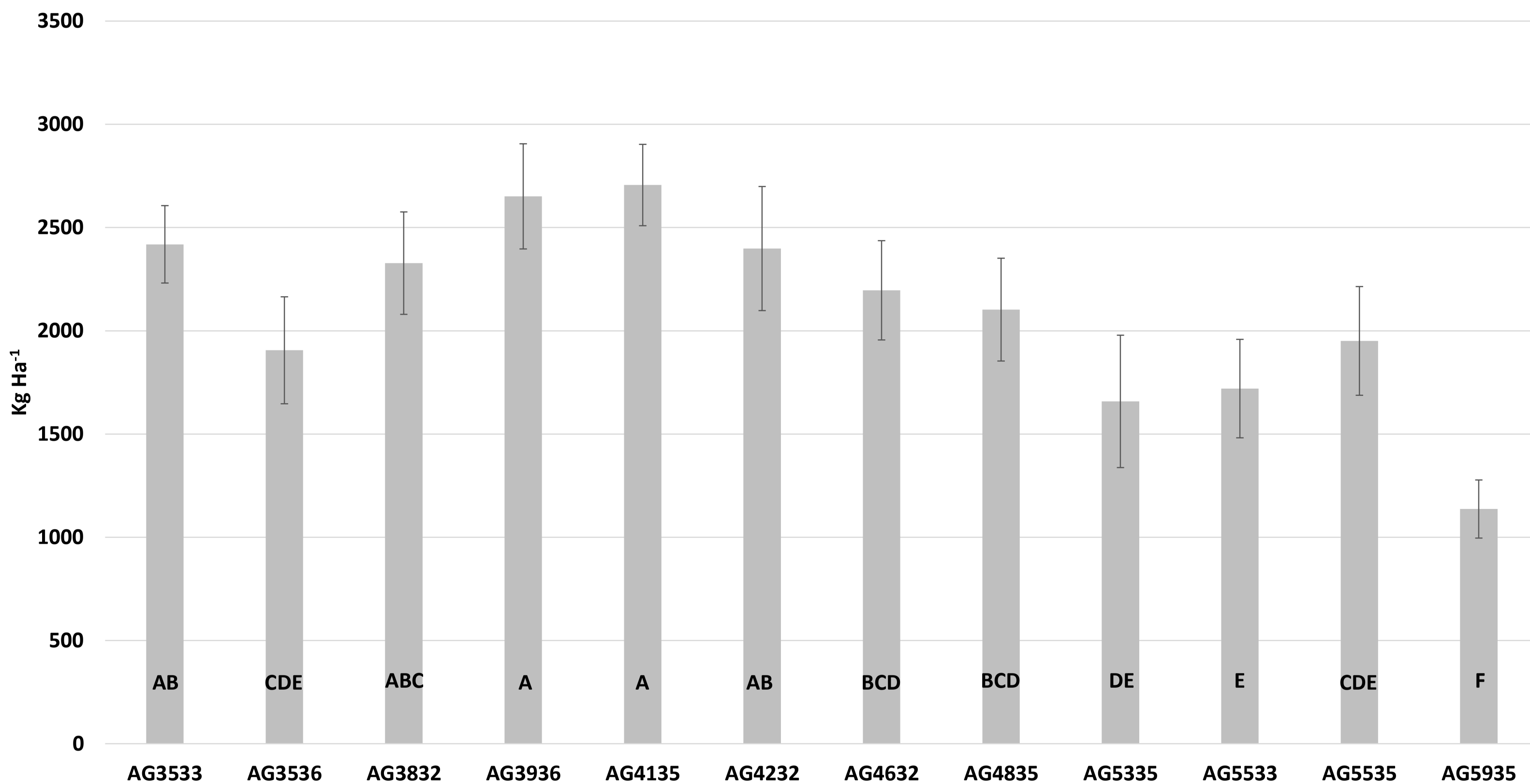


Figure 3.5 Yield of soybeans for Hills region in Mississippi in kg/ha.

Means followed by the same letter are not significantly different ($\alpha$=0.05, Fisher's PLSD).

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# CHAPTER IV

# SUMMARY

Field experiments were conducted during 2016 and 2017 in the Hills and Delta region of Mississippi. These experiments consisted of analyzing nectar for mean total sugar per mg of flower weight during early reproductive stage soybeans. Factors included in these experiments were irrigation practices, planting dates, and maturity groups. These experiments were conducted to determine if certain cultural practices can influence how much mean total sugar is produced per mg of flower weight in soybeans.

Variation in mean total sugar content of nectar was high across all the site years that were tested. Mean total sugar per mg of flower weight did not differ between the locations. Due to major differences in the growing seasons between 2016 and 2017, more mean total sugar was present in 2017. Differences in weather conditions during reproductive stages of soybeans can influence the amount of mean total sugar each year.

Early planted Maturity Group IV soybeans produced the most total sugar in the Hills in 2016. There was a large discrepancy between Maturity Group IV soybeans and Maturity Group V soybeans planted early in the test. The Maturity Group V soybeans planted early produced significantly less mean total sugar. Genetic differences between the two varieties might explain this variability in mean total sugar even though this difference is not observed in the later planting date.

Early planted plots that were irrigated produced the most mean total sugar in the Hills in 2017. Both irrigated and non-irrigated plots that were planted early produced more mean total sugar than the plots that were planted late.  This is consistent with findings from our 2016 test in the Hills suggesting that early planting can produce more mean total sugar.

Late planted soybean plots in the Delta in 2016 produced more mean total sugar than plots planted early. This contradicts observations from the experiments performed in the Hills. This could be explained by the possibility of the growing conditions between the two regions being significantly different. Nighttime temperatures rarely drop below the 22-24°C threshold for nectar production in the later months of summer. Later planted soybeans are usually in reproductive stages around this time.

Early planted Maturity Group V soybean plots in the Delta in 2017 produced more mean total sugar. This data contradicts findings from the Hills region in 2016. Like plots in the 2016 test in the Delta region, the growing conditions can be the only explanation for the major differences.  Later planted soybean plots put on much more total pods than early planted plots in both the Hills location and Delta location. This did not reflect yield though. Early planted soybean plots yielded much higher than the later planted plots. This suggests that many pods were left unfilled in the later planted plots. Like mean total sugar, yield was variable among the different cultural practices.

Studies evaluating the effects of variety selection on mean total sugar in nectar soybean flowers were conducted in 2016 and 2017. Mean total sugar was highly variable for both tested locations. Where some varieties produced significantly more mean total sugar in one location, they sometimes produced significantly less in the other location. Mean total sugar ranged from 82 µg to 143 µg. Later maturing varieties produced significantly more total pods than early maturing varieties. This included a range of indeterminate and determinate varieties. Yield and total number of pods per 1 plant counts were not significantly correlated.

Throughout the studies conducted in 2016 and 2017, mean total sugar was observed to be highly variable. At different site years, there were different interactions at each place. Mean total sugar is influenced by a multitude of factors. Some factors that are not easy to control. Genetic differences between varieties and the conditions they are grown in can have a profound effect on nectar production in soybean. Yield among different varieties is also highly variable. Management practices also affect mean total sugar as well as yield. Early planted plots in the 2017 Delta site and both Hills sites in 2016 and 2017 produced more total sugar than later planted plots. In the 2016 Delta site it was the later planted plots producing the most total sugar. It is difficult to identify exactly what management practices and varieties will benefit beekeepers the most. Since no factor(s) could be identified that consistently influenced sugar content of nectar in soybean, no management recommendations can be made.