

USE AND EFFECTS OF DIPLOID POLLENIZERS FOR TRIPLOID WATERMELON
[*Citrullus lanatus* (Thunberg) Matsumura and Nakai] PRODUCTION

By

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To my wife Lindsey for the sacrifices she has made for me to be here, and to my parents for their unyielding support.

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TABLE OF CONTENTS

	<u>page</u>
ACKNOWLEDGMENTS	4
LIST OF TABLES	7
LIST OF FIGURES	8
ABSTRACT	9
CHAPTER	
1 INTRODUCTION	11
2 REVIEW OF THE LITERATURE.....	13
Review of Diploid Watermelon.....	13
Taxonomy and Botany	13
Watermelon Production Practices	15
Watermelon Production Statistics	18
Review of Triploid Watermelon.....	19
Background Information	19
Seedless Watermelon Market Share.....	20
Triploid Seedless Watermelon Production.....	21
Pollinator Activity and Preference	26
Pollen Effects.....	27
Competition	30
3 CHARACTERISTICS OF DIPLOID POLLENIZERS FOR USE IN TRIPLOID WATERMELON PRODUCTION	32
Introduction.....	32
Materials and Methods	32
Results.....	34
Discussion.....	36
4 POLLEN VIABILITY OF DIPLOID WATERMELON POLLENIZER CULTIVARS.....	40
Introduction.....	40
Materials and Methods	40
Results.....	42
Discussion.....	42

5	DIPLOID WATERMELON POLLENIZER CULTIVARS EXHIBIT VARYING DEGREES OF PERFORMANCE WITH RESPECT TO TRIPLOID WATERMELON YIELD	46
	Introduction.....	46
	Materials and Methods	46
	Results.....	49
	Discussion.....	50
6	COMPETITIVE AFFECT OF IN-ROW DIPLOID WATERMELON POLLENIZERS ON TRIPLOID WATERMELON YIELD	57
	Introduction.....	57
	Materials and Methods	57
	Results.....	58
	Discussion.....	59
7	VARIABILITY IN WATERMELON FLOWER ATTRACTIVENESS TO INSECT POLLINATORS	63
	Introduction.....	63
	Materials and Methods	63
	Results.....	64
	Discussion.....	66
8	VARIABILITY IN POLLEN PRODUCTION BY DIPLOID WATERMELON POLLENIZERS	72
	Introduction.....	72
	Materials and Methods	72
	Results.....	73
	Discussion.....	74
	LIST OF REFERENCES	77
	BIOGRAPHICAL SKETCH	84

LIST OF TABLES

<u>Table</u>	<u>page</u>
3-1 Analysis of variance for pollenizer fruit weight and fruit per plant at Quincy and Live Oak FL during 2005.	37
3-2 Interaction effect of location and watermelon pollenizer cultivars on fruit per plant at Quincy and Live Oak FL during 2005.	37
3-3 Main effects for pollenizer fruit weights combined over experiments conducted in Quincy and Live Oak, FL, during 2005.	38
4-1 Analysis of variance for pollen viability of watermelon pollenizer cultivars tested during the Spring and Fall of 2006 at Quincy, FL.	44
4-2 Influence of diploid watermelon pollenizer cultivar on pollen viability at Quincy, FL during the Spring and Fall of 2006.	45
5-1 Pollenizer cultivar effect on ‘Tri-X 313’ yield at Blackville, SC during 2005.	54
5-2 Pollenizer cultivar effect on ‘Supercrisp’ watermelon yield and average fruit weight at Blackville, SC, Citra, FL, and Quincy, FL during 2006.	54
5-3 Pollenizer cultivar effect on soluble solids concentration of seedless watermelons at Blackville, SC during 2005 and Citra, FL, Quincy, FL, and Blackville, SC during 2006.	55
5-4 Pollenizer cultivar effect on hollowheart disorder in ‘Supercrisp’ watermelon at Quincy, FL, and Blackville, SC combined with Citra, FL during 2006. Means are to be compared within the same column.	56
5-5 Seed sources for various pollenizer cultivars used during 2005 and 2006.	56
6-1 Influence of pollenizer cultivar and spacing on triploid watermelon yield during 2006.	62
8-1 Pollen production by four diploid watermelon pollenizer cultivars at Quincy, FL during the Fall of 2006.	76

LIST OF FIGURES

<u>Figure</u>	<u>page</u>
3-1 Pollenizer staminate flower counts combined over Quincy and Live Oak, FL, during 2005. Means not followed by the same letter are significantly different at ($P \leq 0.05$) by Duncan's Multiple Range Test.	39
5-1 Field diagram for pollenizer experiments at Blackville, S.C., Citra, FL., and Quincy, FL. in 2005 and 2006. Columns represent individual rows. The same design was used for replications three and four.	52
5-2 Individual three-row plot design for pollenizer experiments at Blackville, S.C., Citra, FL, and Quincy, FL in 2005 and 2006. Plot shown is using a pollenizer recommended to be planted at a 1:3 pollenizer to seedless ratio.	53
7-1 Influence of cultivar on pollinator visitation to staminate watermelon flowers at Quincy, FL, Spring 2006. Visitation means are to be compared within sample date. Means not followed by the same letter are significantly different at $P \leq 0.05$	68
7-2 Interaction effect of cultivar and time on pollinator visitation to staminate watermelon flowers at Quincy, FL, on 23 May, 2006. Visitation means are to be compared within sample time. Means not followed by the same letter are significantly different at $P \leq 0.05$	69
7-3 Influence of cultivar on pollinator visitation to staminate watermelon flowers at Quincy, FL, Fall 2006. Visitation means are to be compared within sample date. Means not followed by the same letter are significantly different at $P \leq 0.05$	70
7-4 Interaction effect of cultivar and time on pollinator visitation to staminate watermelon flowers at Quincy, FL, on 23 Sept., 2006. Visitation means are to be compared within sample time. Means not followed by the same letter are significantly different at $P \leq 0.05$	71

Abstract of Dissertation Presented to the Graduate School
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By

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The use of in-row pollenizers has become popular because it allows a greater number of triploid plants to be planted per hectare. Multiple in-row pollenizer cultivars are available but it is unclear if any exhibit superior performance with respect to triploid watermelon yield and if so, what varietal characteristics contribute to a pollenizer's performance.

Field trials were conducted during the Spring and Fall of 2005 and 2006 at various locations in FL. and S.C. to determine the performance of various pollenizers and also what contributed to their success. Of the in-row pollenizers that were tested, 'Sidekick' resulted in the greatest triploid watermelon yields. Yields from plots pollenized by 'Patron', 'SP-1', 'Jenny', and 'Mickylee' were not significantly lower. The use of 'Companion' resulted in significantly lower yields than plots pollenized 'Sidekick', 'Patron', 'SP-1', and 'Jenny'.

Pollen viability can vary between cultivars of a plant species and the pollen viability of four pollenizer cultivars was evaluated. Poor pollen viability from a pollenizer could result in increased fruit abortions and lower yield. No significant differences in pollen viability were detected between pollenizer cultivars tested. The production of staminate flowers is a crucial factor for pollenizers as there must be adequate pollen flow during triploid watermelon fruit set. Flower production for 'Companion', 'Jenny', 'Mickylee', and 'SP-1' was recorded and

‘Companion’ produced as many flowers as ‘Jenny’ or ‘Mickylee’ throughout most of the season. The flowering period of ‘Companion’ does appear to be shorter than other cultivars. Pollinator preference was examined between ‘Companion’, ‘Mickylee’, and ‘SP-1’ and ‘Companion’ was found to be the least attractive of the three. The lack of pollinator visitation to ‘Companion’ appears to be the greatest contributor to its poor performance.

‘Mickylee’ is an attractive option to use as a pollenizer because of its lower seed costs. However, ‘Mickylee’ has a growth habit that is more vigorous than most other pollenizers. Studies comparing ‘SP-1’ and ‘Mickylee’ showed that the ‘Mickylee’ competed more with associated triploid plants and reduced yield. Factors affecting pollenizer performance the most appear to be pollinator preference, staminate flower production, and competitive effect on associated triploids.

CHAPTER 1 INTRODUCTION

Seedless watermelons now account for 78% of the watermelons sold in the United States. Triploid watermelon plants produce seedless fruits however; they do not produce sufficient viable pollen which is necessary for fruit set. To achieve optimal seedless watermelon yields, rows of diploid watermelon cultivars are planted in the same field as the triploids. These diploid cultivars account for 20 to 33% of the watermelon plants per hectare. With low demand for seeded watermelons, it has become less desirable to maintain this much hectareage in diploid plants. Diploid cultivars (pollenizers) designed to be planted within the row of triploid watermelons are now available. The use of these in-row pollenizers requires no dedicated space for the pollenizer plants, thus allowing for an increase in triploid plants per hectare. When in-row pollenizers first became available there were few options, now there are 11 commercially available cultivars. Using in-row pollenizers is a new concept and most of the in-row pollenizer cultivars have been on the market less than two years. The performance of in-row pollenizer cultivars has not been demonstrated, however earlier studies have reported differences in triploid watermelon yields due to pollenizer cultivar. It is suspected that the similar results will be observed with in-row pollenizers.

Available in-row pollenizers have diverse phenotypes and variable plant characteristics and it is unclear how these may effect the cultivar's performance. Characteristics such as flower production, pollen production, pollen viability, attractiveness to pollinators, and plant vigor could have a marked effect on how triploid plants in association with the pollenizers will yield. Pollenizer seed/plant costs are highly variable and it is unclear whether the more expensive pollenizers will provide for greater seedless watermelon yields.

The aim of this body of work is to determine how well in-row pollenizers perform and how their varietal characteristics influence their performance.

CHAPTER 2 REVIEW OF THE LITERATURE

Review of Diploid Watermelon

Taxonomy and Botany

The watermelon [*Citrullus lanatus* (Thunberg) Matsumura & Nakai] was originally described in Africa as *Mamordica lanata* by Carl Thunberg in 1794 (Hara, 1969). Schrader proposed *Citrullus vulgaris* in 1836 which was commonly used until *lanatus* was accepted as the oldest specific epithet. Hara revealed the original description in 1969 that was published by Matsumura and Nakai in 1920 and watermelon has since been designated as *Citrullus lanatus* (Thunberg) Matsumura & Nakai (Hara, 1969).

Watermelon is a dicotyledonous angiosperm in the order Violales and family Cucurbitaceae (Kartesz, 2006). The center of origin for the species is believed to be in southern Africa, where it was first described by Thunberg. David Livingstone reported seeing watermelons growing wild in the Kalahari Desert in 1857 (Wehner, 2006). Watermelon is a warm-season, herbaceous vining annual with angular stems that may reach 9 m long (Wehner, 2006). There are also genotypes that exhibit a bush or dwarf growth habit (due to shorter internodes) that have shorter stems and are less branched. The main stem or crown of the plant develops from the seedling stem and may have few to many lateral branches develop depending on genotype. Most watermelon genotypes have pinnately lobed leaves with three to five pairs of lobes and are born singly on the stem (McCreight, 1996). Cultivars have been developed that have a nearly entire leaf which is due to a gene mutation (Wehner, 2006). Branched tendrils are produced at each node. Watermelon is a monoecious plant with imperfect flowers that are born singly at the leaf axil (Wehner, 2006). Watermelon flowers are small in comparison with other cucurbits such as those in the genera *Cucurbita* and are born on short peduncles. When plants

reach sexual maturity, staminate (male) flowers are produced first and for a period of time before pistillate (female) flowers are produced. Generally, staminate flowers have three anthers and pistillate flowers have three stigmatal lobes. Pistillate flowers have inferior ovaries which vary in size and shape depending on genotype. Staminate flowers are only open for one day and pistillate flowers are only receptive for one day. The corollas, stamens and pistils of watermelon flowers are all yellow.

Watermelons are entomophilous plants that are primarily pollinated in cultivation by honeybee (*Apis mellifera* L.) (Maynard, 2001). Watermelon entomophily is a symbiotic relationship between the insect and plant as plants receive pollination and bees obtain pollen (which is the only protein source for honeybees) and nectar. Pollination is facilitated by the production of a sticky substance on the surface of the pollen grains called pollenkitt. Pollenkitt enables adherence to floral visitors in order to disseminate pollen and is primarily seen in entomophilous angiosperms (Dobson et al., 1996). Pollenkitt is comprised mostly of lipids which also play a role in attraction of pollinators (Dobson, 1988; Pernal and Currie, 2002). Poor pollination may result in fruit abortion or misshapen fruit which is unmarketable (Rubatzky and Yamaguchi, 1997; Stanghellini et al., 1997; Walters, 2005).

Watermelon fruit is a specialized berry with thick skin (rind) known as a pepo (Rubatzky and Yamaguchi, 1997). Watermelon rind thickness is dependent on genotype and can be very thin to thick depending on the intended use of the watermelon. The edible portion of watermelon is the endocarp, although rind and seeds can and are consumed in certain geographic areas. Watermelon fruit are highly variable in size, shape, and color, and although all three are genetically determined, size can be altered through production practices. Watermelons may range from cylindrical to round and may be from two to over 90 kg at maturity (Wehner, 2006).

The largest watermelon recorded was 119 kg (Wehner, 2006). Most cultivars that are currently grown commercially in the U.S. range from 5.5 to 9.0 kg. Consumer preference has changed gradually over time and large fruit (> 11.3 kg) that were once preferred are now virtually unacceptable in most of the U.S. market. Average maturity for watermelon fruit is 80 days from transplant and 110 days from seed. Watermelon seeds are variable in size, shape, and color but are usually flattened, teardrop shaped and brown to black in color (Robinson and Decker-Walters, 1997). Optimum temperatures for seed germination range from 29.4-32.3°C (Olson et al., 2006). Watermelon plants are intolerant to frost and grow best when the average temperature is above 21.1°C (Rubatzky and Yamaguchi, 1997).

Watermelon Production Practices

Growing watermelons and producing optimal yields is dependent on many factors including fertilization, irrigation, soil management, and other cultural practices (Hochmuth et al., 2001a). The pH of the soil can greatly influence nutrient availability and crop growth. Watermelon plants can tolerate a range of pH values from 5.5 to 7.5 but 6.0 to 6.5 is optimum (Hochmuth et al., 2001a). Various forms of lime can be used to correct acidic soil pH, which is common in the highly weathered soils of the southeast. Calcium is important in the growth and development of watermelon because it is necessary to maintain the structural integrity of cell walls. Inadequate calcium in developing watermelon fruit can lead to a disorder known as blossom-end rot (BER) (Taylor and Locascio, 2004). Blossom-end rot occurs when tissue at the blossom-end of the fruit begins to collapse. This collapse is visualized as necrotic areas at the blossom-end which allow pathogens to enter the fruit and eventually leads to decay. Although BER is a calcium deficiency, factors such as inadequate or inconsistent irrigation, elemental competition, high salinity, high temperature and high or low transpiration may induce the problem (Taylor and Locascio, 2004).

Other elements such as nitrogen, phosphorus, potassium, magnesium, and micro-nutrients are also necessary for adequate fruit growth and plant development (Hochmuth et al., 2001a). The amount of these elements needed may vary by soil type and location. Soil testing should be performed in order to understand what the crop requirements are at a given location. Nitrogen, phosphorus, and potassium are generally the three most limiting nutrients and recommendations for the maximum amounts of nutrient addition for watermelon production in Florida are 168N-72.2P-140K kg/ha (Olson et al., 2006).

Irrigation amounts and frequencies required by the crop can vary depending on soil type, its water holding capacity, and environmental conditions (Rubatzky and Yamaguchi, 1997). A minimum of 93 cm of water per hectare is required to successfully grow watermelons (Rubatzky and Yamaguchi, 1997). Irrigation frequencies are important because some portions of growth and development require more water and are more sensitive to moisture stress than others. Water deficit during flowering and early fruit growth can have a greater negative impact on watermelon yield than water stress during other growth periods (Erdem and Yuksel, 2003).

Watermelon fruit growth and development is triggered by hormones produced by developing seeds, which are dependent on pollen transfer from staminate to pistillate flowers (Rubatzky and Yamaguchi, 1997). Pollination can be a yield limiting factor in watermelon production and in many areas, pollination by feral honeybees is no longer reliable due to the recent introduction of two parasitic mite species [*Acarapis woodi* (Rennie) and *Varroa jacobsoni* (Oudemans)] (Delaplane and Mayer, 2000). These mites have reduced feral and domesticated honeybee colonies by 90% and 30% respectively (Stanghellini et al., 1998; Ambrose, 1997; Harbo and Hoopinger, 1997). The reduction of feral honeybee populations necessitates the introduction of domesticated honeybees in large hectareage, high density plantings. Commercial

pollinators are available for rent or purchase and honeybees are the most readily available. Recommendations vary from one to five hives per hectare, which is dependent on hive strength (Olson et al., 2006). Adlerz (1966) reported that in diploid watermelon, a minimum of eight honeybee visits and 1000 pollen grains per pistillate flower were necessary for fruit set. Stanghellini (1997) reported similar findings of a minimum six visits per flower in diploid watermelon. Visitation rates lower than this resulted in significantly greater fruit abortion rates (Stanghellini, 1997).

Cultural practices used in watermelon production have changed over the years but the ones having the most effect are the use of polyethylene mulch and drip irrigation. Traditionally watermelons were grown on bare ground with either overhead or seep irrigation. The use of drip irrigation decreases the amount of water used and increases the use efficiency (Smajstrla et al., 2002). It also allows growers to target fertilizer applications directly to the plants through the use of soluble fertilizer that is pumped through the drip tape (Clark et al., 2005). The use of polyethylene mulch has many advantages including earliness, increased yields, increased profitability, irrigation and fertilizer retention, weed suppression, and fruit protection (Lu et al., 2003; Hochmuth et al., 2001b; Sanders et al., 1999; Lamont, 1993; Bryan, 1966). By covering the ground with black polyethylene mulch, soil temperatures are raised which increases early plant growth rate and thus early yield. Mulch has been shown to decrease harvest time in the spring as much as a month (Bryan, 1966). The earliness of the crop may greatly influence the profitability by allowing producers to enter the market before other producers or producing regions. The use of polyethylene mulch has also allowed for the adoption of soil fumigants which are essential for profitable commercial production in certain areas. The use of greenhouse-grown containerized watermelon seedlings has also increased which is due mostly to

increased seed prices and the benefit of significantly greater early yields (Nettles, 1963).

Implementing polyethylene mulch and the use of transplants has increased the success of producers in Florida, Georgia, and South Carolina as it allows them to enter the market before the July 4th holiday. Generally, watermelon movement in the U.S. is greater and the price is higher before this holiday (USDA, 2006). These three states accounted for 42% of the watermelon hectareage in the U.S. in 2005 (USDA-NASS, 2006).

Watermelon plant spacing has changed over the years; most likely due to adoption of other cultural practices that make production more efficient and a decrease in size preference by consumers (Hochmuth et al., 2001a). Current recommendations for watermelon spacing range from 1.1 to 2.7 m² per plant (Daniello, 2003, Olson et. al, 2006). Recent research has shown that greater yields per hectare may be achieved on polyethylene mulch with spacings as small as 1.0 m² per plant (Goreta et al., 2005; Sanders et al., 1999). Plant spacing is also dependent on plant architecture, as some genotypes have a more vigorous growth habit than others (Edelstein and Nerson, 2002; Kultur et al., 2001; Reiners and Riggs, 1997).

Watermelon Production Statistics

Watermelons are grown throughout the world in tropical and subtropical climates. World watermelon production has ranged from 81 billion kg in 2001 to 93.2 billion kg in 2004 (Arney et al., 2006). The world's largest producer of watermelons is China which usually accounts for over half of the world production (Arney et al., 2006). The U.S. generally ranks fourth in world production and produces on average 1.8 billion kg (Arney et al., 2006).

Florida consistently ranks among the top three watermelon-producing states in the nation in both hectares harvested and crop value and produces about 20% of the watermelons grown in the United States (USDA, 2005). Over the last five years, Florida harvested on average ten thousand hectares of watermelons annually worth an average of 72.1 million dollars. In Florida,

watermelons account for 3.9 % of the cash receipts for vegetables and 0.91% of the cash receipts for agriculture (USDA, 2002). Although watermelon hectareage in the U.S. has declined over the last five years, total production has remained stable as a result of increased production per hectare. One factor that has changed is the types of watermelons that are being produced.

Review of Triploid Watermelon

Background Information

Triploid seedless watermelon was first described in the United States in 1951 based on work that had been conducted in Japan since 1939 (Maynard and Elmstrom, 1992; Kihara, 1951). Seedless watermelons are produced by watermelon plants that are genetically triploid (3n). Watermelon plants are naturally diploid (2n). Triploid plants are grown from triploid seed which is produced by crossing a tetraploid (4n) female parent with a diploid male parent. The female parent is produced by treating a diploid seedling with colchicine, a chemical that binds to tubulin and inhibits the formation and activity of microtubules in plant cells. This inhibits the separation of chromosomes during mitosis which doubles the number of chromosomes in the plant and creates a tetraploid (Wehner, 2006). The triploid offspring are sterile which is why fruits produced by these plants have no seeds. Fruit growth and enlargement in watermelons is signaled by hormones produced by developing seeds. Since there are no developing seeds in triploid fruit, these developmental signals are obtained from pollen tube growth and ovule fertilization (Rhodes et al., 1997; Robinson and Decker-Walters, 1997; Maynard and Elmstrom, 1992). Ovules abort shortly after fertilization but may remain in the flesh as small rudimentary white seeds (Maynard and Elmstrom, 1992; Kihara 1951). Though fruit growth and development in triploids is signaled by pollen tube growth, triploid plants produce little if any viable pollen (Rhodes et al., 1997; Robinson and Decker-Walters, 1997; Maynard and Elmstrom,

1992). A diploid watermelon cultivar must be planted in close proximity to the triploid to provide sufficient viable pollen. This diploid plant is referred to as a pollinizer.

The first hybrid triploid watermelon cultivars produced by Kihara were finished in 1951 (Wehner, 2006). Triploid watermelon cultivars have been commercially available for nearly 35 years but interest from consumers and growers remained low until the late 20th century. There was little interest in early triploid cultivars due to erratic and poor performance in the field and high seed costs as compared to diploid cultivars (Maynard and Elmstrom, 1992). Seed for early triploid cultivars were quoted at \$135 per 1000 seed which was 900 times higher than hybrid diploid cultivars at the time (Maynard and Elmstrom, 1992). Reluctance in adoption of triploids may have also been due to the necessity of using transplants and the increased input costs associated with them. A survey of over 1300 people conducted in 1992 indicated that while 74% knew of seedless watermelons only 31% had ever purchased one (Wiemann, 1992). Marr and Gast (1991) surveyed consumers and indicated that they were willing to pay 50% more for seedless watermelons and that there was no differential preference between taste of seeded and seedless. The authors suggested that the response seen was on appearance alone as participants were shown cut fruit of both types. In 1990, Karst (1990) estimated that 5% of the U.S. watermelon market was seedless but they had the potential to gain up to 50% market share. Since this time, seedless watermelons have gained popularity in the marketplace and also with watermelon growers. Cultivars that would be considered modern cultivars became commercially available in the early 1990s and several are still considered industry standards today (Maynard and Elmstrom, 1992).

Seedless Watermelon Market Share

Before 2002, watermelon market data was not separated into seedless and seeded categories so it is difficult to locate accurate statistics on the production of either. To date, there

is still no separation of the production area grown in the U.S. but there are now reliable data on seedless and seeded watermelons sold in the U.S. Seedless watermelons accounted for 78% of the watermelons sold in the U.S. in 2006 which is up from 50% of the U.S. market in 2002 (USDA, 2006). The portion of Florida's watermelon production that is triploid has increased from 42% in 2002 to 79% in 2006 (USDA, 2006). There is some incentive for growers to produce seedless watermelons because they typically receive 4.5 to 11.0 cents per kg premium and it has become increasingly harder to market seeded watermelons since their market share has decreased (USDA, 2006).

Triploid Seedless Watermelon Production

The cultural practices used in triploid seedless watermelon production are similar to diploid production with respect to plant spacing, fertilization, and irrigation. Average maturity is also similar between the two. Growth habit of triploids is similar to diploids but there are also genotypes that exhibit compact growth and plant spacing may need to be altered to produce fruits of desirable size. Triploid seed are more difficult to germinate than diploids and require precise environmental conditions. While diploid cultivars may germinate in as low as 12.7 °C, triploid seed will not germinate below 26.6 °C and optimum temperature is between 29.4 °C and 32.2 °F (Hochmuth et al., 2001a). Seed coat adherence is also a problem with triploids and may negatively affect seedling growth. To avoid this, seeds must be planted with the radicle end up at 45° to 90° (Maynard and Elmstrom, 1992; Maynard, 1989). With these requirements and the high cost of triploid seed, the use of transplants in spring triploid production is necessary as soil temperatures are too cold for direct seeding.

Pollination is also a necessity in triploid plants and the introduction of domesticated pollinators, such as honeybees or bumblebees, may be more important in triploid production (Walters, 2005). Stanghellini (1997) and Alderz (1966) both reported that pistillate diploid

flowers required a minimum of six to eight honeybee visits for optimal fruit set and visitation rates lower than this significantly increased fruit abortion. Walters (2005) conducted experiments on triploid watermelons in which honeybee visitations were controlled in order to determine the number of visits necessary for optimal fruit set. Research plots contained a 33% pollenizer ratio in order to mimic commercial production. Findings suggested that between 16 and 24 honeybee visits were required to achieve maximum seedless watermelon fruit set. These visitation numbers are two to four times higher than what is needed in diploid plants and it was suggested that this is due to a dilution of viable diploid pollen with non-viable triploid pollen.

The most crucial difference between diploid and triploid production systems is the addition of the diploid pollenizer in the triploid field. Kihara (1951) suggested that one diploid plant should provide enough pollen to achieve adequate fruit set in four to five triploid plants. By these recommendations, 16 to 20% of the plants per hectare should be diploid. Until 2001, there were no scientific data on which pollenizer ratio would maximize seedless watermelon yield. Maynard and Elmstrom (1992) indicated that a pollenizer ratio of 33% had produced acceptable seedless yields and other sources recommended ratios of 20 to 33% (Robinson and Decker-Walters, 1997; Rubatzky and Yamaguchi, 1997). The method for introducing diploid watermelons into the field at this time was to plant solid rows of diploid plants between rows of triploid plants. The diploid cultivar must be different than the triploid in size, shape, or rind pattern in order to facilitate efficient harvest. NeSmith and Duval (2001) used distance of a triploid row from a pollenizer row to make inferences on pollenizer frequencies. ‘Genesis’ triploid was used and ‘Ferrari’ was used as the pollenizer. Their results showed the greatest seedless yields in rows 3.0 m from the pollenizer row with yields in rows farther away declining. Yield estimates produced by NeSmith and Duval (2001) suggested that the greatest seedless

watermelon yield per hectare would be achieved with a 1:4 pollenizer to seedless ratio when 1.5 m between-row spacing is used. In this scenario, pistillate triploid flowers would never be more than 3.8 m away from a staminate diploid flower.

Fiacchino and Walters (2003) conducted the same type of experiment but used isolated fields with different pollenizer ratios. The plot design used by Fiacchino and Walters more closely resembled a commercial watermelon field. Plots consisted of raised, plastic mulched beds with 1.5 m between-row spacing. This experiment also used dedicated rows of pollenizers at ratios of 11, 20, and 33%. In this study, multiple pollenizer cultivars were used to determine if cultivar, as well as frequency, had an effect on seedless watermelon yield. ‘Millionaire’ triploid was used and ‘Crimson Sweet’ and ‘Fiesta’ were used as pollenizers. Fiacchino and Walters (2003) found plots containing a 33% pollenizer ratio did not have greater yield than those with 20%, but both 20 and 33% plots had greater seedless yields than the plots with an 11% pollenizer ratio. Though there was no difference in yield between the 20 and 33% plots, a field with 20% pollenizer ratio would have greater seedless watermelon yield on a per hectare basis due to a higher number of triploid plants. These researchers also reported a significant difference in seedless yield between the two pollenizer cultivars used, with plots pollenized by ‘Crimson Sweet’ having greater yield. When ‘Fiesta’ was used as a pollenizer cultivar there was significantly greater hollow heart disorder present in the seedless watermelons.

Previous to Fiacchino and Walters (2003), pollenizer choice was based on marketing concerns and not how it affected the triploid crop. In this study, watermelon yields per hectare were greatest in plots where pistillate triploid flowers never exceeded 3.8 m away from a staminate diploid flower, which is in agreement with Nesmith and Duval (2001).

The consensus between Nesmith and Duval (2001) and Fiacchino and Walters (2003) reinforced the non-scientific recommendations of Maynard and Elmstrom (1992) who suggested that a 1:2 pollenizer to triploid ratio. The maximum distance between pistillate triploid and staminate diploid flowers was never greater than 3.9 or 4.5 m because 2.75 and 3.0 m between-row spacing's were used (Maynard and Elmstrom, 1989, 1992). This research was conducted when seedless watermelons held less than 50% of the U.S. market so the production scheme of using dedicated pollenizer rows allowed producers to be diversified in the marketplace. During the early years of commercial seedless production, it may have been more economically beneficial for growers to grow at a 1:1 pollenizer to triploid ratio as seedless melons held such a small market share. With the growth in popularity of seedless watermelons and their increased market share, it has become less desirable to grow seeded watermelons. Under previous triploid production schemes, as much as a third of a grower's hectareage needed to be in diploid watermelons.

A new cultural management system has recently been developed that allows for an increase in the number of triploid plants per hectare. New diploid cultivars have been developed specifically for the role of pollenizer and these cultivars, commonly called special pollenizers, are designed to be planted within the row of triploid plants without changing in-row spacing. Special pollenizer cultivars became available in the early 2000s and were used on large hectareage beginning in 2004. As there is no dedicated space for the pollenizer, triploid plants can be planted at 100% stand. Common practices are to punch plant holes and transplant the field solid with triploid seedlings then go back through the field and transplant pollenizers between triploid seedlings at the appropriate density. Diploid cultivars produce flowers sooner than triploid cultivars so transplanting the pollenizer several days later may more closely synchronize

blooming in the two types of plants (Freeman and Olson, 2007). This system of pollenizer arrangement increases triploid plant numbers by 20 to 33% per hectare, thus increasing the number of seedless fruits harvested per hectare. Most of these pollenizer cultivars are not intended to be harvested which, allows for harvest of only seedless fruit. This can avoid the confusion of having multiple types of harvestable watermelons in the field and the added labor costs of multiple harvests. Though most special pollenizers were not intended to be harvested, the following cultivars produce marketable fruit: ‘Jenny’, ‘Minipol’, ‘Pinnacle’, ‘Polimore’. Any small fruited diploid cultivar could be used as a pollenizer if it produces adequate staminate flowers and pollen. Some producers may have a market for the seeded pollenizer fruits and may be able to benefit economically by using one of these cultivars.

There are two different types of special pollenizer cultivars available; highly branched plants with reduced foliage and thin vines or short inter-node bush-type plants. The thin-vine types have foliage and vines that are smaller than standard watermelon plants by varying degrees. These types also exhibit some degree of increased branching which increases the number of terminals and therefore the number of male flowers produced. The reduction in vine and foliage size is intended to reduce the negative effects that may occur when decreasing the area per plant by introducing the pollenizer in-row. Thin-vine special pollenizers currently available are: ‘Increase’ (Southwestern Seeds), ‘Jenny’ (Nunhems USA, Inc., Acampo, CA), ‘Minipol’ (Hazera Seeds, Inc., Coconut Creek, FL), ‘Patron’ (Zeraim Gedera Seed Co, Ltd., Palm Desert, CA), ‘Pinnacle’ (Southwestern Vegetable Seed, LLC., Casa Grande, AZ), ‘Polimore’ (Hazera Seeds, Inc., Coconut Creek, FL), ‘Sidekick’ (Harris Moran Seed Co., Modesto, CA), ‘SP-1’ (Syngenta Seeds, Inc., Boise, ID), ‘SP-4’ (Syngenta Seeds, Inc., Boise, ID). The bush-type pollenizers have a compact growth habit with short internodes and a

branching pattern more similar to standard watermelons. These cultivars also have a nearly entire leaf with highly reduced lobes. Bush-type pollenizers currently available are ‘Companion’ (Seminis, Inc., Oxnard, CA) and ‘Stud’ (Abbott and Cobb, Inc., Feasterville, PA).

Most fruits produced by special pollenizers are small and usually weigh less than 2.5 kg (Freeman and Olson, 2007). Special pollenizer cultivars produce fruits with one of two types of rind patterns, solid grey to light green or light green with a dark green stripe, and vary in shape from round to oblong and blocky. As with the dedicated-row pollenizer arrangement, a pollenizer cultivar that has fruits easily distinguishable from the seedless fruits should be chosen. Most special pollenizer fruits are substantially smaller than medium and large seedless fruits which aids in their distinction. However, when personal size seedless watermelons (< 3.2 kg) are produced, a cultivar with a distinct rind should be chosen as separation based on size may not be possible. The thin-vine pollenizers are recommended to be planted at a 1:3 pollenizer to triploid ratio while the bush-type cultivars are recommended at a 1:2 pollenizer to triploid ratio.

Pollinator Activity and Preference

Cultivated watermelon crops require pollination and domesticated honeybees (*Apis mellifera* L.) are the most important pollinator (Free, 1993). Walters (2005) illustrated that increased honeybee visitation to pistillate triploid flowers is required for fruit set due to the dilution of viable diploid pollen with non-viable triploid pollen. Both triploid and diploid watermelon plants produce visually similar staminate flowers and triploid flowers produce pollen although it is not viable.

Honeybee foraging habits are controlled by both visual and olfactory cues but it is unlikely that they can visually distinguish between triploid and diploid flowers (Butler, 1951; von Frishch 1967). These cues are processed during pre-alighting inspection and determine whether the flower will be foraged. It has been shown that floral structures such as petals, sepals,

gynoecium, and pollen have distinct volatile emissions that are species and genotype specific (Dobson et al., 1996; Dobson, 1991; Dobson et al., 1987). The volatiles from pollen (which are derived from pollenkit) are the most important factors when honeybees decide to forage a flower or not (Pernal and Currie, 2002). Although a hierarchy of pollen preference was shown by Olsen et al. (1979), no differences were observed between the species used by Pernal and Currie (2002). The olfactory cues from pollen also appear to be quantitative and decreasing emissions throughout the day indicate less reward to foragers (Dobson et al., 1996).

It has been suggested that pollen odor may be distinct between male-fertile and male-sterile flowers of the same species (Dobson et al., 1996). Preference for male-fertile over male-sterile potato flowers has been shown in bumblebee which may have been due to pollen odor (Arndt et al., 1990; Batra, 1993). Wolf et al. (1999) conducted pollinator preference experiments in which honeybees were placed in a field with two watermelon cultivars, one *Citrullus colocynthis* accession and one *C. colocynthis* x *C. lanatus* hybrid 'BAG', with the number of visitations being recorded. Significantly greater bee visitation was seen in the watermelon cultivar BAG and the *C. colocynthis* accession, neither of which had nectar volume, pollen quantity, or flower size that was different from the other genotypes tested. Wolf et al. (1999) found a positive correlation between honeybee visitation and sugar concentration of nectar which is what greater visitation was attributed to. This conclusion is in contrast to Pernal and Currie (2002) who illustrated that pollen odor was more important than forage quality for honeybees.

Pollen Effects

Triploid seedless watermelon fruit growth and development is dependent on pollination of the pistillate triploid flowers with viable diploid pollen (Kihara, 1951; Maynard and Elmstrom, 1992). It has been shown in other genera that genotype can have a significant effect on pollen viability and that variations in viability can affect the reproductive success of the individual

receiving the pollen. Parzies et al. (2005) reported significant differences in pollen viability within and between species of barley however, these differences were only evident after the pollen was subjected to incubation treatments. These results indicate more of a difference in pollen longevity as opposed to viability.

Fortescue and Turner (2004) investigated the pollen viability within and among multiple banana species and among ploidy levels within a single species. This study reported significant differences in pollen viability between species, within species, between ploidy levels within a single species, and within ploidy levels of a single species. The differences in pollen viability were as great as 100% between cultivars of the same ploidy level and same species (Fortescue and Turner, 2004). Pollen source has been investigated in mandarin orange and significant effects were reported on fruit quality parameters. Vithanage (1991) investigated the pollen donor effects on 'Ellendale' mandarin using six different pollenizers. Vithanage reported that fruit weights of 'Ellendale' were significantly greater when 'Murcott' and 'Emperor' were used as pollenizers. Wallace and Lee (1999) conducted experiments in which 'Ellenor' mandarin was pollinated by 'Murcott', 'Imperial', and 'Ellenor'. This study found that fruits from 'Ellenor' had significantly greater size and sugar content when 'Murcott' was used as a pollenizer. Lavi et al. (1996) reported significant differences in pollen viability among cultivars of macadamia but found no correlation between pollen viability and fruitlet retention.

Nikkanen et al. (2000) illustrated that pollen viability within *Picea abies* was significantly effected by individual pollen donor and germination conditions, and that there was an interaction between these two factors. These results show that individuals of the same species, within a geographic area, may require specific environmental conditions for reproductive success. Brevis et al. (2006) reported significant differences in pollen viability among rabbiteye blueberry

cultivars, although all cultivars had a high average viability. This study suggested that while pollen viability was statistically significant, it may not be biologically significant and is not thought to contribute to reproductive failure in blueberry. Pollen viability data presented by Brevis et al. (2006) was similar to previous findings in rabbiteye and southern highbush blueberry (Cockerham and Galleta, 1976; Lang and Parrie, 1992).

There is only one published study on pollen viability in the family Cucurbitaceae. Nepi and Pacini (1993) investigated various aspects of pollination in a single cultivar of *Cucurbita pepo*. They reported that average pollen viability at anthesis was 92% which decreased to 75% within 6 h and further decreased to 20% at 11 h after anthesis. This decrease in viability was attributed to dehydration of the pollen grain. There is no published data on pollen viability of watermelon cultivars.

Variation in pollen production has been reported between genera and among species of the same genera in the Poaceae, and among species of the same genera in the Cupressaceae (Hidalgo et al., 1999; Prieto-Baena et al., 2003). In diploid watermelon, Stanghellini and Schultheis (2005) investigated 27 cultivars and found significant differences in production of pollen grains per flower and pollen grains per plant. Pollen production ranged from 134,206 grains per plant per day for ‘Jamboree’ to 264,589 grains per plant per day for ‘Summer Flavor 800’.

The time period over which diploid cultivars produce pollen may be as important as the amount of pollen produced. Diploid watermelon cultivars begin to produce staminate flowers about seven days before triploid plants begin to flower and it is essential that the diploids continue producing staminate flowers throughout triploid fruit set (Freeman and Olson, 2007). Significant differences in total staminate flower production, as well as flowering longevity, have been reported in diploid watermelon cultivars (Freeman and Olson, 2007; Stanghellini and

Schultheis, 2005). Greater staminate flower and pollen production by a diploid cultivar may improve its performance as a pollenizer by reducing the dilution effect of viable pollen that is created by triploid plants.

Competition

Competition has been defined as the negative interaction between two organisms (Connell, 1990). In plants, this competition is for light, water, nutrients, and space, and can be inter-specific (between two species) or intra-specific (between individuals of the same species). Inter-specific competition from weed species as well as intra-specific competition from neighboring crop plants can reduce the survivability of plants and the yield and quality of plant products (Firbank and Watkinson, 1990). Intra-specific competition in cropping systems is regulated by planting density which is intended to maximize production per unit area. Maximum production per unit area occurs when plant population and yield per plant are in correct proportions. Experimental models have shown that plant yields increase with plant density to a maximum point and then plateau or decline as density continues to increase (Holliday, 1960). Intra-specific competition and plant density can also be used as tools to manipulate yield parameters such as size distribution of fruit (Motsenbocker and Arancibia, 2002; Reiners and Riggs, 1999; Sanders et al., 1999).

Intra-specific competition of crop plants is investigated through studies that examine the effect of planting density or spatial arrangement on crop yield. In vining cucurbits such as muskmelon (*Cucumis melo* L.), pumpkin (*Cucurbita pepo* L.), and watermelon, it has been shown that increasing plant density increases total yield but decreases yield per plant (Ban et al., 2006; Duthie et al., 1999a; Duthie et al., 1999b; Goreta et al., 2005; Kultur et al., 2001; Maynard and Scott, 1998; Reiners and Riggs, 1999, 1997; Sanders et al., 1999). The increases in yields per unit area reported in these studies were due to increased fruit numbers per unit area.

Although the use of in-row diploid pollenizers has increased, it has not been determined if the increased competition on neighboring triploid plants will be deleterious. When planted at a 1:3 pollenizer to triploid ratio, the pollenizer will directly impact 2/3 of the plants per hectare by decreasing in-row spacing by 1/4. This would reduce area per plant from 2.2 m² to 1.6 m² for plants grown on 2.4 m between-row spacing and 0.9 m in-row spacing. Though reduced, this area is still greater than the 1.0 m² per plant which has been shown to produce greatest watermelon yields per hectare (Goreta et al., 2005; Sanders et al., 1999). Results from other studies in watermelon do not provide insight as the phenotype of the pollenizer and triploid are different. As pollenizer growth will not impact all triploid plants per hectare, the appropriate study to investigate pollenizer competition effect is the neighborhood (area of influence) study in which the performance of a single individual is measured as a function of distance from the competitor (Radosevich and Roush, 1990).

CHAPTER 3 CHARACTERISTICS OF DIPLOID POLLENIZERS FOR USE IN TRIPLOID WATERMELON PRODUCTION

Introduction

With triploid seedless watermelons now occupying 78% of the United States market, it is suspected that the use of in-row pollenizer cultivars will increase (USDA, 2006). There are no published studies that compare important characteristics of diploid watermelon pollenizers such as staminate flower production, flowering period, and fruit production. Pollenizer flower production may be a strong indicator of how a cultivar will perform and flowering period is critical to the type of production system the pollenizer is used in. Using a pollenizer cultivar with low fruit production and easily distinguishable fruit could also increase efficiency in harvesting operations. The objectives of this study were to determine staminate flower production, flowering period, and quantity and size of fruit production of several commercially available diploid pollenizers.

Materials and Methods

Experiments were conducted at the North Florida Research and Education Center, Quincy, FL, and the North Florida Research and Education Center-Suwannee Valley, Live Oak, FL. In Quincy, the soil type was a Norfolk Loamy Fine Sand (fine-loamy, kaolinitic, thermic, Typic Kandiudults) and in Live Oak the soil was a Lakeland fine sand (thermic, coated Typic Quartzipsamments). At both locations, the experiment was arranged as a randomized complete block design with four replications. Transplants were produced in a greenhouse at Quincy in expanded polystyrene flats of the inverted pyramid design which were $3.75 \times 3.75 \times 6.25$ cm using soil-less media. Prior to the laying of the mulch, pre-plant fertilizer was applied at recommended rates and incorporated into the soil (Olson et al., 2004). All fertilizer was applied pre-plant in Quincy and one-fourth was applied pre-plant in Live Oak. Weekly fertigation was

used to apply the remainder of the fertilizer in Live Oak. Watermelon plants were irrigated as needed.

On 1 Apr. 2005, 5-week-old seedlings of ‘SP-1’, ‘Companion’, ‘Jenny’, and ‘Mickylee’ were transplanted into raised beds fumigated with methyl bromide and chloropicrin (67/33) and covered with black polyethylene mulch. In Live Oak, plots consisted of two rows; beds were 0.6 m wide by 10 m long on 2.1 m centers, with in-row spacing of one meter. Pollenizer cultivars were planted with the triploid cultivar ‘Tri-X 313’. A pollenizer plant was planted at the beginning and end of each plot, and between every third and fourth ‘Tri-X 313’ plant in the plot. Plots at Live Oak consisted of 18 triploid plants and eight pollenizers. In Quincy, plots consisted of two rows; beds were 0.9 m wide by 13.2 m long on 2.4 m centers, with an in-row spacing of one meter. ‘Tri-X 313’ was also used in Quincy and placement and spacing of pollenizer cultivars were the same as for Live Oak. Plots at Quincy consisted of 24 triploid plants and 10 pollenizers.

Plants were sampled after the onset of male flowering by pollenizer cultivars, and data were collected twice a week at both locations. Once 70% of pollenizer plants in each plot had at least one open male flower (flowering threshold), the plot was considered to have begun flowering. Numbers of male flowers per plant were recorded from the beginning of flowering (29 Apr.) until the end of the fruit set period (3 June). Early season flower counts were obtained by counting open male flowers on all pollenizer plants in each plot. However, after plants began to vine heavily (making flower counting difficult), flower counts were obtained from a single row in each plot. Fruit from pollenizers were harvested at or near maturity and weighed on two dates per location. ‘Tri-X 313’ was not harvested for yield and only served to provide intra-specific competition for the pollenizers. Without this competition, growth habit of the

pollenizers would be different and not reflect flower counts that would be seen under commercial production conditions.

Statistical analysis was performed using the GLM procedures of SAS (SAS Institute, Inc., Cary, NC). For statistical analysis, location was added and the experiment was analyzed as a factorial experiment with two factors, location and cultivar. If an interaction was present, LSMEANS analysis and LSD were used to explain results, otherwise means separation was performed using Duncan's multiple range test.

Results

The first pollenizer cultivar to reach the threshold for flowering in all plots was 'SP-1', on 2 May, 2005 in Quincy. All cultivars had reached the flowering threshold by 10 May and 12 May for Quincy and Live Oak, respectively. Peak fruit set by 'Tri-X 313' began 14 May and ended 3 June at both locations. By 1 June, 'Tri-X 313' had set all commercially harvestable fruit, and by 10 June, mature 'Tri-X 313' melons were present at both locations. Location did not significantly affect male flower counts, and there was no significant interaction between location and cultivar ($P > 0.05$). Data were combined over locations and analyzed.

On 29 Apr., 'SP-1' produced 0.65 flowers per plant (fpp), which was not significantly different from 'Companion' or 'Mickylee' which had 0.65 and 0.21 fpp, respectively (Fig. 3-1). 'Jenny' had 0.12 fpp which was significantly lower than that of 'Companion' or 'SP-1', but not 'Mickylee'. On 3 May, 'SP-1' had 0.73 fpp which was greater than 'Companion' at 0.52 fpp or 'Jenny' at 0.38 fpp. 'Mickylee' had 0.25 fpp which was significantly lower than that of 'Companion' or 'SP-1', but not 'Jenny'. On 9 May, 'Jenny' had 1.95 fpp which was greater than 'Companion' at 1.00 fpp. However, fpp for 'Jenny' was not different than 'SP-1' or 'Mickylee', which were 1.65 and 1.50 fpp, respectively. On 13 May, 'SP-1' had 6.36 fpp which was not different than 'Jenny' at 5.76 fpp or 'Mickylee' at 5.52 fpp, however, 'SP-1' was significantly

higher than ‘Companion’, 4.75 fpp. On 16 May (‘Tri-X 313’ had begun producing female flowers), ‘SP-1’ had greater flower counts compared to the other pollenizer cultivars; ‘SP-1’ had 9.72 fpp compared to 7.88, 7.56, and 7.5 fpp for ‘Mickylee’, ‘Companion’ or ‘Jenny’, respectively. This trend continued throughout the remainder of the season as most mid (16 May) to late (1 June) season male flower counts showed that ‘Companion’, ‘Jenny’, and ‘Mickylee’ had similar numbers of flowers but were less than ‘SP-1’. Flower numbers increased and peaked on 26 May when ‘SP-1’ had 35.5 fpp which was greater than ‘Companion’, ‘Mickylee’, and ‘Jenny’ which had 14.50, 13.80 and 12.80 fpp, respectively.

Analysis of pollenizer fruit counts per plot indicated a significant location effect and an interaction between location and cultivar (Table 3-1). Distribution of fruit per plant by cultivars was slightly different at the two locations (Table 3-2). At Quincy, fruit set was greater for ‘SP-1’, with 5.8 melons per plant (mpp) compared to all other cultivars. ‘Companion’ set the least number of fruit, with 1.8 mpp, which was less than ‘Jenny’ at 4.3 mpp or ‘Mickylee’ at 3.7 mpp. At Live Oak, ‘Jenny’ produced 3.4 mpp and ‘SP-1’ produced 3.3 mpp, which were greater than ‘Mickylee’ at 1.7 mpp or ‘Companion’ at 1.2 mpp. The interaction between cultivar and location is due to the different rankings of fruit production by cultivar at each location. In Quincy, fruit production by ‘Jenny’ and ‘Mickylee’ were not significantly different, however both produced less fruit than ‘SP-1’. At Live Oak, fruit production by ‘SP-1’ and ‘Jenny’ were not significantly different but both produced more fruit than ‘Mickylee’. At Quincy, ‘Mickylee’ produced more fruit than ‘Companion’ but at Live Oak fruit production was similar between the two.

Data analysis indicated that location had a significant effect on pollenizer fruit weight, and that fruit weights in Live Oak (avg. = 2.80 kg) were lower than those at Quincy (avg. = 3.06 kg)

($P \leq 0.05$). There was no interaction between location and cultivar. ‘Mickylee’ had the highest fruit weight, 4.34 kg/fruit, which was higher than for ‘Companion’ or ‘Jenny’ which were 2.61 and 2.56 kg/fruit, respectively (Table 3-3). ‘SP-1’ had the lowest fruit weight at 2.21 kg/fruit, which was less ($P \leq 0.05$) than of all other pollenizer cultivars.

Discussion

‘SP-1’ and ‘Companion’ produces non-edible fruits while ‘Jenny’ and ‘Mickylee’ produce edible melons. When used as pollenizers, fruit production is not desirable because melons from pollenizers can confuse the harvesting process (with mixing of seeded and seedless fruit) as well as hinder harvesters from moving efficiently through the field. ‘Companion’ and ‘Mickylee’ produce easily distinguishable melons based on the grey to pale green color and no rind pattern. ‘SP-1’ produces a light green melon with very thin light green broken stripes while ‘Jenny’ produces melons that have a medium green background with dark green stripes. All four pollenizer cultivars would be easily distinguishable by size from most commercial melons, other than personal-size seedless watermelons which generally weigh between 1.8 and 2.2 kg.

All four pollenizers had male flowers present during peak seedless watermelon fruit setting. Results were similar to Dittmar et al. (2005) with ‘SP-1’ producing significantly more male flowers than ‘Companion’, ‘Jenny’, or ‘Mickylee’. Although flower production by ‘SP-1’ was highest other cultivars may also provide more than enough pollen to accomplish optimal seedless watermelon fruit set. Stanghellini and Schultheis (2005) reported that pollen production is also variable between diploid watermelon cultivars so flower production may not be completely indicative of a cultivars male reproductive output. All cultivars appear to be viable options for use as pollenizers in triploid watermelon production as all were producing male flowers during fruit set of seedless watermelons.

Table 3-1. Analysis of variance for pollenizer fruit weight and fruit per plant at Quincy and Live Oak FL during 2005.

Source	df	MS	<i>P</i> -value
Fruit weight			
Replication	3	0.10217	0.848
Location	1	2.67961	0.015
Cultivar	3	35.3234	<0.0001
Location*Cultivar	3	0.17825	0.708
Error	21	0.38212	
Fruit per plant			
Replication	3	0.33208	0.207
Location	1	17.7012	<0.0001
Cultivar	3	14.2712	<0.0001
Location*Cultivar	3	1.75791	0.0006
Error	21	0.20113	

Table 3-2. Interaction effect of location and watermelon pollenizer cultivars on fruit per plant at Quincy and Live Oak FL during 2005.

Location × cultivar	Fruit (no./plant)	
Quincy	SP-1	5.8
	Jenny	4.3 **
	Mickylee	3.7 ^{NS}
	Companion	1.8 **
Live Oak	Jenny	3.4
	SP-1	3.3 ^{NS}
	Mickylee	1.7 **
	Companion	1.2 ^{NS}

^{NS}, *, ** non-significant, or significant at $P \leq 0.05$ or $P \leq 0.01$, Least Squares Means analysis.

Table 3-3. Main effects for pollenizer fruit weights combined over experiments conducted in Quincy and Live Oak, FL, during 2005.

Cultivar	Avg fruit wt (kg)
Mickylee	4.34 a ^z
Companion	2.61 b
Jenny	2.56 b
SP-1	2.21 c

^z Means followed by the same letter are not significantly different at ($P \leq 0.05$) by Duncan's multiple range test.

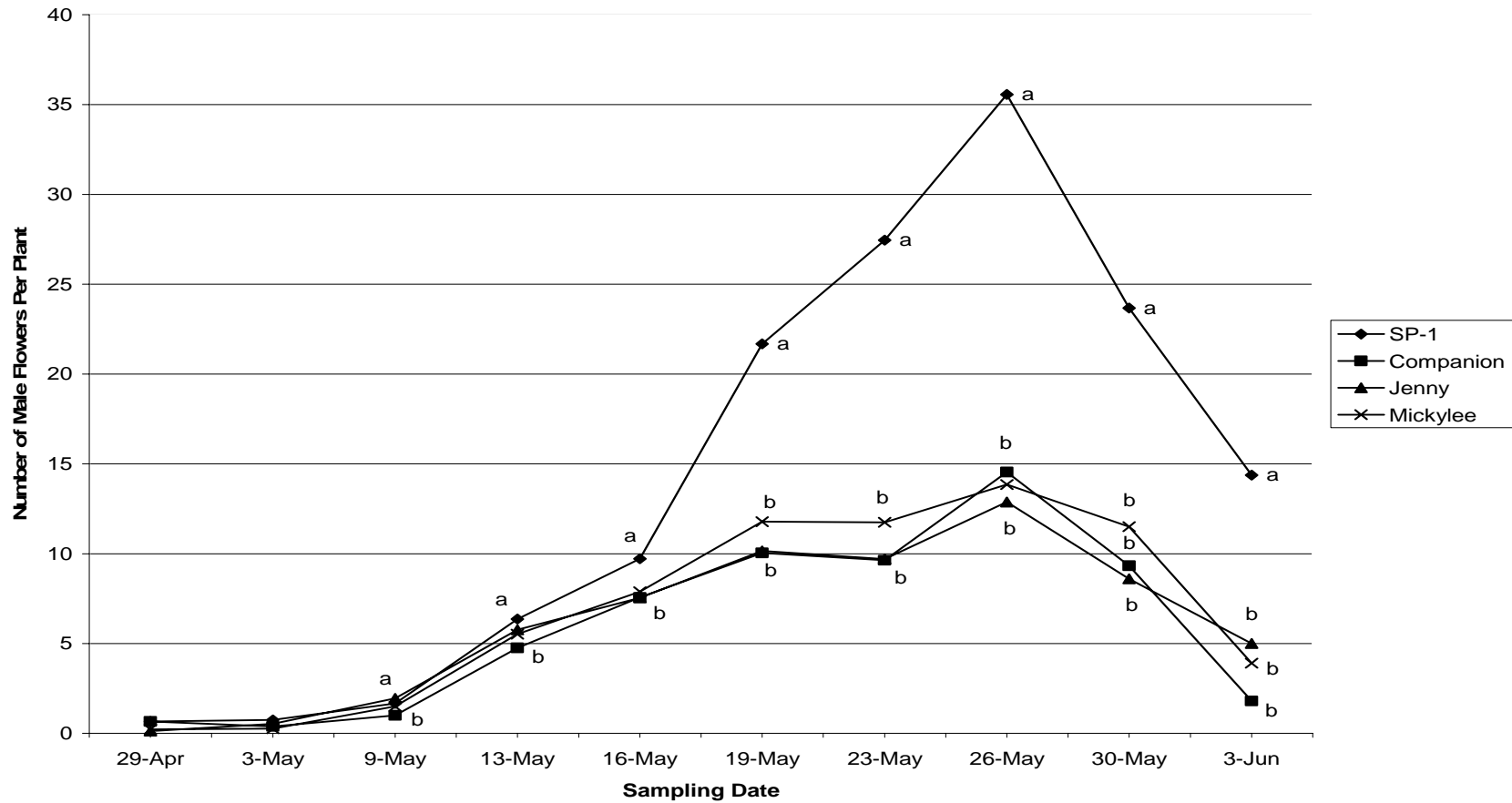


Figure 3-1. Pollenizer staminate flower counts combined over Quincy and Live Oak, FL, during 2005. Means followed by the same letter are not significantly different at ($P \leq 0.05$) by Duncan's multiple range test.

CHAPTER 4 POLLEN VIABILITY OF DIPLOID WATERMELON POLLENIZER CULTIVARS

Introduction

Pollen produced by diploid watermelon pollenizers is important because it is necessary for fruit set and flesh fill in associated triploid watermelon crops (Kihara, 1951; Maynard and Elmstrom, 1992). Viable pollen produced by pollenizers is diluted with non-viable triploid pollen which increases pollinator visitation rates required by pistillate triploid flowers. Pollenizers with poor pollen viability could further increase required visitation from pollinators and reduce stigmatal area available to viable pollen; both of which could have negative effects on seedless watermelon yield and quality. Significant differences in pollen viability have been reported between cultivars of the same species in several genera. The objective of this project was to determine pollen viability of four pollenizer cultivars to investigate possible effects on performance.

Materials and Methods

On 3 Apr. and 1 Aug. 2006, 4-week old watermelon seedlings were transplanted into raised beds. The beds were covered with black polyethylene mulch in the spring and white polyethylene mulch in the fall. Experiments were performed at the North Florida Research and Education Center (NFREC) in Quincy, FL. Soil type present at NFREC is Norfolk loamy sand (fine-loamy, kaolinitic, thermic Typic Kandiudults). Experimental design both seasons was a randomized complete block with four replications. Four diploid pollenizer cultivars, ‘Companion’, ‘Jenny’, ‘Mickylee’, and ‘SP-1’, were used to determine pollen viability. Experimental plots were 4.57 m long with an in-row spacing of 0.91 m and between-row spacing of 2.43 m. Three seedlings were planted in each plot. Fertilization, irrigation, and pesticide

application practices recommended by the University of Florida Institute of Food and Agricultural Sciences were followed (Olson et al., 2006).

Sampling was initiated on 17 May and samples were taken on 24 May and 31 May in the spring. Sampling was initiated on 31 Aug. and samples were taken on 7 Sept. and 14 Sept. in the fall. Sampling was initiated when other triploid watermelons at NFREC that were transplanted on the same dates began to set fruit. The sampling period was scheduled to mimic peak fruit set in triploid watermelons. The fruit set in this time frame would be the majority of the fruit that would be available for commercial harvesting schemes that are typical for FL.

On sampling dates, watermelon flowers were removed from the plant before anthesis. This was to insure that pollinators would not remove pollen and an adequate supply would be available for analysis. Three flowers were removed from each plot and placed into plastic cups and covered to exclude pollinators. Flowers were taken to the lab and allowed to open. After anther dehiscence had occurred (verified with a hand lens) sample analysis was initiated. Pollen was removed from the anthers and placed on a slide. Viability was determined using the diaminobenzidine (DAB) protocol for peroxidase activity in pollen (Dafni et al., 2005). Rodriguez-Riano and Dafni (2000) compared the results of four vital dyes versus pollen germination results and illustrated the superiority of peroxidase tests over other commonly used vital dyes. This test utilizes a dye that creates a color differential between viable and non-viable pollen.

Four 100 pollen grain sub-samples were analyzed from each plot using a compound microscope. A pollen grain was considered viable if it had turned dark brown or black. All pollen samples were analyzed on the same day pollen was collected. It is important with watermelon pollen that the pollen be thoroughly mixed with the dye on the slide in order to have

adequate contact between the pollen grains and dye. If large clumps of pollen are not broken up, dye may not infiltrate the pollen and false negatives may be observed. On each sample date heat killed pollen (two h at 80°C) was used to check the efficacy of the dye. New dye was prepared for each sample date. A square-root transformation was performed on the data and analysis of variance and means separation (Duncan's multiple range test) were performed using the GLM procedures of SAS (SAS Institute Inc., Cary, NC)

Results

There were no sampling dates in either season where pollenizer cultivar had a significant ($P \leq 0.05$) effect on pollen viability (Table 4-1). There was also no significant interaction between pollenizer cultivar and sampling date (Table 4-1). In the spring trial, sampling date had a significant ($P \leq 0.05$) effect on pollen viability with 31 May having greater average viability than 24, or 17 May (Table 4-2). Pollen viability on 31 May was 98.6% which was significantly greater than 97.4 or 97% for 17 May and 24 May, respectively. The average pollen viability over all cultivars and all dates for spring and fall were 97.7 and 97.9%, respectively. There was very little variation within the data and the coefficient of variation was never higher than 0.84%.

Discussion

The results of this study illustrate that there is no significant variation in pollen viability within the cultivars tested and that pollen viability is high and changes very little if any throughout the growing season, at least not within the critical period of triploid fruit set. The sample date in the spring with higher viability is more likely due to environmental conditions than cultivar characteristics. Freeman (2007) observed that seedless watermelon yield was significantly lower when 'Companion' was used as a pollenizer versus 'Jenny' or 'SP-1'. The results of this study suggest that pollen viability was not a contributing factor in the varying degrees of performance of these pollenizers. Factors such as floral attractiveness to pollinators,

timing and total production of staminate flowers, and pollen production may be more important characteristics of pollenizers.

The small amount of variation in pollen viability between the cultivars tested suggests that there may be little within the species or at least in cultivated varieties. Nepi and Pacini (1993) reported that the pollen viability of ‘Greyzini’ (*Cucurbita pepo* L.) averaged 92% which is similar to the findings of this study. Pollen viability appears to play no role in the performance of the pollenizers tested and may not be an important characteristic of pollenizer cultivars.

Table 4-1. Analysis of variance for pollen viability of watermelon pollenizer cultivars tested during the Spring and Fall of 2006 at Quincy, FL.

Source	<i>df</i>	MS	<i>P</i> -value
Spring			
Sampling Date	2	0.02827	0.003
Replication	3	0.00410	0.410
Cultivar	3	0.00856	0.124
Date*Cultivar	6	0.00281	0.669
Error	33	0.00415	
Fall			
Sampling Date	2	0.00302	0.367
Replication	3	0.00290	0.408
Cultivar	3	0.00281	0.422
Date*Cultivar	6	0.00257	0.519
Error	33	0.00292	

Table 4-2. Influence of diploid watermelon pollenizer cultivar on pollen viability at Quincy, FL during the Spring and Fall of 2006.

Pollenizer Cultivar	Pollen viability					
	17 May	24 May	31 May	31 Aug.	7 Sept.	14 Sept.
Mickylee	97.8 ^{NS z}	98.0 ^{NS z}	98.7 ^{NS z}	97.4 ^{NS z}	98.2 ^{NS z}	97.8 ^{NS z}
Companion	97.4	97.2	99.2	97.6	98.1	98.8
Jenny	97.3	97.2	98.3	97.8	97.7	97.0
SP-1	97.2	95.5	98.1	97.9	98.7	97.6
Date Means	97.4 b ^y	97.0 b	98.6 a	97.7 ^{NS y}	98.2	97.8

^z $P = 0.05$ Means are compared within the same column. ^y $P = 0.05$ Means from the same season are compared within the row.

CHAPTER 5
DIPLOID WATERMELON POLLENIZER CULTIVARS EXHIBIT VARYING DEGREES OF
PERFORMANCE WITH RESPECT TO TRIPLOID WATERMELON YIELD

Introduction

With seeded watermelons only holding about 20% of the U.S. market, there is interest in using pollenizers that do not require dedicated field space (USDA, 2006). Traditionally pollenizers occupied 20-33% of the land area in a field. New pollenizers have been developed to be planted in-row with triploid plants without altering spacing. There are now multiple in-row pollenizer cultivars available that exhibit varying growth habits. Previous research has reported differences in production and timing of staminate flowers by pollenizer cultivars. The seed costs of in-row pollenizers are greater than open-pollinated and hybrid diploids and most in-row pollenizers are not intended to be harvested. There is interest in using open-pollinated cultivars in-row to reduce input costs but it is unclear if acceptable yields will result. These experiments were conducted to determine if there was a difference in performance between in-row pollenizer cultivars and if a standard open-pollinated cultivar could be used in-row with similar success.

Materials and Methods

These experiments were performed at one location (Blackville, SC) in 2005 and three locations (Blackville, SC, Citra, FL, Quincy, FL) in 2006. The experimental design used was a randomized complete block with four replications. Experimental plots consisted of three raised bed rows that were spaced 2.43 m center-to-center and covered with black polyethylene mulch. Watermelon plants were spaced 0.91 m in-row. Replications consisted of three rows 116 m long with a 7.62 m buffer between replications. A diagram of the experimental layout is shown in Figures 5-1 and 5-2. The two outside rows were planted with ‘Tri-X Palomar’ and the interior row was planted with ‘Tri-X 313’ in 2005 and ‘Supercrisp’ in 2006. In 2005, pollenizer cultivars used were ‘Jenny’, ‘Mickylee’, and ‘SP-1’ with ‘Tri-X Palomar’ as a control. In 2006, the

pollenizer cultivars used were ‘Companion’, ‘Jenny’, ‘Mickylee’, ‘Patron’, ‘Pinnacle’, ‘Sidekick’, and ‘SP-1’ with ‘Tri-X Palomar’ as a control. Pollenizer seed sources are listed in table 5-5. In order to reduce pollen contamination from neighboring plots, an eight plant buffer (7.3 m) of ‘Tri-X Palomar’ was planted in the center row between each plot (Figure 5-2). It has been demonstrated that distance from a diploid pollenizer of 6.0 m or greater will greatly reduce the triploid fruit set (NeSmith and Duval, 2001). ‘Tri-X Palomar’ was chosen as the buffer cultivar and control plot “pollenizer” because it does not produce viable pollen and its rind coloration is distinctly different than the harvested cultivars, Tri-X 313 and Supercrisp. Eight triploid watermelon plants were transplanted into each plot including the control or check plot. Three plants of a pollenizer cultivar were planted in each plot except the control plot where ‘Tri-X Palomar’ was planted in place of a pollenizer. Control plots were in place in order to observe if pollen was moving from plot to plot. ‘Jenny’, ‘Mickylee’, ‘Patron’, ‘Pinnacle’, ‘Sidekick’, and ‘SP-1’ were planted at a 1:3 pollenizer to triploid ratio while ‘Companion’ was planted at a 1:2 pollenizer to triploid ratio. These ratios are recommended by producers of the various pollenizers. Three plants of the 1:3 ratio pollenizers, and four plants of the 1:2 ratio pollenizer were included in each plot in the same row as the harvested watermelon.

Soil type at the Edisto Research and Education Center (EREC) in Blackville, SC was Dothan loamy sand (fine-loamy, kaolinitic, thermic Plinthic Kandiudults). Soil type at the North Florida Research and Education Center (NFREC) in Quincy, FL was Norfolk loamy sand (fine-loamy, kaolinitic, thermic Typic Kandiudults). Soil type at the Plant Science Research and Education Unit (PSREU) in Citra, FL was Hague sand (loamy, siliceous, semiactive, hyperthermic Arenic Hapludalfs). Drip tapes (1.89 l·min.⁻¹/30.48 m at 68 kPa; 30.48 cm emitter spacing) were laid under and concurrently with the polyethylene mulch. Beds were fumigated

with methyl bromide/chloropicrin 67:33 at a rate of 448 kg·ha⁻¹ broadcast at EREC in 2005, and PSREU and NFREC in 2006. Fertilizer recommendations for EREC were 156N-0P-130.2K kg·ha⁻¹ in 2005 and 2006 (Franklin, 1998). Fertilizer recommendations for PSREU and NFREC were 168N-48P-140.2K kg·ha⁻¹ and 183.6N-24P-152.3K kg·ha⁻¹, respectively (Olson et al., 2006). All fertility recommendations were based on soil test results. Four-week-old watermelon plants were transplanted at EREC on 27 Apr., 2005 and 17 Apr., 2006. Four-week-old seedlings were transplanted at PSREU and NFREC on 21 Mar. and 3 Apr., 2006, respectively.

Plots were sprayed with fungicides and insecticides as recommended (Olson et al., 2006, Sanders et al., 2006). Pesticide applications were timed so that there was minimal effect on pollinators. One honeybee (*Apis mellifera* L.) hive was located near the center of each replication at Citra and Quincy, FL in 2006 while at Blackville, SC in both years a grouping of twenty honeybee hives was maintained 300 ft. north of test plots. At all locations in 2005 and 2006, watermelons were harvested once per week for three weeks. At the last harvest, all marketable melons were harvested. The center of each fruit was sampled for total soluble solids using a hand-held refractometer. Hollow heart measurements were taken by measuring the length and width of hollow cavities in watermelons that had been cut longitudinally from stem end to blossom end. Soluble solids and hollow heart data were taken from three melons per plot during the first harvest at all locations in 2006. Soluble solids data were taken from three watermelons per plot during the first harvest at Blackville, SC in 2005 but no hollow heart data were taken.

Yield and soluble solids data from 2005, and hollow-heart data from 2006 were analyzed using the GLM procedures and means separation was accomplished using Duncan's multiple range test in the SAS system (SAS Institute, Inc., Cary, NC). In 2006, there were multiple

locations and as location was not of primary interest in this study, location was set as a random effect. The MIXED procedure was used to analyze cultivar effect on fruit yield ($\text{kg}\cdot\text{ha}^{-1}$, fruit/ha, and kg/fruit) and soluble solids. This allows for greater inference of the results and how they may relate to many locations as compared with setting location as a fixed effect (Cushman et al., 2003; Schabenberger and Pierce, 2002). Pollenizer cultivar was set as a fixed effect and location, replication, and location by cultivar interaction were set as random effects.

Results

Treatments with pollenizer cultivars had significantly greater yield of triploid watermelons at all locations and in both years compared to the check (Tables 5-1 & 5-2). In addition there were significant differences among pollenizer cultivars in 2006 (Table 5-2). There were no significant differences in triploid watermelon yields among pollenizer cultivars in 2005 (Table 5-1). In 2006, plants pollinated by ‘Sidekick’ yielded $65,242 \text{ kg}\cdot\text{ha}^{-1}$ but were not significantly different than plants pollinated by ‘Patron’, ‘SP-1’, ‘Jenny’, or ‘Mickylee’ which yielded 63,677, 61,766, 61,751, and $59,599 \text{ kg}\cdot\text{ha}^{-1}$, respectively (Table 5-2). Plants pollinated by ‘Companion’ had the lowest yields at $49,976 \text{ kg}\cdot\text{ha}^{-1}$, which were significantly lower than those pollinated by ‘Jenny’, ‘SP-1’, ‘Patron’ or ‘Sidekick’ but not significantly different than plants pollinated by ‘Pinnacle’ or ‘Mickylee’ which yielded 53,333 and $59,599 \text{ kg}\cdot\text{ha}^{-1}$, respectively. Plots containing ‘Pinnacle’ had significantly lower yields than those containing ‘Sidekick’ but were not significantly different than plots containing ‘Mickylee’, ‘SP-1’, ‘Jenny’, or ‘Patron’ (Table 5-2). Pollenizers had a significant effect on number of triploid watermelons compared to the check. All plots with pollenizer cultivars had significantly greater numbers of melons per hectare than the control plots at all locations in both years (Tables 5-1 & 5-2). There were no significant differences in fruit production between the pollenizer cultivars in 2005 and 2006. In 2006, plants pollinated by ‘Patron’ produced 9,616 fruit/ha which was not significantly greater

than ‘Companion’ which produced 7,565 fruit/ha. Pollenizer cultivars had a significant effect on average triploid watermelon fruit weight in 2006, but not in 2005 (Tables 5-1 & 5-2). Pollenizer cultivars did not have a significant effect on soluble solids in both years (Table 5-3). In 2006, pollenizer cultivars did not have a significant effect on hollowheart at the Citra, FL and Blackville, SC locations (Table 5-4). Pollenizer cultivars did have a significant effect on hollow heart at Quincy, FL with all plots with pollenizers having significantly less hollow heart in the triploid watermelons when compared with the control plots (Table 5-4). There were no significant differences in hollow heart incidence between pollenizer cultivars.

Discussion

This research shows that some pollenizer cultivars tested can be expected to perform better than other cultivars, and do so at diverse locations. Similar results were reported by Fiacchino and Walters (2003) in which triploid watermelon yields were significantly different due to pollenizer cultivar used.

The only cultivar that showed questionable performance was ‘Companion’. Due to its growth and flowering habit it may not produce enough staminate flowers and pollen at the end of fruit setting in the triploid crop. ‘Companion’ is a short internode plant that becomes overgrown by triploid plants near the end of the season which may lead to staminate flowers that are not readily detectable by pollinators. Differences in staminate flower production by pollenizer cultivars have been reported however, it does not appear that flower production is the determining factor of a pollenizer’s performance (Dittmar et al., 2005; Freeman and Olson, 2007). In both of these studies, ‘SP-1’ produced greater numbers of staminate flowers when compared to ‘Jenny’ or ‘Mickylee’, however, data presented here indicates no difference in triploid watermelon yields between these pollenizer cultivars. Pollenizers must be able to continue growing and producing flowers throughout the production cycle.

There were significant differences in severity of hollow heart at Quincy between plots containing pollinizer cultivars and the control but not between the pollinizers. Unfortunately, this does not help to elucidate the cause of hollow heart as it may have been caused by reduced pollination in control plots or excessive growth of the few existing watermelons. The incidence of hollow heart at Blackville, SC and Citra, FL was low overall and this may be why there was no effect by the pollinizers. The experimental design was successful in reducing pollen flow out of experimental plots as indicated by minimal fruit set in control plots. This experimental design spaced the triploid watermelon from a pollinizer cultivar by 7.3 m. NeSmith and Duval (2001) illustrated that when distance of a triploid from a pollinizer was six meters or greater, triploid fruit numbers diminished substantially. Triploid pistillate flowers ('Tri-X Palomar') in plot buffers served to filter viable diploid pollen before pollinators entered another plot.

Of the cultivars tested, it appears that the pollinizers 'Jenny', 'Mickylee', 'Patron', 'Pinnacle', 'Sidekick', and 'SP-1' would be good choices. Some of the tested pollinizers ('Mickylee', 'Jenny', 'Pinnacle') can be harvested and sold if the grower has a market for seeded watermelons. If growers have a strong market for seeded melons then there may be no reason to plant pollinizers in-row. The pollinizers' costs vary greatly, so this must also be taken into consideration. Of the pollinizer cultivars that were shown to perform adequately ('Jenny', 'Mickylee', 'Patron', 'Pinnacle', 'Sidekick', 'SP-1'), selection should be based on seed/plant cost and distinctness between pollinizer and market melon.

Replication 1				Replication 2		
B U F F E R	Plot 1	B U F F E R	7.6 m Drive Row	B U F F E R	Plot 1	B U F F E R
	Plot 2				Plot 2	
	Plot 3				Plot 3	
	Plot 4				Plot 4	
	Plot 5				Plot 5	
	Plot 6				Plot 6	
	Plot 7				Plot 7	
	Plot 8				Plot 8	

Figure 5-1. Field diagram for pollenizer experiments at Blackville, SC, Citra, FL, and Quincy, FL in 2005 and 2006. Columns represent individual rows. The same design was used for replications three and four.

B U F F E R	'Tri-X Palomar'	B U F F E R
	'Tri-X Palomar'	
	'Tri-X Palomar'	
	'Tri-X Palomar'	
	Data melon	
	Pollenizer	
	Data melon	
	Data melon	
	Data melon	
	Pollenizer	
	Data melon	
	Data melon	
	Data melon	
	Pollenizer	
	Data melon	
	'Tri-X Palomar'	
'Tri-X Palomar'		
'Tri-X Palomar'		
'Tri-X Palomar'		

Figure 5-2. Individual three-row plot design for pollenizer experiments at Blackville, SC, Citra, FL, and Quincy, FL in 2005 and 2006. Plot shown is using a pollenizer recommended to be planted at a 1:3 pollenizer to seedless ratio.

Table 5-1. Pollenizer cultivar effect on ‘Tri-X 313’ yield at Blackville, SC during 2005

Pollenizer cultivar	Yield (kg·ha ⁻¹) ^z	Fruit (no./ha) ^z	Avg wt (kg/fruit)
Jenny	67,565 a ^y	9,386 a ^y	7.6 ^{NS}
SP-1	63,944 a	9,666 a	6.9
Mickylee	61,759 a	8,966 a	7.0
Tri-X Palomar ^x	10,494 b	1,400 b	8.7

^z Yield estimates are based on plant populations of 4483 plants per hectare. ^y Means with the same letter are not significantly different at ($P \leq 0.05$) by Duncan’s multiple range test. ^x Triploid cultivar serving as check against pollen contamination from neighboring plots.

Table 5-2. Pollenizer cultivar effect on ‘Supercrisp’ watermelon yield and average fruit weight at Blackville, SC, Citra, FL, and Quincy, FL during 2006.

Pollenizer cultivar	Yield (kg·ha ⁻¹) ^z	Fruit (no./ha) ^z	Avg wt (kg/fruit)
Sidekick	65,242 a	9,386 a	7.3 a b
Patron	63,677 a b	9,616 a	7.0 b
SP-1	61,766 a b	9,106 a	7.0 b
Jenny	61,751 a b	9,195 a	6.9 b
Mickylee	59,599 a b c	9,106 a	6.7 b
Pinnacle	53,333 b c	7,845 a	7.2 a b
Companion	49,976 c	7,565 a	6.9 b
Tri-X Palomar ^y	8,545 d	1,074 b	7.8 a
LSD ^x	10,494	2,116	0.7

^z Yield estimates are based on plant populations of 4483 plants per hectare. ^y Triploid cultivar serving as check against pollen contamination from neighboring plots. ^x $P = 0.05$

Table 5-3. Pollenizer cultivar effect on soluble solids concentration of seedless watermelons at Blackville, SC during 2005 and Citra, FL, Quincy, FL, and Blackville, SC during 2006.

Pollenizer cultivar	Soluble solids concentration (%)	
	Blackville, SC 2005	Combined locations 2006
Sidekick		12.2 ^{NS}
Patron		12.1
SP-1	11.0 ^{NS z}	12.3
Jenny	11.6	12.3
Mickylee	11.2	12.3
Pinnacle		12.1
Companion		12.4
Tri-X Palomar ^y	11.6	12.2
LSD ^x		0.7

^z Means with the same letter are not significantly different at ($P \leq 0.05$) by Duncan's multiple range test. ^y Triploid cultivar serving as check against pollen contamination from neighboring plots. ^x $P = 0.05$

Table 5-4. Pollenizer cultivar effect on hollowheart disorder in ‘Supercrisp’ watermelon at Quincy, FL, and Blackville, SC combined with Citra, FL during 2006. Means are to be compared within the same column.

Pollenizer cultivar	Hollowheart area (cm ²)	
	Quincy, FL	Blackville, SC & Citra, FL
Tri-X Palomar ^z	187.0 a ^y	4.7 ^{NS}
Patron	73.3 b	12.2
Jenny	70.2 b	5.8
Sidekick	68.2 b	9.7
Companion	58.1 b	3.3
Mickylee	54.7 b	2.6
SP-1	53.5 b	15.7
Pinnacle	37.5 b	10.7

^z Triploid cultivar serving as check against pollen contamination from neighboring plots ^y Means with the same letter are not significantly different at ($P \leq 0.05$) by Duncan’s multiple range test.

Table 5-5. Seed sources for various pollenizer cultivars used during 2005 and 2006.

Pollenizer cultivar	Company
Patron	Zeraim Gedera Seed Co., Ltd. (Palm Desert, CA)
Jenny	Nunhems USA, Inc. (Acampo, CA)
Sidekick	Harris Moran Seed Co. (Modesto, CA)
Companion	Seminis Vegetable Seed, Inc. (Oxnard, CA)
MickyLee	Many sources
SP-1	Syngenta Seeds, Inc.(Boise, ID)
Pinnacle	Southwestern Vegetable Seed, LLC. (Casa Grande, AZ.)

CHAPTER 6
COMPETITIVE EFFECT OF IN-ROW DIPLOID WATERMELON POLLENIZERS ON
TRIPLOID WATERMELON YIELD

Introduction

Due to the high cost of in-row diploid pollenizer seed, there is interest in using standard watermelon cultivars in-row. One cultivar that is being evaluated for in-row use is ‘Mickylee’ which produces an easily distinguishable fruit, a crucial characteristic for diploid pollenizers. Most in-row pollenizer cultivars have reduced foliage in order to compete less with triploid plants. ‘Mickylee’ is an attractive option as a pollenizer because of the low seed costs but it is unclear whether the more vigorous growth habit will negatively impact seedless watermelon yields. The objective of this study was to determine if pollenizer growth habit and pollenizer to triploid spacing would have an effect on triploid watermelon yield.

Materials and Methods

Experiments were conducted at the North Florida Research and Education Center (NFREC) in Quincy, FL. and the Plant Science Research and Education Unit (PSREU) in Citra, FL. in the Spring of 2006 and also at NFREC in the Fall of 2006. Soil type at NFREC was Norfolk loamy sand (fine-loamy, kaolinitic, thermic Typic Kandiudults) and at PSREU was Hague sand (loamy, siliceous, semiactive, hyperthermic Arenic Hapludalfs). At NFREC (spring and fall) all fertilizer was incorporated pre-plant at a rate of 183.6N-24P-152.3K kg·ha⁻¹. At PSREU two thirds of the fertilizer was applied pre-plant and the remainder of the recommendation was fulfilled through weekly fertigation. Total fertilizer applied at PSREU was 168N-48P-140.2K kg·ha⁻¹. Fertilization was based on soil test results and University of Florida recommendations (Olson et al., 2006). At both locations, irrigation was provided through drip tape (1.89 l·min.⁻¹/30.48 m at 68 kPa; 30.48 cm emitter spacing) which was laid concurrently with black polyethylene mulch in the spring and with white on black polyethylene mulch in the

fall. Soil was fumigated at plastic laying with methyl bromide/chloropicrin 67:33 at a rate of 448 kg·ha⁻¹. Beds were spaced 2.43 m center-to-center.

Experimental design was a factorial with four replications and two factors, pollenizer cultivar and in-row spacing. In-row spacing's between one pollenizer plant and one triploid plant were 0.2, 0.4, 0.6, 0.8, and 1.0 m. Plots consisted of six pairs (one pollenizer, one triploid) of plants with equal pollenizer to triploid spacings. Each pair of plants was spaced 1.0 m in-row from the next pair. On 21 Mar., 3 Apr., and 1 Aug., 4-week old watermelon seedlings were transplanted. 'Sugarheart' triploid was used along with 'Mickylee' (standard vining habit) and 'SP-1' (reduced foliage, thin-vines) as pollenizers. Harvests of spring trials were made on 8 and 19 June at PSREU and on 21 and 28 June, and 7 July at NFREC. Fall harvests were on 11 and 25 Oct. Insecticides and fungicides were applied as needed to maintain plant health (Olson et al., 2006). Groupings of honey bee (*Apis mellifera* L.) hives were placed in close proximity to all experiments. The GLM procedures of SAS (SAS Institute, Inc., Cary, NC) were used to analyze the effects of pollenizer cultivar on 'Sugarheart' watermelon yield and interactions between pollenizer cultivar and spacing. The GLM procedures were also used to analyze the polynomial effects of pollenizer to triploid spacing.

Results

Pollenizer cultivar and plant spacing had significant effects on seedless watermelon yield at Citra and Quincy ($P \leq 0.05$) in the spring but not at Quincy in the fall ($P > 0.05$). Cultivar and plant spacing had significant effects on fruits per plant (fpp) at both locations in the spring, however, the significance at Citra for cultivar and plant spacing were $P = 0.0983$ and $P = 0.0633$, respectively. Plant spacing had significant linear effects on seedless watermelon yield and fpp at both locations in the spring (Table 6-1). No significant effects on average watermelon weight were observed in any of the experiments. Cultivar and plant spacing had no effect on

seedless watermelon yield or yield components at Quincy in the fall. No significant interaction effects were observed between cultivar and plant spacing. A significant interaction between location and pollenizer cultivar was detected in the spring therefore locations will be presented separately.

Seedless watermelon yields from plants paired with ‘Mickylee’ at Citra and Quincy were 24.6 and 33.5 kg/plant, respectively. Yields from plants paired with ‘SP-1’ at Citra and Quincy were 27.7 and 43.3 kg/plant, respectively, which were significantly greater than yields from plants paired with ‘Mickylee’. At Quincy, plants paired with ‘Mickylee’ produced 3.74 fpp which was significantly lower than 4.85 fpp produced by plants paired with ‘SP-1’. At Citra, plants paired with ‘Mickylee’ produced 3.68 fpp which was lower than 4.04 fpp produced by plants paired with ‘SP-1’; however this was only statistically significant at $P = 0.10$.

Seedless watermelon yields and fpp increased linearly with increased spacing at both locations in the spring. Yields from plots with 0.2 m in-row spacing’s were 22.1 and 32.0 kg/plant at Citra and Quincy, respectively. Yields from plots with 1.0 m in-row spacing were 30.8 kg/plant at Citra and 47.8 kg/plant at Quincy.

Discussion

These results appear to indicate that the vigorous growth habit of pollenizer ‘Mickylee’ negatively impacted yield of the triploid cultivar Sugarheart compared to the reduced foliage of pollenizer ‘SP-1’. Seedless watermelon yields (kg/plant) from plants paired with ‘Mickylee’ yielded 11.4 and 22.4% less than plants paired with ‘SP-1’ at Citra and Quincy, respectively. At current pollenizer ratios recommended by seed producers, pollenizer competition could affect two thirds of the triploid plants per hectare. These yield reductions can be expected at all in-row spacings as there was no interaction between cultivar and spacing. Extrapolations of yield to a per hectare basis were not performed because it is unclear how many pollenizer plants would be

necessary at these plant populations which ranged from 10,089 plants·ha⁻¹ at the closest spacing's to 2,017 plants·ha⁻¹ at the widest. There was also no yield data from plants that were considered to be unaffected by pollenizer competition. During the fall experiment at Quincy, there was a severe outbreak of gummy stem blight (*Didymella bryoniae* (Auersw.) Rehm) which was not controlled by the weekly fungicide applications. This no doubt reduced seedless watermelon yields which affected the statistical outcome.

The trends in fruit yield and fruit number observed in this study are similar to other reports in watermelon where increased plant population, and thus increased competition, changes yield through fruit number and not average fruit mass (Brinen et al., 1979; Duthie et al., 1999a, 1999b; NeSmith, 1993). Competition studies investigating weed species effect on watermelon yield have also shown increasing competition lowers watermelon yield as a function of fruit number and not fruit size (Buker et al., 2003; Monks and Schultheis, 1998).

The results from this project are not in agreement with Freeman (2007) who evaluated the performance of in-row pollenizers and found no difference in seedless watermelon yield between plots containing 'Mickylee' or 'SP-1'. The experimental design used was intended to compare pollenizer cultivars as a function of pollen provided by each cultivar and not its competitive effect. This study collected yield data by plot which does not provide insight into pollenizer to triploid competition as some plants were located directly beside pollenizers and some were not. It is unclear why a significant reduction in yield caused by 'Mickylee' in this study is not in agreement with the results reported by Freeman (2007).

'Mickylee' is an attractive option as a pollinizer because of the low seed costs. However, the results of this study indicate that seedless watermelon yields and fruits per plant will significantly decrease when 'Mickylee' is used as a pollinizer as compared with 'SP-1' regardless of in-row spacing.

Table 6-1. Influence of pollenizer cultivar and spacing on triploid watermelon yield during 2006.

Treatment	Location								
	Citra, FL – Spring 2006			Quincy, FL – Spring 2006			Quincy, FL – Fall 2006		
	Fruit (no./plant)	Yield (kg/plant)	Avg. wt. (kg/fruit)	Fruit (no./plant)	Yield (kg/plant)	Avg. wt. (kg/fruit)	Fruit (no./plant)	Yield (kg/plant)	Avg. wt. (kg/fruit)
Cultivar									
SP-1	4.04	27.7	6.8	4.85	43.4	8.8	3.04	19.0	6.2
Mickylee	3.68	24.5	6.7	3.74	33.5	8.8	2.85	17.4	6.0
Significance	*	**	NS	***	***	NS	NS	NS	NS
Spacing (m)									
0.2	3.33	22.1	6.5	3.57	32.0	8.9	2.98	18.2	6.0
0.4	3.68	24.9	6.7	3.46	29.7	8.4	2.85	17.5	6.0
0.6	3.92	26.3	6.7	4.52	40.4	8.8	2.86	18.4	6.4
0.8	4.02	26.6	6.7	4.85	43.9	9.0	3.18	19.3	6.0
1.0	4.35	30.8	7.1	5.28	47.8	9.0	2.86	17.6	6.1
Significance	L***	L***	NS	L***	L***	NS	NS	NS	NS

NS, *, **, *** Non-significant or significant at $P \leq 0.10$, 0.05 or 0.01, respectively.

CHAPTER 7
VARIABILITY IN WATERMELON FLOWER ATTRACTIVENESS TO INSECT
POLLINATORS

Introduction

In a field that is producing seedless watermelons, there must be both diploid and triploid plants (Kihara, 1951; Maynard and Elmstrom, 1992). For fruit set to occur in triploid plants, pollen must be moved from the staminate diploid flower to the pistillate triploid flower. Triploid and diploid plants produce staminate flowers which bear pollen, however, triploid pollen is non-viable. It is unlikely that insect pollinators can visually distinguish between the two and foraging of staminate triploid flowers dilutes the flow of viable pollen within a field. If staminate flowers produced by the diploid pollenizer are more attractive than triploid staminate flowers, a greater proportion of viable pollen could be moved by pollinators which may lead to greater reproductive success in triploid fruits. The floral attractiveness of a pollenizer could also impact its performance. The objective of this study was to determine the floral attractiveness of staminate flowers of three pollenizer cultivars and one triploid cultivar.

Materials and Methods

Field experiments were conducted at the North Florida Research and Education Center (NFREC) in Quincy, FL. during the Spring and Fall of 2006. Soil type at NFREC is Norfolk loamy sand (fine-loamy, kaolinitic, thermic, Typic Kandudults). The experimental design was a randomized complete block with eight replications. Experimental plots were 4.57 m long with in-row spacing of 0.46 m and between row spacing of 4.9 m. The experiment consisted of two rows 73.2 m long. On 3 Apr. and 1 Aug. 2006, 4-week old watermelon seedlings were transplanted into raised beds covered with black polyethylene mulch in the spring and white on black polyethylene mulch in the fall. Three watermelon plants were transplanted into each plot. Number of pollinator visitations was recorded for four watermelon cultivars, 'Companion',

‘Intruder’, ‘Mickylee’ and ‘SP-1’. Three cultivars are diploid pollenizers and one (‘Intruder’) is a triploid. Fertilization, irrigation, and pesticide application practices recommended by the University of Florida Institute of Food and Agricultural Sciences were followed (Olson et al., 2006). A grouping of two honeybee (*Apis mellifera* L.) hives was placed near the center of the experiment.

Sampling was performed on five occasions in the spring and three in the fall. Sampling started when plants began to produce staminate and pistillate flowers. On sampling dates, sampling was initiated at anthesis. Five staminate flowers were chosen in each plot and visitations from honeybees and bumblebees (*Bombus* spp. Cresson) were counted for two minutes. Three to four sampling repetitions were performed on each sampling date and are referred to as sampling time. Previous research has illustrated an interaction between cultivar attractiveness and sampling time, for this reason sampling times were kept succinct and sampling time was considered a main effect. Two individuals recorded visitations in order to keep repetition time under 45 minutes. Analysis of variance was performed using the GLM procedures of SAS (SAS Institute, Inc., Cary, NC) to determine significance of main and interaction effects and Duncan’s multiple range test was used for means separation.

Results

Watermelon cultivar and sampling time had a significant effect ($P \leq 0.05$) on floral visitation by insect pollinators on six of eight sampling dates. Cultivar or sampling time did not influence pollinator visitation on 11 May or 29 Sept. Significant interactions ($P \leq 0.05$) between time and cultivar were detected on 23 May and 22 Sept. Visitation of a diploid cultivar was significantly greater than the triploid cultivar on six of eight sampling dates.

On 16 May, ‘SP-1’ had 2.4 visits per plot (vpp) which was significantly greater than ‘Mickylee’, ‘Companion’, or ‘Intruder’ which had 1.1, 1.0, and 0.8 vpp, respectively (Fig. 7-1).

Pollinator visitation of ‘Mickylee’, ‘Companion’, or ‘Intruder’ was not significantly different. Pollinator visitations to ‘Mickylee’ and ‘SP-1’ were 2.9 vpp which was significantly greater than ‘Companion’ or ‘Intruder’ which had 1.5 and 1.0 vpp, respectively, on 19 May. An interaction between sampling time and cultivar occurred on 23 May. During the first sampling time, ‘SP-1’ had 4.2 vpp which was not significantly greater than ‘Companion’ or ‘Mickylee’ which had 3.0 and 2.75 vpp, respectively (Fig. 7-2). However, all three cultivars had significantly greater visitation than ‘Intruder’ at 0.3 vpp. During the second sampling time, ‘Mickylee’ had 5.5 vpp which was not significantly different than ‘SP-1’ at 2.8 vpp but was significantly greater than ‘Intruder’ and ‘Companion’ at 1.5 and 1.3 vpp, respectively. Visitation of ‘SP-1’, ‘Intruder’, and ‘Companion’ were not significantly different. There were no significant differences between cultivars during the third and fourth sampling times. On 25 May, ‘Mickylee’ had 2.8 vpp which was significantly greater than ‘SP-1’, ‘Companion’, or ‘Intruder’ at 1.8, 1.4 and 0.4 vpp, respectively (Fig. 7-1). ‘SP-1’ and ‘Companion’ were not significantly different but both had greater visitation than ‘Intruder’.

On 15 Sept, ‘SP-1’ had 7.7 vpp which was greater than ‘Mickylee’, ‘Intruder’ or ‘Companion’ at 6.1, 5.8, and 3.7 vpp, respectively (Fig. 7-3). ‘Mickylee’ and ‘Intruder’ were not significantly different but both had greater visitation than ‘Companion’. An interaction between sampling time and cultivar occurred on 22 Sept. During the first sampling time, ‘SP-1’ had 2.7 vpp which was not greater than ‘Mickylee’ at 1.8 vpp but was greater than ‘Companion’ and ‘Intruder’ at 0.7 and 0.5 vpp, respectively (Fig. 7-4). ‘Mickylee’, ‘Companion’, and ‘Intruder’ were not significantly different. During the second sampling time, ‘SP-1’ had 6.8 vpp which was similar to ‘Mickylee’ at 5.2 vpp and both had greater visitation than ‘Companion’ or ‘Intruder’ which had 2.3 and 2.1 vpp, respectively. ‘Companion’ and ‘Intruder’ were not significantly

different. There was no significant difference between cultivars during the third sampling time. On 29 Sept. main effects did not influence pollinator visitation (Fig. 7-3). No data were taken from ‘Companion’ on 29 Sept. because it had ceased producing staminate flowers. Complete floral visitation data are shown in figures 7-1 – 7-4.

Discussion

Either ‘Mickylee’, ‘SP-1’, or both received greater floral visitation than ‘Companion’ or ‘Intruder’. Previous research as shown that staminate flower production by ‘SP-1’ is greater than that of ‘Mickylee’ or ‘Companion’ which were similar (Freeman and Olson, 2007). The number of flowers that were used in sampling was held constant in order to determine the relative attractiveness of each cultivars staminate flower. This research illustrates differential attractiveness between cultivars and ploidy levels. Visitation at the whole plant level for ‘SP-1’ could be higher than the other cultivars due to greater numbers of staminate flowers but a staminate flower produced by ‘SP-1’ is not necessarily more attractive than a staminate flower produced by ‘Mickylee’. Other researchers have reported differences in pollinator visitation between watermelon cultivars and between *Citrullus lanatus* and *Citrullus colocynthis* which was attributed to nectar sugar concentration (Wolf et al., 1999). It has been shown that floral volatiles emitted from pollen are the most important close-range cue during foraging by honeybees, however; visual stimuli are important long-range cues (Pernal and Currie, 2002). ‘Companion’ has a nearly entire leaf with reduced lobes and produces staminate flowers with short peduncles. These factors tend to obstruct the view of ‘Companion’s staminate flowers which may be why ‘Companion’ was generally visited less than ‘SP-1’ and ‘Mickylee’.

A diploid watermelon cultivar was preferred over the triploid on sampling dates where cultivar affected pollinator visitation. Triploid watermelon plants produce mostly non-viable, aborted pollen which may be covered with less pollenkitt than viable pollen. Pollenkitt produces

volatiles that are important in foraging decisions of pollinators. These volatiles are an indicator of pollen reward that is available in a flower and reduced volatile emissions may indicate reduced reward (Dobson et al., 1996). A reduction in pollenkit produced in triploid staminate flowers could represent reduced reward and result in flowers that are less attractive to pollinators. Previous research has shown that *Bombus* spp. preferentially foraged potato flowers that produced viable pollen over ones producing non-viable pollen which may be due to differences in volatile emissions (Batra, 1993).

When the performance of multiple diploid watermelon pollenizer cultivars were compared, triploid watermelon yields from plots pollenized by ‘Companion’ were significantly less than plots pollenized by ‘SP-1’ or ‘Mickylee’. The lower preference of ‘Companion’ may lead to less viable pollen transported by pollinators and thus less fruit on triploid plants. A diploid pollenizer cultivar with staminate flowers which are more attractive than triploid staminate flowers could increase the movement of viable pollen within a field and possibly increase seedless watermelon yield.

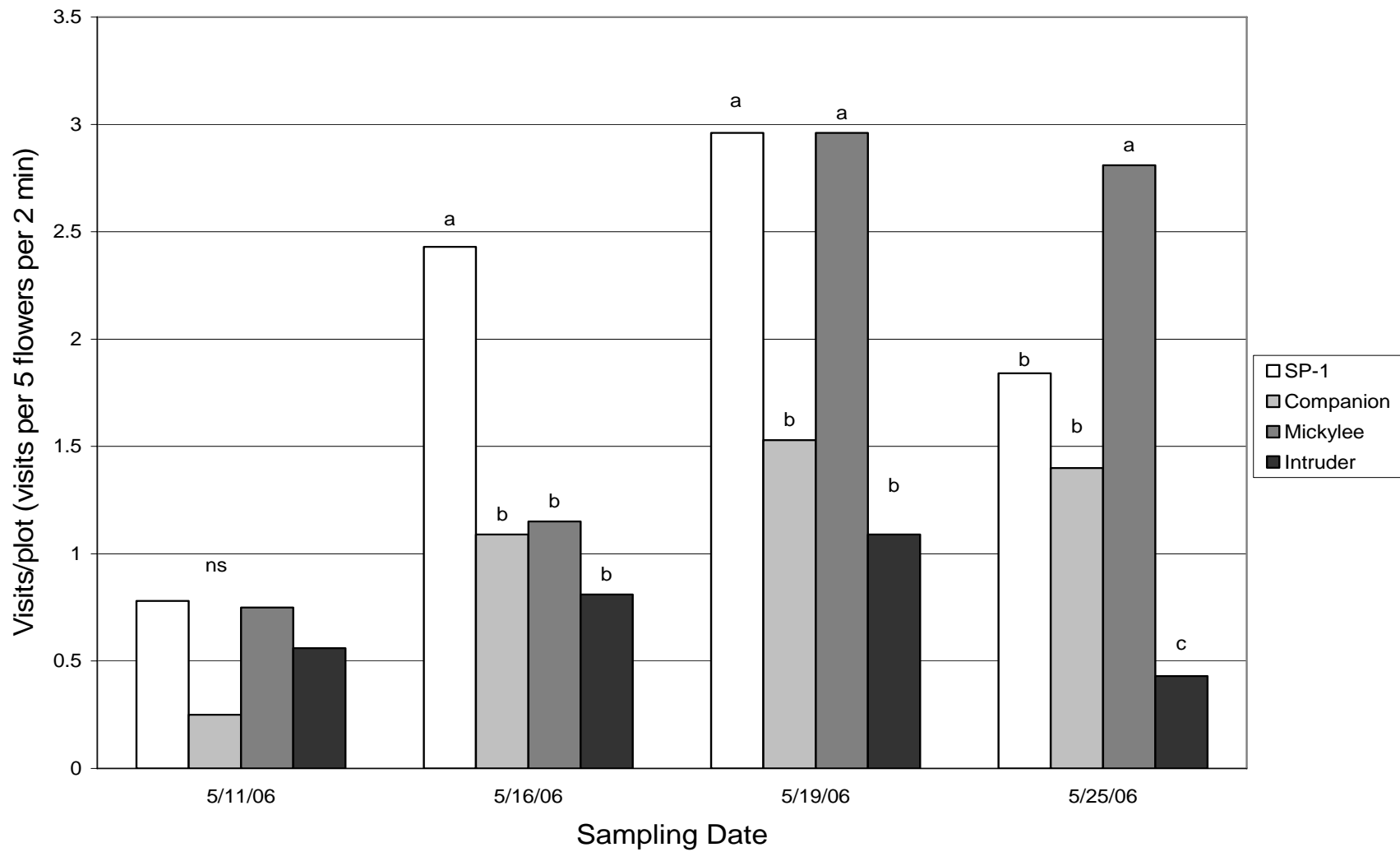


Figure 7-1. Influence of cultivar on pollinator visitation to staminate watermelon flowers at Quincy, FL., Spring 2006. Visitation means are to be compared within sampling date. Means followed by the same letter are not significantly different at $P \leq 0.05$.

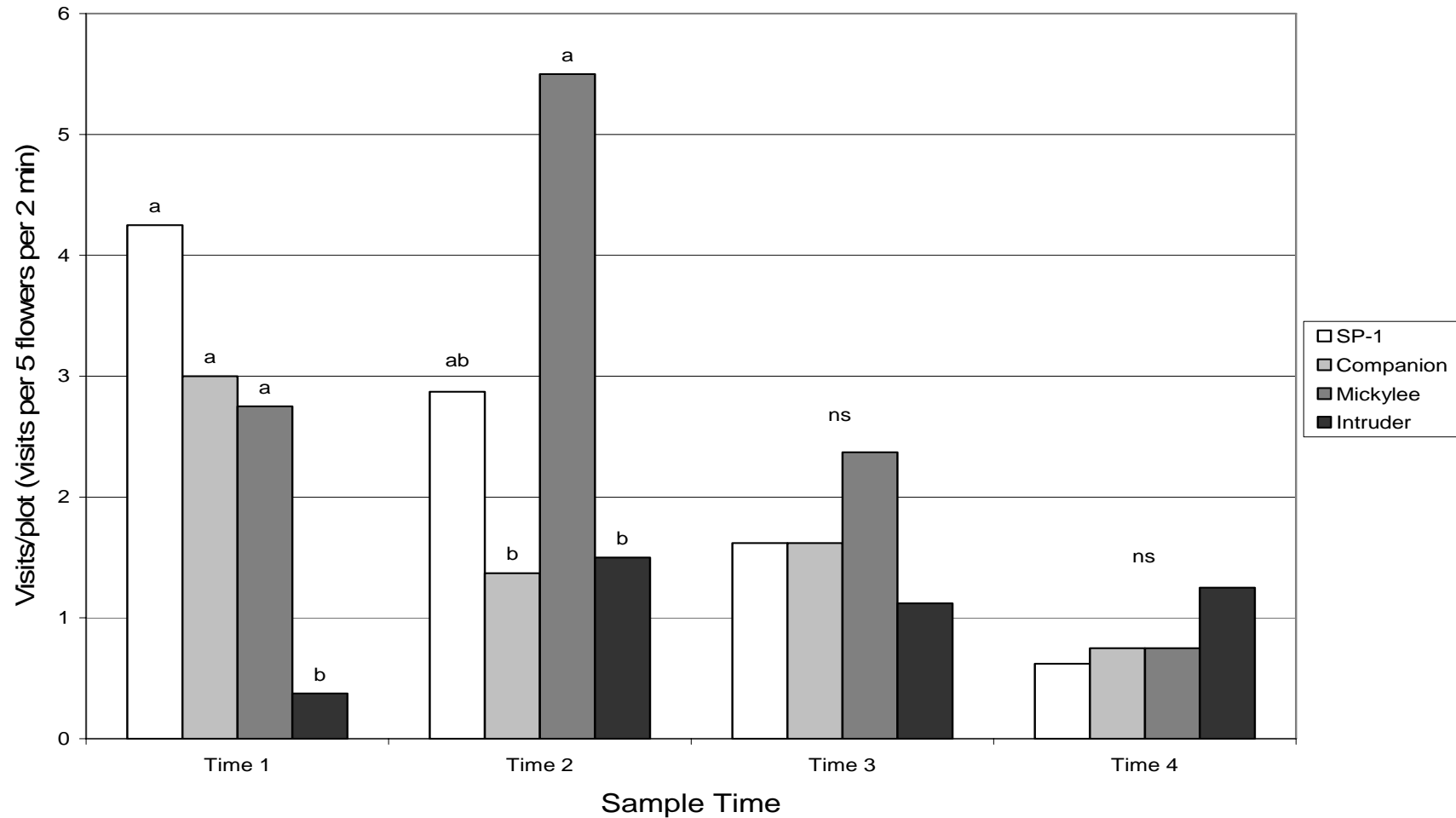


Figure 7-2. Interaction of cultivar and time on pollinator visitation to staminate watermelon flowers at Quincy, FL., on 23 May, 2006. Visitation means are to be compared within sampling time. Means followed by the same letter are not significantly different at $P \leq 0.05$.

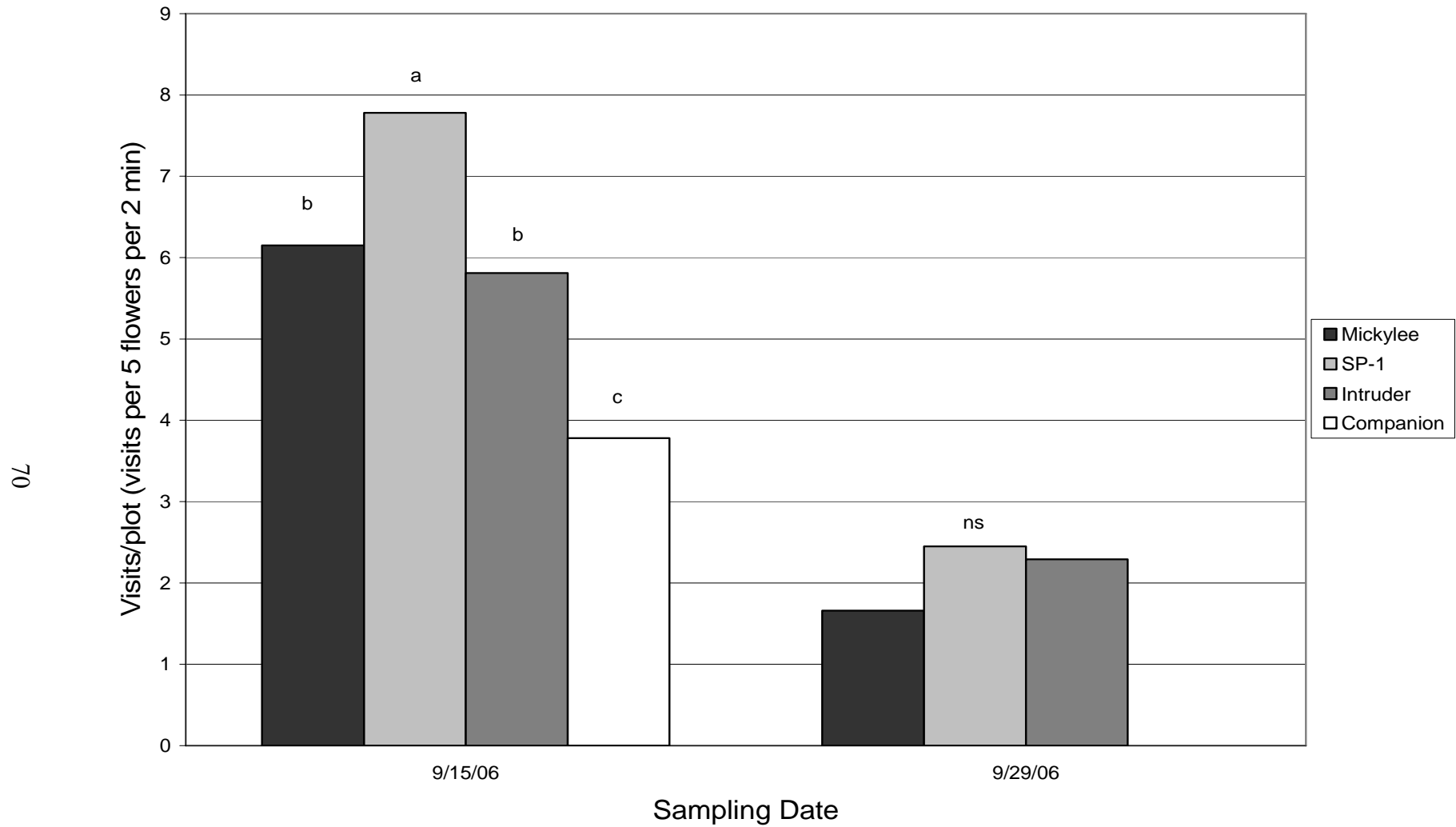


Figure 7-3. Influence of cultivar on pollinator visitation to staminate watermelon flowers at Quincy, FL., Fall 2006. Visitation means are to be compared within sampling date. Means followed by the same letter are not significantly different at $P \leq 0.05$.

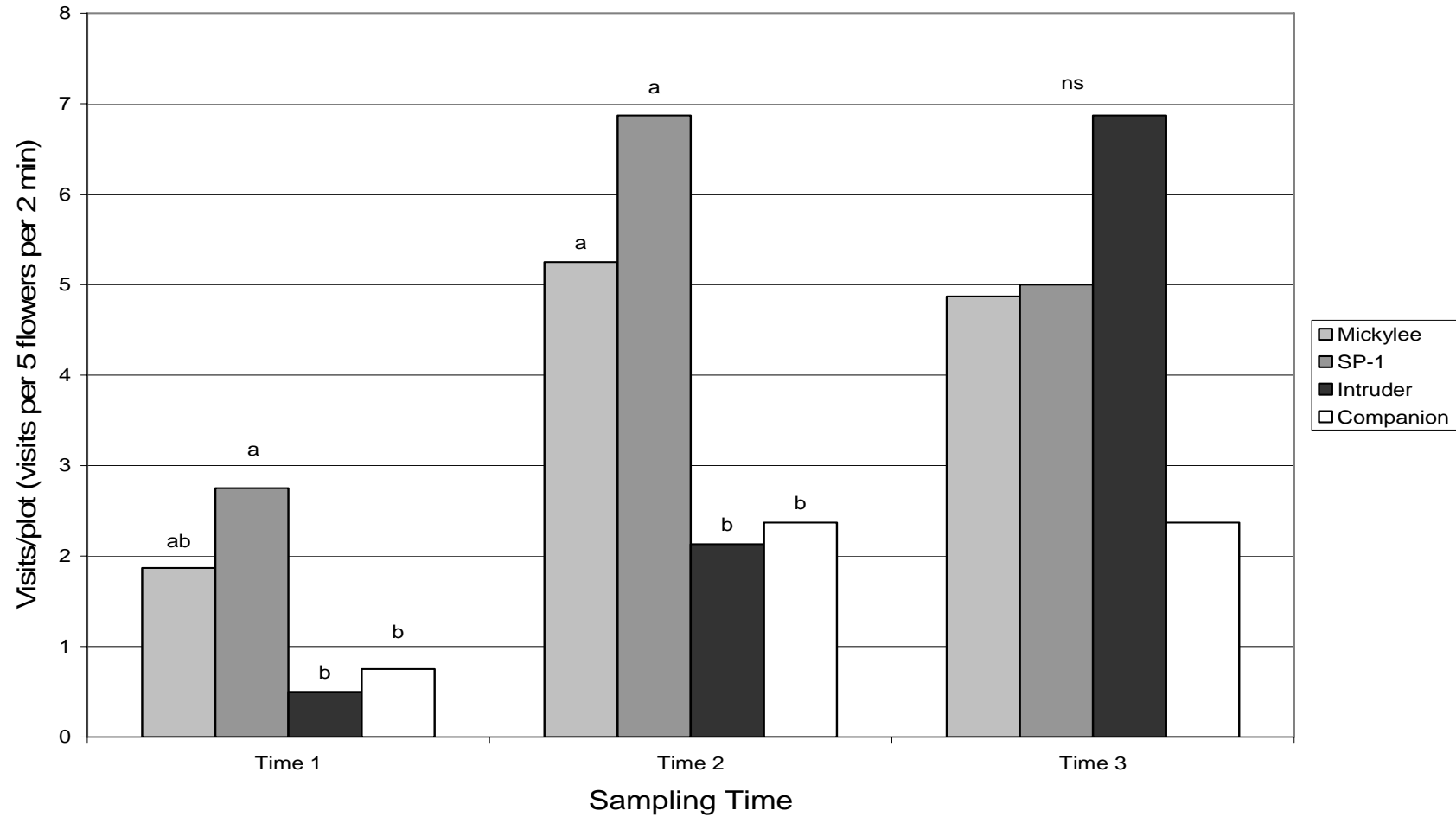


Figure 7-4. Interaction of cultivar and time on pollinator visitation to staminate watermelon flowers at Quincy, FL., on 23 Sept., 2006. Visitation means are to be compared within sampling time. Means followed by the same letter are not significantly different at $P \leq 0.05$.

CHAPTER 8 VARIABILITY IN POLLEN PRODUCTION BY DIPLOID WATERMELON POLLENIZERS

Introduction

Pollen produced by diploid watermelon pollenizers is necessary for fruit set and flesh fill in associated triploid watermelon crops (Kihara, 1951; Maynard and Elmstrom, 1992). Viable pollen produced by pollenizers is diluted with non-viable triploid pollen which increases pollinator visitation rates required by pistillate triploid flowers. It is important that there be adequate viable diploid pollen moved throughout a field and pollenizers that produce the greatest amounts of pollen may perform better with respect to triploid watermelon yield. Significant differences in pollen production have been reported between cultivars of diploid watermelon as well as in other plant species. The objective of this project was to determine the amount of pollen produced by four pollenizer cultivars to investigate possible effects on pollenizer performance.

Materials and Methods

Experiments were conducted at the North Florida Research and Education Center (NFREC) in Quincy, FL during the fall of 2006. Soil type at NFREC is Norfolk loamy sand (fine-loamy, kaolinitic, thermic, Typic Kandiudults). The experimental design was a randomized complete block with four replications. Experimental plots were 4.57 m long with in-row spacing of 0.91 m and between row spacing of 2.43 m. On 1 Aug. 2006, 4-week old watermelon seedlings were transplanted into raised beds covered with white on black polyethylene mulch. Beds were fumigated with methyl bromide/chloropicrin 67:33 at a rate of 448 kg·ha⁻¹ broadcast. Three seedlings were planted in each plot. Fertilization, irrigation, and pesticide application practices recommended by the University of Florida Institute of Food and Agricultural Sciences were followed (Olson et al., 2006).

Sampling was initiated on 31 Aug. with additional samples taken on 8 Sept. and 14 Sept. On sampling dates, watermelon flowers were removed before anthesis, placed into plastic containers and covered to exclude pollinators. Two flowers per cultivar were analyzed from each replication. Flowers were allowed to open and anther dehiscence was verified before flowers were processed. Flower petals were excised with a razor blade and peduncles were removed before flowers were placed into separate vials containing 5 mL of a solution of 70:30 ethanol/ethyl acetate mixture. This solution served to preserve the pollen and also to remove pollenkitt from the exterior of the pollen grains. The removal of pollenkitt was necessary to obtain uniform pollen distribution in the liquid media. Sample vials were sonicated in a water-bath sonicator to remove all pollen from anthers. After the anthers and any other remaining floral structures were removed, vials were sonicated again to ensure the removal of all pollenkitt. Three 50 μ L sub-samples were taken from each vial and pollen grains were counted on a grided microscope slide. Sub-sample values were then extrapolated to obtain total number of pollen grains per flower.

Pollen sub-sample counts were averaged for each flower. Analysis of variance was performed using the GLM procedures of SAS (SAS Institute Inc., Cary, N.C.) and means separation was performed using Duncan's multiple range test.

Results

Pollenizer cultivar and sampling date had significant effects ($P \leq 0.05$) on pollen production per flower. There was no significant ($P > 0.05$) interaction between cultivar and sampling date. On 31 Aug., 'Mickylee' produced 49,975 pollen grains per flower (gpf) which was not significantly greater than 'Companion' at 44,800 gpf but both were greater than 'SP-1' and 'Jamboree' at 37,813 and 36,700 gpf, respectively (Table 1). Pollen production by 'Companion' was not significantly greater than 'SP-1' or 'Jamboree'. On 8 Sept., 'Companion'

produced 62,275 gpf which was not significantly greater than ‘Mickylee’ at 60,250 gpf or ‘SP-1’ at 53.138 gpf but was greater than ‘Jamboree’ at 44,975 gpf. Pollen production by ‘SP-1’ and ‘Jamboree’ were not significantly different on 8 Sept. On 14 Sept., ‘Mickylee’ produced 58,900 gpf which was significantly greater than ‘Companion’, ‘Jamboree’, and ‘SP-1’ at 50,825, 48,475, and 42,250, respectively. Seasonal averages for pollen production are shown in Table 1.

Discussion

The results of this study illustrate that the amount of pollen produced by diploid watermelon plants differs by cultivar and also changes throughout the season. Pooled data for pollen production illustrates that ‘Mickylee’ and ‘Companion’ produce significantly greater amounts of pollen per flower than ‘SP-1’ or ‘Jamboree’. However, the pollen output per flower may not accurately represent the pollen output per plant. Previous research on flower production by pollenizers illustrated that ‘SP-1’ produced significantly greater numbers of staminate flowers when compared to ‘Mickylee’ or ‘Companion’ (Freeman and Olson, 2007). The increased production of staminate flowers by ‘SP-1’ may result in greater pollen output per plant when compared with the other cultivars.

The trend observed was that pollen production starts low, increases and then decreases as the season progresses. These results are in contrast to Stanghellini and Schultheis (2005) who reported that sampling day did not affect pollen production per flower. However, the duration of sampling used by Stanghellini and Schultheis (2005) was not stated and flowers sampled over a shorter time frame may not exhibit the variation observed here. As watermelon plants begin to branch and produce flowers on secondary terminals, it has been observed that flower size decreases. The decrease in flower size apparently correlates with the decrease in pollen production per flower. Although pollen production per flower may decrease as the season

progresses, it is likely that pollen production per plant will increase to a point as the number of staminate flowers produced increases.

In theory, pollenizers that produce the greatest amount of pollen could outperform other pollenizers with lower pollen output. Greater pollen output by a pollenizer may also facilitate the reduction in pollenizer plant numbers used per unit area. However, other experimental results suggest that pollen production alone is not a reliable indicator of pollenizer performance as the use of 'SP-1' resulted in significantly greater triploid watermelon yield when compared to the use of 'Companion'. There are many factors that influence the performance of diploid watermelon pollenizers and pollen production is likely a strong contributor. However, it is not a reliable factor on which to judge a pollenizers potential.

Table 8-1. Pollen production by four diploid watermelon pollenizer cultivars at Quincy, FL during the Fall of 2006.

Cultivar	Pollen grains per flower			
	31 Aug.	8 Sept.	14 Sept.	Combined
Mickylee	49,775 a ^z	60,250 a ^z	58,900 a ^z	56,308 a ^z
Companion	44,800 a b	62,275 a	50,825 b	52,633 a
SP-1	37,813 b	53,138 a b	42,550 c	44,500 b
Jamboree	36,700 b	44,975 b	48,475 b c	43,383 b

^z Means followed by the same letter are not significantly different at $P \leq 0.05$ by Duncan's multiple range test.

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BIOGRAPHICAL SKETCH

Joshua Herbert Freeman was born on October 24, 1980 to Linda and Herbert Freeman of Columbia, S.C. He first became interested in agriculture while working on cattle farm near his home. After graduating from Ridge View High School he attended Clemson University where earned a bachelor's degree in entomology. In the fall of 2002, he enrolled in the University of Florida and pursued a doctor of plant medicine degree. His career aspirations changed and in the fall of 2004, he began working on doctorate of philosophy in horticultural science at the University of Florida.