QUANTIFICATION OF BLACKMARGINED APHID (*MONELLIA CARYELLA* (FITCH)) HONEYDEW PRODUCTION IN PECAN (*CARYA ILLINOINENSIS* (KOCH)) IN TEXAS

A Thesis

by

JESSICA MARIE HONAKER

Submitted to the Office of Graduate Studies of Texas A&M University in partial fulfillment of the requirements for the degree of

MASTER OF SCIENCE

December 2007

Major Subject: Entomology

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Approved by:

Chair of Committee, Marv Committee Members, John Kirk Head of Department, Kevin

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ABSTRACT

Quantification of Blackmargined Aphid (*Monellia caryella* (Fitch)) Honeydew Production in Pecan (*Carya illinoinensis* (Koch)) in Texas. (December 2007)

Jessica Marie Honaker, B.S., Marshall University

Chair of Advisory Committee: Dr. Marvin Harris

Field studies of the blackmargined aphid, Monellia caryella (Fitch), were conducted on three cultivars, 'Cheyenne', 'Kiowa', and 'Pawnee', of pecan, Carya illinoinensis (Koch). Aphid density and natural enemy (lacewings, ladybird beetles, and spiders) densities were determined biweekly by direct inspection of 160 leaves per variety during the 2005 and 2006 growing seasons. Honeydew deposition was measured biweekly using water sensitive cards. Aphid phenologies were similar among cultivars; however, 'Cheyenne' supported higher densities of aphids than either 'Kiowa' or 'Pawnee'. Honeydew production correlated positively with aphid density. Honeydew produced per aphid differed only between 'Cheyenne' and 'Pawnee' in 2006; natural enemies per aphid varied in significance during both seasons. Natural enemy densities increased during initial stages of outbreak on all cultivars in 2006. The asymptote reached on 'Cheyenne' had a lower natural enemy to aphid ratio than that on the other cultivars, indicating that the functional response of natural enemies to increased aphid densities was exhausted sooner on 'Cheyenne than on other cultivars'. Honeydew appears to be an attractant for natural enemies and cost-benefit calculations were made to quantify the loss of photosynthates to aphids for each cultivar versus the gain in natural enemies that occurred. 'Cheyenne' was the least efficient of the three cultivars in the utilization of this defense mechanism. The energy drain per hectare attributable by adult aphid feeding was, 761,197 - 900,312kcal, 266,397 - 237,709kcal, and 138,790 - 134,223kcal for 'Cheyenne', 'Kiowa', and 'Pawnee', respectively. Calculated nut-loss equivalents were 14 – 16kg for 'Cheyenne', 4 - 5kg for 'Kiowa', and 2kg for 'Pawnee'.

ACKNOWLEDGEMENTS

I would like to thank my committee chair, Dr. Harris, and my committee members, Dr. Oswald and Dr. Winemiller, for their guidance and support throughout the course of this research. Additionally, I want to thank Dr. Juan Lopez and Dr. Dan Martin at USDA, and Dr. Leonard Lombardini for their assistance and advice.

Thanks go also to my friends and the departmental faculty and staff at Texas A&M University for providing camaraderie, support and encouragement during my pursuit and completion of this degree, and a special thanks to my lab mates who helped me so much with data gathering. Thanks also to Robert Kwiatkowski and Andrew Sherrod for allowing me to work in their pecan orchards.

Finally, I would like to thank my family for their unconditional love and patience and Kristie Reddick for giving me a reason to believe in myself.

STANDARD ABBREVIATIONS

- LWA lacewing adult
- LWE lacewing egg
- LWL lacewing larva
- LWP lacewing pupa
- SP spider
- LBA lady beetle adult
- LBL lady beetle larva
- G-growth
- E energy
- R respiration
- U-excrement
- *r* rate of increase
- J joules
- kcal kilocalories

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CHAPTER I

INTRODUCTION: PECANS AND BLACKMARGINED APHIDS

The pecan, *Carva illinoinensis*, is one of the most important crops native to the United States (Harris et al. 1986). Its native range extends from Illinois south to Mexico and Texas east to Mississippi. Natural propagation of this monoecious perennial tree obligatorily occurs through seed production primarily by masting every 2-7 years (Harris and Chung 1996). Trees can live for more than 200 years, grow higher than 35 meters, and can make up half or more of the tree canopy in their natural habitats (Maggio et al. 1991). Pecan nuts, or kernels, have a high oil content (>70% fatty acid; Beauchat and Worthington 1978) that both sustains the germinating embryo and attracts scatter hoarders that aid pecan for dispersal (Harris and Chung 1996). The wild pecan was an important component in the diet of Native Americans (Brison 1974) and commercial development of the pecan began late in the 19th Century. The domestication of the pecan and the industry that has developed around it has included the vegetative propagation of cultivars which resulted in increased genetic uniformity and has lead to a crop that is more vulnerable to pathogen and arthropod attack (Harris 1979). Approximately half of the average annual nut production of 350+ million pounds worldwide is still produced by native trees managed in situ. The large pest complex associated with C. illinoinensis includes hundreds of arthropod species, but most do not

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significantly impact pecan nut production. Key pests can and do, however, routinely reach damaging densities (Harris et al. 1992).

The blackmargined aphid, *Monellia caryella* (Fitch), is an important pest of pecan and is often the target of pesticide applications (Harris 1983). It is native to the United States, apparently having coevolved with pecan for millennia, and can be a serious pest in the pecan belt (Liao 1984). This highly fecund insect is capable of producing 16 to 32 generations per year, and, although densities generally average one or fewer aphids per compound leaf, outbreaks peaking at 10-20 aphids/leaf and lasting three to four weeks typically occur at least once during each 32-week growing season (Bumroongsook et al. 1992). Liao and Harris (1986) suggested that the fecundity of *M. caryella* is higher earlier in the growing season, leading to an increased likelihood of outbreaks during that time. Studies conducted in Israel also demonstrated *M. caryella* population peaks early in the growing season (Mansour and Harris 1988). Blackmargined aphid is considered by many to be a limiting factor in pecan production (Bumroongsook and Harris 1992).

Natural Enemies and Their Role in Pecan Systems

While many honeydew-excreting hemipterans have mutualistic relationships with other insects such as ants (Fischer and Shingleton 2001), honeydew serves as an attractant for natural enemies. Although on an annual basis densities of aphids are probably regulated primarily by factors such as seasonality (Sequeira and Dixon 1997), natural enemies are important biological control agents and can significantly influence overall aphid densities during parts of the year. While aphid parasitoids have been studied from the standpoint of aphid control populations in the United States and Israel (Mansour 1988, Bueno and Stone 1985, Mansour et al. 1988), other natural enemies, like spiders, lacewings and lady beetles are among the most important for blackmargined aphid population control (Harris and Li 1996). Spiders are ubiquitous in pecan systems and exhibit a rapid functional response to prospective prey but they have a slow numerical response to elevated aphid densities (Bumroongsook et al. 1992). Lacewings and lady beetles are more rapacious feeders than spiders, though the detectable abundance of these predators is less than that of spiders when aphid densities are low (Bumroongsook et al. 1992). Although the blackmargined aphid has been considered a limiting factor in pecan production, studies on its population dynamics suggest that it also plays an important role in natural enemy attraction in pecan systems because of its honeydew production (Bumroongsook and Harris 1992, Harris and Li 1996). Honeydew also provides an easily accessible energy source for saprophytic fungi. On the forest floor, this energy source may influence the decomposition rates and nutrient recycling of leaves, which may increase the efficiency of production in the ecosystem. The extent to which the honeydew excreted by blackmargined aphids influences the pecan-associated arthropod complex, including the occurrence and abundance of natural enemies has not been studied in detail.

Mortality and Rates of Increase

Understanding how quickly an arthropod pest can reach damaging densities is an important tool in deciding appropriate methods of control. Estimating the rate of increase (r) of an aphid population is complicated because aphids are migratory, highly polymorphic, have a short development time, and a long reproductive life. Most estimates of r for aphid species are made based upon data obtained through laboratory rearing studies. These data are then used to estimate rates of population increase under field conditions. It is doubtful that these methods give accurate estimates of aphid rates of increase (Dixon 1977). Blackmargined pecan aphid is a monoecious, obligatorily alate species, oligophagous on *Carya* spp. Liao (1984) measured r from direct measurements of blackmargined aphids taken from *C. illinoinensis* in the field by meticulously determining instar ratios through time. Although Liao's method is considered accurate, more efficient methods of measuring intrinsic rates of increase and mortality are needed to quickly gauge the true impact of aphid outbreaks.

Energy Drain

Monellia caryella is a phloem-feeding aphid that aggregates on the major veins on the undersides of pecan leaves. Studies on willow aphids have shown that the composition of nutrients in the leaves on which aphids feed influences both aphid growth rate and honeydew excretion rate (Llewellyn et al. 1974). The extent of energy loss in pecan due to the blackmargined aphid is controversial. Wood et al. (1987) argued that blackmargined aphid poses a significant threat to pecan production, even at low densities, while Harris and Li (1996) suggested that the blackmargined aphid-pecan interaction may benefit the pecan. Further characterization of this interaction is needed to reconcile these different views.

Objectives

There were three areas of investigation in this study: 1) the relative susceptibility of selected pecan cultivars to blackmargined aphids, 2) a comparison of honeydew production to densities of selected natural enemies, and 3) determination of whether or not honeydew droplet size could be correlated with aphid instar. The USDA pecan varieties, 'Cheyenne', 'Pawnee', and 'Kiowa' were monitored to determine aphid densities and the volumes of honeydew produced by the aphids. Calculations of energy loss due to aphid honeydew production by variety during the growing season were factored into estimates of expected production loss. Natural enemy data were gathered and compared with honeydew production levels to determine whether volumes of honeydew excreted by blackmargined aphids are correlated with the abundance of three natural enemy taxon groups in pecans. Measurements of honeydew droplet size of all aphid instars were used to determine if direct estimates of mortality and rates of increase could be determined from honeydew droplet data. Honeydew droplet size was also compared with aphid instar to examine whether the size of the anal opening of each instar was correlated with the total amount and droplet size of honeydew excreted.

This study uses proven field techniques together with new computer technologies to help further understanding of the role that honeydew plays in commercial pecan production, pest management, and overall orchard health, as well as advancing our knowledge of the ecology and biology of aphids and their natural enemies.

CHAPTER II

MATERIALS AND METHODS

Aphid and Natural Enemy Counts and Honeydew Collection

The first season of field data was taken from pecan tree varieties 'Pawnee', 'Cheyenne', and 'Kiowa', each tree being replicated four times per variety. Sampled trees were located in the Holmes Orchard in Mumford, Texas (30°44'N, 96°33'W). The second season of data was taken from the same three varieties, with 'Cheyenne' and 'Kiowa' being replicated four times per variety and 'Pawnee' being replicated twice; sampled trees were located in the Royalty Pecans orchard in Caldwell, Texas (30°36'N, 96°33'W). This unplanned change of study location was necessitated by a change of ownership in the Holmes orchard during the study. In both seasons, leaves facing each of the four cardinal directions (N, S, W, and E) were monitored two or three times weekly for adult and immature blackmargined aphid and natural enemy densities on each tree on the selected cultivars. From each direction, ten leaves consisting of ~10 leaflets each were examined. These leaves were located at the bottom of the canopy, within reach of the researcher. The natural enemies of interest were spiders (recorded as 'spider' regardless of whether it was an immature or an adult), lacewings (recorded as egg, larva, pupa, or adult), and ladybird beetles (recorded as egg, larva, pupa, or adult). Each arthropod count was converted from a by-tree number to a by-hectare number by first dividing the total number of arthropods found by the total number of sampled leaves to attain a per-leaf count; then, multiplied by the total number of leaves found in one hectare. This was done to make the measurements consistent with the honeydew measurements.

For both seasons, honeydew was collected two or three times weekly using four 26x76 mm Teejet Water and Oil Sensitive Paper cards (Spraying Systems Co., Switzerland) mounted into fitted depressions cut into a 23x15.25 cm corrugated cardboard holder. To provide a stable and standard placing for these water-sensitivepaper card holders, a wooden platform was constructed using pine boards cut into 16 cm squares on a side and nailed to 46 cm dowel. At the end of each dowel, a double-ended screw was inserted, allowing one end to be drilled into the tree trunk, permanently affixing the platform to the tree. Each wooden platform was used to house one cardboard water-sensitive-paper card holder. The holder fit over a 5 cm screw inserted through the bottom of the platform, and a nut was screwed down over it to prevent the card from moving in the wind. Platforms were placed on sampled trees in such a manner that they avoided overhanging limbs or honeydew drift from other trees, and allowed for direct interception of honeydew falling from the canopy. The water-sensitive cards were placed in the holders using double-stick tape and left for measured periods of time (hrs) twice each week for about 3 months.

Honeydew droplets on the water sensitive cards were analyzed for volume using DropletScan® software (WRK, Inc. and Devore Systems Inc.). This provided a quantification of the honeydew volume collected in gallons per acre (later converted to liters per hectare) on a per-card basis during the exposure time of the cards. These specific mathematical derivations of these volumetric analyses were obtained through the manufacturer's calibration in the software, and unknown to the researcher. This software also provided distribution/abundance data on droplet size for analyzing the abundance of aphids of various sizes.

Aphid and natural enemy counts were converted from aphids or natural enemies/leaf to aphid or natural enemy/ha, based on leaves/ha estimates by Harris and Li (1996). All numerical data were analyzed using SPSS statistical software (Chicago, IL.) Due to the high level of non-normality in the data, the statistical methods used in this study were primarily the Kruskall-Wallis test to calculate the significance among the three cultivars. The Mann-Whitney U test was used when comparing two of the three cultivars.

Honeydew Droplet Size and Aphid Instar

Five adult blackmargined aphids were placed in a clip cage on a pecan leaf and left for 24 hours to allow for the establishment of a cohort of immature aphids. The adults were removed with a paintbrush and an aphid population of known age remained. A water-sensitive card cut to fit the interior of the clip cage was placed in each cage for a measured period of time (hrs) to collect honeydew excretions. To provide a flat surface for accurate honeydew measurement while the water-sensitive card was in the cage, a metal cookie tin was hung from the branch above the chosen leaf and for the time the cards were in the cages, the cages were placed in the cookie tin. Honeydew collections were made daily and taken to the laboratory for analysis with DropletScan® daily until adults emerged. A voucher specimen of one aphid from each cohort was taken daily as well. This procedure was replicated 15-20 times using one tree of a susceptible cultivar. This tree was located at the Entomology Research Lab building on Agronomy Road in College Station, Texas (30°36'N, 96°21'W). Cage placement was in the lower canopy for convenient access from the ground. Honeydew droplet size and volume were measured using DropletScan® software. These data were then analyzed to determine if droplet size and volume could be correlated with aphid instar.

Due to the tendency to 'blue', or darken due to moisture, the water-sensitive cards were somewhat restricted in environments with high humidity. Additionally, in places where excessive honeydew production was present, the card was completely saturated and quantification of honeydew was prevented beyond the upper limit of that detectable by the card technology. Both high humidity and excessive honeydew were observed to give false readings, and those cards judged to be anomalous were excluded from the analysis.

Energy Drain

Estimated energy removal by blackmargined aphid during the 2006 growing season was calculated two different ways. Using aphid density data, the number of adult aphids was plotted against sample date and adult aphid-days/leaf was determined for each cultivar for the 2006 season: one adult on one leaf for one day = one aphid-day (Southwood 1968). The total "adult aphid-days/leaf" for each cultivar was then divided by 14.2 (1/2 the adult longevity period (Wood et al. 1987)) to provide an absolute estimate for the number of blackmargined aphid life-spans that reached the midpoint of

their development; this was converted to per hectare data by using a multiplier of $1.5*10^6$ leaves/ha (from Cutler (1976) and Lozano et al. (1992) who measured ~3 million leaves/ha, which was corrected to 50% canopy coverage). According to Wood et al. (1987), one blackmargined aphid life-span removed 301.41j of energy to complete development. Aphid life-spans/ha * 301.41j was used to calculate the energy removed per adult aphid life-span during the 2006 growing season.

To obtain the energy removed due to nymph presence in the canopy, the number of adult life-spans was multiplied by adult female fecundity of 100 nymphs per aphid (determined from Flores-Flores (1981) and Sears (1985) calculations). This entire initial nymph density was assumed to complete the 1st instar, 75% were assumed to complete the 2nd instar, 50% the 3rd and 25% the 4th; using energy values calculated per instar by Wood et al. (1987), energy removal per instar was calculated by multiplying nymph densities per instar by energy removed for that particular instar. These energy values were then summed to obtain the total energy removed by the nymphs. This value was added to energy removed by adults to estimate total energy removed by the aphid population during the season.

The second way energy removal was calculated was by using honeydew volume. Honeydew volumes used to calculate energy drain in this study were obtained directly from the water-sensitive cards placed in the canopy for the cultivar ratings. The honeydew reaching the cards was analyzed using DropletScan software to determine L/ha, and corrected for canopy coverage and the filtering effect of leaves that intervened between the top of the canopy and the card surface. The latter was aided by leaf area index measurements Honeydew volumes used to calculate energy drain in this study were obtained directly from the water-sensitive cards placed in the canopy for the cultivar ratings. The honeydew reaching the cards was analyzed using DropletScan software to determine L/ha, and corrected for canopy coverage and the filtering effect of leaves that intervened between the top of the canopy and the card surface. The latter was aided by leaf area index measurements using a LiCor 2000 (LI-COR, Lincoln, Nebraska USA 68504-0425).

Honeydew volume in liters per hectare were plotted for each cultivar by sample date and interpolated for each cultivar to determine the total volume of honeydew (in liters) over the course of the 2006 growing season. Auclair (1963) estimated that aphids excrete about 50% of the sugar ingested from photosynthates. Thus, the calculated energy removed by the aphid was doubled to obtain total photosynthates removed. Blackmargined aphids, however, are five percent efficient (Wood et al. 1987) and taking this into account, 5% of the energy removal estimate was calculated, and then added back to the original estimate. Additionally, the "filtering effect" of overlapping leaves in the canopy resulted in only about half of the honeydew produced for the entire tree reaching the water-sensitive cards positioned at the bottom of the leaf canopy. This assumption is based on leaf area index (LAI) data obtained for the sample trees. Taking LAI into account (see below), the energy represented by calculated honeydew was again doubled. Leaf area index data were gathered using LI-COR instrumentation. Sunlight readings were taken at noon in full sun before taking readings of sunlight filtering through the canopy of the sample trees. In this way, a measurement of area of pecan leaf area squared per ground area squared was obtained (Appendix B). The LAI measurements ranged from 3-5 and the leaves are positioned at an angle to the ground depending in part on the angle of incidence of the sun into the canopy. Honeydew measurements were obtained during the early afternoon so that a leaf inclination of 10-20 degrees would have resulted in maximum surface area exposure to sunlight. Since all infested leaves in the lower canopy were directly contributing to honeydew deposition on the cards, the "filtering effect" was only of concern for honeydew emanating from infested leaves higher in the canopy. Given the LAIs from 3-5, the minimum deposition range would be 20-33.3% of total honeydew if all the leaves were oriented parallel to the ground and perfectly overlapped so that all honeydew produced above the lower canopy leaves was intercepted before it could reach the cards. This would be an underestimate since the leaves are known to orient toward the sun as much as possible so that the orientation of the lower surfaces to the ground increases (note that if leaves were perpendicular to the ground that 100% of honeydew could reach the ground). We lack direct measurements for this calculation, and estimate that 50% of honeydew produced in the canopy reached the ground in our study.

The photosynthates caloric values as calculated above using both honeydew volume and aphid densities were converted to grams of nut tissue using Harris and Chung (1996) value of 11.15 Kcal/gm to convert photosynthate to nut tissue.

CHAPTER III

RESULTS

Cultivar Ratings

Aphid Density

Monellia caryella densities increased in June with a smaller, secondary peak occurring later (Figs. 1-2). Peak densities of blackmargined aphid on the 'Cheyenne' variety were consistently higher than those on 'Kiowa', and 'Kiowa' maintained a slightly higher density of *M. caryella* than 'Pawnee' throughout the sampling period.

Honeydew Collection

The volume of honeydew collected per cultivar peaked early in the growing season in 2005, with the relationship in honeydew deposition being 'Cheyenne'>'Kiowa'>'Pawnee'. A secondary peak occurred, most noticeably on 'Cheyenne'. In 2006, similar patterns were found to those in 2005, with a greater volume of honeydew being collected from 'Cheyenne' than from the other two varieties (Figs. 3-4).



Figure 1. Mean blackmargined aphid density on three pecan cultivars over the 2005 growing season.



Figure 2. Mean blackmargined aphid density on three pecan cultivars over the 2006 growing season.



Figure 3. Mean honeydew production for three pecan cultivars during the 2005 growing season.



Figure 4. Mean honeydew production for three pecan cultivars during the 2006 growing season.

Honeydew per Aphid

Honeydew produced on a per-aphid basis did not differ significantly among the three cultivars sampled in either year with the exception being at 'Cheyenne' vs. 'Pawnee' in 2006 (Table 1).

Cultivar	2005	2006
'Cheyenne' vs. 'Kiowa'	0.827	0.114
'Cheyenne' vs. 'Pawnee'	0.073	0.020*
'Kiowa' vs. 'Pawnee'	0.168	0.443

 Table 1. P values of honeydew volume produced per aphid among cultivars for the 2005 and 2006 growing seasons.

* indicates a significant difference in measured parameters.

Aphid and Natural Enemy Counts and Honeydew Collection

Aphid Density and Honeydew Volume

'Cheyenne' was found to have significantly higher relative aphid densities and higher levels of honeydew than either 'Kiowa' or 'Pawnee' while 'Kiowa' and 'Pawnee' aphid densities and honeydew did not significantly differ from one another in both seasons (Table 2).

	2005		2006	
	Mean Honeydew	Mean Aphid	Mean Honeydew	Mean Aphid
	Production	Density	Production	Density
	(L/ha/day)		(L/ha/day)	
'Cheyenne' vs.	0.027*	0.001*	>0.01*	>0.01*
'Kiowa'				
'Cheyenne' vs.	>0.01*	>0.01*	0.001*	0.002*
'Pawnee'				
'Kiowa' vs.	0.069	0.082	0.403	0.069
'Pawnee'				

Table 2. *P* values of mean honeydew production and mean aphid densities compared among cultivars during the 2005 and 2006 growing seasons.

* indicates a significant difference in measured parameters.

Relative densities of natural enemies (lacewing larva, ladybird beetles, and spiders) were monitored throughout the sampling period among the three pecan varieties. Relative mean spider densities among the cultivars did not differ significantly over the course of the growing season in 2005 or 2006. Lacewing egg, lacewing larvae and ladybird beetle mean densities were not significantly different in 2005 but were in 2006 (Table 3).

Table 3. *P* values of measured parameters for three pecan cultivars ('Cheyenne', 'Kiowa', and 'Pawnee') during the 2005 and 2006 growing seasons.

PARAMETER	2005	2006
Mean Aphid Density	<0.01*	<0.01*
Mean Honeydew Volume	<0.01*	<0.01*
Mean Spider Density	0.382	0.477
Mean Lacewing Egg Density	0.725	<0.01*
Mean Lacewing Larva Density	0.406	<0.01*
Mean Lady Beetle Density	0.524	0.003*

*indicates a significant difference in measured parameters

Natural Enemy Density - Trends

An early season peak in mean natural enemy density that coincided with primary

the aphid outbreak occurred in 2005 and 2006 (Figs. 5-6).



Figure 5. Mean number of natural enemies per hectare for three pecan cultivars during the 2005 growing season.



Figure 6. Mean number of natural enemies per hectare for three pecan cultivars during the 2006 growing season.

Natural Enemy Density by Taxon Group

When mean natural enemy densities were compared among taxon groups, spiders had the highest density, followed by lacewing (larvae), then ladybird beetles (adults & larvae) (Figs. 7-8). When the ratios of natural enemies-to-aphids were compared, variation was found by both cultivar and by year.



Figure 7. Mean densities of three natural enemies over the 2005 growing season.



Figure 8. Mean densities of three natural enemies over the 2006 growing season.

There was a significant difference in spider density (p< 0.05 for all cultivars) per aphid among cultivars in 2005; in 2006, spider densities per aphid among 'Cheyenne' and 'Kiowa' differed significantly but densities on 'Pawnee' did not differ significantly from either 'Cheyenne' or 'Kiowa'. In 2005, there was a significant difference in lacewing egg deposition among cultivars for all cultivar comparisons, and no significant difference in the density of lacewing larva or ladybird beetle. In 2006, there was a significant difference among 'Cheyenne' and 'Kiowa' in lacewing egg density; a significant difference among 'Cheyenne' and 'Kiowa', and 'Cheyenne' and 'Pawnee' in ladybird beetle density, and a difference among all three cultivars in lacewing larval density (Table 4).

	<u>, , , , , , , , , , , , , , , , , , , </u>			
	Lacewing	Lacewing	Ladybird	Spider
	Egg	Larva	Beetle	
		2005		
'Cheyenne' vs. 'Kiowa'	>0.01*	0.657	1.00	0.030*
'Cheyenne' vs. 'Pawnee'	>0.01*	0.417	0.417	> 0.01*
'Kiowa vs. 'Pawnee'	0.014*	0.209	0.369	>0.01*
2006				
'Cheyenne' vs. 'Kiowa'	0.017*	0.031*	0.022*	>0.01*
'Cheyenne' vs. 'Pawnee'	0.057	>0.01*	0.021*	0.342
'Kiowa vs. Pawnee'	0.961	0.047*	0.790	0.107

Table 4. P values of natural enemy-to-aphid ratios during the 2005 and 2006 growing seasons.

*indicates a significant difference in measured parameters

Natural Enemies by Date

A comparison of measured parameters (aphid density, honeydew volume, and natural enemy densities) by sample dates during the 2005 and 2006 growing season was made by comparing data taken on the same day grouped from all three cultivars with data gathered on other sampling dates (Figs. 9-10). In 2005, there was no significant difference (p > 0.05) in any of the parameters measured during the sampling period, except relative spider densities, which differed significantly (p < 0.05) between 23 June and 1 August from the rest of the season (Fig. 7). In 2006, there was also no significant difference in the parameters of interest when the cultivar data was combined by date, except spider densities from other natural enemies (Fig. 8), which differed significantly on six of the sampling dates between 13 June and 4 August.



Figure 9. Mean natural enemy to aphid ratio for three pecan cultivars for the 2005 growing season.



Figure 10. Mean natural enemy to aphid ratio for three pecan cultivars over the 2006 growing season.

Measurements obtained during the 2005 season were begun in the middle of the early-season peak in aphid densities, and thus represent the latter half of the outbreak. When the data gathered in the mid-season (July to August) were compared, the same patterns of early season peaks were found.

Honeydew Droplet Size and Aphid Instar

Output from the proprietary DropletScan software used to scan and quantify honeydew data reports droplet number by droplet size class in the form of histogram plots; the droplet size was ranked according to the pre-programmed measuring parameters of the software. Those droplets measuring 0-100 μ m, 101-200 μ m, 201-300 μ m, 301-400 μ m, 401-500 μ m, and 501-600 μ m were ranked as 1-6, respectively. The high humidity inside the clip cage stained the water-sensitive papers slightly. All measurements ranked as 1 were discarded, because this measurement was present even when no honeydew was collected on the cards. Pearson's correlation of honeydew droplet size to aphid instar was non-significant (p = 0.074), despite apparent visual evidence of changing droplet size in later instars (Fig. 11).



Figure 11. Comparison of honeydew droplet sizes for one generation of blackmargined aphid.

While the DropletScan technology may ultimately have application for determining aphid instar from honeydew droplet size, measurement constraints in the configuration of the present software prevents sufficient discrimination of the relevant droplet sizes to identify such a correlation. Additionally, the measurement of honeydew volume collected on the cards proved to be difficult, as the high humidity in the clip cages darkened the cards and likely made any volume readings inaccurate.

Energy Drain

Energy drain calculated from aphid density data range from 138,790 kcal to 900,312 kilocalories over the 2006 growing season (Table 5). The calculated nut equivalents for these values are 80.74kg ('Cheyenne'), 23.89kg ('Kiowa') and 12.44kg ('Pawnee') nut loss per hectare.

Table 5. Energy (in kcal) removed by blackmargined aphid in three cultivars of pecan over the 2006 growing season based on aphid density data.

Cultivar	Aphid (adult) Energy Removal (kcal)	Aphid (nymph) Energy Removal (kcal)	TotalEnergyRemoval (kcal)
Cheyenne	23,661	876,651	900,312
Kiowa	6,994	259,396	266,397
Pawnee	3,647	135,142	138,790

Energy removal based on honeydew volume data collected in 2006 growing season ranged from 134,223 kcal to 761,197 kcal (Table 6).

Table 6. Energy (in kcal) removed by blackmargined aphid in three pecan cultivars over the 2006 growing season based on honeydew volume data.

Cultivar	Energy Removal in kcal/ha
'Cheyenne'	761,197
'Kiowa'	237,709
'Pawnee'	134,223

Nut equivalents calculated using energy removed values from both honeydew volume and aphid density data are as follows: 'Cheyenne', 68 - 81kg, 'Kiowa', 21 - 24kg, and 'Pawnee', 12kg per hectare, assuming 100% of photosynthates in the tree are allocated to nut production. These photosynthates removed by the blackmargined aphid in June-July are primarily sugars that are used to satisfy the entire physiological needs of the tree. Realistically, only about 20% of energy available as photosynthate can be allocated to nut production, so these values were adjusted accordingly to attain a more accurate estimate of actual nut loss. Adjusted nut loss (in kg) was, for 'Cheyenne' 13 - 16kg, 'Kiowa' 4 - 5kg, and 'Pawnee' 2.5kg.

CHAPTER IV

CONCLUSIONS

Of the three pecan cultivars considered in this study, 'Cheyenne' had both higher aphid densities and greater collected honeydew volume. Despite the variation in significant differences upon cultivar comparison in natural enemy densities, the patterns of density increase and decrease for natural enemies generally track that of aphid population and honeydew volume fluctuations. Therefore, the cultivar rating of 'Cheyenne' would rank lower in desirability (because of the high densities of blackmargined aphids supported and the greater volume of honeydew collected) than 'Kiowa', and 'Pawnee' ranking higher in desirability than the other two cultivars with regard to both aphid densities and honeydew collection.

Because the volumes of honeydew collected from 'Cheyenne' in both seasons were significantly higher than those collected on 'Kiowa' or 'Pawnee', except in the 2006 growing season, the question of what factor(s) promotes this difference in deposition is raised. The examination of honeydew volume-per-aphid shows there is no significant difference in honeydew per aphid by cultivar (except 'Cheyenne' and 'Pawnee' in 2006; Table 2), indicating the energy drain per aphid on all three varieties is generally similar on a per aphid basis; 'Cheyenne' simply supports more aphids for a longer time period than the other two cultivars.

Difficulties in the droplet size-to-aphid correlation study hindered the gathering of desired data for this section of the study. However, this method would be more effective in areas of lower humidity and, perhaps, clip cages with better ventilation. Software designed specifically for measuring individual droplets is also crucial in the gathering of this data. Though no concrete data on a correlation between honeydew droplet size and aphid instar could be attained for this study, the importance of such a measurement should in no way be underestimated. Better understanding of the population dynamics of this arthropod can allow pecan growers to decide whether or not it is economical to spray for blackmargined aphid when outbreaks occur.

Calculations of energy removal based on the aphid density and honeydew volume data over the 2006 growing season provided estimates of nut-loss well below the average annual harvest of ~1000kg per hectare.

Financial loss due to feeding by blackmargined aphid in this study is far outweighed by the cost of spraying pesticides to eliminate them. The current action level for blackmargined aphid in pecan systems is 25 aphids per compound leaf, and for a brief period in 2006 that limit was exceeded. The economic threshold level should be four times the cost of treatment – that is, the economic loss should be four times greater than the cost of applying pesticide. At \$2.50 per kg, the profit loss per hectare is \$34 – 41 for 'Cheyenne', \$10 – 12 for 'Kiowa', and \$6 for 'Pawnee'. Comparatively, the cost of spraying pesticide is ~\$50 per hectare; at the economic threshold level, the three cultivars in this study are only 1/6, 1/19, and 1/32 of threshold levels for 'Cheyenne', 'Kiowa', and 'Pawnee' varieties, respectively.

Based on the data for the 2006 season, negligible financial loss due to blackmargined aphid presence (even with densities exceeding action levels) suggests that economic threshold levels for the insect should be re-evaluated and increasing the threshold levels to 30+ aphids/leaf should be considered.

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APPENDIX A



A-1. Overhead view of Mumford pecan orchard in Mumford, Texas (photo by Google Earth, 2007).



A-2. Overhead view of Royalty Pecans Orchard in Caldwell, Texas (photo by Google Earth, 2007).

APPENDIX B

Lincoln, Nebraska USA 68504-0425).			
Cultivar	Tree	LAI	
'Cheyenne'	1	3.64	
	2	3.62	
	3	5.05	
	4	5.51	
'Kiowa'	1	4.58	
	2	4.92	
	3	6.53	
	4	5.43	
'Pawnee'	1	4.70	
	2	5.71	

B-1. Leaf area index measurements for sampled cultivars in 2006 taken using a LiCor 2000 (LI-COR, Lincoln, Nebraska USA 68504-0425).

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