Abundance and Species Diversity of Thrips (*Thysanoptera: Thripidae*) in Cotton, Soybean, and Peanut in Southeast Virginia, and Evaluation of Cyantraniliprole for Thrips Management

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ABSTRACT

Thrips are major agricultural pests throughout much of the United States. More information is needed about sampling methods, management practices, and insecticide susceptibility to help better control this pest. A two year survey was conducted to determine the species present in southeast Virginia and the population characteristics of those species. Thrips were monitored using yellow sticky traps. Tobacco thrips, *Frankliniella fusca*, were the most abundant species. In general thrips populations began to build up beginning in April, peaked in August, and then started to decline. Differences in this trend were observed between species.

A study was conducted in seedling soybean to evaluate the within-plant location of thrips, whether a plant subsample could be used for thrips monitoring, and to determine the thrips species complex present. Soybean thrips, *Neohydatothrips variabilis*, were the most prominent species present. The greatest density of thrips larvae was located in the terminal bud of the seedling and suggests that immature thrips aggregate. Neither of the proposed subsamples of plant material explained the variability in immature thrips numbers and at this time we recommend whole-plant sampling for obtaining the most accurate estimate of thrips populations in seedling soybean.

Tobacco thrips, *F. fusca*, as well as a complex of other thrips species attack cotton and peanut seedlings and can cause significant yield loss to these crops in the mid-Atlantic U.S. Experiments were conducted in these two crops to assess the efficacy of a novel diamide insecticide cyantraniliprole applied as a liquid in-furrow at planting and post-plant emergence broadcast spray treatment to control thrips. In both cropping systems cyantraniliprole significantly reduced the number of immature thrips and reduced thrips feeding injury to the plants. In several instances cyantraniliprole treatments resulted in increased yield as compared

to the non-insecticide treated control and yields which were statistically similar to those obtained with standard thrips control insecticides.

Laboratory bioassays were conducted to evaluate the toxicity (LC_{50} values) of cyantraniliprole and two conventional insecticides against *F. fusca* adults. Results of these assays were inconclusive. At times *F. fusca* adults were susceptible to the insecticides, but the results could not be replicated consistently.

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Research is never a singular effort but requires time and dedication from a number of contributors. I have been blessed to work alongside several excellent technicians and graduate students and without their help this research would still be in its infancy. My deepest thanks to Dr. Sean Malone, Mike Arrington, Rebecca McGrath, and David Owens. It has been a pleasure to work and learn from you all.

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Introduction

Thrips (Thysanoptera: Thripidae) are worldwide crop pests causing damage to a wide array of commodities ranging from row crops to fruit to ornamentals. Much of the injury they cause is through direct feeding or oviposition, but there are several species which also transmit viruses that are harmful to certain plants. Cotton, peanut, and soybean are just a few of the crops in Southeastern Virginia which are adversely affected by thrips. Their populations build steadily and rapidly early in the growing season and susceptible crops if not treated will incur high populations that cause injury to the plants, delayed maturity, stunted growth, and yield loss.

There are many different insecticides with efficacy against thrips. In cotton and peanut carbamates like aldicarb and organophosphates like acephate are common standards. Pyrethroids are also frequently used, but the increasing incidence of resistance and general lack of efficacy to this class of chemicals is forcing many researchers and growers to abandon it for thrips control. New insecticides with different modes of action and lower toxicity to mammals and beneficial insects are being developed which will offer growers and researchers some alternatives. If used properly, these novel chemistries will help lessen the occurrence of resistance. A portion of the research discussed in this thesis examines both standard and novel insecticides and their efficacy for thrips control.

Before making any management decision, it is important to know the species of insects present in a crop. Not all species of thrips are equally susceptible to a given insecticide; western flower thrips (*Frankliniella occidentalis*) for example are known to be resistant or to develop resistance rapidly to many standard insecticides used for thrips control. In southeast Virginia, little documentation existed about the common species found infesting row crops. This lack of information was cause for concern because it meant that growers might be making management decisions without full knowledge of the pests they were trying to combat. Therefore, research presented herein monitored and recorded the species found in Southern Virginia; it also examined the population dynamics of several of the most commonly documented species in an effort to gather important information for both growers and researchers that could be used to improve thrips management programs.

Chapter One

Literature Review

Thrips Biology and Anatomy

Thrips are minute insects on average reaching no more than 1mm in length. Those with wings are weak fliers, but their wings are fringed with long setae allowing them to be transported great distances by the wind. Even wingless thrips can take advantage of wind transport because all thrips have a large surface area to volume ratio (Mound 1996). Thrips vary in color from pale white to yellow, dark brown, or black. While most are solidly colored a few species have bands or dark spots. This coloring enables them to blend in well with their environment and avoid detection by predators; light colored thrips may take on a green appearance from their food hosts reflection while dark colored thrips, aided by their small size, resemble a piece of dirt or sand.

Thrips have unusual asymmetrical mouthparts with the right mandible being vestigial and greatly reduced. They have two well-developed maxillary stylets which join together to form a sucking tube. These parts are enclosed and protected by a mouth cone formed by the labrum and labium. Earlier literature stated thrips were rasping-sucking feeders (Huckaba and Coble 1991, Childers and Achor 1995, Kirk 1995). Newer evidence shows them to be piercing-sucking feeders. To feed, most thrips press their mouth cone against the plant surface; this is held in place by the labral pad. The mandible is used to pierce through the substrate; it is quickly removed and replaced by the maxillae which locate an individual plant tissue cell and suck out the contents (Chisholm and Lewis 1984, Childers and Achor 1995, Kirk 1995, Mortz 1997). When feeding on pollen grains, thrips use their forelegs and/or palps to hold the grain while they pierce it (Childers and Achor 1995).

Feeding

Most thrips are phytophagous, but a few genera will feed on fungi or are considered predatory feeding on other thrips, mite adults and eggs, scale insects, and whiteflies (Mound

and Teulon 1995). Phytophagous thrips will feed on the flowers, leaves, fruit, pollen, and nectar of plants. Because thrips must pierce through the outer tissue wall to reach the cells, they prefer to feed on young seedling plants which have not yet developed tough outer tissue walls and/or defense mechanisms such as wax layers.

Direct and Indirect Plant Damage

Thrips preference to feed on young plants and their ability to transmit diseases through their saliva while feeding makes them detrimental early season pests to many different crops. Larvae tend to cause more damage than adults because they occur in larger numbers and some species are gregarious (Childers 1997). Their damage to crops includes stunted plant growth, leaf stippling, distortion, blemishes, slowed maturity, plant death, and reduction in yield and quality. Quite often, thrips damage is not readily apparent because effects are delayed; the pest may not even be present by the time damage is noticeable. Oviposition and feeding injury cause direct damage to crops (Childers 1997). In 2008, thrips caused an estimated loss of 2, 625 bales of cotton in Virginia (Herbert et al. 2009). At the end of the 2007 tomato season, several large commercial growers reported millions of dollars in losses due to thrips feeding injury (Herbert et al. 2009). In 2009, research showed a 12% yield loss from thrips in non-insecticide treated Virginia peanuts (Herbert et al. 2009).

Thrips may also cause indirect damage by transmitting viruses or as passive carriers of fungal and bacterial spores (Childers and Achor 1995). For example, tobacco thrips, *Frankliniella fusca* (Hinds), (Johnson et al. 1995) and western flower thrips, *F. occidentalis* (Pergande), are major vectors for tomato spotted wilt virus (Bunyaviridae: *Tospovirus)* (TSWV) (Morsello et al. 2008). In Virginia, North Carolina, and Georgia tobacco thrips accounted for a large majority of the vector species in peanuts, tobacco, and tomato (McPherson and Beshear 1990, Johnson et al. 1995, Groves et al. 2002, Groves et al. 2003, Nault et al. 2003, Herbert et al. 2007). TSWV has a host range of greater than 600 plant species. Not all of the hosts support thrips reproduction however and some may be a dead end for the virus (Groves et al. 2002). In crops, TSWV affects not only tomatoes, but peppers, peanuts, and tobacco as well as many ornamental species. Thrips acquire the virus from infected plants while feeding. Once the virus

has successfully passed through the thrips gut it will migrate to the cells of the salivary gland (Kritzman et al. 2002). There the virus can replicate and pass into a new plant host through the thrips salivary secretions during feeding. There is a 3-7 day latent period after which thrips remain competent vectors for the rest of their lives (Groves et al. 2002). While thrips can become infected with TSWV throughout their life, for transmission, the virus must be acquired during the larval stage while feeding on infected plant tissues. The highest incidence of successful acquisition occurs during the first instar (Kritzman et al. 2002).

Finally, it has been shown that feeding by certain thrips species will cause stress induced ethylene production by the injured plant. These species saliva also contains ethylene which is injected into the plant. Excess ethylene is detrimental to the plant; in Arizona cotton over 90 per cent of early bud death is attributed to excess stress ethylene (Childers 1997).

Thrips as Opportunists

Mound and Teulon (1995) have demonstrated that the large majority of thrips species can be considered opportunists. This enables them to accumulate large host ranges and adapt to rapidly changing environments. In the general sense, most predators and parasitoids have a very limited, restricted host range. Predatory thrips, however, have remained opportunistic and have been documented feeding on prey outside of their normal host preference. The predatory species, *Aeolothrips intermedius* (Bagnall) will even consume pollen if their natural prey source is limited. Western flower thrips, tobacco thrips, and onion thrips (*Thrips tabaci* Lindeman) which are primarily thysanopteran crop pests, have been documented eating mites (Boykin et al. 1984, Milne and Walter 1998, Zhi et al. 2006). In another less extreme example, species which are considered host-specific to one type of plant have been reported as host-specific on a different plant type in another region. The ability of opportunistic thrips to quickly adapt to a new environment, colonize, and establish large populations further contributes to their ability to become significant agricultural pests.

Reproduction

Thrips are also aided in their ability to harm crops by their high fecundity and rapid reproductive rates. All thrips have a haploid-diploid reproductive strategy. Females result from fertilized eggs and unfertilized eggs produce male progeny. Also in some species, females can reproduce without males (parthenogenesis) by doubling chromosomes within the egg to produce a female; a process known as thyelytoky. In a few asexual species it is believed temperature or microbe presence or absence determines the offspring's sex (Mortz 1997). The length of one full reproductive cycle varies between species; on average it is reported to be 30 days but in warm temperatures it may be as short as two weeks (Zhang et al. 2007). Eggs are laid in or less commonly on plant tissue; the larva emerges and begins to feed upon the plant. Thrips drop to the ground and seek out a dark sheltered hiding place in which they pupate. The pre-pupal and pupal stages are non-feeding and immobile (unless disturbed) (Thoeming et al. 2003). Cold temperatures will not immediately kill thrips, but will render them temporarily immobile. During the winter months, some thrips species survive on various winter weed hosts continuing to feed and reproduce on days when the temperature is moderately warm.

Thrips Species

Although there are numerous thrips species, two of the most important in the Southeastern United States are the tobacco thrips and the western flower thrips. Tobacco thrips are of average size (~1mm) and usually dark brown in color, but may also be dark yellow. This is one of the most abundant species collected in several Southeastern United States crops (McPherson and Beshear 1990, Cook et al. 2003). It is known to overwinter in the southeast and has been reported to achieve high numbers on late winter alfalfa and clover. It has also been determined that tobacco thrips winter populations contain mainly brachypterous (wingless) adult females, however macropterous (winged) females become dominant as spring approaches (Johnson et al. 1995). Western flower thrips are large in size (~2mm) compared to other thrips species and have a milky yellow appearance. In Virginia and North Carolina, they are known to occur in cotton, soybean, and tomato, but their occurrence is currently highly variable from field to field. Like tobacco thrips, western flower thrips are also known to

overwinter in the southeast (Cho et al. 1995). However, their population size is still closely monitored due to their propensity for developing resistance to most common pesticides (Eger et al. 1998, Jensen 2000, Espinosa et al. 2002). While most researchers and growers consider western flower thrips to be a pest species, they are occasionally considered to be beneficial in California because they eat spider mites on young cotton seedlings (Wilson et al. 1991, Dreistadt et al. 2009).

Management Challenges

A common problem in pest management is the extensive over use of pesticides leading to resistance development (as is the case with western flower thrips), secondary pest flares, and increased cost to producers and consumers. Some of the most heavily treated crops, like tomatoes may have pesticides applied weekly during peak pest times. Further exacerbating this problem is the promotion of what appears to be unnecessary pesticide treatments. In soybean, for example, there is promotion of the use of seed which has been pretreated with pesticides (fungicides and insecticides) and the use of in-season foliar pesticide sprays. However, research in Suffolk Virginia over the past 9 years (unpublished data) and research conducted in North Texas (Heitholt et al. 2006) has shown that these treatments do not increase or lend an advantage to total yield. Resistance of western flower thrips to commonly used organophosphate, carbamate and pyrethroid insecticides has been documented following their overuse (Eger et al. 1998, Bielza 2008) as well as a few in other classes including abamectin (Immaraju et al. 1992), DDT (Zao et al. 1995), and spinosad (Broadbent and Pree 1997). The resistance development to spinosad is particularly alarming because this is one of the few insecticides that has been shown to successfully control western flower thrips (Eger et al. 1998).

Opportunistic species, like thrips, have traits such as: short life spans, fast generation turnovers, and swift colonization ability, which may enable them to develop resistance more quickly than non-opportunistic species. It has also been shown that at least some thrips females will mate with their own male offspring. This effectively bottle-necks the gene pool and allows for amplification of resistance genes (Mound and Teulon 1995). These and other behavioral habits along with inherent biological characteristics have and will continue to allow thrips to

survive and prosper as crop pests. In order to effectively combat these tiny, but difficult insects we must continue to evaluate new insecticide chemistries that will provide control, and develop management strategies that will adequately compete with their opportunistic lifestyles and ability to quickly develop resistance.

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Chapter Two

Monitoring of thrips (Thysanoptera: Thripidae) using yellow sticky cards: species diversity and population dynamics in southeast Virginia.

Abstract

Thrips species diversity and abundance were monitored throughout 2010 and 2011 in Southeast Virginia. Adults were trapped using yellow sticky cards placed in and on the perimeters of peanut and cotton fields. Traps were replaced weekly from May 2010 to December 2011. The total number of adult thrips on each card was counted and recorded and a subsample of the adults was identified to species. Results showed that 11 species of thrips were trapped; thrips from the suborder *Tubulifera* and thrips in the genus *Haplothrips* were also encountered but not identified to species. Thrips populations began to increase in March and April, generally peaked by August, and then declined. There were differences in the magnitudes of thrips numbers between years with 2010 having higher numbers than 2011. Individual species abundance also varied between years with some species being more abundant in 2010 and others more abundant in 2011. The months of the peak trap capture for the top five most commonly encountered species indicated that each has a unique peak flight period and for some species it varied between years. Information from this study could be useful in improving thrips management decisions for local growers.

Throughout the United States thrips are considered to be economically important because multiple species are crop pests causing damage through feeding, oviposition, and virus transmission (Childers and Achor 1995, Childers 1997, Mound 1997, Osekre et al. 2009). In Southeastern Virginia thrips are well known pests of cotton, peanut, tomatoes, and multiple other crops (Herbert et al. 2011). Thrips feeding on cotton causes a delay in maturity, which due to the shortened growing season in Virginia, means these plants may never reach their full yield potential (Herbert 1998, Faircloth et al. 2002). In peanut and tomato crops, thrips transmit tomato spotted wilt virus (TSWV) (*Bunyaviridae Tospovirus*) (Kritzman et al. 2002), which can

limit yield potential and in many cases lead to plant death (Womack et al. 1981, Drake et al. 2009).

It is important to know what species of thrips are present in a given area or cropping system because of the differences in insecticide susceptibility and injury causing potential among different species (Mound 2001). One common method for monitoring thrips and other small insects is through the use of yellow sticky traps (Lewis 1973, Boykin et al. 1984, Nault et al. 2003). Sticky cards are useful for catching adult thrips and can track population movements into and out of fields and wild host plants. Typically, traps are placed in the area of interest (e.g., a greenhouse or field edge), the yellow color may act as an attractant or the adult insects may passively fly or be blown on the card via wind transportation. Researchers are then able to identify and monitor the species and abundance of local thrips populations.

The objective of this study was to monitor adult thrips populations in and around peanut and cotton fields in Suffolk, Virginia using yellow sticky trap cards to determine species composition and to gain insight into temporal changes in population size throughout the year.

Materials and Methods

Beginning on 4 May, 2010 and ending 12 December, 2011, yellow sticky trap cards (7.62 x 12.7 cm) (Olson products, Inc., Medina, Ohio) were placed in each of two field locations, Field 15 and Field 36, at the Virginia Tech Tidewater Agricultural Research and Extension Center (TAREC) in Suffolk, Virginia. In 2010, Field 15 contained peanuts and Field 36 contained cotton, In 2011 the crops were switched between these two fields. Cards were replaced weekly in each field. During the spring and summer seasons of 2010 and 2011, two sets of five cards were placed in both fields, but beginning in October 2011 and continuing until the study was concluded in December 2011 only one set of five cards was placed weekly in Field 15. Within each field, the five cards were placed in an "X" pattern; one card was located at the end of each line, about 1 meter into the field, and one at the intersection.

Each card was affixed to the top of a small metal pole about 7.62 cm above the ground. During times of high thrips abundance (spring and summer months) cards were left in the field for 48 hours; during times of low thrips abundance (fall and winter months) cards were left out

for 168 hours (7 days). For consistency, all data are presented based on a 48 hour sampling period. In cases where cards were left in the field for longer than 48 hours a simple mathematical formula was used to rescale the data [(# of thrips/hours in field)*48h].

The total number of adult thrips was tallied for each card, and when this number was greater than 30, a random subsample of 30 was identified to species using a stereo dissecting microscope (90x magnification); if the total number of adult thrips did not exceed 30 then all adults were identified to species. Data were analyzed to compare the mean number of thrips trapped between the two years, the mean number of thrips trapped monthly, and the differences in species trapped monthly using ANOVA and least significant means differences (LSM) Student's t-test statistical procedures, $\alpha = 0.05$. In the ANOVA, month was considered a random effect. Statistical analyses were performed using JMP™ software, version 8 (SAS Institute Inc., Cary, NC, 1989-2007).

Results

A total of 39,364 thrips were trapped and 15,404 of those were identified to species. Eleven different species were identified; the most commonly encountered species were: *Frankliniella fusca* (Hinds), *F. tritici* (Fitch), *F. occidentalis* (Pergande), *Thrips tabaci* (Lindeman), *Neohydatothrips variabilis* (Beach), and *Chirothrips texanus* (Andre). Other thrips species identified included *Limothrips cerealium* (Haliday), *Caliothrips fasciapennis* (Pergande), *Scolothrips sexmaculatus* (Pergande), *Plesiothrips perplexus* (Beach), and *Aeolothrips bicolor* (Hinds). In addition, multiple specimens from the suborder *Tubulifera* were encountered, but not identified to species; and at least one specimen from the genus *Haplothrips* was believed to have been found, but further identification is needed.

Across both years, based on the mean thrips identified from all cards (405 total cards), *F. fusca* was the most common species ($P = \leq 0.0001$, $F = 81.07$, df= 4, $\bar{x} = 6.16$) and comprised the greatest proportion of adult thrips (40.13%). *F. tritici* was second most common ($\bar{x} = 4.60$) and accounted for 28.02% of adult trips identified. *N. variabilis*, *F. occidentalis*, and *T. tabaci* were also frequently encountered ($\bar{x} = 2.37$, $\bar{x} = 1.59$, and $\bar{x} = 0.99$, respectively) and accounted for 18.03, 6.38, and 6.67 percent of identified adult thrips, respectively.

No significant difference was found between the mean number of thrips trapped at both field locations between 2010 and 2011. In these analyses, only the months of May through December were analyzed, because they were sampled in both years whereas the months of January through April were not sampled in 2010.

There were significant differences in the number of adult thrips trapped monthly (P = ≤0.0001, F = 30.55, df = 7). From the LSM Student's t-test, August and September were found to have the highest mean (± SE) catches of adult thrips ($\bar{x}$ = 104.59 ± 7.1, $\bar{x}$ = 107.35 ± 7.9, respectively) while May and July had the second highest adult thrips catches (Table 2.1).

There were significant differences in the mean (± SE) number of individuals trapped monthly from a particular species (Table 2.2). Peak catches for *F. fusca* adults occurred in the month of July ($\bar{x}$ = 16.93 ± 0.8) (P = ≤0.0001, F = 53.90, df = 7). *Thrips tabaci* adult catches peaked in May ($\bar{x}$ = 4.23 ± 0.3) (P = ≤0.0001, F = 21.04, df = 7); while the peak capture of *F. occidentalis* occurred in June ($\bar{x}$ = 5.68 ± 0.5) (P = ≤0.0001, F = 14.19, df = 7). The capture of *F. tritici* adults was equally abundant in both August ($\bar{x}$ = 11.20 ± 0.5) and September ($\bar{x}$ = 11.28 ± 0.6) (P = ≤0.0001, F = 67.67, df = 7). Finally, *N. variabilis* adults were trapped in the greatest numbers in May ($\bar{x}$ = 6.35 ± 0.6) and again in September ($\bar{x}$ = 6.65 ± 0.7) (P = ≤0.0001, F = 15.85, df = 7).

Discussion

Thrips appear to be a constant early season agricultural pest in Southeast Virginia. The more we learn about these insects, the better our chances of successfully mitigating their damage to important crops. The data presented here will help researchers and farmers in Virginia to better understand native thrips populations. Results show that there are several species in this pest complex and although not all species are considered crop pests, many are. For example, *F. fusca* and *F. occidentalis* are major early season pests of cotton and peanut, *T. tabaci* are pests of vegetable crops and *N. variabilis* are pests of soybean and some vegetable crops in southeastern Virginia. Knowing the species present is a key step in determining what control measure or specific insecticide a grower or researcher should implement. For example, the most common species recorded, *F. fusca*, is susceptible to most insecticides used for its control; however, *F. occidentalis* is known to be less susceptible to many insecticides and is also

known to develop resistance quickly (Immaraju et al. 1992, Eger et al. 1998, Bielza 2008). From this research we now know that both species are present in Southeast Virginia and we have a better understanding of the peak flight times for each species. Peak flight numbers may be related to adult movement between host plants or (although less likely) adult mating flights; more research is needed to further elucidate that relationship. Regardless of the reason for the mass flight, knowledge of when it may occur for a given species can aid in determining which species are present and possibly causing crop damage. It must be stressed, however, that sticky card sampling and knowledge of peak flights should not replace direct sampling of the crop of interest when investigating direct pest injury. Instead this knowledge should act as supplemental information. Directly sampling the crop of interest is the only sure way to know what thrips species are present and causing damage. This is due to the fact that sticky cards will capture any adult thrips that are flying around or being blown by the wind. It does not necessarily mean those thrips species are feeding in a particular crop. However, in the event that an immediate decision is needed and direct plant sampling cannot be performed, knowledge of the thrips species composition and population movement will help ensure a more accurate deduction. Future research is needed to help understand the motivation behind peak flights and also what the variability in a species timing of that flight between years is related to. Also, monitoring for thrips populations under different vegetative environments than the ones used in this study may reveal even more native thrips species present in Virginia, again aiding in our overall knowledge and understanding of these insects.

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Table 2.1. Mean (± SE) total thrips trapped monthly in 2010 and 2011, Field 15 and Field 36, Virginia Tech TAREC, Suffolk, Virginia.

Month	Mean Thrips (± SE)
May	69.6 ± 7.0 bc
June	58.4 ± 7.9 c
July	82.3 ± 7.9 b
August	104.6 ± 7.1 a
September	107.4 ± 7.9 a
October	8.4 ± 7.8 d
November	1.5 ± 8.3 d
December	0.2 ± 10.2 d

Means within a column followed by the same letter(s) are not significantly different (P=0.05).

Table 2.2. Mean (± SE) monthly thrips species trapped per five sticky cards in peanuts and cotton in Suffolk, VA in 2010 and 2011.

Month	*Frankliniella fusca*	*Frankliniella tritici*	*Neohydatothrips variabilis*	*Frankliniella occidentalis*	*Thrips tabaci*
May	8.7 ± 0.7 c	2.4 ± 0.5 c	6.4 ± 0.6 a	1.8 ± 0.4 c	4.2 ± 0.3 a
June	9.7 ± 0.8 bc	7.7 ± 0.6 b	2.5 ± 0.7 bc	5.7 ± 0.5 a	2.1 ± 0.3 b
July	16.9 ± 0.8 a	7.9 ± 0.6 b	1.5 ± 0.7 cd	1.5 ± 0.5 cd	0.8 ± 0.3 c
August	10.7 ± 0.7 b	11.2 ± 0.5 a	3.8 ± 0.6 b	3.1 ± 0.4 b	0.9 ± 0.3 c
September	9.4 ± 0.8 bc	11.3 ± 0.6 a	6.7 ± 0.7 a	2.4 ± 0.5 bc	0.6 ± 0.3 c
October	0.6 ± 0.8 d	1.4 ± 0.6 cd	0.6 ± 0.7 d	0.3 ± 0.5 d	0.0 ± 0.3 c
November	0.1 ± 0.8 d	0.6 ± 0.6 d	0.1 ± 0.7 d	0.1 ± 0.5 d	0.0 ± 0.4 c
December	0.0 ± 1.0 d	0.0 ± 0.7 d	0.0 ± 0.9 d	0.0 ± 0.6 d	0.0 ± 0.4 c

Means within a column followed by the same letter(s) are not significantly different (P=0.05).

Chapter Three

Within plant distribution and species complex of thrips (Thysanoptera: Thripidae) on soybean seedlings in Virginia.

Abstract

Several thrips species are known to feed on soybean and can cause yield reductions if injury to seedlings is exacerbated by other environmental factors. There is little information available about where thrips are located on soybean seedlings, and whether specific plant parts can be used to accurately estimate whole plant populations. A study was conducted in seedling soybean to evaluate the within-plant location of thrips, whether a subsample could be used for thrips monitoring, and to determine the thrips species complex present. Four soybean seedlings randomly selected from eight locations per field. The soybeans were sectioned into three parts. Thrips larvae and adults were counted on the Terminal; the Trifoliate; and the Remainder of each plant; in addition, the plant material was analyzed to determine the leaf area.

The most abundant species of thrips found were soybean thrips, *Neohydatothrips variabilis* (Beach). Larval density was highest in the Terminal section, while the highest total numbers occurred on the Remainder of the plant. There was no significant relationship between leaf area and number of larvae in each section. The number of larvae on the Remainder of the plant had the strongest relationship to whole plant counts ($R^2 = 0.88$) followed by the Trifoliate section ($R^2 = 0.54$). There was no significant relationship between counts of thrips from the Terminal section and the whole plant counts ($R^2 = 0.06$).

Soybean is the leading value export commodity for the United States and the second largest crop in cash sales (Ash et al. 2006). The United States also accounts for more than 50% of the world's soybean production (USEPA 2009), with 30.9 million hectares planted in 2009 (NASS 2010a, 2010b). Increased production of soybean, however, is in part hampered by the effects of multiple insect pests including various thrips species that colonize seedlings early in

the season (Kumar et al. 1998). Under high thrips densities and certain conditions such as when plants are stressed by drought or poor canopy development, thrips feeding can cause significant damage and stunted growth (D.A.H., Unpublished data).

In the southern and central United States where much of the soybean is planted earlier than in other regions, the crop is at higher risk to damage and yield reductions from infestations of thrips and other early season insect pests. Many growers in those regions rely on insecticide seed treatments to help mitigate this damage (Stewart 2008a, 2008b). A strong integrated pest management approach should first begin with knowledge of the pest complex present and their distribution in the plant to help determine the proper treatment methods. However, methods for monitoring and sampling thrips in soybean are poorly developed; most of the research related to this topic was published more than 30 years ago.

There are multiple methods that can be used to monitor thrips in seedling soybean, subsampling is one such method. Subsampling saves the observer time and money during sampling, and is a less destructive assessment method than whole plant sampling. Along with the ability to reduce plant growth and yield, other important reasons for monitoring thrips populations in seedling soybean include their status as vectors of at least one virus in soybean, tobacco ringspot virus (*Comoviridae: Nepovirus*), which can cause lowered seed quality and yield loss; as a major portion of the fauna in soybean fields; and as attractive prey for certain predatory species of insects, which are important for natural control of subsequent infestations of other soybean pests (Irwin and Yeargan 1980). The purpose of this research was to determine the thrips species present on soybean in Virginia, the within plant distribution of immature thrips on seedling soybean, and whether subsamples of plant material are adequate for monitoring within-plant populations of thrips.

Materials and Methods

This two year study was conducted in Suffolk, Virginia at the Virginia Tech Tidewater Agricultural Research and Extension Center (TAREC). NK brand cultivar S56-L5 (Syngenta, Wilmington, DE) and Asgrow brand cultivar 5605RR (Monsanto, St. Louis, MO) soybean were planted on 5 May, 2010 and AG5531 (Asgrow®, Monsanto, St. Louis, MO) soybean was planted

on 9 May, 2011 using a 91.4 cm row spacing. Plant and thrips samples were taken on 21 June 2010 and 7 June 2011, when plants were in the V3-V4 growth stage (3–4 fully expanded trifoliate leaves) (McWilliams et al. 1999). This growth stage was selected because previous experience has shown that this is a good time to compare efficacy of different thrips control practices (e.g., insecticide seed treatments) as thrips injury to seedlings is fully apparent, and thrips populations are abundant. All sampling occurred between 0700 and 0730 h. EST when thrips adults and larvae were observed to be less active (Aliakbarpour and Rawi 2010).

A stratified random sampling method was used in which eight non-insecticide-treated 12.2 m long rows were partitioned into four 3.1 m strata. One soybean plant was randomly selected from each 3.1 m stratum and then systematically divided into three sections: the plant terminal, which we defined as the uppermost non-expanded trifoliate leaf (Terminal); the uppermost fully expanded trifoliate leaf (Trifoliate); and the remainder (Remainder) of the plant (consisting of two to three trifoliate leaves, the unifoliate leaves, and cotyledons), which was cut at the base just above the soil surface. Each plant section was placed immediately into a separate 950 ml glass jar containing 450 ml of a mildly soapy water solution. Thrips larvae and adults were washed from the leaf material by swirling the material vigorously for 30 seconds (Aliakbarpour and Rawi 2010); vacuum filtration was then used to remove the water, the number of immature and adult thrips was recorded and adult thrips were identification to species. After thrips were removed, the leaf material from each plant section was immediately analyzed to determine the leaf area using a desk-top Li-Cor 3100 (LI-COR Environmental, Lincoln NE) leaf area meter calibrated with a 50-cm metal disk. Non-expanded leaves from the terminal of the plant were unfolded and laid flat on the machine to allow for more accurate determination of leaf area.

For greater insight into the species complex, adult thrips were also collected over time from non-insecticide treated soybean at two field locations (TAREC) in 2010 and 2011. Unlike the method described above, the seedlings were not sectioned. Ten whole plants per replicate were randomly selected and placed into jar containing a mild soapy water solution. Extraction and identification of adult thrips was performed as described previously.

Statistical analyses were performed using either SAS™ software, version 9.2 of the SAS System for Microsoft Windows 7® copyright © 2010, SAS Institute Inc., or JMP™ software, version 8, SAS Institute Inc., Cary, NC, 1989-2007. Proportionally, there were much higher numbers of larvae compared with adults, so only larval counts were used for with-in plant distribution analysis.

A mixed model ANOVA, followed by pairwise comparisons of least square means, was used to test for differences between larval counts per plant section and density of larvae per plant section. In the ANOVA, plant section was considered fixed, and block, block*plant section, rows within plots and samples within rows were all considered random factors.

Regression analyses based on the mean counts for each replicate were used to determine the strength of dependence between counts of larvae in each plant section with the whole plant count (sum of the three plant section samples), and whether the relationship was linear or quadratic. Our method followed from previous work in which the means of count data rather than individual counts were used and found to provide better correlations (Rudd and Jensen 1977, Luna et al. 1982, Browde et al. 1992, Zalom et al. 1993, Knott et al. 2006). Whole plant counts (Y variable) were reciprocally transformed for this analysis. Although reciprocal transformations are more commonly used to transform data for ANOVA, they can also be used to transform data for regressions (Zar 1984); In addition, some authors have cautioned against using the more common log transformations on count data (O'Hara and Kotze 2010).

Regressions were calculated separately for each plant section to determine the relationship between leaf area and numbers of thrips larvae. This allowed us to assess whether the number of thrips larvae present on a plant section was influenced by the amount of leaf area. In addition, a two-way t-test was used to test for differences in leaf area between years. All analyses were performed using an $\alpha = 0.05$.

Results

Thrips species identification from a one-time sample was determined based on 71 adults collected in 2010 and 137 adults in 2011. In 2010, soybean thrips (*Neohydatothrips variabilis,* Beach) made up more than 50% of the species followed by flower thrips (*Frankliniella*

tritici, Fitch), which comprised almost 24% (Table 3.1). In 2011, soybean thrips were more abundant making up 70% of the species complex, followed by onion thrips (*Thrips tabaci*, Lindeman) at 23% (Table 3.1). These percentages were consistent with species identified overtime in other thrips samples taken from concurrent, but non-related soybean studies at TAREC (Table 3.2). Flower thrips were more abundant in 2010 than 2011 and onion thrips were more abundant in 2011 than 2010. It is also worth noting that based on samples over time from these non-related studies, tobacco thrips (*F. fusca*) comprised a major portion of soybean fauna early, but over the course of a few weeks their numbers declined and the number of soybean thrips increased (Table 3.2). Larval thrips are extremely difficult if not impossible to accurately identify to species. Previous attempts to rear field collected larval thrips to species were unsuccessful due to high mortality. Therefore, an assumption was made that the larval thrips species collected during the study are consistent with the adult thrips which were positively identified and presented in Table 3.1.

From the mixed model ANOVA, followed by pairwise comparisons of least square means analysis (adjusted for density) it was determined that there was a significant difference in the number of larval thrips among soybean seedling sections in both years (Table 3.3). The Terminal had the highest density (count/leaf area) of thrips larvae (F = 37.50; df = 6; P = 0.0004) while the Remainder of the plant had the highest absolute count when density was not a factor (F = 53.87; df = 6; P = 0.0001) (Table 3.3).

The strongest regression relationships between thrips counts on plant sections and whole plant, based on significant P-values and R^2-values, were quadratic. There was no significant correlation between Terminal larval counts and whole plant larval counts (R^2 = 0.20, P = 0.23) (Figure 3.1). However, Trifoliate counts had a significant relationship to whole plant counts (R^2 = 0.54, P = 0.01) (Figure 3.2).

The regressions between the number of larvae and leaf area were extremely weak for all plant sections (Terminal: R^2 = 0.07, P = 0.04; Trifoliate: R^2 = 0.02, P = 0.32); therefore, we elected not to adjust larval counts for leaf area differences.

Using a two-tailed t-test, we determined that there was no difference between years in leaf area in the Trifoliate plant section (t = 1.49, df = 14, P = 0.16). The mean leaf area for the

Trifoliate plant section was 40.69 cm^2 . There was a difference between years in leaf area in the Terminal plant sections. In 2010 the leaf area of the Terminal sections was 5.29 cm^2 and in 2011 it was 3.05 cm^2 (t = -6.15, df = 14, P = ≤0.0001).

Discussion

At the V3-V4 growth stage, soybean thrips are the most abundant species colonizing Virginia soybean. In most cases this trend continues as the plant matures. However, as seen from the data presented in Table 2, in the cotyledon (VC) and V1 growth stages tobacco thrips are most abundant. Tobacco thrips can therefore be regarded as the initial colonizers of soybean in Virginia, but they quickly migrate out of the crop as the plants mature. Insecticide seed treatments and/or foliar sprays used by growers to control thrips in the early growth stages should be selected to have efficacy against tobacco thrips, soybean thrips, or a combination of the two.

The smallest sample area (the terminal leaflets) had the highest density of larvae suggesting that immature thrips have an aggregated distribution. Irwin et al. (1979) found similar evidence for an aggregated distribution of thrips on soybean plants in Illinois, Missouri, and Kentucky. They also discerned that *F. tritici* and *Neohydatothrips (Sericothrips) variabilis* (Beach) were the two most abundant species on soybean in the three aforementioned states; both species were unevenly distributed on the plant especially during the vegetative growth stages. *S. variabilis* adults were most abundant on the upper leaves while the larvae appeared most commonly on different trifoliates depending on the instar. *Frankliniella tritici* adults and larvae were most abundant in the terminal buds. Irwin et al. (1979) also found that there were no differences in total number among plants. Other studies have also found evidence that thrips generally occur in aggregated groups both within and among plants (Salguero Navas et al. 1994, Cho et al. 1995, Joost and Riley 2004).

In 2011 there was significantly less leaf area in the terminal sections than in 2010. This difference could have been due to differences in growing conditions, or differences in the varieties between years. We found that a quadratic model and not a linear model best fit our data. In a similar study describing sampling patterns for *Dicyphus hesperus* Knight (Heteroptera:

Miridae) on greenhouse tomatoes, Sanchez et al. (2002) also found that a quadratic model provided the best fit to their data. Based on the weak regressions between the number of larvae and leaf area, and the president set in similar studies examining sampling, spatial distribution, and population distribution of Homopteran and Hemipteran pests of cotton, we did not adjust for leaf area differences (Suh and Westbrook 2010, Naranjo and Flint 1995).

Results of our study showed that the number of thrips on the uppermost fully opened trifoliate are related to the total number per seedling by a strong correlation value (0.74). This suggests that a subsample consisting of the first fully opened trifoliate can be used to monitor thrips in V3-V4 stage soybean. Subsampling may be useful in studies where thrips population estimates are needed, or when monitoring in areas where thrips infestations and plant injury are more closely related to yield reductions in soybean (e.g., early planted soybean in the mid-south; Stewart 2008a, 2008b).

We should also stress that due to the aggregated distribution of thrips larvae, when sampling in soybean one should be careful not to overestimate the thrips population by sampling only the areas of highest density (the terminals) or underestimate the population by sampling in a lower density area only (the lower trifoliates). Also as with any sampling protocol, care should be taken to avoid dislodging larval thrips or allowing adults to escape during sampling. Our sampling recommendations are partially contradictory to the sampling techniques proposed by Irwin et al. (1979). In their study they recommend that the sampling unit be either a fully opened trifoliate or terminal bud depending on the thrips species of interest. While this might be useful for species specific monitoring, we caution against sampling the terminal bud at this time due to inaccurate population estimation as presented above. At this time, we do not know whether damage from thrips feeding is equally detrimental on all parts of the plant or if feeding on a specific section (e.g. the terminal) has a greater impact on the plant. One must take in to account the specific objectives of a study to determine the relevance of any sampling recommendations. More work is needed to improve sampling techniques for thrips in soybean and other sampling methods need to be investigated. Boll et al. (2007) and Palumbo (2003) proposed other thrips sampling techniques that they found to be adequate for monitoring infestations on cucumber, rose, and lettuce crops. In both studies they

investigated the accuracy of visually counting thrips on the plant. Palumbo (2003) also evaluated the efficacy of beat pans, sticky traps, and plant washes. These alternative sampling techniques may prove to be beneficial in soybean as well, but further research is needed.

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Table 3.1. Percentage of thrips by species collected in a onetime sample from non-insecticide treated V3-V4 stage soybean seedlings at TAREC, Suffolk, VA. Samples consisted of four whole plants per replicate.

Year	Date	% *N. variabilis*	% *F. tritici*	% *F. fusca*	% *T. tabaci*	% *F. occidentalis*	n
2010	21 June	57.8	23.9	9.9	7.0	1.4	71
2011	7 June	70.1	0	5.1	22.6	2.2	137

Full species names: Neohydatothrips variabilis, Frankliniella tritici, Frankliniella fusca, Thrips tabaci, Frankliniella occidentalis.

Table 3.2. The percentage thrips by species collected over time from non-insecticide treated soybean seedlings at two field locations. TAREC, Suffolk, VA. Samples consisted of 10 whole plants per replicate.

Year and Location	Date	Growth Stage	% *N. variabilis*	% *F. tritici*	% *F. fusca*	% *T. tabaci*	% *F. occidentalis*	n
2010								
Location 1	7 June	V1	31.4	2.9	65.7	0	0	35
	14 June	V2	68	4	24	4	0	25
	21 June	V3-V4	71.4	0	14.3	14.3	0	7
	29 June	V6-R1	85.7	14.29	0	0	0	7
Location 2	7 June	V1	9.4	15.6	59.4	12.5	3.1	32
	14 June	V2	88	0	8	4	0	25
	21 June	V3-V4	66.7	25	0	8.3	0	12
	29 June	V6-R1	80	20	0	0	0	10
2011								
Location 3	17 May	VC	3.1	1.5	89.2	4.6	1.5	65
	26 May	V1	40.3	2.6	24.7	24.7	7.8	77
	3 June	V2	28.6	0	0	71.4	0	7
	10 June	V4	100	0	0	0	0	1
Location 4	17 May	VC	3.4	0	92.1	2.3	2.3	88
	26 May	V1	46.4	2.9	24.6	18.8	7.25	69
	3 June	V2	62.5	0	12.5	12.5	12.5	8
	10 June	V4	76.7	0	0	23.3	0	30

Full species names: Neohydatothrips variabilis, Frankliniella tritici, Frankliniella fusca, Thrips tabaci, Frankliniella occidentalis.

Table 3.3. Means comparison (± SE) by year for numbers of larval thrips per soybean plant section non-adjusted and adjusted for density (count/leaf area).

Sample Unit	Year	Mean Number of thrips per Sample Unit	Mean Number of Thrips per cm^2
Terminal	2010	3.05a ± 1.03	1.41a ± 0.30
Trifoliate		2.29a ± 0.66	0.37b ± 0.02
Remainder		4.52b ± 2.45	0.47b ± 0.03
Terminal	2011	1.41a ± 0.58	0.87a ± 0.18
Trifoliate		2.92b ± 1.11	0.46b ± 0.03
Remainder		5.26c ± 3.53	0.59b ± 0.04

Means within each column for the same year followed by the same letters are not significantly different (P>0.05).

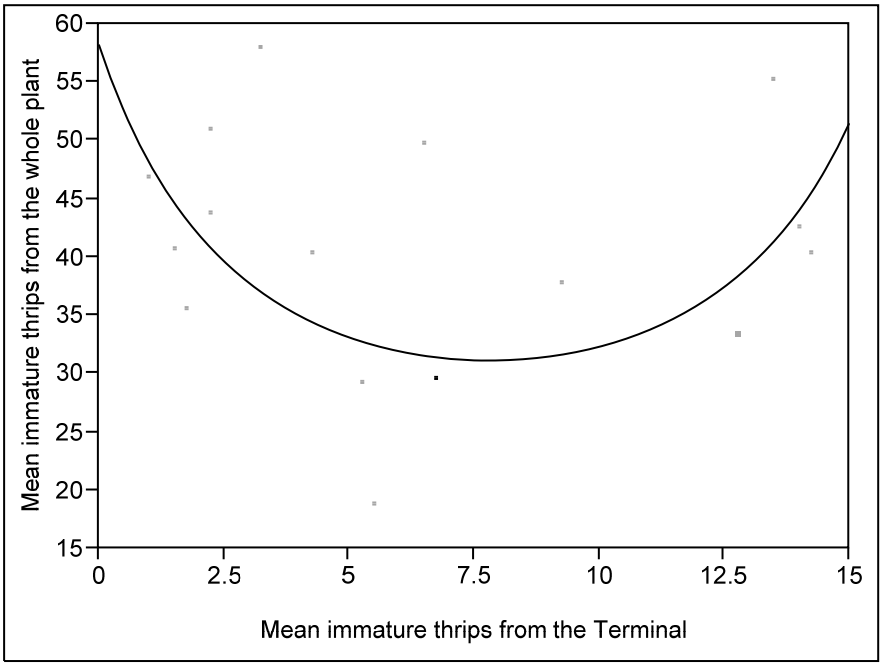

Figure 3.1. Regression between mean larval thrips counts from the Terminal of the plant and mean totals from the whole plant. Equation of the line:

$\frac{1}{Y} = 0.03 - (0.0006 * \bar{X}) + 0.0003 * (\bar{X} - 6.5)^2$**, where Y is the mean of the whole plant larval counts and X is the mean of the Terminal larval counts;** R^2 **= 0.20;** ***P*****= 0.23.**

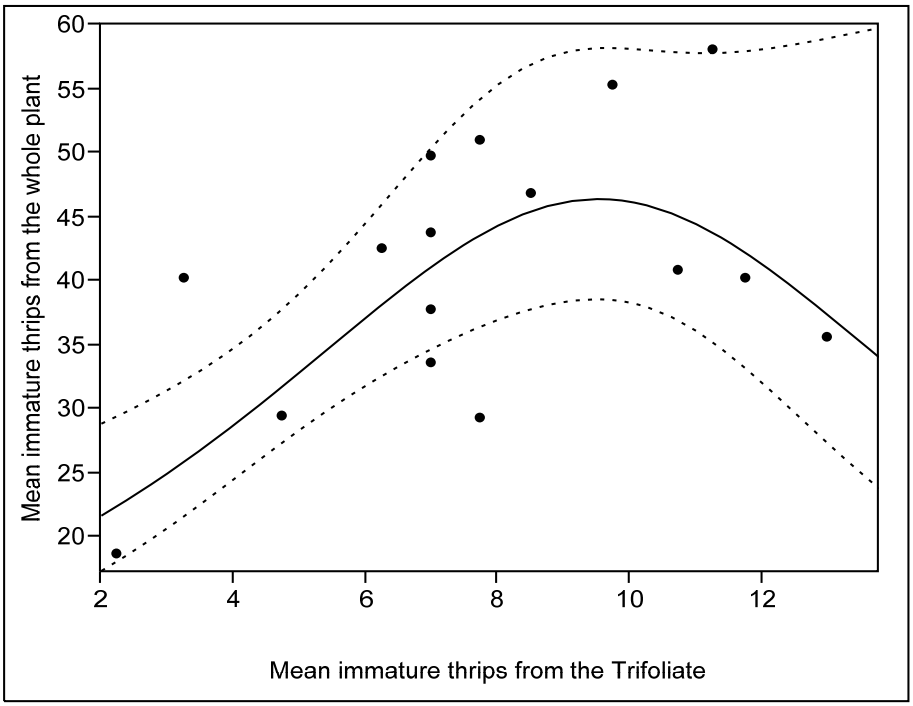

Figure 3.2. Regression between mean larval thrips counts from the Trifoliate of the plant and mean totals from the whole plant. Equation of the line:

$\frac{1}{y} = 0.03 - (0.002 * X) + 0.0004 * (X - 7.81)^2$, **where Y is the mean of the whole plant larval counts and X is the mean of the Trifoliate larval counts;** $R^2 = 0.54$; $P = 0.0063$.

Chapter Four

Field efficacy of cyantraniliprole, a novel diamide insecticide, against thrips (Thysanoptera: Thripidae) on cotton and peanut seedlings.

Abstract

Tobacco thrips, *Frankliniella fusca*, as well as a complex of other thrips species attack cotton and peanut seedlings and can cause significant yield loss to these crops in the mid-Atlantic U.S. Experiments were conducted in Suffolk, Virginia in 2010 and 2011 to assess the efficacy of a novel diamide insecticide, cyantraniliprole, applied to these two crops as a liquid in-furrow or broadcast spray treatment for thrips control. In both cropping systems cyantraniliprole significantly reduced the number of immature thrips and also reduced thrips feeding injury to the plants. In several instances the application of cyantraniliprole for thrips control resulted in yields that were higher than the untreated control and statistically similar to yields obtained with the standard thrips control insecticides of aldicarb 15G, acephate 97, and phorate 20G. These results indicate that cyantraniliprole is a promising new insecticide for thrips management in cotton and peanut crops.

Approximately 87 species of thrips are crop pests capable of causing damage and economic loss (Mound 1997). In Virginia and North Carolina, tobacco thrips, *F. fusca*, is one of the most abundant thrips pests on cotton, peanut, and a number of other crops (Groves et al. 2001, 2002; Nault et al. 2003; Herbert et al. 2007). In cotton (*Gossypium hirsutum* L.), thrips feeding can lead to excessive branching, delayed maturity, and reductions of fruit size and lint quality (Gaines 1934, Dunnam and Clark 1937, Watts 1937, Lei and Wilson 2004, Greenberg et al. 2009). In peanut (*Arachis hypogaea* L.) thrips feeding may adversely affect plant growth including malformation of leaves, reduction in leaf area, delayed maturity, and lowered pod yield (Womack et al. 1981, Herbert et al. 2007, Drake et al. 2009). Thrips cause injury through feeding and oviposition and in the case of certain species through disease and virus transmission (e.g., *Fusarium* hardlock and *Bunyaviridae Tospovirus*) (Childers 1997, Childers and

Achor 1995, Osekre et al. 2009). There is some debate about the necessity of thrips control in cotton with multiple studies citing the plants ability to recover from initial stunting later in the season (Sadras and Wilson 1998, Lei and Wilson 2004). However, other authors disagree (Herbert 1998, Faircloth et al. 2002), and in Virginia, yield losses of up to 50% in cotton and 30% in peanut from thrips injury have been documented (Herbert et al. 2007).

There are multiple management techniques that can help mitigate thrips damage to plants. Proper removal of alternative or overwintering host plants, which sustain populations of multiple thrips species, may help lessen spring migration into cropping systems (Northfield et al. 2008). The adoption of winter cover crops and strip tillage techniques has been shown to lessen thrips populations and subsequent damage (Olson et al. 2006, Toews et al. 2010). Nonetheless, chemical control using broad spectrum insecticides such as aldicarb and acephate, is the most commonly used and recommended management approach (Greenberg et al. 2009, Herbert et al. 2011); there are concerns, however, regarding the toxicity of these insecticides to beneficial species, the development of resistance, and in some cases, the lack of efficacy (Zhao et al. 1995, Allen et al. 2005, Duso et al. 2008, Herron et al. 2008). These concerns have led to the development of new insecticides which have different modes of action, greater host specificity, and less toxicity to beneficially important species (Isayama et al. 2005, Bruck et al. 2009, Cameron et al. 2009).

Cyantraniliprole (DuPont Crop Protection, Wilmington, DE) is a novel insecticide that has been shown to reduce thrips feeding injury and tomato spotted wilt virus transmission (*Bunyaviridae Tospovirus*) by tobacco thrips, *Frankliniella fusca* (Jacobson and Kennedy 2011). Cyantraniliprole is an anthranilic diamide insecticide that acts as an agonist targeting ryanodine receptors in insects affecting calcium release during muscle contraction (IRAC Group 28) (Cordova et al. 2006, Sattelle et al. 2008, IRAC 2011). Insects treated with cyantraniliprole exhibit rapid feeding cessation, muscle paralysis, and ultimately death (Hannig et al. 2009). Chlorantraniliprole, a similar chemical to cyantraniliprole, has demonstrated tremendous efficacy against a variety of lepidopteran pests, whiteflies, and beetles. It has demonstrated excellent efficacy against Colorado potato beetle in the field (Kuhar and Doughty 2009, 2010; Sewall and Alyokhin 2009). Cyantraniliprole, like chlorantraniliprole, is also xylem-mobile for

root uptake providing systemic control of insect pests. The objective of this research was to evaluate the effectiveness of cyantraniliprole for controlling thrips in cotton and peanut, and the effects on plant injury and crop yields.

Materials and Methods

Cotton

A two-year study was conducted in Suffolk, Virginia at the Virginia Tech Tidewater Agricultural Research and Extension Center (TAREC). In 2010, 'DP 0920B2RF' cotton was planted 5 May, and in 2011, 'PHY 499 WRF' cotton was planted 2 May, using 91.4 cm row spacing. Experimental treatments consisted of various combinations of in-furrow (IF) applications of cyantraniliprole 20SC and aldicarb 15G (Bayer Crop Science, Research Triangle Park, NC), and foliar applications of cyantraniliprole 10OD and acephate 97 (Amvac, Los Angeles, CA) (Table 4.1). In-furrow applications of cyantraniliprole 20SC were made diluted in water into the seed furrow at 46.75 L/ha using a microtube mounted between the disc openers, pressurized by CO_2 at 351.63 kPh. Aldicarb 15G was applied into the seed furrow using calibrated, tractor-mounted inverted jars. Foliar applications of cyantraniliprole 10OD and acephate 97 were broadcast at first true leaf stage (BC @ 1st tl) on 20 May with a CO_2-pressurized backpack sprayer at 133.7 L/ha and 115.83 kPa through 8002VS nozzles spaced 45.72 cm apart on the spray boom.

A randomized complete block experimental design with 4 replicates was used; plots were 4 rows by 12.19 meters in length. Thrips injury to cotton plants was determined by visually rating injury using a 0 to 5 scale, where 0=no thrips induced plant injury, 1 = 10% leaves showing injury, no bud injury; 2 = 25% injured leaves, no bud injury; 3 = 75% injured leaves, and 0 to 25% buds injured; 4 = 90% injured leaves, and greater than 25% buds injured; and 5 = dead plants (Herbert and Malone 1999 b). Two raters individually examined each treatment plot. The final rating for the plot consisted of the average of the two independent ratings. Thrips were counted on four sample dates by collecting five plants per plot and placing them in Mason jars containing 500 ml soapy water. Following vacuum filtration into a Büchner funnel, thrips

were counted and identified under a stereoscope. Yield was determined by harvesting the 2 center rows of each plot (21.34 row meters) using a commercial 2-row cotton picker. Plots were harvested on 12 October, 2010 and 21 October, 2011. Sub-samples (454 g) were ginned to determine lint versus seed and trash weight. Gross yields were reduced by 57.9% to account for seed and trash. Immature thrips counts and yield data were analyzed using ANOVA and Fisher's Protected LSD statistical procedures, $\alpha = 0.05$. Thrips injury ratings were analyzed using Kruskal-Wallis and Wilcoxon Each Pairs tests, $\alpha = 0.05$. Statistical analyses were performed using either SAS™ software, version 9.2 of the SAS System for Microsoft Windows 7® copyright © 2010, SAS Institute Inc., or JMP™ software, version 8, SAS Institute Inc., Cary, NC, 1989-2007.

Peanut

'CHAMPS' peanut was planted 30 April 2010, and 'Phillips' peanut was planted 3 May 2011 in two separate field locations at TAREC, using 91.44 cm row spacing. The two separate field locations in 2011 are referred to as Peanut Trial 1 and Peanut Trial 2 for the remainder of this document. Liquid in-furrow (IF) insecticide (cyantraniliprole 20SC, acephate 97, and phorate (Amvac, Los Angeles, CA)), fungicide, and inoculant treatments were applied into the seed furrow at 46.75 L/ha using a microtube mounted between the disc openers, pressurized by CO_2 at 379.21 kPa. Aldicarb 15G was applied using the same method as in cotton. Foliar treatments (cyantraniliprole 10OD and acephate 97) were broadcast at late ground cracking on newly emerging seedlings (BC @ late GC) on 23 May and seven to ten days after late ground cracking (when seedlings were emerged and plants were about 15 cm in diameter) 27 May (Peanut Trial 1, treatment 10 only) using the same method as in cotton. A complete treatment and applied peanut rates list can be seen in Table 4.2. A randomized complete block experimental design was used with 4 replicates; plots were 4 rows by 12.19 meters in length. Thrips injury to plants was determined by visually rating injury using a 0 to 10 scale, where 0 = no injury, 1 = 10% leaves injured, 2 = 20% leaves injured, 3 = 30% leaves injured, 4 = 40% leaves injured, 5 = ≥ 50% leaves injured and ≤ 5% terminal buds injured, 6 = ≥ 50% leaves injured and 25% terminal buds injured, 7 = ≥ 50% leaves injured and 50% terminal buds injured, 8 = ≥ 50% leaves injured and 75% terminal buds injured, 9 = ≥ 50% leaves injured and 90% terminal buds

injured, and 10 = dead plants (Herbert and Malone 1999 a). The visual injury ratings in peanut are determined using the same procedure as described for cotton. Thrips counts from selected treatments were determined by collecting ten unopened terminal leaflets per plot in vials containing 30 ml soapy water, teasing the leaflets open, and counting thrips under a stereoscope following suction filtration. Adult thrips from selected treatments were identified to species using a stereoscope. Peanuts were dug on 23 September, 2010 and 6 October, 2011. Yields were determined by combining and weighing peanut pods and kernels from the 2 center rows of each plot on 7 October, 2010, 14 October 2011 (Peanut Trial 1), and 11 October 2011 (Peanut Trial 2) and adjusting to 7% moisture. Data were analyzed using the same procedures as described for cotton.

Results and Discussion

Cotton

In 2010, thrips species identification was determined on 620 adults. Tobacco thrips (*Frankliniella fusca,* Hinds) comprised 68% of the species collected, onion thrips (*Thrips tabaci*, Lindeman) 14%, flower thrips (*Frankliniella tritici*, Fitch) 15%, and soybean thrips (*Neohydatothrips variabilis,* Beach) comprised 3%. The best treatments (those treatments with the greatest numerical improvement from the control) had 4.5 to 156.1 fewer immature thrips per five plants than the non-treated control on four out of five sampling dates (24 May: F = 4.39, df = 9, P = 0.004; 31 May: F = 73.80, df = 9, P = ≤0.0001; 9 June: F = 6.23, df = 9, P = 0.0005; 16 June: F = 2.80, df = 9, P = ≤0.0001) (Table 4.3). Cyantraniliprole treatments consistently reduced immature thrips numbers comparable to the standard of aldicarb 15G, especially cyantraniliprole treatments three and four (0.20 kg ai/ ha IF and 0.15 kg ai/ha IF followed by 0.10 kg ai/ha BC). The best treatments also had thrips injury rating which were reduced by 2.69 and 4.33 points relative to the non-treated control (28 May: χ^2 = 22.94, df = 6, P = 0.0008; 1 June: χ^2 = 24.28, df = 6, P = 0.0005; 7 June: χ^2 = 25.88, df = 6, P = 0.0002; 17 June: χ^2 = 21.53, df = 6, P = 0.0015) (Table 4.4). All cyantraniliprole treatments resulted in reduced thrips injury compared with the non-treated control; however, the injury ratings were slightly higher than in the standard treatment. On average, the treatments that provided the best protection from

thrips resulted in higher yields of 1071.4 kg lint/ha (F = 21.53, df = 9, P = ≤0.0001) (Table 4.4). Cyantraniliprole treatments had yields that were statistically similar to the standard treatment of aldicarb 15G (IF) + acephate 97 (BC) and were higher compared with aldicarb (IF) alone.

In 2011, thrips species identification was determined on 744 adults. Tobacco thrips comprised 59% of the species collected, onion thrips 25%, western flower thrips (*Frankliniella occidentalis*, Pergande) 12%, soybean thrips 3%, and flower thrips 1%. The best preforming treatments reduced immature thrips numbers on two of four sampling dates (24 May: F = 8.03, df = 7, P = 0.001; 31 May: F = 4.72, df = 7, P = 0.0094). On average those reductions ranged from 58 to 61 fewer immature thrips per five plants compared with the non-treated control; cyantraniliprole treatments were statistically similar to the standard of acephate (IF) + acephate (BC) on both sampling dates (Table 4.5). The best treatments also resulted in reduced thrips injury between 2.87 and 3.81 points lower than the non-treated control based on the plant injury scale (24 May: χ^2 = 27.54, df = 7, P = 0.0003; 31 May: χ^2 = 29.65, df = 7, P = 0.0001; 8 June: χ^2 = 28.45, df = 7, P = 0.0002). There was no significant treatment effect on yield, but on average all treatments resulted in a modest yield increase of 145.9 kg lint/ha (Table 4.5).

Results from studies conducted in 2011 were not as clearly defined as those from 2010. While all treatments reduced immature thrips numbers and thrips injury to plants, there were no statistical differences in lint yields. Also, the reduction in the number of immature thrips and reduction in visible injury was not as great as in 2010. We believe these inconsistencies resulted from a lack of soil moisture at and the weeks following planting and harsh weather during the growing season and prior to harvest. Cyantraniliprole, like many insecticides applied as liquid in furrow treatments, needs adequate soil moisture to move down in the soil profile to the rooting zone of the growing seedling (E.I. Du Pont 2008, 2010). In 2011, we received just over 5.1 cm of precipitation in May; in 2010 we received almost 17.8 cm. The lack of precipitation at planting in 2011 was compounded by drought-like conditions during the majority of the growing season. Southeastern Virginia crops were also damaged by a hurricane on 27 August, followed by a delay of harvest due to an overabundance of precipitation. When considering the hardships faced by the cotton crop in 2011 it is impressive that the insecticide

treatments still reduced immature thrips numbers and injury to plant ratings; this could be viewed as a testament to the strength of these treatments when faced with excessive environmental conditions.

Cyantraniliprole has a novel mode of action, low toxicity to mammals and beneficial insects, longer residual activity against target pests, and increased plant mobility with respect to other diamide insecticides (Lahm et al. 2009, Hardke et al. 2011). These attributes combined with the positive performance of cyantraniliprole in 2010 and its ability to perform well under the harsh growing conditions of 2011, make it a promising new tool for the management and control of thrips in cotton.

Peanut

Thrips species identification was determined on 764 adults. Tobacco thrips compromised 94% of the species collected, flower thrips 4%, onion thrips 2%, western flower thrips 0.5%, and soybean thrips comprised 0.1%. In 2010, results in the peanut trials were similar to the 2010 cotton trials. All treatments were similar in their reduction of the number of immature thrips per ten peanut leaflets on two separate sample dates. Cyantraniliprole treatments were statistically similar to the standard treatment of aldicarb applied in furrow followed by a broadcast spray of acephate 97 at late ground crack. On average the number of immatures was reduced by either 11.03 or 58.44 immature thrips per ten leaflets compared to the non-treated control (25 May: $F = 3.30$, df = 10, $P = 0.0101$; 1 June: $F = 4.77$, df = 10, $P = 0.0013$) (Table 4.6). On all sample dates, the thrips injury was reduced by the best treatments between 1.98 and 7.12 points relative to the non-treated control. The treatment of cyantraniliprole 0.15 kg a.i./ha (IF) + cyantraniliprole 0.10 kg a.i./ha (BC) preformed as well as the standard treatment(s) (phorate IF, aldicarb If, and aldicarb IF plus acephate BC) on all rating dates (21 May: $\chi^2 = 25.46$, df = 7, $P = 0.0006$; 28 May: $\chi^2 = 26.27$, df = 7, $P = 0.0005$; 1 June: $\chi^2 = 27.63$, df = 7, $P = 0.0003$; 7 June: $\chi^2 = 25.92$, df = 7, $P = 0.0005$; 17 June: $\chi^2 = 28.66$, df = 7, $P = 0.0002$). There was no significant difference between pod yields among treatments, however, on average treated plots had 518.2 kg/ha more pods compared to the non-treated control (Table 4.7).

In 2011, Peanut Trial 1, thrips species identification was determined on 554 adults. Tobacco thrips comprised 70% of the species collected, onion thrips 17%, western flower thrips 13%, flower thrips 0.5%, and soybean thrips 0.2%. Peanut trials occurring in 2011 faced the same harsh growing conditions as our cotton trials. All treatments were statistically similar in their reduction of the number of immature thrips per ten peanut leaflets on two of five sample dates. On average the number of immatures was reduced in the treated plots between 5.0-14.4 immatures compared to the non-treated control (25 May: $F = 6.10$, df = 8, $P = 0.0014$; 3 June: $F = 3.81$, df = 8, $P = 0.0124$) (Table 4.8). On all sample dates, the plant injury was reduced by the best treatments between 2.81 and 5.75 points relative to the control (24 May: $\chi^2 = 39.08$, df = 10, $P = \leq 0.0001$; 31 May: $\chi^2 = 37.84$, df = 10, $P = \leq 0.0001$; 9 June: $\chi^2 = 38.97$, df = 10, $P = \leq 0.0001$; 15 June: $\chi^2 = 36.39$, df = 10, $P = \leq 0.0001$). A dual broadcast application of cyantraniliprole resulted in an identical plant injury rating as the non-treated control. The plants in that cyantraniliprole treatment received their first application on 23 May. Until that time they were identical to the non-treated control plots receiving the same thrips pressure without any protection from insecticides. Consequently the injury ratings for these two treatments on 24 May were similar. On subsequent rating dates the appearance of the plants in the dual broadcast only cyantraniliprole plot quickly improved and was reflected in the later ratings. Only treatment five, a high rate (0.84 kg ai/ha) of acephate 97 applied in furrow followed by broadcast spray (0.41 kg ai/ha) on 23 May, resulted in a statistically significant higher yield (1429.4 kg/ha) ($F = 3.57$, df = 13, $P = 0.0019$); however, all treatments resulted in numerically higher yields (Table 4.9).

In Peanut Trial 2, thrips species identification was determined on 289 adults. Tobacco thrips comprised 75% of the species collected, onion thrips 11%, western flower thrips 13%, and flower thrips 0.7%. There was no significant difference among treatments in the reduction of immature thrips. On all four sample dates, the plant injury rating was reduced by the best treatments between 2.25 and 5.81 points relative to the non-treated control (24 May: $\chi^2 = 40.19$, df = 11, $P = \leq 0.0001$; 31 May: $\chi^2 = 41.34$, df = 11, $P = \leq 0.0001$; 9 June: $\chi^2 = 42.13$, df = 11, $P = \leq 0.0001$; 15 June: $\chi^2 = 42.19$, df = 11, $P = \leq 0.0001$). Only treatment one, a high rate (1.12 kg ai/ha) of acephate in furrow, resulted in a statistically significant higher yield (472.6 kg/ha) ($F =$

3.28, df = 14, *P* = 0.003), however, all treatments resulted in numerically higher yields (Table 4.10).

In these studies, most cyantraniliprole treatments had a positive impact on reducing immature thrips numbers and thrips injury to peanut seedlings. However, the results are somewhat inconsistent across years and trials. Because of these inconsistencies, we recommend further investigation of cyantraniliprole use for thrips control in peanuts. With future studies and more insight on cyantraniliprole's effectiveness for thrips control in peanut, cyantraniliprole may prove to be a useful new insecticide for this crop.

Although cyantraniliprole is a relatively new diamide insecticide, it has captured the attention of the scientific community. Unlike its predecessors, chlorantraniliprole (DuPont Crop Protection, Wilmington, DE) and flubendiamide (Bayer Crop Science, Research Triangle Park, NC), which are mainly active against lepidopteran pests, cyantraniliprole has a wider range of insecticidal activity, which includes Lepidoptera, Coleoptera, Hemiptera, and Thysanoptera pests (Lahm et al. 2009, Fettig et al. 2010, Kuhar et al. 2010, Hardke et al. 2011). Its low mammalian toxicity is attributed to specific selectivity for insect ryanodine receptors. Also, no cross-resistance with other available insecticides has been found (Lahm et al. 2009). Based on the evidence available to date, cyantraniliprole should fit well into existing IPM practices and its novel mode of action, when rotated properly with different chemistries, could help mitigate resistance development in target pest populations.

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Table 4.1. Foliar broadcast spray (BC) and in-furrow (IF) applied products on cotton in 2010 and 2011. Tidewater AREC, Suffolk, VA.

Year of application	Material and application method	Rate: kg a.i./ha
2010	Cyantraniliprole 20SC (IF) + Cyantraniliprole 10OD (BC)	0.10 + 0.10
2010	Cyantraniliprole 20SC (IF)	0.15
2010	Cyantraniliprole 20SC (IF)	0.20
2010	Cyantraniliprole 20SC (IF) + Cyantraniliprole 10OD (BC)	0.15 + 0.10
2010	Aldicarb 15G (IF)	0.84
2010	Aldicarb 15G (IF) + Acephate 97 (BC)	0.84 + 0.27
2011	Cyantraniliprole 20SC (IF)	0.15
2011	Cyantraniliprole 20SC (IF)	0.20
2011	Cyantraniliprole 20SC (IF) + Cyantraniliprole 10OD (BC)	0.15 + 0.1
2011	Acephate 97 (IF) + Acephate 97 (BC)	0.84 + 0.41

Table 4.2. Foliar broadcast spray (BC) and in-furrow (IF) applied products on peanut in 2010 and 2011. Tidewater AREC, Suffolk, VA.

Year of application and test name where applicable	Material and application method	Rate: kg a.i./ha
2010	Cyantraniliprole 20SC (IF) + Cyantraniliprole 10OD (BC)	0.10 + 0.10
2010	Cyantraniliprole 20SC (IF)	0.15
2010	Cyantraniliprole 20SC (IF)	0.20
2010	Cyantraniliprole 20SC (IF) + Cyantraniliprole 10OD (BC)	0.15 + 0.10
2010	Phorate 20G (IF)	1.12
2010	Aldicarb 15G (IF)	1.18
2010	Aldicarb 15G (IF) + Acephate 97 (BC)	1.18 0.27
2011 thrips trial 1	Phorate 20G (IF)	1.12
2011 thrips trial 1	Acephate 97 (IF) + Acephate 97 (BC)	0.84 0.41
2011 thrips trial 1	Cyantraniliprole 10OD (BC) + Cyantraniliprole 10OD (BC)	0.10 + 0.10
2011 thrips trial 1	Cyantraniliprole 20SC (IF) + Acephate 97 (BC)	0.20 + 0.41
2011 thrips trial 1	Cyantraniliprole 20SC (IF) + Cyantraniliprole 10OD (BC)	0.20 + 0.10
2011 thrips trial 2	Acephate 97 (IF)	1.12
2011 thrips trial 2	Cyantraniliprole 20SC (IF)	0.15
2011 thrips trial 2	Cyantraniliprole 20SC (IF)	0.20

Table 4.3. Mean (± SE) number of immature thrips per 5 plants, cotton 2010. Tidewater AREC, Suffolk, VA, 2010. In-furrow applications were made on May 5. Broadcast at first true leaf applications were made on May 21.

Treatment[1]	Rate: (kg a.i./ha)	May 24 (1st true leaf stage)	May 31 (2-3 true leaf stage)	June 9 (5-6 true leaf stage)	June 16 (6th true leaf stage)
Cyantraniliprole 20SC (IF) + Cyantraniliprole 10OD (BC)	0.10 + 0.10	0.3 ± 0.25 b	4.0 ± 2.04 c	3.0 ± 0.71 bc	5.8 ± 0.85 b
Cyantraniliprole 20SC (IF)	0.15	0.8 ± 0.25 b	23.3 ± 8.86 b	3.8 ± 1.11 bc	8.5 ± 1.89 b
Cyantraniliprole 20SC (IF)	0.20	0.5 ± 0.5 b	12.3 ± 1.75 bc	1.5 ± 0.50 c	2.8 ± 2.14 b
Cyantraniliprole 20SC (IF) + Cyantraniliprole 10OD (BC)	0.15 + 0.10	0.5 ± 0.29 b	2.3 ± 1.44 c	3.0 ± 1.35 bc	5.3 ± 3.68 b
Aldicarb 15G (IF)	0.84	0.3 ± 0.25 b	17.5 ± 7.59 bc	7.0 ± 2.48 b	3.0 ± 0.82 b
Aldicarb 15G (IF) + Acephate 97 (BC)	0.84 + 0.27	0.3 ± 0.25 b	3.3 ± 0.95 c	1.5 ± 0.65 c	7.0 ± 2.04 b
Non-treated	---	4.5± 1.55 a	159.3 ± 7.83 a	13.0 ± 1.96 a	19.5 ± 7.51 a

Means within a column followed by the same letter(s) are not significantly different (Protected LSD, P>0.05).

[1] *IF = In-furrow application, BC = Broadcast application*

Table 4.4. Mean rank (± SE) thrips injury to plants ratings[1] and mean (± SE) lint yield, cotton 2010. Tidewater AREC, Suffolk, VA, 2010. In-furrow applications were made on May 5. Broadcast at first true leaf applications were made on May 21.

Treatment[2]	Rate: (kg a.i./ha)	Thrips injury to plants rating				Yield kg/ha[3]
		May 28 (2nd true leaf stage)	Jun 1 (2-3 true leaf stage)	June 7 (4th true leaf stage)	June 17 (6th true leaf stage)	
Cyantraniliprole 20SC (IF) + Cyantraniliprole 10OD (BC)	0.10 + 0.10	2.19 ± 0.52 b	1.19 ± 0.06 b	1.06 ± 0.06 c	1.69 ± 0.06 b	1651 ± 56.95 ab
Cyantraniliprole 20SC (IF)	0.15	1.94 ± 0.13 bc	1.44 ± 0.06 b	1.69 ± 0.06 b	0.75 ± 0 c	1462 ± 59.31 b
Cyantraniliprole 20SC (IF)	0.20	1.75 ± 0 bc	1.19 ± 0.06 b	1.56 ± 0.06 b	0.94 ± 0.12 cd	1735 ± 58.61 a
Cyantraniliprole 20SC (IF) + Cyantraniliprole 10OD (BC)	0.15 + 0.10	1.69 ± 0.06 bc	1.13 ± 0.07 b	0.75 ± 0 d	0.75 ± 0.25 cd	1773 ± 36.24 a
Aldicarb 15G (IF)	0.84	1.50 ± 0.14 c	0.75 ± 0 c	0.94 ± 0.06 c	1.25 ± 0.10 d	1511 ± 52.84 b
Aldicarb 15G (IF) + Acephate 97 (BC)	0.84 + 0.27	1.06 ± 0.06 d	0.50 ± 0 d	0.50 ± 0 e	0.81 ± 0.12 c	1786 ± 47.74 a
Non-treated	---	3.75 ± 0 a	4.63 ± 0.07 a	4.83 ± 0.03 a	4.80 ± 0.03 a	693 ± 107.57 c

Means and mean ranks within a column followed by the same letter(s) are not significantly different (Protected LSD, P>0.05).

[1] *Thrips injury based on a 0-5 scale, 0 = no injury and 5 = dead plants.*

[2] *IF = In-furrow application, BC = Broadcast application*

[3] *Cotton was harvested on October 12. Gross yields were reduced by 57.7% to account for seed and trash.*

Table 4.5. Mean (± SE) number of immature thrips per 5 plants, mean rank (± SE) thrips injury to plant ratings, and mean (± SE) lint yield, cotton 2011. Tidewater AREC, Suffolk, VA, 2011. In-furrow applications were made on May 2. Broadcast at 1st true leaf applications were made on May 20.

Treatment[2]	Rate (kg a.i./ha)	Immature thrips per 5 plants		Thrips injury to plant rating[1]			Yield kg/ha[3]
		May 24 (1-2 true leaf stage)	May 31	May 24	May 31	June 8	
Cyantraniliprole 20SC (IF)	0.15	19.50 ± 5.48 b	35.75 ± 6.98 b	2.44 ± 0.16 b	3.69 ± 0.06 b	2.81 ± 0.06 b	1098 ± 26.13
Cyantraniliprole 20SC (IF)	0.20	18.25 ± 7.25 b	18.75 ± 6.84 bc	2.31 ± 0.19 b	3.44 ±0.12 b	2.75 ± 0 b	1064 ± 36.11
Cyantraniliprole 20SC (IF) Cyantraniliprole 10OD (BC)	0.15 + 0.1	10.50 ± 3.57 b	23.25 ± 3.64 bc	2.13 ± 0.22 b	3.25 ±0.18 b	2.69 ± 0.06 b	1011 ± 57.95
Acephate 97 (IF) + Acephate 97 (BC)	0.84 + 0.41	2.75 ± 0.25 b	4.50 ± 1.85 c	0.75 ± 0.10 d	1.50 ± 0 c	0.63 ± 0.07 c	1029 ± 60.51
Non-treated		73.75 ± 15.46 a	62.50 ± 12.71 a	3.50 ± 0.10 a	4.50 ± 0 a	4.31 ± 0.12 a	935 ± 17.67

Means and mean ranks within a column followed by the same letter(s) are not significantly different (Protected LSD, P>0.05).

[1]*Thrips injury based on a 0-5 scale, 0 = no injury and 5 = dead plants.*

[2]*IF = In-furrow application, BC = Broadcast application*

[3]*Cotton was harvested on October 21. Gross yields were reduced by 57.9% to account for seed and trash.*

Table 4.6. Mean (± SE) immature thrips per ten terminal leaflets, peanut 2010. Tidewater AREC, Suffolk, VA, 2010. In-furrow applications were made on April 30. Broadcast at late ground cracking applications were made on May 21.

Treatment[1]	Rate: (kg a.i./ha)	Thrips per 10 terminal leaflets[2]	
		May 25	June 1
Cyantraniliprole 20SC (IF) + Cyantraniliprole 10OD (BC)	0.10 + 0.10	0.0 ± 0 b	3.8 ± 1.65 b
Cyantraniliprole 20SC (IF)	0.15	2.5 ± 2.18 b	7.8 ± 3.36 b
Cyantraniliprole 20SC (IF)	0.20	0.0 ± 0 b	5.5 ± 2.22 b
Cyantraniliprole 20SC (IF) + Cyantraniliprole 10OD (BC)	0.15 + 0.10	0.3 ± 2.18 b	2.5 ± 0.87 b
Phorate 20G (IF)	1.12	0.5 ± 1.12 b	4.3 ± 1.97 b
Aldicarb 15G (IF)	1.18	0.0 ± 0 b	4.0 ± 1.87 b
Aldicarb 15G (IF) + Acephate 97 (BC)	1.18 0.27	0.0 ± 0 b	0.5 ± 0.29 b
Non-treated	---	11.5 ± 4.87 a	62.5 ± 24.03 a

Means within a column followed by the same letter(s) are not significantly different (Protected LSD, P>0.05)

[1] *IF = In-furrow application, BC = Broadcast application*

[2] *Ten leaflets were sampled per plot on each date.*

Table 4.7. Mean rank (± SE) thrips injury to plants ratings[1] and mean (± SE) yield, peanut 2010. Tidewater AREC, Suffolk, VA, 2010. In-furrow applications were made on April 30. Broadcast at late ground cracking applications were made on May 21.

Treatment[1]	Rate: (kg a.i./ha)	Thrips injury to plant rating[2]					Yield kg/ha[3]
		May 21	May 28	June 1	June 7	June 17	
Cyantraniliprole 20SC (IF) + Cyantraniliprole 10OD (BC)	0.10 + 0.10	2.13 ± 0.13 b	1.69 ± 0.06 c	2.13 ± 0.81 b-d	3.38 ± 0.75 bc	2.31 ± 0.16 b	4817 ± 409.70
Cyantraniliprole 20SC (IF)	0.15	1.44 ± 0.16 c	1.44 ± 0.26 bc	2.44 ± 0.26 b	3.63 ± 0.26 b	1.13 ± 0.13 cd	5203 ± 190.93
Cyantraniliprole 20SC (IF)	0.20	0.69 ± 0.16 d	1.44 ± 0.19 bc	1.69 ± 0.06 c	3.63 ± 0.32 b	1.31 ± 0.06 c	4746 ± 201.56
Cyantraniliprole 20SC (IF) + Cyantraniliprole 10OD (BC)	0.15 + 0.10	0.75 ± 0.10 d	1.19 ± 0.12 b	0.94 ± 0.12 d	1.25 ± 0.42 cd	0.69 ± 0.06 e	5296 ± 291.48
Phorate 20G (IF)	1.12	0.75 ± 0 d	0.75 ± 0 d	0.75 ± 0 de	0.88 ± 0.07 d	0.56 ± 0.06 e	5280 ± 46.09
Aldicarb 15G (IF)	1.18	0.56 ± 0.12 de	0.94 ± 0.06 bd	0.69 ± 0.12 de	0.94 ± 0.12 d	0.88 ± 0.07 de	5365 ± 349.55
Aldicarb 15G (IF) + Acephate 97 (BC)	1.18 0.27	0.50 ± 0 de	0.75 ± 0 d	0.50 ± 0 e	0.69 ± 0.12 d	0.69 ± 0.06 e	5233 ± 429.14
Non-treated	---	2.63 ± 0.13 a	3.75 ± 0 a	6.50 ± 0 a	7.81 ± 0.12 a	3.94 ± 0.06 a	4616 ± 304.40

Means and mean ranks within a column followed by the same letter(s) are not significantly different (Protected LSD, P>0.05)

[1] *IF = In-furrow application, BC = Broadcast application*

[2] *Thrips injury rated on a 0-10 scale, 0 = no injury and 10 = dead plants.*

[3] *Yield based on weight of peanut with moisture content of 7%. Dig date = September 23 and harvest date = October 7.*

Table 4.8. Mean (± SE) number of immature thrips per 10 terminal leaflets, peanut trial 1, 2011. Tidewater AREC, Suffolk, VA, 2011. In-furrow applications were made on May 3. Broadcast at late ground cracking applications were made on May 23 and May 27.

Treatment[1]	Rate: (kg a.i./ha)	Thrips per 10 terminal leaflets[2]	
		May 25	June 3
Phorate 20G (IF)	1.12	0.25 ± 0.25 b	6.25 ± 1.11 b
Acephate 97 (IF) + Acephate 97 (BC)	0.84 0.41	0.50 ± 0.29 b	4.25 ± 1.11 b
Cyantraniliprole 10OD (BC)+ Cyantraniliprole 10OD (BC)	0.10 + 0.10	5.50 ± 1.04 b	10.50 ± 3.59 b
Cyantraniliprole 20SC (IF) + Acephate 97 (BC)	0.20 + 0.41	2.00 ± 0.71 b	11.25 ± 4.35 b
Cyantraniliprole 20SC (IF) + Cyantraniliprole 10OD (BC)	0.20 + 0.10	1.25 ± 0.48 b	9.50 ± 3.01 b
Non-treated		14.00 ± 4.20 a	22.75 ± 5.54 a

Means within a column followed by the same letter(s) are not significantly different (Protected LSD, P>0.05).

[1] *IF = In-furrow application, BC = Broadcast application*

[2] *Ten leaflets were sampled per plot on each date.*

Table 4.9. Mean rank (± SE) thrips injury to plant ratings[1] and mean (± SE) yield, peanut trial 1, 2011. Tidewater AREC, Suffolk, VA, 2011. In-furrow applications were made on May 3. Broadcast at late ground cracking applications were made on May 23 and May 27.

Treatment[2]	Rate: (kg a.i./ha)	Thrips injury to plant rating				Yield kg/ha[3]
		May 24	May 31	June 9	June 15	
Phorate 20G (IF)	1.12	1.81 ± 0.16 b	1.69 ± 0.06 c	0.75 ± 0 d	0.75 ± 0 e	5037 ± 82.97 b-e
Acephate 97 (IF) + Acephate 97 (BC)	0.84 0.41	0.63 ± 0.07 c	1.38 ± 0.07 d	0.69 ± 0.06 d	1.19 ± 0.06 d	5896 ± 479.78 ab
Cyantraniliprole 10OD (BC)+ Cyantraniliprole 10OD (BC)	0.10 + 0.10	3.50 ± 0 a	3.56 ± 0.06 b	1.25 ± 0 c	1.38 ± 0.13 d	4650 ± 313.15 c-e
Cyantraniliprole 20SC (IF) + Acephate 97 (BC)	0.20 + 0.41	2.06 ± 0.06 b	3.56 ± 0.06 b	2.75 ± 0.20 b	2.06 ± 0.26 c	4525 ± 165.48 de
Cyantraniliprole 20SC (IF) + Cyantraniliprole 10OD (BC)	0.20 + 0.10	1.88 ± 0.16 b	3.50 ± 0.18 b	3.44 ± 0.16 b	3.75 ± 0.20 b	4480 ± 114.98 de
Non-treated		3.50 ± 0 a	5.69 ± 0.12 a	6.50 ± 0.10 a	6.19 ± 0.19 a	4261 ± 139.99 e

Means and mean ranks within a column followed by the same letter(s) are not significantly different (Protected LSD, P>0.05).

[1]Thrips injury rated on a 0-10 scale, 0 = no injury and 10 = dead plants. Peanut was planted on May 3.

[2] IF = In-furrow application, BC = Broadcast application

[3]Yield based on weight of peanut with moisture content of 7%. Dig date = October 6 and harvest date = October 14.

Table 4.10. Mean rank (± SE) thrips injury to plant ratings[1] and mean (± SE) yield, peanut thrips trial 2, 2011. Tidewater AREC, Suffolk, VA, 2011. All treatments were applied as liquid in-furrows at planting on May 3.

Treatment[2]	Rate: (kg a.i./ha)	Thrips injury to plant rating				Yield kg/ha[3]
		May 24	May 31	June 9	June 15	
Acephate 97 (IF)	1.12	0.50 ± 0 c	1.50 ± 0.10 c	1.00 ± 0 c	1.63 ± 0.07 d	5232 ± 92.65 b
Cyantraniliprole 20SC (IF)	0.15	1.50 ± 0.10 b	3.06 ± 0.24 b	3.56 ± 0.12 b	3.69 ± 0.26 b	4604 ± 125.51 ab
Cyantraniliprole 20SC (IF)	0.20	1.50 ± 0.10 b	2.81 ± 0.21 b	3.69 ± 0.21 b	2.81 ± 0.12 c	4824 ± 153.78 ab
Non-treated		2.75 ± 0.14 a	4.94 ± 0.16 a	6.81 ± 0.12 a	6.31 ± 0.12 a	4414 ± 166.97 a

Means and mean ranks within a column followed by the same letter(s) are not significantly different (Protected LSD, P>0.05).

[1] *Thrips injury rated on a 0-10 scale, 0 = no injury and 10 = dead plants. Peanut was planted on May 3.*

[2] *IF = In-furrow application, BC = Broadcast application*

[3] *Yield based on weight of peanut with moisture content of 7%. Dig date = October 6 and harvest date = October 11.*

Chapter Five

Acute toxicity of cyantraniliprole, lambda-cyhalothrin, and acephate against *Frankliniella fusca* (Thysanoptera: Thripidae).

Abstract

Leaf-dip assays were conducted to help elucidate the relative toxicity of cyantraniliprole, lambda-cyhalothrin, and acephate to tobacco thrips. Non-insecticide treated peanut leaves were dipped into specific concentrations of insecticides and then air dried. Small leaf disks were cut from the larger leaves and placed into 1.5 ml micro-centrifuge tubes; then tobacco thrips adults were placed into the tube. Mortality was recorded every 24 hours for three days. Toxicity data were highly variable. However, cyantraniliprole produced replicable results in most trials with an average LC_{50} of 178.5 ppm. The results for the other two insecticides were inconclusive and at this time we cannot accurately determine the lethal concentration for fifty percent of the test population (LC_{50}). Further testing is needed to verify this result and to obtain results for the other two test compounds.

Thrips have long been recognized as pests in multiple cropping systems. They cause injury to plants through feeding, oviposition, and virus transmission (Childers and Achor 1995, Mound, 1997, Childers 1997, Osekre et al. 2009). In Virginia, thrips are recognized as potentially devastating early season pests of cotton and peanut crops (Herbert et al. 2011). In both cases their injury can lead to economic damage by reducing plant vigor leading to yield losses (Herbert 1998, Faircloth et al. 2002). In peanut, *Frankliniella fusca* and *F. occidentalis* thrips are known vectors of tomato spotted wilt virus (TSWV) (*Bunyaviridae Tospovirus*). This virus causes and malformations of fruit pods, reducing yield and fruit quality, and in severe cases will cause whole plant death (Womack et al. 1981, Drake et al. 2009). Thrips injury to cotton seedlings leads to delayed maturity, fewer bolls, and lowered fiber quality (Gaines 1934, Dunnam and Clark 1937, Watts 1937, Lei and Wilson 2004, Greenberg et al. 2009).

The most common management strategy for thrips is chemical control using broadspectrum (carbamate, organophosphate or pyrethroid) insecticides (Olsen et al. 2006, Greenberg et al. 2009, Toews et al. 2010, Herbert et al. 2011). This approach has also been shown to lead to outbreaks of secondary pests, toxicity to beneficial species, development of resistance in the target insect, and potential toxicity for the applicator (Zhao et al. 1995, Allen et al. 2005, Duso et al. 2008, Herron et al. 2008). These concerns have led to the development of new insecticides that have stronger target specificity, reduced toxicity for both beneficial insects and human applicators, and new modes of action reducing the potential for resistance development (Isayama et al. 2005, Bruck et al. 2009, Cameron et al. 2009).

One such novel insecticide is cyantraniliprole (DuPont Crop Protection, Wilmington, DE), which has been shown to reduce thrips feeding injury and TSWV transmission by tobacco thrips, *Frankliniella fusca* Hinds (Jacobson and Kennedy 2011). Cyantraniliprole is an anthranilic diamide insecticide that acts as an agonist targeting ryanodine receptors in insects (IRAC group 28) (IRAC 2011, Sattelle et al. 2008). Similar to other ryanodine receptor modulator insecticides, cyantraniliprole causes muscle impairment in the insect, which leads to lethargy, cessation of feeding, reduced reproduction, and eventually death.

Relatively little data exist on the relative toxicity of insecticides against tobacco thrips. Most research on insecticide toxicity to thrips has focused on western flower thrips, *F. occidentalis* Pergande and onion thrips *Thrips tabaci* Lindeman, or the authors did not identify the thrips species being investigated (e.g., Bielza et al. 2008, Lopez et al. 2008). The objective of this study was to determine the relative toxicity of selected insecticides against tobacco thrips using a leaf dip bioassay method.

Materials and Methods

Leaf dip bioassays were conducted during 2010 and 2011 to determine the relative toxicity of cyantraniliprole, acephate (Amvac, Los Angeles, CA), and lambda-cyhalothrin (Syngenta Crop Protection LLC, Greensboro, NC) to tobacco thrips. A colony of tobacco thrips was acquired from North Carolina State University and maintained at the Virginia Tech Tidewater Agricultural Research and Extension Center (TAREC), Suffolk, VA. The insects were

reared in 1.9-liter plastic tubs. The tubs had approximately 5-cm diameter holes cut into the top and bottom of the container covered with thrips-proof screening to aid in air circulation (100 micron, Midwest Filter Corporation, Lake Forest, IL). The colony was maintained on a diet of green beans (*Phaseolus vulgaris*), which were washed in a 1% bleach solution and rinsed with water prior to administration, and kept on a 16:8 (L:D) h. cycle at 29.45°C +/- 3°C with 85 to 90% relative humidity.

A total of five bioassays, each performed on different dates, were conducted for each insecticide. A series of dilutions with multiple replicates (4-8) were prepared for each insecticide. Table 5.1 provides the active ingredient in parts per million (ppm) that were tested for cyantraniliprole, lambda-cyhalothrin, and acephate, respectively. Acephate and lambda-cyhalothrin were chosen because they are standard insecticides used for thrips control in peanut, while cyantraniliprole was chosen because it is a novel insecticide being investigated for thrips control. In the first two assays we used concentrations based on the lowest labeled field rate for each product based on the Virginia Peanut Production Guide (Wilson et al. 2010) along with several other dilutions, as seen in the tables. For subsequent bioassays we adjusted the concentrations and dilutions based on the previous bioassays to accurately determine the LC_{50} values for each insecticide. In cases where the previous bioassays were unsuccessful, concentrations were either not adjusted or adjusted to the best of our ability based on the little information available from the assays. For example, if mortality was high in all tested concentrations of an insecticide then the concentrations for the next bioassay would be lowered below the lowest rate used in that initial bioassay.

Greenhouse grown, non-insecticide treated 'CHAMPS' variety peanut leaves were used as the substrate for insecticide exposure to the insects. The leaves were dipped into one of the insecticide dilutions, then allowed to drip dry for one hour or until the leaf surface appeared dry. Once dry, two 6.35-mm diameter disks were cut from the leaves and placed into labeled 1.5-ml, sterile micropipette tubes. Once all of the leaf disks were in place, 4-6 adult thrips were allocated to the tubes, and maintained at room temperature (22.8°C +/- 3°C). The control tubes followed the same procedure as above except that the peanut leaves were not dipped into an insecticide solution. Thrips mortality was assessed every 24 hours for three consecutive days.

Adult thrips were not handled during these assays. To get the adult thrips into the micropipette tubes, the thrips were allowed to crawl up the sides of their rearing container and when the testing tube was placed just below their location they would readily jump into the tube on their own. To assess mortality the tube was tapped multiple times with a pencil; thrips which did not move after agitation were considered dead, the slightest movement of a leg or antennae however designated the insect as alive. Data on mortality recorded at 72 hours were analyzed using the U.S. EPA's probit analysis program available to the public on their website (EPA, 2006). Mathematical computations in the program are based on Finney (1971) with data corrected for mortality in the control (if present) using Abbott's formula. All statistical tests were carried out at $\alpha = 0.05$.

Results

A total of 2,397 thrips were tested among the three insecticides with a total of 28 replicates per insecticide. The mean mortality in the non-treated control, across all five tests was 20.08%. Mortality in the control occurred at a rate of 21.2 percent. The test-specific mortality was 29.2 %, 37.5 %, 12.5%, and 0% for tests one through five, respectively.

A total of five bioassays were performed with cyantraniliprole, concentrations ranged from 0.1 to 1000 ppm (Table 5.1). In test one, a rate of 177.3 ppm (slope = 0.92, $\chi^2 = 2.61$) cyantraniliprole was found to kill fifty percent of the tested population (LC_{50}). The LC_{50} value determined from test two was 73.7 ppm (slope = 0.62, $\chi^2 = 3.85$), almost 100 ppm lower than the rate ascertained in test one. In test three cyantraniliprole at a rate of 174.8 ppm (slope = 3.09, $\chi^2 = 0.31$) killed fifty percent of the test population, which is consistent with results from test one. The proposed LC_{50} of 183.4 ppm (slope = 1.44, $\chi^2 = 10.48$) from test four is within a consistent range of the LC_{50} values proposed by test one and test three (177.3 and 174.8, respectively) for cyantraniliprole. Finally, test five did not produce usable data for any of the tested compounds (Table 5.2).

Five different tests were conducted with lambda-cyhalothrin with concentrations ranging from 0.06 to 1000 ppm (Table 5.1). Lambda-cyhalothrin had an LC_{50} of 0.13 ppm (slope

= 0.79, χ^2 = 0.81) in test one (Table 5.2). The first bioassay was the only test which yielded usable data for this compound.

The concentrations of acephate tested in five different trials ranged from 0.1 to 2042.5 ppm. In test three, the LC_{50} for acephate was found to be 297.1 ppm; however, because of the unusually wide confidence interval, and the fact that the analysis program determined 297.1 ppm to be not only the LC_{50}, but also the LC_{10} and $LC_{15,}$ we do not believe this result to be accurate. No other tests with acephate provided usable data.

Discussion

Although every effort was made to obtain consistent, documentable results, the assays were as a whole unsuccessful. Only the LC_{50} for cyantraniliprole was able to be replicated, but not in all five tests. We feel the average LC_{50} of 178.5 ppm for cyantraniliprole should receive more thorough testing before it is deemed accurate. However, cyantraniliprole's apparent toxicity against tobacco thrips in these bioassays, a result which has also been shown in small-plot field trials, leads us to conclude that this insecticide may be an appropriate recommendation for thrips control. No clear discernible LC_{50} was obtained for either lambda-cyhalothrin or acephate and those individual assays that produced LC_{50} estimates could not be replicated and therefore cannot be deemed accurate.

There are multiple possibilities why these assays preformed so poorly. One was that the type of assay used was inappropriate. Lopez et al. (2008) used the adult vial technique to test relative toxicity of selected insecticides against undetermined thrips spp. With this method, the inside of a vial is thoroughly coated with the insecticide of interest. Once dry, the insects are introduced to the vial, and in the case of thrips, they crawl around the vial coming in contact with the insecticide treated surface. However, Nehare et al. (2010) and others have successfully used a leaf-dip method similar to the one used in my study to look at resistance and cross resistance patterns in diamondback moth, *Plutella xylostella* L. (Shelton et al. 1993a, 1993b; Cordero and Kuhar 2007). Another source for error lies in the experimenter and/or the equipment used. Although proper protocol was followed to help ensure accuracy, natural human error could have occurred. Finally, the natural vitality of the thrips colony used should

be called into question. On all but one occasion there was mortality recorded in the control vials. The colony used has been reared in the laboratory for many years and that may have reduced fitness, although it does not explain why fitness varied between tests. There have been reports of differing susceptibility found in laboratory reared insects as opposed to wild-caught insects, differing susceptibility between generations, and natural variation within a population that can pose complications when performing bioassays (Robertson et al. 1976, Savin et al. 1977, Robertson et al. 1995).

Regardless of the reasons these assays were unsuccessful, the original objective should not be forgotten. Consistent data for relativity toxicity of insecticides against tobacco thrips is still missing from the literature. Thrips are and will continue to be a major pest species of concern in agriculture for the foreseeable future and every effort should be made to obtain information which may help mitigate this pest's damage to our crops.

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Table 5.1. Percent active ingredient (a.i.) in parts per million (ppm) of cyantraniliprole, lambda-cyhalothrin, and acephate tested against adult tobacco thrips. Tidewater AREC, Suffolk, VA, 2010 and 2011.

Product	Rates: ppm, 2010 Trials	Rates: ppm, 2011 Trials		
	Test 1 & 2	Test 3	Test 4	Test 5
Cyantraniliprole	0.1 1 10 100 738.1 1000	94.8 142.2 213.3 320 480	30.7 46.1 69.1 103.7 155.6 233.3 350	39.5 59.3 88.9 133.3 200
Lambda-cyhalothrin	0.1 1 10 100 174.5 1000	0.06 0.09 0.13 0.20 0.30	0.4 0.7 1.0 1.5 2.2 3.3	0.9 1.3 2.0 3.0 4.4 6.7 10
Acephate	0.1 1 10 100 1000 2042.5	35.1 52.7 79.0 118.5 117.8	26.3 39.5 59.3 88.9 133.3 200 300	87.8 131.7 197.5 296.3 444.4 666.7 1000

Table 5.2. Tabulated lethal concentration in parts per million (PPM) for fifty percent (LC_{50}) of the tested population of adult tobacco thrips and associated 95% confidence intervals. Tidewater AREC, Suffolk, VA, 2010 and 2011.

Product	LC_{50} in PPM (χ^2; 95% Confidence Interval)				
	Test 1	**Test 2**	**Test 3**	**Test 4**	**Test 5**
Cyantraniliprole	177.3 (2.61; 25.5-465.5)	73.7 (3.85; 1.8-370.6)	174.8 (0.31; 69.8-251.9)	183.4 (10.48; 103-342.5)	Slope not significantly different from zero
Lambda-cyhalothrin	0.13 (0.81; 0.003-0.583)	Iterations are not converging[1]	Slope not significantly different from zero	Slope not significantly different from zero	Iterations are not converging
Acephate	Slope not significantly different from zero	Slope not significantly different from zero	297.1[2] (-1.34; 90.3-362430.8)	Slope not significantly different from zero	Iterations are not converging

[1] *This usually means that only one concentration is on the linear portion of the slope.*
[2] *Unusually large confidence interval, value not believed to be accurate.*

Conclusion

Thrips monitoring in Suffolk, Virginia revealed the presence of eleven different species; the most commonly encountered species were: *Frankliniella fusca* (Hinds), *Frankliniella tritici* (Fitch), *Frankliniella occidentalis* (Pergande), *Thrips tabaci* (Lindeman), *Neohydatothrips variabilis* (Beach), and *Chirothrips texanus* (Andre). Other thrips species identified included *Limothrips cerealium* (Haliday), *Caliothrips fasciapennis* (Pergande), *Scolothrips sexmaculatus* (Pergande), *Plesiothrips perplexus* (Beach), and *Aeolothrips bicolor* (Hinds). In addition, multiple specimens from the suborder *Tubulifera* were encountered, but not identified to species; and at least one specimen from the genus *Haplothrips* was believed to have been found, but further identification is needed. Sticky card monitoring also determined that adult thrips catches peaked in August and September, but individual peak catches of a specific species varied monthly (Chapter 2). Future studies should focus on understanding the motivation behind peak flights and also what the variability in a species timing of that flight is related to. Also, monitoring for thrips populations under different vegetative environments than the ones used in this study may reveal even more native thrips species present in Virginia, aiding in our overall knowledge and understanding of these insects.

Results from studies conducted in soybean to examine the within plant distribution and species complex present showed that *F. fusca* is the initial colonizer of soybean seedlings, but they move quickly out of the crop and *N. variabilis* becomes the dominate species. Immature thrips were most dense in the terminal section of the plant, which is consistent with the aggregated distribution found by Irwin et al. (1979) and other researchers (Salguero Navas et al. 1994, Cho et al. 1995, Joost and Riley 2004). Results of our study showed that the number of thrips on the uppermost fully opened trifoliate is related to the total number per seedling, but only about 50 percent of the variation in numbers of thrips can be explained by this sampling unit. The remainder of the plant accounted for 88% of the variation in thrips numbers. Using the Remainder of the plant as a sample unit would be possible; however, in our study the Remainder was comprised of two to three fully opened trifoliates out of a total of three or four trifoliates on the entire plant. We do not feel this is a practical sample unit as it would require more time (the sectioning of the seedlings) than sampling the whole plant (Chapter 3). More

work is needed to improve sampling techniques for thrips in soybean and other sampling methods need to be investigated.

Field trials in both cotton and peanut with the novel insecticide cyantraniliprole, demonstrated its efficacy for thrips control. Results were more consistent in cotton across two years, but in both crops cyantraniliprole reduced immature thrips numbers below those in the non-treated control and it reduced the visual rating of thrips injury to plants (Chapter 4). Cyantraniliprole has a novel mode of action, low toxicity to mammals and beneficial insects, longer residual activity against target pests, and increased plant mobility with respect to other diamide insecticides (Lahm et al. 2009, Hardke et al. 2011). These attributes combined with the positive performance of cyantraniliprole in 2010 and its ability to perform well under the harsh growing conditions of 2011, make it a promising new tool for the management and control of thrips. Future research should focus on improving our knowledge about and use of this new insecticide; as well as determining the best use of this product in an integrated pest management program.

A series of bioassays was conducted using cyantraniliprole, lambda-cyhalothrin, and acephate to determine the lethal concentration for 50% (LC_{50}) of adult thrips from the species *F. fusca*. Assays with cyantraniliprole revealed the LC_{50} to be 178.5 ppm; assays with lambda-cyhalothrin and acephate were inconclusive (Chapter 5). The ability to replicate results across different bioassay tests was an issue in this study; therefore we recommend further research before drawing any definitive conclusions from this work.

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