## ARTHROPOD MANAGEMENT & APPLIED ECOLOGY

# The Effect of Neonicotinoids on Thrips Oviposition, Immatures, and Cotton Injury

Jessica B. Mahas, Giovani Rossi, Anitha Chitturi, Bernardo Chaves-Cordoba, Scott H. Graham, and Alana L. Jacobson\*

#### **ABSTRACT**

Thrips are one of the major pests of seedling cotton (Gossypium hirsutum L.) in the U.S. Whereas previous studies have investigated the effects of neonicotinoids on reducing adult and immature thrips on cotton, their effect on oviposition in the field is understudied. In this study the effect of three neonicotinoid insecticide seed treatments on the number of thrips adults, immatures, eggs, plant injury, and plant biomass were evaluated. The experiment was replicated six times over weekly planting dates. Insecticides reduced thrips oviposition and immature thrips on seedlings. Seedlings from plantings in April had lower oviposition and number of immature thrips than seedlings planted in May, and the efficacy of insecticides against oviposition and immature thrips was reduced as seedlings reached four to five true leaves. Oviposition was not reduced by insecticides in seedlings planted after 2 May that were exposed to larger numbers of adult thrips. Insecticides reduced plant injury, which was variable across planting dates and growth stages. Differences in biomass were observed only on the last planting date and were likely due to more favorable growing conditions. The efficacy of these insecticide seed treatments to reduce thrips oviposition and numbers of immature insects depends upon pest population pressure, growth stage of the cotton, and growing conditions of the crop.

Upland cotton, Gossypium hirsutum L., is a major crop across the southeastern U.S., contributing nearly \$6 billion in production to the economy in 2022 (USDA NASS, 2023). The 13 states that produce the bulk of cotton in the U.S. are referred

J.B. Mahas, G. Rossi, A. Chitturi, S.H. Graham, and A.L. Jacobson\*, Auburn Univ. Dept. of Entomology and Plant Pathology, 301 Funchess Hall, Auburn, AL 36849; and B. Chaves-Cordoba, Auburn Univ. College of Agriculture Research Office, 101 Comer Hall, Auburn, AL 36849. \*Corresponding author: alj0043@auburn.edu

to as the Cotton Belt and extend from Texas to Virginia. This region produced 11.1 million acres in 2022 (USDA NASS, 2023): 11 million acres of *G. hirsutum* and the remaining acreage was *Gossypium barbadense* L. Thrips are a primary pest of seedling cotton because their feeding causes injury that can lead to deformed plant growth, reduced yield, and delayed maturation (Cook and Threet, 2022; Cook et al., 2011). Management programs aim to reduce feeding injury primarily caused by immature thrips with the use of at-plant or foliar applications of insecticide made before three to four true leaves.

Thrips are small (1-2 mm) insects that feed on plant epidermis and mesophyll cells. In the southeastern U.S., the primary species of thrips found on cotton is the tobacco thrips, Frankliniella fusca Hinds; other species that occasionally appear on cotton include Frankliniella occidentalis Pergande, Frankliniella tritici Fitch, Thrips tabaci Lindeman, and Neohydatothrips variabilis Beach (Osekre et al., 2009; Reay-Jones et al., 2017; Samler, 2012; Stewart, 2013; Toews et al., 2010; Wang et al., 2018). Wind facilitates dispersal of adult thrips and can result in thrips being present in fields before seedlings are planted (Lewis, 1997; Smith et al., 2016). Immediately after cotton seedlings emerge, adult thrips begin feeding on and ovipositing in cotyledons and true leaves, both of which injure plant cells. The juvenile thrips that hatch from these eggs cause the most injury to seedlings, feeding on hosts for up to 13 days before pupation (Cook et al., 2011). Cotton is susceptible to thrips feeding injury during the seedling stage, and injury can have a lasting impact on cotton growth and development. Thrips feeding ruptures mesophyll cells and removes sap and nutrients from newly developing leaves (Cook et al., 2011). This causes growing leaves to appear crinkled, sometimes with a silvery sheen caused when air fills punctures in the cells (Reed and Reineke, 1990; Telford and Hopkins, 1957). Feeding on apical meristems can cause forked growing points and, in severe infestations, seedling death and stand reductions (Cook et al., 2011; Gaines, 1934; Ritchie et al., 2007). Seedlings are most vulnerable to thrips until four true leaves. Once this stage is reached, cotton growth rate increases, and plants can outpace thrips feeding (Krob et al., 2022; Lei and Wilson, 2004). Thrips have the potential to decrease cotton lint yield by more than 112 kg ha<sup>-1</sup> and can delay the maturity of plants (Bourland et al., 1992; Copeland et al., 2016; Davis et al., 1966; Dunnam and Clark, 1937; Gaines, 1934; Layton and Reed, 2002; Lentz and Austin, 1994; Micinski et al., 1990; Race, 1961).

Management of thrips has relied on insecticides either applied as seed treatments, an at-plant liquid or granular formulation in the furrow, and/or as a foliar spray after seedlings emerge (Allen et al., 2018; Cook et al., 2011; Williams, 2011, 2012, 2013, 2014). Seed treatments are standard practice in the southeastern U.S. and typically last two to three weeks compared to in-furrow applications that can last four to five weeks (D'Ambrosio et al., 2018). The level of protection varies, depending on the timing of planting to the time and magnitude of infestation, soil types, and environmental conditions that influence cotton growth (Copeland et al., 2016; Kerns et al., 2019). Common insecticides for seed-based applications include neonicotinoids (IRAC classification group 4a), which are an insecticidal class containing seven active ingredients, of which only imidacloprid and thiamethoxam have been registered for the management of thrips in cotton and are applied in the methods mentioned above (North et al., 2018). Since their registration in the 1990s, these chemicals have been widely used in cotton production as one of the primary and most effective tools for the control of early infestations of thrips (Allen et al., 2018; Douglas and Tooker, 2015; Williams, 2011, 2012, 2013, 2014) due to their low application rates and broad-spectrum activity (Elbert et al., 2008).

At the time of this study, reports of thrips resistance to neonicotinoid seed treatments were an emerging concern in the southeastern and midsouthern U.S. production regions (Darnell-Crumpton et al., 2018; Huseth et al., 2016, 2018; Kerns et al., 2019). This prompted research into the efficacy of active ingredients and combinations thereof that could be used to manage thrips (Adams et al., 2013; Catchot et al., 2013; D'Ambrosio et al., 2020; Kerns et al., 2019; Stewart, 2013). Insecticides have been evaluated primarily for their ability to reduce adult and immature populations. Reductions in oviposition also have been observed for some active ingredients (Huseth et al., 2017) but have been evaluated only

under field conditions in one study (D'Ambrosio et al., 2018). The objective of this study was to quantify the effects of three insecticide seed treatments on thrips oviposition, number of adult and immature thrips, injury ratings, and plant dry biomass 42 days after planting.

#### **MATERIALS AND METHODS**

Experimental Design and Treatments. The experiment was performed at the Prattville Agricultural Research Unit (PARU), in Prattville, AL, in 2016. Phytogen 390 W3FE (Corteva Agriscience, Indianapolis, IN) was planted in a reduced tillage field at a standard planting rate of 9.8 seeds m-1. The experiment was organized in a split-plot design with four replications. Planting date was designated as the main plot factor (six levels), and insecticide seed treatment was the subplot factor (four levels). Replications of subplots were arranged in randomized complete blocks within each planting date. For each planting date, 16 plots (4 rows wide and 9.14 m long each) were separated by a 2.13-m alley on each end. Cotton seed received one of four treatments: 1) control, no insecticide seed treatment; 2) Avicta Elite (0.375 mg imidacloprid + 0.340 mg thiamethoxam + 0.150 mg abamectin per seed; Syngenta, Greensboro, NC); 3) Avicta Duo (0.150 mg abamectin + 0.375 mg thiamethoxam per seed; Syngenta, Greensboro, NC); or 4) Aeris (0.375 mg imidacloprid + 0.375 mg thiodicarb per seed; Bayer CropScience, Research Triangle Park, NC). Instead of replicating this study across years, experimental replications were performed across six planting dates during the same growing season. Studies show the risk of cotton injury fluctuates within a window of several weeks each spring as thrips disperse from overwintering hosts, and growing conditions for cotton change seedling susceptibility (Chappell et al., 2020; Kerns et al., 2019). Therefore, evaluating treatment effects across the cotton planting window, while thrips are dispersing, should capture similar effects to what a multi-year study should. The Thrips Infestation Predictor (Chappell et al., 2020) was used to select a range of planting dates when adult thrips flights responsible for crop colonization events were predicted to occur. This allowed a higher confidence that the intensity of natural thrips infestations would be high enough to test for treatment differences. Replicating across a window of time during the same growing season also allowed us to test for

treatment effects across variable infestation levels (Kerns, 2018).

Data Collection. Dispersing thrips were monitored in plots using yellow sticky traps (12.7  $\times$  7.6 cm; Great Lakes IPM, Inc., Vestaburg, MI). One trap was placed in the middle of each plot, for a total of 16 per planting date. Traps were suspended just above the cotton canopy with a binder clip attached to a 1.2-m pole; the height of the binder clip was adjusted using clothespins to support the bottom of the binder clip. Traps were collected and replaced weekly, starting from 20 April through 14 June, for 9 wk. Collected traps were sandwiched between the front and back of clear plastic sheet protectors with 27.94 × 21.59-cm inserts (Avery Products Corporation, Brea, CA) and stored at 4 °C, as in Chitturi et al. (2018). The total number of adult thrips on each trap was counted under a dissecting microscope and recorded.

On each sampling date, 10 random seedlings in the middle two rows of each plot were selected and rated for injury. Leaf injury was visually estimated using a 1 to 5 injury scale (1= minor, visible thrips injury; 5 = seedling death) (modified from Vineyard et al., 2017; Graham and Stewart, 2018). Cotton seedlings were destructively sampled for eggs and immature and adult thrips weekly, starting at the cotyledon stage and ending at four to five true leaves (Table 1). During each sampling date, 10 random, but similarly sized (i.e., same leaf stage), seedlings per plot (five seedlings each from the first and fourth rows) were cut with scissors at the soil line and quickly placed into a wide-mouthed glass jar with soapy water (approximately 1.3 mL L<sup>-1</sup> of dish soap).

In the laboratory, seedlings were dipped into soapy water several times to dislodge immature and adult thrips and then saved in 0.94-L plastic storage bags in a -20 °C freezer until thrips eggs were

Table 1. Planting dates and sampling dates for thrips in cotton at various growth stages in Prattville, AL, during 2016

Planting Date	Growth Stage During Sampling Dates				
	Cotyledon-1 true leaf	2–3 true leaf	4–5 true leaf		
April 11	4/26	5/3	5/10		
April 18	5/3	5/10	5/17		
April 25	5/3	5/10	5/17		
May 2	5/24	5/31	6/7		
May 9	5/24	5/31	6/7		
May 16	5/31	6/7	6/14		

stained and counted (below). Thrips were collected from soapy water onto filter paper using vacuum filtration; a Buchner funnel lined with a 9-cm filter paper (Cytiva, Marlborough, MA) sat on top of a 1.0-L flask that was connected to a vacuum pump. Filter papers containing thrips were sandwiched between two pieces of Saran Wrap and stored at -20 °C until thrips counts were performed using a dissecting microscope. The total number of immature and adult thrips per 10 seedlings was recorded for each plot.

To count eggs, plants were defrosted, and a lactophenol acid fuchsin staining solution (prepared as in Riley et al., 2007) was brought to a boil in a fume hood and removed from a hot plate. Immediately, leaves were slowly dipped in this solution and allowed to sit until the solution cooled (3-5 h). Leaves were removed from the main stem, rinsed with warm water to remove excess stain, and transferred to 100 × 15-mm (Thermo Fisher Scientific, Waltham, MA) petri dishes containing warm water (Parrella and Robb, 1982). Dishes were then placed under a Leica M60 dissecting microscope (Leica Microsystems Inc., Deerfield, IL) using 40x magnification. The total number of eggs in each leaf was recorded, and leaf totals were summed to determine the total number of eggs per plant.

Finally, dry biomass was measured to assess any potential delays in plant growth among the treatments. Forty-two days after planting, five random plants per plot were collected from rows one and four by cutting plants at the soil line and placing plants into paper bags. Plants were dried at 60 °C in an oven (Grieve SA-400, Round Lake, IL) until the weight was constant for two consecutive reading days, and then weighed using a scale (model ML6001E, Mettler-Toledo, LLC, Columbus, OH). Total biomass in grams per five plants was recorded.

Statistical Analyses. Data were analyzed using PROC GLIMMIX in SAS (Version 9.4; SAS Institute, Inc., Cary, NC). The factors sampling date, planting date, insecticide treatment, interaction between insecticide treatment and sampling date, and interaction between insecticide and planting date were modeled as fixed effects; whereas the interaction among block, sampling date, and planting date was included as a random effect. Sampling dates were standardized across planting dates using growth stage as a categorical variable instead of calendar/sampling dates (Table 1). Data from injury rating were modeled using the multinomial distribution and clogit link function. For all other

dependent variables (number of dispersing thrips, immature thrips, adult thrips, thrips eggs, and plant dry weight) the negative binomial distribution was used, and when interaction terms were significant between fixed effects, the SLICE option was used to examine simple effects while controlling for the other factor. Tukey's LSD test ( $p \le 0.05$ ) was used as post hoc mean comparisons.

#### RESULTS AND DISCUSSION

The magnitude of thrips dispersal over the nine consecutive weeks of this study was evaluated from 401 sticky traps that captured a total of 25,931 adult thrips. The numbers of dispersing adult thrips at this location were significantly different among weeks

(Table 2). The highest number of dispersing thrips was observed during the 24 May and 31 May collections, whereas the lowest numbers occurred the first week of sampling on 26 April (Fig. 1A). Numbers of thrips were statistically different but generally similar all other weeks.

Insecticide treatments did not significantly reduce adult populations collected from cotton compared with the control, but adult captures were overall very low. A total of 2,710 cotton seedlings were sampled to determine the number of adult thrips per 10 plants (Fig. 1B). Only planting date and insecticide significantly affected the number of adult thrips on cotton seedlings (Table 2). Although there was a trend for an increasing number of adult thrips from cotyledon to five true leaves, these values were not significantly dif-

Table 2. Type III effects for separate analyses conducted on thrips dispersal, number of adult thrips, number of thrips eggs, number of immature thrips, injury ratings, and plant dry weights collected from seedling cotton in Prattville, AL, during 2016

Dependent Variables <sup>z</sup>	Independent Variables	Num DF	Den DF	F	P
Thrips dispersal	Sample Date	8	389	139.15	<.0001
Adult thrips	Sample Date	2	60	3.01	0.0567
	Planting Date	5	60	7.52	<.0001
	Insecticide	3	179	3.31	0.0215
	Sample Date*Insecticide	6	179	0.93	0.4763
	Planting Date*Insecticide	15	179	0.84	0.6299
Oviposition	Sample Date	2	162	48.88	<.0001
	Planting Date	5	162	90.18	<.0001
	Insecticide	3	486	23.5	<.0001
	Sample Date*Insecticide	6	486	10.47	<.0001
	Planting Date*Insecticide	15	486	3.95	<.0001
Immature thrips	Sample Date	2	60	65.38	<.0001
	Planting Date	5	60	22.77	<.0001
	Insecticide	3	179	23.19	<.0001
	Sample Date*Insecticide	6	179	4.52	0.0003
	Planting Date*Insecticide	15	179	1.63	0.0700
Injury rating	Sample Date	2	60	3.3	0.0436
	Planting Date	5	60	8.73	<.0001
	Insecticide	3	2621	174.43	<.0001
	Sample Date*Insecticide	6	2621	13.8	<.0001
	Planting Date*Insecticide	15	2621	9.83	<.0001
Dry weight	Planting Date	5	18	5.08	0.0045
	Insecticide	3	54	8.21	0.0001
	Planting Date*Insecticide	15	54	1.68	0.0831

Abbreviations: Num DF: numerator degrees of freedom; Den DF: denominator degrees of freedom; F: F statistic; P: p-value associated with the F-statistic.

<sup>&</sup>lt;sup>z</sup>Response data were fitted with different models: adult thrips, oviposition, immature thrips, dry weights (negative binomial), and injury ratings (multinomial).

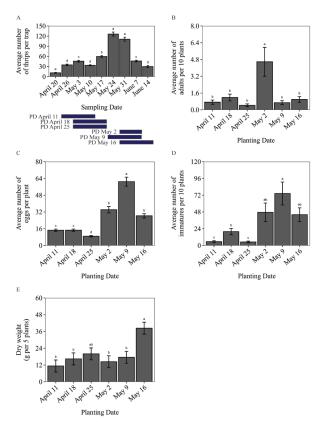


Figure 1. The average  $\pm$ standard error of the number of dispersing thrips captured on sticky traps on each sample date (A), number of adult thrips collected from 10 cotton seedlings (B), number of thrips eggs per cotton seedling (C), number of immature thrips collected from 10 cotton seedlings (D), and dry weight of five cotton plants collected 42 days after planting (E). For panel A, numbers are averaged across all plots and the bars under the x-axis indicate the weeks data were collected from plots established during each planting date (PD) from emergence until seedlings reached 4–5 true leaves. For panels B–E, numbers are averaged across all sampling dates and insecticide treatments. Means with the same letter are not significantly different ( $p \le 0.05$ , Tukey's LSD).

ferent (Fig. 2A). The number of adult thrips collected from cotton was significantly higher during the fourth planting date (Fig. 1B), whereas all other planting dates were similar and under 1.2 individuals per 10 seedlings. Differences in the number of adult thrips between insecticide treated cotton seedlings were only observed between Aeris and Avicta Elite; no significant differences were observed between the control and insecticide treatments. The efficacy of insecticides did not change between sampling dates (Table 2).

The number of eggs found on control seedlings over time shared a similar trend with thrips dispersal data because increases in oviposition occurred when numbers of dispersing adult thrips increased. Thrips

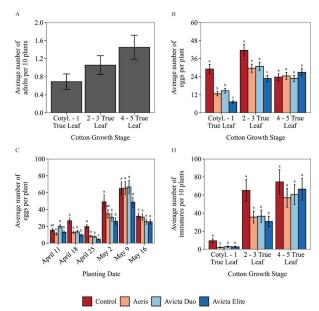


Figure 2. The average  $\pm$  standard error of the number of adult thrips collected from 10 cotton seedlings (A), number of thrips eggs per cotton seedling (B and C), and the number of immature thrips collected from 10 cotton seedlings (D). For panel A, numbers are averaged across all planting dates and insecticide treatments. For panels B and D, numbers are averaged across all planting dates, and means are compared by growth stage or plant date, respectively. For panel C, numbers are averaged across all sampling dates and are means compared by growth stage. Means with the same letter are not significantly different ( $p \le 0.05$ , Tukey's LSD).

eggs were counted from a total of 680 cotton seedlings. Sampling date, planting date, insecticide, and their interactions significantly affected oviposition of thrips on cotton seedlings (Table 2). The number of eggs per plant averaged across treatments increased from 14.1  $\pm$  0.87 on cotyledon/one true leaf stage cotton to 30.5  $\pm$  1.56 on seedlings with two to three true leaves, then decreased to  $24.5 \pm 1.27$  on seedlings with four to five true leaves. Oviposition remained relatively low during the first three planting dates, increased significantly on the fourth and sixth planting dates, and was highest on the fifth planting date (Fig. 1C). When averaged across sampling dates, insecticides significantly reduced oviposition on cotton. Avicta Elite (16.4  $\pm$  0.94) was the most efficacious, with a reduction of 46.3% compared with the control (30.6  $\pm$  1.69). Aeris (21.0  $\pm$  1.18) and Avicta Duo  $(21.8 \pm 1.21)$  reduced oviposition by 30.1%compared with the control. The efficacy of insecticides to reduce oviposition was significantly affected by the sample date and planting date (Table 2). Reduced oviposition was observed in the first two sampling dates on 26 April and 3 May, but after seedlings reached the four to five true leaf stage, insecticides failed to reduce oviposition (Fig. 2B). All insecticides statistically or numerically reduced oviposition during the first four planting dates (Fig. 2C), but these reductions were not observed for the last two planting dates, which experienced the highest infestations of dispersing adult thrips (Fig. 1A). No consistent differences in oviposition reduction were observed among treatments across sampling dates (Fig. 2C).

A total of 2,710 cotton seedlings were sampled to determine the average number of immature thrips per 10 plants. Sampling date, planting date, insecticide, and the interaction between insecticide and sampling date significantly affected the number of immature thrips on cotton seedlings (Table 2). The highest number of immature thrips occurred during the fourth, fifth, and sixth planting dates, whereas the lowest numbers were seen during the first, second, and third planting dates (Fig. 1D). All insecticide treatments were equally effective in reducing immature thrips counts, but, similar to oviposition, reductions in immatures associated with insecticides were only observed during the first two sampling dates of 26 April and 3 May and not on seedlings at four to five true leaves (Fig. 2D). The number of immature thrips averaged across planting dates and insecticides significantly increased from 3.6  $\pm$  0.75 at cotyledon/one true leaf to  $40.3 \pm 6.33$  at two to three true leaves, and to  $64.4 \pm 10.06$  on seedlings with four to five true leaves; the increase in immatures from the second sampling date to the third was not significantly different. When averaged across planting and sampling dates, insecticides significantly decreased the number of immatures on cotton by 50.8% (between  $16.6 \pm 2.09$  to  $18.5 \pm 2.28$ ), compared with the control  $(35.8 \pm 4.14)$ .

The degree of injury ratings caused by thrips to seedlings was significantly affected by sampling date, planting date, insecticide, and their interactions (Table 2). Higher injury ratings were generally observed on planting dates in which the highest number of eggs and immatures were observed on cotton (Figs. 1C, 1D, 3). The injury ratings observed in this study ranged from 1.0 to 5.0, but an injury rating of 2.0 was most common across all sampling dates (Fig. 3A), planting dates (except May 9th, score 3.0) (Fig. 3B), and in plots receiving insecticide seed treatments (Fig. 3C). An injury rating of 3.0 was most common for non-treated cotton seedlings, and they were between 3.3 and 10.0 times more likely to have higher plant injury ratings than insecticide treated seedlings.

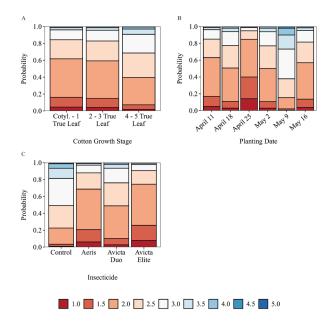


Figure 3. The cumulative probability of seedlings receiving injury ratings (on a scale of one to five, one indicating little to no injury, and five indicating seedling death) analyzed by sampling dates (A), planting dates (B), and insecticide treatment (C).

Aeris and Avicta Elite reduced plant injury more than Avicta Duo (Fig. 3C). When analyzed by planting date, seedlings from the third planting date had lower injury ratings than all other planting dates; the odds of injury from the other planting dates were between 3.3 (first planting date) and 30.6 (fifth planting date) times greater (Fig. 3B). When analyzed by growth stage, differences in plant injury from cotyledon to three true leaves stages were small and not significant. Seedling injury at four to five true leaves was 2.2 to 2.5 times greater than the odds of injury to seedlings during cotyledon to three true leaves, possibly because time would allow more eggs to hatch and increase immature thrips feeding on the true leaves present. Previous research also showed neonicotinoid seed treatment concentration and efficacy decrease as cotton seedlings age (D'Ambrosio et al., 2018; Zhang et al., 2011). Environmental factors likely contributed to the variation in efficacy to reduce injury among treatments because cotton growing conditions changed across planting dates, which affected how long seedlings remained in susceptible growth stages (Cook et al., 2011; Copeland et al., 2016; Kerns et al., 2019; Krob et al., 2022; Lei and Wilson, 2004; Micinski et al., 1990; Parajulee et al., 2006; Slosser, 1993). Although faster cotton growth occurred during planting dates four and six, compared with planting dates one and two, the injury ratings were similar among them. There were fewer thrips immatures during planting dates one and two, but the cotton remained in a susceptible growth stage for a longer time. Seedlings grew faster during planting dates four and six, but they were exposed to a higher number of immature thrips (Figs. 1D, 3B). Greater thrips populations likely increased injury ratings on plants from the fifth planting date, regardless of the favorable cotton growing conditions.

Overall, seedlings treated with insecticides had higher biomass than non-treated seedlings, but no significant differences in biomass were observed among treatments for each planting date (Table 2). A total of 480 cotton seedlings were collected to determine dry weight (grams per five seedlings) 42 days after planting. The highest biomass was observed the sixth week of planting  $(38.4 \pm 4.27)$ , and it was significantly different from seedlings in the first, second, fourth, and fifth planting dates (Table 2, Fig. 1E). When averaged across planting dates and sampling dates, all insecticides significantly increased biomass from 25.3 to 45.2%, compared with non-treated plots, in which the average biomass was  $15.8 \pm 1.96$ ; no significant differences between insecticides were observed. There was no significant interaction between planting date and insecticide treatment. Environment could have influenced these results because the most favorable cotton growing conditions occurred during the sixth planting date (personal observation).

In this year and location, F. fusca was the most dominant species collected from cotton, representing more than 90% of the adults and immature thrips collected and identified using PCR-based methods (Wang et al., 2018). This local population of F. fusca has been characterized as exhibiting resistance to both imidacloprid and thiamethoxam, as demonstrated by bioassays conducted in 2014 and 2015 (Huseth et al., 2016). It would be valuable to better understand if the reduction in the efficacy of insecticides to reduce oviposition and immatures on the fourth, fifth, and sixth planting dates (Fig. 1A) was due to the high population size of thrips or resistant individuals in the population. Although a decrease in susceptibility to these two active ingredients could have influenced these results, measuring susceptibility was beyond the scope of this study, and it is difficult to determine any effects due to resistance. Many of the changes in efficacy to reduce oviposition and immature populations in this study appear to have been influenced by their interactions with planting date and sampling date (Table 2). Changes in population numbers, injury, and plant growth of cotton across planting dates have been well documented in other studies in the U.S. Cotton Belt (Cook et al., 2011; Copeland et al., 2016; Kerns et al., 2019; Micinski et al., 1990; Parajulee et al., 2006; Slosser, 1993) and the interactions between planting date, cotton seedling growth, and thrips population pressure are key factors in determining risk of cotton injury (Chappell et al., 2020).

Knowledge about how the risk of plant injury changes across cotton planting dates and the use of neonicotinoid seed treatments continue to be important components of thrips management in early season cotton. Our results contribute new information about the efficacy of seed treatments containing multiple active ingredients to reduce thrips oviposition and immature life stages. Insecticide use has increased in response to the emergence of neonicotinoid resistant populations (Huseth et al., 2016), which includes using seed treatments with multiple active ingredients to improve efficacy of thrips management. The three insecticide seed treatments used in this study were generally effective at reducing the average number of thrips eggs and immatures on cotton seedlings across sampling dates and planting dates, but efficacy was variable across the six planting dates of this trial. This was related to fluctuations in thrips population sizes, as adults dispersed from senescing weeds in the landscape to newly planted crops. This supports previous research showing cotton injury is a function of environmental factors that influence thrips population size and cotton growth stage (Chappell et al., 2020; Copeland et al. 2016; Kerns, 2018). Furthermore, resistance to neonicotinoids in local populations may have also contributed to decreases in efficacy when thrips populations increased, but additional work is needed to better understand how changes in thrips population size versus changes in insecticide susceptibility influence product efficacy to reduce plant injury.

### **ACKNOWLEDGMENTS**

We thank staff at the Prattville Agricultural Research Station for providing assistance with maintenance of field plots, and Charles Ray, Ian Ingram, Emily Ingram, Adrian Pekarcik, Sara Rains, Wade Turner, and Emanii Striker for their assistance with plot maintenance, sample collection, slide mounting, and insect identification. We also thank the Agricultural Research Service, U.S. Department of

Agriculture (Agreement No. 58-6010-0-011), Cotton Incorporated (Agreement No. 15-979), and Alabama Hatch Project 1006125 for their financial support of this project.

#### REFERENCES

- Adams, B., A. Catchot, J. Gore, D. Cook, F. Musser, and D. Dodds. 2013. Impact of planting date and varietal maturity on tarnished plant bug (Hemiptera: Miridae) in Cotton. J. Econ. Entomol. 106:2378–2383. https://doi.org/10.1603/EC12330.
- Allen, K.C., R.G. Luttrell, T.W. Sappington, L.S. Hesler, and S.K. Papiernik. 2018. Frequency and abundance of selected early-season insect pests of cotton. J. Integr. Pest Manag. 9:20. https://doi.org/10.1093/jipm/pmy010.
- Bourland, F.M., D.M. Oosterhuis, and N.P. Tugwell. 1992. Concept for monitoring the growth and development of cotton plants using main-stem node counts. J. Prod. Agric. 5:532–538. https://doi.org/10.2134/jpa1992.0532.
- Catchot, A., J. Gore, and D. Cook. 2013. Tobacco thrips in mid-south resistant to thiamethoxam [Online]. Available at https://www.mississippi-crops.com/2013/11/07/tobacco-thrips-in-mid-south-resistant-to-thiamethoxam (verified 22 May 2025).
- Chappell, T.M., R.V. Ward, K.T. DePolt, P.M. Roberts, J.K. Greene, and G.G. Kennedy. 2020. Cotton thrips infestation predictor: a practical tool for predicting tobacco thrips (*Frankliniella fusca*) infestation of cotton seedlings in the south-eastern United States. Pest Manage. Sci. 76:4018–4028. https://doi.org/10.1002/ps.5954.
- Chitturi, A., K. Conner, E.J. Sikora, and A.L. Jacobson. 2018. Monitoring seasonal distribution of thrips vectors of soybean vein necrosis virus in Alabama soybeans. J. Econ. Entomol. 111:2562–2569. https://doi.org/10.1093/ jee/toy237.
- Cook, D.R., and M. Threet. 2022. 2021 Cotton insect losses estimates. p. 145–200 *In* Proc. Beltwide Cotton Conf., San Antonio, TX. 4-6 Jan. 2022. Natl. Cotton Counc. Am., Memphis, TN.
- Cook, D., A. Herbert, D.S. Akin, and J. Reed. 2011. Biology, crop injury, and management of thrips (Thysanoptera: Thripidae) infesting cotton seedlings in the United States. J. Integr. Pest Manag. 2:1–9. https://doi.org/10.1603/IPM10024.
- Copeland, J.D., D.M. Dodds, A.L. Catchot, J. Gore, and D.G. Wilson. 2016. Evaluation of pre herbicides and seed treatment on thrips infestation and cotton growth, development, and yield. Agron. J. 108:2355–2364. https://doi.org/10.2134/agronj2016.03.0151.

- D'Ambrosio, D.A., A.S. Huseth, and G.G. Kennedy. 2018. Temporal efficacy of neonicotinoid seed treatments against *Frankliniella fusca* on cotton. Pest Manage. Sci. 74:2110–2115. https://doi.org/10.1002/ps.4907.
- D'Ambrosio, D.A., G.G. Kennedy, and A.S. Huseth. 2020. Frankliniella fusca and Frankliniella occidentalis response to thrips-active Cry51Aa2.834\_16 Bt cotton with and without neonicotinoid seed treatment. Crop Prot. 129:105042. https://doi.org/10.1016/j.cropro.2019.105042.
- Darnell-Crumpton, C., A.L. Catchot, D.R. Cook, J. Gore,
   D.M. Dodds, S.C. Morsello, and F.R. Musser. 2018.
   Neonicotinoid insecticide resistance in tobacco thrips
   (Thysanoptera: Thripidae) of Mississippi. J. Econ. Entomol. 111:2824–2830. https://doi.org/10.1093/jee/toy298.
- Davis, J.W., W.C. Watkins, C.B. Cowan, R.L. Ridgway, and D.A. Lindquist. 1966. Control of several cotton pests with systemic insecticides. J. Econ. Entomol. 59:159– 162. https://doi.org/10.1093/jee/59.1.159.
- Douglas, M.R., and J.F. Tooker. 2015. Large-scale deployment of seed treatments has driven rapid increase in use of neonicotinoid insecticides and preemptive pest management in U.S. Field Crops. Environ. Sci. Technol. 49:5088–5097. https://doi.org/10.1021/es506141g.
- Dunnam, E.W., and J.C. Clark. 1937. Thrips damage to cotton. J. Econ. Entomol. 30:855–857. https://doi.org/10.1093/jee/30.6.855.
- Elbert, A., M. Haas, B. Springer, W. Thielert, and R. Nauen. 2008. Applied aspects of neonicotinoid uses in crop protection. Pest Manage. Sci. 64:1099–1105. https://doi.org/10.1002/ps.1616.
- Gaines, J.C. 1934. A preliminary study of thrips on seedling cotton with special reference to the population, migration, and injury. J. Econ. Entomol. 27:740–743. https://doi.org/10.1093/jee/27.4.740.
- Graham, S.H., and S.D. Stewart. 2018. Field study investigating Cry51Aa2.834\_16 in cotton for control of thrips (Thysanoptera: Thripidae) and tarnished plant bugs (Hemiptera: Miridae). J. Econ. Entomol. https://doi.org/10.1093/jee/toy250.
- Huseth, A.S., T.M. Chappell, A. Chitturi, A.L. Jacobson, and G.G. Kennedy. 2018. Insecticide resistance signals negative consequences of widespread neonicotinoid use on multiple field crops in the U.S. Cotton Belt. Environ. Sci. Technol. 52:2314–2322. https://doi.org/10.1021/acs. est.7b06015.

- Huseth, A.S., T.M. Chappell, K. Langdon, S.C. Morsello, S. Martin, J.K. Greene, A. Herbert, A.L. Jacobson, F.P. Reay-Jones, T. Reed, D.D. Reisig, P.M. Roberts, R. Smith, and G.G. Kennedy. 2016. Frankliniella fusca resistance to neonicotinoid insecticides: an emerging challenge for cotton pest management in the eastern United States. Pest Manage. Sci. 72:1934–1945. https:// doi.org/10.1002/ps.4232.
- Huseth, A.S., D.A. D'Ambrosio, and G.G. Kennedy. 2017. Responses of neonicotinoid resistant and susceptible Frankliniella fusca life stages to multiple insecticide groups in cotton. Pest Manage. Sci. 73:2118–2130. https://doi.org/10.1002/ps.4590.
- Kerns, C.D. 2018. The effects of planting date and varietal susceptibility of *Gossypium hirsutum* L. on thrips. MS Thesis. Clemson Univ., Clemson, SC.
- Kerns, C.D., J.K. Greene, F.P.F. Reay-Jones, and W.C. Bridges. 2019. Effects of planting date on thrips (Thysanoptera: Thripidae) in cotton. J. Econ. Entomol. 112:699–707. https://doi.org/10.1093/jee/toy398.
- Krob, J.L., S.D. Stewart, and S.A. Brown. 2022. Effects of plant density, seed spacing, and seed treatment on thrips injury to cotton. Crop Prot. 161:106059. https://doi. org/10.1016/j.cropro.2022.106059.
- Layton, B., and J.T. Reed. 2002. Biology and control of thrips on seedling cotton. Publ. 2302 (POD-3-20). Mississippi State Univ. Ext. Serv., Mississippi State, MS.
- Lei, T.T., and L.J. Wilson. 2004. Recovery of leaf area through accelerated shoot ontogeny in thrips-damaged cotton seedlings. Ann. Bot. (Oxford, U. K.) 94:179–186. https://doi.org/10.1093/aob/mch125.
- Lentz, G.L., and N.B. Austin. 1994. Control of early season thrips on cotton with Gaucho (NTN 33893) seed treatments. p. 847–849 *In Proc. Beltwide Cotton Conf.*, San Diego, CA. 5-8 Jan. 1994. Natl. Cotton Counc. Am., Memphis, TN.
- Lewis, T. 1997. Thrips as Crop Pests. CAB International, University Press, Cambridge, UK.
- Micinski, S., P.D. Colyer, K.T. Nguyen, and K.L. Koonce. 1990. Effects of planting date and early-season pest control on yield in cotton. J. Prod. Agric. 3:597–602. https://doi.org/10.2134/jpa1990.0597.
- North, J.H., J. Gore, A.L. Catchot, S.D. Stewart, G.M. Lorenz, F.R. Musser, D.R. Cook, D.L. Kerns, and D.M. Dodds. 2018. Value of neonicotinoid insecticide seed treatments in mid-south cotton (*Gossypium hirsutum* [Malvales: Malvaceae]) production systems. J. Econ. Entomol. 111:10–15. https://doi.org/10.1093/jee/tox324.

- Osekre, E.A., D.L. Wright, J.J. Marois, and J. Funderburk. 2009. Population dynamics and within-plant distribution of *Frankliniella* spp. thrips (Thysanoptera: Thripidae) in cotton. Environ. Entomol. 38:1205–1210. https://doi.org/10.1603/022.038.0430.
- Parajulee, M.N., R.B. Shrestha, and J.F. Leser. 2006.
  Influence of tillage, planting date, and Bt cultivar on seasonal abundance and within-plant distribution patterns of thrips and cotton fleahoppers in cotton. Int. J. Pest Manage. 52:249–260. https://doi.org/10.1080/09670870600774240.
- Parrella, M.P., and K.L. Robb. 1982. Technique for staining eggs of *Liriomyza trifolii* within chrysanthemum, celery, and tomato leaves. J. Econ. Entomol. 75:383–384. https://doi.org/10.1093/jee/75.2.383.
- Race, S.R. 1961. Early-season thrips control on cotton in New Mexico. J. Econ. Entomol. 54:974–976. https://doi.org/10.1093/jee/54.5.974.
- Reay-Jones, F.P.F., J.K. Greene, D.A. Herbert, A.L. Jacobson, G.G. Kennedy, D.D. Reisig, and P.M. Roberts. 2017. Within-plant distribution and dynamics of thrips species (Thysanoptera: Thripidae) in cotton. J. Econ. Entomol. 110:1563–1575. https://doi.org/10.1093/jee/tox131.
- Reed, J.T., and J. Reinecke. 1990. Western flower thrips on cotton: plant damage and mite predation preliminary observations. p. 309–310 *In* Proc. Beltwide Cotton Conf., Las Vegas, NV. 10-13 Jan. 1990. Natl. Cotton Counc. Am., Memphis, TN.
- Riley, D.G., A. Chitturi, and A.N. Sparks. 2007. Does natural deposition of pine pollen affect the ovipositional behavior of *Frankliniella occidentalis* and *Frankliniella fusca*? Entomol. Exp. Appl. 124:133–141. https://doi.org/10.1111/j.1570-7458.2007.00561.x.
- Ritchie, G.L., C.W. Bednarz, P.H. Jost, and S.M. Brown. 2007. Cotton growth and development. Publ. B1252. University of Georgia, College of Agricultural and Environmental Sciences, Athens, GA.
- Samler, J.A. 2012. Abundance and species diversity of thrips (Thysanoptera: Thripidae) in cotton, soybean, and peanut in Southeast Virginia, and evaluation of cyantraniliprole for thrips management. MS Thesis. Virginia Polytechnic Institute and State University, Blacksburg, VA.
- Slosser, J.E. 1993. Influence of planting date and insecticide treatment on insect pest abundance and damage in dryland cotton. J. Econ. Entomol. 86:1213–1222. https://doi.org/10.1093/jee/86.4.1213.
- Smith, E.A., E.J. Shields, and B.A. Nault. 2016. Impact of abiotic factors on onion thrips (Thysanoptera: Thripidae) aerial dispersal in an onion ecosystem. Environ. Entomol. 45:1115–1122. https://doi.org/10.1093/ee/nvw089.

- Stewart, S. 2013. Tobacco thrips resistance to Cruiser [Online]. Available at https://news.utcrops.com/2013/11/tobacco-thrips-resistance-to-cruiser/ (verified 22 May 2025).
- Telford A.D., and L. Hopkins. 1957. Arizona cotton pests. Arizona Agric. Expt. Sta. Bull. 86.
- Toews, M.D., R.S. Tubbs, D.Q. Wann, and D. Sullivan. 2010. Thrips (Thysanoptera: Thripidae) mitigation in seedling cotton using strip tillage and winter cover crops. Pest Manage. Sci. 66:1089–1095. https://doi.org/10.1002/ps.1983.
- United States Department of Agriculture, National Agricultural Statistics Service [USDA NASS]. 2023.
  Upland cotton production 2022 summary [Online].
  Available at https://www.nass.usda.gov/Statistics\_by\_Subject/result.php?16E617FB-D9FE-3945-A104-8CE4A1287B28&sector=CROPS&group=FIELD%20CROPS&comm=COTTON (verified 22 May 2025).
- Vineyard, C.J., H. Kelly, L. Steckel, and S. Stewart. 2017. Potential interaction of pre-emergence herbicides and the efficacy of insecticide and fungicide seed treatments in cotton. J. Cotton Sci. 21:284–295. https://doi. org/10.56454/BTIN7241.
- Wang, H., G.G. Kennedy, F.P.F. Reay-Jones, D.D. Reisig, M.D. Toews, P.M. Roberts, D.A. Herbert, S. Taylor, A.L. Jacobson, and J.K. Greene. 2018. Molecular identification of thrips species infesting cotton in the Southeastern United States. J. Econ. Entomol. 111:892–898. https:// doi.org/10.1093/jee/toy036.
- Williams, M.R. 2011. Cotton insect losses—2010. p. 896–940
  In Proc. Beltwide Cotton Conf., Atlanta, GA. 4-7 Jan. 2011. Natl. Cotton Counc. Am., Memphis, TN.
- Williams, M.R. 2012. Cotton insect losses estimates—2011.
  p. 1001–1012 *In* Proc. Beltwide Cotton Conf., Orlando,
  FL. 3-6 Jan. 2012. Natl. Cotton Counc. Am., Memphis,
  TN.
- Williams, M.R. 2013. Cotton insect losses—2012. p. 546–586
  In Proc. Beltwide Cotton Conf., San Antonio, TX. 7-10
  Jan. 2013. Natl. Cotton Counc. Am., Memphis, TN.
- Williams, M.R. 2014. Cotton insect losses estimates—2013. p. 798–812 *In Proc. Beltwide Cotton Conf.*, New Orleans, LA. 6-8 Jan. 2014. Natl. Cotton Counc. Am., Memphis, TN.
- Zhang, L., S.M. Greenberg, Y. Zhang, and T. Liu. 2011. Effectiveness of thiamethoxam and imidacloprid seed treatments against *Bemisia tabaci* (Hemiptera: Aleyrodidae) on cotton. Pest Manage. Sci. 67:226–232.