EVALUATING INTERACTIONS BETWEEN ONION THRIPS AND ASSOCIATED PLANT PATHOGENS AND METHODS TO IMPROVE MANAGEMENT IN ONION

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EVALUATING INTERACTION BETWEEN ONION THRIPS AND ASSOCIATED PLANT PATHOGENS AND METHODS TO IMPROVE ONION PRODUCTION

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The success of integrated pest management (IPM) programs relies on pest biology and ecology, and the tactics to manage damage caused by those pests. In onion production, onion thrips (*Thrips tabaci*) and their associated plant pathogens are primary constraints to crop production. The objectives of this work were to 1) evaluate IPM tactics to reduce damage caused by onion thrips and associated plant pathogens, 2) further characterize the relationship between onion thrips and iris yellow spot virus (IYSV), and 3) determine the success of extension programming to increase grower adoption of insecticide resistance management and IPM tactics for onion thrips. In chapters 1 and 2, a combination of different IPM tactics (host plant resistance, fertility regimes, and insecticide programs) was evaluated to reduce onion thrips densities and severity of associated plant diseases, namely IYS disease and bacterial bulb rot. In these trials, fertility regime did not consistently affect onion thrips densities, IYS disease, or bacterial bulb rot. Insecticide use consistently reduced onion thrips densities, IYSV disease, but not the incidence of bacterial bulb rot. Additionally, a thrips resistant cultivar ('Avalon') experienced lower thrips densities and IYS disease severity but suffered from greater levels of bacterial rot. In chapter 3, there is discussion about the potential role that different habitats within the onion production system may have as a source for IYSV inoculum (viruliferous onion thrips). In these trials, transplanted onion fields accounted for 49-51% of the total estimated numbers of viruliferous thrips, which may generate inoculum for late-season outbreaks of IYSV. In chapter 4, I describe a laboratory study that evaluated the effect of IYSV infection on the reproduction and mortality of adult onion thrips. Reproduction did not differ between groups, but viruliferous adults lived 1-6 days longer than non-viruliferous adults. Lastly, in chapter 5, the effectiveness of an extension-based program was investigated to increase grower adoption of IPM tactics for onion thrips. The program was successful, and growers increased use of insecticide class rotation by 31% and use of the action threshold by 44%. These studies improved our understanding about the biology and ecology of onion thrips and IYSV and described methods that will improve onion thrips management in onion.

BIOGRAPHICAL SKETCH

Ashley Leach was born in Traverse city, MI to Michele Ruman and Steven Leach. She attended Michigan State University and earned a B.S. degree in Entomology from Lyman Briggs College. After graduation, she was employed as an agricultural scout and consultant which inspired her to participate in applied agricultural research. She initially enrolled as a M.S. student in the Entomology department at Cornell University in 2014 but has since continued her education as a PhD student.

To my family, who will never read this.

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INTRODUCTION

Onion, *Allium cepa*, production

Onion, *Allium cepa*, belongs to the Alliaceae family. Notable cultivated representatives include chives (*Allium schoenoprasum*), green onion (*Allium chinense*), onion (*Allium cepa*), leek (*Allium ampeloprasum*) and garlic (*Allium sativum*). However, onion is the most widely grown *Allium* species with over 9.2 million acres planted globally (Brewster 2008).

Approximately 60 million tons of onions are produced annually in over 170 countries. Major producers of onions worldwide include China, India, and the United States (FAO 2018). China accounts for approximately 26% of the world's dry bulb onion production, yielding over 22 million tons every year. India ranks second in onion production, and the United States third. Within the United States, onion is the third largest fresh vegetable in production. Approximately 125,000 acres of onions are planted annually, amounting to 6.2 billion tons each year. Onions are grown commercially in 20 different states. Washington, Oregon, Idaho, California, Georgia and New York lead the nation in production and acreage. Nationally, the value of these onions varies depending on demand and yield, but the annual farm gate value in the United States is approximately $1 billion. Value added products can amount to values exceeding $6 billion (NASS 2014).

Onion, *A. cepa*, production in New York

New York is a top ten producer of onions in the United States, with over 7,000 acres of onion planted each year. The average farm gate value of onions in New York ranges from 40 to 60 million dollars. The onion industry accounts for about 10% of the state's vegetable production value (NASS 2014). Onions grown in New York are long-day cultivars and most are yellow cooking onions, but red and sweet onion cultivars are planted as well. Onions are grown throughout the state, most acreage is concentrated in Orange, Oswego, Genesee, Cayuga, Madison, Wayne, Steuben, Yates, and Orleans counties. Onion acreage in New York is grown on fertile 'muck' soils in in these counties. 'Muck' soil is unique as it consists of 20-80% organic matter (NRCS, 2016; Wilson and Townsend, 1931), which provides substantial amounts of nitrogen to supplement plant growth throughout the growing season (Haynes 2012). The remaining acreage is grown on sandy loam type soils throughout the state.

Onion is a cool-season crop that grows best at temperatures ranging from 55ºF to 75ºF (Brewster 2008). In New York, onions are planted in the spring from late March to early May. Most of the acreage is direct seeded, but approximately 20% of onions are transplanted. Most onions that are transplanted arrive from the southwestern United States and are planted using hand labor. Other onion transplants are sourced from local greenhouses and transplanted as plugs or a new system called PlantTape. Although, more expensive than the seeds, transplants offer a premium price for their earlier harvest and large size grades, thus offsetting their initial cost. Direct-seeded onions are typically planted using a precision seeder.

Onion Thrips (*Thrips tabaci* Lindeman) as a major pest of onion

As a significant crop in the United States and specifically within New York, onions are intensively managed in order to produce a high-value crop (Brewster et al. 2008). Onions face a range of arthropod pests throughout the growing season. In New York, there are many arthropods that can significantly damage onions: onion maggot and seedcorn maggot (*Delia antiqua* and *Delia platura*, respectively); bulb mites (*Rhizoglyphus spp*.); cutworms (*Noctua spp*.); and onion thrips (*Thrips tabaci*) (New York State IPM Program 2018). Seedcorn maggot and onion maggot feed directly on the roots, stem and base of leaves of onion plants as they emerge (Schwartz and Mohan 2008). Although less prevalent in New York, bulb mites also feed and damage onion bulbs (Schwartz and Mohan 2008). Onion mites are found underground and feed within the developing onion bulb. Occasionally, cutworms can impact onion stand establishment in the spring by feeding on onion seedlings (Brewster 2008, Nault and Shelton 2015). A new invasive pest of Allium crops, the Allium leafminer (*Phytomyza gymnostoma* Loew), may negatively impact onion, but no economic losses have been reported yet. Although the aforementioned pests can sporadically cause significant damage to onion, none cause the consistent and widespread damage associated with onion thrips, *Thrips tabaci* Lindeman. Due to their short-generation time, high reproductive capacity, parthenogenic nature, small size, and ability to transmit and worsen plant pathogens, onion thrips pose the most significant threat to onion growers in New York (Diaz-Montano et al. 2011).

Onion thrips is an indirect pest of onion, and feeds on onion leaves as well as transmitting or worsening plant diseases. Onion thrips remove sap from plant cells using rasping-sucking mouthparts (Lewis 1997). Onion thrips feed on mesophyll cells, which ultimately deplete chlorophyll in leaves (Boateng et al. 2014), and damaged onion leaves appear white and silvery. Feeding by onion thrips reduces photosynthetic potential of the onion plant, thereby reducing bulb size. Onion plants are most vulnerable to thrips feeding during the prebulbing and bulbing stages, when plants are most rapidly growing (Brewster 2008 and Gill et al. 2015).Extensive thrips damage can contribute to yield losses between 30-36% in New York (Nault and Huseth 2016).

Onion thrips can transmit bacterial pathogens and worsen fungal diseases in onion (Schwartz and Mohan 2008). Onion thrips feeding creates openings that allow for the introduction of pathogens that cause diseases such as purple blotch (*Alternaria porri*) and bacterial center rot (*Pantoea ananatis, P.agglomerans*) (Cartwright et al. 1995; Dutta et al. 2014). These pathogens have substantial impact on onion plants and associated diseases reduce yields by 39 to 75% (Schwartz and Mohan 2008; Stiver 1997). *Alternaria porri*, which causes purple blotch enters leaves via stomatal openings or wounds, thereby infecting the plant. Diseased leaves form white lesions that gradually turn purple and coalesce (Schwartz and Mohan 2008). Cartwright et al. (1995) reported that higher densities of onion thrips increase the incidence of purple blotch disease. Bacterial bulb rot is caused by *Pantoea spp.* infects the plant and compromises the internal integrity of the bulb and reducing marketable yield. Studies have also indicated that onion thrips may play a role in bacterial bulb rot (Grode et al. 2019; Grode et al. 2016; Dutta et al. 2014).

Onion thrips is also the primary vector of the economically significant iris yellow spot virus (IYSV) (Peribunyaviridae), a tospovirus that infects *Allium* species (Bag et al. 2015). Initially described on irises in the Netherlands, the virus has been reported to infect over 30 plant species worldwide (Cortês et al. 1998). IYSV is now globally widespread on onion and has been found in Asia, South America, Europe, North America, Africa, Australia, and New Zealand (Bag et al. 2015; Gent et al. 2006). Once infected, onion leaves will form straw-colored lesions that can coalesce and girdle onion plants. IYSV can spread quickly, and if plants are infected early in the season, growers can face yield losses upwards of 60% in the United States. In 2003, Colorado onion growers were unable to control the virus and lost an estimated 5 million dollars in yield (Gent et al. 2006). Although sporadic in New York, the virus still poses a considerable threat if it becomes widespread. Currently, there are no onion cultivars resistant to the virus (Bag et al. 2015). Onion thrips management is the primary mean to control IYS disease in onions (Gent et al. 2006).

Previous studies have indicated that IYSV is not transmitted via seed or though mechanical inoculation in onion (Bag et al. 2015; Kritzman et al. 2001). Common to most tospoviruses, IYSV is acquired and transmitted principally by thrips. IYSV is circulative and propagative within its thrips vector, and adults transmit the virus until death (Whitfield et al. 2005). Tospoviruses are acquired only by first and second instars (Whitfield et al. 2005): acquisition rates decrease as larvae mature (Ullman et al. 2002) as a mid-gut barrier develops, which prevents viral infection (Nagata et al. 1999). Contrary to studies in other pathosystems with other thrips species, onion thrips are not believed to be affected by IYSV infection (Birthia et al.2013; Inoue et al.2010). Inoue et al.(2010) reported that onion thrips mortality, development, and reproduction were not significantly different between groups feeding on IYSV-infected and healthy (non-infected) tissue. Similarly, Birithia et al. (2013) found no significant difference in the mortality rates between onion thrips feeding on IYSV-infected tissue and healthy tissue. However, these studies only monitored impacts on onion thrips for short periods and conducted the studies using less preferred hosts. It is not known how IYSV impacts the lifespan of onion thrips adults nor the numbers of their progeny produced.

Adult onion thrips dispersal and IYSV patterns in onion fields may provide insight into IYSV epidemics (Ullman et al.2002), especially in New York. Previous studies have identified three different sources of inoculum, which host both onion thrips and IYSV within onion production systems including, onion plants imported from the southwestern US and then transplanted elsewhere, certain weed species, and volunteer onions in cull piles (Gent et al. 2006; Hsu et al. 2010; Hsu et al. 2011; Schwartz et al. 2014; Smith et al. 2011). However, the relative contribution of habitats containing these various sources of IYSV and its vector on IYSV epidemics in onion agroecosystems is not known.

Biology and ecology of onion thrips (*T. tabaci*)

Thrips are taxonomically classified in the order Thysanoptera, which describes small, fringe-winged insects with elongate bodies. This order is divided into suborders Tubulifera, those thrips that lay their eggs outside of plant tissue and have two pupal stages, and Terebrantia, thrips that insert eggs into plant tissue and have only one pupal stage (Mound and Kibby 1998). Onion thrips are organized within the Terebrantia suborder, and then further placed into family Thripidae, genus *Thrips*. Onion thrips can be distinguished from other North American species, as *Thrips tabaci* has light grayish brown ocellar crescents, a medially reticulated metanotum with no sensilla, and a well-developed posteromarginal comb with prominent microtrichia on the eighth tergite (Nakahara 1994) (Figure 1a).

Onion thrips are found throughout North America, but likely originated from the Mediterranean (Lewis 1997). Compared to other thrips species, onion thrips has a wide host range and feed on more than 300 plant species (Diaz-Montano et al.2012). Onion thrips is a pest of many vegetable crops including cabbage, carrots, cucumber, and onion. However, as their namesake indicates, onion thrips prefer onion foliage (Lewis 1997; Doderline et al.1993).

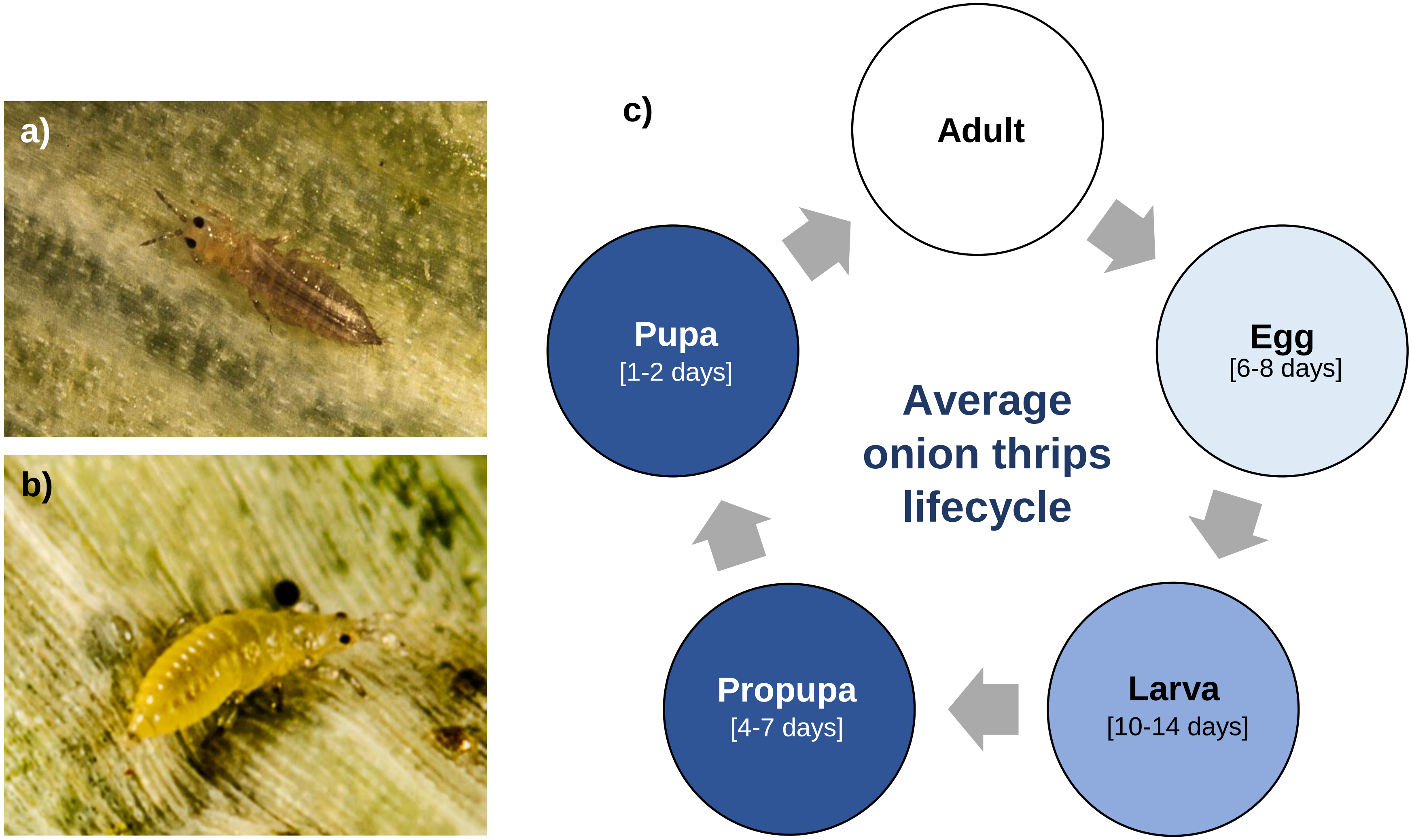

Figure 1: Onion thrips (*Thrips tabaci*) **a)** adult and **b)** larva. **c)** average life cycle of onion thrips.

Onion thrips are hemimetablous insects and typically produce six to eight generations in New York. Typically, three to four of those generations occur on onion, and remaining generations occur on other crops or weedy hosts (Hoffman et al.1996). Onion thrips have five described life stages: egg, larva (2 instars), propupa, pupa, and adult. Females insert eggs into leaf tissue which are small (0.2 mm long), white, and rounded. Eggs mature in six to eight days. After hatching from the egg, first and second instars will feed on plant tissue. Larvae are 0.3-0.4 mm long and range in color from light yellow to brown. Onion thrips are thigmotactic and preferentially aggregate within the "pseudostem" of the onion where thrips are protected by leaf folds. Larvae develop in approximately ten to fourteen days (Gill et al.2015; Lewis 1997) (Figure 1b). Following the second instar, onions thrips enter propupal and pupal stages. Propupae and pupae look similar to a large second instar, with the exception of wing pad development. These life stages do not feed and live in the soil. Adults emerge in five to nine days. Adult onion thrips are 1.3 mm long, yellow to brown in color, and have fringed wings. Adults, unlike the larvae, are highly mobile and will disperse throughout the onion field and surrounding area (Smith et al.2015). Similar to the first and second instars, adults will feed on foliage until pupation (Lewis 1997) (Figure 1b). Onion thrips adults overwinter in the soil within onion fields or under plant debris in weedy areas adjacent to onion fields (Larentzaki et al. 2007 and North and Shelton 1986). Onion thrips can complete one generation in fifteen to twenty days. In cooler, wetter summers, onion thrips populations are low and sometimes only reach three generations during the growing season. Conversely, hot and dry summers support onion thrips populations that grow quickly and can reach eight or more generations (Gill et al.2015).

In New York, onion thrips typically emerge in early spring and feed on weed hosts (Smith et al. 2011). In May to early June, onion thrips will begin to colonize transplanted onions, and then secondarily move to direct-seeded onion fields (Hsu et al.2010). Thrips densities build throughout the onion growing season and reach economic thresholds in mid-June to early July. Onions need to be managed for onion thrips from June until mid to late August. After onions are harvested, onion thrips will move onto weedy hosts and continue to reproduce until they overwinter.

Onion thrips exhibit three modes of reproduction: thelytoky (unfertilized eggs yield female progeny), arrhenotoky (unfertilized eggs yield males and fertilized eggs females), and deuterotoky (unfertilized eggs yield both males and females) (Nault et al.2006). However, there is some variability in these reproductive modes, as some thelytokous females can occasionally produce a male and some arrenotokous females will produce a female (Jacobson et al.2016). In New York, onion thrips primarily reproduce via thelytoky, but arrhenotokous populations also can be found.

Management of onion thrips in onion

Integrated pest management (IPM) is the primary paradigm to manage pests in agriculture, including onion thrips in onion. IPM combines management tactics with the aim of reducing pest damage, maximizing crop yield and limiting negative off-target effects (Pedigo et al.1986; Stern et al.1959). In the United States, management of onion thrips is necessary to produce a marketable onion crop. Chemical, cultural, and biological management options are available to control onion thrips (Gill et al.2015). These tactics differ in their efficacy and efficiency but can be combined to optimize the management of onion thrips.

There are many biological control options to suppress and manage thrips. Onion thrips have been controlled by entomopathogenic nematodes, entomopathogenic fungi, parasitoids and predators (Brodsgaard and Hansen 1992, Maniania et al. 2003, Wu et al. 2013). Biological control of onion thrips has been documented within the onion agroecosystem in New York. Fok et al. (2014) found eight predator species in small-scale and large-scale onion fields, including predaceous thrips species, *Aeolothrips fasciatus*. Due to the application of many broad-spectrum insecticides and lack of natural resources or reservoirs, natural enemies are either killed and the use of the biological control is generally not supported in many commercial onion fields.

Cultural control tactics like manipulating row spacing and planting rate impact onion thrips densities. Malik et al. (2003) found that onion plants spaced 40 centimeters apart had approximately 60% fewer thrips than plants spaced 20 centimeters. These results were consistent, regardless of insecticide treatment. However, due to the high value of muck soil in New York, many onion growers are unwilling to reduce their planting rate or row spacing. Buckland et al. (2013) also showed that rotating onions with corn reduced densities of onion thrips in onion. While crop rotation may reduce pest pressure and diversify onion thrips management programs, the reduction in pest pressure does not equate to the revenue lost by taking onion out of production in those fields. Therefore, crop rotation is also not a current viable option for many commercial onion growers.

Mulching is another cultural control that has shown to reduce onion thrips densities. Straw mulches interfere with the pupation and emergence of onion thrips, and can reduce thrips densities 45-54% when compared to onion fields with bare soil (Larentzaki et al. 2008 and Schwartz et al. 2009). However, mulches are expensive and labor intensive, which limits their application in commercial onion production.

Commercial onion growers, including those in New York, would benefit from an IPM program that optimized the usage of insecticide regimes, fertilizer application, and resistant onion cultivars to control onion thrips. These tactics can be easily incorporated into current onion growing practices. Most importantly, they offer promise to reducing onion thrips densities quickly and can offer season-long control.

Effect of insecticide regime, nitrogen rate, and onion cultivar on onion thrips management in onion

Insecticide program

Insecticides are unparalleled in their ease of use, efficacy, and efficiency when compared to cultural and biological management tactics alone. As such, growers have relied on insecticide applications to control onion thrips. However, insecticides are often over-used, which can lead to resistance issues and environmental contamination. Therefore, insecticide application should be harmonized with economic threshold information and pest biology (Pedigo et al.1986).

Insecticides are the most common management tactic to control onion thrips in commercial onion production in the United States. Both synthetic and botanical insecticides have been identified to control onion thrips in onion. Synthetic insecticides from chemical classes including anthranilic diamides, avermectins, spinosyns, and tetramic acids are currently most effective against onion thrips in large-scale commercially produced onion fields (Table 1). Some registered products from classes:

organophosphates, carbamates, pyrethroids and neonicotinoids are no longer effective against onion thrips for a variety of reasons (Table 1). Although not used widely in the United States due to their lower efficacy, some botanical insecticides made from various fruits, seeds, and latex infusions have been applied to control onion thrips (Malik et al.2003). In New York, abamectin, spirotetramat, spinetoram, and cyantraniliprole are the most effective chemistries to reduce and maintain low levels of onion thrips (Nault and Hessney 2010, 2011). Because many of the newer insecticides are either systemic or translaminar in nature, co-application with a penetrating surfactant significantly improves the level of onion thrips control (Nault et al. 2013).

Table 1: Insecticides commonly applied in onion to control onion thrips

Chemical class	Mode of action (IRAC)	Active ingredient	Trade name(s)	Comments
Anthranilic diamides	Ryanodine receptors modulators	Cyantranilporole	Exirel	
Avermectin	Chloride channel activators	Abamectin	Agri-mek	
Carbamate	Acetylocholinesterase inhibitors	Methomyl	Lannate	Resistance has developed in New York (Shelton et al.2006).
Organophosphate		Methyl-parathion	Penncap-M	
Neonicotinoid	Nicotinic acetylcholine receptor agonists	Acetamiprid	Assail	
Pyrethroid	Sodium channel modulators	Permethrin	Pounce, Ambush	
		Cypermethrin	Ammo	
		Zeta-cypermethrin	Mustang	
		Lambda-Cyhalothrin	Warrior with Zeon	Resistance has developed in New York (Shelton et al.2006).
Spinosyns	Nicotonic acetylcholine receptor allosteric activators	Spinosad	Entrust, Success	Organic product.
		Spineotram	Radiant SC	Apply when onion thrips are at their highest (Nault and Shelton 2010). Residual activity of <7 days (Nault et al.2012).
Tetramic acid	Inhibitors of acetyl coa carboxylase	Spirotetramat	Movento	Apply initially to control onion thrips for longer pest suppression. Works best on larval thrips. Residual activity of <10 days (Nault et al.2012).

Insecticide resistance in onion thrips populations in onion have developed in multiple regions throughout the world. Globally, onion thrips have become resistant to organophosphates, carbamates, and pyrethroids (Herron et al. 2008, MacIntyre Allen 2004, Martin et al. 2003). In New York, onion thrips have developed resistance to lambda-cyhalothrin and methomyl (Shelton et al. 2006). In 2005, New York onion growers were unable to control onion thrips populations due largely in part to insecticide resistance. Onion yields were reduced by more than 30% when compared to previous years. Thus, many formerly effective insecticides are no long effective against onion thrips (Diaz-Montano et al.2008).

Surveys in New York have shown that many growers are already utilizing some IPM and insecticide resistance management (IRM) tactics. Currently, approximately 52% of onion growers rotated between insecticide classes, and only 40% of those growers used an action threshold. However, many use the most efficacious chemical in accordance with onion thrips biology. Almost 95% of respondents begin their thrips control with applications of spirotetramat (Movento) and finish with applications of spinetoram (Radiant) later in the growing season. About 88% of growers stated that they either personally scout their fields or hire a professional crop advisor. More than half of respondents also apply fewer insecticides now when compared to 15 years ago. While many growers have reduced their insecticide applications, approximately 50% have reported that they have not changed the number of insecticides applications to control onion thrips (Nault and Hoepting 2015).

Fertility regime

Nutrient input can increase the incidence and population growth of insect pests, thus increasing damage to a crop (Altieri and Nicholls 2003). Nutrient input imposes a complex balance between increasing the overall attractiveness of a plant to insect pests and providing the necessary nutrients for plant growth and immunity. Therefore, nutrient input should be considered when developing a pest management program.

Nitrogen and phosphorus are essential nutrients for onion production. If levels of either nutrient are too low during the “bulbing” phase, onions will be significantly undersized and yield a lower profit. Studies have shown that current rates of nitrogen application often exceed the necessary amount for onion growth and development (Hoepting 2009; Brewster et al.2008). Reducing rates of phosphorus and nitrogen fertilizer could have economic and environmental benefits in addition to decreasing thrips populations and limiting insecticide applications.

Previous studies have shown that onion thrips populations in onion decrease between 23-70% with decreased rates of nitrogen (Buckland et al.2013; Malik et al.2009), while Chen et al. (2004) found 2.3 times fewer thrips (*Frankliniella spp.)* on plants on impatiens flowers (*Impatiens wallerana*) when fertigated with lower rates of phosphorus (1.28 mM P vs. 0.32 mM P.). Thus, a reduction in nitrogen and phosphorus fertilizers also may be an effective cultural control tactic for onion thrips in onion.

In New York, onions grown with high rates of nitrogen had significantly more larval and adult onion thrips than those with lower nitrogen rates (Hsu et al.2010). The benefits of reducing nitrogen are numerous as growers can save money, limit surface runoff and ground water pollution, and reduce onion thrips densities. Surveys show that approximately 32% of New York onion growers have already reduced their nitrogen rates based on earlier work done by Hsu et al. (2010). Those who have reduced their nitrogen input have decreased rates by an average of 56%. Current grower nitrogen application rates are approximately 98 lbs per acre, compared to a former rate of 212.5 lbs per acre (Nault and Hoepting 2015). Thus, nitrogen fertilizer programs in commercial onion production can be modified to prevent onion thrips population growth while still optimizing onion growth. The current recommendation is 125 lbs/acre, but more research is needed to confirm that lower rates (e.g., 98 lbs/acre) will not only reduce thrips populations but will not reduce bulb yield.

Cultivar resistance

Cultivar resistance is one of the most important components of pest management, since it can eliminate or drastically reduce the need for control measures. Further, if used appropriately, resistant cultivars can provide durable control in the long term (Mundt 2014). The application of this technique can also be harmonized with other pest management strategies like biological, chemical, and cultural control tactics to further reduce insect pest populations.

Currently, there are no onion cultivars completely resistant to onion thrips feeding, but some cultivars have shown to withstand feeding with low to zero effect on yield. Although the variables conferring tolerance to onion thrips feeding is still being examined, plant color, waxiness, and architecture appear most important. Onion cultivars with blue-green leaves rather than yellow-green ones are more resistant to onion thrips feeding (Diaz-Montano et al. 2012 and Boateng et al.2014). Glossy and semi-glossy onion cultivars have lower amounts of onion thrips as well. Damon et al.

(2014) suggested that the higher amount of hentriacontanone-16 in the epicuticular waxes may be responsible for the increased density of thrips.

Additionally, cultivars with an open architecture, in which onion leaves are angled away from each other, tend to have lower levels of thrips feeding damage. This is likely due to thigmotactic nature of thrips, which preferentially colonize leaves that are close together. Boateng et al.(2014) also found that cultivars with fewer leaves and earlier harvest had lower amounts of thrips. The reason for this finding could be two-fold, as thrips may be less attracted to plants with fewer leaves, and thus have lower rates of colonization. Secondarily, the earlier maturing onions will have lower amounts of thrips, as they are removed from the field before onion thrips populations build to high densities in late summer and early fall.

While some thrips-resistant cultivars, such as cv. ‘Advantage ‘, are available to onion growers, the 120 days or more to harvest makes these cultivars less attractive to growers in New York. Onions that mature over 120 days from planting are not ideally suited for the Northeast climate because they may not properly mature in time before harvest in late summer/early fall. More research is needed to identify thrips-resistant cultivars that mature in less than 120 days from planting for New York.

Growers in New York have expressed interest in incorporating thrips-resistant cultivars into their onion production (Nault and Hoepting 2015), but research on these cultivars in the Northeast is limited. Most growers plant thrips-susceptible onion cultivars including “Braddock”, “Red wing”, and “Milestone” (Nault and Hoepting 2015). These cultivars have known thrips-susceptible characteristics such as blue-green leaves and high levels epicuticular wax. Virtually none of the onion growers who were surveyed in 2015 planned on planting a thrips-resistant cultivar (Nault and Hoepting 2015); presumably because there is a risk that they would not mature in time to be harvested. One grower transplanted some fields with a partially thrips-resistant onion, cv. 'Delgado', which is a late maturing cultivar. Transplanting this cultivar, rather than direct seeding, truncates the maturity period and ensures that the crop will be harvested in time.

Grower adoption of IPM and IRM programs

The effectiveness of integrated pest management and insecticide resistance management is largely predicated on grower decision and compliance (Siegfried et al.1998; Hurley and Mitchell 2008). However, our understanding of the implementation and adoption of IRM and related IPM practices is relatively limited (Peshin and Karla 2009). Growers tend to adopt practices that are not risky, easy to implement, and save money (Peshin 2013; Peshin and Karla 2009; Trumble 1998), which can put some IPM and IRM practices at a disadvantage because many are complicated and time-consuming to implement. Consequently, the adoption of some IPM practices have been slow to progress as compared with other agricultural technologies (Zalucki et al.2009; Kogan and Bajwa 1999). Further research is needed to identify those methodologies that can successfully increase adoption of IRM and related IPM tactics to mitigate the onset of insecticide resistance.

Poor insecticide resistance management has resulted in pest control failures worldwide. In onion production systems, insecticide resistance in onion thrips populations has led to significant yield losses (Herron et al. 2008, MacIntyre-Allen et al. 2005, Martin et al. 2003, Shelton et al. 2003, 2006). Previous research has identified two pest management practices that should mitigate insecticide resistance and control onion thrips populations; using an action threshold (Nault and Huseth 2016; Nault and Shelton 2010) and following an insecticide sequence that rotates insecticide classes (Nault 2015; Nault and Shelton 2010). The use of thresholds is an important component to insecticide resistance management programs (IRAC International 2016). In onion production, an action threshold of one thrips per leaf has been effective in controlling thrips populations without reducing yield, which can reduce insecticide applications between 30-50% (Nault and Huseth 2016). Recent research has identified effective thrips management using season-long sequences of insecticides belonging to different classes that are rotated (Nault 2015; Nault and Shelton 2010). Onion thrips typically complete a generation in 14-21 days on onion (Jamieson et al.2012), thus no more than two consecutive sprays of the same mode of action is recommended. These two approaches should reduce exposure of an insecticide to multiple generations of onion thrips and slow the potential onset of insecticide resistance (Espinosa et al.2002; Immaraju et al.1992; Immaraju et al.1990).

Research goals and justification of future research

Onion production is challenged by a number of pests, however onion thrips and their associated plant pathogens, iris yellow spot disease and bacterial bulb rots, are primary constraints to production. The goal of my research is to provide additional information on the ecology of onion thrips and their associated plant pathogens, as well as describe methods to improve onion thrips management in onion. The objectives of this work were to 1) evaluate IPM tactics to manage onion thrips and associated plant diseases, 2) further characterize the relationship between onion thrips and iris yellow spot virus

(IYSV), and 3) determine the success of extension programming to increase grower adoption of insecticide resistance management and IPM tactics for onion thrips.

Justification for future research to improve onion thrips management

Onion thrips management will not likely be sustainable if growers rely solely on insecticide applications. In New York, fertility regime, cultivar selection, and insecticide program offer the most potential for reducing onion thrips densities while producing a profitable onion crop. Excess nitrogen can be associated with higher levels of both larval and adult onion thrips (Hsu et al.2011). Nitrogen is often applied in excess in agricultural areas throughout New York State, but pollution of nitrogen can be especially severe in intensively managed vegetable crops (Hoepting 2009). Therefore, reducing nitrogen rates could reduce thrips densities, resulting in lower levels of damage. In addition, some onion cultivars (cv. ‘Avalon’, ‘Advantage’) have tolerance towards thrips feeding, and incur little to no yield loss (Nault 2014). These cultivars are not being planted commercially in New York but have potential to reduce the impact of onion thrips on bulb yield. Lastly, insecticides are a significant tool for onion growers, but are best optimized when applied according to specific economic thresholds and are rotated to best minimize insecticide resistance (Nault and Shelton 2010). This insecticide program will not only control onion thrips but should preserve effective insecticides for future use.

Objectives:

The objectives of this study were to 1) examine the effect of an integrated pest management program that combined thrips management techniques (reduced fertility regimes, thrips-resistant onion cultivar, and an action threshold-based insecticide program) on onion thrips densities and onion yield, and 2) examine the effect of this integrated pest management program on plant diseases associated with onion thrips.

Hypothesis:

In this study, the following hypotheses were tested: a reduced fertility regime paired with an action-threshold based insecticide program would provide effective thrips and disease management without compromising marketable yield. Moreover, the greatest reduction in agrichemical input (lower amount of fertilizer and fewer insecticide applications) in the cultivar with the highest resistance to thrips.

Justification for future research evaluating onion thrips interactions with iris yellow spot virus (IYSV)

Prevalence of viruliferous thrips in different onion habitats

In New York state, IYSV is a sporadic, significant disease of onions. Further research is needed to understand which habitat(s) may be most influential in fostering IYSV epidemics in New York onion fields. Previous research has indicated that transplanted onion fields, weedy areas near onion fields and onion cull piles may be important sources of IYSV inoculum as these habitats contain both IYSV host plants and its vector, onion thrips. Further research should address the abundance of viruliferous onion thrips captured in these habitats early to mid-season to determine which habitat may be most likely to contribute to IYSV epidemics later in the season.

Objective:

The objective of this study was to determine which habitat (transplanted onion fields, weedy areas, onion cull piles) contributed the greatest to early-season viruliferous thrips populations.

Hypothesis:

In this study, the following hypothesis was tested: onion fields established with transplants imported from the southwestern US would generate the greatest numbers of viruliferous thrips early to mid-season compared to the other habitats. In this case, secondary spread of IYSV would occur into adjacent onion fields (especially direct-seeded) and weedy habitats because onion thrips adults are known to disperse from maturing transplanted onion fields in search of other suitable habitats later in the season (Smith et al. 2017).

Effect of IYSV infection on mortality and reproduction of adult thrips

In addition to understanding the broad implication of thrips abundance on the epidemiology of IYSV, further research is needed to address the impact of IYSV infection of onion thrips biology. Previous research has indicated that thrips are impacted by tospovirus infections (Stafford-Banks et al.2014, Shrestha et al.2012, Stafford et al.2011, Stumpf and Kennedy 2005, and DeAngelis et al.1993), and many of these studies suggest that thrips are positively benefitted by a tospovirus infection. Previous literature indicates that IYSV infection does not impact the reproduction or mortality of thrips when monitored for the first week after eclosion (Birthia et al.2013; Inoue et al.2010); however, no studies have examined the long-term effects of IYSV on the lifespan and numbers of progeny produced by onion thrips.

Objective:

The objective of this study was to examine the effect of IYSV infection on the lifespan and fecundity of onion thrips.

Hypothesis:

In this study, the following hypothesis was tested: viruliferous thrips would positively benefit from IYSV infection by living longer and producing more offspring.

Justification for future research to advance grower adoption of insecticide resistance management and integrated pest management practices

Integrated pest management and insecticide resistance management are core paradigms guiding modern pest management. However, our understanding about why growers may or may not adopt these management practices is lacking. Specifically, further research is needed to identify those methodologies that can successfully increase adoption of IRM and related IPM tactics to mitigate the onset of insecticide resistance. In onion production, onion thrips have a high capacity for developing resistance due to their short-generation time, high reproductive rates and polyphagy. Furthermore, current survey data indicate a low grower adoption of IRM practices. Only 52% of growers claimed to rotate between insecticide classes and even fewer (40%) used an action threshold.

Objective:

The objective of this study was to improve the adoption of research-based IRM tactics (use of the action threshold and insecticide class rotation) to manage onion thrips in onion

Hypothesis:

In this study, the following hypotheses were tested: the use of action thresholds and rotation of insecticide classes would increase over the three-year program, and conservatively estimated that growers would collectively increase their use of both tactics by 10% annually. Furthermore, growers who adopted these tactics would positively benefit by applying fewer insecticide applications, reducing total insecticide cost, while successfully managing onion thrips infestations.

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CHAPTER 1

EVALUATING INTEGRATED PEST MANAGEMENT TACTICS FOR ONION THRIPS AND PATHOGENS THEY TRANSMIT TO ONION

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Abstract

Onion thrips (Thrips tabaci) is a significant pest of onion worldwide, causing both direct and indirect damage to the crop. Integrated pest management of onion thrips should improve profitability and sustainability of onion production. Promising management approaches include reducing nitrogen application rates, using thrips-resistant cultivars and implementing action threshold-based insecticide programs. However, the impact of these integrated pest management approaches on thrips densities and damage, crop yield, and thrips-associated plant diseases like iris yellow spot (IYS) (caused by Iris yellow spot virus) and bacterial center rot (caused by Pantoea agglomerans and P. ananatis) remains largely unknown. In a two-year field trial in New York, combinations of varying levels of nitrogen applied at planting (67, 101 and 140 kg ha-1) and different insecticide programs (standard weekly insecticide program and action threshold-based insecticide program) were evaluated for onion thrips management in onion cultivars that had moderate resistance ('Avalon'), low resistance ('Delgado') and no resistance ('Bradley') to onion thrips. Results indicated that regardless of cultivar, nitrogen did not affect larval thrips densities, onion yields, IYS or bacterial center rot. Across all cultivars, insecticide use (both programs) significantly reduced larval thrips densities and damage, IYS severity and incidence, and increased onion yield. Insecticide use did not consistently affect the incidence of bacterial center rot. Both insecticide programs reduced onion thrips larval densities by 60-81% relative to the untreated control, but the action threshold-based application program used 2.8 fewer applications than the standard program. 'Avalon' had low thrips densities and IYS disease but required the same number of insecticide applications as 'Bradley'. Onion yields in both insecticide programs were statistically similar in both years, and bulb weights averaged 10-54%

more than those in the untreated control. Our results indicated that growers can reduce nitrogen levels at planting and insecticide use without compromising control of either onion thrips or IYS disease or onion bulb yields.

Key Words: *Thrips tabaci*, *Allium cepa*, Iris yellow spot virus, bacterial center rot, host-plant resistance, nitrogen fertilizer

Highlights

- Multiple tactics did not improve thrips control compared with insecticide use only.
- An action threshold-based program required 3 fewer applications than the standard.
- A similar number of insecticide applications were required for all cultivars.
- Nitrogen rates at planting did not impact onion thrips management or bulb yields.
- Thrips control reduced Iris yellow spot disease but did not affect bacterial rot.

1. Introduction

Integrated insect pest management often addresses the direct effects of insect feeding damage to a crop but does not consider the impacts of indirect effects such as those arising from plant pathogen-insect interactions. Onion thrips (*Thrips tabaci* Lindeman) is an example that exacts both direct and indirect effects on its host, onion (Allium cepa L.). Severe infestations of onion thrips can account for substantial onion yield reductions if unmanaged (Fournier et al., 1995; Nault and Shelton 2008; Rueda et al., 2007). As a direct pest, onion thrips adults and larvae feed on onion leaves, decreasing photosynthetic potential, and thereby reducing bulb size (Boateng et al., 2014; Lewis 1997). Damage to leaves also induces physiological stress, which accelerates leaf senescence (Kendall and Bjostad 1990; Levy and Kedar 1970) and reduces bulb size. Bulb weight losses as high as 60% have been reported from onion thrips damage (Rueda et al., 2007), which tends to vary based on location, severity of infestation, and environmental stress (see review by Gill et al., 2015).

As an indirect pest of onion, onion thrips has been associated with an array of viral, bacterial and fungal plant pathogens (Dutta et al., 2014; Gent et al., 2006; McKenzie et al., 1993). Onion thrips is the principal vector of the economically significant *Tospovirus*, Iris yellow spot virus (IYSV) (genus Tospovirus, family Bunyaviridae), which reduces size and quality of bulbs (Gent et al., 2004; Muñoz et al., 2014). Under severe IYSV infections, lesions coalesce and girdle onion leaves, thus inhibiting onion development. Damage by IYSV can range from insignificant to complete yield loss (i.e., no marketable bulbs) (Gent et al., 2006). In a study conducted in Colorado, annual incidence of IYSV varied from 6 to 73% over three years (Gent et al., 2004). Similarly, in New York, Hsu et al., (2010) reported varying IYSV incidences from 0% to 97% over two years. Managing the vector, onion thrips, is currently the primary means for reducing IYSV incidence and severity (Bag et al., 2015; Gent et al., 2006).

Onion thrips also transmits bacterial center rot pathogens (Pantoea agglomerans and P. ananatis) to onion (Dutta et al., 2014). Center rot is a significant disease that can impact onions in the field and storage. Dutta et al. (2014) isolated both bacterial species in the midgut and feces of adult onion thrips. Subsequent transmission experiments indicated that adults could successfully transmit the pathogen to onion seedlings, with approximately 30 to 70% of seedlings becoming infected. Even when thrips do not directly transmit bacteria, their feeding creates wounds in which pathogenic bacteria likely enter. While bacterial center rot incidence can be variable, bulb yield losses upwards of 75% have been reported in New York (Stivers 1999). The role that onion thrips management has on the incidence and severity of onion diseases like iris yellow spot (IYS) and bacterial bulb rots has not been thoroughly examined.

Insecticide use is the most common management practice to control onion thrips in commercial onion production (Gill et al., 2015). In many cases, insecticides are exclusively relied upon to manage onion thrips infestations. However, in the past two decades, onion thrips have developed resistance to three chemical classes: pyrethroids, carbamates, and organophosphates. Resistance to these insecticides has been observed in many countries including the United States, Canada, New Zealand, and Australia (Herron et al., 2008; MacIntyre-Allen et al., 2005; Martin et al., 2003; Shelton et al., 2003, 2006). Utilizing multiple management techniques should not only slow the onset of insecticide resistance in onion thrips populations, but also limit harmful environmental effects that may arise from excessive insecticide applications. There are many different pest management techniques that have been reported to control onion thrips infestations (Gill et al., 2015). However, in commercial onion production, the amount of nitrogen applied, cultivar selection, and the type and frequency of insecticides applied have offered the greatest potential for reducing damage by onion thrips and associated plant diseases. Moreover, these management tactics are practical and most likely to be adopted by growers.

Appropriate levels of nitrogen during the growing season are critical to the establishment and development of the onion crop. However, excessive amounts of nitrogen fertilizer have been associated with greater onion thrips densities (Buckland et al., 2013; Malik et al., 2009). Buckland et al. (2013) found that onions treated with 134 kg N ha-1 had 23-31% fewer onion thrips than those onions treated with 402 kg N ha^{-1}. Similarly, Malik et al. (2009) reported nearly twice as many thrips on onions supplemented with 200 kg N ha-1 compared with 50 to 150 kg N ha-1. Thus, applying low levels of nitrogen fertilizer at onion planting may be an integral component of an onion thrips management program.

Currently, there are no onion cultivars that are completely resistant to onion thrips feeding, but some cultivars are partially resistant and suffer less feeding damage with little to no effect on bulb size. Both leaf waxiness and color have been reported to affect onion thrips densities. Cultivars with yellow-green leaves tend to be ‘semi-glossy’ and support fewer onion thrips, whereas those ‘waxy’ cultivars with blue-green leaves tend to have greater levels of epicuticular wax and are highly susceptible to onion thrips (Boateng et al., 2014; Diaz-Montano et al., 2012a). Damon et al. (2014) found that cultivars with blue-green leaves typically had a high amount of cuticular wax containing the ketone hentriacontanone-16 (H16), and onions with yellow-green, semi-glossy leaves had less cuticular wax and low levels of the H16 ketone. Thus, yellow-green, ‘semi-glossy’ onion cultivars should be included in an onion thrips management program.

The use of thresholds to manage onion thrips in onion has been examined for the past three decades (Fournier et al., 1995; Nault and Huseth, 2016; Rueda et al., 2006; Shelton et al., 1987). Consistently, researchers have reported that insecticides applied following action thresholds can provide effective thrips control. Hoffmann et al. (1995) found that an action threshold-based insecticide program provided equivalent thrips control as a standard insecticide program, but the action threshold-based program reduced insecticide applications by 37%. Nault and Huseth (2016) also compared an action threshold-based insecticide program with a standard insecticide program (weekly applications) and found equal levels of thrips control, but the action threshold-based program reduced insecticide applications between 34 and 46%. Additionally, onion bulb weights were equivalent following the standard and action threshold-based programs.

The purpose of our study was to 1) examine the effect of an integrated pest management program that combined the aforementioned thrips management techniques (low nitrogen rate at planting, thrips-resistant onion cultivar, and an action threshold-based insecticide program) on onion thrips densities, damage and onion yield, and 2) examine the effect of this integrated pest management program on the incidence and severity of two thrips-associated plant diseases, iris yellow spot and bacterial rot, in onion. We hypothesized that a reduced rate of nitrogen paired with an action threshold insecticide program would provide effective thrips and disease management without compromising marketable yield. Moreover, we expected the greatest reduction in agri-chemical input (lower amount of nitrogen and fewer insecticide applications) in the cultivar with the highest resistance to thrips.

2. Materials and Methods

2.1 Experimental design

Field studies were conducted on a commercial onion farm near Elba, NY in 2015 and 2016. Soil type at the test sites was 'Carlisle' muck (NRCS, 2016). Three onion cultivars ranging from moderate levels of resistance to no resistance to onion thrips were chosen based on their leaf waxiness and color (Damon et al., 2014; Diaz-Montano et al., 2012a). 'Avalon' (Crookham Co., Caldwell, ID) has yellow-green, semi-glossy foliage and has a moderate level of resistance to thrips, while 'Delgado' (Bejo Seeds, Inc., Oceano, CA) has green, semi-glossy foliage and has a low level of resistance to thrips. 'Bradley' (Bejo Seeds, Inc., Oceano, CA) has blue-green, waxy foliage and is highly susceptible to thrips. All cultivars are intermediate to long-day, yellow onions with similar days to harvest; 'Avalon' matures in 115 days, 'Delgado' in 118 days and 'Bradley' in 118 days. Fields were planted using a vacuum seed planter with approximately 646,000 onion seeds per hectare on 28 Apr 2015 and 16 Apr 2016. Seeds were treated with FarMore FI500 (mefenoxam [0.15 g ai/kg of seed], fludioxonil [0.025 g ai/kg of seed], azoxystrobin [0.025 g ai/kg of seed], spinosad [0.2 mg ai/seed] and thiamethoxam [0.2 mg ai/seed]) and Pro-Gro (carboxin [7.5 g ai/kg of seed] and thiram [12.5 g ai/kg of seed]) to improve plant establishment by protecting seedlings from maggots (Delia spp.) and seedling diseases.

Because each cultivar has a different yield potential, bulb yields were not compared among cultivars. Therefore, each cultivar was planted into separate blocks that were 28 m x 40 m. All three blocks were contiguous and separated from each other by only 1-3 m. Within each cultivar, there were nine treatments in a 3 (nitrogen rate) x 3 (insecticide program) factorial. Nitrogen rates were 67, 101 and 140 kg ha-1; insecticide programs were standard weekly applications, applications based on an action threshold and an untreated control. Nitrogen rates were chosen in accordance to current grower practices and management guidelines in New York: 140 kg N ha-1 (standard rate), 101 kg N ha-1 (28% reduction from the standard rate), and 67 kg N ha-1 (52% reduction from the standard rate) (Reiners and Seaman 2015). Treatments were replicated five times and arranged in a randomized complete block design, amounting to 45 experimental plots per cultivar. Experimental plots were 1.5 m wide x 6 m long and consisted of 5 rows of onion plants. Urea nitrogen (46-0-0) was incorporated into plots at planting. Experimental plots were also supplemented at planting with the appropriate rates of potassium (potassium chloride; 0-0-60; N-P-K) and phosphorus (triple superphosphate; 0-46-0; N-P-K) per current soil tests and corresponding fertility guidelines. All experimental plots were surrounded by either 1.5 m of bare ground or non-nitrogen treated onions to minimize the chances that nitrogen would move between plots. Soil nitrate levels were tested in all fields prior to planting to ensure that soil did not have excessively high levels of soil nitrate; all fields tested were within the low to normal range of soil nitrate (15-50 ppm) (Hoepting 2009).

Treatments receiving the standard insecticide program were sprayed every week, while those receiving the action threshold program were sprayed only when the onion thrips population met or surpassed an action threshold of 1 larva per leaf (Nault and Huseth 2016; Nault and Shelton 2010). The untreated control did not receive foliar-applied insecticides. Insecticide applications were made in accordance with current insecticide resistance management recommendations and guidelines (Reiners and Seaman 2015). All insecticide programs were initiated when treatments reached a mean density of approximately 0.8 larvae per leaf. Plots were scouted weekly beginning on 24 Jun 2015 and 21 Jun 2016, and insecticide program treatments were initiated on 15 Jul 2015 and 5 Jul 2016. Standard insecticide programs concluded on 25 Aug 2015 and 8 Aug 2016. Action threshold insecticide program treatments concluded on 18 Aug 2015 and 8 Aug 2016.

Four insecticides, each with a different mode of action, were rotated such that no insecticide was applied more than twice within a growing season. Insecticides were applied with the following sequence and rates: spirotetramat at 0.08 kg (AI) ha-1 (Movento; Bayer CropScience, Research Triangle Park, NC), abamectin at 0.02 kg (AI) ha-1 (Agri-Mek SC; Syngenta, Greensboro, NC), spinetoram at 0.07 kg (AI) ha-1 (Radiant SC; Dow AgroSciences, Inc., Indianapolis, IN), and cyantraniliprole at 0.1 kg (AI) ha-1 (Exirel; DuPont, Wilmington, DE). Insecticides were applied with a CO2-pressurized backpack sprayer with four, twin flat-fan nozzles (TJ-60-8003VS; TeeJet Technologies Harrisburg, PA). All insecticides were co-applied with an adjuvant at 0.5% v:v (Induce; Helena, Collierville, TN) to increase efficacy (Nault et al., 2013).

There were no other insect pests that damaged the onions in this experiment. Weeds and plant pathogens were managed according to Cornell vegetable management guidelines and recommendations (Reiners and Seaman 2015).

2.2 Nitrogen assessments

Foliar nitrogen assessments were completed at three developmental stages: pre-bulbing (3-5 leaves per plant), bulbing (5-8 leaves per plant), and post-bulbing (9+ leaves per plant). Ten randomly selected leaves per plot were collected to create an average composite sample. Leaves were transported to the New York State Agricultural Experiment Station in Geneva, NY. Leaves were washed with distilled water, dried at 70oC for at least 48 hours and ground through a 40-mesh screen. Soil samples were submitted to Cornell Nutrient Analysis Laboratory in Ithaca, NY where total carbon, nitrogen, and hydrogen were determined using combustion analysis (Kalra 1998).

Plant growth was monitored throughout the growing season. Leaf length was measured twice during each developmental stage, and number of leaves per plant was recorded weekly. The number of green leaves was counted on 15 randomly selected onion plants. To estimate leaf length, the tallest leaf on 15 randomly selected plants in each plot was taken.

2.3 Onion thrips sampling and damage

Numbers of onion thrips adults and larvae were counted every week in every plot. Fifteen plants, randomly selected from the inner three rows, were visually examined for thrips. Counts began after colonization, which occurred when plants had approximately 4-5 leaves, and continued until 80% or more of the plants had lodged. Thrips were monitored for 11 weeks in 2015, and 9 weeks in 2016. Voucher specimens are held at the New York State Agricultural Experiment Station in Geneva, NY.

Onion thrips damage was assessed when most plants had matured. Each plot was assigned a rating between 0-100 based on thrips feeding damage (modified from Nault and Shelton 2010). The rating scale was continuous, and ratings were assigned using the following reference points: 0: leaves devoid of thrips feeding, 50: 50% of leaves appear white due to thrips feeding, 100: complete damage, 100% of leaves appear white from thrips feeding. Damage ratings were completed on 22 Aug 2016; no damage ratings were collected in 2015 because a late-season outbreak of Stemphylium leaf blight obstructed thrips damage symptoms.

2.4 Iris yellow spot virus (IYSV)

Fifteen plants per plot were visually examined for characteristic IYS symptoms from the inner three rows of onions. Symptoms included leaves exhibiting lesions that were either tan or straw colored (Schwartz and Mohan 2008). Plants were assessed based on the presence or absence of IYS disease symptoms. In 2015, plots were evaluated on two dates during the growing season, 29 Jul and 29 Aug. Because IYS was more severe in 2016, sampling intensity increased to five dates: 24 Jul, 1 Aug, 8 Aug, 15 Aug, and 22 Aug.

Severity of IYS was determined using a scale from 0-4 as described in Schwartz and du Toit (2005). Fifteen plants per plot, randomly selected from the inner three rows of onions, were visually assessed and each given a rating: 0= no lesions, 1= 1-2 small lesions per leaf, 2= 1-2 medium sized lesions per leaf, 3= 25% of leaves with lesions that were coalescing, or 4= 50% or more of the leaves had coalesced lesions. Onions were assessed on 1 Sept 2015 and 24 Aug 2016. An outbreak of Stemphylium leaf blight in 2015, which obstructed IYS symptoms late in the season, precluded IYS severity ratings to be taken in two of the three cultivars; only ‘Delgado’ was assessed. All cultivars were assessed for IYS severity in 2016.

While IYS disease has very characteristic symptoms and is not commonly confused with other diseases or physiological problems in onion in New York State, we wanted to confirm our visual assessments using RT-PCR on a subset of plants that were symptomatic following the protocol described in Hsu et al. (2011). Thus, we randomly selected ten plants expressing IYS symptoms in 2015 and again in 2016 and all were confirmed positive.

2.5 Bacterial bulb rot

Onion bulbs were assessed for bacterial rot at harvest and another set of bulbs were assessed three months after harvest while in storage. Onions were cured in the field, and then transported to the New York State Agricultural Experiment Station in Geneva, NY. To reduce the potential confounding effect of bacterial rot on bulb size, only standard-sized (diameter of 4.9 cm to 7.6 cm, weight of 90 g to160 g) bulbs were assessed for rot. Approximately 50 standard-sized bulbs per plot were assessed for rot at harvest and an additional 50 bulbs were assessed three months after harvest. All onion bulbs were cut longitudinally and examined for bacterial rot. Bacterial bulb rot was classified based on symptoms when possible. Onion bulbs were considered to have ‘center rot’ when rot was present only in the inner scales of the onion, and ‘outer rot’ when rot was present in the outer scales of the onion. Sub-samples of onion bulbs that were stored for three months were placed in nylon bags, and stored in a ventilated, temperature controlled building. Onions were stored between 0-3o C and 60-75% relative humidity. Number of rotten bulbs at harvest were added to number of rotten bulbs three months after harvest to create an estimate of total rotten onion bulbs for a given plot. Bacterial species were identified from a random sub-sample of 20 onion bulbs per treatment that were symptomatic for bulb rot. Bacteria from symptomatic bulbs were recovered using a semi-selective onion extract medium (Zaid et al., 2012). Bacteria known to cause bacterial rot of onion were identified by PCR.

2.6 Onion bulb yield

Bulbs were harvested when 80% or more of the plants had lodged for each cultivar. Onion plants were undercut, and cured in the field for a week prior to harvest. Onions were harvested on 6 Sept 2015 and 25 Aug 2016. After curing, onions were placed in nylon bags, and transported to the New York State Agricultural Experiment Station in Geneva, NY. Any remaining dried leaves on onion bulbs were mechanically removed, and bulbs weighed. Bulbs were classified according to bulb diameter and assigned a size class of either ‘boiler’ (2.5 cm-4.8 cm), ‘standard’ (4.9 cm-7.6 cm), or ‘jumbo’ (≥7.7 cm). Bulbs that were either ‘standard’ or ‘jumbo’ were considered marketable, and ‘boiler’ bulbs unmarketable. Marketable yields for treatments were then extrapolated to estimate mean tons per hectare based on onion stand counts recorded in each cultivar in 2015 and 2016.

2.7 Statistical analysis

Data for each cultivar were analyzed independently based on the rationale mentioned earlier and data within each year were analyzed separately because environmental conditions were extremely different (Table 2.1). Data were analyzed using a generalized linear mixed model (SAS PROC GLIMMIX, 2016; SAS Institute, Cary, NC). Nitrogen rate and insecticide program were treated as fixed effects and replicate as a random effect.

All count data, including seasonal mean number of adult and larval onion thrips per leaf and mean number of onion leaves per plant were analyzed assuming a negative binomial distribution. Leaf length, percent total nitrogen, and marketable yield were analyzed assuming a normal distribution. IYS severity data was log-transformed prior to analysis to normalize the data and homogenize variation, and then analyzed assuming a normal distribution. Bacterial rot incidences were analyzed as a binomial distribution (n rotten onion bulbs/total onion bulbs, n bulbs with center rot/total rotten bulbs). A low amount of bacterial center rot in ‘Bradley’ precluded its inclusion in the analysis for center rot incidence in 2015. IYS incidence was also analyzed as a binomial distribution (n plants expressing IYS symptoms/ total plants examined). IYS incidence was only analyzed when it was above 0% or below 100%. Thus, analysis of IYS incidence was completed for 22 Aug 2015, 25 Jul 2016, and 1 Aug 2016. Treatments in each analysis were compared using least squared means ($P<0.05$).

3. Results

3.1 Nitrogen assessments

Foliar nitrogen assessments. Total nitrogen levels in onion leaves at pre-bulbing, bulbing, and post-bulbing were not significantly affected by nitrogen rate, insecticide treatment, or the interaction between nitrogen rate and insecticide treatment in any cultivar in both years ($P>0.05$) (data not shown). Foliar nitrogen ranged from 3.5 to 5.9% over the course of the growing season in all cultivars. Percent nitrogen in onion leaves decreased at each developmental stage, with highest values recorded at pre-bulbing and lowest at post-bulbing in both years.

Length and number of leaves. In all cultivars, mean leaf length and total number of leaves were not significantly different in any of the treatments in either year ($P>0.05$) (data not shown). Mean number of leaves and leaf length increased over the duration of the season in both years and reached maximum lengths and counts in early to mid-August in every cultivar.

3.2 Onion thrips densities and damage

Onion thrips larvae. Although differences among cultivars were not statistically compared, ‘Avalon’ had the fewest seasonal mean number of thrips larvae per leaf in untreated plots. There was 0.5-1 fewer larva per leaf in ‘Avalon’ than in ‘Delgado’ and ‘Bradley’ in 2015 and 2016.

Onion thrips larvae were more abundant than adults. Larvae accounted for 65-82% of total mean thrips per leaf in 2015 and 65-73% in 2016. The seasonal mean larval densities were significantly affected by the insecticide program in both years (Table 2.2), but not by nitrogen rate nor the interaction between nitrogen rate and insecticide program ($P>0.05$) (data not shown). The highest seasonal mean densities of larvae occurred in untreated controls and exceeded the economic threshold of 2.2 thrips per leaf in all cultivars in both years (Fournier et al., 1995).

In 2015 for all cultivars, larval densities in the action threshold and standard insecticide programs were significantly lower than those in the untreated control (Avalon: $P<0.0001$, $F_{2,\ 32}=21.7$, Delgado: $P<0.0001$, $F_{2,\ 32}=19.6$ and Bradley $P<0.0001$, F2, 32=32.4), but larval densities were statistically similar between the two insecticide programs (Table 2.2). Larval densities in ‘Avalon’, ‘Delgado’ and ‘Bradley’ were reduced by 66, 70, and 83%, respectively, using either an action threshold or standard insecticide program. Similarly, in 2016, larval densities in the action threshold and standard insecticide programs were significantly lower than those in the untreated control (Avalon: $P=0.002$, $F_{2, 32}=7.4$, Delgado: $P<0.0001$, $F_{2, 32}=18.9$, Bradley: $P=0.0008$, $F_{2, 32}=8.9$) (Table 2). Larval densities in the action threshold and standard insecticide treatments reduced larval densities by 40-83% in comparison with untreated control (Table 2.2). In ‘Avalon’ and ‘Delgado’, larval densities in the action threshold and standard insecticide programs were statistically similar, whereas in ‘Bradley’ larval densities in the standard insecticide program were significantly lower than in the action threshold-based program (Table 2.2).

In all cultivars, larval onion thrips densities peaked in late July to early August in 2015 and 2016, respectively (Figure 2.1). Peaks in larval onion thrips densities in untreated controls were preceded by peaks in adult densities in every cultivar in both years. In 2015, larval densities peaked in action threshold and standard insecticide treatments on 22 July (Figure 2.1). However, the largest larval population densities were recorded on 29 Jul in untreated controls, with mean maximums of 21.5, 22.5, and 33.3 larvae per leaf in ‘Avalon’, ‘Delgado’, and ‘Bradley’, respectively. In 2016, the highest numbers of thrips larvae were recorded on 8 Aug in action threshold treatments in cv. ‘Avalon’ and ‘Bradley’, and untreated control in cv. ‘Delgado’, with peak densities of 5.6, 13.6, and 8.6 respectively. Densities of onion thrips larvae in all treatments and cultivars decreased in mid-August and remained below 2 thrips per leaf until harvest in 2015 and 2016.

Onion thrips adults. Fewer adults were recorded in 2016 than in 2015. In both years, mean number of adults per leaf was not significantly impacted by nitrogen rate, insecticide program, or the interaction between insecticide program and nitrogen rate in any cultivar ($P>0.05$) (data not shown). Consistently in 2015 and 2016, ‘Avalon’ had the lowest mean number adult thrips per leaf, 0.5 and 0.4 respectively, and ‘Delgado’ had the highest mean number of adult thrips per leaf both years, 0.9 and 0.8 respectively. ‘Bradley’ had seasonal means of 0.6 and 0.7 adults per leaf in 2015 and 2016, respectively.

In 2015 and 2016, adult onion thrips colonized onion fields in early to mid- June and densities remained low, below 1 adult per leaf, until mid- to late-July when densities peaked (Figure 2.1). In 2015, the largest numbers of adults were recorded between 13 Jul and 22 Jul. Adults reached maximum densities of 2.6, 4.0, and 3.1 adult thrips per leaf in ‘Avalon’, ‘Delgado’, and ‘Bradley’, respectively. In 2016, adult densities peaked from 19 Jul to 1 Aug, with maximum densities of 0.9, 2.2, and 1.6 adult thrips per leaf in ‘Avalon’, ‘Delgado’, and ‘Bradley’ respectively. In both years and all cultivars, adult densities decreased in early August and remained below one adult per leaf until harvest.

Onion thrips damage. Damage ratings were significantly affected by the interaction between nitrogen rate and insecticide program in all cultivars (Avalon: $P=0.036$, $F_{4, 32}=2.9$, Delgado: $P=0.015$, $F_{4, 32}=3.6$ and Bradley $P=0.0002$, $F_{4, 32}=7.8$) (Figure 2.2). For every cultivar, the most damage was recorded in the untreated control, and the least in standard insecticide treatments. Damage ratings in standard insecticide treatments in ‘Avalon’, ‘Delgado’, and ‘Bradley’ were 37, 67, and 75% lower, respectively, than ratings in the untreated control. While damage levels in the untreated controls did not vary much across nitrogen rates, damage levels in the action threshold treatments that received 140 kg N ha-1 in ‘Delgado’ and ‘Bradley’ tended to be higher than those at 67 kg N ha-1. Additionally, higher levels of damage were recorded in standard insecticide treatments supplemented with 140 kg N ha-1 in ‘Avalon’ compared to other nitrogen rates (Figure 2.2).

Insecticide applications. Fewer insecticide applications were consistently made following the action threshold programs compared with the standard insecticide programs (Table 2.2). In 2015, frequency of insecticide applications in action threshold treatments decreased by 47% in ‘Avalon’ and ‘Bradley’, and 33% in ‘Delgado’ compared with the frequency of applications in the standard insecticide programs. In 2015, larval densities surpassed the action threshold of 1 thrips larva per leaf on four dates in ‘Avalon’ and ‘Bradley’, and five dates in ‘Delgado’. In 2016, frequency of insecticide applications in action threshold treatments decreased by 50% in ‘Avalon’, 33% in ‘Delgado’, and 45% in ‘Bradley’ compared with the frequency of applications in the standard insecticide programs. Larval onion thrips densities exceeded the action threshold on three dates in ‘Avalon’, and on four dates in ‘Delgado’ and ‘Bradley’. Overall, numbers of insecticide applications were similar across the various nitrogen rates within each cultivar.

3.3 Iris yellow spot virus (IYSV)

IYS Incidence. IYS incidence reached very high levels by the end of each season. In 2015 for all cultivars, the incidence of IYS (% plants exhibiting IYS disease) was not influenced by nitrogen rate, insecticide program or an interaction ($P>0.05$). No plants exhibited symptoms on 24 Jul, but by the end of the season 60-80% of the plants had IYS symptoms (Figure 2.3). In contrast in 2016 for all cultivars, the incidence of IYS was significantly influenced by insecticide program on 25 Jul (Figure 2.3). IYS incidence was significantly affected by insecticide treatments in all cultivars (Avalon: $P=0.0005$, $F_{2, 32}=9.9$, Delgado: $P<0.0001$, $F_{2, 32}=36.1$ and Bradley $P=0.014$, $F_{2, 34}=3.7$). In late July, more onion plants in untreated control plots displayed IYS symptoms than those in action threshold and standard insecticide treatments. On 25 Jul, symptoms of IYS in ‘Avalon’, ‘Delgado’ and ‘Bradley’ were first detected 14%, 19%, and 37% (overall mean) of plants exhibiting IYS symptoms, respectively. By 15 Aug, 100% of all onions in every cultivar displayed IYS symptoms (Figure 2.3).

IYS severity. IYS symptoms were less severe in 2015 than in 2016. In 2015, ‘Delgado’ had a mean severity value of 1.3±0.3 (on a scale of 0-4) and displayed few, small- to medium-sized, IYS lesions on leaves. Severity of IYS was not statistically different in any treatments in ‘Delgado’ in 2015 ($P>0.05$) (data not shown). Conversely in 2016, IYS severity averaged 2.9, 3.0, and 3.1 in ‘Avalon’, ‘Delgado’, and ‘Bradley’, respectively (Figure 2.4). Most assessed plants exhibited leaf dieback and lesion coalescence from the IYSV infection. In ‘Avalon’ and ‘Delgado’, IYS severity was only significantly impacted by insecticide program (Avalon: $P=0.0005$, $F_{2, 32}=9.9$, Delgado: $P<0.0001$, $F_{2, 32}=36.1$). IYS severity in action threshold and standard insecticide programs were statistically similar, and had 16 and 30% lower severity levels, respectively, compared with levels in the untreated control. For ‘Bradley’, IYS severity was significantly impacted by the interaction of insecticide program and nitrogen rate ($P=0.014$, $F_{4, 34}=3.7$) (Figure 2.5). IYS severity in untreated controls and standard insecticide programs were similar across all nitrogen rates. However, in action threshold treatments treated with 140 kg N ha-1 had higher levels of IYS severity as compared to action threshold treatments treated with 67 and 101 kg N ha-1.

3.4 Bacterial rot incidence

Multiple bacterial species were identified by PCR in rotten bulbs including Enterobacter ludwigii, Klebsiella pneumoniae, Klebsiella oxytoa, Burkholderia cepacia, Serratia marcescens, Pantoea agglomerans, Lactococcus lactis, and Rahnella spp. However, the incidence of bacterial center rot caused by Pantoea agglomerans was not significantly affected by any treatment in 2015 or 2016 ($P>0.05$) (data not shown).

In 2015 and 2016, total incidence of bacterial bulb rots was significantly affected by the interaction of nitrogen rate and insecticide treatment in all cultivars (2015: Avalon: $P=0.0003$, $F_{4,32}=7.1$, Delgado: $P<0.0001$, $F_{4,28}=13.8$ and Bradley $P=0.0056$, $F_{4,32}=4.5$; 2016: Avalon: $P=0.021$, $F_{4,32}=3.3$, Delgado: $P=0.0187$, $F_{4,32}=3.5$ and Bradley $P=0.0324$, $F_{4,32}=3$) (Table 2.3). Incidences of bacterial rot at harvest and after three months after harvest varied between treatments; however, no consistent trends were observed (Supplemental Tables 2.1 and 2.2). In ‘Avalon’ in 2015, standard insecticide programs paired with 140 kg N ha-1 had significantly higher amounts of total bacterial rot compared with all other treatments. However, in ‘Avalon’ in 2016, untreated controls and action threshold treatments had the highest incidences of rot. In ‘Delgado’ in 2015, the highest levels of bacterial rot were recorded in the untreated control that received 140 kg N ha-1. In ‘Delgado’ in 2016, the highest levels of rot occurred in untreated controls supplemented with either 67 or 101 kg N ha-1 and in standard insecticide programs paired with 101 kg N ha-1. Bacterial rot levels in ‘Bradley’ in 2015 or 2016 ranged from 0.6-6.7% across all treatments.

In all cultivars in both years, bacterial bulb rot incidence increased greatly three months after harvest (Figure 2.6). In 2015, averaging across all treatments, there was a 45, 1861, and 417% increase in bacterial bulb rot three months after harvest in ‘Avalon’, ‘Delgado’, and ‘Bradley’, respectively. In 2016, there was only a 1203% increase in bacterial bulb rot three months after harvest in ‘Avalon’. In both years, ‘Avalon’ had the highest amount of bacterial bulb rot both at harvest and three months later, with 10% and 22% of total bulbs rotten in 2015 and 2016, respectively. ‘Delgado’ and ‘Bradley’ had lower levels of bacterial rot, with 7% and 4% of total bulbs rotten, respectively. The same trend persisted in 2016, with 2% of bulbs rotten in ‘Delgado’ and 1% rotten in ‘Bradley’.

3.5 Onion yield

Marketable bulb yields in all three cultivars were impacted by insecticide program treatments in 2015 and 2016, but not by nitrogen rate or an interaction between the two (Figure 2.7). In 2015, marketable yields in ‘Delgado’ and ‘Bradley’ that received insecticide treatments were significantly higher than those in the untreated control, averaging 12.7 tons/hectare more than the control (Delgado: $P=0.0107$, $F_{2, 29}=5.4$ and Bradley: $P=0.0002$, $F_{2, 32}=11.3$). Yields in ‘Avalon’ followed the same trend, but differences were not significant ($P>0.05$). In 2016, marketable yields in all three cultivars that received insecticide treatments were significantly greater than those in the untreated controls (Avalon: $P=0.0089$, $F2, 32=5.5$, Delgado: $P=0.0002$, $F2, 32=11.7$, and Bradley $P<0.0001$, $F2, 32=16.9$). Yields were 7.9, 10.7, and 12.1 tons/hectare greater in insecticide treated plots of ‘Avalon’, ‘Delgado’ and ‘Bradley’, respectively, compared with yields in the untreated controls. Moreover, for each cultivar, yields were similar between insecticide programs.

Table 2.1: Weather conditions in 2015 and 2016 near Elba, NY.

Month	Minimum temp. (C°)		Maximum temp. (C°)		Mean temp. (C°)		Total rainfall (cm)	
	2015	2016	2015	2016	2015	2016	2015	2016
May	1.1	3.3	31.7	32.2	17.2	15	8.4	3.1
June	7.2	6.7	28.9	32.2	18.9	20	12.8	3.3
July	10	12.8	32.8	32.8	21.7	23.3	6.1	4.6
August	11.1	13.9	31.7	33.3	21.1	24.4	11.2	10.6

Table 2.2: Mean densities of larval onion thrips populations during the season in three onion cultivars varying in susceptibility to onion thrips and treated following different insecticide programs. Studies were conducted in commercial fields near Elba, NY in 2015 and 2016. Insecticide applications were made weekly in the standard program and only when thrips densities ≥1 larva/leaf in the action threshold-based program. Means within the same cultivar and year that share the same letter are not significantly different (P>0.05; LSmeans).

Cultivar	Insecticide program	Seasonal mean (± SE) number of onion thrips larvae/ leaf		Mean number of insecticide applications	
		2015	2016	2015	2016
Avalon	Untreated control	3.5 ± 0.5 a	2.4 ± 0.4 a	____	
	Action threshold	0.8 ± 0.1 b	1.3 ± 0.3 b	3.7	3
	Standard	0.7 ± 0.1 b	0.6 ± 0.2 b	7	6
Delgado	Untreated control	4.0 ± 0.5 a	3.6 ± 0.5 a	____	
	Action threshold	1.3 ± 0.2 b	0.9 ± 0.3 b	4.7	4
	Standard	1.1 ± 0.2 b	0.6 ± 0.2 b	7	6
Bradley	Untreated control	4.9 ± 0.8 a	3.2 ± 0.4 a	____	
	Action threshold	0.9 ± 0.1 b	1.9 ± 0.5 b	3.7	3.3
	Standard	0.8 ± 0.1 b	0.7 ± 0.2 c	7	6

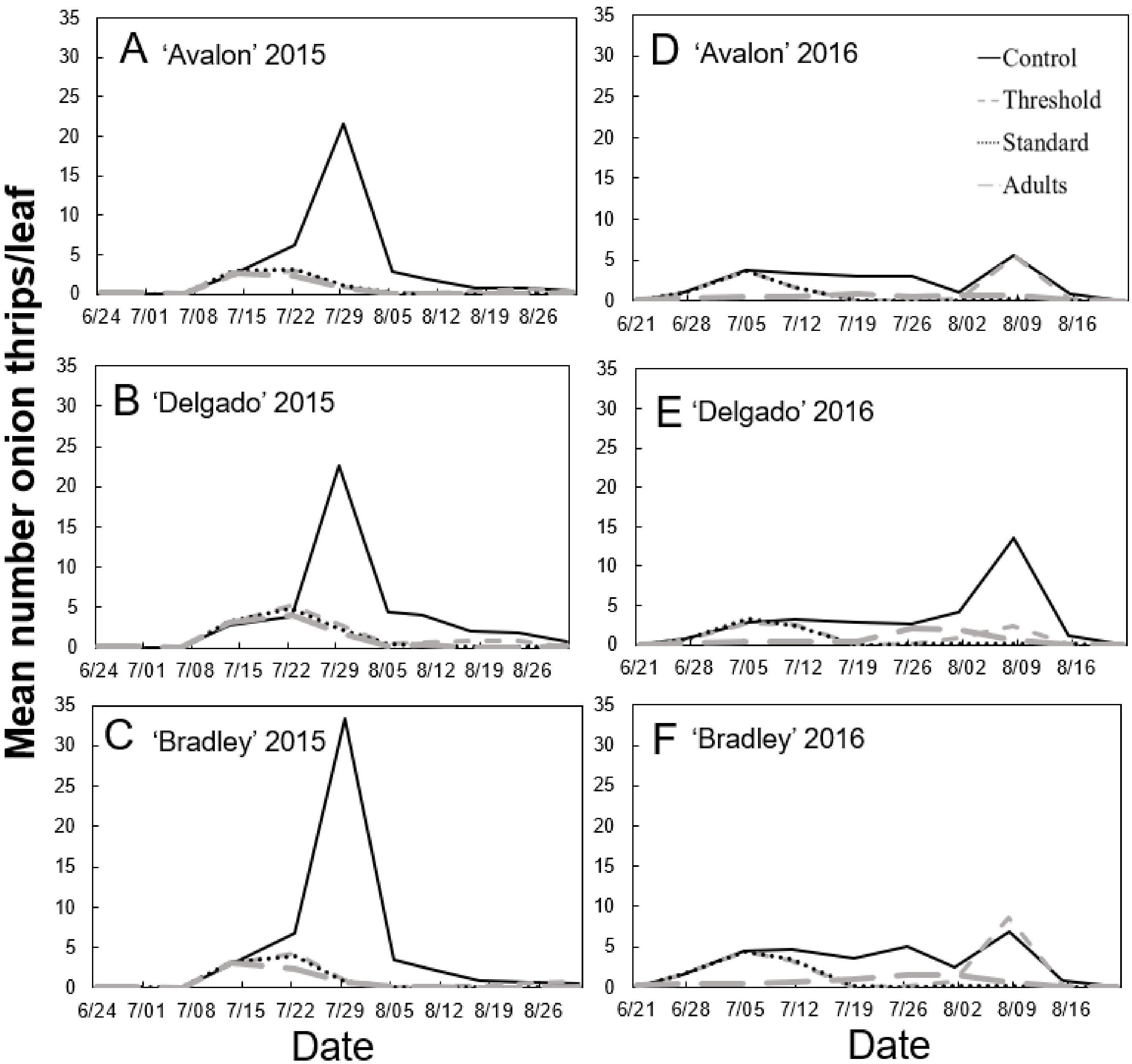

Figure 2.1: Mean densities of onion thrips during the season in onion cultivars with varying susceptibility to onion thrips, 'Avalon' (A and D), 'Delgado' (B and E), and 'Bradley' (C and F) in 2015 (A-C) and 2016 (D-F). Densities of larvae are shown for plots that were either not treated with insecticides (control) or treated following either a standard or threshold-based insecticide program, whereas densities of adults are shown for plots pooled across all insecticide treatments. Studies were conducted in commercial fields near Elba, NY. Monitoring thrips densities began when onions had 4-5 leaves, and concluded near harvest. Standard and action threshold-based insecticide programs were initiated on 15 July 2015 and 5 July 2016. Insecticide applications were made weekly in the standard program and only when thrips densities were ≥1 larva/leaf in the action threshold program. See Table 2 for average number of insecticide applications.

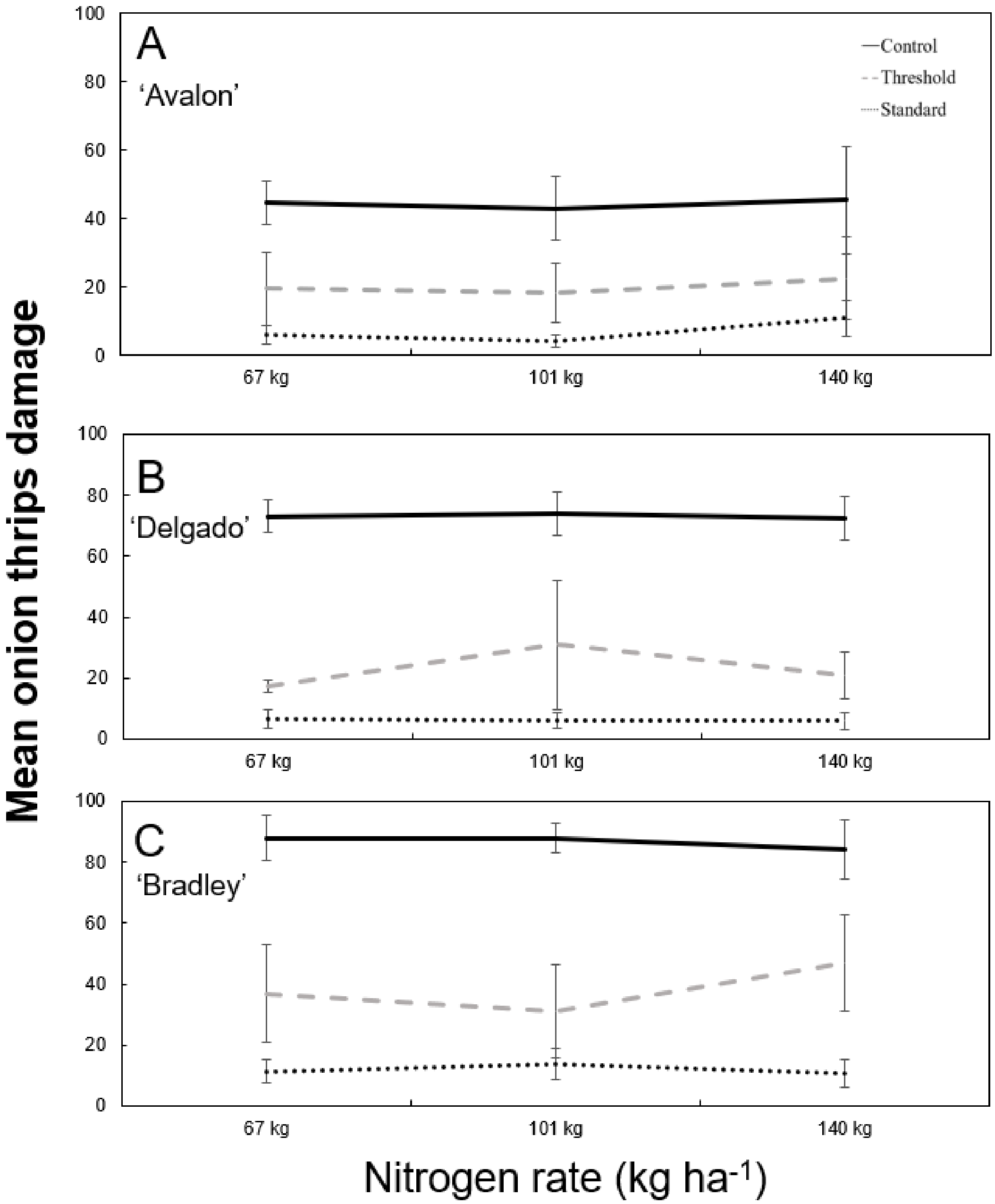

Figure 2.2: Mean (± SE) onion thrips damage ratings (0-100 scale) for onions that received different combinations of nitrogen fertilizer at planting and insecticide programs for managing onion thrips for each of three onion cultivars, 'Avalon' (A), 'Delgado' (B), and 'Bradley' (C). Studies were conducted near Elba, NY in 2016. Standard and threshold-based insecticide programs were initiated on 5 July 2016. Insecticide applications were made weekly in the standard program and only when thrips densities were ≥1 larva/leaf in the action threshold program.

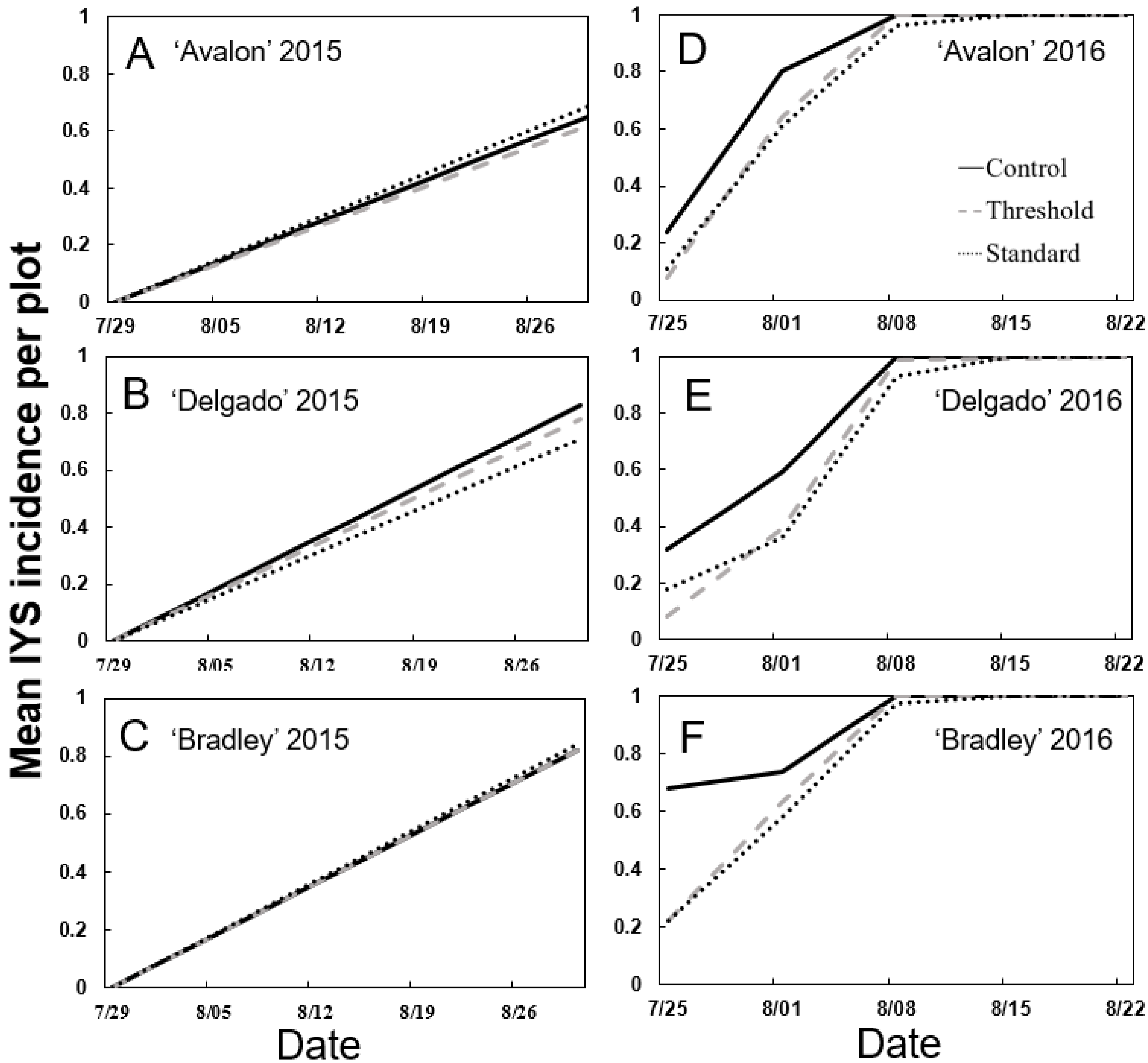

Figure 2.3: Mean IYS incidence (proportion of plants with symptoms) in onions treated with different insecticide programs for managing onion thrips in each of three onion cultivars that varied in susceptibility to onion thrips, 'Avalon' (A and D), 'Delgado' (B and E), and 'Bradley' (C and F) in 2015 (A-C) and 2016 (D-F). Studies were conducted in commercial fields near Elba, NY. Monitoring IYS incidence began when virus symptoms were first detected and concluded near harvest. Standard and threshold-based insecticide programs were initiated on 24 July 2015 and 25 July 2016. Insecticide applications were made weekly in the standard program and only when thrips densities ≥1 larva/leaf in the action threshold-based program.

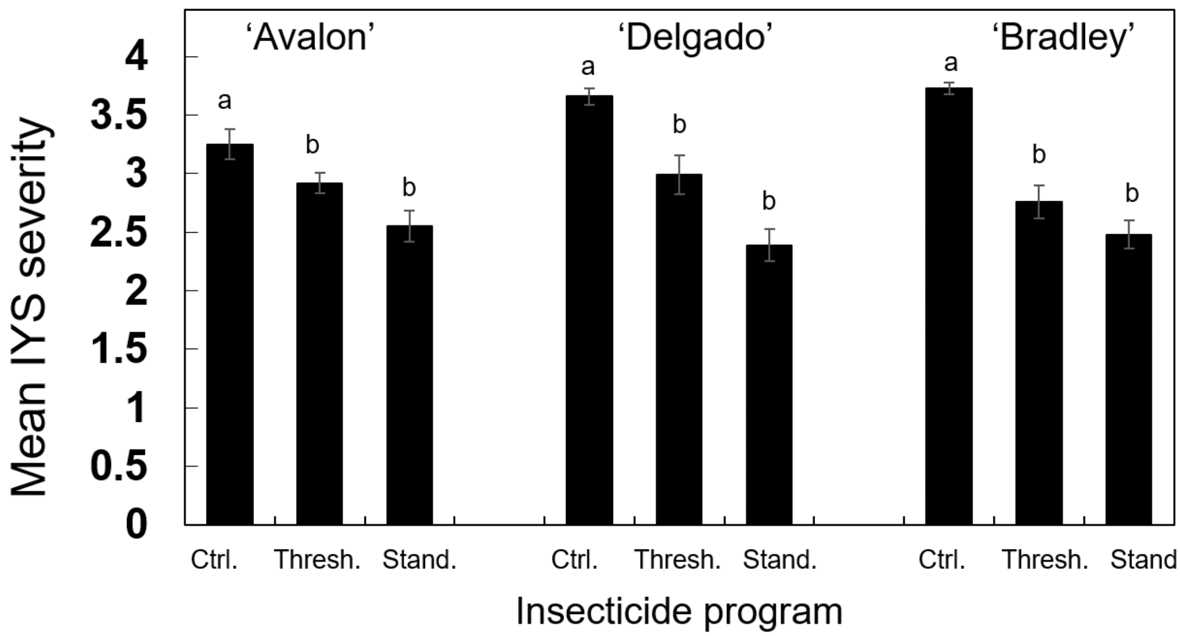

Figure 2.4: Mean (± SE) severity of IYS symptoms (1-4 scale) for onion cultivars that vary in susceptibility to onion thrips and that were either not treated with insecticides (Ctrl.) or treated with a threshold-based insecticide program (Thresh.) or a standard (Stand.) insecticide program. Studies were conducted in commercial fields near Elba, NY. Insecticide applications were made weekly in the standard program and only when thrips densities ≥1 larva/leaf in the action threshold-based program.

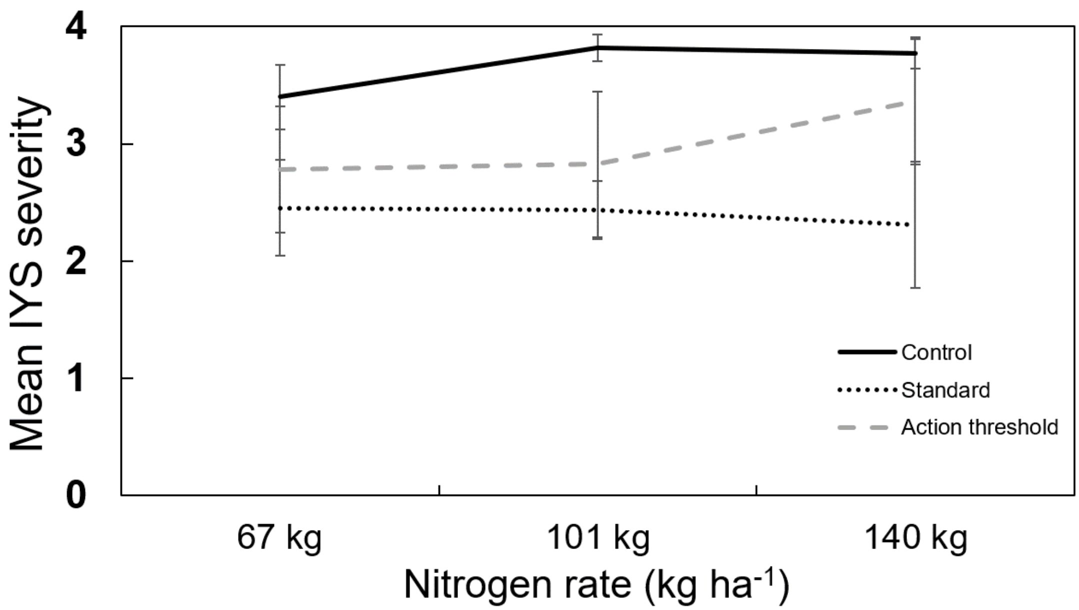

Figure 2.5: Mean (± SE) severity of IYS symptoms (1-4 scale) per plot for 'Bradley' in 2016 under various combinations of nitrogen fertilizer at planting and insecticide programs for managing onion thrips. Studies were conducted in commercial fields near Elba, NY. Insecticide applications were made weekly in the standard program and only when thrips densities ≥1 larva/leaf in the threshold-based program.

Table 2.3: Mean percent of bulbs with bacterial rot for onion cultivars varying in susceptibility to onion thrips that received various combinations of nitrogen fertilizer at planting and insecticide treatments for managing onion thrips. Studies were conducted near Elba, NY in 2015 and 2016. Insecticide applications were made weekly in the standard program and only when thrips densities ≥1 larva/leaf in the action threshold-based program. Means within the same cultivar and year that share the same letter are not significantly different (P>0.05; LSmeans).

Cultivar	Treatment		Mean % (± SE) bacterial incidence	
	Insecticide program	Nitrogen rate (kg ha^{-1})	2015	2016
Avalon	Untreated control	67 kg	18.5 ± 5.6 d	9.0 ± 3.8 abc
		101 kg	25.6 ± 4.8 bc	12.9 ± 3.7 a
		140 kg	17.7 ± 1.8 d	7.7 ± 1.5 bc
	Action threshold	67 kg	16.7 ± 2.9 d	12.6 ± 3.3 a
		101 kg	20.4 ± 3.9 ab	10.5 ± 2.5 ab
		140 kg	27.3 ± 5.9 cd	12.6 ± 3.8 a
	Standard	67 kg	21.4 ± 2.6 d	7.4 ± 1.5 bcd
		101 kg	21.6 ± 3.6 cd	4.5 ± 2.1 d
		140 kg	31.6 ± 8.8 a	6.8 ± 1.7 cd
Delgado	Untreated control	67 kg	4.9 ± 2.1 cd	4.5 ± 2.5 a
		101 kg	3.9 ± 2.9 d	3.4 ± 0.7 a
		140 kg	17.9 ± 9.6 a	2.2 ± 1.0 ab
	Action threshold	67 kg	8.5 ± 2.2 b	2.4 ± 1.1 ab
		101 kg	9.4 ± 3.0 b	0.8 ± 0.5 b
		140 kg	4.8 ± 1.6 cd	2.3 ± 1.1 ab
	Standard	67 kg	4.4 ± 1.3 cd	0.8 ± 0.8 b
		101 kg	7.2 ± 1.1 bc	3.1 ± 0.9 a

		140 kg	5.1 ± 3.9 cd	1.2 ± 0.5 b
Bradley	Untreated control	67 kg	6.7 ± 3.8 a	1.5 ± 0.9 b
		101 kg	2.3 ± 1.2 de	2.1 ± 1.1 ab
		140 kg	5.9 ± 2.9 ab	1.1 ± 0.5 b
	Action threshold	67 kg	3.1 ± 2.4 cd	0.7 ± 0.3 b
		101 kg	1.6 ± 1.0 de	1.7 ± 1.1 b
		140 kg	1.4 ± 0.7 de	1.8 ± 0.8 b
	Standard	67 kg	1.2 ± 0.8 e	1.5 ± 0.5 b
		101 kg	3.4 ± 1.5 bcd	0.6 ± 0.3 b
		140 kg	4.9 ± 0.8 abc	4.0 ± 3.1 a

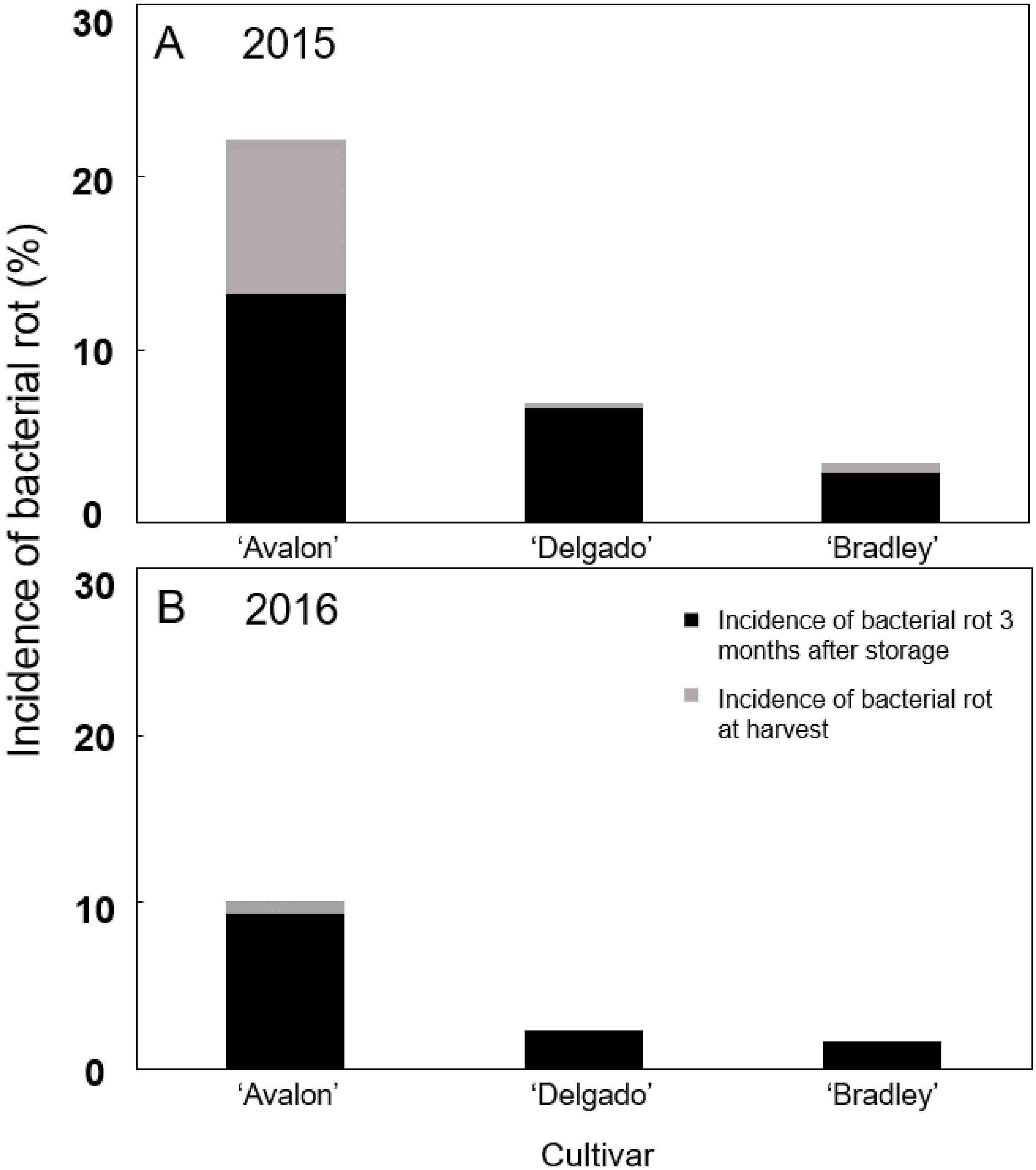

Figure 2.6: Mean total percentage of bulbs with bacterial rot for three onion cultivars that vary in susceptibility to onion thrips in field trials near Elba, NY in 2015 (A) and 2016 (B). Subsamples of bulbs were assessed for rot at harvest and again three months after harvest; these results show the combination of both assessments.

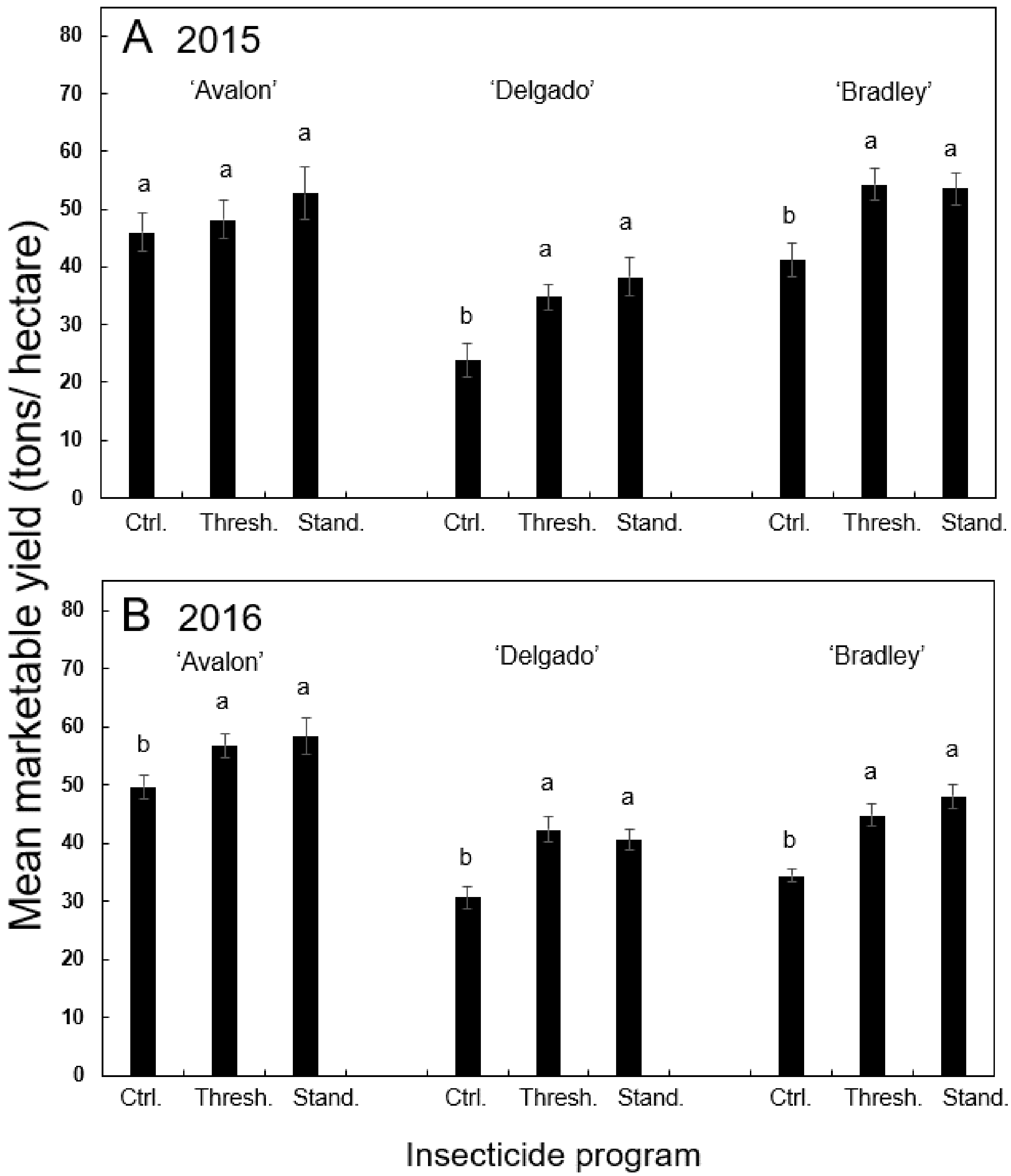

Figure 2.7: Mean marketable bulb yield (±SE) for onion cultivars that vary in susceptibility to onion thrips and that were either not treated with insecticides (Ctrl.) or treated with a threshold-based insecticide program (Thresh.) or a standard insecticide program (Stand.). Studies were conducted near Elba, NY in 2015 (A) and 2016 (B). Insecticide applications were made weekly in the standard program and only when thrips densities ≥1 larva/leaf in the threshold-based program. Means within the same cultivar that share the same letter are not significantly different ($P>0.05$; LSmeans).

4. Discussion

Insecticide use had the greatest impact on reducing larval onion thrips densities, IYS severity and incidence, and increasing bulb yields, while nitrogen rate did not have a substantial impact on any of the variables examined. Standard and action threshold-based insecticide programs were equivalent in reducing larval thrips densities and IYS disease suppression and produced similar bulb yields. Yet, one-third to one-half fewer insecticide applications were needed following the action threshold-based program compared with the standard program, indicating that growers can adopt action thresholds and increase profits. Contrary to our expectation, a similar number of insecticide applications were required in the moderate-thrips resistant 'Avalon' as the thrips-susceptible 'Bradley'.

4.1 Onion thrips densities

Larval onion thrips comprised the greatest proportion of the thrips population, indicating that adults may contribute less to direct crop damage and loss. Multiple studies have reported similar ratios of larvae and adults as our study. Buckland et al. (2013) reported that adults composed approximately 20% of the total thrips, while Hsu et al. (2010) found that adults comprised less than 50% of the total thrips population at any given time during the growing season. Similarly, Coudriet et al. (1979) suggested that larvae may be the best predictors of crop damage and loss, and thus should be preferentially sampled. Our results continue to assert that larvae are the most damaging life stage in onion fields, and consequently the most important to control.

In contrast with other studies, we did not see reductions in onion thrips densities using lower rates of nitrogen at planting (Buckland et al., 2013; Malik et al., 2009). We also did not observe an increased amount of plant growth or leaf nitrogen in plots supplemented with higher rates of nitrogen. Differences in application timing of nitrogen and soil type in our study differed from those in previous studies and may explain the discrepancy in results. Buckland et al. (2013) and Malik et al. (2009) examined the effect of differing nitrogen rates applied at multiple times throughout the growing season on onion thrips densities, whereas our study examined the effect of nitrogen rates only applied at planting, which is the typical practice in New York. At-plant or pre-plant rates of nitrogen are vulnerable to biological and physical processes including leaching, run-off, and volatilization (Haynes 2012). Therefore, nitrogen applied at planting may not be present later in the season for onion plant uptake. Our study was conducted on 'muck' soil, which differs from the mineral soil types studied in Buckland et al.(2013) and Malik et al.(2009). 'Muck' soil is characteristically nutrient-rich and can consist of 20-80% organic matter (NRCS, 2016; Wilson and Townsend, 1931). These high levels of organic matter can provide substantial amounts of nitrogen to supplement plant growth throughout the growing season (Haynes 2012). Furthermore, Gonzalez et al. (2016) found that onions grown in histosol soil types can have differing responses to nitrogen amendments, with some requiring very low amounts of nitrogen. Perhaps, the currently recommended nitrogen rates for onion production in muck soils are too high. Thus, the rates evaluated in our study may still have been too high to detect noticeable differences in plant growth, thus resulting in a lack of significant differences in thrips densities.

Larval densities were significantly impacted by insecticide program. The lowest larval densities were recorded in action threshold-based and standard insecticide programs in all cultivars, and in most cases, the insecticide programs preformed equivalently.

Densities of larvae were reduced by up to 83% in plots treated with insecticides compared with untreated controls. These results are consistent with past and recent reporting on action thresholds to manage onion thrips in onion (Hoffmann et al., 1995; Nault and Huseth 2016). As predicted, fewer insecticide applications were made in action threshold treatments. Across all cultivars, frequency of insecticide applications was reduced between 33-50%. The function of an action threshold is generally not to provide better or even equivalent control as that provided by a standard (or weekly) insecticide program, rather it is to maintain pest densities below an economic injury level (Parrella and Lewis 1997; Pedigo et al., 1986; Stern et al., 1959). Thus, the difference in insecticide application frequency between standard and action threshold treatments can be considered excessive (and maybe even unnecessary) as it does not provide substantially better control of onion thrips. Our results continue to support that timing of insecticide applications based on an action threshold can provide effective control of onion thrips.

The least amount of thrips damage was consistently observed in standard insecticide programs in comparison with action threshold and untreated control treatments, suggesting that weekly-applied insecticide applications reduced visual damage on onion plants. These trends are consistent with previous records of visual feeding damage (Nault and Shelton 2010, Nault and Huseth 2016). In one case in ‘Bradley’, we observed significantly higher levels of damage in plots treated with insecticides following an action threshold and supplemented with higher rates of nitrogen. This result was not consistent between years.

Although, statistical comparisons were not made among data sets for different cultivars, low numbers of onion thrips were observed in ‘Avalon’, the moderately thrips-resistant cultivar, while high numbers of larval thrips were recorded in ‘Bradley’, the most thrips-susceptible cultivar. Our results corroborated those in previous studies that showed reduced onion thrips densities on yellow-green onion cultivars that had low levels of cuticular wax as is characteristic of ‘Avalon’ (Boateng et al., 2014; Damon et al., 2014; Diaz-Montano et al., 2012a). In 2016, larval densities in action threshold treatments in ‘Bradley’ were significantly higher than in standard insecticide treatments. These results may suggest that thrips-susceptible cultivars like ‘Bradley’ will foster onion thrips densities that build more rapidly and reach higher levels (even within the span of a week) compared with those like ‘Avalon’.

4.2 IYSV severity and incidence

IYS differed between 2015 and 2016, as earlier symptom incidence and greater severity was recorded in 2016. Variability in symptom expression and incidence of IYS among years is common (Diaz-Montano et al., 2012b; Muñoz et al., 2014). While specific IYSV isolates can impact symptom expression (Bag et al., 2012; Bulajić et al., 2009), we believe that the variable incidence was attributed to the hot, dry weather in 2016 (Table 1). Additional stress to the plants, especially limited soil moisture, may increase the presence of virus symptoms (Gent et al., 2006). Therefore, to reduce IYS symptom incidence and severity, thrips management will be more important when environmental conditions are unfavorable for onion growth.

Insecticide program generally had the largest impact on IYS severity and incidence, indicating that either program (action threshold or standard) will delay IYS incidence and reduce severity of IYS. Conversely, onions that do not receive protection are likely to develop IYS sooner and with greater severity by the end of the season. Management of IYSV is currently lacking control strategies (Gent et al., 2006), and as a result many growers have adopted more conservative insecticide programs. However, our results indicate that growers can continue to use action thresholds and not experience greater levels of IYS compared with more insecticide-intensive strategies.

In ‘Bradley’, nitrogen rate significantly impacted IYS severity in the action threshold treatment supplemented with 140 kg N ha-1. We suspect that this increase in severity was associated with more onion thrips larvae in the same treatment (Figure 2C).

4.3 Bacterial rot

The incidence of bacterial rot was not consistently impacted by levels of nitrogen applied at planting nor the type of insecticide program followed. While high rates of nitrogen can predispose onions to bacterial rot (Pfeufer et al., 2015; Wright 1993), we did not consistently observe this trend in our study. Additionally, leaf nitrogen levels were nearly identical among nitrogen treatments at several phenological stages, indicating that nitrogen rate at planting did not play a significant role in bacterial rot development. Initially, we hypothesized that onions that did not receive insecticide application may have a higher risk of developing bacterial rot because thrips would either directly transmit the bacteria or create entry wounds for the bacteria (Dutta et al., 2014). However, this relationship was not observed, as insecticide program did not have a consistent effect on bacterial rot incidence in any cultivar in both years. Additionally, the causal organisms of bacterial center rot, P. agglomerans or P. ananatis, were uncommon and not detected at greater levels in untreated plots than treated ones.

Rather, multiple other bacterial species were isolated from rotten bulbs in our study and there was no trend for a particular species to be associated with a particular treatment. As many others have suggested, bacterial rot and blights are caused by a complex array of many variables including climatic conditions, irrigation, mulches, fertilizer rate and type, storage time and temperature, and curing time (Batal et al., 2015; Gitaitis et al., 2004; Schroeder et al., 2012; Schroeder and du Toit 2010; Schwartz et al., 2003; Teviotdale et al., 1989; Vahling-Armstrong et al., 2016). Our results indicated that onion thrips management is unlikely to impact the incidence of bacterial bulb rot in New York.

Bacterial bulb rot levels increased three months after harvest, especially in 'Avalon'. Similar to other reports, we observed a consistent positive relationship between bacterial rot incidence and time in storage, with almost 18 times more rotten bulbs when compared with levels at harvest in some cases (Gitaitis et al., 2004; Schroeder and du Toit 2010; Schroeder et al., 2012). High levels of rot were recorded in 'Avalon', with some treatments reaching as high as 30%. The tolerance for bacterial rot in commercial onion production is very low; levels greater than 5% are unacceptable. Multiple cultivar trials have determined that 'Avalon' has a greater predisposition to bulb rot when compared with other cultivars (McDonald et al., 2013; Shock et al., 2015). While no studies have indicated a reason for this predisposition, we suggest that the difference may be due to a low level of cuticular wax on 'Avalon'. Increased disease susceptibility has been reported in onion cultivars with lower levels of wax. Mohan and Molenaar (2005) found higher levels of powdery mildew (Leveillula taurica) infection on onion cultivars with lower amounts of epicuticular wax. Additionally, we observed that Avalon had high levels of leaf dieback near harvest, with 90% and 39% of leaves dead in 2015 and 2016, respectively (data not shown). Thus, premature plant mortality due to leaf dieback may make plants more vulnerable to pathogenic bacteria. This finding underpins the importance of holistically evaluating an integrated pest management program for insects and diseases before commercial implementation, as certain components may improve pest or disease control, but may negatively impact other pests or pathogens in a production system.

4.4 Onion bulb yield

Yields were similar between years, even with drought conditions in 2016. Consistently, yield was only significantly impacted by insecticide program. Greatest yields were recorded in action threshold and standard insecticide programs. Similar to those results reported by Hoffmann et al. (1995) and Nault and Huseth (2016), bulb yield weights were statistically similar when following action threshold and standard insecticide programs. Of particular note was the lack of yield differences in the two insecticide programs in ‘Bradley’ in 2016. Action threshold treatments had statistically higher densities of larvae, approximately 2.5 times more thrips per leaf, compared with the standard insecticide program, but did not experience a yield reduction. Thus, onion thrips densities and damage can be successfully maintained below economic thresholds (i.e., 2.2 thrips larvae per leaf [Fournier et al.1995]) using action thresholds to determine if and when an insecticide application is necessary.

In 2015, yields in ‘Avalon’ were not significantly affected by any treatment. The lack of significant differences between treatments is likely due to a late-season outbreak of Stemphylium leaf blight (caused by Stemphylium versicarium), a serious, emerging disease of onion in New York. The disease has been reported to cause losses in onion between 80-85% (Tomaz and Lima 1986). In 2015, we recorded high levels of Stemphylium leaf blight lesions and leaf dieback late in the season in ‘Avalon’ (data not shown), compared with the other cultivars. Therefore, we believe the disease confounded our ability to see significant differences in marketable yield in ‘Avalon’ in 2015.

Because onion thrips feeding and IYSV infection occur simultaneously, we were unable to distinguish the impact of each on yield loss. Yield reductions were likely caused by a combination of IYSV and thrips feeding. We did observe a negative association between IYS severity and bulb yield. Specifically, we observed reduced yields in those onions that displayed higher severity ratings (data not shown). Lowest yields were recorded in untreated controls where thrips surpassed a seasonal mean of 2.2 thrips larvae per leaf. This is consistent with economic threshold levels reported from trials conducted with onions grown on ‘muck’ soil types in the Great Lakes region, which suggests thrips densities per leaf greater than 2.2 would result in yield reductions (Fournier et al., 1995). Yield reductions in untreated controls may not only be caused by the amount of onion thrips feeding, but also when feeding occurs during the development of the crop. Consistently, we observed peaks in onion thrips larval densities mid to late in the growing season when onions were actively bulbing (onions between 4-7 leaves), which has been reported to be a vulnerable time for onion bulb development (Kendall and Capinera 1987; Waiganjo et al., 2008).

Various at-plant rates of nitrogen did not have a significant impact on either larval onion thrips densities or onion bulb yield. As indicated above, the lack of positive yield responses to increased nitrogen rates is likely due to fertilizer application timing and soil type. Typically, commercial onion growers in New York apply 112 kg N ha-1 to 168 kg N ha-1 at planting. However, according to our results, at-plant nitrogen rates should be reduced as increased rates of nitrogen did not increase yield. Previous fertility studies have found that lower rates of nitrogen fertilizer, 50 to 120 kg N ha-1, are needed on muck soil types in comparison to mineral soil types (Harmer and Lucas 1956).

5. Conclusions

This study provides evidence that onion thrips and certain associated plant pathogens can be managed effectively in onion with reduced insecticide input. Consistently, we reported that an action threshold-based insecticide program provided equivalent levels of thrips control, IYS suppression, and marketable bulb yields as compared to those following a standard (weekly) insecticide program. Yet, 33-50% fewer insecticide applications were made in the action threshold-based program than the standard program. Additionally, nitrogen levels at planting can be reduced as there was no evidence that marketable yields were improved using the current recommended rates. Although benefits of reducing thrips damage with lower rates of nitrogen applied at planting were not observed in our study, growers can benefit by using less nitrogen at planting without compromising yield, which will decrease input costs. Most importantly, adoption of action thresholds and reduced levels of nitrogen at planting could reduce harmful non-target effects and slow the onset of insecticide resistance, thus contributing to the long-term sustainability of onion production.

‘Avalon’, the moderately thrips-resistant cultivar, had low seasonal mean densities of onion thrips larvae and severity and incidence of IYS. However, the percentage reduction in insecticide applications following the action threshold treatment relative to the standard insecticide program was similar to those for the other cultivars, suggesting that despite the moderate thrips resistance, insecticide application frequency may not be reduced. ‘Avalon’ also had high rates of bacterial rot. Future screening of cultivars for thrips and IYSV resistance should consider additional plant pathogens to comprehensively assess its best fit for commercial adoption.

Acknowledgements

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CHAPTER 2

OPTIMIZING IPM: A CASE STUDY MANAGING ONION THRIPS AND BACTERIAL BULB ROT IN ONION

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Abstract (400 words):

Most agricultural production systems face challenges with multiple pests, but few studies address the impact of multiple management tactics to control multiple insects and/or plant pathogens. Onion thrips and bacterial bulb rot are primary constraints to onion production, and choice of onion cultivar, fertility regime and insecticide use may be important tactics for both pests. Identifying the optimal combination of onion cultivar, fertility regime and insecticide use to manage onion thrips and bacterial bulb rot is not known. In a two-year study in New York, field trials independently evaluated the effect of a nitrogen and phosphorous fertilization regime combined with different onion cultivars and insecticide use patterns on onion thrips infestations and bulb rot incidence. In one study, five rates of nitrogen (0 kg ha^{-1}, 67 kg ha^{-1}, 84 kg ha^{-1}, 118 kg ha^{-1}, and 151 kg ha^{-1}) were combined with either a moderately thrips-resistant cultivar ('Avalon') or a thrips susceptible cultivar ('Bradley') and two season-long insecticide use patterns (untreated control or action threshold-based insecticide program). In a second study, four rates of phosphorus (0 kg ha^{-1}, 56 kg ha^{-1}, 112 kg $ha^{-1,}$ and 168 kg ha^{-1}) were combined with the same onion cultivars and insecticide programs mentioned above. In both years, 'Avalon' experienced lower thrips densities, but suffered 58% more bacterial rot, which reduced onion yields overall by 9%. Nitrogen and phosphorus fertilizer had limited impact on onion thrips, bacterial rot, and onion yield. Thrips densities were not affected by phosphorus fertilizer and reduced rates of nitrogen only marginally reduced densities in 'Avalon', but not in 'Bradley' in 2017. Nitrogen fertilizer consistently impacted bacterial bulb rot in 2017, and plants fertilized with nitrogen had 12 times the bacterial rot compared to unfertilized plants. In both years, low rates of nitrogen and phosphorus fertilizer (67 kg/ha N or 56 kg/ha P) produced statistically similar yields to plants supplemented with highest rates of fertilizer. Insecticide use reduced thrips densities and increased bulb yield in both years but did not consistently reduce bacterial bulb rot. Our results suggest that growers should adopt reduced fertility regimes paired with an action-threshold based insecticide program to optimize onion production, but we caution the use of thrips-resistant onion cultivars in our production system as they may be more susceptible to bacterial rot. Furthermore, our results indicated that IPM programs should be evaluated to consider multiple pests within an agricultural production system as IPM tactics can be counterproductive.

Keywords: IPM, *Thrips tabaci*, *Allium cepa*,

Highlights (85 characters)

- ‘Avalon’ experienced fewer thrips, but greater levels of bacterial rot
- Fertilizer amendments had little impact on onion thrips, bacterial rot, and yield
- Insecticide use reduced thrips and increased yield, but did not impact bacterial rot
- IPM program evaluation should consider the effect of tactics on multiple pests

1. Introduction

Integrated pest management (IPM) is the primary paradigm to manage pests in agriculture. IPM combines management tactics with the aim of reducing pest damage, maximizing crop yield and limiting negative off-target effects (Pedigo 1989; Pedigo et al.1986; Stern et al.1959). However, IPM strategies for a crop typically focus on a single insect or plant pathogen, with little consideration for other pests or pathogens. An agricultural production system is dynamic and a crop within a system faces challenges with multiple insect pests and plant pathogens simultaneously in a season. Consequently, management tactics that focus on a single insect or plant pathogen could create or exacerbate problems managing other pests or pathogens of that crop. Such a scenario would be a disservice to practitioners of IPM (i.e. growers, land managers) and ultimately hinder the sustainability of agriculture (Kogan 1998). Therefore, it is critical to develop an IPM program that reduces economic damage caused by multiple pests to provide the greatest overall benefit to sustainable crop production (Kogan 1998).

While addressing the impact of an IPM program on multiple pests would be ideal, the complexity of doing so is typically logistically prohibitive. Rather, a more reasonable approach would be to focus on the most significant pests and pathogens. In onion production, several major pests and pathogens damage the crop (Schwartz and Mohan 2008; Brewster 2007), but onion thrips (*Thrips tabaci*) and bacterial bulb rots (many spp.) are the most destructive and difficult to control. Onion thrips feed directly on leaf tissue and use their rasping-sucking mouthparts to remove mesophyll tissue. Onion thrips feeding can reduce bulb yields by 60% as well as transmit and spread plant pathogens and exacerbate plant diseases (Rueda et al. 2006; Gill et al. 2015; Leach et al. 2017). Bacterial bulbs rots are significant plant diseases that cause yield losses as high as 75% (Stivers 1999). Most onion diseases limit photosynthesis and reduce bulb size, but bacterial rots compromise the internal integrity of bulbs, rendering them unmarketable. Moreover, onion thrips have been positively associated with bacterial bulb-rot causing species, *Pantoea ananatis* and *Pantoea agglomerans*, and studies suggest thrips may play a critical role in the epidemiology of bacterial leaf blight and bacterial center rot in onion fields (Dutta et al.2012; Grode et al.2016; Grode et al.2019). However, bacterial bulb rot is caused by a complex of bacterial species, which vary based of the onion-production region. In New York (USA), *Burkholderia spp*., *Enterobacter cloacae*, *Pantoea ananatis* and *Rahnella spp.* have been identified as the primary bacterial pathogens of onion (Beer et al.2010).

Host plant resistance is a cornerstone of IPM, as it aims to prevent insect and pathogen damage (Pedigo et al.1989). Many studies have evaluated the performance of onion cultivars against onion thrips infestations and damage (Ferreira et al. 2017; Njau et al. 2017; Boateng et al., 2014; Damon et al. 2014; Diaz-Montano et al., 2012). No cultivars have been identified as completely resistant to thrips feeding, but some have moderate resistance and support lower densities and less feeding damage than others. Two onion cultivar characteristics positively related to thrips resistance include leaf waxiness and leaf color. Cultivars with semi-glossy wax and yellow-green leaves tend to have fewer thrips than those with waxy, blue-green leaves (Boateng et al., 2014; Diaz-Montano et al., 2012). Damon et al. (2014) identified a ketone, hentriacontanone-16 (H16), that is positively associated with epicuticular wax production in onion and higher levels of H16 may be responsible for thrips preference for waxier cultivars. Onion cultivars differ greatly in susceptibility to bacterial rot (Stumpf et al. 2017; Wohleb and Waters 2016; Schroeder et al. 2010); however, no plant characteristics have been identified as responsible for this variation. Some studies have postulated that epicuticular wax may play a significant role in onion disease susceptibility (Mohan and Molenaar 2005; Leach et al. 2017).

Crop fertilization can impact the attractiveness and susceptibility of crops to pests and pathogens (Abawi and Widmer 2000; Altieri et al. 2003). Previous studies have shown that onion thrips populations in onion decrease between 23-70% with decreased rates of nitrogen (Buckland et al. 2013; Malik et al. 2009), while Chen et al. (2004) found 2.3 times fewer thrips (*Frankliniella spp.*) on plants on impatiens flowers (*Impatiens wallerana*) when fertigated with lower rates of phosphorus (1.28 mM P vs. 0.32 mM P.). Thus, a reduction in nitrogen and phosphorus fertilizers also may be an effective cultural control tactic for onion thrips in onion.

Increased nitrogen fertilization can increase the incidence of bacterial bulb rots and reduce onion bulb quality (Wright et al. 1993; Diaz-Perez et al. 2003). Pfeufer et al. (2018) found that early-season nitrate levels as well as foliar nitrogen values in onion were positively related to the incidence of bacterial bulb rots. Thus, nitrogen fertilizer amendments may be important to consider when managing bacterial bulb rots. Currently, the relationship between phosphorus levels and bacterial bulb rot is understudied, although some research has indicated that bulb rot may increase with increasing rates of phosphorus fertilizer (Shock et al. 2014; Bekele et al. 2018).

Insecticide use is the primary tool for managing onion thrips in onion and multiple active ingredients and season-long program guidelines are available (Nault and Shelton 2010;

Nault 2015; Werling and Szendrei 2015). Previous studies have evaluated the integration of insecticide use and onion cultivar for onion thrips management. Nault and Huseth (2016) showed that integrating partially thrips-resistant cultivars into insecticide programming resulted in 36% fewer insecticide applications compared with managing thrips with insecticides on a thrips-susceptible cultivar. The use of insecticides to reduce onion thrips damage has been touted as a potential means of indirectly reducing bacterial bulb rot in onion (Grode et al. 2019). Further research is needed to determine if insecticide use will indirectly and successfully reduce the incidence of bacterial bulb infections.

While previous studies have evaluated management tactics for either onion thrips or bacterial bulb rot alone, none have considered the impact of multiple management tactics in concert to manage both. There is a need to identify a robust IPM program using insecticides, thrips-resistant cultivars and reduced rates of fertilizer that will effectively manage both of these major biotic constraints to onion production. The purpose of our study was to evaluate the effect of reduced fertility regimes paired with different onion cultivar and season-long insecticide use combinations on 1) onion thrips densities 2) bacterial bulb rots, and 3) onion bulb yield. We hypothesized that reduced levels of nitrogen and phosphorous paired with a thrips-resistant cultivar ('Avalon') and an action-threshold based insecticide program would provide optimal management of onion thrips and bacterial bulb rot, thereby increasing marketable bulb yield.

2. Materials and Methods

2.1 Experimental design

Trials were conducted on a commercial onion farm in 2017 and 2018 (Orleans County, NY) ('Carlisle' muck soil type, NRCS, 2016). Two independent trials were executed simultaneously to evaluate either the effect of nitrogen with different onion cultivar and insecticide use combinations or the effect of phosphorous with different onion cultivar and insecticide use combinations. Field sites for nitrogen and phosphorus trials were selected based on low initial values of soil nitrate and soil phosphorus, respectively. Two onion cultivars that differ in their resistance to onion thrips were selected based on leaf waxiness and color (Damon et al., 2014; Diaz-Montano et al., 2012a). 'Avalon' (Crookham Co., Caldwell, ID) is moderately resistant to thrips feeding and has yellow-green, semi-glossy foliage, whereas 'Bradley' (Bejo Seeds, Inc., Oceano, CA) has blue-green, waxy foliage that is susceptible to thrips (Leach et al. 2017). Both cultivars are intermediate to long-day, yellow onions with similar days to harvest. Nitrogen and phosphorus trials were planted with a vacuum seed planter (646,000 onion seeds per hectare) on 15 Apr 2017 and 21 Apr 2018 (see 2.5 for management of other pests and pathogens).

2.1.1 Nitrogen trial

A total of 20 treatments (2 onion cultivars x 5 nitrogen rates x 2 insecticide treatments) were replicated 5 times. Onion cultivars were 'Bradley' and 'Avalon'; nitrogen rates were 0, 67, 84, 118 and 151 kg ha-1; insecticide treatments were treated with insecticide and an untreated control. The treatments were arranged in a split-split plot design in which cultivar was the main plot factor, nitrogen fertilizer was the sub-plot factor and insecticide was the sub-sub-plot factor. Cultivars were arranged in long rows across the field. Nitrogen treatments were randomly assigned to plots nested within each cultivar row and each plot was bisected into subplots randomly assigned to the insecticide treatments.

Urea nitrogen (46-0-0) was applied twice, at-planting and during the pre-bulbing stage (3-5 leaves per plant). Rates and timings were 0 kg ha^{-1} (no nitrogen applied), 67 kg ha^{-1} (67 kg ha^{-1} applied at planting), 84 kg ha-1 (split into two applications, 67 kg ha^{-1} applied at planting and 17 kg ha^{-1} applied pre-bulbing), 118 kg ha-1 (split into 67 kg ha^{-1} applied at planting and 51 kg ha^{-1} applied pre-bulbing), and 151 kg ha^{-1} (split into 67 kg ha^{-1} applied at planting and 84 kg ha^{-1} applied pre-bulbing). To reduce the chance of urea fertilizer volatilizing, 3 cm of overhead irrigation was applied immediately after the fertilizer was applied. Experimental plots were supplemented at planting with the appropriate rates of potassium (potassium chloride; 0-0-60; N-P-K) and phosphorus (triple superphosphate; 0-46-0; N-P-K) per current soil tests and corresponding fertility guidelines. Fertilizers were broadcast and raked in. Each experimental plot was 1.5 m wide x 9.1 m long and consisted of five rows of onion plants, and subplots within each plot were 1.5 m wide x 4.55 m long. The entire experiment was 32 m wide x 52 m long. Experimental plots were surrounded by either 1.5 m of bare ground or unfertilized onions to minimize movement of fertilizer between plots.

Experimental subplots receiving insecticide were sprayed when the onion thrips population met or surpassed an action threshold of 1 larva per leaf (Nault and Huseth 2016; Nault and Shelton 2010). The untreated control was never sprayed with insecticides. Subplots were scouted weekly beginning on 19 Jun 2017 and 19 Jun 2018, and insecticide program treatments were initiated when treatments reached a mean density of approximately 1 larva per leaf on 8 Aug 2017 and 1 Jul 2018.

Insecticide applications were made in accordance with current insecticide resistance management guidelines (Leach et al. 2018). Decisions to apply insecticides were made on a weekly basis. The following sequence of insecticides and rates were used during each experiment: spirotetramat at 0.08 kg (AI) ha-1 (Movento; Bayer CropScience, Research Triangle Park, NC), cyantraniliprole at 0.1 kg (AI) ha-1 (Exirel; DuPont, Wilmington, DE), and spinetoram at 0.07 kg (AI) ha-1 (Radiant SC; Dow AgroSciences, Inc., Indianapolis, IN). Each insecticide was applied no more than twice consecutively if the thrips density exceeded the action threshold; if the action threshold was not exceeded for a week, no insecticide was applied. Insecticides were applied with a CO_2-pressurized backpack sprayer with four, twin flat-fan nozzles (TJ-60-8003VS; TeeJet Technologies Harrisburg, PA) calibrated to deliver 140 liters per acre at 276 kPa. All insecticides were co-applied with an adjuvant at 0.5% v:v (Induce; Helena, Collierville, TN) to increase efficacy (Nault et al., 2013).

2.1.2 Phosphorus trial

A total of 16 treatments (2 onion cultivars x 4 phosphorus rates x 2 insecticide treatments) were replicated 5 times. The same onion cultivars and insecticide treatments evaluated in the nitrogen trial were included in this trial. Similarly, the treatments were arranged in a split-split plot design in which cultivar was the main plot factor, phosphorous fertilizer was the sub-plot factor and insecticide was the sub-sub-plot factor. Arrangement of the cultivars, phosphorus treatments and insecticide applications were the same as described in the nitrogen trial. Phosphorus rates were 0, 56, 112, and 168 kg ha^{-1}. Triple superphosphate (0-46-0; N-P-K) was applied at planting. Experimental plots were supplemented at planting with the appropriate rates of nitrogen (Urea; 46-0-0; N-P-K) and potassium (potassium chloride; 0-0-60; N-P-K) per current soil tests and corresponding fertility guidelines. All fertilizers were broadcast and then incorporated into the soil. Plots were the same size and orientation as those in the nitrogen trial; the total area of the trial was 32 m wide x 41 m.

Insecticide applications and initiation of the insecticide sequence was executed in the same manner as described in the nitrogen trial. Subplots were scouted weekly beginning on 19 Jun 2017 and 19 Jun 2018, and insecticide program treatments were initiated when treatments reached a mean density of approximately 1 larva per leaf on 8 Aug 2017 and 1 Jul 2018. In 2017, 'Avalon' did not surpass the action threshold at any point, and thus no insecticide was applied.

2.2 Nitrogen and phosphorus assessments

Soil nitrate and phosphorus assessments were completed at three developmental stages: pre-bulbing (3-5 leaves per plant), bulbing (5-8 leaves per plant), and post-bulbing (9+ leaves per plant). A total of ten soil samples per plot were collected from the surface (0-20 cm) and subsurface (20-30 cm) using a soil probe. Soil samples from each plot were homogenized and submitted for testing within 24 hr of sampling (Dairy One, Lansing, NY). Soil nitrate was determined using the RQflex® Reflectometer method (EMD Chemicals Inc., One International Plaza, Suite 300, Philadelphia, PA) and soil phosphorus with the Bray and Kurtz (1945) method.

Onion plant growth was evaluated in the nitrogen and phosphorus trials during each developmental stage. Three plants from each plot were randomly selected and removed at each stage (pre-bulbing, bulbing, and post-bulbing) (n=15 plants per treatment/developmental stage). The number of leaves, length of longest leaf, and weight of each plant were measured.

2.3 Onion thrips population assessments

In both fertility trials, the number of larvae were counted weekly in every subplot. Only onion larvae were recorded, as previous studies have suggested that adults do not significantly contribute to crop damage (Leach et al. 2017; Coudriet et al. 1979). Ten plants, randomly selected from the inner three rows, were visually examined for thrips larvae. Counts began early in the growing season, when plants had approximately 3-4 leaves, and concluded when 80% or more of the plants matured. Thrips were monitored for the same duration in both the nitrogen and phosphorus trials, 10 weeks in 2017 and 8 weeks in 2018. Numbers of onion thrips larvae were binned into three sampling periods based on onion development; pre-bulbing (19 June to 10 Jul 2017, 19 June to 10 Jul 2018), bulbing (11 Jul to 7 Aug 2017; 11 Jul to 31 Jul 2018), and post-bulbing (8 Aug to 28 Aug 2017; 1 Aug to 15 Aug 2018). Voucher specimens are held at Cornell AgriTech in Geneva, NY.

2.4 Bacterial rot assessment

Within-season assessment. In mid to late season, plants in the nitrogen and phosphorus trial were visually examined for bacterial rot symptoms. Plants were selected from the inner three rows of onions in each subplot and the number of infected plants counted. Onions displaying typical bacterial rot symptoms including, bleached, wilted inner leaves were considered infected (Schwartz and Mohan 2008). Subplots were evaluated on two dates during the growing season, 30 Jul 2017 and 15 Aug 2017;

1 Aug 2018 and 15 Aug 2018. In 2017, the number of onions with bacterial rot symptoms was only assessed in untreated control subplots, whereas all insecticide treatment sub plots were assessed for bacterial rot in 2018.

At harvest assessment. Onions were cured in the field for at least one week before they were evaluated for bacterial bulb rot (see details about harvest below). A subsample of 40 bulbs (diameter of < 5 cm, weight of < 90g) were cut longitudinally and inspected for bacterial decay. Incidence of bacterial rot was determined for each subplot (*n* rotten onion bulbs/total onion bulbs). Bacterial species were identified from a random subsample of 20 onion bulbs per treatment that were symptomatic for bulb rot. Bacteria from a subset of symptomatic bulbs were recovered using a semi-selective onion extract medium (Zaid et al., 2012), and pathogenic bacteria were identified by PCR (Asselin et al. 2016).

2.5 Management of other pests and pathogens

Onion plants in both the nitrogen and phosphorus trials were managed to reduce damage by other pests in the production system. To ensure crop establishment, seeds were treated with FarMore FI500 (Mefenoxam (0.15 g ai/kg), Fludioxonil (0.025 g ai/kg), Azoxystrobin (0.025 g ai/kg), Spinosad (0.20 mg ai/seed), Thiamethoxam (0.2 mg ai/seed)) and Pro-Gro (Carboxin (7.50 g ai/kg) and Thiram (12.50 g ai/kg)). This seed treatment package does not impact either onion thrips or bacterial rot. Other than onion thrips, no other insect pests damaged onions in this experiment. Symptoms of iris yellow spot disease, which is caused by iris yellow spot virus and transmitted by onion thrips, was nearly absent in 2017 and very low in 2018. Weeds and foliar plant pathogens like botrytis leaf blight (*Botrytis* spp.) and Stemphylium leaf blight

(*Stempylium vesicarium*) were successfully managed using pesticides following Cornell vegetable management guidelines and recommendations (Reiners et al. 2017).

2.6 Onion bulb yield

For each fertility trial, onion plants were undercut when 80% or more of each onion cultivar had senesced or died and then cured in the field for at least one week before harvest. Onions were harvested on 30 Aug 2017 and 18 Aug 2018. Bulbs were graded by bulb diameter and assigned a size class of either ‘boiler’ (2.5 cm-4.8 cm), ‘standard’ (4.9 cm-7.6 cm), or ‘jumbo’ (≥7.7 cm) and then weighed. Bulbs that were either ‘standard’ or ‘jumbo’ were considered marketable, and ‘boiler’ bulbs unmarketable. Marketable yields were estimated on a mean metric ton per hectare basis by multiplying mean bulb weight in each size class by the density of plants in the plots, which was determined previously via onion plant stand counts for each cultivar each year. Adjusted marketable yields were calculated for each subplot by subtracting the percent of bulbs with bacterial rot from the estimated marketable yield (see 2.4 for bacterial rot assessment).

2.7 Statistical analysis

Data within each year were analyzed independently since environmental conditions and thrips pressure were different between years in both fertility trials (Supplemental table 3.1). Data were analyzed using a generalized linear mixed model (R package; ‘lme4’) (Bates et al. 2015). Generalized linear mixed models were fit with fixed effects of onion cultivar, fertilization rate, insecticide use, and all their interactions, and included random effects of row and plot nested within row as well as subplot nested within plot nested within row.

The number of larvae per leaf (total for the season, pre-bulbing, bulbing, and postbulbing densities) were analyzed assuming a negative binomial distribution. Plant weight, plant leaf length, number of leaves per plant, soil nitrate, marketable yield, and adjusted marketable yield were analyzed assuming a normal distribution. The number of onion plants with bacterial rot within the growing season was analyzed using a Poisson distribution. Bacterial rot incidences were analyzed as a binomial distribution (*n* rotten onion bulbs/total onion bulbs). No insecticide applications were applied in the 'Avalon' in the phosphorus trial in 2017, which precluded the three-way analysis between phosphorus rate, onion cultivar, and insecticide use. Treatments in each analysis were compared using least squared means ($P<0.05$) ('emmeans', Lenth et al. 2018).

3. Results

3.1 Nitrogen and Phosphorous levels in the soil and plants

3.1.1 Nitrogen trial

Nitrogen assessments in soil and plants. Soil nitrate levels were higher in 2017 than 2018 (35.2 ppm and 18.2 ppm in 2017 and 2018, respectively), but soil nitrate was positively associated with the amount of urea applied during each developmental stage (Supplemental table 3.2). Plots that received the highest rate of nitrogen, cumulative amount of 151 kg ha^{-1}, had the highest soil nitrate levels at every sampling period. Similarly, plots that did not receive nitrogen fertilizer had the lowest levels of soil nitrate throughout the growing season.

Onion plant growth characteristics were not significantly different between ‘Avalon’ and ‘Bradley’ in either year and data were pooled across cultivars. Overall, nitrogen fertilizer had a limited impact on plant growth. In 2017, onion plants fertilized with nitrogen were significantly heavier and had longer leaves compared with unfertilized plants (Supplemental table 3.2). Additionally, fertilized onions had a greater number of leaves (~7.3 leaves per plant) compared with those unfertilized (~6.2 leaves per plant) throughout the growing season, but this was only nearly significant (*P*=0.0547). However, in 2017, increased rates of nitrogen did not increase plant growth, as onions that received 67 kg ha^{-1} were statistically similar across all plant characteristics to onions that received 151 kg ha^{-1} (Supplemental table 3.2). In 2018, nitrogen fertilizer impacted plant growth during the prebulbing stages, but not bulbing or postbulbing stages (Supplemental table 3.2).

3.1.2 Phosphorus trial

Phosphorus assessments in soil and plants. Soil phosphorus levels were highest early in the growing season, during the prebulbing and bulbing stages, and lowest in the postbulbing stage (Supplemental table 3.3). Levels of phosphorus were higher in 2017 than 2018 (83.3 ppm in 2017 vs. 61.0 ppm in 2018), but levels were positively associated with the amounts of phosphorus fertilizer applied, although phosphorus treatments were not always statistically different (Supplemental table 3.3).

Similar to the nitrogen trial, onion plant growth characteristics were not significantly different between cultivars and data were pooled. Plant growth was not significantly impacted by phosphorus fertilization at any developmental stage ($P>0.05$) (Supplemental table 3.3). Mean number of leaves, leaf length, and plant weight tended to increase over the duration of the season in both years and reached maximum values in the post-bulbing or bulbing stages in 2017 and 2018, respectively.

3.2 Onion thrips densities

3.2.1 Nitrogen trial

Larval onion thrips densities increased as the season progressed both years; however, onion thrips pressure was greater in 2018 than 2017 (season total mean of 0.7 larvae/leaf in 2017 vs. season total mean of 10.6 larvae/leaf in 2018). On average, onion thrips densities were lowest during the pre-bulbing and bulbing stages and peaked during the post-bulbing stage (Supplemental figure 3.1).

Season-long effect, 2017. Season total onion thrips densities were significantly impacted by cultivar, nitrogen rate, insecticide use and the interaction of cultivar and nitrogen rate (Table 3.1). Unfertilized ‘Avalon’ and ‘Bradley’ had the highest mean season total number of thrips per leaf (0.8 thrips/leaf), and ‘Avalon’ fertilized with 67 kg ha^{-1} had the lowest mean season total larval density (0.2 thrips/leaf) (Figure 3.1a). Insecticide use also significantly impacted onion thrips (Table 3.1), and higher season mean total densities were recorded in the untreated control (0.59 ± 0.02 thrips per leaf) as compared with the insecticide treatment (0.48 ± 0.01 thrips per leaf).

Within-season effects, 2017. Onion cultivar, nitrogen rate, and the interaction of onion cultivar and nitrogen rate significantly impacted onion thrips densities during all developmental stages in 2017 (Table 3.2) (Supplemental figure 3.1a-d). During the prebulbing stage, onion thrips densities increased with increasing rates of nitrogen (Supplemental figure 3.1a) and thrips densities in ‘Bradley’ were significantly greater than those in ‘Avalon’ (Supplemental figure 3.1b). During the bulbing stage, onion thrips were significantly affected by the interaction of onion cultivar and nitrogen rate, and more thrips were recorded in unfertilized ‘Avalon’ and ‘Bradley’ than ‘Avalon’ fertilized with 67 kg/ha, 84 kg/ha or 118 kg/ha (Supplemental figure 3.1c). During the post-bulbing stage, results were similar to those during the bulbing stage where onion thrips in unfertilized ‘Avalon’ and ‘Bradley’ were significantly greater than fertilized ‘Avalon’ treatments (Supplemental figure 3.1d). Onion thrips densities remained below the action threshold for most of the growing season, and only one insecticide was applied during the postbulbing stage. Thus, insecticide use only significantly impacted onion thrips densities during postbulbing and significantly fewer thrips were recorded in the treated plots as compared with the untreated control (Supplemental figure 3.1e).

Season-long effect, 2018. Season total onion thrips densities were significantly impacted by insecticide and the interaction of cultivar and insecticide use, but not by either cultivar or nitrogen rate alone (Table 3.1). ‘Avalon’ treated with insecticide had the lowest seasonal thrips density (1.6 thrips/leaf) compared with insecticide-treated ‘Bradley’ (2 thrips/leaf) and untreated ‘Avalon’ and ‘Bradley’ (6.7 thrips/leaf) (Figure 3.1b).

Within-season effects, 2018. Onion thrips densities were significantly impacted by onion cultivar and insecticide use during the bulbing and postbulbing stages, but not the prebulbing stage (Table 3.2). During the bulbing stage, onion thrips were significantly impacted by the interaction of insecticide use and onion cultivar, and significant effects were similar to those reported for the seasonal means (Supplemental figure 3.1f). Onion cultivar and insecticide use each independently impacted onion thrips densities in the postbulbing stage (Table 3.2) (Supplemental figure 3.1g, 3.1h). Insecticides were applied 3 out of 3 weeks during the postbulbing stage when thrips populations were peaking, which resulted in significantly fewer thrips in treated plots compared to untreated plots. Fewer thrips were also reported in 'Avalon' as compared to 'Bradley' (Supplemental figure 3.1h).

3.2.2 Phosphorus trial

Similar to the nitrogen trial, larval onion thrips densities increased throughout the season in both years, but infestation levels were much higher in 2018 than 2017 (season total mean of 0.4 thrips larvae/leaf in 2017 vs. season total mean of 12.1 in 2018). Densities of thrips were lowest in prebulbing stages and highest in the postbulbing stages.

Season-long effect, 2017. Season total onion thrips densities was impacted by onion cultivar, but not by phosphorus rate, insecticide or any interactions (Tables 3.3). Thrips densities were reduced in 'Avalon' compared to 'Bradley' (Figure 3.1c).

Within-season effects, 2017. Only cultivar significantly impacted onion thrips densities during the growing season (Table 3.4). Significantly more thrips were recorded in 'Bradley' compared to 'Avalon' during the bulbing and postbulbing stages, but not prebulbing stages (Supplemental figure 3.2a, 3.2b). No insecticides were applied in 'Avalon' since weekly densities remained below the action threshold of 1 thrips per leaf. One insecticide application was applied in 'Bradley', and significantly fewer thrips were recorded in treated plots (0.4 ± 0.06) compared to untreated plots (0.6 ± 0.07) ($F_{1,28}=92.4$, $p<0.0005$).

Season-long effect, 2018. Season total onion thrips densities was significantly impacted by onion cultivar and insecticide use, but not by phosphorous rate or any of the interactions (Table 3.3.). Mean season total number of larvae per leaf in ‘Avalon’ was significantly lower than the season total for ‘Bradley’ (Figure 3.1c). Mean season total number of larvae per leaf in insecticide-treated plots was significantly lower than those not treated (Figure 3.1d).

Within-season effects, 2018. Onion thrips densities were significantly impacted by cultivar and insecticide use in the bulbing and postbulbing stages (Table 3.4). Similar to 2017, onion thrips densities were not significantly impacted by any IPM tactic in the prebulbing stage. During the bulbing stage, onion thrips were significantly impacted by the interaction of onion cultivar and insecticide use, and highest densities were recorded in untreated controls for both cultivars and lowest densities in ‘Avalon’ treated with insecticide (Supplemental figure 3.2c). Onion cultivar and insecticide use impacted onion thrips densities in the postbulbing stage (Table 3.4) (Supplemental figure 3.2d, 3.2e). ‘Avalon’ had fewer thrips per leaf as compared to ‘Bradley’. Additionally, insecticide use significantly reduced onion thrips densities during the postbulbing stage.

3.3 Bacterial rot

3.3.1 Nitrogen trial

Incidences of bacterial rot differed between years, and overall incidences of rot at harvest pooled across all treatments were 6.6% and 9.3% in 2017 and 2018, respectively. In 2017, the following bacterial species were detected: *Klebsiella oxytoca, Rahnella spp., Rouxiella spp., Pseudomonas spp., Pantoea agglomerans,* *Enterococcus spp., Lactobacillus plantarum,* and *Leuconostoc pseudomesenteroides* were detected. In 2018, the following bacterial species were detected: *Enterobacter ludwigii, Kosakonia cowanii, Burkholderia spp.,* and *Rahnella spp*.

Within-season. Onion cultivar significantly impacted the number of plants with bacterial infection in both years and nitrogen rate impacted numbers of infected plants in 2017; insecticide use had no impact in either year (Table 3.1). On average, twice as many 'Avalon' plants displayed symptoms of bacterial infection compared with 'Bradley' (Figure 3.2a). In 2017, the number of plants displaying bacterial symptoms increased with increasing rates of nitrogen (Figure 3.2b). The greatest number of infected plants was recorded in the two highest rates of nitrogen, followed by the intermediate rates and finally the unfertilized control.

At harvest. In 2017, nitrogen rate and insecticide use significantly impacted the percent bulbs with bacterial rot at harvest, but not cultivar (Table 3.1). Bulbs fertilized with nitrogen (67 kg ha^{-1} N or higher) experienced significantly greater levels of bacterial rot as compared with unfertilized onions (Figure 3.3a). Insecticide use also significantly impacted the incidence of bacterial rot, as insecticide treated plots had twice the amount of bacterial rot as compared with the percentage of rot in untreated plots (Figure 3.3b).

In 2018, onion cultivar and insecticide use significantly impacted bacterial bulb rot (Table 3.1). 'Avalon' had twice the amount of bacterial rot as 'Bradley' (Figure 3.3c). Insecticide use also influenced bacterial rot; however, the relationship was opposite as that observed in 2017. Plots treated with insecticide had significantly less rot as compared with levels in the untreated controls (Figure 3.3d).

3.3.2 Phosphorus trial

Greater levels of bacterial rot were detected in 2018 compared to 2017 (2017: 2.8% incidence of rot at harvest; 2018: 10.1% incidence of rot at harvest). Many bacterial species were isolated from bulb samples in 2017: *Rahnella spp., Pseudomonas spp., Pantoea agglomerans, Enterococcus spp.,* and *Lactobacillus plantarum*. In 2018, *Enterobacter ludwigii, Kosakonia cowanii, Burkholderia spp.,* and *Rahnella* species were detected.

Within-season. In both years, only onion cultivar significantly impacted the number of plants exhibiting bacterial rot symptoms. Phosphorus rate and insecticide use, and their interactions had no impact on plants with bacterial rot symptoms (Table 3.3). Overall, ‘Avalon’ had greater numbers of plants with bacterial rot symptoms in both years (Figure 3.2c).

At harvest. In 2017, onion cultivar, phosphorus rate, insecticide use, and the interactions between cultivar and phosphorous had no impact on the incidence of bacterial rot (Table 3). While only ‘Bradley’ was treated with an insecticide in 2017, interactions among some of the main effects on bacterial rot were omitted from the analyses. Nevertheless, insecticide use in ‘Bradley’ did not significantly impact the incidence of bacterial rot in 2017.

In 2018, percent bacterial rot was significantly affected by onion cultivar, but not phosphorus rate or insecticide use (Table 3.3). Greater percentage of bacterial rot was recorded in ‘Avalon’ (13.7 ± 1.2%) compared with ‘Bradley’ (6.4 ± 0.9%) (Figure 3.3e).

3.4 Onion yield

3.4.1 Nitrogen trial

In 2017, marketable yield was significantly affected by nitrogen rate and not onion cultivar or insecticide use (Table 3.1). Marketable yields were 88% greater in plots that received nitrogen fertilizer compared with those that did not, but all treatments with nitrogen fertilizer were statistically similar (Figure 3.4a). Adjusted marketable yield was significantly impacted by cultivar and nitrogen rate, but not insecticide use (Table 3.1). The effect of onion cultivar on adjusted marketable yield was low and adjusted marketable yields in ‘Bradley’ were 9% higher than those in ‘Avalon’ (Figure 3.5a). Fertilized treatments had 74% greater adjusted marketable yields compared to unfertilized treatments (Figure 3.5b).

In 2018, insecticide use but not onion cultivar or nitrogen rate significantly impacted marketable yields. Onion yield increased by 46% when treated with insecticide as compared to untreated controls (Figure 3.4b). Adjusted marketable yields were significantly impacted by cultivar, insecticide use and the interaction between onion cultivar and insecticide use. Untreated controls for both cultivars had the lowest adjusted marketable yield, followed by ‘Avalon’ treated with insecticide and then ‘Bradley’ treated with insecticide (Figure 3.5c).

3.4.2 Phosphorus trial

3.2.4 Onion yield

In 2017, marketable yield and adjusted marketable yield were significantly impacted only by phosphorus rate, but not onion cultivar, insecticide program or their interactions

(Table 3.2). Unfertilized controls had the lowest yields, whereas plots treated with 56 and 168 kg ha-1 P had 10-12% higher yields (Figure 3.4c, Figure 3.5d).

In 2018, yield was significantly affected by insecticide and the interaction between onion cultivar and insecticide (Table 3.3). Highest marketable yields were reported in treated plots for both cultivars, followed by untreated ‘Avalon’ and lastly untreated ‘Bradley’ (Figure 3.5b). Adjusted marketable yield was only impacted by insecticide treatment (Table 3.3). Treated plots had 42% greater adjusted marketable yields compared with untreated controls (Figure 3.5d)

Table 3.1: Summary ANOVA testing fixed effects of nitrogen, onion cultivar, and insecticide use and all interactions on onion thrips, bacterial bulb rot, and onion yield.

		Response variable									
		Onion thrips		Bacterial bulb rot				Onion yield			
Year	**Fixed effect**			Within season		At harvest		Marketable yield		Adjusted yield	
		df	*f*	*df*	*f*	*df*	*f*	*df*	*f*	*df*	*f*
2017	Cultivar	1, 76	12.7*	1, 36	**5.5***	1, 76	0.9	1, 76	1.8	1, 76	**5.2***
	Nitrogen	4, 76	24.3**	4, 36	**67.7****	4, 76	**19.7****	4, 76	**244.4****	4, 76	**97.5****
	Insecticide	1, 76	**101.2****	n/a	n/a	1, 76	**13.4***	1, 76	0.3	1, 76	2.1
	Cultivar x Nitrogen	4, 76	**11.4***	4, 36	7.9	4, 76	0.8	4, 76	5.0	4, 76	2.8
	Cultivar x insecticide	1, 76	0.1	n/a	n/a	1, 76	0.6	1, 76	0.7	1, 76	1.2
	Nitrogen x insecticide	4, 76	5.3	n/a	n/a	4, 76	0.9	4, 76	3.3	4, 76	4.4
	Cultivar x Nitrogen x insecticide	4, 76	3.3	n/a	n/a	4, 76	0.9	4, 76	8.8	4, 76	7.2
2018	Cultivar	1, 76	1.5	1, 76	**8.4***	1, 76	**12.6***	1, 76	0.2	1, 76	4.2*
	Nitrogen	4, 76	2.3	4, 76	0.7	4, 76	0.6	4, 76	1.8	4, 76	1.2
	Insecticide	1, 76	850.1**	1, 76	1.1	1, 76	**5.7***	1, 76	**264.1****	1, 76	246.2**
	Cultivar x Nitrogen	4, 76	2.7	4, 76	5.3	4, 76	0.3	4, 76	4.0	4, 76	5.8
	Cultivar x insecticide	1, 76	**5.9***	1, 76	0.3	1, 76	2.9 †	1, 76	1.8	1, 76	**4.9***
	Nitrogen x insecticide	4, 76	2.2	4, 76	2.4	4, 76	0.1	4, 76	2.4	4, 76	5.6
	Cultivar x Nitrogen x insecticide	4, 76	2.8	4, 76	1.3	4, 76	1.4	4, 76	1.7	4, 76	2.7

* and ** indicates significance at 0.05, <0.0005 respectively. † indicates marginal significance, 0.05-0.1.
Random effect structure included the effects of row (cultivar), plot nested within row (nitrogen rate), and subplot (insecticide use) nested within plot nested within row. Degrees of freedom calculated using the Satterthwaite approximation.

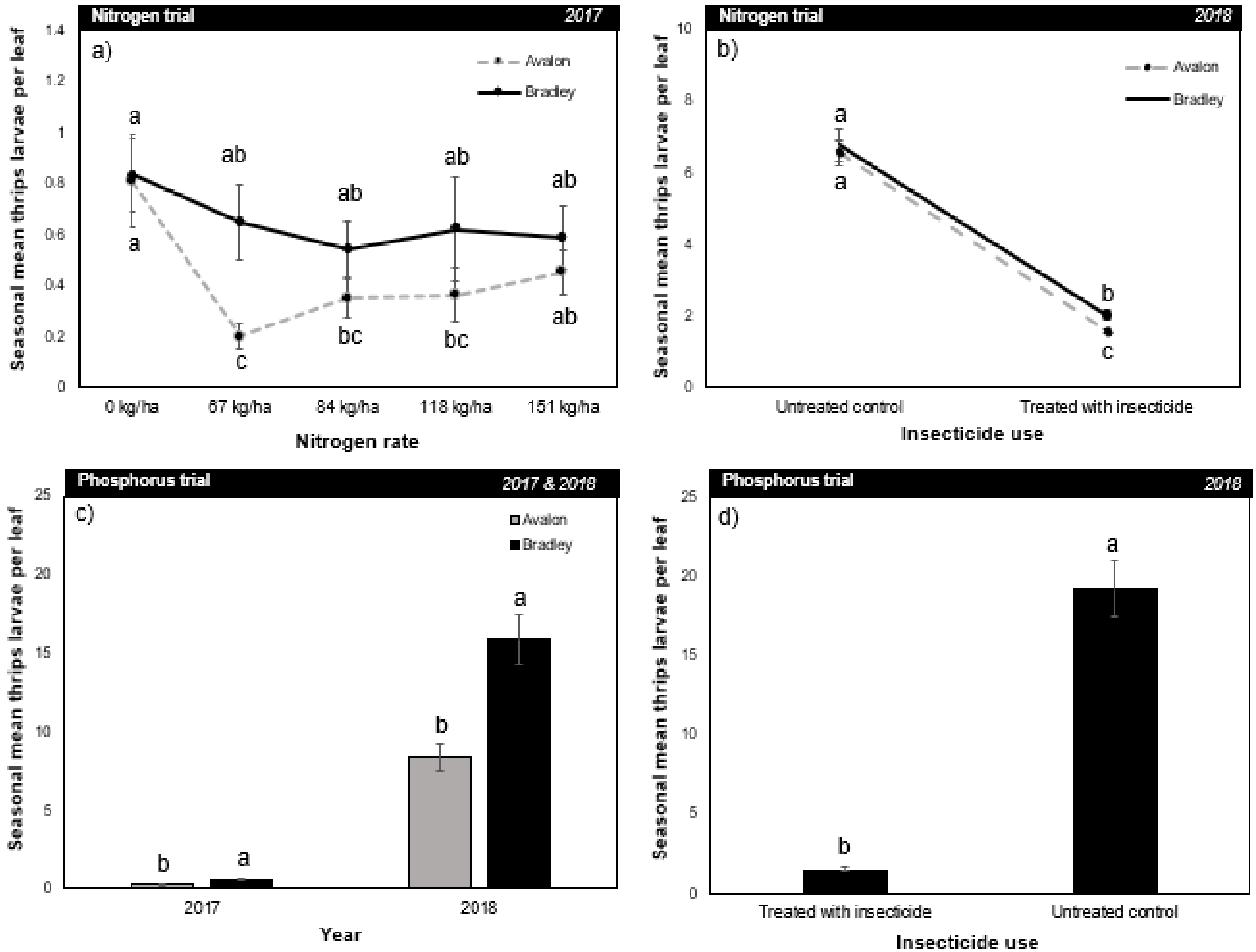

Figure 3.1: Significant effects of the interaction of cultivar and insecticide (a), and interaction of onion cultivar and nitrogen rate (b), onion cultivar (c), insecticide use (d) on seasonal larval onion thrips densities in 2017 (a, c) and 2018 (b, d). Two independent trials evaluated the effect of nitrogen with different onion cultivar and insecticide use combinations or the effect of phosphorous with different onion cultivar and insecticide use combinations. Five different rates of nitrogen were applied through the growing season; 0 kg ha-1, 67 kg ha-1, 84 kg ha-1, 118 kg ha-1 and 151 kg ha-1. Phosphorus rates were 0, 56, 112, and 168 kg ha-1. Fertilizer treatments were paired with two onion cultivars and two insecticide treatments. 'Avalon' is moderately thrips resistant and 'Bradley' is susceptible to thrips feeding. Every fertilizer treatments and onion cultivar combination were either treated with insecticide or left untreated. Significant effects determined by Type II ANOVA and means separated using LSMEANS at a 0.05 significance level.

Table 3.2: Summary ANOVA testing fixed effects of nitrogen, onion cultivar, and insecticide use and all interactions on onion thrips densities within season during three onion developmental stages, prebulbing, bulbing, and bulbing in 2017 and 2018.

		Developmental stage					
Year	**Fixed effect**	Prebulbing		bulbing		postbulbing	
		df	*f*	*df*	*f*	*df*	*f*
2017	Cultivar	1, 36	**7.4****	1, 36	19.0**	1, 76	25.4**
	Nitrogen	4, 36	**34.5***	4, 36	6.4	4, 76	32.1**
	Insecticide	n/a	n/a	n/a	n/a	1, 76	**196.4****
	Cultivar x Nitrogen	4, 36	4.0	4, 36	**16.1***	4, 76	**21.9****
	Cultivar x Insecticide	n/a	n/a	n/a	n/a	1, 76	0.4
	Nitrogen x Insecticide	n/a	n/a	n/a	n/a	4, 76	9.2 †
	Cultivar x Nitrogen x Insecticide	n/a	n/a	n/a	n/a	4, 76	2.6
2018	Cultivar	1, 36	1.2	1, 76	4.6*	1, 76	**84.1****
	Nitrogen	4, 36	3.0	4, 76	0.3	4, 76	0.5
	Insecticide	n/a	n/a	1, 76	1660.2**	1, 76	**2313.5****
	Cultivar x Nitrogen	4, 36	6.3	4, 76	2.2	4, 76	8.8 †
	Cultivar x Insecticide	n/a	n/a	1, 76	**6.3***	1, 76	0.1
	Nitrogen x Insecticide	n/a	n/a	4, 76	1.7	4, 76	0.1
	Cultivar x Nitrogen x Insecticide	n/a	n/a	4, 76	2.6	4, 76	2.9

* and ** indicates significance at 0.05, <0.0005 respectively. † indicates marginal significance, 0.05-0.1.
Random effect structure included the effects of row (cultivar), plot (phosphorus rate) nested within row, and subplot (insecticide use) nested within plot nested within row. Degrees of freedom calculated using the Satterthwaite approximation.

Table 3.3: Summary ANOVA testing fixed effects of phosphorus, onion cultivar, and insecticide use and all interactions on onion thrips, bacterial bulb rot, and onion yield.

		Response variable									
		Onion thrips		Bacterial bulb rot				Onion yield			
Year	Fixed effect			Within season		At harvest		Marketable yield		Adjusted yield	
		df	*f*	*df*	*f*	*df*	*f*	*df*	*f*	*df*	*f*
2017	Cultivar	1, 49	**28.2****	1, 49	**5.2***	1, 49	1.1	1, 49	0.4	1, 49	0.1
	Phosphorus	3, 49	3.4	3, 49	4.6	3, 49	3.1	3, 49	**13.1***	3, 49	**16.4****
	Insecticide	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a
	Cultivar x Phosphorus	3, 49	1.5	3, 49	7.2†	3, 49	1.8	3, 49	2.4	3, 49	1.8
	Cultivar x Insecticide	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a
	Phosphorus x Insecticide	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a
	Cultivar x Phosphorus x Insecticide	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a
2018	Cultivar	1, 59	**50.1***	1, 59	**8.1***	1, 59	**25.4****	1, 59	4.8	1, 59	0.5
	Phosphorus	3, 59	0.9	3, 59	1	3, 59	1.3	3, 59	5.7	3, 59	5.9
	Insecticide	1, 59	**256.6****	1, 59	0.1	1, 59	1.4	1, 59	242.6**	1, 59	**169.7****
	Cultivar x Phosphorus	3, 59	1.8	3, 59	3.2	3, 59	2.9	3, 59	2.0	3, 59	0.6
	Cultivar x Insecticide	1, 59	3.3†	1, 59	0.5	1, 59	0.5	1, 59	**4.1***	1, 59	2.8 †
	Phosphorus x insecticide	3, 59	1.4	3, 59	1.4	3, 59	0.1	3, 59	1.4	3, 59	0.8
	Cultivar x Phosphorus x Insecticide	3, 59	2.2	3, 59	0.1	3, 59	1.1	3, 59	3.4	3, 59	3.5

* and ** indicates significance at 0.05, <0.0005 respectively. † indicates marginal significance, 0.05-0.1.
Random effect structure included the effects of row (cultivar), plot (phosphorus rate) nested within row, and split plot (insecticide use) nested within plot nested within row. Degrees of freedom calculated using the Satterthwaite approximation.

Table 3.4: Summary ANOVA testing fixed effects of phosphorus, onion cultivar, and insecticide use and all interactions on onion thrips densities within season during three onion developmental stages, prebulbing, bulbing, and bulbing in 2017 and 2018.

Year	Fixed effect	Developmental stage: Prebulbing		bulbing		postbulbing	
		df	*f*	*df*	*f*	*df*	*f*
2017	Cultivar	1, 28	1.3	1, 28	**34.7****	1, 28	**11.5****
	Phosphorus	3, 28	7.6 †	3, 28	2.1	3, 28	1.9
	Insecticide	n/a	n/a	n/a	n/a	n/a	n/a
	Cultivar x Phosphorus	3, 28	1.8	3, 28	1.2	3, 28	0.9
	Cultivar x Insecticide	n/a	n/a	n/a	n/a	n/a	n/a
	Phosphorus x Insecticide	n/a	n/a	n/a	n/a	n/a	n/a
	Cultivar x Phosphorus x Insecticide	n/a	n/a	n/a	n/a	n/a	n/a
2018	Cultivar	1, 60	0.1	1, 60	15.8**	1, 60	**51.8****
	Phosphorus	3, 60	0.6	3, 60	0.7	3, 60	3.6
	Insecticide	n/a	n/a	1, 60	718.4**	1, 60	**2504.3****
	Cultivar x Phosphorus	3, 60	0.8	3, 60	1.8	3, 60	3.9
	Cultivar x Insecticide	n/a	n/a	1, 60	**14.8****	1, 60	0.1
	Phosphorus x Insecticide	n/a	n/a	3, 60	2.1	3, 60	0.9
	Cultivar x Phosphorus x Insecticide	n/a	n/a	3, 60	1.3	3, 60	5.6

* and ** indicates significance at 0.05, <0.0005 respectively. † indicates marginal significance, 0.05-0.1.
Random effect structure included the effects of row (cultivar), plot (phosphorus rate) nested within row, and subplot (insecticide use) nested within plot nested within row. Degrees of freedom calculated using the Satterthwaite approximation.

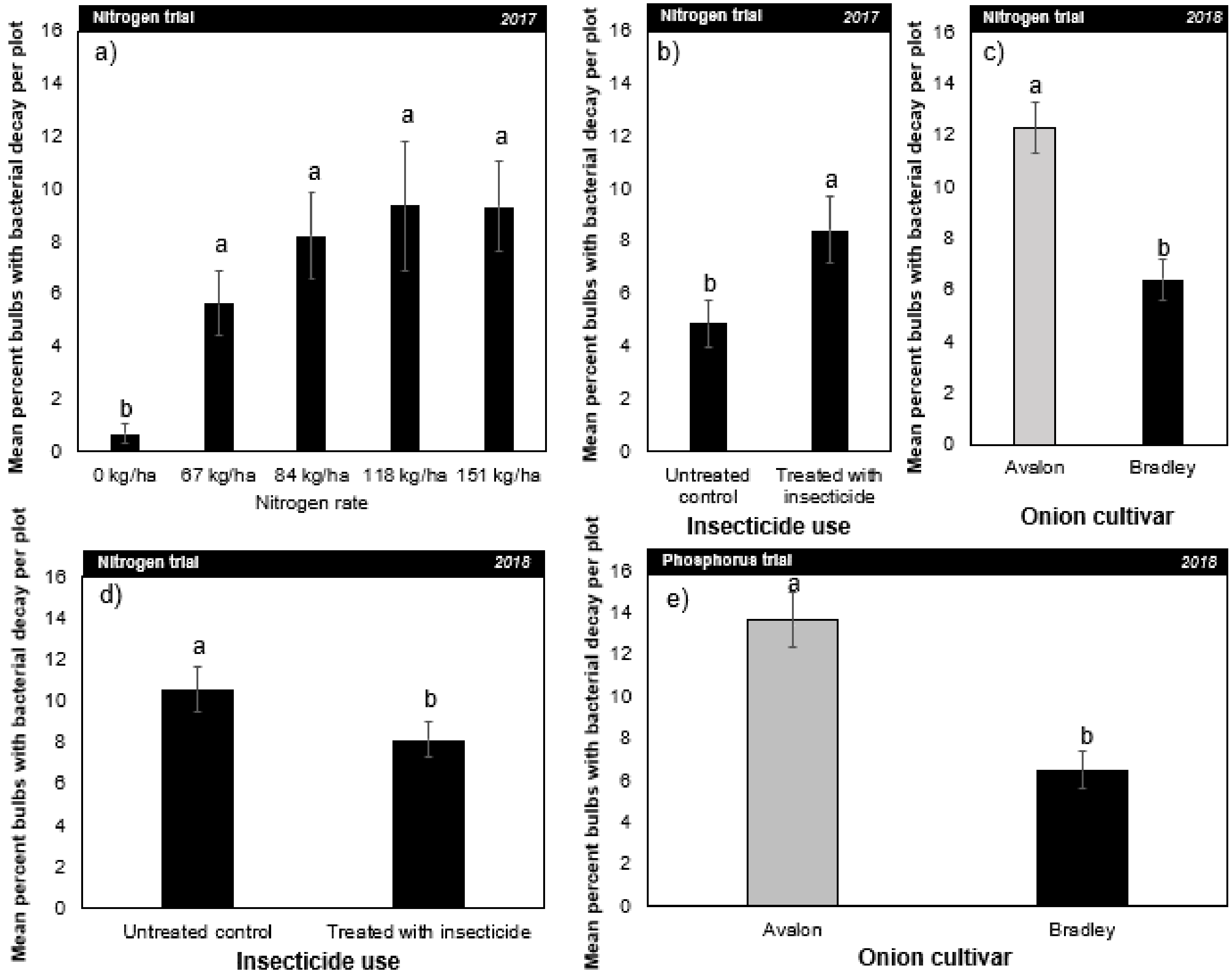

Figure 3.3: Significant effects of nitrogen rate (a), insecticide use (b, c), and onion cultivar (d, e) on the incidence of bacterial rot in marketable onions in 2017 (a, b) and 2018 (c, d, e). Two independent trials evaluated the effect of nitrogen with different onion cultivar and insecticide use combinations or the effect of phosphorous with different onion cultivar and insecticide use combinations. Five different rates of nitrogen were applied through the growing season; 0 kg ha-1, 67 kg ha-1, 84 kg ha-1, 118 kg ha-1 and 151 kg ha-1. Phosphorus rates were 0, 56, 112, and 168 kg ha-1. Fertilizer treatments were paired with two onion cultivars and two insecticide treatments. 'Avalon' is moderately thrips resistant and 'Bradley' is susceptible to thrips feeding. Every fertilizer treatments and onion cultivar combination were either treated with insecticide or left untreated. Significant effects determined by Type II ANOVA and means separated using LSMEANS at a 0.05 significance level.

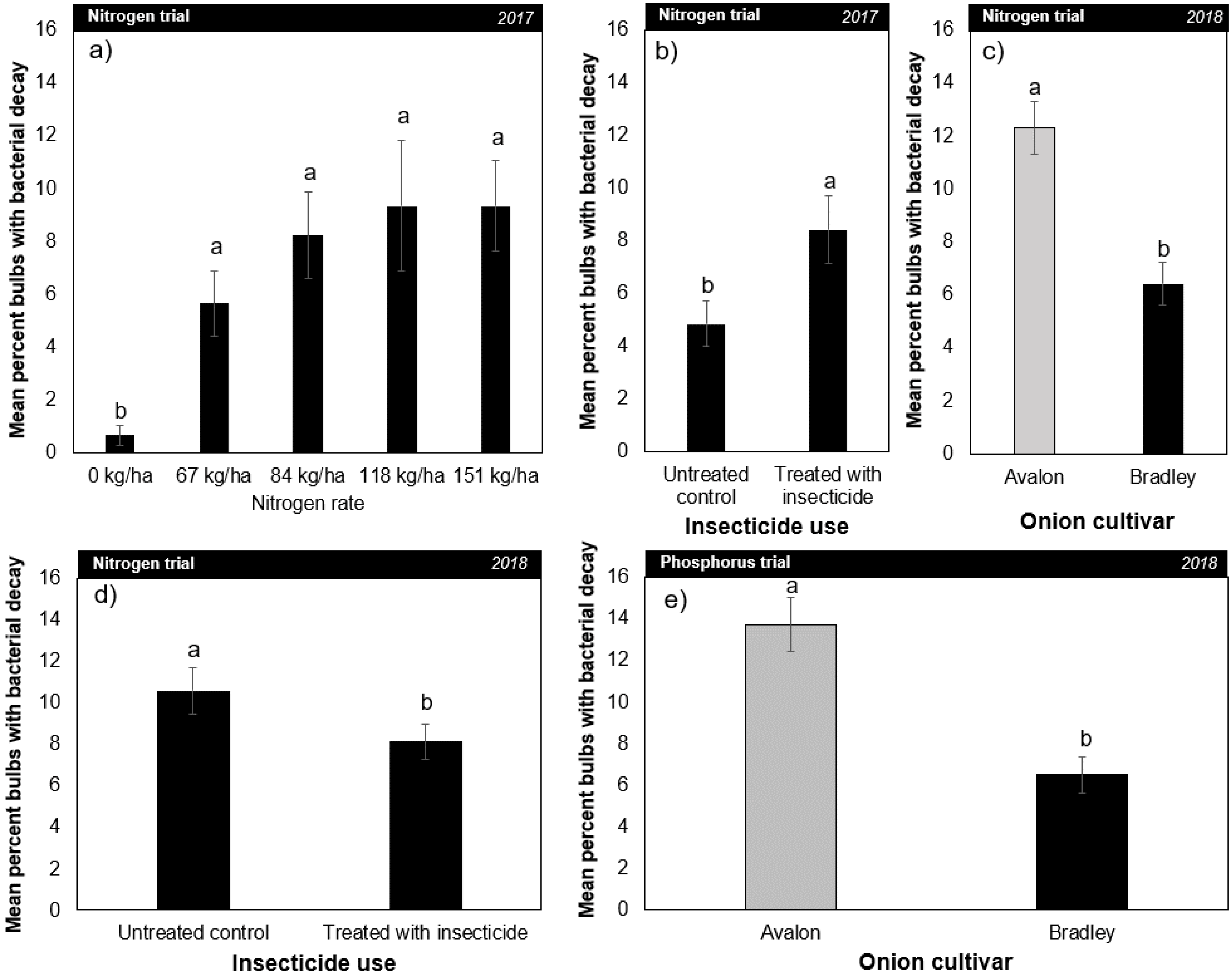

Figure 3.3: Significant effects of nitrogen rate (a), insecticide use (b, c), and onion cultivar (d, e) on the incidence of bacterial rot in marketable onions in 2017 (a, b) and 2018 (c, d, e). Two independent trials evaluated the effect of nitrogen with different onion cultivar and insecticide use combinations or the effect of phosphorous with different onion cultivar and insecticide use combinations. Five different rates of nitrogen were applied through the growing season; 0 kg ha-1, 67 kg ha-1, 84 kg ha-1, 118 kg ha-1 and 151 kg ha-1. Phosphorus rates were 0, 56, 112, and 168 kg ha-1. Fertilizer treatments were paired with two onion cultivars and two insecticide treatments. 'Avalon' is moderately thrips resistant and 'Bradley' is susceptible to thrips feeding. Every fertilizer treatments and onion cultivar combination were either treated with insecticide or left untreated. Significant effects determined by Type II ANOVA and means separated using LSMEANS at a 0.05 significance level.

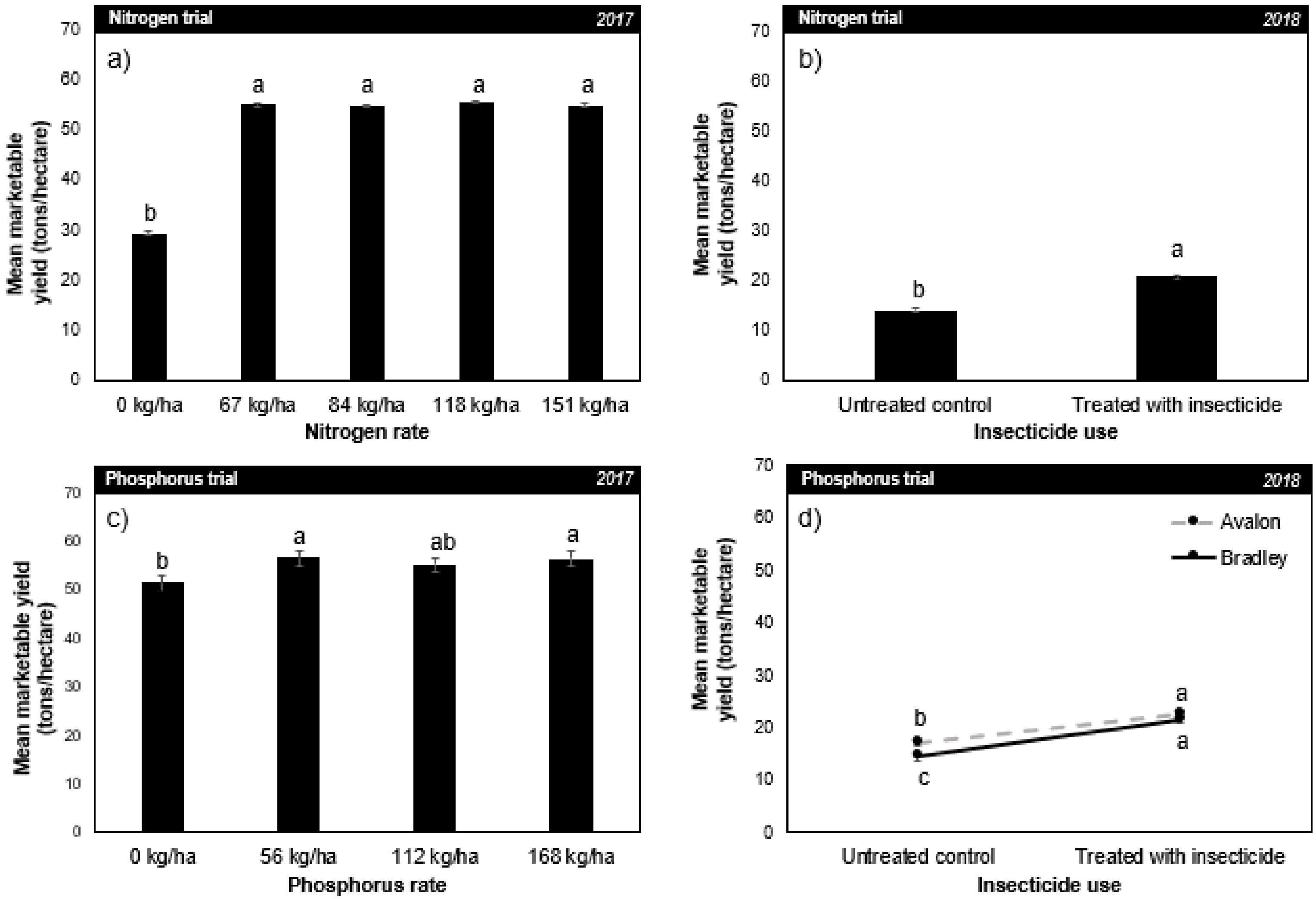

Figure 3.4: Significant effects of fertilizer rate (a, c), insecticide use (b), and the interaction of onion cultivar and insecticide use on marketable yield and adjusted marketable yield in 2017 (a, c) and 2018 (b, d). Two independent trials evaluated the effect of nitrogen with different onion cultivar and insecticide use combinations or the effect of phosphorous with different onion cultivar and insecticide use combinations. Five different rates of nitrogen were applied through the growing season; 0 kg ha-1, 67 kg ha-1, 84 kg ha-1, 118 kg ha-1 and 151 kg ha-1. Phosphorus rates were 0, 56, 112, and 168 kg ha-1. Fertilizer treatments were paired with two onion cultivars and two insecticide treatments. 'Avalon' is moderately thrips resistant and 'Bradley' is susceptible to thrips feeding. Every fertilizer treatments and onion cultivar combination were either treated with insecticide or left untreated. Significant effects determined by Type II ANOVA and means separated using LSMEANS at a 0.05 significance level.

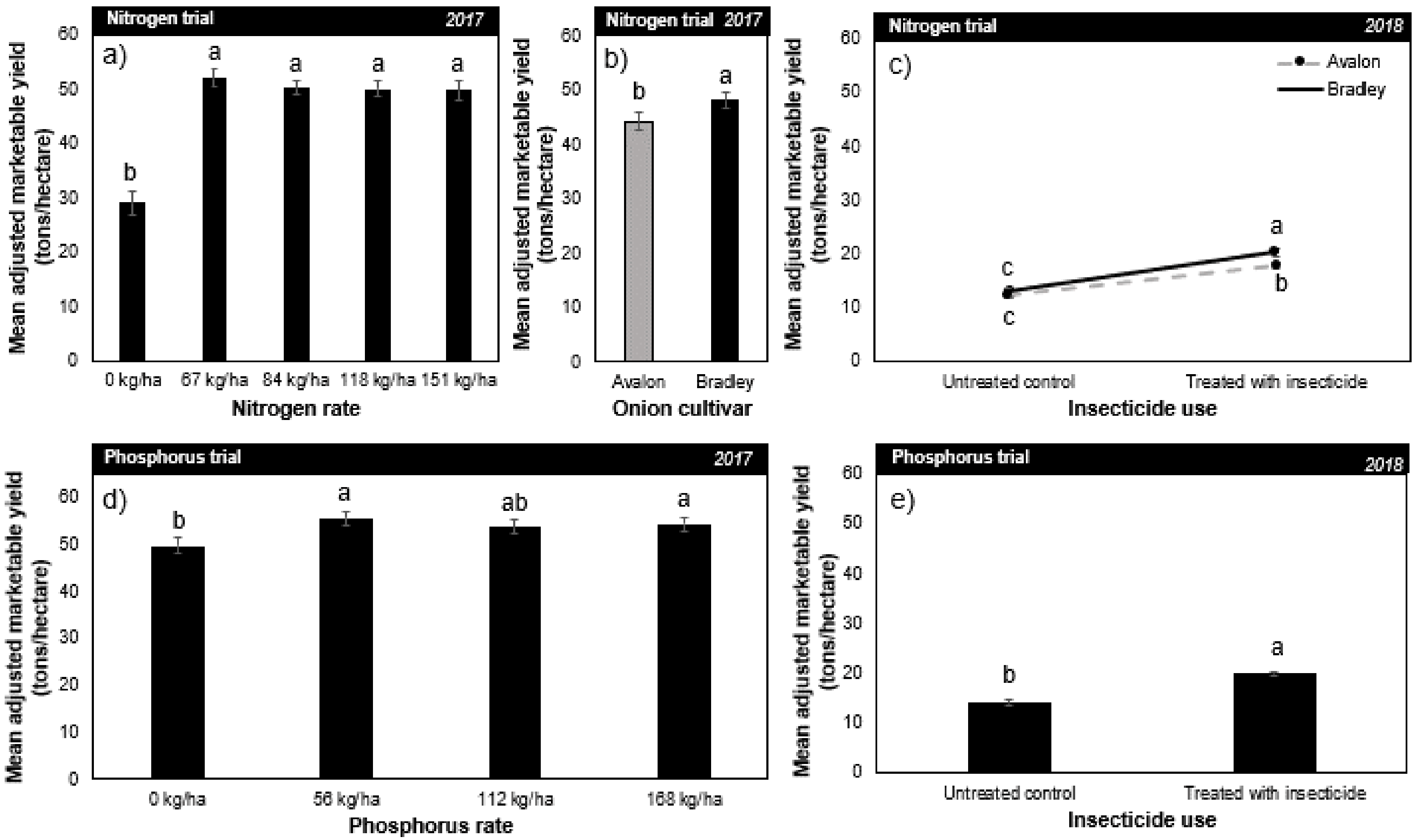

Figure 3.5: Significant effects of fertilizer rate (a, d), onion cultivar (b), interaction of onion cultivar and insecticide use (c), and insecticide use (e) on adjusted marketable yield in 2017 and 2018. Two independent trials evaluated the effect of nitrogen with different onion cultivar and insecticide use combinations or the effect of phosphorous with different onion cultivar and insecticide use combinations. Five different rates of nitrogen were applied through the growing season; 0 kg ha-1, 67 kg ha-1, 84 kg ha-1, 118 kg ha-1 and 151 kg ha-1. Phosphorus rates were 0, 56, 112, and 168 kg ha-1. Fertilizer treatments were paired with two onion cultivars and two insecticide treatments. 'Avalon' is moderately thrips resistant and 'Bradley' is susceptible to thrips feeding. Every fertilizer treatments and onion cultivar combination were either treated with insecticide or left untreated. Significant effects determined by Type II ANOVA and means separated using LSMEANS at a 0.05 significance level.

Table 3.5: Summary of the net effects of three IPM tactics implemented to manage onion thrips and bacterial bulb rot in onion in New York.

IPM tactic	Thrips densities	Bacterial bulb rot	Onion yield	Recommended practice for onion growers
Thrips-resistant cultivar	**Decrease**	**Increase**	**Decrease**	**No**
Reduced rate of fertilizer	**Decrease** [a]	**Decrease** [b]	**Decrease**[c]	**Yes**
Insecticide use	**Decrease**	**Increase/Decrease** [b]	**Increase**	**Yes**

Color indicates the number of trials in which IPM tactic was significant

0/4 **1/4** **2/4** **3/4** **4/4**

[a] Thrips-susceptible onion cultivar, ‘Bradley’, did not experience fewer thrips at lower fertility regimes
[b] Insecticide use increased the incidence of bacterial rot in 2017, but decreased increased in 2018
[c] Only observed decrease was in unfertilized treatments compared with fertilized ones.

4. Discussion

Onion production is challenged by multiple pests and onion thrips and bacterial rot are two of the most important constraints to producing marketable yields. In a multipartite IPM program, the use of an onion-thrips resistant cultivar, reduced fertility regimes and an action-threshold based insecticide application program were evaluated to reduce onion thrips densities and bacterial bulb rot. We hypothesized that a thrips-resistant cultivar combined with a reduced fertility regime and an action-threshold based insecticide program would have the greatest success in managing onion thrips and reducing bacterial bulb rot, thereby increasing marketable yields. The combination of the thrips-resistant cultivar ('Avalon') and action threshold-based insecticide program significantly reduced thrips densities, but the reduction in fertility (nitrogen and phosphorous) had little impact on reducing thrips densities. Despite reducing thrips densities in 'Avalon', it consistently had greater levels of bacterial rot and reduced marketable yields. Overall, reduced fertility regimes had a limited impact on reducing thrips densities and bacterial bulb rot; however, marketable bulb yields did not differ between the lowest and highest rates of fertilizer (excluding the unfertilized control) (Table 3.5). Insecticide use had the greatest impact on reducing thrips densities and increasing marketable yield but had little to no benefit in reducing bacterial rot (Table 3.5). Our study exemplifies the importance of selecting IPM tactics that optimize management for multiple pests and pathogens within a production system.

4.1 Onion thrips

Host plant resistance shows great promise as a preventative tactic for onion thrips management in onion. The onion-thrips resistant cultivar significantly reduced onion thrips densities during all onion developmental stages throughout the growing season. Consistently, ‘Avalon’ had fewer onion thrips than ‘Bradley’ regardless of any additional management tactic implemented (insecticide use or fertility regime). Many studies have evaluated onion cultivars for resistance to thrips feeding damage (Boateng et al., 2014; Damon et al., 2014; Diaz-Montano et al., 2012). Findings from these studies indicate that thrips prefer onion cultivars with blue-green leaves and high amounts of epicuticular wax, which is characteristic of ‘Bradley’. Conversely, ‘Avalon’ has yellow-green leaves and presumably a lower amount of epicuticular wax, which is likely the reason why we observed lower densities of thrips (Damon et al. 2014).

Epicuticular waxes are important for onions to resist foliar plant pathogens. Mohan and Molenaar (2005) reported that onion cultivars with lower amounts of epicuticular wax (glossy leaf phenotypes) were more vulnerable to powdery mildew caused by *Leveillula taurica* than waxier cultivars. In our study, ‘Avalon’ consistently had fewer thrips per leaf as compared with ‘Bradley’, but it also had greater levels of bacterial rot. Thus, the slight to moderate advantage that ‘Avalon’ had for reducing thrips damage was surpassed by its greater disadvantage of succumbing to moderate to high levels of bacterial rot. Therefore, ‘Avalon’ may have limited utility in onion production system in humid climates like those in the Great Lakes region of the US (Table 5).

Plant fertilization may not be an effective cultural control tactic to manage onion thrips in muck onion production. Studies conducted on mineral soil report a reduction in onion thrips densities with decreasing rates of nitrogen. Malik et al. (2009) found that onions treated with reduced rates of nitrogen (<150 kg/ha) had 70% fewer thrips than plants treated with high rates of nitrogen (>150 kg/ha). Similarly, Buckland et al. (2013) found that larval onion thrips densities were reduced by 25% in onions treated with 134 kg N ha $^{-1}$ compared to a standard rate of 407 kg N ha $^{-1}$. In our study, onion thrips densities were negatively influenced by a reduction in fertilizer levels in only one of four studies. Specifically, in the 2017 nitrogen trial, highest onion thrips densities were initially recorded in fertilized onions during the prebulbing stage, but as the season progressed, highest densities were recorded in unfertilized onions. Ultimately, these densities during the bulbing and postbulbing stages were similar to the overall seasonal effects, and the seasonal mean onion thrips densities were greatest in unfertilized treatments and approximately four times greater than the lowest nitrogen treatment (‘Avalon’ fertilized with 67 kg/ha).

Previous studies with muck soil types have indicated that onion thrips are unaffected by nitrogen fertilizer. For example, Westerveld et al. (2008) did not observe differences in onion thrips feeding damage in onion treated with nitrogen at rates: 0, 90, and 180 kg/ha. Similarly, Leach et al. (2017) found no significant differences in thrips densities in onion treated with 67, 101 and 140 kg of nitrogen/ha. Thus, results from our study are consistent with those previous reports and suggest that a reduction in thrips densities by reducing fertilizer is not a consistent or reliable management tactic for onion growers who produce onions on muck soil.

Phosphorus fertilizer did not significantly impact seasonal mean larval densities in 2017 or 2018. Chen et al. (2004) reported a 40% decrease in the number of western flower thrips (*Frankliniella occidentalis*) on Impatiens flowers (*Impatiens wallerana)* when fertigated with a 0.32 millimolar (mM) rate/pot of phosphorus compared with those fertilized with the 1.28 mM rate/pot. In our study, thrips were not significantly impacted by phosphorus during any of the onion developmental stages. Phosphorus amendments did not significantly impact plant growth, which may explain why we failed to find differences in onion thrips densities. To the authors' knowledge, this is the first study to evaluate the effect of phosphorus fertilizer on onion thrips in onion. However, further evaluation is needed to determine if phosphorus may be an effective cultural control of onion thrips in mineral onion production.

4.2 Bacterial bulb rot

Bacterial bulb rot is one of the most significant plant diseases in onion production, as infections may either kill plants during the season or render bulbs unmarketable. Previous studies have suggested that choice of onion cultivar, reduction in nitrogen fertilizer and increased insecticide use can reduce the incidence of bacterial rot in onion (Grode et al. 2019; Pfeufer and Gugino 2018; Stumpf et al. 2017; Dutta et al. 2014; Diaz-Perez et al. 2003; Wright et al. 1993). Some studies have suggested that onion thrips have a significant role in transferring *Pantoea spp.*, which can cause bacterial bulb rot in onion (Dutta et al. 2014; Grode et al. 2016; Grode et al. 2019); however, in our 2-yr study we observed that insecticide use reduced the incidence of bacterial rot in only one of four trials. Yet, in our trials, neither *Pantoea ananatis* nor *Pantoea agglomerans*, the bacterial pathogens responsible for bacterial symptoms in previous studies, were isolated from rotten bulbs in 2018, and only 5% of the bulbs in 2017. The low incidence of *Pantoea spp.* may explain why we failed to confirm a relationship between onion thrips densities and bacterial bulb rot. Nevertheless, it should be noted that only one study thus far has connected onion thrips and bacterial bulb rot (Dutta et al. 2014), and all other reports have only identified a relationship between bacterial leaf blight and onion thrips (which was not examined in our study) (e.g. Grode et al. 2019; Grode et al. 2016). Therefore, it is possible that thrips contribute to foliar bacterial diseases, such as those reported in Grode et al. (2016) and Grode et al. (2019), but do not necessarily increase the incidence of bacterial bulb rot.

In our study, the significance of the onion-thrips resistant cultivar ‘Avalon’ being highly susceptible to bacterial bulb rot was far more important and consistent than the slight benefits it had in reducing thrips densities. Across both years and trials, levels of bacterial bulb rot in ‘Avalon’ were considerably greater than those in ‘Bradley’. Previous studies have shown that red onion and Spanish onion cultivars tend to have a higher incidence of bacterial rot than other cultivar types and may be predisposed to these pathogens in certain climates (Stumpf et al. 2017; Wohleb and Waters 2016; Pfeufer et al. 2015 Schroeder et al. 2010). While unreported in this study, we consistently observed differences between ‘Avalon’ and ‘Bradley’ including variations in plant development and maturity and susceptibility to foliar plant pathogens. These differences may explain, in part, the predisposition of ‘Avalon’ to bacterial rot, as other studies have indicated the importance of onion development and curing in the incidence of bacterial rot (Wright et al. 2001). Nevertheless, further research should address the mechanisms behind cultivar susceptibility to bacterial rots.

The impact of nitrogen fertilizer on levels of bacterial bulb rot differed between years in our study. In 2017, levels of bacterial rot in fertilized onions were significantly greater than those in unfertilized ones. These results are consistent with previous reports (Diaz-Perez et al. 2003; Batal et al. 1994; Wright et al. 1993). In contrast, nitrogen fertilizer did not impact bacterial bulb rot levels in our study in 2018, which may be due to the differing weather conditions between the years. In our study, split applications of nitrogen did not significantly impact incidence of bacterial rot at harvest; however, within the season, lowest levels of infected plants were observed in treatments with low rates of nitrogen at the second application. While there were numerical increases in the incidence of bacterial rot in higher rates of nitrogen, we did not record a significant difference between plants fertilized with 67 kg/ha, 84 kg/ha, 118 kg/ha, or 151 kg/ha. Therefore, it may benefit growers to reduce nitrogen application rates to 67 kg/ha, as greater amounts of nitrogen fertilizer may significantly increase bacterial bulb rot in certain years. Phosphorus fertilizer did not significantly impact bacterial bulb rot; however, we did not observe any differences in plant and minimal differences in onion yield. Thus, if bacterial rot is significantly impacted by plant growth, further evaluation should address phosphorus fertilizer amendments when plants are responsive to the phosphorus fertilization.

4.3 Onion yield

Low rates of nitrogen and phosphorus fertilizer (67 kg/ha N 56 kg/ha P) produced statistically similar yields to plants fertilized with highest rates of fertilizer in both years. In fact, we found that adjusted marketable yields decreased by 8-10% in high rates of nitrogen (84 kg/ha, 118 kg/ha, or 151 kg/ha) due to increased incidence of bacterial bulb rots in 2017. Muck soils are unique as they are rich in organic matter, and naturally high in nitrogen (Lucas 1982). Multiple studies have documented that less fertilizer is typically needed in muck agriculture (Haynes et al. 2012; Gonzalez et al. 2016), and current recommended rates of nitrogen can be as low as 67 kg/ha (Reiners et al. 2017; Warncke et al. 2004). However, in New York, growers regularly fertilize with approximately 118 kg/ha N annually (Nault and Hoepting 2014, unpublished). Our study suggests that a large majority of fertilizer remains in the soil, as we consistently observed higher rates of soil nitrate with higher rates of nitrogen fertilizer, which is similar to other studies (Boyhan et al. 2007). Therefore, growers should critically evaluate their soil fertility programs to maximize yields, but also reduce fertilizer loss from leaching or runoff.

5. Conclusions

Pest management in agricultural production systems, like onion, is inherently complex as these systems are challenged by multiple pests and pathogens. Kogan (1998) argued that the progress of IPM relies on the integration of multiple pest management tactics at increasing agricultural scales. Recently, the relevance of IPM has been questioned (Peterson et al. 2018), with many urging researchers to create programs that will manage multiple pest interactions within an agroecosystem. Our study illustrates the importance of curating an integrated pest management program to address multiple pests in a production system (i.e. onion thrips and bacterial rot). In our case, we found that an integrated pest management tactic (thrips-resistant onion cultivar ‘Avalon’) was effective in reducing densities of an important onion insect pest, but highly susceptible to bacterial rot pathogens. Additionally, an integrated pest management tactic (reducing fertilizer levels) that reduced insect densities in other onion production systems did not consistently reduce densities in our system. However, we found decreasing rates of fertilizer did not compromise levels of marketable yield, and in one year it decreased the incidence of bacterial rot. Future research should continue to develop pest management programs that holistically evaluate their impact on major pests and pathogens within production systems, such that growers can observe maximum benefits from the programs and increase sustainability.

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CHAPTER 3

IMPORTANCE OF TRANSPLANTED ONIONS CONTRIBUTING TO LATE-SEASON IRIS YELLOW SPOT VIRUS EPIDEMICS IN NEW YORK

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Abstract

Iris yellow spot virus (IYSV) is an economically significant tospovirus of onion transmitted by onion thrips (*Thrips tabaci* Lindeman). IYSV epidemics in onion fields are common in New York; however, the role of various habitats contributing to viruliferous onion thrips populations and IYSV epidemics is not known. In a two-year field study in New York, the abundance of dispersing onion thrips, including those determined to be viruliferous via reverse-transcriptase polymerase chain reaction (RT-PCR), was recorded in habitats known to harbor both IYSV and its vector. Results showed viruliferous thrips were encountered in all habitats; however, transplanted onion sites accounted for 49-51% of the total estimated numbers of viruliferous thrips. During early to mid-season, transplanted onion sites had 9 to 11 times more viruliferous thrips than the other habitats. These results indicate that transplanted onion fields are the most important habitat for generating IYSV epidemics in all onion fields (transplanted and direct-seeded) in New York. Our findings suggest that onion growers should control onion thrips in transplanted fields early in the season to minimize risk of IYSV epidemics later in the season.

KEY WORDS: onion, onion thrips, *Thrips tabaci*, IYSV, tospovirus

1. Introduction

Iris yellow spot virus (IYSV) (genus *Tospovirus*, family *Bunyaviridae*) is transmitted by onion thrips (*Thrips tabaci* Lindeman) and can cause extensive economic damage to onion. IYSV was originally described by Cortes et al. (1998) on Dutch iris (*Iris hollandica* Tub.) in the Netherlands. Since its first identification, IYSV has been isolated from 61 plant species in 27 countries (Bag et al. 2015; Gent et al. 2006). IYSV has a great economic impact on the commercial onion bulb and seed industry in which yield losses can range between 60-100% annually (Pozzer et al. 1999; Gent et al. 2006). Once infected with IYSV, diamond-shaped lesions appear on onion scapes and tan or straw-colored necrotic lesions form on leaves. In severe infections, these lesions coalesce, girdling the leaf or stem and causing dieback (De Avila et al. 1981). In an economic analysis conducted in 2003, onion growers in Colorado reported annual losses of approximately $2.5-5 million due to IYSV infection (Gent et al. 2006).

Previous studies have indicated that IYSV is not seed transmitted and mechanical inoculation is largely unsuccessful in onion (Bag et al. 2015; Kritzman et al. 2001). Thus, spread of IYSV is dependent on the acquisition and transmission of IYSV by onion thrips. Similar to other tospoviruses, IYSV is both circulative and propagative within its thrips vector, allowing adults to transmit the virus until death (Whitfield et al. 2005). Tospoviruses are acquired only by first and second instars (Whitfield et al. 2005): acquisition rates decrease as larvae mature (Ullman et al. 2002) because a mid-gut barrier develops, which prevents viral infection (Nagata et al. 1999). Unlike larvae, adults can disperse great distances and may infect multiple plants. Consequently, understanding dispersal of thrips adults provides insight into the epidemiology of tospoviruses (Ullman et al. 2002).

Onion thrips is the only species known to transmit IYSV to onion and transmission efficiencies have been recorded as high as 76% or greater (Srinivasan et al., 2012; Birithia et al. 2013). A positive relationship between IYSV incidence in onion fields and onion thrips densities has been documented by Kritzman et al. (2001) and Schwartz et al. (2009). Onion thrips has a strong preference for onion, despite their utilization of over 300 plant species as hosts (Doederlein and Sites 1993). Additionally, their populations can increase quickly, with seven or more generations produced in a year (Hoffman et al. 1996). These traits of host specificity and rapid population growth are critical factors influencing IYSV epidemics in onion fields (Gent et al. 2006). In addition to transmitting IYSV, onion thrips feeding also causes significant bulb yield reductions, ranging from 43-60% (Fournier et al. 1995; Rueda et al. 2006).

Currently, there are no IYSV-resistant onion cultivars (Cramer et al. 2014; Diaz-Montano et al. 2012). Virus management efforts are then focused on reducing onion thrips populations during the growing season to reduce IYSV epidemics (Bag et al. 2015; Gent et al. 2006). Within the Unites States, additional efforts have been made to identify sources of IYSV inoculum in onion production systems to better understand its epidemiology and develop management strategies. Thus far, three different sources of inoculum within onion production systems have been identified: onion plants imported from the southwestern US and then transplanted elsewhere, certain weed species, and volunteer onions in cull piles (Gent et al. 2006; Evans et al. 2009; Hsu et al. 2010; Hsu et al. 2011; Nischwitz et al. 2012; Sampangi et al. 2007; Schwartz et al. 2014; Smith et al. 2011; Szostek and Schwartz 2015). The relative contribution of habitats containing these various sources of IYSV and its vector on IYSV epidemics in onion agroecosystems is not known.

Young onion plants imported from the southwestern US and then transplanted and grown in commercial fields could be an important habitat affecting IYSV epidemics in all onion fields later in the season. In New York, some onion growing areas are not established with transplants, whereas others may have as much as 35% of the area established with transplants. Gent et al. (2006) found 50% of onion transplant lots tested positive for IYSV and were also infested with onion thrips. Hsu et al. (2011) assayed over 1,000 onion plants imported from the southwestern US and found 0.5% infected with IYSV. Infection levels as low as 0.5% could create severe IYSV epidemics later in the season. Additionally, fields of transplanted onions are preferentially colonized over direct-seeded onions early in the season and can host large populations of onion thrips (Hsu et al. 2011). In New York, onion fields established with imported transplants from the southwestern US, and were isolated from major onion producing areas, had severe epidemics of IYSV (over 75% of plants with symptoms) (B. Nault, *personal observation*).

Habitats containing weeds near onion fields could be important contributors to IYSV epidemics in onion fields. At least 61 weed species have tested positive for IYSV, and approximately 30% are commonly encountered in onion production systems (Gent et al. 2006; Schwartz et al. 2014; Smith et al. 2011). Smith et al. (2011) identified four weed species (i.e., dandelion [*Taraxacum officinale,* G.H. Weber ex Wiggers], common burdock [*Arctium minus,* Bernh.], curly dock [*Rumex crispus,* L.], and chicory [*Cichorium* *intybus,* L.]) that were suitable hosts for both IYSV and onion thrips; therefore, these perennial or biennial weed species may provide a 'green bridge' for IYSV between onion growing seasons in New York. Similar results with other plant species have been presented by Nischwitz et al. (2012) and Schwartz et al. (2014).

Habitats where onion bulb cull piles are located may be important to IYSV epidemiology in onion fields. While cull piles are dominated by decomposing onion bulbs, bulbs that produce leaves also occur (i.e., volunteer onion). Volunteer onion plants, which grow from bulbs leftover from the previous year's onion crop, may enable IYSV to persist between growing seasons. Indeed, volunteer onion plants from cull piles have tested positive for IYSV in multiple studies (Gent et al. 2006; Hsu et al. 2011; Schwartz et al. 2014). In New York, 50% of onion cull piles examined had volunteer onion plants that tested positive for IYSV (Hsu et al. 2011). Furthermore, the probability of detecting viruliferous onion thrips in onion cull piles is largely dependent on the presence of volunteer onions, as Szostek and Schwartz (2015) failed to detect viruliferous onion thrips in cull piles composed of only decaying onion bulbs.

The purpose of this study was to gain insight into which habitat(s) may be most influential in fostering IYSV epidemics in New York onion fields. To examine this question, we considered the abundance of viruliferous onion thrips captured in a habitat early to mid-season as a proxy for identifying the relative contribution of that habitat to IYSV epidemics later in the season. Habitats sampled included those known to contain IYSV and its vector (i.e., transplanted onion fields, weedy areas near onion fields and onion cull piles) as well as direct-seeded onion fields, which served as an early-season control because IYSV is not seed transmitted. We hypothesized that onion fields established with transplants imported from the southwestern US would generate the greatest numbers of viruliferous thrips early to mid-season compared to the other habitats. Such a scenario would create an opportunity for secondary spread of IYSV into adjacent onion fields (especially direct-seeded) and weedy habitats because onion thrips adults are known to disperse from maturing transplanted onion fields in search of other suitable habitats later in the season (Smith et al. 2017).

2. Materials and Methods

The study was conducted on the ‘Elba muck’ near Elba, NY in 2014 and 2015. The Elba muck is in northwestern New York and spans two counties, Orleans and Genesee. The Elba muck is nearly 2,200 hectares and approximately 50% of the area is planted annually to onion and about 35% is transplanted with onions imported from the southwestern US. Onions are direct seeded from early April through mid-May or transplanted from early April through early June. Onions are harvested from July to September. Most onion fields in the Elba muck are not rotated from year to year because such land is a premium for onion production. The Elba muck was chosen as the study area because it is one of the largest onion production areas in the eastern US and IYSV is frequently encountered, sometimes at very high levels (Hsu et al. 2010; Smith et al. 2015).

Sampling sites. Populations of adult onion thrips were monitored at a total of sixteen sites representing the four habitat types: 4 weedy areas, 4 culled onion piles, 4 onion fields established with imported transplants and 4 fields that were direct-seeded (Fig. 4.1). ‘Weedy areas’ were located at least 10 meters from an onion field and at least 60% of the area was dominated by weeds (Fig. 4.2a). ‘Weedy area’ sites were also preferentially selected based on presence of weed species known to be suitable hosts for both onion thrips and IYSV, and in areas where IYSV had been identified previously in perennial and biennial weed hosts (Smith et al. 2011). Areas designated as 'onion cull piles' were located within approximately 2 km of onion fields and were dominated by culled onion bulbs and volunteer onions annually (Fig. 4.2b). Some of these onion cull piles previously had volunteer onions that tested positive for IYSV (Hsu et al. 2011). 'Transplanted' onion sites were in fields transplanted with onions that originated from a farm in the southwestern US (Fig. 4.2c). These imported plants had stems approximately 1.5 to 2 cm in diameter and 2-3 leaves at the time of transplanting in May. Each year, a subsample of imported onion plants was taken prior to transplanting from each of these field sites and then tested for IYSV using double antibody sandwich enzyme-linked immunosorbent assay (DAS-ELISA) with commercially available antibodies and following the manufacturer's protocol (Agdia, Inc., Elkhart, IN). 'Direct-seeded' onion fields were included as an early-season "negative" control because IYSV is not seed transmitted and consequently does not serve as an initial source of IYSV inoculum for thrips. Direct-seeded onion fields were seeded into fields in late April. Both transplanted and direct-seeded onion fields were devoid of volunteer onions, which were either absent or removed before the experiment was initiated. Transplanted onion sites were preferentially selected to feature fields with comparable maturation times as direct-seeded onion sites, based on cultivar and planting date, such that all sites were monitored for a similar period.

Sampling methods. Yellow sticky cards (Scentry MultiGuard; Great Lakes IPM, Vestaburg, MI) were used to monitor onion thrips flight activity in the various habitats. At each site, four yellow sticky cards (7.6 cm x 12.7 cm) were placed 25 m apart along a transect spanning 100 m. The sticky cards were mounted on wooden stakes and suspended approximately 60-92 cm from the ground (Fig. 4.2d). Yellow sticky cards were replaced weekly and stored at -20ºC until onion thrips could be morphologically identified and recorded (Moritz et al. 2011). Sampling of adult onion thrips was initiated in all habitats when transplanted onions had approximately 4-5 leaves and concluded when onions were harvested. In 2014, sampling began on 10 June and concluded on 4 September, while in 2015 sampling began on 1 June and ended 3 September. Numbers of adult onion thrips were binned into three sampling periods that approximately represented initial colonization of onion fields by onion thrips (early-season: 10 to 30 June 2014, 1 June to 1 July 2015), dispersal of the first generation of onion thrips formed within the onion crop (mid-season: 2 to 28 July 2014, 2 to 30 July 2015), and dispersal of subsequent generations of onion thrips (late-season: 1 August to 4 Sept 2014, 1 August to 3 September 2015). For each sampling period, the mean total number of adults/card/site was determined. Voucher specimens are maintained at the Department of Entomology at the New York State Agricultural Experiment Station in Geneva, NY.

Plant species composition in weedy areas. Weed species and their prevalence were assessed at each weedy area. At each site, 10 quadrats of 1 m^2 each were randomly placed immediately adjacent to the area where the yellow sticky cards were located (10 m by 100 m). All weed species were identified and botanically classified (family and species), including those known to be hosts for IYSV and onion thrips. The dominance of each species within each quadrat was visually assessed on a scale from 0-100%, based on the area covered by that species; thus, 1% indicated a weed species only covered 1% of the area and 100% indicated that the weed species covered 100% of the area within the quadrat. Weedy areas were assessed 17 Jul 2016 and 23 Aug 2016. All weedy areas were permanent and were dominated by the same weed species in each year of the study.

IYSV detection in onion transplants. A subsample of imported onion plants was obtained from New York onion growers' warehouses prior to transplanting at each field site. Onion plants were imported from the southwestern US, where IYSV is known to occur (Gent et al. 2006; Miller et al. 2006). Imported onion plants were grown in a greenhouse for three months in attempt to increase IYSV titer levels. All onion plants were planted into pots (7.6 cm diameter x 31 cm tall) containing Cornell potting mix. Plants were treated with spinetoram at 1.9 g/L (AI) (Radiant™, Dow AgroSciences, Inc., Indianapolis, IN) and spirotetramat at 1.9 g/L (AI) (Movento™, Bayer CropScience, Research Triangle Park, NC) to ensure that plants were thrips-free and then grown in thrips-proof cages in the greenhouse. After three months, plants were tested for IYSV using DAS-ELISA. All samples were composites of leaf tissue from four onion plants weighing 1 gram. Leaf tissue was cut from inner leaves of the onion plant to increase the likelihood of detecting IYSV (Kritzman et al. 2001). ELISA outputs were analyzed with a BioTek ELx 800 platereader (BioTek, Winooski, VT). Samples were duplicated and the mean optical density reading for each sample was used to determine the sample absorbance. Mean absorbance values two times the negative control was deemed positive for IYSV. A positive composite sample was conservatively estimated to represent a single infected plant because there was a low frequency of samples testing positive for IYSV. Samples were tested 12 August 2014 and 30 July 2015.

IYSV detection in adult onion thrips. Thrips were tested for IYSV using reverse-transcriptase polymerase chain reaction (RT-PCR) assays, as done in previous studies (Smith et al. 2015). Onion thrips adults were randomly selected and extracted from yellow sticky cards using a fine tipped paint brush and 1 ml of solvent (De-SolvIt, Orange-Sol Household Products, Inc., Gilbert, AZ). Paintbrushes were washed with ethanol between extractions to limit any potential contamination between onion thrips. Once removed, individual thrips were placed in a 0.5 ml centrifuge tube (USA Scientific, Ocala FL) and kept in a freezer (-80^{o} C) until processing. A subsample of six adult onion thrips was collected from each site (across all cards from that site) during each of the three sampling periods. Thus, at each of the 16 sites, a total of 18 onion thrips adults were tested for IYSV each year.

Total RNA was isolated from individual thrips using modified procedures from the Omega MicroElute RNA Kit (Omega Bio-Tek, Norcross, GA). Individual thrips were processed by adding a working solution of TRK lysis buffer and β-mercaptoethanol (β -me; 200 ml TRK buffer and 4.0 ml β-me per sample) paired with RNase-free, acid-washed, glass beads (Sigma-Aldrich, Glass Beads, Acid-Washed 425- 600 mm, cat # G8772-10G). Thrips were homogenized at 30 Hz for 2 min using a Qiagen TissueLyser (Qiagen, Valencia, California).

The diagnostic primers used to detect IYSV were IYSV-N402F 5'-ACTCACCAATGTCTTCAAC-3' and IYSV-N402R 5'-GGCTT CCTCTGGTAAGTGC-3', which were designed from the N gene of several IYSV isolates collected in New York in

2007–2008. To confirm the identity of onion thrips and quality of total RNA extracts, primers ThMCOI-F 5'-CGGGAACGGGATGAACAG-3' and ThMCOI-R 5'-GGTCCCCTCCCC CTCTA-3' designed in the mitochondrial cytochrome oxidase subunit I gene sequence (GenBank accession no. DQ228494) were used in a multiplex RT-PCR. Extracted total RNA was tested using the Qiagen one-step RT-PCR kit in a final volume of 12.5 µl containing total thrips RNA (1 µl), IYSV primers (1.25 µl, 1 mM each), onion thrips MCOI primers (0.625 µl, 0.1 mM each), dNTPs (0.5 µl, 10 mM stock), RNasin (0.1 µl), 5X buffer (2.5 µl), enzyme mix (0.5 µl), and sterile RNAse free water (4.15 µl). The thermal cycling conditions were as follows: 50°C for 30 min (1 cycle), 95°C for 15 min followed by 40 cycles of 94°C for 30s, 50°C for 1 min, 72°C for 1 min, and a final extension at 72°C for 10 min (BioRad ThermalCycler). RT-PCR products (402 bp for IYSV and 325 bp for onion thrips) were stained with GelRed (Biotium, Hayward, CA) following electrophoresis on 1.5% agarose gels, and then imaged using ultra-violet illumination.

Ten thrips that tested positive for IYSV in RT-PCR were randomly selected in each year to characterize their N gene amplicons by sequencing after processing with ExoSAP-IT. Sequences (a total of 20 N gene nucleotide sequences) were analyzed and compared using the DNASTAR Lasergene software (version 14.1) (DNASTAR, Madison, Wisconsin). This work confirmed the viruliferous nature of selected onion thrips.

Estimated number of viruliferous adult onion thrips. Detection of the IYSV N gene in an individual thrips suggested that it was viruliferous. Past research has indicated a positive association between thrips testing positive for the non-structural protein (NSs) gene, which indicates virus replication within the vector, and the coat protein (N) gene

(Birithia et al. 2013). To assess the relative importance of each habitat contributing to IYSV epidemics, the number of viruliferous onion thrips was estimated at each habitat type and sampling period each year. To estimate the number of viruliferous adults ($V_{s|p}$) for each site and sampling period, the total number of onion thrips adults per card ($\sum t$) within a site during a particular sampling period (*s*|*p*) was multiplied by the incidence of viruliferous adult thrips ($\%I$) within each site and sampling period (*s*|*p*).

$$V_{s|p} = \sum t_{s|p} * \%I_{s|p}$$

Viruliferous adults were estimated for each site over three sampling periods in 2014 and 2015 for a total of 96 data points (16 sites x 3 sampling periods x 2 years= 96). Season total estimated viruliferous adult thrips per card was also determined for every site by summing the number of viruliferous thrips adults across all three sampling periods.

Statistical analysis. Data for each year were analyzed independently because weather and growing conditions were substantially different. Data were analyzed using a generalized linear mixed model (SAS PROC GLIMMIX, 2016; SAS Institute, Cary, NC). Habitat type was treated as a fixed effect and site replicate as a random effect.

All insect count data were analyzed using a negative binomial distribution. Total viruliferous thrips per card (thrips testing positive for IYSV by RT-PCR) were analyzed using a binomial distribution (*n* viruliferous thrips /total onion thrips captured). Differences in habitat types within each analysis were compared using least squared means (P<0.05).

3. Results

Abundance of adult onion thrips across habitats. More adult onion thrips were captured in 2014 (total = 696 thrips per site) than in 2015 (total = 468 thrips per site). Total mean numbers of adults differed significantly among the habitat types in 2014 (P=0.0008, $F_{3, 44}$=6.66) and 2015 (P=0.0142, $F_{2, 43}$=3.95) (Table 4.1). Greatest numbers of thrips were recorded in transplanted onion fields in both years (Table 4.1). However, numbers of thrips adults in transplanted fields were only significantly greater than those in culled onion sites. Numbers of adults in weedy areas were second highest followed by those in direct-seeded onion fields. Sites with culled onions had the fewest number of adult onion thrips (Table 4.1).

Consistently, across all habitats and years, fewer adults were captured early in the season compared with mid to late season (Fig. 4.3). In 2014 and 2015 early in the season, the total mean number of adults captured among habitats did not differ ($P>0.05$) (Fig. 4.3). In 2014 and 2015 in the middle of the season, the total mean numbers of adults captured in transplanted onion fields were greater than those in the other habitats, although the difference was only significant in 2014 (P=0.0017, $F_{3, 12}$=4.15) (Fig. 4.3). Abundance of adults in the middle of the season did not differ between other habitat types. In 2014 and 2015 late in the season, total mean numbers of adults in transplanted onions, weedy areas and direct-seeded onions were greater than those in culled onions, but this difference was significant only in 2014 (P=0.0032, $F_{3, 12}$=8.12) (Fig. 4.3). Abundance of thrips in culled onion sites remained low for the entirety of the growing season, and never surpassed 30 thrips per card per sampling period.

Plant species composition in weedy areas. Between 11 and 36 plant species were recorded at each weedy area site (data not shown). The most common species were pigweed [*Amaranthus spp.*], followed by goldenrod [*Solidago spp.*] and wild raspberry [*Rubus spp*.]. *Amaranthus spp.* was the most dominant weed species and covered approximately 21% ±11% (mean ± standard error) of the area sampled over the two dates. Of those four species capable of hosting onion thrips and IYSV in New York (Smith et al. 2011), only dandelion [*Taraxacum officinale*], common burdock [*Arctium minus*], and curly dock [*Rumex crispus*] were identified. The most common IYSV weed host encountered was dandelion, which occurred at 75% of the sites. Overall, known plant host species for both IYSV and onion thrips were not numerous, and only covered 6% of the total area sampled.

IYSV detection in imported onion plants. Most of the imported onion plants tested negative for IYSV in 2014 and 2015 (Table 4.2). In 2014, no onions (0/829) tested positive for IYSV, while 0.9% (7/798) tested positive in 2015 when three out of six cultivars tested positive for IYSV: 1.5% (3/194) for 'Brandt', 1.3% (3/233) for 'Red Defender' and 0.6% (1/155) for 'Festival'.

Onion thrips testing positive for IYSV. All 20 IYSV N gene sequences from viruliferous thrips that were determined in this study shared 99% nucleotide identity with IYSV sequences available in GenBank, including previous entries from New York (GenBank JQ973065.1), Washington State (GenBank JQ973066.1), Idaho (GenBank KF263487.1), Georgia (GenBank DQ838593.1), and Colorado (GenBank KF263484.1).

Overall, a total of 576 individual thrips was tested for IYSV by RT-PCR (18 thrips per site x 4 habitat types x 4 replications per habitat type x 2 years = 576 thrips). Incidence of adult onion thrips testing positive for IYSV was much lower in 2014 than in 2015. The overall mean incidence of viruliferous onion thrips across all habitats was 6% and 18% in 2014 and 2015, respectively. In 2014, there were no significant differences in overall incidence of thrips testing positive for IYSV among the habitats. In 2015, overall incidence of viruliferous thrips captured in transplanted onion fields (31% infected) was significantly greater than incidence of those from weedy areas and cull piles (P=0.0107, $F_{3, 39}$=4.26), but not direct-seeded onion sites (21% infected) (Table 4.1). The seasonal mean incidences of IYSV in thrips from transplanted onion and direct-seeded onion were 1.5 to 3 times greater than those captured in cull piles or weedy areas.

The percentage of viruliferous adults captured in this study tended to be lower early in the season than mid to late season in both years, but the trend was more obvious in 2015 (Fig. 4.4). In 2014, the percentage of viruliferous thrips captured among habitats did not differ during any of the sampling periods (P>0.05) (Fig. 4.4). Early in the 2015 season, the percentage of viruliferous thrips captured among habitats did not differ (P>0.05). In the middle of the 2015 season, the percentage of viruliferous thrips in transplanted onion fields was numerically higher than those in the other habitats, but the difference only approached significance (P=0.08) (Fig. 4.4). Percentages of viruliferous thrips in the other habitats did not differ. Late in the season in 2015, the percentage of viruliferous thrips in transplanted onion fields and direct-seeded onion fields were significantly greater than those in the other habitats (P=0.0370, $F_{3, 11}$=4.03).

Estimated number of viruliferous adult onion thrips. Although there were more thrips captured in 2014 than 2015, estimated numbers of viruliferous adults were higher in 2015 (39 per card) than in 2014 (15 per card). Despite 10- to 20-fold differences in total estimated numbers of viruliferous thrips among some habitat types in 2014, none were significant (*P*>0.05) (Table 4.1). In 2014, there were overall numerically more viruliferous thrips estimated from transplanted onion fields than in the other habitats. In 2015, estimated numbers of viruliferous thrips in transplanted onion fields were significantly greater than those in culled onions and similar to those in weedy areas and direct-seeded onion fields (P=0.0094, $F_{3, 42}$=4.34) (Table 4.1). Transplanted onion fields had the greatest total number of estimated viruliferous thrips and accounted for 49-51% of total estimated numbers of viruliferous thrips in 2014 and 2015 compared to the other habitat types (Table 4.1). In both years, the lowest seasonal total estimates of viruliferous thrips were from cull piles, which only accounted for 4 to 5% of the total.

The fewest estimated numbers of viruliferous individuals occurred early in the season each year and there were no differences among habitat types (*P*>0.05) (Fig. 4.5). In 2014 and 2015 in the middle of the season, the estimated numbers of viruliferous adults in transplanted onion fields were greater than those in the other habitats, but this difference was only significant in 2015 (P=0.0296, $F_{3, 11}$=4.36) (Fig. 4.5). In 2014 and 2015 late in the season, the estimated total numbers of viruliferous adults in transplanted onion fields, weedy areas and direct-seeded onion fields were greater than those in culled onions, but none of the differences were significant (*P*>0.05) (Fig. 4.5). Temporally, there were numerical differences between estimated numbers of viruliferous thrips within the season (Fig. 4.5). Early in the season, cull pile sites had the greatest number of viruliferous thrips per card and accounted for between 65-86% of total viruliferous thrips. Transplanted onion sites had the greatest number of estimated viruliferous thrips during the mid-season period and accounted for 83 and 76% of the total in 2014 and 2015, respectively. Late in the season in both years, direct-seeded onion fields had highest densities of estimated viruliferous thrips and accounted for the highest percentages of estimated viruliferous thrips (48% in 2014 and 62% in 2015).

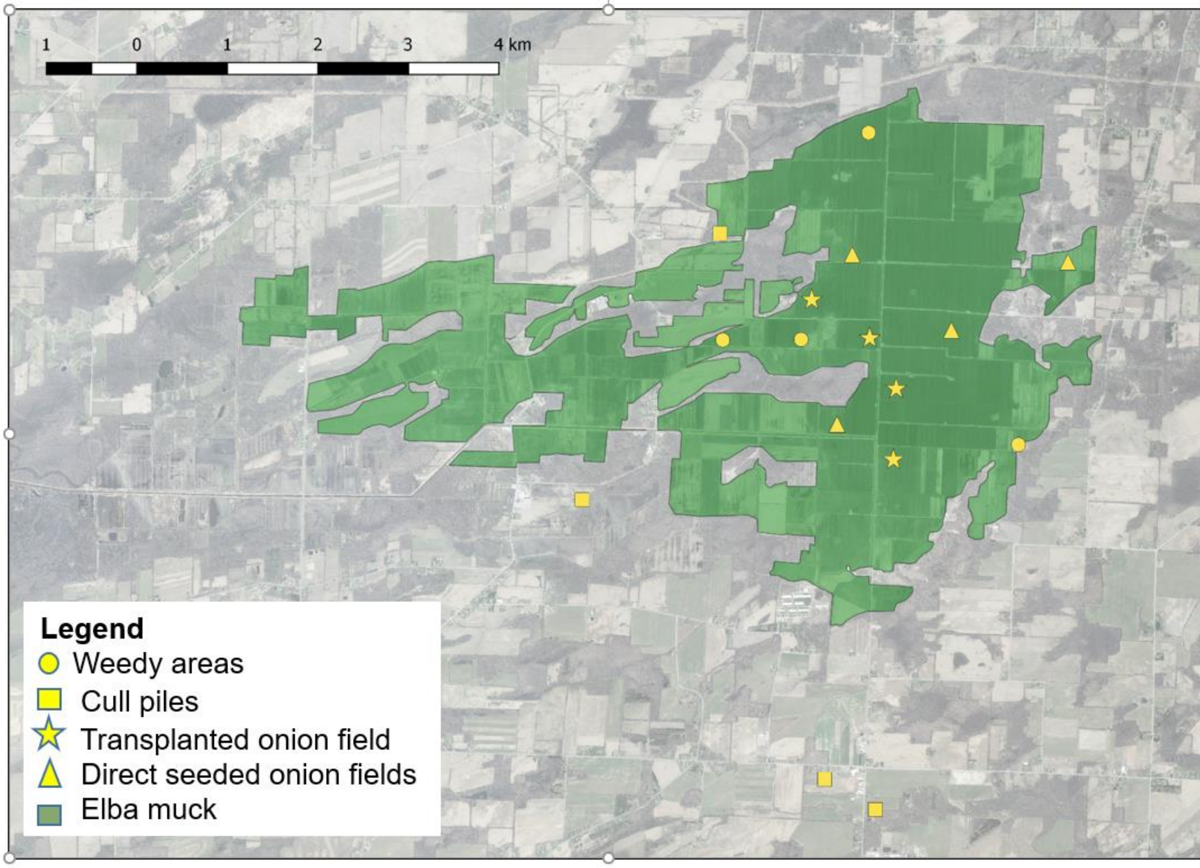

Figure 4.1: Sites monitored for adult onion thrips within the 'Elba muck' near Elba, NY in 2014 and 2015.

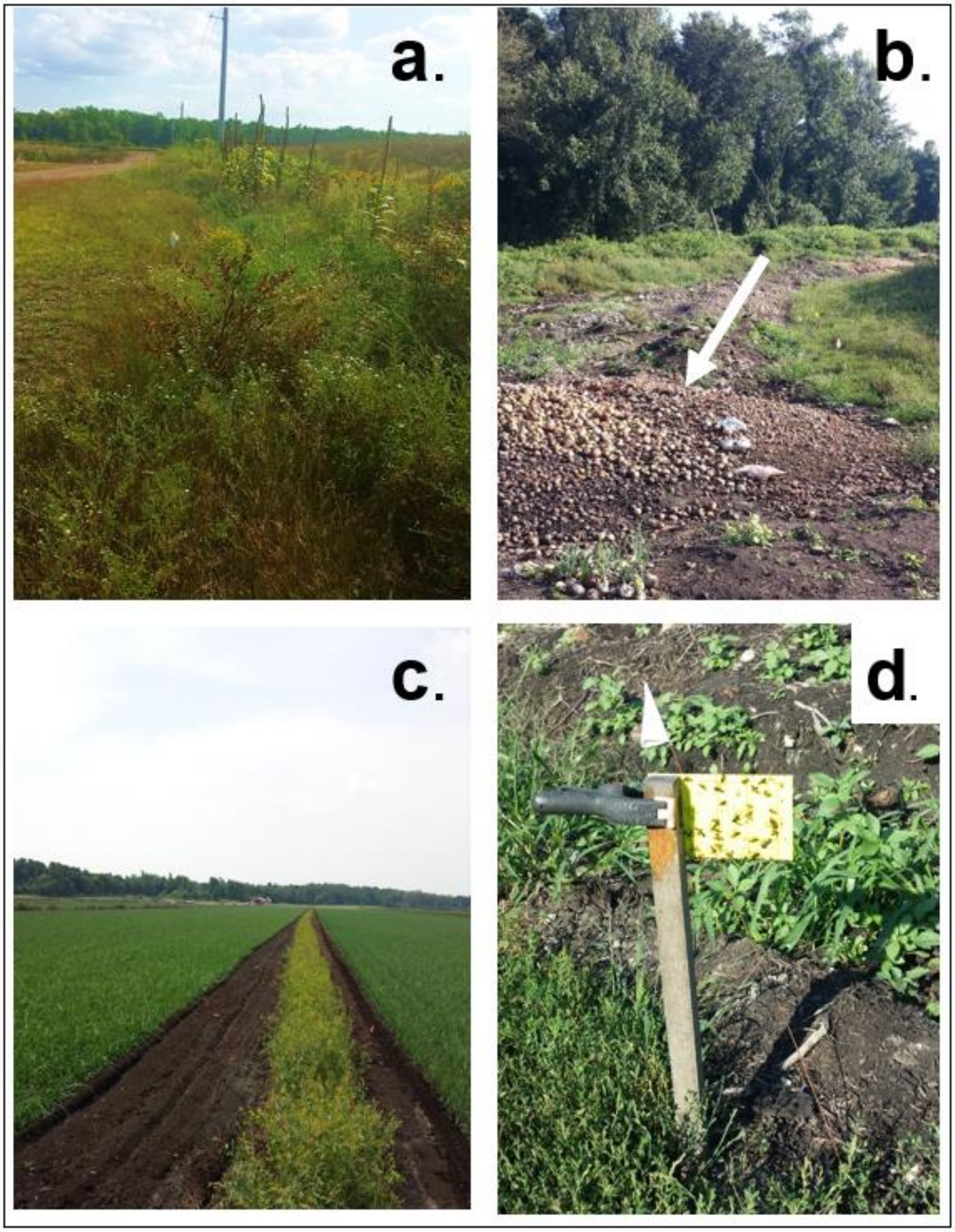

Figure 4.2: Habitat types sampled for onion thrips adults. Weedy areas (a), onion cull piles (b), and transplanted and direct-seeded onion fields (c) were monitored in western New York from June to early September in 2014 and 2015. Adult onion thrips were monitored weekly using yellow sticky cards (d).

Table 4.1: Total seasonal number of onion thrips adults, percent viruliferous thrips, and number of viruliferous onion thrips in culled onions, transplanted onion fields, weedy areas and direct-seeded onion fields near Elba NY in 2014 and 2015.

Year	Habitat type	Number of onion thrips adults per card [a,b]	Seasonal percent viruliferous thrips (%) [a,b]	Number of estimated viruliferous onion thrips adults per card [a,b]
2014	Culled onions	47 ± 5 b	4.5 ± 1.6 a	2 ± 1 a
	Transplanted onion fields	450 ± 55 a	6.4 ± 2.0 a	29 ± 17 a
	Weedy areas	203 ± 29 a	4.8 ± 1.9 a	10 ± 5 a
	Direct-seeded onion fields	172 ± 20 a	9.7 ± 1.8 a	18 ± 9 a
2015	Culled onions	44 ± 6 b	11.4 ± 1.1 b	5 ± 1 b
	Transplant onion fields	258 ± 45 a	30.6 ± 2.6 a	79 ± 14 a
	Weedy area	229 ± 72 a	11.1 ± 1.5 b	25 ± 19 a
	Direct-seeded onion fields	219 ± 66 a	20.8 ± 1.6 ab	46 ± 40 a

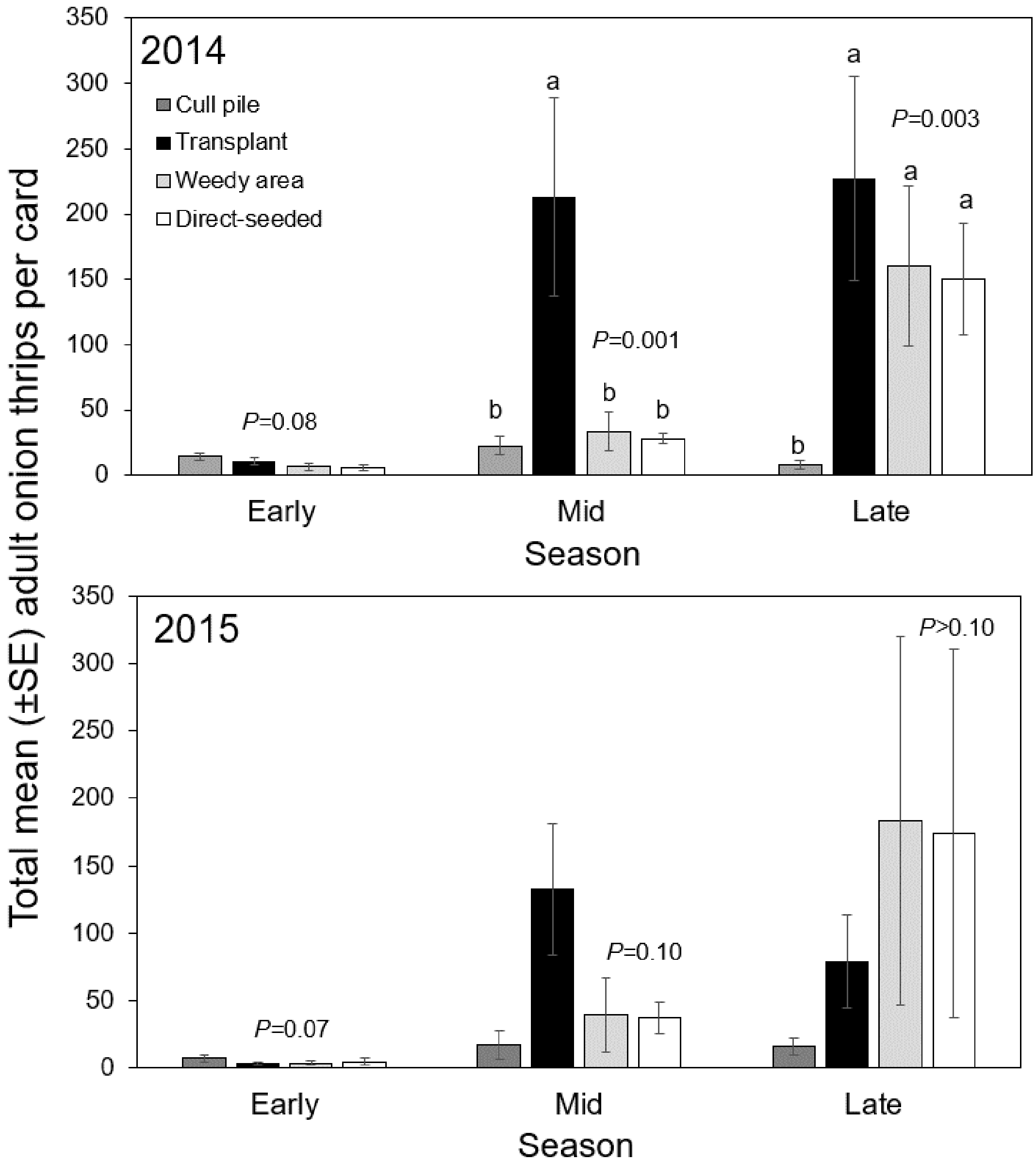

Figure 4.3: Mean (± SE) number onion thrips adults per card per sampling period within four habitat types (onion cull piles, transplanted onion fields, weedy areas, and direct-seeded- onion fields) near Elba, NY in 2014 and 2015. Thrips were morphologically identified to species. In both years, sampling was initiated when onions had 1-4 leaves, and concluded with onion harvest.

Table 4.2: Number of imported onion plants testing positive for IYSV using DAS-ELISA after being maintained in thrips-proof cages in a greenhouse for 3 months in 2014 and 2015. Plants were obtained before transplanting in the field.

Onion cultivar	Number of samples tested for IYSV			
	2014		2015	
	Total tested	Positive IYSV	Total tested	Positive IYSV
Brandt	173	0	194	3
Red Defender	204	0	233	3
Delgado	181	0	216	0
Festival	167	0	155	1
Moondance	104	0	-	-
Total tested	829	0	798	7

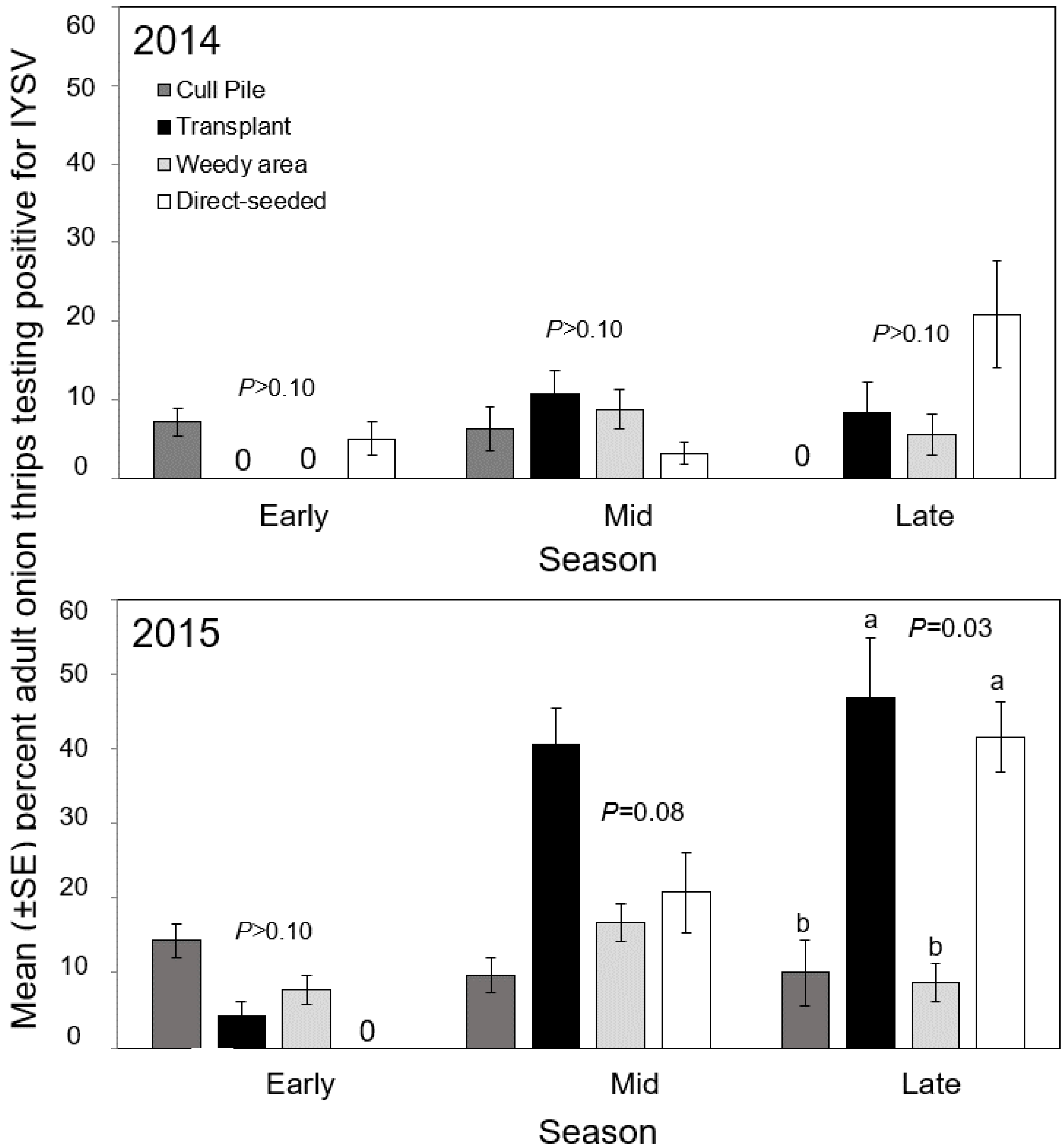

Figure 4.4: Mean (± SE) percent onion thrips adults testing positive for IYSV per sampling period within four habitat types (onion cull piles, transplanted onion fields, weedy areas, and direct-seeded- onion fields) that may foster IYSV epidemics near Elba, NY in 2014 and 2015. In both years, sampling was initiated when onions were 1-4 leaves, and concluded with onion harvest.

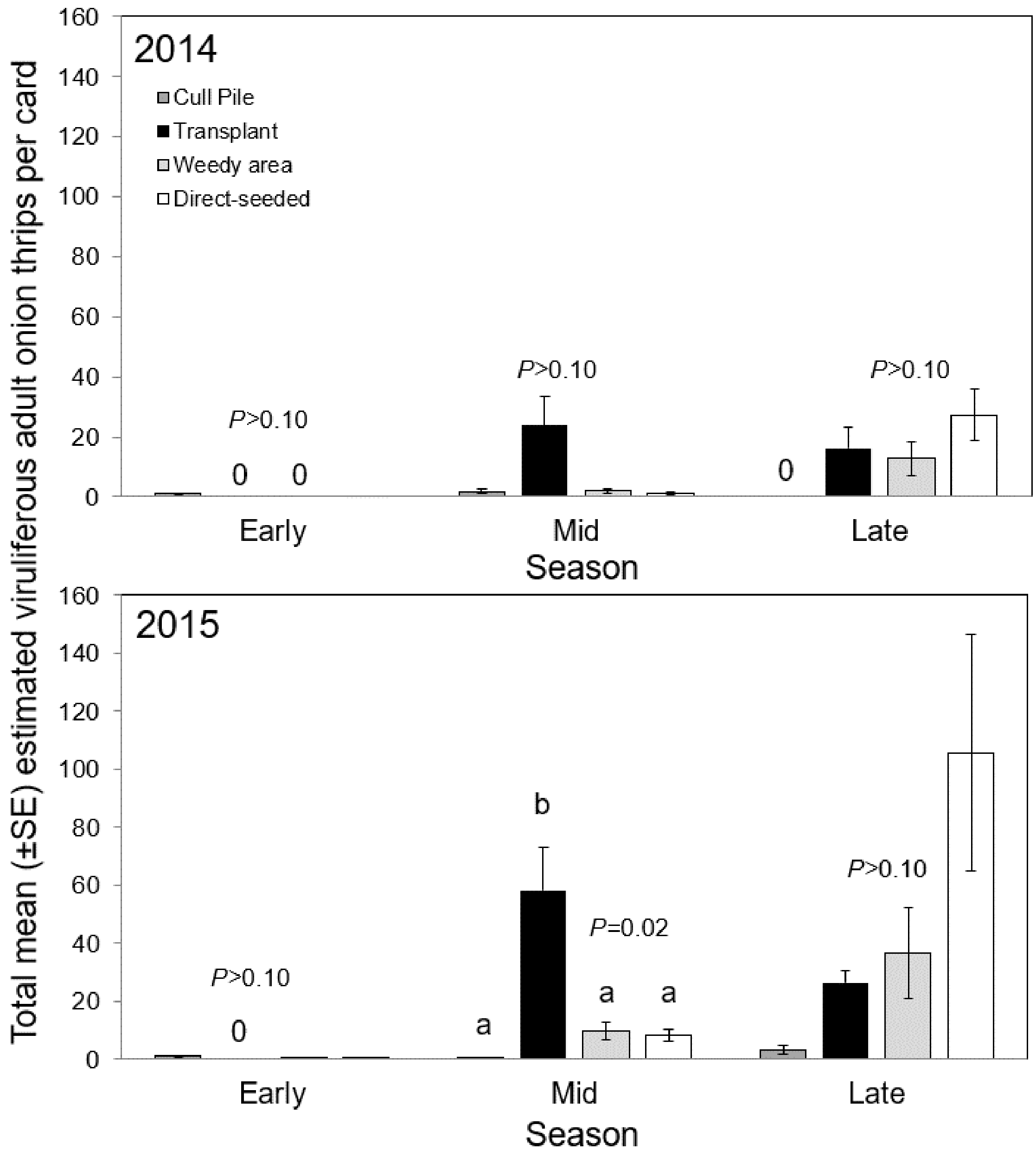

Figure 4.5: Mean (± SE) estimated number of viruliferous adult onion thrips early, middle and late in the growing season (June, July and August, respectively) within four habitat types (onion cull piles, transplanted onion fields, weedy areas, and direct-seeded onion fields) that may contribute to IYSV epidemics near Elba, NY in 2014 and 2015. In both years, sampling was initiated when onions were 1-4 leaves, and concluded with onion harvest.

4. Discussion

Onion thrips adults and IYSV were detected in all habitat types over the duration of the study. However, fields transplanted with imported onion plants had the greatest densities of onion thrips and highest seasonal incidences of viruliferous adults compared with other potential IYSV source habitats, weedy areas and culled onion sites. Moreover, transplanted onion sites accounted for 49-51% of the total estimated numbers of viruliferous thrips. From early to mid-season, transplanted onion fields had 9 to 11 times more viruliferous thrips compared to the other habitats. Because onion thrips adults colonize and reproduce in transplanted onion fields in late May and June, viruliferous adult thrips captured from these fields during mid-season in July were likely the progeny from the original colonizers. As we hypothesized, the overwhelming abundance of viruliferous adult thrips in transplanted onion fields during mid-season compared with those in culled onion sites and weedy areas strongly suggests that transplanted onions are the most important habitat for generating IYSV epidemics in all onion fields (transplanted and direct-seeded) later in the season.

The incidence of thrips testing positive for IYSV was three times greater in 2015 than 2014. One potential reason for this difference in viruliferous onion thrips populations may be the number of imported transplants that were infected with IYSV. Multiple studies have suggested that onion transplants may re-introduce IYSV annually into the onion production system (Gent et al. 2006; Hsu et al. 2010; Schwartz et al. 2014). Low levels of IYSV found in transplants prior to planting may supply inoculum to initiate epidemics later in the season (Gent et al. 2006; Hsu et al. 2011). In 2014, subsamples of transplants coming into New York from the southwestern US all tested negative for

IYSV (0 out of 829 transplants). In contrast in 2015, 7 out of 798 transplants (0.9%) tested positive for IYSV. While this is a low initial percentage of infected plants, it would have provided enough initial inoculum to foster the higher IYSV incidences in onion thrips observed in 2015. For example, New York onion growers often transplant approximately 642,500 onions per hectare. If 0.9% of the plants arrived already infected with IYSV, there would be a starting inoculum level of 578 IYSV-infected plants per ha. In the Elba muck, land is partitioned into a series of 4 hectare-fields, which would create a series of transplanted onion fields each starting with an estimated 2,300 IYSV-infected plants. Since a large portion of the Elba muck is planted using imported onion transplants, this can further increase IYSV inoculum in the onion production system.

Regardless of initial infection of imported onion plants, transplanted onion fields are likely an important habitat for IYSV inoculum, as it serves as a highly attractive homogenous IYSV host early in the season. Notably, zero onion plants tested positive for IYSV in 2014; however large populations of viruliferous thrips were estimated in transplanted onion fields. In early to mid-season, over 60% of thrips were captured in fields planted with imported transplants, and only 15% in weedy areas, 10% in culled onions, and 14% in direct-seeded onion fields. Hsu et al. (2010) reported that onions in transplanted fields had approximately 20% more onion thrips than those in direct-seeded fields early in the growing season. Preferential colonization of transplanted onion fields by onion thrips early in the season may have important epidemiological consequences when combined with the presence of IYSV-infected transplants, as it likely contributes to a large population of thrips acquiring IYSV early in the growing season.

In both years of this study, a temporal increase in the number of viruliferous adult thrips was observed in direct-seeded onion fields later in the season, which was likely due to immigration of adult thrips from adjacent, senescing transplanted onion fields. Our results showed that numbers of viruliferous adults peaked in transplanted sites in mid-season of each year, while the number of viruliferous thrips in direct-seeded sites peaked a month later in August. Approximately, 76-83% of the estimated viruliferous thrips were recorded in transplanted onion fields mid-season. However, in August, the majority (55%) of viruliferous thrips occurred in direct-seeded onion fields. Higher onion thrips populations at the end of the growing season in direct-seeded fields compared to transplanted onion fields have been previously reported in the Elba muck by Hsu et al. (2010) and Smith et al. (2017). Smith et al. (2011) also reported spikes in populations of onion thrips in weedy areas late in the season when onion fields were harvested. Since direct-seeded onions are not an initial source of IYSV (Kritzman et al. 2001), the high number of viruliferous adults in direct-seeded onion fields later in the season suggests secondary spread of the virus from initial sources of IYSV inoculum from transplanted fields. As the season progresses, viruliferous adults likely disperse to new sites that contain attractive hosts. In this production system, transplanted onion fields are planted adjacent to and simultaneously with direct-seeded fields, which facilitates movement of thrips between fields of differing developmental stages. Therefore, transplanted onion fields foster secondary spread of IYSV into nearby direct-seeded onion fields.

Viruliferous onion thrips were recorded in weedy areas and may also contribute to IYSV epidemics. However, we found that seasonal incidence of IYSV in weedy areas was lower than incidence in transplanted onion fields. Two potential reasons for this finding include fewer IYSV plant hosts in weedy areas and reduced fecundity of onion thrips on non-onion plant hosts. In New York, only four biennial/perennial plant species (i.e., dandelion, common burdock, curly dock, and chicory) have been demonstrated to be hosts of both onion thrips larvae and IYSV (Smith et al. 2011). In our study, we found that three of those species were present, and only comprised 6% of the weedy areas sampled. Thus, onion thrips were much less likely to encounter a suitable IYSV plant host in a weedy area versus an onion field. Additionally, studies have shown that thrips numbers are lower on weedy IYSV hosts compared to numbers on onion plants (Nischwitz et al. 2012; Smith et al. 2011). Notably, we found that *Amaranthus spp.* were common in the weedy areas sampled. Although *Amaranthus spp.* are hosts of IYSV (Sampangi et al. 2007), it is a poor-quality host for onion thrips (Schwartz et al. 2014; Smith et al. 2011). Smith et al. (2011) found that of the 25 weed species sampled, none exceeded greater than six thrips per plant over the course of the growing season. In onion, onion thrips densities can easily exceed 100 per leaf (Fournier et al. 1995).

At the end of each growing season, onion thrips likely migrate to weedy areas adjacent to onion fields. Like Smith et al. (2011), we observed large abundances of adult onion thrips in weedy areas on the last date of sampling. In New York, onion thrips can produce 6 to 8 generations and 2 to 3 of those generations occurring exclusively on non-onion plant hosts (Hoffmann et al. 1996). Therefore, as adult onion thrips move into weedy areas from onion fields, viruliferous thrips populations may decrease as thrips are less likely to encounter an IYSV plant host on which to feed and oviposit. Even if an adult successfully colonizes a plant that is a host for both the virus and vector, fewer progeny will likely be supported on that plant, which might reduce the overwintering population of viruliferous thrips the subsequent season. However, it should be noted that we did detect viruliferous adults in weedy areas as early as June, indicating that weeds may still provide a green bridge by which IYSV can persist (Hsu et al. 2010, Nischwitz et al. 2012, Schwartz et al. 2014, Smith et al. 2011).

The fewest numbers of adult onion thrips and those estimated to be viruliferous were captured near culled onions in both years of the study. Cull piles are unlikely to greatly contribute to IYSV inoculum via viruliferous onion thrips populations. These results are like those found by Szostek and Schwartz (2015), who reported few to zero thrips in cull piles. Like Szostek and Schwartz (2015), the cull pile sites in our study were likely poor habitats for onion thrips populations as they were dominated by decaying onions. While cull piles do not appear to contribute greatly to the overall amount of viruliferous adults in the landscape, thrips captured near cull piles had relatively high levels of IYSV early in the growing season. High percentages of viruliferous thrips were captured near cull pile sites (65% and 86%) early in the season in 2014 and 2015, respectively, and could potentially initiate virus epidemics if they emigrated to nearby onion fields. However, a relatively low percentage of adult onion thrips engage in long-distance dispersal, and even fewer do so early in the onion growing season (Smith et al. 2015). Nevertheless, cull piles should be spatially separated from onion fields to limit risk of viruliferous thrips migrating into onion fields.

Our results add to the growing body of literature addressing factors influencing the risk and development of iris yellow spot disease in onion. Our study is the first to estimate the relative significance of habitats containing IYSV sources in the landscape by surveying populations of viruliferous adult onion thrips. While we detected viruliferous adult thrips in all habitat types known to harbor IYSV, fields planted with imported onion transplants had the highest incidence of viruliferous onion thrips. Additionally, we consistently observed that transplanted onion fields had the greatest abundance of dispersing viruliferous thrips in the middle of the growing season (July). Therefore, onion thrips control in transplanted onion fields may be a priority for onion growers, especially early in the season, to reduce risk of IYSV spread to other onion fields later in the season. To potentially reduce the risk of viruliferous thrips dispersing from maturing transplanted fields into direct-seeded onion fields, growers may consider spatially isolating onion fields planted with imported onion transplants from direct-seeded onion fields.

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CHAPTER 4

IRIS YELLOW SPOT VIRUS PROLONGS THE ADULT LIFESPAN OF ITS PRIMARY VECTOR, ONION THRIPS (*THRIPS TABACI*)

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Abstract: Iris yellow spot virus (IYSV) from the genus Tospovirus, family Peribunyaviridae, reduces yield in several crops, especially Allium spp. IYSV is primarily transmitted by onion thrips (Thrips tabaci), but little is known about how IYSV impacts the biology of its principal vector. In a controlled experiment, the effect of IYSV on the lifespan and fecundity of onion thrips was examined. Larvae were reared on IYSV-infected onions until pupation. Individual pupae were confined until adults eclosed, and the lifespan and total progeny produced per adult were monitored daily. Thrips were tested for the virus in reverse-transcriptase polymerase chain reaction using specific primers to confirm the presence of IYSV. Results indicated that 114 and 35 out of 149 eclosing adults tested positive (viruliferous) and negative (non-viruliferous) for IYSV, respectively. The viruliferous adults lived 1.1 to 6.1 days longer (average of 3.6 days) than non-viruliferous adults. Fecundity of viruliferous and non-viruliferous onion thrips was similar with 2.0 ± 0.1 and 2.3 ± 0.3 offspring produced per female per day, respectively. Fecundity for both viruliferous and non-viruliferous thrips also was significantly positively correlated with lifespan. These findings suggest that the longer lifespan of viruliferous onion thrips adults may allow this primary vector of IYSV to infect more plants, thereby exacerbating IYSV epidemics.

Keywords: Onion thrips, Iris yellow spot virus, Tospovirus, Lifespan, Fecundity

1. Introduction

Viruses from the genus Tospovirus, family Peribunyaviridae are economically significant plant viruses responsible for annual yield losses of many agronomic crops (Pappu et al. 2009). These viruses are transmitted by thrips (Thysanoptera), which acquire the virus during larval development and remain viruliferous until death. Tospoviruses are persistent and propagative in their vector, replicating within the thrips midgut and associated digestive organs including the salivary glands (Wijkamp et al. 1993, Birithia et al. 2013). Like many plant viruses, tospoviruses alter the biology and behavior of their insect vectors. Studies have documented positive and negative effects of these viruses on their thrips vectors (DeAngelis et al. 1993, Stumpf and Kennedy 2005, Stafford et al. 2011, Shrestha et al. 2012, Stafford-Banks et al. 2014). However, tospovirus infection tends to increase vector fitness. For example, viruliferous Frankliniella spp. typically have more offspring and longer life spans than those not infected (Maris et al. 2004, Stumpf and Kennedy 2005, Shrestha et al. 2012, Ogada et al. 2013, Zheng et al. 2014, Keough et al. 2016).

Most research on tospoviruses and thrips has focused on describing the relationship between Frankliniella spp. and tomato spotted wilt virus (e.g. DeAngelis et al. 1993, Stumpf and Kennedy 2005, Stafford et al. 2011, Shrestha et al. 2012, Stafford-Banks et al. 2014); although interactions with other important thrips vectors have been examined (Birithia et al. 2013, Chen et al. 2014, Keough et al. 2016). Iris yellow spot virus (IYSV) from the genus Tospovirus significantly reduces yield in Allium crops (Pozzer et al. 1999, Gent et al. 2006). Onion thrips (Thrips tabaci) is the primary vector of IYSV, but there is limited information on the impact of IYSV on the fitness of onion thrips. Some studies have indicated that IYSV infection does not impact the reproduction or mortality of thrips when monitored for the first week after eclosion (Inoue et al. 2010, Birthia et al. 2013); however, no studies have examined the long-term effects of IYSV on the total lifespan and production of progeny of onion thrips. Knowledge of the impact that IYSV has on the lifespan and fecundity of onion thrips could provide better insight into the epidemiology of IYSV in Allium crops. For example, a longer lifespan and increased fecundity of viruliferous thrips or both could accelerate the spread of IYSV, thereby increasing IYS disease in agricultural systems. The purpose of this study was to examine the effect of IYSV infection on the lifespan and fecundity of onion thrips. We predicted that viruliferous thrips would positively benefit from IYSV infection by living longer and producing more offspring.

2. Materials and Methods

Plant and thrips collection

Onion transplants (cv. 'Bradley') exhibiting typical IYSV symptoms, including straw-colored diamond shaped lesions, were collected from an onion field in Elba, NY. All plants collected were similar in size (approximately six leaves and weighed 60 ± 10 g). Plants were free of any additional plant diseases and not treated with any insecticides or fungicides. All plants were collected early in the onion growing season (10 Jun 2017), when onion thrips populations are typically low to absent; therefore, infection likely occurred prior to transplantation. After collection, onion plants were transported to Cornell AgriTech in Geneva, NY, cleaned with ethanol to remove any thrips that might have been on the plants and then placed singly into thrips-proof cages ("2120F", BioQuip, Rancho Dominguez, CA) with a damp paper towel on the bottom of the cage. Plants were monitored for 14 days to ensure no thrips larvae emerged. These were considered our source onion plants. Onion thrips adults used in this study were acquired from a laboratory colony originally established from individuals collected from a non-IYSV infected onion field in Elba, NY in 2017. All subsequent thrips generations were reared on cabbage, which is not a host plant for IYSV (Smith et al. 2011).

Thrips and data collection

Approximately 25-30 adults from the laboratory colony were placed on the source onion plants and caged on the plants in a controlled environment and maintained at 25 ± 1oC with 60 ± 5% relative humidity and a photoperiod of 16 hours light and 8 hours dark. Adults laid eggs and larvae developed on these onions until pupation.

Pupae were removed from the cages and then placed singly into Falcon™ dishes (150 × 25 mm; Falcon™, item #353025, BD, Franklin Lakes NJ, US) containing a single cabbage leaf disc (5 cm diameter, ~6 cm3 volume). Cabbage is a highly desired host for onion thrips, but not a host for IYSV. Therefore, cabbage was an ideal food source and ovipositional medium for onion thrips in our study.

Observations of adult lifespan and fecundity began as soon as adults eclosed and thrips were monitored every 24 hours until death. Because this was a thelytokous population of onion thrips, all individuals were female and reproduced parthenogenetically (= referred to as mother from here on). At 5-day intervals, cabbage discs in each dish were examined for larvae. Mothers were transferred to new falcon dishes containing a new cabbage disk every 5 days. Progeny were counted on each disk, and then summed to determined total progeny per mother. Mothers were deemed ‘alive’ if they moved when observed or gently prodded with a paintbrush tip. If a mother died, she was placed into a 0.5 ml centrifuge tube and stored at -80o C until tested for IYSV with reverse-transcriptase polymerase chain reaction (RT-PCR) to determine her vector status. Any mother that died within the first 24 hours of observation was excluded from analysis, which resulted in a total of 149 thrips mothers monitored and tested for IYSV in this study.

IYSV testing

All thrips (n=149) were tested for the IYSV nucleoprotein (N) gene using RT-PCR and total RNA isolated from individual thrips using modified procedures from the Omega MicroElute RNA Kit (Omega Bio-Tek, Norcross, GA), as previously described (Leach et al. 2018).

The diagnostic primers used to detect IYSV were IYSV-N402F 5’-ACTCACCAATGTCTTCAAC-3’ and IYSV-N402R 5’-GGCTT CCTCTGGTAAGTGC-3’, which were designed from the N gene of several IYSV isolates collected in New York (Leach et al. 2018). Primers ThMCOI-F 5’-CGGGAACGGGATGAACAG-3’ and ThMCOI-R 5’-GGTCCCCTCCCC CTCTA-3’ (designed in the mitochondrial cytochrome oxidase subunit I gene sequence, GenBank accession no. DQ228494) were used in a multiplex RT-PCR to confirm the nature of onion thrips and ensure quality of RNA extracts. Non-viruliferous thrips, which were reared exclusively on cabbage, were included in RT-PCR testing to protect against false negatives. RT-PCR was carried out with the Qiagen one-step kit in a final volume of 12.5 µl and the following thermal cycling conditions: 50°C for 30 min (1 cycle), 95°C for 15 min followed by 40 cycles of 94°C for 30s, 50°C for 1 min, 72°C for 1 min, and a final extension at 72°C for 10 min (BioRad ThermalCycler). RT-PCR products (402 bp for IYSV and 325 bp for onion thrips) were stained with GelRed (Biotium, Hayward, CA) following electrophoresis on 1.5% agarose gels, and then imaged using ultra-violet illumination.

Statistical analysis

The lifespan of each mother and numbers of her offspring were analyzed using R statistical software and packages ‘lme4’ (Bates et al. 2015) and ‘emmeans’ (Lenth et al. 2016). Lifespan data were analyzed with a normal distribution, and fecundity data (number of progeny and progeny per day) were analyzed using a Poisson distribution. Vector status (viruliferous or non-viruliferous) was treated as a fixed effect, individual thrips nested by individual source plant (specific onion plant that the mother was originally reared on as a larva) as the random effect, and a weight term was included to correct for the differing sample sizes between status groups. Differences within each analysis were compared using a one-way ANOVA. Differences within each analysis were compared using least square means ($P<0.05$).

3. Results

All symptomatic plants used in this experiment yielded onion thrips that tested both positive and negative for IYSV. Many of the mothers tested positive for IYSV (77%; n = 114) and were considered viruliferous, whereas the remainder did not (23%; n= 35) and were considered non-viruliferous. Vector status significantly impacted the lifespan of the onion thrips mothers ($P= 0.02459$, $F_{1,144}=5.02$) (Table 5.1). Viruliferous adults lived for 20.2 ±1.6 days, which was 1.1 to 6.2 days longer (average of 3.6 days) than non-viruliferous adults (16.6 ± 0.9 days). Differences in survival were observed early in the data collection, as 28.6 ± 0.9% of non-viruliferous thrips died within the first 5 days, which was significantly greater than the percentage of viruliferous thrips that died at that point (19.3 ± 0.4%) ($P<0.001$, $F_{1,144}=132.4$).

Fecundity of viruliferous and non-viruliferous thrips was similar ($P>0.05$) (Table 5.1). Viruliferous thrips produced an average of 40.4 ± 6.9 offspring per female and non-viruliferous produced an average of 38.2 ± 3.2 offspring per female. The number of larvae produced per day was not significantly different between mothers who were or were not viruliferous, as both groups produced approximately 2 larvae per day (Table 5.1). Most thrips produced the greatest number of progeny between 7-21 days from adult emergence. Fecundity of thrips from both groups also was significantly positively correlated with lifespan ($P<0.001$, $F_{1,144}= 595.1$) (Figure 5.1).

Table 5.1: Mean lifespan and fecundity of adult onion thrips infected (viruliferous) and not infected (non-viruliferous) with iris yellow spot virus. Females were monitored daily until death, and total progeny per female counted. Thrips infected with IYSV were confirmed with RT-PCR.

Vector status	*n*	Mean lifespan (days) ± SE*	Mean progeny (emerged larvae) ± SE*	Mean progeny per day ± SE*
Viruliferous	114	20.2 ± 1.6 a	40.4 ± 6.9 a	2.0 ± 0.1 a
Non-viruliferous	35	16.6 ± 0.9 b	38.2 ± 3.2 a	2.3 ± 0.3 a

*Significant values determined by LSMEANS at a 0.05 significance level.

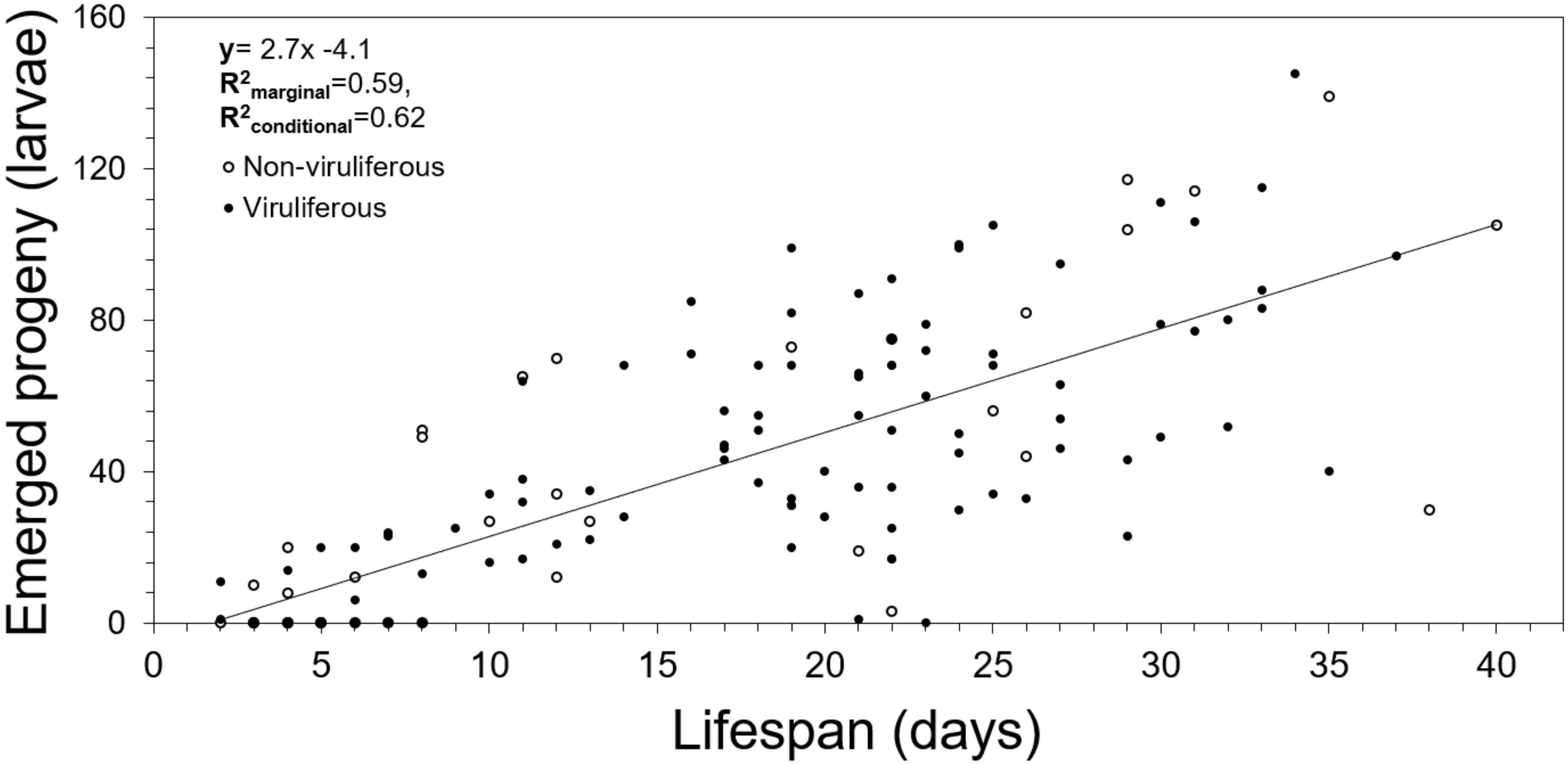

Figure 5.1: Relationship between an onion thrips mother's lifespan and her total progeny (number offspring over lifespan) produced for mothers that were either viruliferous with iris yellow spot virus or were not viruliferous. Each data point represents a single mother and the total number of her progeny. A total of 149 thrips were monitored in this study, 114 were viruliferous and 35 non-viruliferous.

4. Discussion

Onion thrips were positively impacted by IYSV infection, as viruliferous thrips lived almost four days longer than those not infected. Our results contrast from previous studies that reported no significant effect of IYSV infection on onion thrips mortality, reproduction or development (Inoue et al. 2010, Birithia et al. 2013). Inoue et al. (2010) reported that onion thrips mortality, development, and reproduction were not significantly different between groups feeding on infected and healthy tissue and noted that IYSV-exposed thrips had numerically higher mortalities than unexposed thrips. Similarly, Birithia et al. (2013) found no significant difference in the mortality rates between onion thrips feeding on IYSV-infected tissue and healthy tissue (virus-free). The difference between our results and those mentioned above may be methodological. Previous studies did not confirm the vector status of thrips tested. Rather, there was an assumption that the thrips would be viruliferous after feeding for 16 hours on IYSV-positive plant tissue (Inoue et al. 2010, Birithia et al. 2013). However, acquisition of tospoviruses by thrips can vary (Bautista et al. 1995, Hunter et al. 1995, Srinivasan et al. 2012). Variable virus acquisition may confound experimental results as numbers of non-viruliferous thrips may be underestimated, thereby reducing the likelihood of finding significant differences between treatment groups. In our trial, we observed that only 77% of thrips acquired IYSV after feeding on symptomatic plants during larval development; thus, larval feeding on IYSV-infected onion plants did not guarantee IYSV infection. Therefore, it was important to confirm the vector status of each thrips to correctly associate effects of a tospovirus infection with onion thrips reproduction and mortality.

Another difference in methodology between our study and those described in Birithia et al. (2013) and Inoue et al. (2010) was that onion thrips was reared on fabaceous hosts including soybean (Glycines max) and snow pea (Pisum sativum var. saccharatum). These two species are sub-optimal hosts of T. tabaci compared with Alliums and Brassicas (Doederlein et al. 1993, Lewis et al. 1997). Therefore, it is possible that host plant quality may significantly impact the effect of tospoviruses on adult thrips biology, thereby masking differences in lifespan between viruliferous and non-viruliferous adults.

In our study, viruliferous thrips lived longer. This may increase the rate of IYSV spread in onion fields. While there are no studies that have documented the daily movement of an individual thrips over time, studies have shown that populations of adult thrips are very mobile (Smith et al. 2015). Thrips move readily both within and between plants (Lewis 1997) and are known to disperse long distances, in some cases hundreds of kilometers under the right environmental conditions (Laughlin 1977, Lewis 1997). Studies in New York onion fields (Elba, NY) showed that onion thrips tended to disperse short distances, but some engage in long-distance dispersal (Smith et al. 2015). A longer lifespan may provide adults with more time to disperse and feed on multiple host plants, thereby increasing the number of plants infected with IYSV, and consequently accelerating epidemics.

IYSV infection may have additional impacts on onion thrips biology. Indeed, other studies have identified many effects of tospoviruses on thrips biology and ecology including increased development time, changes in probing behaviors, and differences in dietary preferences (Stumpf and Kennedy 2005, Stafford et al. 2011, Stafford-Banks et al. 2014, Zheng et al. 2014). Further studies are needed with larger sample sizes, and different IYSV isolates and thrips populations to fully evaluate the impact of IYSV on these aspects of onion thrips biology.

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CHAPTER 5

GROWER ADOPTION OF INSECTICIDE RESISTANCE MANAGEMENT PRACTICES INCREASE WITH EXTENSION-BASED PROGRAM

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Abstract

BACKGROUND: Insecticide resistance management (IRM) practices that improve the sustainability of agricultural production systems are developed, but few studies address the challenges with their implementation and success rates of adoption. This study examined the effectiveness of a voluntary, extension-based program to increase grower adoption of IRM practices for onion thrips (*Thrips tabaci*) in onion. The program sought to increase the use of two important IRM practices: rotating classes of insecticides during the growing season and applying insecticides following an action threshold.

RESULTS: Onion growers (n=17) increased their adoption of both IRM practices over the three-year study. Growers increased use of insecticide class rotation from 76% to 100% and use of the action threshold for determining whether to apply insecticides from 57% to 82%. Growers who always used action thresholds successfully controlled onion thrips infestations, applied significantly fewer insecticide applications (1-4 fewer applications) and spent $148/hectare less on insecticides compared with growers who rarely used the action threshold. Growers who regularly used action thresholds and rotated insecticide classes did so because they were primarily concerned about insecticide resistance development in thrips populations.

CONCLUSION: Implementation of the IRM education program was successful, as adoption rates of both practices increased within three years. Growers were surprisingly most receptive to adopting these practices to mitigate insecticide resistance as opposed to saving money. Developing extension-based programs that involve regular and interactive meetings with growers may significantly increase the adoption of IRM and related integrated pest management tactics.

KEY WORDS: Insecticide resistance, management, extension, adoption, onion thrips

1. Introduction

The development of insecticide resistance is a threat to many agricultural production systems where insecticides are applied. Over 500 insect species have developed resistance to one or more insecticides (Sparks and Nauen 2015), which has contributed to a global yield loss of 1.5 billion dollars (USD) annually (Pimentel and Burgess). This loss can be further exacerbated by the lack of new, readily available insecticides. New active ingredients are costly to develop and can take between 10 to 15 years until they are commercially available (Sparks 2013). Thus, insecticide resistance management (IRM) tactics, including chemical class rotation, using thresholds, and other non-chemical control measures are needed to maintain the profitability and stability of agricultural systems. Numerous research efforts have identified IRM and related integrated pest management (IPM) tactics to slow the onset of insecticide resistance in a variety of agricultural production systems (Huseth et al. 2014; Bielza 2008; Palumbo et al. 2001; Tabashnik et al. 1991; Haynes et al. 1987). While the efficacy and application of IRM is dependent on the specific pest biology and agricultural production system, the goal of these techniques is to reduce the selection pressure of a given active ingredient on an insect pest, thus prolonging the active ingredient's efficacy (IRAC 2017).

The effectiveness of IRM and related IPM practices to delay the onset of insecticide resistance is largely predicated on grower decision and compliance (Hurley and Mitchell 2008; Siegfried et al. 1998). However, our understanding of the implementation and adoption of IRM and related IPM practices is relatively limited (Peshin and Karla 2009). Previous studies and surveys on general IPM practices reveal that rates of grower adoption vary from 30-99% depending on region and commodity (Farra et al. 2016;

Blake et al. 2007; Kaine and Beswell 2008; Vandeman et al. 1994; Fernandez-Cornejo et al. 1994). Currently, the USDA estimates that 70% of US cropland is managed using some level of IPM (GOA 2001), however the use of IRM tactics is unknown. Growers tend to adopt practices that are not risky, easy to implement, and save money (Farrar et al. 2016; Khan and Damalas 2014; Trumble 1998), which can put some IRM and related practices at a disadvantage because many are complicated and time-consuming to implement. Consequently, the adoption of some IPM practices have been slow to progress as compared with other agricultural technologies (Zalucki et al. 2009; Kogan and Bajwa 1999). Adoption of IPM practices has been associated with many factors including farm size and age of grower (Punete et al. 2011; Wearing 1988), but grower education and inexperience remain the greatest impediments for IPM and IRM practice adoption (Farrar et al. 2016). Many studies have evaluated the effect of different educational programs on grower's knowledge (Thomas et al. 2018; Landis et al. 2016; Van den Berg and Jiggins 2007) and adoption of IPM (Stephens et al. 2017; Allahyari et al. 2016; Kabir and Rainis 2015). Nevertheless, further research is needed to identify those methodologies that can successfully increase adoption of IRM and related IPM tactics to mitigate the onset of insecticide resistance.

Poor insecticide resistance management has resulted in pest control failures worldwide. In onion production systems, insecticide resistance in onion thrips (*Thrips tabaci*) populations has led to significant yield losses. Onion thrips has developed resistance to pyrethroids, organophosphates, and carbamates (Herron et al. 2008; Shelton et al. 2006; MacIntyre et al. 2005; Martin et al. 2003; Shelton et al. 2003). Previous research has identified two pest management practices that should mitigate insecticide resistance and control onion thrips populations; using an action threshold (Nault and Huseth et al. 2016; Nault and Shelton 2010) and following an insecticide sequence that rotates insecticide classes (Nault 2015). The use of thresholds is an important component to insecticide resistance management programs (IRAC 2017). In onion production in the Great Lakes region, an action threshold of one thrips per leaf has been effective in controlling thrips populations without reducing yield (Nault and Huseth 2016; Leach et al. 2017). Implementing an action threshold to control thrips in onion production can reduce the frequency of insecticide applications between 30-50%, thereby reducing exposure of insecticides to onion thrips populations (Nault and Huseth 2016; Leach et al. 2017). Recent research also has identified effective thrips management using season-long rotation sequences of insecticides belonging to different classes (Nault 2015; Werling and Szendrei 2015; Nault et al. 2012; Nault and Shelton 2010). Onion thrips typically complete a generation in approximately 14 days on onion (Jamieson et al. 2012), thus no more than two consecutive sprays of the same mode of action is recommended. As such, proposed insecticide sequences include multiple products with different modes of action applied twice 7-10 days apart (Nault 2015; Werling and Szendrei 2015; Nault et al. 2012). This approach should reduce exposure of an insecticide to multiple generations of onion thrips and slow the potential onset of insecticide resistance (Espinosa et al. 2002; Immaraju et al. 1992; Immaraju et al. 1990). Recent onion grower survey results in New York revealed that only 52% of growers rotated insecticide classes, and even fewer (40%) used an action threshold to determine when to make an insecticide application (Nault BA, unpublished). Therefore, an opportunity existed to help onion growers improve their adoption of action thresholds and rotation of insecticide classes following research-based IRM tactics, while maintaining acceptable levels of onion thrips control.

The purpose of this study was to improve the adoption of research-based IRM tactics for onion thrips in onion. We developed an extension program entitled, “IRM adoption program” to increase onion grower adoption of 1) an action threshold to make decisions about insecticide use, and 2) a rotation of insecticide classes in a season-long sequence that adhered to resistance management principles. We hypothesized that the use of action thresholds and rotation of insecticide classes would increase over the three-year program, and conservatively estimated that growers would collectively increase their use of both tactics by 10% annually. Furthermore, we anticipated that growers who adopted these tactics would positively benefit by applying fewer insecticide applications, reducing total insecticide cost, while successfully managing onion thrips infestations.

2. Materials and Methods

2.1 Thrips management approaches prior to the IRM adoption program

2.1.1 Grower participants. Onion growers from four of the major onion-producing counties in New York participated in this program, and all were familiar with Cornell Entomology and Cornell Cooperative Extension. Invitations to participate in the scouting program were sent to all known commercial onion growers from each county (n~22). Those growers who responded to the invitations were selected as participants for the ‘IRM adoption program’. The counties included Orleans, Wayne, Orange, and Oswego. In 2015, 15 growers participated in the program. In 2016, 2 additional growers joined the program for a total of 17, and in 2017, 14 growers continued to participate in the program (Supplemental figure 6.1).

2.1.2 Farm demographics and onion thrips management practices. Prior to initiating the IRM program, a survey was given to all participating growers to obtain baseline information about their farm demographics as well as the tactics they used for managing onion thrips (Supplemental table 6.1 and Supplemental figure 6.2). All growers who participated in the IRM adoption program were commercial vegetable producers and farmed between 22 and 2023 hectares of onions annually. Growers who participated in this study collectively managed 45 to 60% of the total onion hectarage in New York from 2015 to 2017 and represented 28% of the commercial onion growers in the state. The average grower participant operated a 51-hectare farm (Supplemental figure 6.2).

Most growers responded that they implemented IPM tactics on their farm to control onion thrips populations (Supplemental table 6.1). Approximately 76% of growers stated that they implemented a cultural pest management tactic, but none used either biological or physical controls to reduce onion thrips infestations. Approximately 88% of growers either scouted their own onion fields or had a professional crop consultant scout their fields. Many growers (65%) claimed to use an action threshold to determine when to apply an insecticide. However, most growers made between seven and eight insecticide applications each season specifically targeting onion thrips, which typically follows a standard or weekly insecticide program (Nault and Shelton 2010). Most growers (94%) claimed to effectively rotate insecticides in an effective season-long sequence, and only made two sequential applications of one mode of action before rotating to a new insecticide.

2.2 IRM adoption program

All growers who participated in this program received free, weekly scouting information from personnel affiliated with either Cornell Cooperative Extension or the Department of Entomology. All scouts had previous experience scouting agricultural crops for insect pests and had been properly trained to correctly identify and count onion thrips on onion prior to program initiation. Each scout was assigned a location within the state (Supplemental figure 6.1) where he or she would work with a sub-set of onion growers from that county. Each grower selected one onion field ranging from 4-8 hectares that was scouted weekly for the entire onion growing season. Initiation and conclusion of scouting depended on the phenology of the crop, not on previous history of thrips infestations in that field. Scouting typically began in early to mid-June and concluded in late August for a total of approximately 10 to 13 weeks.

Scouts randomly sampled onion plants within fields and visually assessed plants for onion thrips adults and larvae (Reiners and Seaman 2015). Within 24 hours of sampling fields, scouts sent a report to each onion grower documenting the infestation level of onion thrips in their field, whether the population exceeded an action threshold of one thrips per leaf (including both adults and larvae), and if so, what insecticide product and rate to use. In most cases, growers and scouts met and discussed this scouting information and recommendation. All scouts were unified in providing the same advice throughout the season.

A minimum of one week between applications was recommended. Insecticide products, rates and the sequence for applying these products were as follows: 1) Movento® at 5 fl oz. per acre (350 g per ha) (spirotetramat) (Bayer CropScience, Research Triangle Park, NC), 2) Agri-mek SC® at 3.5 fl oz. per acre (245 g per ha) (abamectin) (Syngenta, Greensboro, NC), 3) a co-application of Lannate® LV at 48 fl oz. per acre (3360 g per ha) (methomyl) (DuPont Crop Protection, Wilmington, DE) and Warrior® at 1.9 fl oz. acre (140 g per ha) (lambda-cyhalothrin) (Syngenta, Greensboro, NC), and 4) Radiant® SC at 8 to 10 fl oz. per acre (560-700 g per ha) (spinetoram) (Dow AgroSciences, Inc., Indianapolis, IN). In 2016, Exirel® (cyantraniliprole) (DuPont Crop Protection, Wilmington, DE) also was recommended at 13.5 fl oz. per acre (945 g per ha) as a substitution for the Lannate® LV and Warrior® combination. In 2017, Minecto™ Pro (premix formulation of cyantraniliprole and abamectin) was registered in New York for controlling onion thrips on onion and was consequently included as an insecticide option provided to growers. Minecto™ Pro was recommended at 7 to 10 fl. oz. per acre (490-700 g per ha) (abamectin and cyantraniliprole) (Syngenta, Greensboro, NC). Movento® (spirotetramat), Radiant® SC (spinetoram), Exirel® (cyantraniliprole), and Minecto™ Pro (premix formulation of cyantraniliprole and abamectin) provide excellent control of onion thrips larvae. Agri-mek® SC, Lannate® LV, and Warrior® are less effective insecticides, however they often provide suppression or limited control, and thus are still recommended at specific times throughout the season. Agri-mek (abamectin) offers only thrips suppression. While onion thrips populations in New York have developed resistance to both methomyl and lambda-cyhalothrin, the mixture of the two insecticides has been shown to provide better thrips control than the level of control provided by either product alone (Reiners and Seaman 2015). Growers were encouraged, but not required, to follow the action threshold recommendations and insecticide sequences provided by the scouts.

At the end of each growing season, every grower supplied pesticide application records for fields sampled by the scout (i.e., products, rates, dates of application). Pesticide application records were compared with weekly thrips density data to determine whether the grower complied with the IRM guidelines (i.e., following the action threshold and/or the insecticide sequence that rotated chemical classes). Additionally, annual post-season meetings between scouts and all growers within each county were held, where scouts discussed all insecticide records with the group. All 17 participating growers, who collectively represent between 45-60% of the onion acreage in New York, completed a survey describing their experience participating in the program (Supplemental figure 6.3).

2.3 Measurement of IRM adoption and definitions of associated metrics

Every insecticide application made by participating onion growers was analyzed based on its compliance with the action threshold and an insecticide rotation sequence. An insecticide application complied with the action threshold if applied when onion thrips densities exceeded the action threshold of one thrips per leaf. Applications were noncompliant if applied below the action threshold. Insecticide applications complied with insecticide rotation requirements if no more than two consecutive insecticide applications of a single mode of action or insecticide group was applied. Conversely, an insecticide application was considered noncompliant if more than two insecticide applications of a given class were applied and if the same insecticide was not applied consecutively. For each participating grower, the number of compliant insecticide applications from either IRM tactic was compared with the total number of insecticide applications made in every year to determine overall adoption success of each tactic.

In response to recent research (Nault BA, unpublished), all growers were recommended to apply two sequential applications of Movento® (spirotetramat) early in the growing season either before onions were bulbing (4-6 leaves) or when onion thrips densities reached 0.5 thrips per leaf. Therefore, an application of Movento® (spirotetramat) at this lower density was considered as compliant in 2017; no other times or for no other insecticides was this lower threshold compliant. Total insecticide cost per hectare was estimated using prices obtained from local agrichemical dealers. The costs of surfactants and other spray adjuvants were not included in overall cost estimates because they are routinely used and similarly priced. Insecticides were characterized as either inexpensive (<$24 (USD)/hectare) or expensive (>$72 (USD)/hectare). Movento® (spirotetramat), Radiant® SC (spinetoram), and Exirel® (cyantraniliprole) insecticide applications were considered expensive, whereas Warrior® (lambda-cyhalothrin) and Agri-mek® SC (abamectin) insecticide applications were considered inexpensive. Insecticides priced between $24-72/hectare (Lannate® (methomyl) mixed with Warrior® (lambda-cyhalothrin) and Minecto™ Pro (Minecto™ Pro (premix formulation of cyantraniliprole and abamectin)) were infrequently used and excluded from this analysis. All insecticides were characterized based on chemical class and the number of applications from each insecticide class was counted for every grower in each year.

2.4 Statistical analysis

2.4.1 Adoption analysis. Data were fit using generalized linear mixed effect models (GLMER, LMER) using the R library lme4 package (Bates et al. 2015). Adoption data (i.e., percentage of insecticide applications made when thrips density exceeded the action threshold of the total applied; percentage of insecticide applications that were rotated properly of the total applied) were analyzed with the lme4 package and function glmer() for binomial regression. Years in program (participating years) was treated as a fixed effect and grower within county as a random effect. The ‘IRM adoption program’ was initiated in 2015; however, the number of participating growers differed between years, which affected the number of years a grower participated in the program. This was accounted for by generating a new variable (participating years) that was used in the analysis rather than calendar year (e.g., 2015, 2016, 2017). Differences in adoption data between years were determined using ANOVA, and differences separated using Tukey’s HSD ($P<0.05$).

2.4.2. Post-hoc analysis of metrics associated with IRM adoption. Analyses were conducted to determine if adoption of either IRM tactic (independent variable) significantly affected seasonal onion thrips densities, number of insecticide applications, and costs and types of insecticides (expensive or inexpensive) used. These metrics were analyzed using adoption data (same as mentioned previously) as fixed effects. Growers within county were treated as a random effect. Seasonal onion thrips densities, number of insecticide applications, and costs of insecticides data were normally distributed, and analyzed using function lmer() for linear regression. Numbers of products and counts of expensive and inexpensive insecticides were analyzed using a Poisson distribution with function glmer(). Additional analysis identified the relative thrips abundance over the three-year period, which was analyzed using function lmer() with participating year as a fixed effect and growers within county as a random effect. Differences in treatments (seasonal onion thrips densities, number of insecticide applications or products, and costs of insecticides etc.) were determined using ANOVA, and differences separated using Tukey's HSD ($P<0.05$). Marginal and conditional R-squared values were determined using package, MuMIn, and function r.squaredGLMM() (Barton 2009).

3. Results

3.1 Onion thrips pressure

Onion thrips densities were slightly higher in years 1 and 2 compared to year 3, but this difference was only marginally significant ($p=0.059$, $F_{2,39}=5.64$). In years 1 and 2, seasonal densities of onion thrips were 0.6± 0.1 and 0.8± 0.2 thrips per onion leaf respectively, which was greater than densities in year 3, 0.4± 0.1 thrips per leaf. Onion thrips densities in onion fields were significantly different across counties (Table 6.1). Across all years, onion fields in Orleans County tended to have the greatest average number of thrips per leaf (1.1±0.2), which was significantly greater than densities in Oswego (0.3±0.1), but not Wayne (0.4±0.1) or Orange (0.6±0.1) counties ($p=0.003$, $F_{3,39}=13.5$).

No growers reported reduced onion bulb yields from onion thrips damage in this study using either the action threshold or rotating insecticide classes. Most growers stated that they effectively controlled thrips in all three years. Growers who regularly used the action threshold did not express lower satisfaction with their thrips control and did not report any “poor” or “failed” control of thrips in any year of the program. In year 1, approximately 94% (16/17) of growers stated that they had “good” or “excellent” control of onion thrips. Similarly, in Year 2, most (88%, 15/17) growers said that they had “good” or “excellent” control of thrips. Some growers reported having slightly reduced onion thrips control in year 2, as 12% said that they had “average” control of thrips, as compared with year 1 when only 6% (1/17) of growers reported having had “average” control of thrips. In year 3, growers across the state experienced high levels of thrips control, with most growers (83%, 10/12) having excellent control, 17% (2/12) having “good” control, and none (0/12) having ‘average’ control.

3.2 Adoption of the action threshold.

3.2.1 Adoption frequency of the action threshold. Growers significantly increased their use of the action threshold over the three-year program (Figure 6.1a) ($p= 0.006$, $F_{2, 41}=9.98$). More insecticide applications were applied following an action threshold in year 3 as compared with year 1 (82% and 57% respectively) (Figure 6.1a). Specifically, there were large increases in complete adoption of the action threshold (100% of insecticide applications made in accordance to the action threshold) by individual growers from year 1 to year 3. Only 23% (4/17) of growers used the action threshold for every insecticide application in year 1, but in year 3, 58% (7/12) of growers used the action threshold for every insecticide application.

Growers in Orleans County tended to have the highest, consistent rates of action threshold adoption, whereas growers in Oswego County tended to have the lowest (Table 6.1); however, these differences were not significant (p=0.158) (Table 6.1). Growers increased adoption of thresholds in all counties in years 2 and 3 compared to year 1, except Orange County whose growers only participated in the program for the first two years.

3.2.2 Onion thrips populations. Overall, seasonal mean onion thrips densities were greater in fields that used the action threshold more frequently (Figre 6.2). This relationship was consistent in years 1 and 2, but not year 3 (Supplemental table 6.2). On average, growers who always used the action threshold (100% compliance) had between 3 to 9 times more thrips per leaf as compared with growers who did not use the action threshold (less than 15% compliance) (Figure 6.2). While populations of thrips were higher in fields with greater adoption of the action threshold, all growers successfully controlled onion thrips. Over all three years, 97% (46/47) of the onion fields had mean season densities below the economic threshold of 2.2 thrips per leaf (Fournier et al. 1995) (Figure 6.2).

3.2.3 Insecticide applications. Overall, growers who used the action threshold more often made significantly fewer insecticide applications (Figure 6.3a) ($p=0.00014$, $F_{1,40}=14.81$). This trend occurred consistently in years 2 and 3, but not in year 1 (Supplemental table 6.2). Growers who always used action thresholds (100% compliance) made between one and four fewer insecticide applications per season compared with growers who did not follow the action threshold (less than 15%

compliance) (Figure 6.3a). Overall, most growers (59%, 10/17) reduced the number of insecticide applications in years 2 and 3 as compared with year 1, 29% (5/17) applied the same number of applications and 12% (2/17) increased the number of applications. The total number of products applied throughout the growing season was not significantly related to action threshold use.

3.2.4 Insecticide cost. Insecticide costs decreased with increased use of the action threshold (Figure 6.4). However, the statistical significance of this relationship differed between years (Supplemental table 6.2). Growers who used the action threshold for every insecticide application (100% compliance) saved approximately $148 per hectare as compared with those growers who rarely used the action threshold (less than 15% of their insecticide applications) (p= 0.016, $F_{1,\ 22}$=5.7). The use of inexpensive insecticides was negatively correlated with action threshold use (p= 0.034, $F_{1,40}$ =4.49) (Figure 6.5), suggesting that growers who rarely followed the action threshold were making more applications with inexpensive products. Specifically, greater numbers of applications of lambda-cyhalothrin were negatively associated with action threshold use (p=0.02, $F_{1,40}$=5.31) (Supplemental figure 6.4a). There were no significant relationships between the use of expensive insecticide products and adoption of the action threshold.

3.3. Adoption of insecticide class rotation.

3.3.1 Adoption frequency of insecticide (mode of action) rotation. Over the three-year program, there was a significant increase in the percentage of insecticide applications that successfully rotated insecticide classes (P= 0. 009 $F_{2,\ 41}$=9.35) (Figure 6.1b). Adoption of insecticide class rotation was relatively high across all years but increased

31% from year 1 to year 3. A total of 44 insecticide applications did not comply with proper insecticide rotation recommendations over the three-year program; 29 of the non-compliant applications (66%) included more than two insecticide applications of a given insecticide class. The remaining 34% (15/44) of non-compliant insecticide applications involved an insecticide that was not applied consecutively, thereby exposing more than one onion thrips generation to a given insecticide class.

There were no significant differences between counties and insecticide class rotation (p=0.192); however, rates of adoption differed numerically among years (Table 6.1). In years 1 and 2, at least 60% of growers from all counties adopted the insecticide rotation recommendations and Orleans County growers tended to have the highest levels of adoption (Table 6.1). In year 3, 100% of growers in all counties followed the insecticide rotation recommendation.

3.3.2 Onion thrips populations. Onion thrips populations did not differ based on insecticide class rotation (p=0.546) (Supplemental table 6.2). Numerically, growers who did not rotate insecticide classes appropriately tended to have slightly lower thrips densities than those that consistently rotated between insecticide classes. Overall, growers who properly rotated insecticide classes for every application (100% of insecticide properly rotated) had 0.6 thrips/leaf, whereas the growers with lowest rates of insecticide class rotation (33% of insecticide properly rotated) averaged 0.4 thrips/leaf.

3.3.3 Insecticide applications. Overall, growers who rotated insecticide classes more frequently made significantly fewer insecticide applications (Figure 6.3b) (p=0.00014,

$F_{1,40}$= 14.45). Growers with the lowest levels of insecticide class rotation (33% of insecticide properly rotated) made 1-3 more insecticide applications as compared with those growers who properly rotated every insecticide application. A variety of products were used to control onion thrips populations (Table 6.2). On average, growers applied between 2-4 different insecticide products each season, but some growers used as many as 5 products and others as little as 1 product to control onion thrips. There was no significant relationship between the number of different products used throughout the growing season and insecticide class rotation (p= 0.201). Most growers followed the rotation sequence recommended by the scouts and began their thrips management program with spirotetramat followed in succession by abamectin, co-applications of methomyl and lambda-cyhalothrin or cyantraniliprole, and then spinetoram. Of the 44 insecticide applications that did not comply with the insecticide rotation recommendations, most involved applications of lambda-cyhalothrin. There was a significant negative association between increased lambda-cyhalothrin use and insecticide rotation (p= 0. 001 $F_{1,40}$=10.14), indicating that lambda-cyhalothrin tended to be used more frequently by growers who were less likely to follow the insecticide rotation recommendation (Supplemental figure 6.4b).

3.3.4 Insecticide cost. Insecticide class rotation was not significantly associated with total insecticide cost (p= 0. 215). Regardless of cost, growers created effective season-long sequences of insecticides that successfully rotated classes. While there was no significant relationship between expensive insecticides (>$72/hectare) and use of insecticide rotation, there was a significant negative relationship between the use of inexpensive insecticides (<$24/hectare) and adoption rates of insecticide rotation (p= 0.

008 $F_{1, 40}$=7.03) (Figure 6.5b). The least expensive insecticide applied, lambda-cyhalothrin (at <$7/hectare), was commonly used in a non-compliant manner (Supplemental figure 6.4b).

3.5 Grower opinions of the IRM adoption program

All growers surveyed stated that they followed the insecticide sequences provided by the scouts. Growers typically began their onion thrips management program with spirotetramat and concluded with applications of spinetoram with a variety of other products in between. Growers cited a multitude of reasons for not using the action threshold regularly. However, growers most commonly cited that the risk of forgoing a week without an insecticide application was greater than the price of applying an insecticide, despite the thrips population being below that action threshold (Table 6.3). Secondarily, growers cited that their weekly insecticide program was effective, and therefore did not feel the need to adopt action thresholds. Growers also expressed concern that the action threshold of 1 thrips per leaf was too high and that it didn't adequately accommodate for hot, dry weather conditions. Conversely, those growers who used the action threshold regularly did so because they believed that fewer insecticide applications would slow the onset of insecticide resistance (Table 6.4). Growers also attributed their usage of the action threshold to their individual scouts, as 65% of growers said that they trusted their scout, and therefore were likely to value his or her recommendation.

3.6 Value of IRM adoption program to growers

The majority (94%) of onion growers stated that they benefited from the IRM adoption program. Growers reported making between 0 and 5 fewer insecticide applications, with most replying they made two fewer insecticide applications per year from participating in the program. Most growers responded that the scouting program provided a valuable second opinion to their onion thrips management and onion production. Growers described the scouting program as an educational opportunity that provided them with a better understanding of how to implement the action threshold and effectively rotate insecticides on their farm. Growers appreciated the connection they developed with the scout, and many growers followed recommendations because they trusted their scout (Table 6.4). Growers who participated in the 'IRM adoption program' received all scouting information and recommendations free of cost, but most (94%) stated that they would pay to continue the program. Growers suggested a wide range of prices they would pay to continue the program: between $0 and $123 per hectare per week. Most growers (65%) stated that they would pay $24 per hectare/week for a scout to continue to provide IRM recommendations.

Table 6.1: Thrips density (thrips/leaf), insecticide use and cost, and adoption of insecticide resistance management (IRM) tactics by onion growers in four major onion growing counties in New York over three years.

County	Year participating in program	*n*	Onion thrips density (thrips/leaf)	Number of insecticide applications	Insecticide cost per acre (USD)[a]	Percent (%) of insecticide applications made in accordance to the action threshold	Percent (%) of insecticide applications made in accordance to insecticide rotation restrictions
Orange	1	3	0.7±0.1	5.3±1.2	190±5	68±26	73±20
	2	3	0.6±0.3	6.3±1.3	158±23	48±29	75±16
	3	0	n/a	n/a	n/a	n/a	n/a
Wayne	1	4	0.5±0.3	3.5±0.3	122±13	50±22	85±9
	2	4	0.4±0.1	5±0.4	134±24	57±17	78±16
	3	4	0.4±0.1	2±0.5	65±13	100	100
Orleans	1	5	1.3±0.4	5.6±0.7	162±28	80±20	87±6
	2	5	1.5±0.6	6±0.7	163±24	85±15	94±4
	3	4	0.8±0.1	4.8±1	107±13	96±4	100
Oswego	1	5	0.4±0.1	7.2±0.5	163±15	38±15	60±8
	2	5	0.6±0.3	5.4±1	154±27	65±15	89±5
	3	4	0.1±0.02	4.3±1	109±45	49±12	100

[a] Costs of insecticides were estimated based on prices provided by commercial pesticide dealers in New York from 2015-2017.

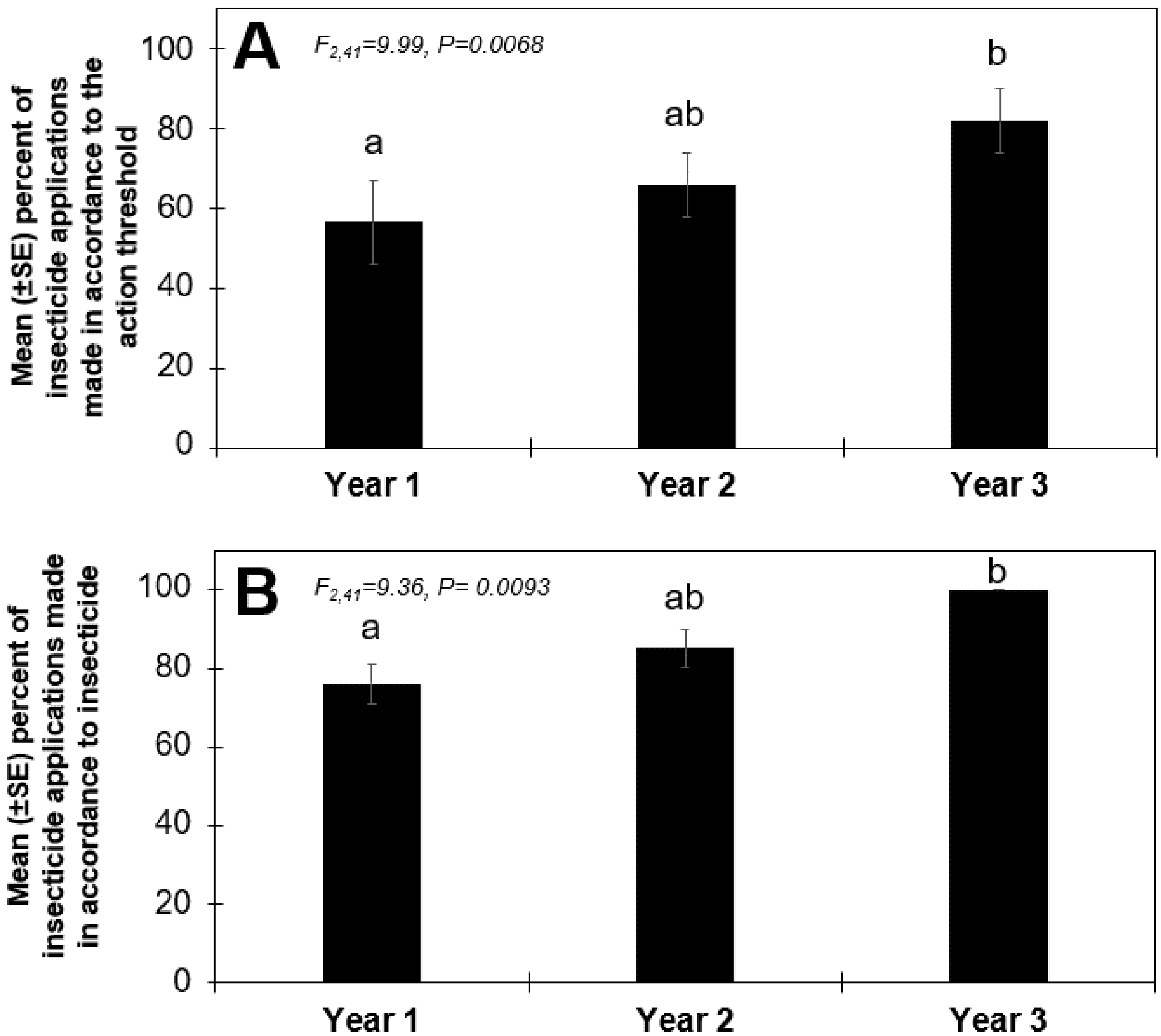

Figure 6.1: Adoption of insecticide resistance management tactics to manage onion thrips for growers who participated in our program over three years in New York. The average percentage of insecticide applications made using the action threshold of one thrips per leaf (A), and the percentage of insecticide applications rotating between insecticide classes (B). The scouting program was voluntary and growers could choose whether or not to follow the scouting recommendation of using the action threshold and rotating between insecticide classes.

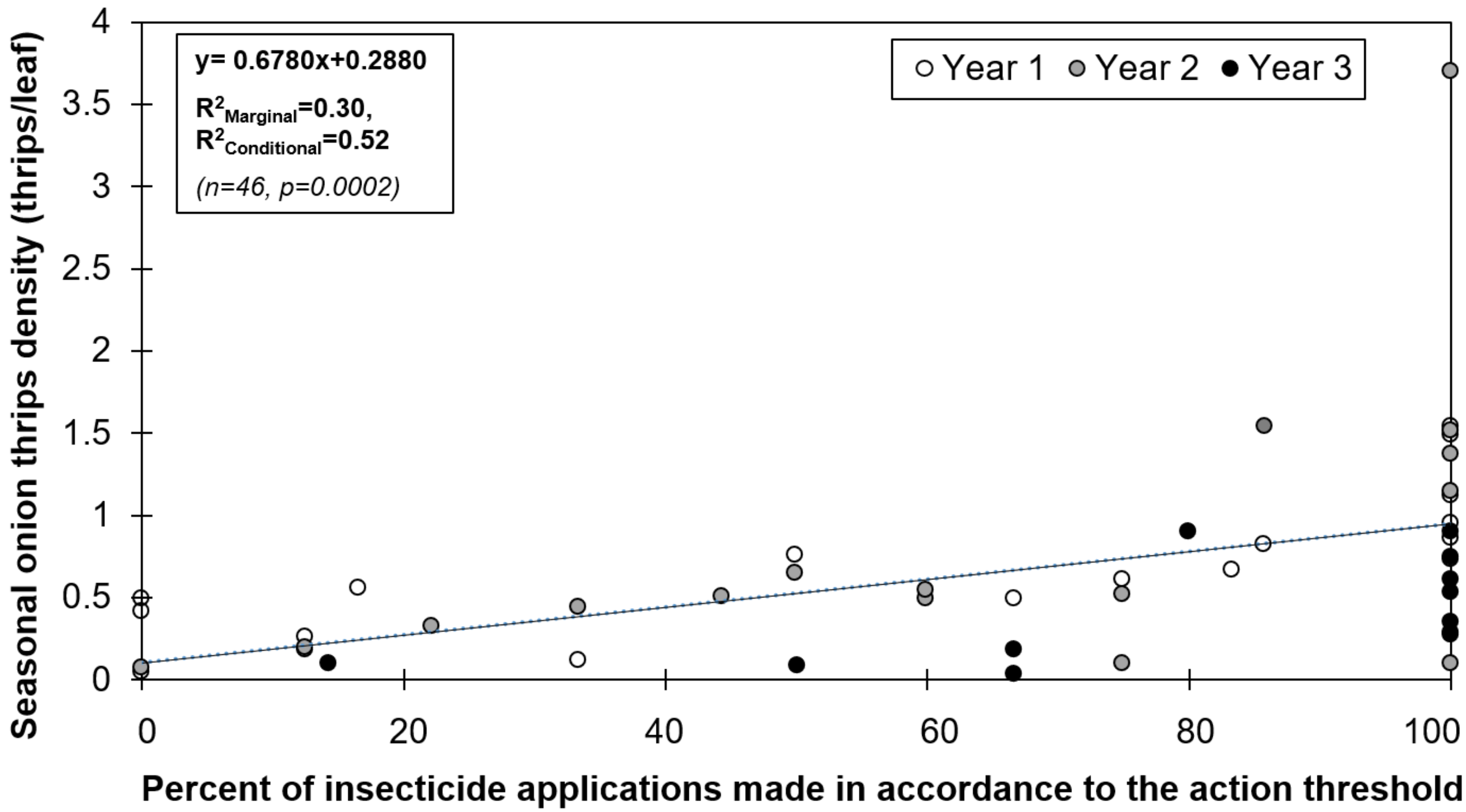

Figure 6.2: Relationship between use of the action threshold (1 thrips/leaf) and seasonal onion thrips densities over the three year period that the IRM adoption program was implemented. Each point represents one onion field that was scouted for thrips and managed by an onion grower in our program. Points with various colors correspond with a particular year.

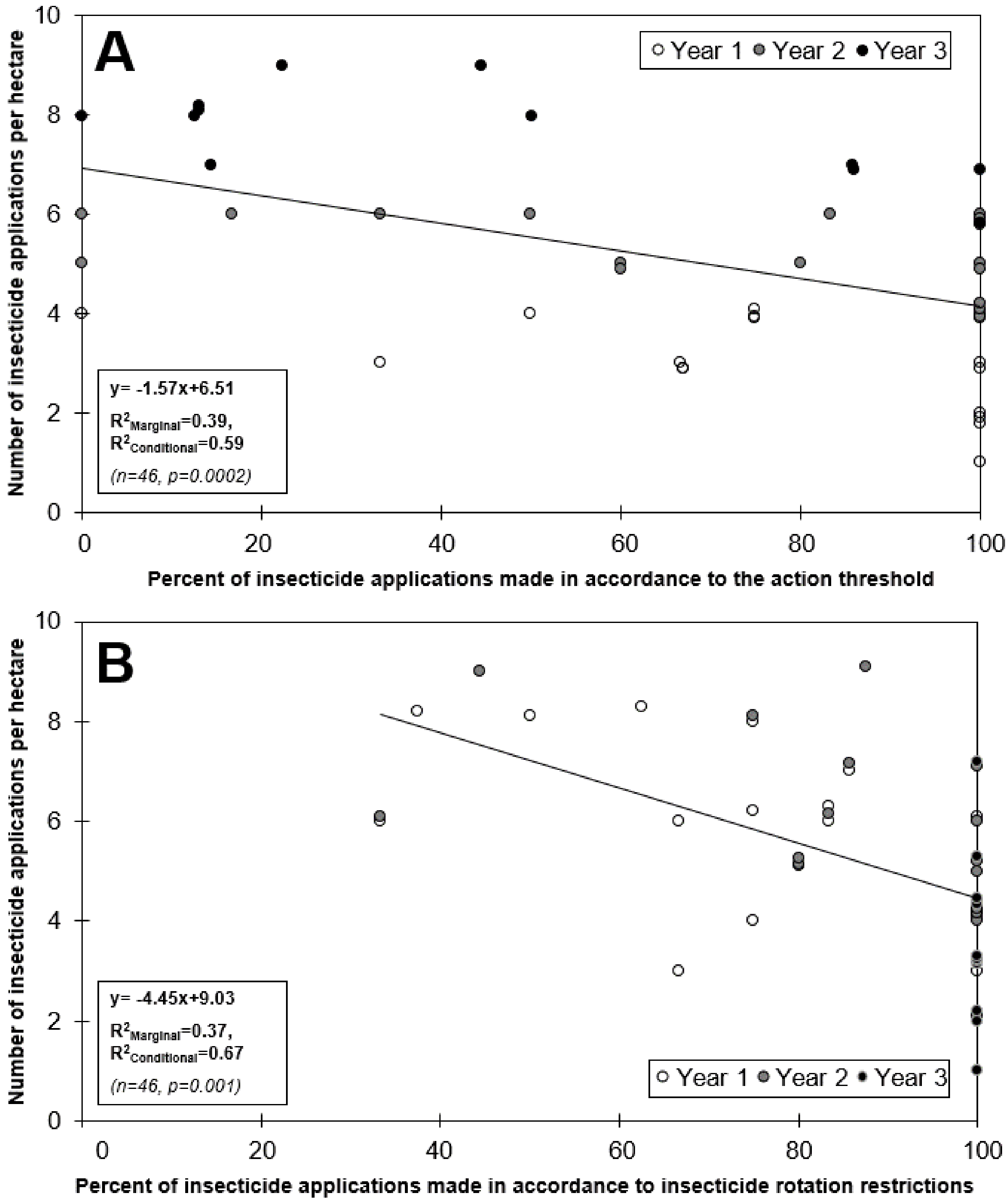

Figure 6.3: Relationship between use of the action threshold (1 thrips/leaf) and number of insecticide applications per acre over the three year period that the IRM adoption program was implemented (A). Relationship between insecticide rotation and number of insecticide application per hectare over the three year period the IRM adoption program was implemented (B). Each point represents one onion field that was scouted for thrips and managed by an onion grower in our program. Points with various colors correspond with a particular year.

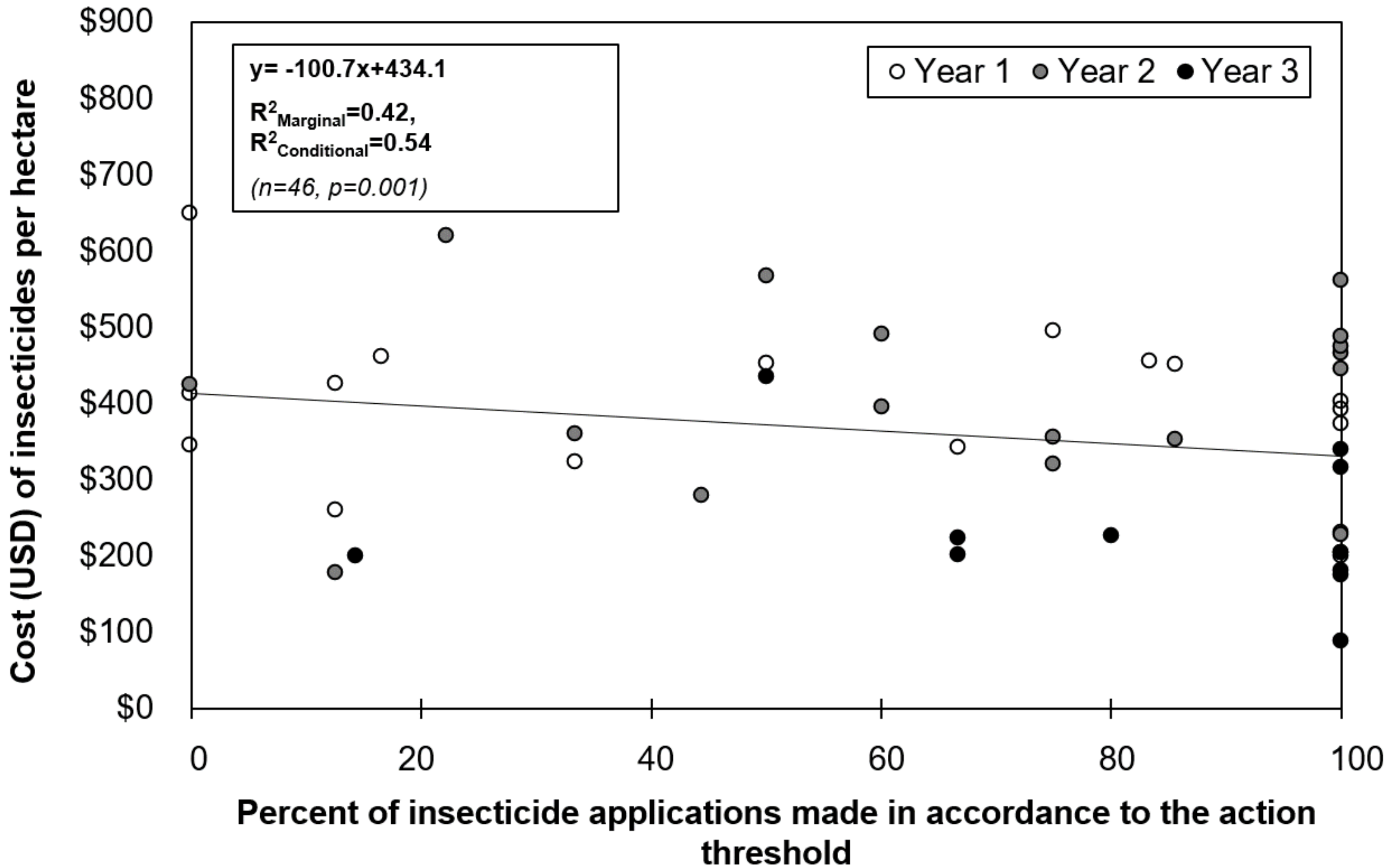

Figure 6.4: Relationship between use of the action threshold (1 thrips/leaf) and total cost of insecticides per acre over the three year period that the IRM adoption program was implemented. Each point represents one onion field that was scouted for thrips and managed by an onion grower in our program. Points with various colors correspond with a particular year.

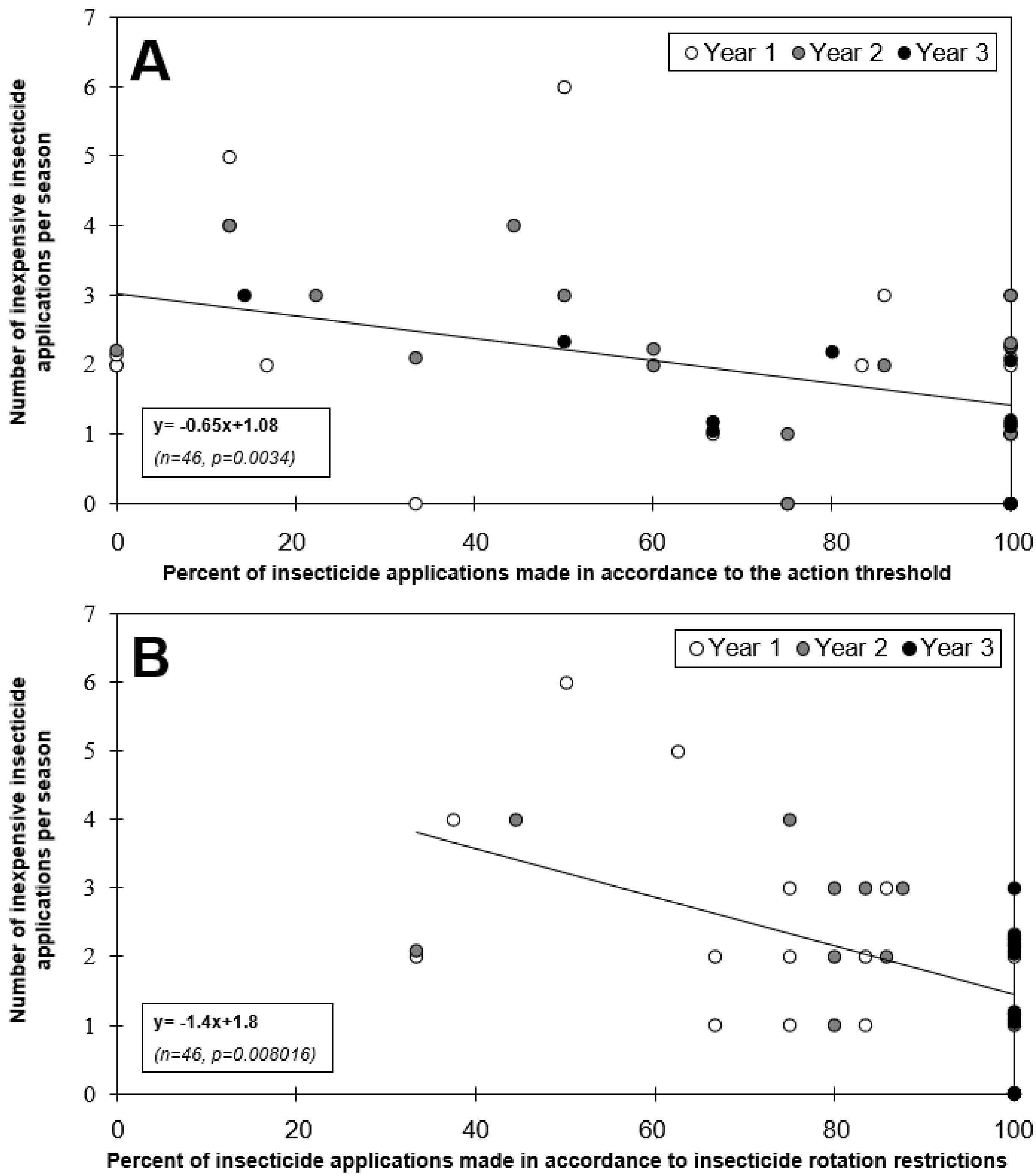

Figure 6.5: Relationship between use of the action threshold (1 thrips/leaf) and total number of inexpensive insecticide applied per acre over the three year period that the IRM adoption program was implemented (A). Relationship between insecticide class rotation and total number of inexpensive insecticide applied per acre over the three year period that the IRM adoption program was implemented (B). Each point represents one onion field that was scouted for thrips and managed by an onion grower in our program. Points with various colors correspond with a particular year.

Table 6.2: Percentage and use of different insecticide classes to control thrips over the three-year IRM adoption program.

Insecticide use	Year	Insecticide classes: Tetronic and Tetramic acid derivatives	Avermectins	Pyrethroids	Carbamates	Diamides	Spinosyns
Total percent applied	1	28.8%	23.1%	17.3%	5.8%	1.9%	23.1%
	2	33.7%	21.8%	14.9%	3.0%	4.0%	22.8%
	3	57.5%	30.0%	2.5%	0.0%	0.0%	10.0%
	All years	35.5%	23.7%	13.9%	3.7%	2.4%	20.8%
Average number applications (±SE)	1	1.8±0.1	1.4±0.2	1.0±0.3	0.3±0.1	0.1±0.1	1.2±0.1
	2	2±0.1	1.4±0.2	0.9±0.2	0.2±0.1	0.2±0.1	1.4±0.2
	3	1.9±0.1	1.0±0.2	0.08±0.008	0±0	0±0	0.3±0.1
	All years	1.9±0.1	1.3±0.1	0.7±01	0.1±0.1	0.1±0.1	1.1±0.1

Table 6.3: Survey results describing why growers who participated in the IRM adoption program used the action threshold to manage onion thrips populations in New York.

Reason grower implemented the action threshold	**Percent of growers** (number responding/total respondents)
• I am concerned about insecticide resistance and want to preserve the useful life of the current insecticides	71% (12/17)
• I trust my Cornell scout and Cornell-based recommendations and value his/her opinion	65% (11/17)
• Using fewer insecticide sprays is less harmful to the environment	47% (8/17)
• The Cornell scout's recommendation to spray or not to spray confirmed what I was going to do anyway	47% (8/17)
• I want to save money on insecticide sprays	33% (5/17)
• Other	12% (2/17)
• Other growers in New York State use the Action-threshold based management program and it has been effective for them	6% (1/17)
• Does not apply-- I never did	6% (1/17)

Table 6.4: Survey results describing why growers who participated in the IRM adoption program did not use the action threshold to manage onion thrips populations in New York.

Reason grower implemented the action threshold	Percent of growers (number responding/total respondents)
• The cost of an insecticide application is less than the risk of the onion thrips population building when I skip an application	59% (10/17)
• My insecticide program is effective, and I did not want to change it	24% (4/17)
• Other	24% (4/17)
• I have had years where I have trouble controlling thrips, and I don't want to experience that again	18% (3/17)
• Does not apply-- I always followed the Cornell scout's recommendations.	12% (2/17)
• I did not have time to consult with a scout or read scouting reports for thrips every week	0% (0/17)
• I trust my chemical company representative recommendations for making insecticide applications more than the Cornell scout's recommendations	0% (0/17)
• I did not trust the Cornell scouting recommendations	0% (0/17)

4. Discussion

Onion growers increased their use of both IRM tactics over the duration of this study. As hypothesized, there were significant increases in the percentage of insecticide applications made following the action threshold (43%) and in the percentage that successfully rotated insecticide classes (31%). No growers reported a yield loss from adopting the IRM tactics and 97% of fields had seasonal mean densities of thrips below the regional economic injury level of 2.2 thrips per leaf (Fournier et al. 1995). Growers who increased usage of the action threshold made 12-50% fewer insecticide applications in year 3 as compared with year 1. Furthermore, growers who regularly used the action threshold saved approximately $148 per hectare as compared with growers who did not use the threshold. Therefore, this extension-based program effectively increased IRM education and practice and provided measured benefits to participating growers. Undoubtedly, sustainability the 'IRM adoption program' will depend on growers who value the program and will make thrips control decisions based on scouting information. Survey data from 2014 revealed that many onion growers (80%) in New York scout or pay for a scouting service and receive weekly information on onion thrips densities (Nault BA, unpublished). Therefore, the resources needed to successfully continue this program are already in place. Nevertheless, ongoing communication between extension educators, crop consultants and growers will be needed to ensure long-term success of this program.

Research on action thresholds and insecticide sequences to manage onion thrips populations in New York has been ongoing for the past three decades (Nault and Huseth 2016; Hoffmann et al. 1995; Shelton et al. 1897). However, results from grower surveys in New York in 2014 indicated a relatively low adoption of either practice, with approximately 40% of growers using an action threshold and 52% rotating between chemical classes (n=45) (Nault BA, unpublished). After one year of working with growers in our study, adoption of both IRM tactics was higher than levels in the 2014 survey. The adoption of a given tactic or innovation depends on many characteristics, including the ability to observe or experiment with an innovation or tactic (Rogers 2003). In this study, we sought to increase the opportunities for growers to experiment with either IRM tactic on a portion of their farm and to observe the success of other growers implementing these IRM tactics through annual meetings. Most growers (94%) stated that they positively benefitted from participating in the program. Growers stated that participation in our program enabled them to better understand when to spray for onion thrips, and what types of products would be most effective. Furthermore, many growers stated that they trusted their scout, and valued their scout's time and communication. Studies have suggested the importance of face-to-face contact in strengthening the relationship between growers and extension educators to increase IPM adoption (Mohammadrezaei and Hayati 2015; Pilcher 2009; Peshin and Karla 2001), and this study further verifies the importance of intensive interactions between growers, researchers and extension educators in increasing the adoption of management practices.

Specifically, onion growers who participated in the 'IRM adoption program' gained experience with new, recently registered insecticides. Prior to 2008 in New York, most insecticides used to manage onion thrips in onion were contact insecticides (e.g. organophosphates, carbamates and pyrethroids), and provided one week of onion thrips control. Since 2008, multiple insecticides have been registered that have either translaminar or systemic activity (e.g. spirotetramat, spinetoram, cyantraniliprole) and greater efficacy against onion thrips compared with older insecticides (Nault and Hessney 2011a; Nault and Hessney 2011b; Nault and Hessney 2008). These new insecticides have residual activity ranging from 5-14 days (Nault et al. 2012) and can offer weeks of onion thrips control in onion. For example, the systemic insecticide spirotetramat can provide 2-3 weeks of onion thrips control after one application. Consequently, growers do not necessarily need to make an insecticide application every week as they needed to in the past. However, the prices of these newer insecticides are approximately 2-4 times more expensive than the older insecticides. Presumably, the higher costs of the newer insecticides inhibited growers from experimenting and regularly applying these newer products. The ‘IRM adoption program’ enabled growers to observe and experiment with these newer, more effective insecticides.

Our study documents further evidence that extension-based programs can significantly impact the actions of growers. Functionally, extension educators are a conduit between growers and researchers and extension’s communication of research findings can be a major factor determining IPM adoption (Pannell 1991; Wearing 1988). Consistently in our study, growers from specific counties tended to manage thrips on their farms similarly, although this was not statistically significant. For example, growers in Orleans County consistently followed the action threshold and adherence to the recommended insecticide sequence and rotation restrictions in all years of the program. Research and extension conducted by Cornell Cooperative Extension educators and Cornell entomologists have had a strong presence in Orleans County over the past decade, and growers and Cornell personnel frequently and openly communicate (i.e. weekly meetings between growers and Cornell extension). Conversely, we observed that Oswego County onion growers, who had much lower levels of extension and research involvement on their farms, had the lowest initial level of adoption of either IRM tactic. Our case study showed that the installment of greater extension resources and communication with growers led to fewer insecticides being applied to manage onion thrips. In year 1 in Oswego County, only 38% of applications made by growers followed the action threshold, but approximately 57% of the applications followed the action threshold in years 2 and 3.

Interestingly, our study identified a potential synergy between the two IRM practices implemented. Significant reductions in insecticide applications were recorded with increased use of the action threshold and insecticide class rotation. Specifically, those growers with fewer insecticide applications were more likely to successfully rotate between insecticide classes. On average, use of an action threshold reduced the number of insecticide applications in most agricultural production systems when compared with a standard (or weekly) insecticide program (Leach et al. 2017; Nault and Huseth 2016; Hoffmann et al. 1995). Fewer insecticide applications present fewer opportunities for growers to incorrectly rotate insecticide products. Therefore, use of an action threshold may facilitate insecticide class rotation. This finding highlights the potential importance of fully evaluating IRM programs such that returns can be maximized to the grower and the onset of insecticide resistance is slowed.

The use of inexpensive insecticides may be a significant barrier in IRM adoption. Consistently, increased numbers of inexpensive insecticide applications were negatively associated with percentage adoption of either the action threshold or insecticide rotation. Interestingly, the use of inexpensive lambda-cyhalothrin was also negatively associated with proper insecticide rotation. Many onion thrips populations in New York are resistant to lambda-cyhalothrin (Shelton et al. 2006; Shelton et al. 2003); however, some growers still apply this insecticide with hopes to reduce thrips infestations. Inexpensive insecticides, regardless if they are effective or not (as is the case with lambda-cyhalothrin), are unlikely to incentivize the adoption of IRM tactics, especially in high-value commodities. The perception of risk imposed by the insect pest will often supersede recommendations from an action threshold (Pannell 1991). The cost of pesticides has been implicated as a potential barrier to the adoption of resistance management practices in other systems as well (Hurley and Frisvold 2016; Forrester 1990). Thus, IRM programs should dissuade growers from repeatedly applying inexpensive insecticides because overuse may result in insecticide resistance.

Adoption of IRM and associated IPM practices can be challenging in high-value commodities, where losses in yield can be economically devastating (Farrar et al. 2016). In our study, the primary reason growers declined using the action threshold was, “the insecticide price was lower than risk of the thrips population building [and not being controllable in the future]”. In many cases, growers also mentioned that they had experienced “bad years” in which they had great difficulty managing thrips, and thus were more averse to the risk of skipping a weekly insecticide application routine. Additionally, growers responded that the cost savings generated by using an action threshold was not perceived as a large benefit, as only 33% of growers indicated that reducing their insecticide bill was a reason for adopting the action threshold. Because onion is a high-value crop, the cost savings of eliminating an insecticide application is marginal. For example, assuming an average value of $16/cwt with an average yield of 864 cwt/hectare would amount to a gross revenue of $13,824 (USDA NASS 2015). Therefore, even a 1% loss in yield would amount to a loss of $138, which is similar to the average cost of insecticide savings we have demonstrated in this study ($148/hectare). The economic incentives of using an action threshold to determine pest control decisions in a high-value crop are less compelling than benefits like slowing the onset of insecticide resistance by making fewer applications. The primary reason onion growers in our study cited for following the action threshold was to slow the onset of insecticide resistance and thereby preserve the efficacy of currently labeled insecticides. Therefore, New York onion growers appeared to be responsive to adopting IRM tactics that are predicated largely on IRM principles, which is consistent with other studies[20]. Therefore, this study further verifies the need for IRM and related programs to appeal to resistance management rather than economics for high-value commodity farmers.

5 Conclusion

The 'IRM adoption program' successfully increased grower education of insecticide resistance management tactics. As a result, participating growers substantially increased usage of both the action threshold and rotation of insecticide classes, which reduced numbers of insecticide applications and saved them money. However, since the ‘IRM adoption program’ was free to the grower, long-term impacts and sustainability of this effective program will depend on the complexity of the IRM tactics and returned benefits to the grower. Action thresholds are notoriously difficult to implement, as they can be complicated and time consuming to the grower or practitioner (Peshin and Karla 2009). Scouting incurs a cost to growers, either through their time spent scouting or paying for a scouting service, which can further limit the economic incentive of implementing an action threshold. Therefore, further innovation and technology is needed to address this issue to ensure that growers can implement these tactics in a timely and affordable manner.

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CONCLUSIONS AND FUTURE RESEARCH QUESTIONS

Integrated pest management (IPM) is the primary paradigm to manage pests within agricultural production systems (Radcliffe et al. 2009). Most IPM programs are designed to manage a single pest, but most, if not all, productions systems face challenges from *multiple* pests. In order for IPM to provide the greatest service to agriculture, multiple pests and management tactics should be considered simultaneously. Kogan (1998) suggests that IPM progress is inherently connected to the scale in which we evaluate and implement IPM tactics. Therefore, the most progressive and effective IPM would theoretically occur when we manage multiple agricultural pests at a landscape level (Kogan 1998). While this approach may be logistically prohibitive, IPM programs should attempt to maximize the scale in which IPM practitioners can implement management tactics.

In onion production, there are multiple pests that limit marketable yield including onion thrips, bacterial bulb rots and iris yellow spot disease (Schwartz and Mohan 2008; Gill et al. 2015). Several IPM tactics have been suggested to manage these pests, including the use of different onion cultivars, fertility regimes, and insecticide programs.

In IPM trials (chapters 1 and 2), we consistently observed that onion thrips densities were impacted by an onion thrips-resistant cultivar and action-threshold based insecticide program, but not fertility regime. In our studies, thrips densities were unaffected by nitrogen and phosphorus fertilizer amendments; however, plant growth was also unresponsive to fertilizer amendments in most trials (5/6 trials). Previous literature has shown that thrips increase with increasing rates of fertilizer, and researchers have posited that thrips may be responding to increased onion plant vigor

(Malik et al. 2009; Buckland et al. 2013). The effect of plant growth metrics on onion thrips populations is currently understudied, but may have significant implications on onion thrips management. Further research should identify those plant characteristics associated with thrips colonization including: Does increased size of a specific plant part (e.g. length of leaves, width of onion pseduostem, number of leaves) predict thrips colonization and ovipositional preference? Does ovipositional preference predict larval survival on these plant parts? Does soil fertility influence epicuticular wax development in onion, and is it possible that increased fertility increases thrips attraction to certain onion cultivars?

Recent research has suggested that onion thrips significantly contribute to the incidence of bacterial bulb rot, primarily those rots caused by *Pantoea spp.* (Dutta et al. 2014; Grode et al. 2016; Grode et al. 2019); however, our study did not observe increased levels of bacterial rot with increased thrips densities. Further research should specifically address this relationship, as continuous speculation regarding the contribution of thrips to the development of bacterial bulb rot may cause growers to unnecessarily increase insecticide use. Future research questions might include: Does thrips control (using different insecticide programs) impact the incidence of bacterial bulb decay caused by *Pantoea spp.*? Does the effect of thrips control significantly differ based on causal bacterial species? And, if there is a relationship between onion thrips and bacterial bulb rot, what is the mechanism by which thrips increase the incidence of bacterial rot (increased leaf dieback and/or transmission of bacteria through frass)? Should we modify current action thresholds (i.e. 1 thrips per leaf) to successfully manage both thrips and bacterial bulb rot?

In our IPM studies, we found that tactics to manage onion thrips positively and negatively influenced thrips-associated plant diseases. For example, in chapter 1, we found that thrips control reduced IYS disease, as insecticide use reduced the incidence and severity of IYS disease. However, in chapter 2, a thrips-resistant onion cultivar (‘Avalon’) had lower thrips densities but greater levels of bacterial rot. These studies highlight the importance of holistically evaluating IPM programs, as management tactics can influence multiple pests within a production system. Peterson et al. (2018) recently criticized the approach and utility of modern IPM and claimed the practice may have, “lost its way”. Nevertheless, the authors suggest that IPM can be improved by “recommitting to Kogan’s levels of IPM”. Our findings corroborate the importance of this approach, as we found that IPM tactics influenced pests contrary to our initial hypotheses. Thus, IPM programs should consider the concerted effect of multiple tactics on multiple pests within an agricultural production system. Whenever possible, future studies evaluating IPM programs should address other influential abiotic and biotic components within agricultural production systems.

In order to effectively develop IPM tactics for agricultural pests, the pest biology and ecology must be fully understood. Iris yellow spot virus (IYSV), transmitted principally by onion thrips, is a significant tospovirus which causes iris yellow spot disease in onion. While research efforts to manage IYSV in onion production have been ongoing since the virus’s discovery (Gent et al. 2006), a number of questions regarding the relationship between thrips and IYSV remain. In our studies, we examined the relative contribution of three habitats to provide IYSV inoculum. Transplanted onion fields had the greatest percentage (50%) of viruliferous onion thrips, and thus we postulated that these fields may provide the greatest amount of inoculum for late-season outbreaks of IYS disease. This study raises interesting questions regarding the relationship between landscape ecology and plant viruses like IYSV. A considerable amount of research has been devoted to understanding how fungal plant pathogens are influenced by the landscape (Plantegenest et al. 2007); however, there are fewer studies applying this context to plant viruses. As the landscape changes and agricultural land intensifies production, the effects of landscape composition should be considered for plant virus management programs. Further questions regarding IYSV and the landscape include; Will modifying the orientation of transplanted onion fields significantly impact the incidence of IYS disease (as compared to current planting orientations that do not consider the impact of transplanted onion fields contributing to IYSV inoculum)? Given the heterogeneity of weedy areas and low occurrence of IYSV plant hosts, do weedy areas significantly increase or decrease overwintering viruliferous thrips populations? Furthermore, how does the surrounding habitat composition impact adult thrips dispersal and subsequent IYSV outbreaks?

Many tospoviruses have been shown to impact their thrips vector, and multiple studies have demonstrated that thrips positively benefit from tospovirus infection (DeAngelis et al. 1993, Stumpf and Kennedy 2005, Stafford et al. 2011, Shrestha et al. 2012, Stafford-Banks et al. 2014). In laboratory trials, we found that viruliferous thrips lived 3-4 days longer than non-viruliferous thrips. This was an interesting finding as previous work had indicated that onion thrips are unaffected by IYSV infection (Inoue et al. 2010, Birthia et al. 2013). Future research should continue to define the effects of IYSV on onion thrips populations. Do viruliferous thrips have different developmental timing as compared to non-viruliferous thrips? Furthermore, what is the effect of IYSV on thrips feeding behaviors? Does IYSV infection increase fitness of the vector, and would increased fitness confer a competitive advantage in interspecific interactions?

Lastly, in chapter 5, we examined the effect of an extension-based program to increase grower adoption of two management tactics (insecticide class rotation and action threshold). We found that growers increased use of both management tactics by 31-44% over the three years the program was implemented. Furthermore, growers who increased use of the action threshold successfully managed onion thrips densities but applied fewer insecticides and spent less on insecticide cost. Importantly, this study acknowledges that the success of Insecticide resistance management (IRM) and IPM does not rely on the researcher developing IRM/IPM tactics, but on the grower, who *chooses* to implement IRM/IPM. Gould et al. (2018) recently described the development of pesticide resistance as a, “sociobiological dilemma” in which social factors including grower adoption and coordination of management programs are tantamount to the success of insecticide resistance management. Research should continue to address the methods that successfully increase grower adoption of IPM and IRM tactics. Future research questions include: What is the current level of IRM/IPM adoption by growers within the United States? Does the value of the crop influence levels of IRM adoption? Does grower education correspond to grower adoption (e.g. if a grower understands a tactic, is he/she likely to implement that tactic)? Are there other sociological considerations that better explain when and why a grower adopts a management tactic? Do extension-based programs sustain long-term grower adoption of IRM/IPM practices?

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Appendix

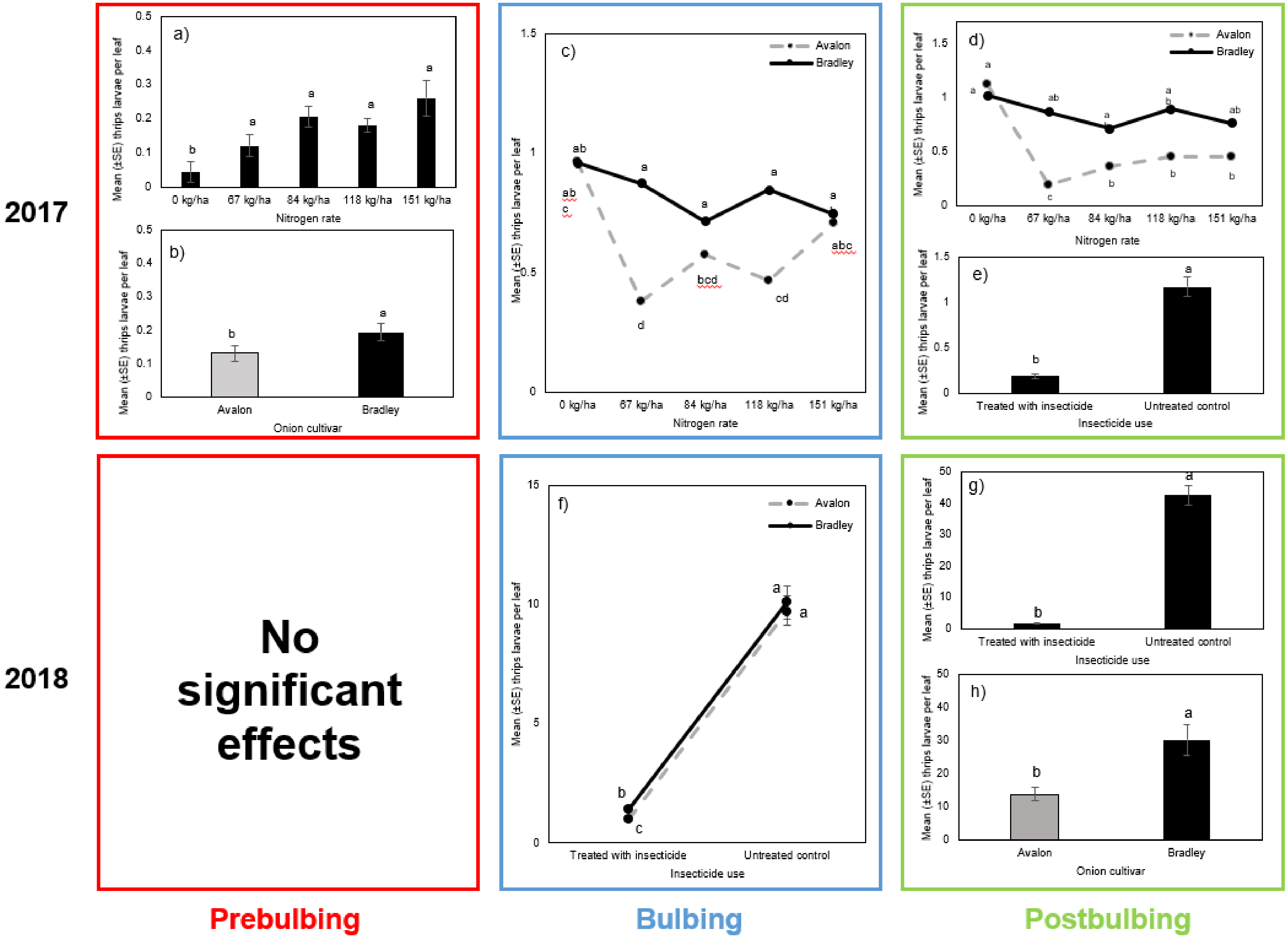
I. SUPPLEMENTAL FIGURES

II. SUPPLEMENTAL TABLES

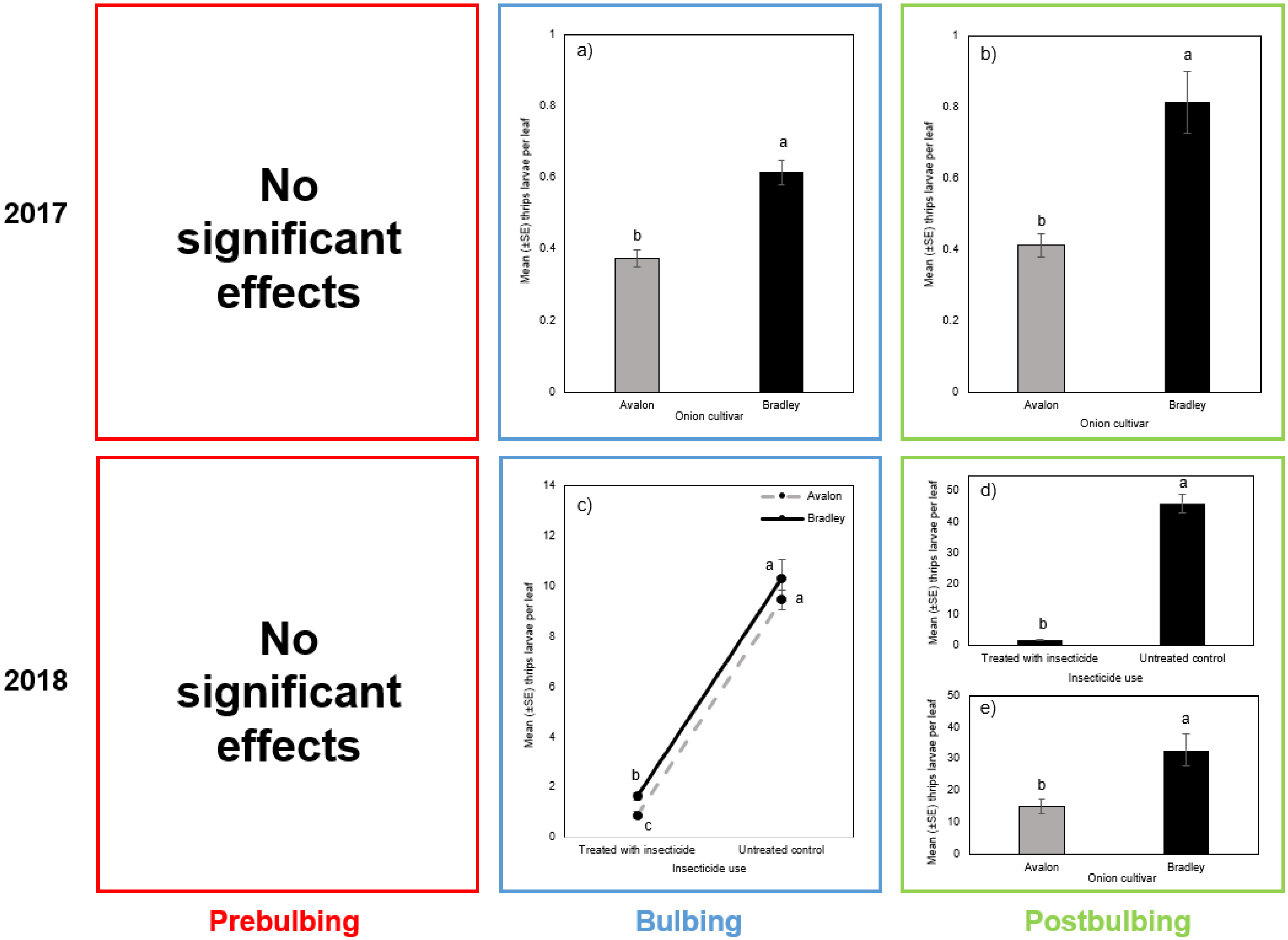
III. RELATIONSHIP BETWEEN ONION THRIPS (THRIPS TABACI) AND STEMPHYLIUM VESICARIUM IN THE DEVELOPMENT OF STEMPHYLIUM LEAF BLIGHT IN ONION

Appendix I

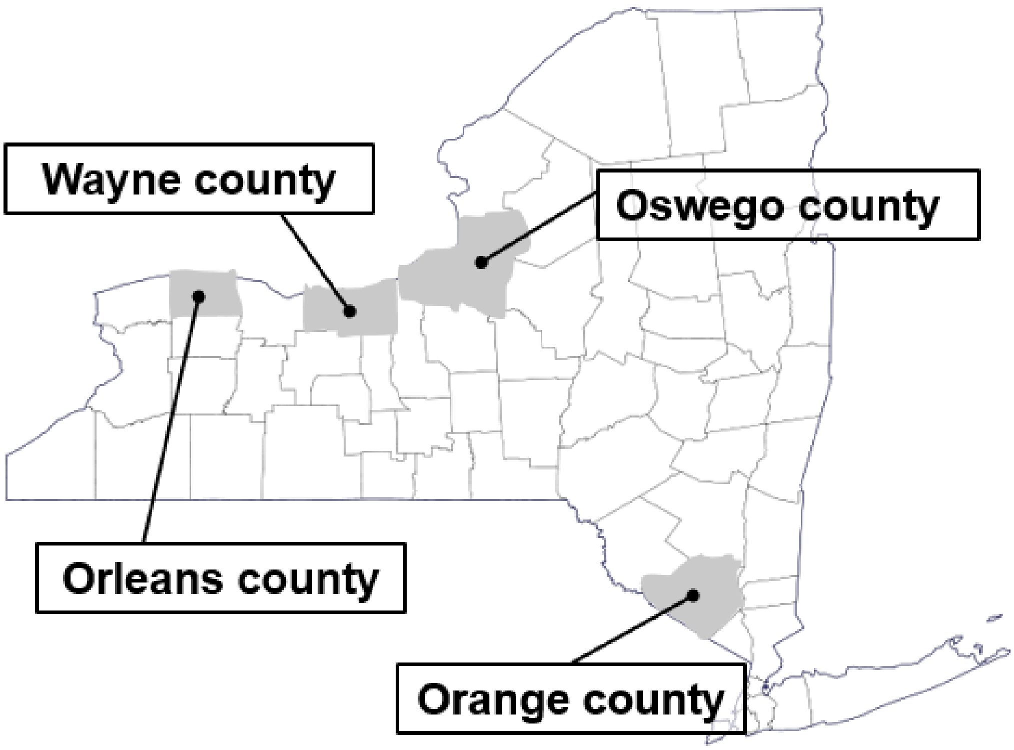
SUPPLEMENTAL FIGURES



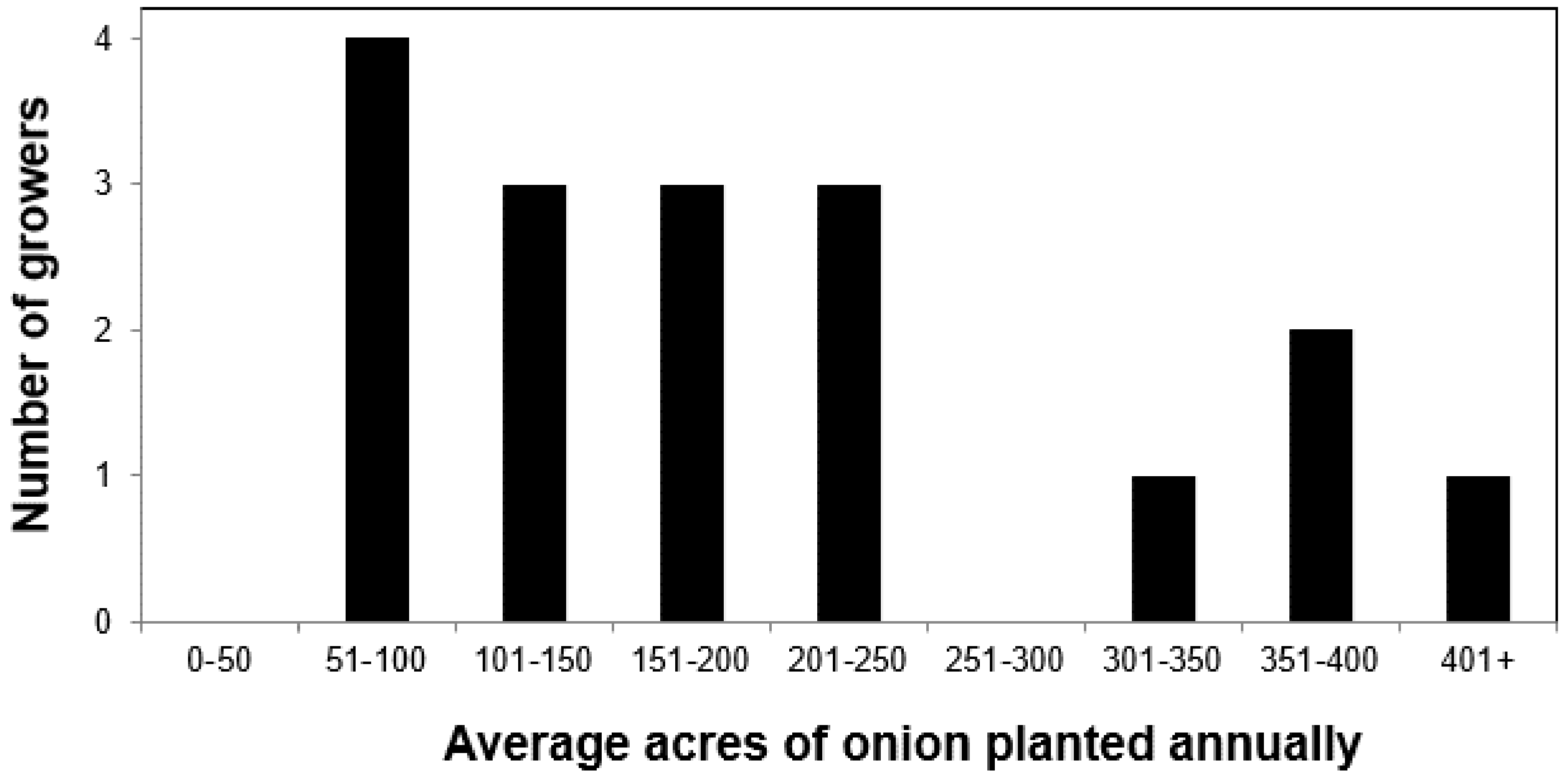
Supplemental figure 3.1: Significant effects of different treatments combining nitrogen fertilizer, onion cultivar, and insecticide use on larval onion thrips densities during the three developmental stages of onions: Prebulbing (a, b), bulbing (c, f), and postbulbing (d, e, g, h) in 2017 and 2018. No treatments significantly affected onion thrips densities during the prebulbing stage in 2018. Five different rates of nitrogen were applied through the growing season; 0 kg ha-1 (no nitrogen applied during the growing season), 67 kg ha-1 (67 kg ha-1 applied at planting), 84 (67 kg ha-1 applied at planting and 17 kg ha-1 applied pre-bulbing), 118 (67 kg ha-1 applied at planting and 51 kg ha-1 applied pre-bulbing), and 151 kg ha-1 (67 kg ha-1 applied at planting and 84 kg ha-1 applied pre-bulbing). Nitrogen rates were paired with two onion cultivars and two insecticide treatments. 'Avalon' and 'Bradley' differ in thrips susceptibility, and 'Avalon' is moderately thrips resistant 'Bradley' is susceptible to thrips feeding. Every nitrogen and onion cultivar combination was either treated with insecticide or left untreated. When plots exceeded the action threshold, treatments were sprayed with an insecticide. Onion thrips densities exceeded the action threshold in 1 out of 10 weeks of sampling in 2017 and 5 out of 8 weeks in 2018. Onion thrips were sampled weekly beginning in early June and concluding in mid-August.



Supplemental figure 3.2: Significant effects of different treatments combining different onion cultivars and insecticide use on larval onion thrips densities during developmental stages of onions: bulbing (a, c), and postbulbing (b, d, e) in 2017 and 2018. Phosphorus treatments did not significantly impact onion thrips densities during any developmental stage in either year. No treatments significantly affected onion thrips densities during the prebulbing stage in 2017 or 2018. 'Avalon' and 'Bradley' differ in thrips susceptibility, 'Avalon' is moderately thrips resistant 'Bradley' is susceptible to thrips feeding. Plots were treated with insecticide when thrips densities exceeded the action threshold (1 thrips/leaf). Onion thrips densities exceeded the action threshold in 1 out of 10 weeks of sampling in 'Bradley' and 0 out of 10 weeks in 'Avalon' in 2017. In 2018, the action threshold was exceeded 5 out of 8 weeks in 2018. Onion thrips were sampled weekly beginning in early June and concluding in mid-August in each year.



Supplemental figure 6.1: Statewide distribution of onion growers who participated in the onion thrips IRM management program in 2015, 2016, and 2017 in New York. The counties included Orleans, Wayne, Orange, and Oswego. In 2015, 16 growers participated in the program: 5 in Orleans County, 4 in Oswego County, 3 in Orange County, and 4 in Wayne County. In 2016, 2 additional growers participated in the program and the totals per county were as follows: 6 in Orleans County, 5 in Oswego County, 3 in Orange County, and 4 in Wayne County. In 2017, 14 growers participated in the program: 5 in Orleans County, 5 in Oswego County, and 4 in Wayne County; none from Orange County



Supplemental figure 6.2: Mean acreage of onion planted annually by growers who participated in the scouting program in 2015, 2016, and/or 2017.

Question 1- Describe your overall ability to control onion thrips in 2017 (please check one):

___Excellent
___Very Good
___Average
___Poor
___Failed

Question 2- Describe your overall ability to control onion thrips in 2016 (please check one):

___Excellent
___Very Good
___Average
___Poor
___Failed

Question 3- Describe your overall ability to control onion thrips in 2015 (please check one):

___Excellent
___Very Good
___Average
___Poor
___Failed

Question 4- Do you think your yield was reduced as a result from using either the action threshold or insecticide class rotation (rotating between insecticide classes)?

___Yes
___No
___I don't know

Question 5- As compared to two years ago, how often do you use the action threshold to determine your insecticide applications? Please check one.

___I use action thresholds MORE
___ My use of action thresholds HAS NOT CHANGED
___I use action thresholds LESS

Question 6- How often did you follow the Cornell scout's recommendations to spray or not spray for thrips? Check one.

___Always
___Most of the time
___About half of the time
___Sometimes
___Never

Question 7- Why did you follow the Cornell scout's recommendations? Check all that apply.

___I never did
___To save money on insecticide sprays
___I am concerned about insecticide resistance and want to preserve the useful life of the current insecticides
___Other growers in New York State use the Cornell Onion Thrips Management program and it has been effective for them
___I trust my Cornell scout and Cornell-based recommendations and value his/her opinion
___Using fewer insecticide sprays is less harmful to the environment
___The Cornell scout's recommendation to spray or not to spray confirmed what I was going to do anyway
___Other, please specify: _______________

Question 8- If you did not always follow the Cornell scout's recommendations, what were your reasons? Check all that apply.

___Does not apply – I always followed the Cornell scout's recommendations.
___The cost of an insecticide application is less than the risk of the onion thrips population building when I skip an application
___My insecticide program works great, and I did not want to change it
___I experienced a bad year trying to control onion thrips, and I didn't want that to happen again.
___I did not have time to consult with a scout or read scouting reports for thrips every week
___I trust my chemical company representative recommendations for making insecticide applications more than the Cornell scout's recommendations
___I did not trust the Cornell scouting recommendations
___Other, please specify: ______________________

Question 9- By participating in the onion thrips scouting program, how many insecticide sprays per season did you save?

In 2015, I saved ________ insecticide sprays per season
In 2016, I saved ________ insecticide sprays per season
In 2017, I saved ________ insecticide sprays per season

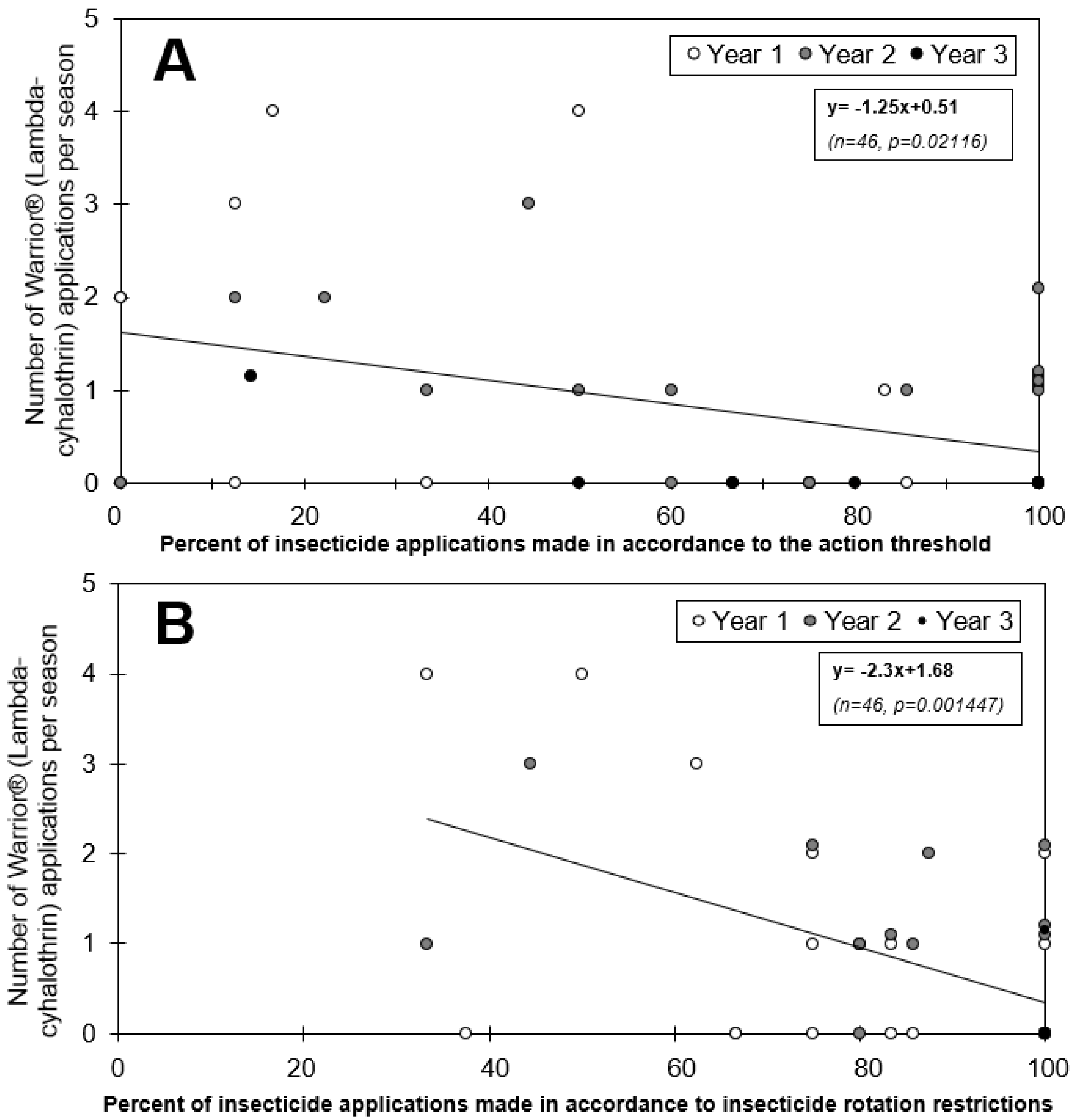
Question 10- Implementing the "IRM adoption program" requires scouting information (thrips counts per field). How much would you pay to get the scouting information?
$________ per acre

Question 11- Did you benefit from participating in this Project?

___No
___Yes
If yes, how so?

Question 12- What was the most important thing that you learned from participating in the Cornell Onion Thrips Management Program?

Supplemental figure 6.3: Survey was completed by all growers who participated in the 'IRM adoption program' to evaluate growers' opinions and perceptions of the program.



Supplemental figure 6.4: Relationship between use of the action threshold and total number of Warrior® (Lambda-cyhalothrin) applications applied per acre over the three year period that the IRM adoption program was implemented (A). Relationship between insecticide class rotation and total number of Warrior® (Lambda-cyhalothrin) applications applied per acre over the three year period that the IRM adoption program was implemented (B). Each dot represents one grower field per year.

Appendix II

SUPPLEMENTAL TABLES

Supplemental table 2.1: Mean percent of bulbs with bacterial rot at harvest for onion cultivars varying in susceptibility to onion thrips that received various combinations of nitrogen fertilizer at planting and insecticide treatments for managing onion thrips. Studies were conducted near Elba, NY in 2015 and 2016. Insecticide applications were made weekly in the standard program and only when thrips densities ≥1 larva/leaf in the action threshold-based program. Means within the same cultivar and year that share the same letter are not significantly different (P>0.05; LSmeans).

Cultivar	Treatment		Mean % (± SE) bacterial incidence	
	Insecticide program	Nitrogen rate (kg ha^{-1})	2015	2016
Avalon	Untreated control	67 kg	7.0 ± 3.1 c	0 ± 0
		101 kg	12.4 ±2.3 b	1.7 ± 0.9
		140 kg	6.8 ± 1.3 c	0.4 ± 0.3
	Action threshold	67 kg	5.3 ± 0.8 c	1.2 ± 0.5
		101 kg	8.8 ± 1.9 c	0.7 ± 0.6
		140 kg	6.13 ± 1.9 c	1.2 ± 0.8
	Standard	67 kg	8.2 ± 2.8 c	0.8 ± 0.5
		101 kg	8.3 ± 2.1 c	0.2 ± 0.2
		140 kg	19.3 ± 7.8 a	0.3 ± 0.2
Delgado	Untreated control	67 kg	0 ± 0	0 ± 0
		101 kg	0 ± 0	0 ± 0
		140 kg	1.7 ± 1.6	0 ± 0
	Action threshold	67 kg	0 ± 0	0 ± 0
		101 kg	0 ± 0	0 ± 0
		140 kg	0.5 ± 0.2	0 ± 0
	Standard	67 kg	0.3 ± 0.3	0 ± 0
		101 kg	0.8 ± 0.7	0 ± 0
		140 kg	0 ± 0	0 ± 0

Bradley	Untreated control	67 kg	1.7 ± 1.2	0 ± 0
		101 kg	0.4 ± 0.3	0 ± 0
		140 kg	1.4 ± 1.2	0 ± 0
	Action threshold	67 kg	0.5 ± 0.32	0 ± 0
		101 kg	0.2 ± 0.2	0 ± 0
		140 kg	0.7 ± 0.5	0 ± 0
	Standard	67 kg	0 ± 0	0 ± 0
		101 kg	0 ± 0	0 ± 0
		140 kg	0 ± 0	0 ± 0

Supplemental table 2.2: Mean percent of bulbs with bacterial rot after three months in storage for onion cultivars varying in susceptibility to onion thrips that received various combinations of nitrogen fertilizer at planting and insecticide treatments for managing onion thrips. Studies were conducted near Elba, NY in 2015 and 2016. Insecticide applications were made weekly in the standard program and only when thrips densities ≥1 larva/leaf in the action threshold-based program. Means within the same cultivar and year that share the same letter are not significantly different (P>0.05; LSmeans).

Cultivar	Treatment		Mean % (± SE) bacterial incidence	
	Insecticide program	Nitrogen rate (kg ha^{-1})	2015	2016
Avalon	Untreated control	67 kg	11.5 ± 4.1	9.0 ± 3.8 abc
		101 kg	13.2 ± 3.9	11.2 ± 3.3 a
		140 kg	10.9 ± 1.4	7.3 ± 1.6 bc
	Action threshold	67 kg	11.4 ± 3.0	11.4 ± 3.2 a
		101 kg	11.6 ± 5.8	9.8 ± 2.0 ab
		140 kg	21.2 ± 4.6	11.4 ± 3.0 a
	Standard	67 kg	13.2 ± 2.0	6.6 ± 1.5 bcd
		101 kg	13.3 ± 2.7	4.3 ± 2.1 d
		140 kg	12.3 ± 1.4	6.5 ± 1.6 cd
Delgado	Untreated control	67 kg	4.9 ± 2.1 c	4.5 ± 2.5 a
		101 kg	3.9 ± 2.9 c	3.4 ± 0.7 a
		140 kg	16.2 ± 8.0 a	2.2 ± 1.0 ab
	Action threshold	67 kg	8.5 ± 2.2 abc	2.4 ± 1.1 ab
		101 kg	9.4 ± 3.0 ab	0.8 ± 0.5 b
		140 kg	4.3 ± 1.6 c	2.3 ± 1.1 ab
	Standard	67 kg	4.1 ± 1.4 c	0.8 ± 0.8 b
		101 kg	6.4 ± 1.2 bc	3.1 ± 0.9 a
		140 kg	5.1 ± 3.9 c	1.2 ± 0.5 b

Bradley	Untreated control	67 kg	5.0 ± 2.7 a	1.5 ± 0.9 b
		101 kg	1.9 ± 0.9 b-e	2.1 ± 1.1 ab
		140 kg	4.5 ± 1.9 abc	1.1 ± 0.5 b
	Action threshold	67 kg	2.6 ± 2.2 a-e	0.7 ± 0.3 b
		101 kg	1.4 ± 0.9 cde	1.7 ± 1.1 b
		140 kg	0.7 ± 0.4 e	1.8 ± 0.8 b
	Standard	67 kg	1.2 ± 0.8 de	1.5 ± 0.5 b
		101 kg	3.4 ± 1.5 a-d	0.6 ± 0.3 b
		140 kg	4.9 ± 0.8 ab	4.0 ± 3.1 a

Supplemental table 3.1: Weather conditions in 2017 and 2018 near Elba, NY.

Year	Month	Average temperature (c)	Max temperature (c)	Min temperature (c)	Total precipitation (cm)
2017	May	13.1	28.3	0.6	16.1
	June	14.6	30.0	7.8	5.6
	July	21.0	28.9	10.6	11.9
	August	19.9	29.4	7.2	8
2018	May	17.6	31.7	8.0	6.9
	June	19.1	31.7	7.8	7.1
	July	23.2	33.9	12.8	5.3
	August	22.6	33.4	11.7	9.1

Supplemental table 3.2: Plant growth traits and soil nitrate levels in plots receiving different rates of nitrogen in 2017 and 2018. Rates and timings were 0 kg ha-1 (no nitrogen applied), 67 kg ha-1 (67 kg ha-1 applied at planting), 84 kg ha-1 (split into two applications, 67 kg ha-1 applied at planting and 17 kg ha-1 applied pre-bulbing), 118 kg ha-1 (split into 67 kg ha-1 applied at planting and 51 kg ha-1 applied pre-bulbing), and 151 kg ha-1 (split into 67 kg ha-1 applied at planting and 84 kg ha-1 applied pre-bulbing).Soil nitrate levels were tested at three different phenological time points: pre-bulbing (June), bulbing (July), and post-bulbing (August). Every plot was sampled, for a total of five replicates. Means within a column with same letters are not significantly different (P>0.05; LSmeans). † indicates near significance, 0.05-0.1.

			Plant growth metric			
Year	Developmental stage	Nitrogen treatment	Length of leaves (cm)	Plant weight (g)	Number of leaves	Soil nitrate (ppm)
2017	Prebulbing	0 kg/ha	24.6 ± 4.3 b	3.5 ± 1.9 b	3.1 ± 0.3	11.2 ± 0.7 b
		67 kg/ha	36.3 ± 1.8 a	8.9 ± 0.9 a	4.0 ± 0.1	31.7 ± 3.5 a
	Bulbing	0 kg/ha	55.3 ± 1.5 b	41.6 ± 3.6 b	6.2 ± 0.2	8 ± 1.1 c
		67 kg/ha	70.4 ± 1.4 a	88.4 ± 4.9 a	7.8 ± 0.2	9.6 ± 1.8 bc
		84 kg/ha	69.9 ± 1.4 a	83.2 ± 5.9 a	7.5 ± 0.2	10.1 ± 1.9 bc
		118 kg/ha	72.2 ± 1.3 a	93.2 ± 4.2 a	7.8 ± 0.2	21.5 ± 4.8 ab
		151 kg/ha	73.0 ± 1.3 a	91.6 ± 4.5 a	7.7 ± 0.1	37.3 ± 6.5 a
	Postbulbing	0 kg/ha	72.3 ± 1.8 a	124.7 ± 8.9 a	7.8 ± 0.2	8.5 ± 1.1 c
		67 kg/ha	85.4 ± 1.1 a	233.2 ± 7.4 b	8.9 ± 0.2	12.8 ± 2.2 bc
		84 kg/ha	82.1 ± 1.4 a	206.1 ± 8.0 b	8.7 ± 0.2	13.2 ± 1.7 bc
		118 kg/ha	87.1 ± 1.2 a	219.2 ± 10.6 b	8.7 ± 0.2	19.1 ± 3.5 b
		151 kg/ha	83.8 ± 1.3 a	221.7 ± 9.0 b	9.1 ± 0.1	36.4 ± 5.9 a
2018	Prebulbing	0 kg/ha	13.7 ± 1.4 b	28.7 ± 2.5 b	4.9 ± 0.5	41.9 ± 0.8 b
		67 kg/ha	18.3 ± 0.7 a	39.5 ± 4.1 a	5.5 ± 0.6	46.4 ± 1.4 a
	Bulbing	0 kg/ha	55.8 ± 5.6	102.9 ± 0.9	7.5 ± 0.2	16.9 ± 2.6 e
		67 kg/ha	58.9 ± 5.4	110.8 ± 0.9	7.4 ± 0.2	29.1 ± 5.6 d
		84 kg/ha	56.8 ± 4.6	102.1 ± 0.9	7.6 ± 0.2	39.3 ± 5.1 c

	118 kg/ha	56.5 ± 5.4	97.1 ± 1.2	7.1 ± 0.2	60.1 ± 4.1 b
	151 kg/ha	57.5 ± 5.5	109.1 ± 0.9	7.8 ± 0.2	114.1 ± 15.1 a
Postbulbing	0 kg/ha	38.0 ± 1.5 †	164.5 ± 5.9	7.2 ± 0.2	12.9 ± 0.5 c
	67 kg/ha	28.3 ± 1.7 †	176.4 ± 6.4	7.1 ± 0.2	17.5 ± 1.5 c
	84 kg/ha	40.6 ± 1.3 †	174.6 ± 6.8	7.1 ± 0.2	18.9 ± 2.1 b
	118 kg/ha	33.9 ± 1.7 †	185.6 ± 29.35	7.1 ± 0.1	27.8 ± 2.4 ab
	151 kg/ha	37.4 ± 1.6 †	172.1 ± 8	7.3 ± 0.2	32.8 ± 3.3 a

Supplemental table 3.3. Plant growth traits and soil phosphorous levels in plots receiving different rates of phosphorous in 2017 and 2018. Four different rates of phosphorus were at planting 0, 56, 112, and 168 kg ha-1. Soil phosphorus levels were tested at three different phenological time points: pre-bulbing (June), bulbing (July), and post-bulbing (August). Every plot was sampled, for a total of five replicates. No metrics were statistically different between phosphorus treatments (*P*>0.05; LSmeans). † indicates near significance, 0.05-0.1.

Year	Developmental stage	Phosphorus treatment	Length of leaves (cm)	Plant weight (g)	Number of leaves	Soil phosphorus (lb./ac)
2017	Prebulbing	0 kg/ha	38.0±2.2	9.9±1.3	4.1±0.1	66.8 ± 4.2 b
		56 kg/ha	37.0±1.8	9.9±0.9	4.1±0.1	75.9 ± 5.7 b
		112 kg/ha	38.5±1.8	11.3±1.4	4.2±0.1	107.5 ± 8.1 a
		168 kg/ha	37.5±1.7	10.2±1.2	4.1±0.2	109.3 ± 6.3 a
	Bulbing	0 kg/ha	74.4±2.2	99.9±9.2	7.9±0.02	59.4 ± 6.2 b
		56 kg/ha	74.9±2.6	100.8±6.9	7.9±0.02	88.6 ± 8.4 ab
		112 kg/ha	75.3±1.8	106.0±8.3	8.1±0.02	87.8 ± 7.8 a
		168 kg/ha	76.5±2.6	112.5±9.1	8.3±0.02	110.7 ± 6.3 a
	Postbulbing	0 kg/ha	83.6±1.3	200.0±7.8	8.7±0.2	50.6 ± 5.8 b
		56 kg/ha	82.2±1.2	220.2±7.5	9.1±0.2	67.8 ± 6.1 ab
		112 kg/ha	82.8±1.1	201.6±11.3	8.9±0.2	81.2 ± 7.3 a
		168 kg/ha	84.7±1.3	217.8±10.7	8.7±0.2	92 ± 5.7 a
2018	Prebulbing	0 kg/ha	45.8 ± 1.1 †	16.8 ± 1.2	5.3 ± 0.1	52 ± 4.2 b
		56 kg/ha	48.5 ± 1.5 †	19.5 ± 1.1	5.5 ± 0.1	52 ± 3.2 ab
		112 kg/ha	46.1 ± 1.1 †	18.2 ± 1.2	5.3 ± 0.1	64.6 ± 7.2 a
		168 kg/ha	48.1 ± 1.2 †	19.2 ± 1.0	5.5 ± 0.1	81.7 ± 8.9 a
	Bulbing	0 kg/ha	57.9 ± 5.5	114.4 ± 0.9	7.4 ± 0.2 †	46.9 ± 4.6 c
		56 kg/ha	61.3 ± 5.2	115.1 ± 0.9	7.9 ± 0.2 †	61.6 ± 6.4 c
		112 kg/ha	58.8 ± 4.6	108.4 ± 1.1	7.4 ± 0.1 †	90.6 ± 13.5 b
		168 kg/ha	59.9 ± 6.6	121.4 ± 1.1	7.7 ± 0.2 †	124.2 ± 21.2 a
	Postbulbing	0 kg/ha	31.7 ± 1.1	176.9 ± 11.5	7.2 ± 0.1	32.9 ± 3.2 b

	56 kg/ha	32.8 ± 1.1	182.1 ± 10.1	7.1 ± 0.1	33.8 ± 3.2 ab
	112 kg/ha	29.3 ± 1.1	181.8 ± 9.8	7.3 ± 0.1	44.8 ± 6.3 a
	168 kg/ha	30.9 ± 1.1	181.4 ± 7.1	7.4 ± 0.1	47.7 ± 8.2 a

Supplemental table 6.1: Baseline survey data from participating onion growers prior to the initiation of the IRM adoption program.

Management tactic	Percent of growers who use the tactic (number responding/total respondents)
• I use integrated pest management on my farm to manage onion thrips populations (e.g., use multiple chemical, cultural, biological, and/or physical approaches)	88% (15/17)
• I use cultural management tactics to control onion thrips (e.g., reducing nitrogen rates, planting less thrips-susceptible onion cultivars, or removing volunteer onions)	76% (13/17)
• I use physical management tactics to control onion thrips (e.g. physical barriers)	0% (0/17)
• I use biological control management tactics to control onion thrips (e.g. use of natural enemies and/or release of natural enemies)	0% (0/17)
• I scout my field for onion thrips, or pay for a scouting/consulting service	88% (15/17)
• I use action thresholds to manage onion thrips populations.	65% (11/17)
• I rotate insecticide classes, and only make two sequential applications of one mode of action before rotating to a new insecticide	94% (16/17)

Supplemental table 6.2: Relationships between the adoption of action threshold on seasonal onion thrips densities, total number of insecticide applications, and total insecticide costs in each year the program was implemented.

Fixed effect	Variable	Year	Marginal R^2 value	Conditional R^2 value	Chi-square	P-value
Adoption of action threshold	Onion thrips densities	1	0.54	0.69	9.3	0.002283
		2	0.26	0.28	6.0	0.01415
		3	0.0002	0.90	0.0023	ns, 0.9244
	Number of Insecticide applications	1	0.09	0.63	2.3	ns, 0.1292
		2	0.23	0.27	4.9	0.02654
		3	0.59	0.94	34.1	p<0.001
	Total insecticide cost	1	0.08	0.31	1.8	ns, 0.189
		2	0.15	0.23	3.2	ns, 0.07556
		3	0.52	0.91	19.2	p<0.001
Adoption of insecticide class rotation	Onion thrips densities	1	0.007	0.37	0.14	ns, 0.7073
		2	0.008	0.13	0.15	ns, 0.6982
		3	n/a	n/a	n/a	n/a
	Number of Insecticide applications	1	0.15	0.61	4.72	0.02974
		2	0.22	0.23	4.54	0.03298
		3	n/a	n/a	n/a	n/a
	Total insecticide cost	1	0.04	0.15	0.83	ns, 0.3616
		2	0.07	0.17	1.31	ns, 0.2519
		3	n/a	n/a	n/a	n/a

Appendix III

RELATIONSHIP BETWEEN ONION THRIPS (*THRIPS TABACI*) AND *STEMPHYLIUM VESICARIUM* IN THE DEVELOPMENT OF STEMPHYLIUM LEAF BLIGHT IN ONION

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Abstract

Stemphylium leaf blight caused by Stemphylium vesicarium and onion thrips (Thrips tabaci) are two common causes of leaf damage in onion production. Onion thrips is known to interact synergistically with pathogens to exacerbate plant disease. However, the potential relationship between onion thrips and Stemphylium leaf blight is unknown. In a series of controlled laboratory and field trials, the relationship between thrips feeding and movement on the development and severity of Stemphylium leaf blight disease were examined. In laboratory assays, onions (cvs. 'Avalon' and 'Ailsa Craig') with varying levels of thrips feeding damage were inoculated with S. vesicarium. Pathogen colonization and leaf dieback were measured after two weeks. In pathogen transfer assays, thrips were exposed to S. vesicarium conidia, transferred to onion and leaf disease development was monitored. In field trials, insecticide use was examined as a potential indirect means to reduce Stemphylium leaf blight disease and pathogen colonization by reducing thrips damage. Results from laboratory trials revealed that a reduction in thrips feeding decreased S. vesicarium colonization of onion leaves, and decreased leaf dieback. Additionally, onion thrips were capable of transferring S. vesicarium conidia to onion plants (albeit at a low frequency 2-14% of plants inoculated). In field trials, the symptoms and colonization of Stemphylium leaf blight were reduced with the use of insecticide to control thrips. These results suggest that onion thrips may play a significant role in the development of Stemphylium leaf blight, and increased thrips control may reduce disease in commercial onion fields.

Key words: *Allium*, *Pleospora allii*, thrips movement, plant pathogen-insect interaction

1. Introduction

Stemphylium leaf blight (SLB), caused by *Stemphylium vesicarium* (E.G. Simmons), is an important disease of onion worldwide. Symptoms of the disease in onion include excessive leaf dieback (necrotic leaf tissue) and water-soaked lesions along leaves, which reduce size and quality of onion bulbs (Shishkoff and Lorbeer; 1989; Basallote-Ureba et al. 1999). In Portugal, SLB was shown to reduce bulb yield up to 85% (Tomaz and Lima 1986). In New York, USA, onions protected from SLB with fungicides had bulbs that were 33-40% larger than those in the untreated control (Hoepting 2018a, 2018b), and control of SLB increased weight of Jumbo grade bulbs (> 7 cm in diameter) by 29% (Hoepting 2018b).

Stemphylium leaf blight is most severe and prevalent in warm, humid growing conditions, which are favorable for the development of the pathogen (Basallote-Ureba et al., 1999; Prados-ligero et al. 2003). The pathogen has an asexual stage (*Stemphylium vesicarium*), which produces large numbers of conidia on leaves during the season, and a sexual stage, *Pleospora allii*, which produces ascospores in pseudothecia. Ascospore release from pseudothecia plays an important role in the epidemiology of SLB in other countries (Misawa and Yasuko 2012). Although pseudothecia are produced on onion leaves in New York, ascospore production within these structures has yet to be confirmed. *S. vesicarium* can invade through dead tissue or live tissue with appressoria (Aveling and Snyman 1993).

Stemphylium leaf blight impacts the physiological maturity of the plant, inhibits natural development, and increases the likelihood of bacterial bulb decay (Wright et al.1993;

Hoepting 2016). A previous study reported that plants dying prematurely from SLB were twice as likely to have bacterial bulb decay as compared to plants protected from the disease and senesced naturally (Hoepting 2016). In New York, the disease has become especially problematic in recent years because *S. vesicarium* has developed resistance to several commonly used fungicide active ingredients such as azoxystrobin, boscalid, and cyprodinil (Hay et al. 2018). As an emerging disease of prominence in New York, research efforts have concentrated on the epidemiology of SLB and control practices to reduce its impact on onion production. Management of SLB requires a comprehensive understanding of how the pathogen interacts with its abiotic and biotic environment, including how the pathogen and the disease are affected by herbivorous insect pests in the onion production system.

Thrips have been associated with a variety of plant diseases, and their capacity to transmit or spread significant plant pathogens often rivals their role as herbivores (Ullman et al. 1997). While thrips are recognized for their role as major vectors of tospoviruses, thrips also are associated with spreading fungal plant pathogens. Yarwood (1943) reported that powdery mildew (*Erysiphe necator*) infection on grape leaves increased with thrips densities and speculated that thrips may transmit powdery mildews to an array of crops including grape, strawberry, cantaloupe, clover, and rose. Western flower thrips (*Frankliniella occidentalis*) abundance was associated with fusarium ear rot (caused by *Fusarium verticillioides*) in corn and insecticide use reduced thrips populations and the severity of ear rot disease (Farrar and Davis 1991). Similarly, Mailhot et al. (2007) found that flower thrips (*Frankliniella spp*.) abundance was consistently associated with *Fusarium verticillioides*, and insecticide use decreased severity of hardlock in cotton. In addition to feeding damage, thrips also may transfer fungal plant pathogens within and between plants. For example, *Thrips obscuratus* has been identified as a potential vector of *Botrytis cinerea* in kiwifruit, and up to 17% of adults were found to be naturally contaminated with the pathogen (Fermaud and Gaunt, 1995). Marullo (1995) identified viable conidia from multiple species on the body surface and gut of adult thrips. Thrips species have been identified as potential vectors of fungal pathogens, suggesting, thrips feeding and their movement on plants may impact the severity and success of fungal plant pathogens.

Onion thrips (*Thrips tabaci* Lindeman) is a primary insect pest of onion and feeds directly on onion leaf tissue. Onion thrips infestations have been associated with the development of purple blotch disease of onion, which is caused by *Alternaria porri* (Bhangale and Joi 1983; McKenzie et al. 1993). McKenzie et al. (1993) found that onion plants infested with thrips had more severe purple blotch disease, with greater amounts of leaf dieback and lesion lengths, than those not infested with thrips. Additionally, thrips feeding injury provided an alternate entrance into the onion leaf. Without thrips injury, *A. porri* entered through the stomates, but when damaged by thrips, *A. porri* also entered through wounds created by the feeding damage.

Infestations of onion thrips and infection of *S. vesicarium* often co-occur in onion fields, with thrips damage preceding the onset of SLB. Furthermore, sporulating lesions are commonly found during peak onion thrips infestations in New York. Thrips may be transferring this pathogen within and among neighboring onion plants. *S. vesicarium* may also take advantage of thrips feeding damage to invade onion leaf tissue; however, this relationship is currently unstudied. The purpose of this project was to describe the contribution that onion thrips has on the epidemiology of SLB in onion. In a series of laboratory and field experiments, the relationship between onion thrips movement and feeding on *S. vesicarium* infection were investigated. We hypothesized that thrips feeding and movement would increase the success of *S. vesicarium* infection of onion leaves, and that thrips feeding on leaves would increase the leaf area colonized by *S. vesicarium*. We also surmised that thrips would passively transfer *S. vesicarium* conidia and infect healthy onion plants. Moreover, we hypothesized that relationships identified in our laboratory studies would be consistent with those in commercial onion field trials, and that insecticides used to reduce thrips feeding damage would reduce the severity of Stemphylium leaf blight disease in onion.

2. Materials and Methods

Thrips feeding damage impact on *S. vesicarium* incidence in laboratory trials

Assay design and preparation. To test if thrips feeding damage promoted the development of SLB, onion plants were infested with varying levels of thrips densities and inoculated with conidia of *S. vesicarium*. Onion plants (cvs. 'Avalon' and 'Ailsa Craig') were seeded and grown in a greenhouse free from *S. vesicarium* and onion thrips. All onions were seeded into 72-cell round propagation trays (TO Plastics INC. item #59-5010) with superfine germination mix (Pro-mix item #20-200400) and then transplanted at the 2-leaf stage into pots (7.6 cm diameter × 31 cm tall) containing Cornell potting mix (peat, perlite and vermiculite in a 4:1:1 ratio). When onions produced 4-5 leaves, plants were infested with differing numbers of thrips adults. Thrips were reared in the laboratory on cabbage, which is not a host of *S. vesicarium* (Köhl et al.

2009). Plants were either infested with high levels of thrips (25-30 thrips per plant), low levels of thrips (2-3 thrips per plant), or no thrips. Thrips-infested onion plants were placed into thrips-proof cages ('1462W' bug dorm, Bio Quip, Rancho Dominguez, CA) for 7 days to establish varying levels of feeding damage. Plants with 'high thrips feeding damage' had damage on all leaves covering ≥60% of the total leaf area, whereas plants with 'low thrips feeding damage' had damage covering approximately 10-20% of the total leaf area. Plants with 'no feeding damage' had no thrips feeding on any leaves.

Independent and identical studies were conducted for each cultivar and studies were repeated three times (= trial). For each cultivar study, the experiment was designed as a 3 x 2 factorial in which thrips feeding had three levels ('no feeding damage', 'low feeding damage', and 'high feeding damage') and inoculum had two levels (inoculated with *S. vesicarium* and untreated). For each trial, 30 plants were selected for each thrips feeding level and half (15 plants) were inoculated with a conidial suspension containing *S. vesicarium* and the remaining half (15 plants) were untreated. Therefore, for each cultivar study, there was a total of 270 plants (90 plants per trial x 3 trials).

S. vesicarium inoculation. *S. vesicarium* cultures were initiated from diseased onions from a commercial field in Elba, NY. Cultures were grown on V8 agar in petri plates for 4 weeks under 12 h fluorescent light/12 h dark. Plates were flooded with approximately 15 ml of sterile water and a surfactant (Tween 20 (1 drop Tween 20/100 ml water)) and conidia were scraped into suspension with a spatula. The suspension was stirred on a magnetic stirrer for 20 min to detach conidia from conidiophores and sieved through a 150 µm sieve to remove large pieces of mycelium. The resulting conidial suspension was counted using a haemocytometer and adjusted to 400 ml to give a mean conidial count of 19,000/ml. The conidial suspension was sprayed directly onto onion plants using a spray bottle (~8 ml/plant) and each plant was incubated in a plastic bag (Uline poly bags item# s-7498) for 48 hr to maintain leaf wetness for germination and infection. Untreated control plants were sprayed with sterile water and Tween 20 (1 drop Tween 20/100 ml water) and incubated singly in the bags for 48 hours. After 48 hours, all plants were removed from bags and placed into controlled growth chambers (25° C, 50% RH, 16:8 L:D photoperiod).

S. vesicarium disease assessments. Plants were visually assessed for SLB disease symptoms (leaf dieback and lesion presence) 14 d after *Stemphylium* inoculation. The amount of dead tissue was measured (cm) and compared with the amount of green tissue (cm) on every leaf/plant to estimate the percent of leaf dieback. The presence and number of lesions were recorded. Additionally, one leaf was randomly excised from each plant, and placed into a gallon-sized plastic bag containing a wet paper towel. After incubating for 7 days, leaves were observed under a dissecting microscope (× 40) for the presence of *Stemphylium vesicarium* conidia and *Pleospora allii* pseudothecia. The total length of leaf exhibiting sporulation by *S. vesicarium* was marked on the bag, measured with a ruler and expressed as a percent of total leaf length.

Thrips transfer of *S. vesicarium* conidia in laboratory trials

To test if thrips could physically transfer *S. vesicarium* conidia, thrips adults were exposed to conidia of *S. vesicarium* and then transferred to healthy onion plants. Onion thrips were obtained from a colony reared on cabbage. Thrips adults were placed onto

V8 agar in petri dishes containing colonies of sporulating *S. vesicarium* for 30 mins (same isolate as used in the previous experiment). While in the petri dish, onion thrips were observed using a dissecting scope and Zeiss Stemi 508 Stereo Microscope (Carl Zeiss Microscopy, Thornwood, NY) to determine if conidia were attached to the thrips exterior body. After 30 min, thrips were individually placed into 10µl tubes, and each thrips was transferred to a thrips-proof cage containing one healthy onion plant (cv. 'Avalon'). Onions were grown and maintained as described above). To reduce the chance of inadvertently contaminating the plant with conidia, the 10µl tube containing the thrips was inverted until the thrips independently exited the tube onto the plant. Negative controls containing thrips exposed to only V8 agar in a petri plate were transferred onto healthy onion plants in thrips-proof cages in the same manner. After two weeks, all plants were removed, incubated for 48 hours in gallon-sized plastic bags containing a moist paper towel, and examined under a dissecting microscope to determine if *S. vesicarium* colonized the tissue. The experiment consisted of two treatments, thrips exposed to *S. vesicarium* and thrips exposed to only V8 agar, and each treatment was replicated 50 times. The experiment was repeated 4 times for a total of 200 plants per treatment.

Relationship between thrips feeding damage and Stemphylium leaf blight in field trials

Experimental design and applications. To determine if reducing thrips damage would reduce Stemphylium leaf blight disease, an experiment was conducted in a commercial onion field in 2017 (cv. 'Fortress') and 2018 (cv. 'Pocono'). The experiment was designed as a 2 x 2 factorial experiment, with 2 levels of fungicide use (no fungicide, fungicide) and 2 levels of insecticide use (no insecticide, insecticide). The four treatments were arranged in a randomized complete block design with each treatment replicated 4 times. Experimental plots were 1.5 m x 6 m and consisted of five rows of onions. Treatments that included insecticide use were treated with spinetoram at 0.07 kg (AI) ha^{-1} (Radiant SC; Corteva AgriSciences, Indianapolis, IN), and treatments that included fungicide were treated with fluopyram/pyrimethanil (0.24 kg (AI) ha-1/0.73 kg (AI) ha-1 respectively) (Luna Tranquility; Bayer CropScience, Research Triangle park, NC). Insecticide and fungicide products were chosen based on their superior control of thrips and Stemphylium leaf blight, respectively (Reiners et al. 2019). Pesticides were applied weekly with a CO_2-pressurized backpack sprayer with four, twin flat-fan nozzles (TJ-60-8003VS; TeeJet Technologies Harrisburg, PA). Pesticides were co-applied with an adjuvant at 0.5% v:v (Induce; Helena, Collierville, TN) to increase efficacy (Nault et al. 2013). Trials in 2017 and 2018 were initiated on 2 Aug 2017 and 18 July 2018 respectively, and concluded on 22 Aug 2017 and 15 Aug 2018, respectively. Trials were initiated when onions were bulbing (6-7 leaves per plant) and concluded when onions naturally senesced (total of 4 and 6 sprays in 2017 and 2018, respectively).

Trial set up and maintenance. Fields were planted using a vacuum seed planter with 650,000 onion seeds per hectare on 28 Apr 2017 and 16 Apr 2018. Seeds were treated with FarMore FI500 (mefenoxam [0.15 g ai/kg of seed], fludioxonil [0.025 g ai/kg of seed], azoxystrobin [0.025 g ai/kg of seed], spinosad [0.2 mg ai/seed] and thiamethoxam [0.2 mg ai/seed]) and Pro-Gro (carboxin [7.5 g ai/kg of seed] and thiram [12.5 g ai/kg of seed]) to improve plant establishment by protecting seedlings from maggots (*Delia* spp.) and seedling diseases. Prior to the initiation of the field experiment, fungicide applications were applied to control botrytis (*Botrytis squamosa*);

however, the products used for botrytis control are known not to significantly impact Stemphylium leaf blight disease and they were applied 4 weeks earlier. There were no other insect pests or plant pathogens that damaged the onions over the duration of the experiment in either year. Weeds were managed according to Cornell vegetable management guidelines and recommendations (Reiners et al. 2019).

Visual Stemphylium leaf blight disease assessments. Ten plants per plot were randomly chosen and assessed for the number of onion thrips larvae, as well as common symptoms of SLB including, leaf dieback and number of water-soaked lesions. All leaves on the onion plant were assessed, and leaf dieback was categorized as either “full dieback” where the entire leaf was necrotic, “partial dieback” in which < 50% of the leaf was necrotic, and “no dieback” in which none of the leaf area was necrotic. Any lesions characteristic of Stemphylium leaf blight (Basallotte-Ureba et al. 1999) were identified and counted. Data were collected weekly for 3 weeks in 2017, and for 5 weeks in 2018.

S. vesicarium assessments. Leaves were removed from the field trial to confirm pathogen presence and estimate colonization. In each plot, 10 leaves were randomly chosen and excised to determine the severity of *S. vesicarium* colonization. Leaves were placed singly into plastic bags (Uline poly bags item# s-7498) containing a moist paper towel and incubated at room temperature for 7 days. After incubation, the incidence of *S. vesicarium* conidia and *Pleospora allii* pseudothecia were recorded. The total leaf length and length of leaf colonized by *S. vesicarium* were measured using a dissecting microscope to determine the percent of leaf colonized by *S. vesicarium* in each treatment. Leaves were collected at two times in each year, on 25 Jul and 8 Aug 2017, and 18 Jul and 1 Aug 2018 (80 leaves total per treatment/year).

Onion senescence and yield. Onion senescence and yield data were collected in 2018, but not in 2017. Onions naturally undergo plant senescence in which the onion pseudostem weakens due to decreasing levels of photosynthate to foliage, and as a result the foliage collapses (Brewster 2008). The percentage of plants naturally senescing were recorded in each plot on 22 Aug 2018. Onion plants were deemed as naturally senesced if foliage had lodged, necks were soft, and roots dead. Onions were harvested on 29 Aug 2018. Any remaining dried leaves on onion bulbs were mechanically removed, and bulbs graded and weighed. Bulbs were classified according to bulb diameter and assigned a size class of either 'boiler' (2.5 cm-4.8 cm), 'standard' (4.9 cm-7.6 cm), or 'jumbo' (≥7.7 cm). Bulbs that were either 'standard' or 'jumbo' were considered marketable, and 'boiler' bulbs unmarketable. Marketable yields for treatments were then extrapolated to estimate mean tons per hectare based on final onion stand counts in 2018.

Statistical analyses

Thrips and *S. vesicarium* laboratory studies. Data for each cultivar were analyzed independently. Data were analyzed using a generalized linear mixed model (GLMER) with the R library lme4 package (Bates et al. 2015). Feeding damage ('no damage', 'low damage', 'high damage') and *S. vesicarium* inoculation (inoculated, uninoculated) were treated as fixed effects and plant within trial as a random effect. Leaf dieback measurements, *Stemphylium* colonization, and incidence of *S. vesicarium* conidia and

P. allii pseudothecia were analyzed assuming a binomial distribution. Low incidence of lesions in all trials and *P. allii* pseudothecia in 'Ailsa Craig' precluded its inclusion in statistical analysis. Treatments in each analysis were compared using least squared means (P<0.05) using R package 'emmeans' (Lenth et al. 2018).

Thrips and *S. vesicarium* field studies. Data (number of thrips larvae, number of lesions per plant, leaf dieback measurements, Stemphylium colonization, and incidence of *S. vesicarium* conidia and *P. allii* pseudothecia) from 2017 were statistically similar to those in 2018 and thus pooled for statistical analysis. Data were analyzed using a generalized linear mixed model (GLMER, LMER) (Bates et al. 2015). Insecticide use (insecticide or no insecticide) and fungicide use (fungicide or no fungicide) were treated as fixed effects and replicated within year as a random effect. All count data, including the number of thrips larvae and number of lesions per plant, were analyzed assuming a Poisson distribution. Leaf dieback measurements, incidence of *S. vesicarium* conidia and *P. allii* pseudothecia and *Stemphylium* colonization were analyzed assuming a binomial distribution. Onion yield in 2018 was analyzed using a normal distribution. Treatments in each analysis were compared using least squared means (P<0.05) using R package 'emmeans' (Lenth et al. 2018).

3. Results

Thrips feeding damage impact on *S. vesicarium* incidence in the laboratory

Leaf dieback. Naturally low levels of leaf dieback were present in untreated controls with no thrips feeding damage (3-11%), but this dieback only occurred in the tips of leaves and did not impact overall plant health. The greatest amounts of leaf dieback were consistently associated with thrips feeding damage and *S. vesicarium* inoculation; however, results differed between onion cultivars. In ‘Avalon’, leaf dieback was significantly impacted by the interaction between thrips feeding class and *Stemphylium* inoculation ($F_{2,261}$= 10.4, *p=0.005*) (Table 1). Plants with high thrips feeding damage that were inoculated with *S. vesicarium* had the highest amount of leaf dieback (46.6%), whereas those with no feeding damage that were not inoculated had the lowest amount of dieback (11.1%). Similarly, in ‘Ailsa Craig’, leaf dieback was affected by the interaction of thrips feeding damage and *S. vesicarium* inoculation ($F_{2,261}$= 42.3, $p < 0.001$). Plants with the highest amount of feeding damage and were inoculated with *S. vesicarium* had the greatest amount of leaf dieback (37.1%) (Table 1). Uninoculated plants with no feeding damage had lowest level of leaf dieback (3.2%) (Table 1).

Stemphylium vesicarium infection. Stemphylium leaf blight disease in controlled laboratory experiments was significantly impacted by the amount of thrips feeding prior to inoculation. In cv. ‘Avalon’, thrips feeding damage significantly impacted the likelihood of *S. vesicarium* colonization ($F_{2,129}$= 14.4, *p= 0.0007)*, and onions with ‘high feeding damage’ were two times more likely to be infected with *S. vesicarium* as compared with undamaged plants (Table 2). Incidence of *S. vesicarium* in ‘Ailsa Craig’ tended to increase with thrips feeding, but was not statistically significant ($F_{2,129}$= 4.9, *p= 0.08)* (Table 2). ‘Avalon’ plants with ‘high feeding damage’ had a greater percentage of plants with *P. allii* as compared with undamaged controls ($F_{2,129}$= 11.9, *p= 0.002*) (Table 2). Data on the presence of *P. allii* was not collected in all trials using ‘Ailsa Craig’, which precluded statistical analysis.

Consistently, in both onion cultivars, plants with higher levels of thrips feeding damage had higher percentages of leaf area colonized by *S. vesicarium* ('Avalon': $F_{2,129}$= 34.2, $p < 0.001$; 'Ailsa Craig': $F_{2,129}$= 8.12, $p=0.017$) (Figure 1). In 'Avalon', plants with high thrips feeding damage had the highest leaf area colonized by *S. vesicarium* (51.8%), which was 3-4 times greater than undamaged leaves (13.4%) (Figure 1). While 'Ailsa Craig' had lower levels of infection, plants with high levels of thrips feeding damage had 2-3 times more leaf area colonized by *S. vesicarium* (15.8%) than undamaged plants (5.8%) (Figure 1).

Thrips transfer of *S. vesicarium* conidia in the laboratory

Conidia were observed attached to the bodies of both onion thrips larvae and adults (Figure 2). Thrips successfully transferred *S. vesicarium* conidia to onion plants in 3 out of 4 trials. In trial 1, 14% (7/50) of plants were infected with *S. vesicarium* after being inoculated with an onion thrips exposed to *S. vesicarium*, 0% in trial 2, 6% (3/50) in trial 3, and 2% (1/50) in trial 4.

Relationship between thrips damage and Stemphylium leaf blight disease in field trials

Visual estimates of onion thrips densities and Stemphylium leaf blight disease. Thrips densities and SLB symptoms increased as the season progressed in both years of the field trial (Supplemental Figure 1). However, insecticide use reduced the number of thrips and symptoms of SLB. Onion thrips densities were significantly lower in plots treated with insecticide, but not fungicide ($F_{1,23}$= 136.1, $p<0.001$) (Table 3). Plots treated with insecticide had a seasonal mean total of 13.2 thrips per plant, whereas plots without insecticide had 6 times more thrips per plant (84.9 thrips/plant).

Mean seasonal percent leaf area with dieback was significantly impacted by both insecticide ($F_{1,23}$= 16.1, *p<0.001*) and fungicide use ($F_{1,23}$= 3.8, *p=0.04*), but not by the interaction of the pesticides. Numerically, insecticide appeared to have the greatest impact on the amount of leaf area with dieback, and treatments with insecticide had the lowest amount of leaf area with dieback (Table 3). The number of lesions per plant was significantly affected by both insecticide and fungicide use ($F_{1,23}$= 13.7, *p<0.001* and $F_{1,23}$= 24.2, *p<0.001* respectively), but not the combination of the pesticides. Treatments with either fungicide or insecticide had approximately 2.1 lesions per leaf, whereas plants without fungicide or insecticide had 2.8 lesions per leaf (Table 3).

Impact of pesticides on *S. vesicarium* infection. Thrips control significantly impacted *S. vesicarium* colonization, but not the incidence of *S. vesicarium* or *P. allii*. The proportion of plants infected with *S. vesicarium* was high (Table 4), and there were no significant differences in the proportion of plants with *S. vesicarium* conidia following incubation in plastic bags in any of the treatments (*p>0.05*) (Table 4). The proportion of plants infected with *P. allii* following incubation was significantly impacted by only fungicide use, and plants treated with fungicide had less *P. allii* (0.92) as compared with those that were untreated (0.98).

The percent leaf area colonized by *S. vesicarium* was significantly impacted by sampling date ($F_{1,468}$= 56.04, *p<0.001*), but trends in data remained consistent between the sampling dates (Figure 3). Percent leaf area colonized by Stemphylium was significantly impacted by both fungicide and insecticide use. Overall, plants treated with fungicide had 39% less leaf area colonized by *S. vesicarium* as compared with untreated plants. Similarly, plants treated with insecticide had 17% less leaf area colonized by *S. vesicarium* as compared with the untreated plants (Figure 3).

Onion senescence and yield. In 2018, onion senescence and marketable yield were significantly impacted by pesticide use. Onion senescence was significantly impacted by the interaction of insecticide and fungicide use ($F_{1,10}$= 4.84, p=*0.02*), and all treatments were significantly different from one another. Untreated controls had the lowest percentage of plants senescing successfully (2.7% ± 0.4%), followed by fungicide use only (21.7% ± 20.1%), insecticide use only (45.2% ± 24.8%), and the combination of both insecticide and fungicide use (71.2% ± 22.2%). Marketable bulb yield was significantly impacted by insecticide use, but not fungicide use or the interaction between the two. Insecticide use increased bulb weights by 86% as compared with those not treated with insecticide (Table 3). While fungicide use was not statistically significant, bulb weights were numerically larger in fungicide-treated plots as compared with the untreated control (33.3 tons/hectare v. 28.4 tons/hectare).

Table 1: Mean (±SE) leaf dieback in the onion cultivars 'Avalon' and 'Ailsa Craig' that were either inoculated with S. vesicarium or untreated after experiencing three different levels of onion thrips feeding damage in a controlled environment. Within each cultivar, means with differing letters indicate significant differences among Stemphylium inoculation x feeding damage levels, as determined by LSMEANS at a 0.05 significance level.

Onion cultivar	Onion thrips feeding damage level	Stemphylium inoculation	Mean (±SE) percent plant dieback
'Avalon'	No	-	11.1 ± 4.7 c
		+	28.7 ± 6.7 bc
	Low	-	23.9 ± 6.4 c
		+	34.5 ± 7.1 b
	High	-	29.3 ± 6.8 bc
		+	46.6 ± 7.4 a
'Ailsa craig'	No	-	3.2 ± 2.6 d
		+	22.8 ± 7.6 bc
	Low	-	17.4 ± 5.6 cd
		+	25.1 ± 6.5 ab
	High	-	19.1 ± 5.8 bc
		+	37.1 ± 7.1 a

Table 2: Mean (±SE) proportion of onion plants with S. vesicarium conidia and P. allii pseudothecia for cultivars ‘Avalon’ and ‘Ailsa Craig’ that had different levels of onion thrips feeding damage and then were either inoculated with S. vesicarium or untreated. Within each cultivar, means with differing letters indicate significant differences among feeding damage levels as determined by LSMEANS at a 0.05 significance level. Low incidence of lesions in all trials and P. allii pseudothecia in ‘Ailsa Craig’ precluded their inclusion in statistical analysis.

Onion cultivar	Onion thrips feeding damage level	Incidence of *S. vesicarium* conidia	Incidence of *P. allii* pseudothecia
‘Avalon’	No	0.46 ± 0.09 b	0.30 ± 0.08 b
	Low	0.76 ± 0.07 a	0.48 ± 0.08 a
	High	0.91 ± 0.04 a	0.74 ± 0.07 a
‘Ailsa Craig’	No	0.30 ± 0.08 ns	--
	Low	0.53 ± 0.09 ns	--
	High	0.56 ± 0.09 ns	--

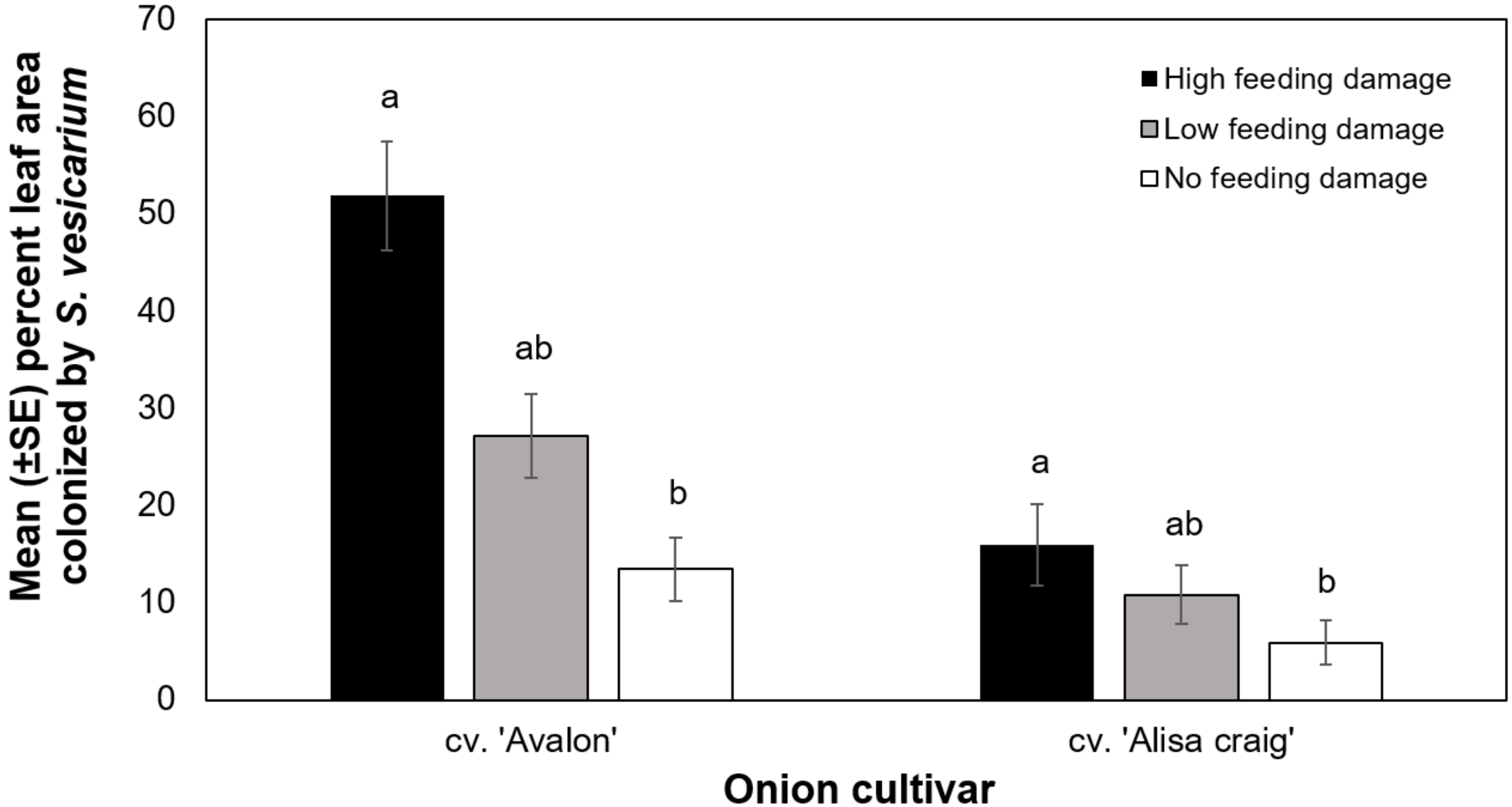


Figure 1: Amount of *Stemphylium vesicarium* colonization on leaves that had different levels of onion thrips feeding damage. For each cultivar ('Avalon' and 'Ailsa Craig'), means with differing letters indicate significant differences among feeding damage levels as determined by LSMEANS at a 0.05 significance level.

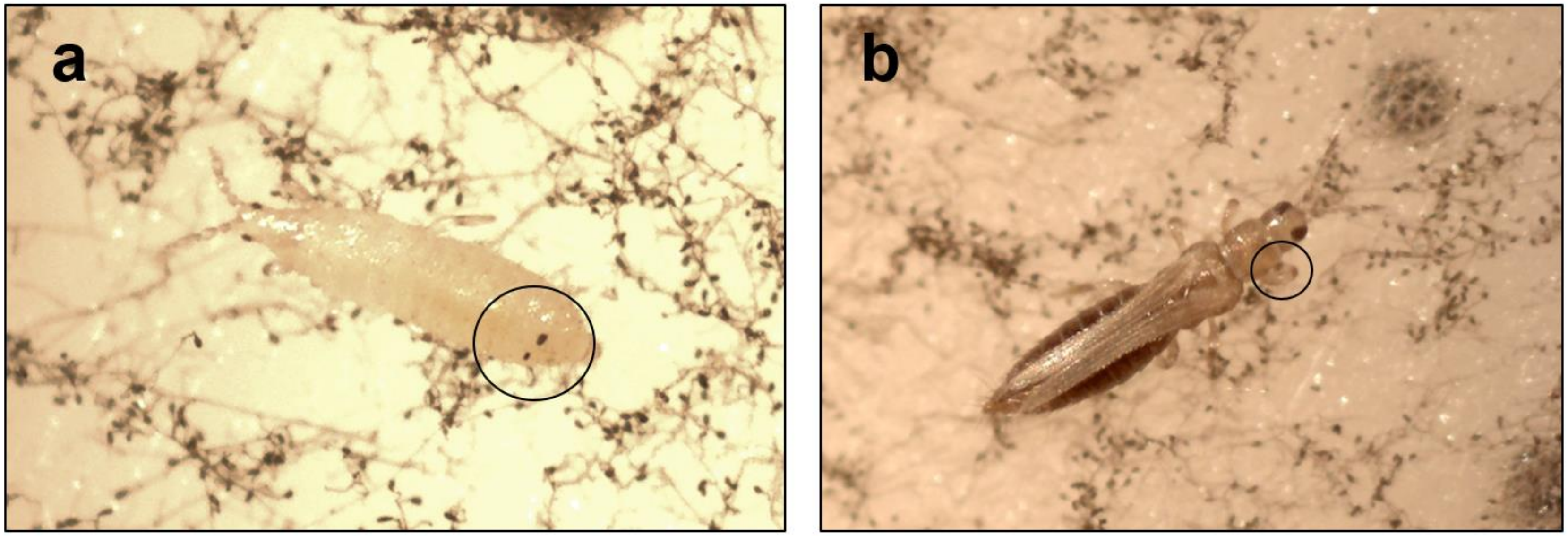


Figure 2: Thrips larva (a) and adult (b) with *Stemphylium vesicarium* condiospores attached to the abdomen and prothorax respectively (circled in black).

Table 3. Visual assessment of Stemphylium leaf blight disease in the field. Combinations of fungicide and insecticide were applied, and their effect on the number of thrips, lesions, leaf dieback, and marketable bulb yield were measured. Data below were generated by combining years; insecticide and fungicide main effects were significant, but not the interaction. Means within a pesticide main effect with differing letters indicate significant differences as determined by LSMEANS at a 0.05 significance level.

Pesticide use	Mean (SE) thrips per plant	Mean (SE) lesions per plant	Mean (±SE) percent dieback per plant	Mean (±SE) marketable yield
Fungicide use	51.1± 4.7 ns	2.1 ± 0.1 b	73.8 ± 0.01 b	33.3± 5.2 ns
No fungicide	47.1± 4.3 ns	2.9 ± 0.1 a	76.0 ± 0.01 a	28.4 ± 3.1 ns
Insecticide use	13.2± 0.7 b	2.2 ± 0.1 b	71.7 ± 0.1 b	41.6 ± 2.8 a
No Insecticide	84.9± 6.1 a	2.8 ± 0.1 a	78.1 ± 0.1 a	22.3 ± 3.4 b

Table 4. Mean (±SE) proportion of plants with either S. vesicarium conidia or P. allii pseudothecia that were treated with combinations of an insecticide and a fungicide in the field. Data below were generated by combining years; insecticide and fungicide main effects were significant, but not the interaction. Means within a pesticide main effect with differing letters indicate significant differences as determined by LSMEANS at a 0.05 significance level.

Pesticide use	Mean proportion of plants with *S. vesicarium* conidia			Mean proportion of plants with *P. allii* pseudothecia		
	Week 1	Week 2	Total	Week 1	Week 2	Total
Fungicide use	0.96 ± 0.09 ns	0.97 ± 0.09 ns	0.97 ± 0.09 ns	0.92 ± 0.09 ns	0.94 ± 0.09 ns	0.92 ± 0.09 b
No fungicide	0.98 ± 0.07 ns	0.99 ± 0.07 ns	0.98 ± 0.07 ns	0.97 ± 0.07 ns	0.99 ± 0.07 ns	0.98 ± 0.07 a
Insecticide use	0.96 ± 0.04 ns	0.97 ± 0.04 ns	0.97 ± 0.04 ns	0.95 ± 0.04 ns	0.95 ± 0.04 ns	0.95 ± 0.04 ns
No Insecticide	0.98 ± 0.08 ns	0.99 ± 0.08 ns	0.98 ± 0.08 ns	0.93 ± 0.08 ns	0.97 ± 0.08 ns	0.95 ± 0.08 ns

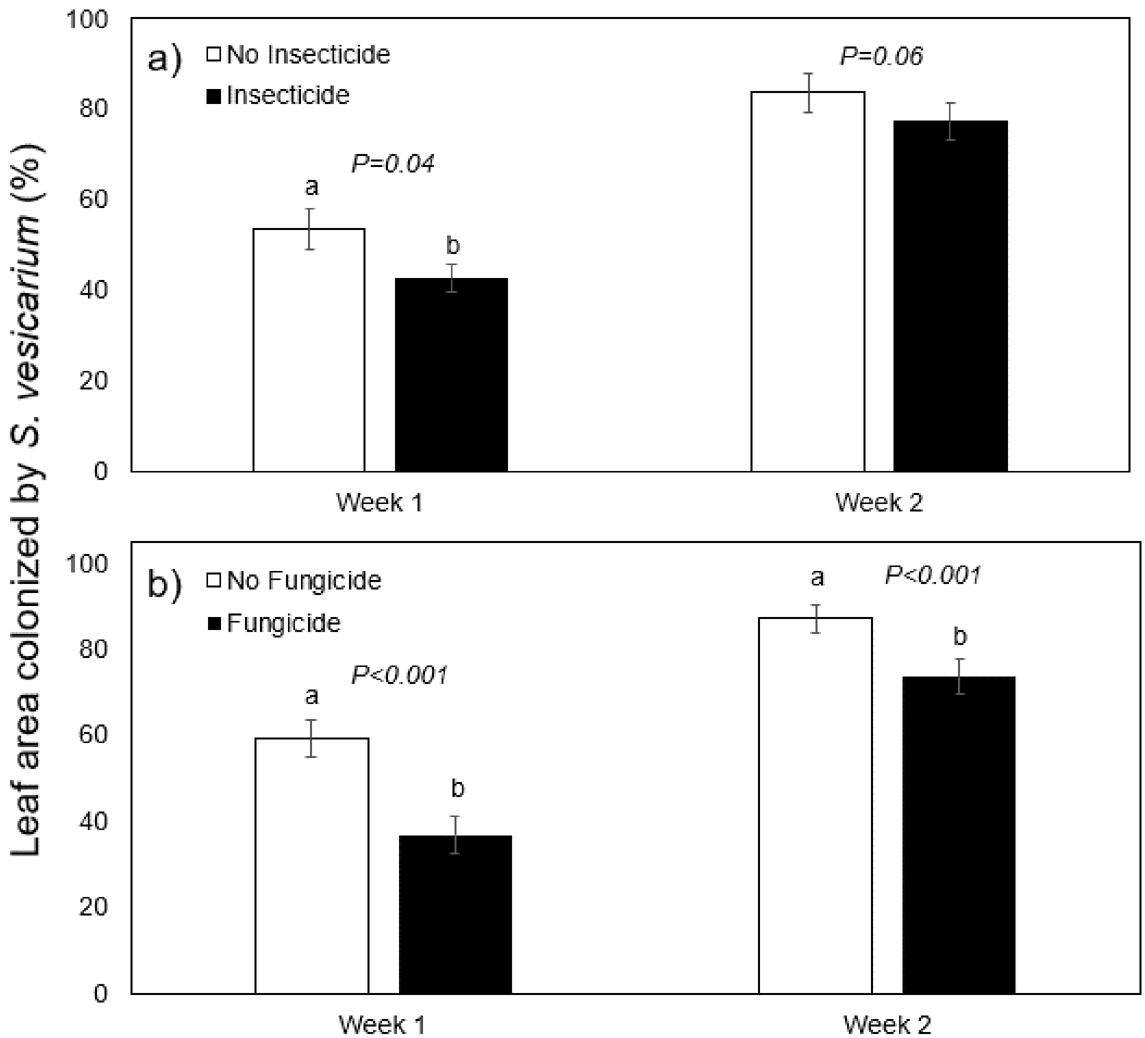
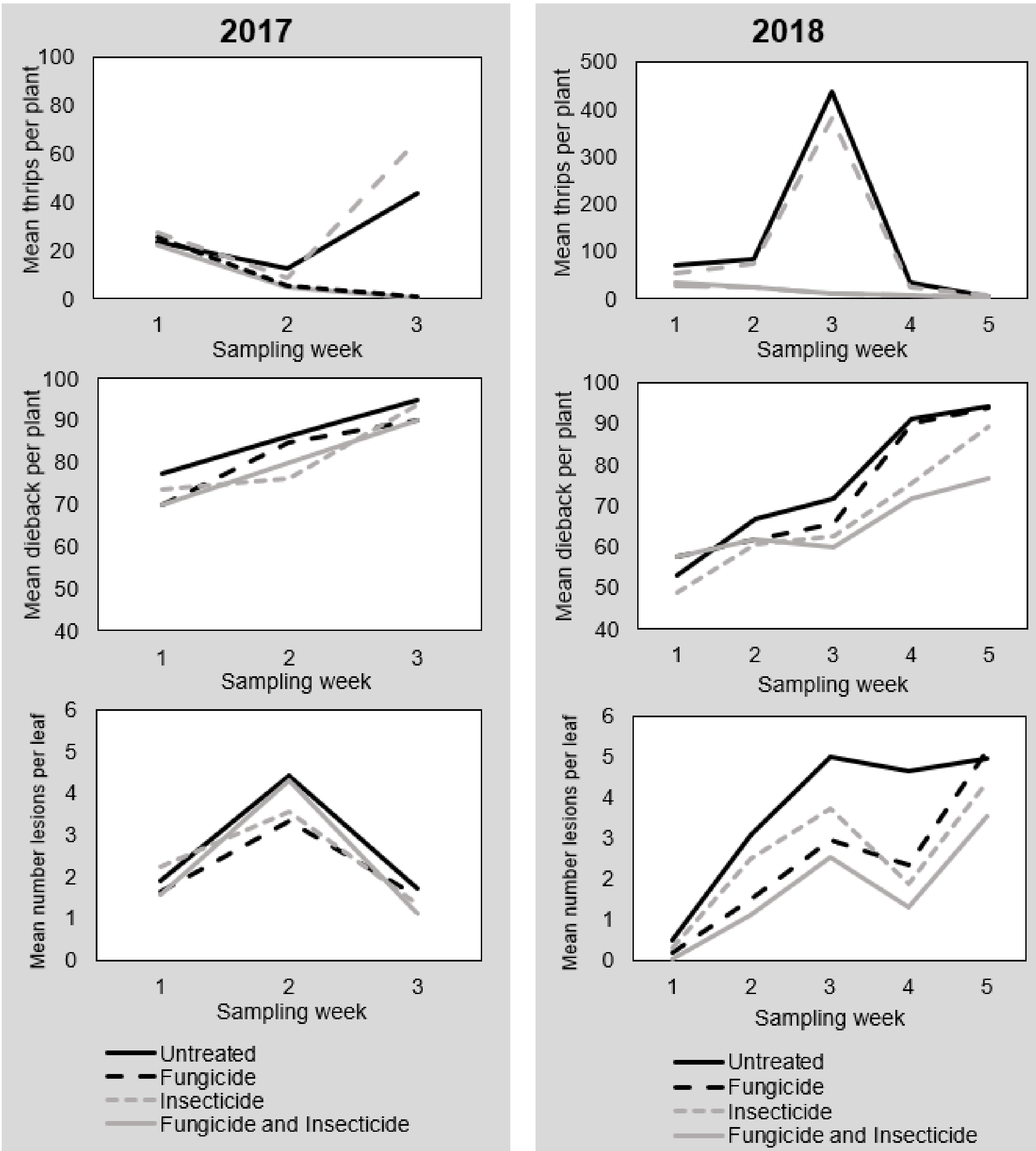


Figure 3: Mean (±SE) percent leaf area colonized by *S. vesicarium* treated with combinations of pesticides, a) insecticide treatments and b) fungicide treatments over two sampling periods (week 1 and week 2) in 2017 and 2018. Results were generated by combining years; insecticide and fungicide main effects were significant, but not the interaction. Means within a pesticide main effect with differing letters indicate significant differences as determined by LSMEANS at a 0.05 significance level.



Supplemental figure 1: Progression of thrips populations and Stemphylium leaf blight disease over time in 2017 and 2018 field trials in Elba, NY. Mean thrips densities per plant in 2017 (a) and 2018 (d), mean plant dieback in 2017 (b) and 2018 (e), and mean lesions per leaf in 2017 (c) and 2018 (f). Data was taken during 3 sampling dates in 2017, beginning on 9 Aug (1), 15 Aug (2), and 22 Aug (3). In 2018, data was collected over 5 sampling dates: 18 Jul (1), 24 Jul (2), 1 Aug (3), 7 Aug (4), and 15 Aug (5). Untreated plots did not received any pesticide, whereas treated plots received weekly applications of either insecticide, fungicide, or a combination of both. Fungicide treated plots received fluopyram/pyrimethanil (0.58 kg (AI) ha-1), insecticide treatments spinetoram (0.07 kg (AI) ha-1), and fungicide and insecticide treated plots a combination of fluopyram/pyrimethanil (0.58 kg (AI) ha-1) and spinetoram at (0.07 kg (AI) ha-1)

4. Discussion

Stemphylium leaf blight can have a major deleterious impact on the yield and quality of onion (Tomaz and Lima 1986; Hoepting 2018ab). Understanding the epidemiology of *S. vesicarium* within the onion production system is critical for developing effective management tactics to mitigate effects of the disease. In our controlled laboratory experiments and field trials, onion thrips infestations consistently exacerbated the development of SLB in onion. In controlled experiments, plants with thrips feeding damage had a greater area colonized by *S. vesicarium*, and thrips successfully transferred conidia to healthy onion plants, leading to successful infection and disease development. In replicated field trials, percent leaf dieback and number of lesions and leaf area colonized by *S. vesicarium* decreased with insecticide use. These results are consistent with our hypotheses and suggest that control of onion thrips is important not only to obviate thrips feeding damage, but also to reduce the impact of SLB in onion.

Thrips are known to exacerbate plant diseases caused by fungal pathogens in multiple cropping systems (Yarwood 1943; Bhangale and Joi 1983; Farrar and Davis 1991; McKenzie et al. 1993; Mailhot et al. 2007; Osekre et al. 2009). Thrips possess unique asymmetrical mouthparts, which enable them to pierce and suck plant tissue (Chisholm 1984). Thrips may damage plants by directly feeding on tissue, removing cell contents, or through probing the plant to determine host suitability (Kindt et al. 2003). Damage caused by thrips feeding can provide alternate entry points for plant pathogens (McKenzie et al. 1993). McKenzie et al. (1993) observed this relationship in the development of purple blotch in onion, and *Alternaria porri* was observed entering though thrips feeding injury on onion leaves. Similar to McKenzie et al. (1993), our study associated higher thrips feeding damage with greater levels of pathogen colonization and disease severity, which may indicate that *S. vesicarium* is taking advantage of thrips feeding injury to infect the onion plant. Although *S. vesicarium* can invade healthy tissue (Suheri and Price 2000), the pathogen often invades dead and dying onion tissue including necrotic leaf tips, purple blotch and downy mildew lesions and injured or senescent tissue (Miller and Schwartz 2008). *Stemphylium vesicarium* is also known to live saprophytically in necrotic plant tissue (Rossi et al. 2005; Köhl et al. 2009). Furthermore, these results imply that any physical damage to the plant (e.g., herbicide injury) may significantly impact *S. vesicarium* colonization of onion and Stemphylium leaf blight disease.

In our field trial, we observed that effective thrips control reduced SLB, but the difference was slight (17%) compared with treatments that did not use insecticide. This was surprising given that our laboratory results consistently showed plants with no or low levels of thrips feeding damage had the lowest levels of Stemphylium colonization compared with plants with high levels of thrips infestation. In our field trial, seasonal densities of thrips in insecticide-treated plots averaged 13 thrips per plant. However, weekly densities of thrips per plant fluctuated throughout the growing season, and plots treated with insecticide had weekly means ranging between 0.8 thrips per plant to 33 thrips per plant. Feeding damage from one week may have increased the plant’s susceptibility to *S. vesicarium*, thereby limiting the effect of subsequent insecticide applications. Further research should address timing as it relates to thrips control and Stemphylium leaf blight disease severity. Previous research has shown that managing thrips with insecticides reduces the incidence of two fungal diseases, hard lock and ear rot in cotton and corn, respectively (Farrar and Davis 1991; Mailhot et al. 2007). In New York, onion thrips colonization precedes the onset of Stemphylium leaf blight disease and early season thrips feeding damage may predispose onions to *Stemphylium* infection.

An additional reason that may explain why we observed a minimal effect of insecticide reducing *Stemphylium* colonization in the field trials were high levels of Stemphylium leaf blight inoculum and plant age. While we did not trap for ascospores or conidia in our field trial, previous research has shown that these spores are present throughout the growing season but reach peaks in late June to Mid-August (Misawa and Yasuoka 2012; Tayviah 2017). Therefore, the high amount of inoculum present in the environment may have negated any potential practical benefit of insecticide use. Our trial was also initiated during the bulbing and postbulbing stages, and age of plant has been shown to impact the plant's susceptibility to *S. vesicarium*. Shishkoff and Lorbeer (1989) found that older leaves were 3.5 times more susceptible to *S. vesicarium* compared with younger leaves. Thus, the impact of pesticides in the reduction of Stemphylium leaf blight during the bulbing and postbulbing stages may be negligible due to the conducive environmental conditions and susceptible plant physiology.

S. vesicarium may differentially colonize leaf tissue based on cultivar susceptibility and thrips damage. While the lack of cultivar replication in our lab trials precluded the inclusion of cultivar in our statistical analysis, we consistently observed that 'Ailsa Craig' was less susceptible to *S. vesicarium* and limited the effect of thrips feeding on *S. vesicarium* colonization compared with 'Avalon'. Previous studies have identified differences in onion cultivar and SLB (Pashtak et al. 2001; Tayviah 2017). However, it is possible that this susceptibility may be mediated by damage to leaves. For example, in our laboratory assays, we observed that ‘Avalon’ with no thrips feeding damage had similar levels of *S. vesicarium* colonization as ‘Ailsa Craig’ with high thrips feeding injury. Therefore, resistance in onion cultivars to SLB may depend on the pesticide programs used to control physical damage to leaves in the field. Nevertheless, our results indicate that onion cultivar is an important consideration when managing SLB in onion.

The role of thrips movement on the spread of *S. vesicarium* may impact the epidemiology of SLB in onion fields. While *S. vesicarium* is primarily dispersed aerially (Prado-Ligero et al. 2003; Rossi et al. 2005), thrips also may play a role in the pathogen’s dispersal. Thrips are highly mobile in cropping systems and move readily both within and between plants (Lewis 1991). While less common, thrips also have been known to disperse long distances, and may travel hundreds of kilometers in the right environmental conditions (Lewis 1991; Laughlin 1977). Studies in New York onion fields showed that most onion thrips tended to disperse short distances (trivial movement), but that some engage in long distance dispersal (Smith et al. 2015). In laboratory trials, we found that thrips could transfer conidia and infect healthy plants with *S. vesicarium*. The success rate of thrips transferring *S. vesicarium* conidia was low (2-14%), but our assays only examined the effect of a single thrips, and the success rate of infection may be higher with greater densities of onion thrips and necrotic leaf tissue. Thrips populations can reach high densities on onion during the growing season (e.g., 262 thrips/plant) (Nault and Hessney 2010) and *S. vesicarium* lesions can contain

200 conidia/cm^2 (Miller and Schwartz 2008), and conceivably even a small proportion of thrips inadvertently carrying conidia may amount to high levels of *S. vesicarium* transmission and infection. Onion thrips are present on onion leaves for most of the growing season (Gill et al. 2015), and are likely to contact most, if not all, foliar plant pathogens of onion. Further study may address the contribution of onion thrips moving fungal spores within onion fields.

Stemphylium leaf blight disease is a challenge for onion growers to manage, especially since *S. vesicarium* has developed resistance to multiple fungicides (Hay et al. 2018). As a result, growers are limited in the number of efficacious fungicides they can use to control the disease on their farm. Our trials showed that onion thrips have a positive relationship with *S. vesicarium* and can significantly worsen the disease. However, one potential way to mitigate this disease is to apply insecticides, which will reduce thrips abundance, thus limiting feeding damage and potentially slowing the spread of the disease. In our field trials, we observed a decrease in Stemphylium leaf blight with insecticide use, but further optimization of pesticide application timing may increase the effect of both insecticide and fungicide use in the management of Stemphylium leaf blight disease.

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