

Evaluating Low Tunnel Plastics for
Day-Neutral Strawberry (*Fragaria ×ananassa* Duchesne)
Production in Minnesota

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Abstract

American consumers place high value on local agriculture and direct market sales, particularly for fruits and vegetables. Growers who supply local strawberries, especially organic, have a competitive edge in the direct-to-consumer market. New developments in extended season strawberry production offer new opportunities for growers in the Upper Midwest to meet this demand for local, organic strawberries using low tunnel protective structures in an annual day-neutral strawberry production system. A range of specialty tunnel plastics that modify the light around plants are now available as well, but there is little information on how these products influence strawberry growth and performance in the field. We tested the effects of experimental UV-blocking and UV-transmitting plastics on light and microclimate in low tunnel environments and on fruit yield and fruit quality in the day-neutral strawberry 'Albion'. We also assessed changes in UV transmittance levels of the plastics over time and evaluated their use in the context of organic insect pest management. We collected data on the presence of the insect pest species *Lygus lineolaris* (tarnished plant bug) and *Tetranychus urticae* (two-spotted spider mite) in the field and tested the effectiveness of the microbial-based organic biopesticides Entrust SC (AI: spinosad), Mycotrol WPO (AI: *Beauveria bassiana*), and PFR-97 (AI: *Isaria fumosorosea*) for control of *Drosophila suzukii* (spotted wing drosophila) in semi-field bioassays. This research was conducted on USDA-certified organic land at the Minnesota Agricultural Experiment Station (MAES) in St. Paul, Minnesota in the 2016 and 2017 growing seasons. We found that both UV-transmitting and UV-blocking plastics improved fruit yield and quality compared to an open control, and the plastics maintained their spectral properties over the course of one season. There were no distinct differences in results observed between the UV-transmitting and UV-blocking treatments. Covering type did not affect the presence of *L. lineolaris* or *T. urticae* in the field, nor did it influence the efficacy of the biopesticides for control of *D. suzukii* in semi-field bioassays.

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Literature Review: Optimizing Protected Tunnel Cropping and Integrated Pest Management for Organic Production of Day-Neutral Strawberries in the Upper Midwest

Introduction

American consumers place high value on local agriculture and direct market sales, creating strong demand around the country for locally produced fruits and vegetables (Howard and Allen, 2010; Jensen and Malter, 1995; Tourte et al., 2016). However, this demand is generally not reflected in the distribution of production – many horticultural crops are concentrated in limited regions of the U.S. One of the most popular fruits in the U.S. is the strawberry (*Fragaria ×ananassa* Duchesne) (Nielsen, 2015; Zillman, 2014). Locally produced out-of-season strawberries are highly sought and command a price premium. Thus, growers who supply local strawberries outside the normal season, especially organic, have a competitive edge in the direct-to-consumer market (Gu et al., 2017; Kadir et al., 2006a; Petran et al., 2017; Rowley et al., 2011). This paper will review developments in extended season strawberry production, focusing on the use of low tunnel protected cropping for day-neutral strawberries in the Upper Midwest and on integrated pest management strategies for organic growers.

Strawberry Supply and Demand in the United States

Berries are currently the most popular fruits in the U.S. based on fresh sales by dollar at grocery stores (Nielsen, 2015; Zillman, 2014). In 2016, strawberries were the third most valuable non-citrus fruit crop, valued at \$2.3 billion (USDA NASS, 2017). Though the U.S. is one of the major world producers of strawberries (Wu et al., 2012), it has been a net importer of fresh strawberries since 2012 due to steadily increasing demand (USDA ERS, 2017). Between 1980 and 2016, annual consumption of fresh strawberries in the U.S. increased from 2 pounds per person to 8 pounds per person (Ferreira and Perez, 2017). This trend may be linked in part to more year-round availability of strawberries from domestic production and imports, as well as use of improved varieties, but the increase in consumer demand is likely fueled by greater understanding about healthy diets (Cook, 2011; USDA ERS, 2016; Zillman, 2014). Berries can be a good source of fiber, vitamins, minerals, and other

bioactive compounds that contribute to good nutrition and build a body's defenses against certain chronic illnesses (Nile and Park 2014, Seeram, 2010). Information about the health benefits of berries and other fruits is widely disseminated by government programs to encourage healthy eating (CDC, 2015; USDA, 2015) and is subsequently shared by berry promotion programs such as the National Berry Crop Initiative (Cook, 2011; Seeram, 2010).

Climate plays a major role in determining regional and site suitability for strawberries (Rysin et al., 2015). Currently, the U.S. strawberry industry is concentrated in California and Florida, which together accounted for 98 percent of total production in 2015 (USDA NASS, 2016). Depending on the cultivar, strawberries can be sensitive to variables such as late spring frosts, low winter minimum temperatures, and short growing seasons (Carroll et al., 2015). There are three types of strawberry cultivars, categorized by their flowering and fruiting habits: June-bearing, everbearing, and day-neutral (Darrow and Waldo, 1933; Gu et al., 2017). June-bearing cultivars produce fruit for several weeks in early summer and are typically grown in a perennial system with matted rows (Hoover et al., 2014; Solomon et al., 2001). These cultivars induce flower buds during the shortening day lengths of fall and are dormant in winter. Warming temperatures and lengthening days stimulate flowering in the spring (Darrow and Waldo, 1933). Everbearing cultivars flower under longer photoperiods but are not produced commercially (Petran, 2016; Sønsteby and Heide, 2007). Day-neutral cultivars flower and fruit continuously, regardless of photoperiod (Durner et al., 1984).

Nationally, commercial strawberry production favors day-neutral cultivars for their longer season and higher yield potential compared to June-bearing cultivars, but historically, day-neutral cultivars have not performed well in northern regions of the U.S. such as the Upper Midwest (Darrow and Waldo, 1933; Petran et al., 2017). Currently available day-neutral cultivars originated from breeding programs in California, the Eastern U.S., and the United Kingdom and have not been developed for northern U.S. growing conditions (Dale et al., 2002; Hoashi-Erhardt et al., 2013). Therefore, despite the increasing national demand for access to locally produced foods, commercial strawberry production in the Upper Midwest has been mostly limited to lower-yielding June-bearing cultivars (Hoover et al., 2014; Petran, 2016;

Wold and Hutchison, 2003a). These are better-adapted to overwintering and producing in short growing seasons. However, recent developments in protected agriculture systems have created new opportunities for producing day-neutral strawberries as annuals in some regions of the U.S. where conditions have traditionally been considered unsuitable (Hoover et al., 2014; Petran et al., 2017; Solomon et al., 2001).

Protected Agriculture Systems for Strawberry Production

Protected agriculture refers to any system of modifying the natural environment around a crop to improve its growth and performance. Modifications made to both root and aerial environments include anything from mulches and row covers to tunnels and greenhouses (Jensen and Malter, 1995). Growing crops under protection can be beneficial; particularly well-documented are the benefits of high tunnel protective structures for berries. The methods recommended for growing berries in high tunnels closely follow those recommended for field production, with only minor management adjustments needed (Heidenreich et al., 2007; Jett, 2007; Lamont et al., 2003). Shielded from rain and wind, fruits sustain less damage under high tunnels (Jett, 2007). Berries are also cleaner with less surface moisture at harvest (Karlsson and Werner, 2011). Tunnels can increase the amount of time at which plants are held at optimal growing temperatures (Kadir et al., 2006a; Rowley et al., 2011) and protect plants from some early end-of-season frost events (Demchak and Hanson, 2013; Karlsson and Werner, 2011).

Kadir et al. (2006a) found that overwintering June-bearing strawberries in Kansas under high tunnels resulted in less cold damage to plant crowns than overwintering in an open field. Furthermore, during the growing season, high tunnel production resulted in earlier flowering and fruiting in strawberries compared to results from open field production. Whereas field conditions promoted runner development and vegetative growth, high tunnels promoted branch-crown development, increasing fruit yield and quality. Findings from the research of Nes et al. (2017) on organic strawberry production methods in southern Norway indicate that strawberries grown in high tunnels are less susceptible to changing weather conditions than strawberries grown in open field production. The more diffuse light conditions under tunnels may result in better penetration of light to lower leaves,

thereby increasing a plant's photosynthesis (Baeza and López, 2012; Demchak, 2009).

Grower interest in high tunnel berry production is strong. Where land is expensive or limited and where inclement weather makes production risky, growers are keenly aware of the value protected cropping can offer (Demchak and Hanson, 2013). In the U.S., the federal government promotes high tunnel production as an environmentally sound method of extending the growing season for high value crops. Through the Natural Resources Conservation Service (NRCS) Environmental Quality Incentives Program (EQIP), producers can receive financial and technical assistance to construct high tunnels (USDA NRCS, n.d.). The combined effects of the numerous high tunnel benefits can lead to significantly higher yields (Kadir et al., 2006a; Karlsson and Werner, 2011; Rowley et al., 2011), extended seasons (Demchak, 2009; Demchak and Hanson, 2013; Kadir et al., 2006a), and higher fruit quality with longer shelf life (Demchak, 2009; Kadir et al., 2006a; Karlsson and Werner, 2011; Nes et al., 2017).

Similar to high tunnels but relatively less utilized are low tunnels. These structures allow for a more efficient use of space for growing low-stature plants like strawberries under protection. In the low tunnel system, strawberries are grown on raised beds with plastic mulch. Steel hoops spaced evenly down the length of a bed support a plastic covering roughly 0.6 meters off the ground. Landscape fabric is often used for weed control on walkways between beds (Demchak and Hanson, 2013; Gu et al., 2017; Hoashi-Erhardt et al., 2013; Kadir et al., 2006a; Lewers et al., 2017). Strawberries are planted directly in the plastic mulch and watered via drip irrigation, which is more efficient than overhead irrigation. The plastic mulching discourages weeds – often a significant challenge since herbicides alone do not provide sufficient weed control. Excessive cultivation to remove weeds can be harmful to soil health by causing erosion, reducing soil organic matter, and breaking down soil structure. Left uncontrolled, weeds compete for water and nutrients, provide hosts for pests, and interfere with planting and harvesting. Thus, proper weed management is imperative (Carroll et al., 2015).

Low tunnel production comes with some unique advantages over high tunnel production. Long-term high tunnel growers have identified soil compaction and

quality as issues in their systems (Demchak and Hanson, 2013). Low tunnels, which are not permanent structures, can easily be moved to new fields annually, reducing the risk of soil compaction. This also gives growers more flexibility in adjusting the scale of production from year to year. Furthermore, strawberries are highly sensitive to soil salinity, and since tunnel environments prevent rainfall from leaching salts out of root zones (Jett, 2007), moving tunnel sites each year may reduce the risk of accumulating harmful levels of salt in the soil. Other problems growers have observed with high tunnels include building and maintenance costs (Lewers et al., 2017), difficulties with temperature management, and loss of tunnels in extreme weather (e.g. severe winds, excessive snow) (Demchak and Hanson, 2013). Low tunnel materials can be expensive initially, but the hoops can be re-used over multiple years, and actual tunnel construction is relatively simple. Temperature management is fairly easy with low tunnels since they do not require complex venting schemes – the sides can be opened or closed manually, and some tunnel plastics incorporate ventilation holes that run the length of the plastic. As with high tunnels, low tunnels can also offer some frost protection when closed (Gu et al., 2017).

Tunnels and Organic Disease Management

Strawberries are highly susceptible to pests and diseases (Andrade et al., 2016; Nes et al., 2017), and so it is important to have multiple effective options for pest prevention and control. This is particularly true in organic systems where growers must rely first on preventive and cultural control methods before resorting to a limited set of permitted pest control products (Caldwell et al., 2013; Fanning et al., 2017; Marques-Francovig, 2014). Most strawberry acreage in the United States is non-organic, but demand for organic products is on the rise, fueled by consumer concern over pesticide residues on conventionally grown fruits and vegetables (Daugaard, 1999; Gu et al., 2017; Hoover et al., 2014). Among fruits, strawberries are one of the top-selling organic products in the U.S. and brought in \$89 million in sales in 2014 (USDA NASS, 2015). In light of this, the development of new strategies for organic pest management has become increasingly relevant.

One way that tunnels can contribute to organic disease management for strawberries is by altering the abiotic environmental conditions that encourage

disease spread, such as temperature, moisture, and light (Carroll et al., 2015). For example, by eliminating rainfall on plants, tunnels create an environment where less moisture accumulates on leaves, reducing the occurrence of certain fungal diseases in strawberries (Burlakoti et al., 2014; Carroll et al., 2015; Demchak, 2009). Burlakoti et al. (2014) observed that under high tunnels, the incidence of *Colletotrichum acutatum* (anthracnose fruit rot) in strawberries was consistently very low compared to the incidence in outdoor field plots. Increased leaf wetness and relative humidity created greater risk of infection for plants in open fields. Another economically significant strawberry disease, *Botrytis cinerea* (gray mold) is typically controlled by chemical fungicides during flowering, though it is developing resistance to these fungicides. Fungicide resistance and growing demand for organic production necessitates alternative, non-fungicidal control methods (Daugaard, 1999). Along these lines, Nes et al. (2017) found that tunnels reduced the incidence of *B. cinerea* in strawberries when compared with the incidence in open field plots in southern Norway, even without the use of fungicides.

Compared to high tunnels, low tunnels may offer even more successful disease management. Because low tunnels are only about 1.2 meters wide, it is easy to achieve good airflow with the sides open. This helps with management of *Sphaerotheca macularis* (powdery mildew), a common problem in strawberries under high tunnels due to restricted air circulation (Karlsson and Werner, 2011). Relative humidity is an important factor in the life cycle of *B. cinerea* as well; good aeration around plants in low tunnels is likely to reduce disease incidence. Air circulation coupled with protection from rain means that foliage, flowers, and fruit remain dry for longer periods of time, which can reduce the duration and frequency of disease infection periods (Carroll et al., 2015; Daugaard, 1999).

The mobility of low tunnels facilitates better management of certain soil-borne diseases as well, such as Verticillium wilt, which is caused by the soil-borne fungi *Verticillium albo-atrum* and *Verticillium dahliae*. When these fungi are widespread, the damage to a strawberry crop can be catastrophic. Once present in the soil, the fungi can survive and re-infect strawberry plants year after year. In conventional systems, synthetic soil fumigants can be used to control for the disease, but in organic systems, growers must rely on crop rotation or biofumigation. Susceptible

crops such as strawberries must be kept out of Verticillium-infected soils for five years to break the cycle of disease (Rysin et al., 2015; UIL, 1997).

Tunnels and Organic Insect Pest Management

In addition to impeding disease development, low tunnel strawberry production may offer new avenues for insect pest management (Baeza and López, 2012; Krizek et al., 2005). The range of strawberry insect pests includes many generalist herbivores with alternate wild and cultivated hosts, making control or elimination difficult. The most detrimental pests of strawberries globally are insects in the Miridae family (capsid bugs) (Solomon et al., 2001). Among these, *Lygus lineolaris* (tarnished plant bug) is a common pest of strawberries in Minnesota (Wold and Hutchison, 2003a). It feeds on the achenes of developing fruits, rendering otherwise good fruit unmarketable by causing distinctive deformities often referred to as “cat-faced” or “button” berries (Carroll et al., 2015; Day and Hoelmer, 2012; Solomon et al., 2001). *Tetranychus urticae* (two-spotted spider mite), another primary strawberry pest in the U.S. and around the world, begins feeding on the leaves of strawberry plants in early spring (Demchak and Hanson, 2013; Solomon et al., 2001; Wold and Hutchison, 2003b). With heavy infestation, the damage it causes can result in sparse new growth and reduced quality and quantity of fruit (Carroll et al., 2015; Livinali et al., 2014). *Drosophila suzukii*, commonly known as the spotted wing drosophila, is a more recent pest in strawberries (Goodhue et al., 2011). It has become one of the most serious pests in soft fruit, including raspberries, blueberries, grapes, blackberries, and cherries, as well as strawberries (Gong et al., 2016; Lee et al., 2011). The female’s serrated ovipositor allows her to deposit eggs in firm, ripening fruit. Fall fruits of day-neutral strawberries in the northern U.S. are at considerable risk from *D. suzukii* and *L. lineolaris*, due to crop phenology that overlaps with high population densities of these insect pests (Carroll et al., 2015; Demchak and Hanson, 2013).

Though synthetic chemical pesticides are commonly applied as a primary means of control for insect pests and plant pathogens in conventional strawberry systems, pesticide usage has many known drawbacks for farmers, consumers, and the environment. Broad-spectrum insecticides can harm insect pollinators and reduce populations of other beneficial, predatory insects (Baeza and López, 2012;

Hamburg and Guest, 1997; Mullin et al., 2010). Recognizing that sole reliance on pesticides is not sustainable, many growers seek to incorporate a greater array of tools, developing Integrated Pest Management (IPM) plans (Baeza and López, 2012; Carroll et al., 2015). Beyond chemical control of a pest, growers can utilize cultural and biological strategies and tools to reduce pest problems organically. IPM relies on a combination of such methods in a more holistic, ecosystem-based approach to pest prevention and control (Radcliffe et al., 2017). Both the crop and the environment must be conducive to the maintenance of a pest before considerable crop damage will occur; therefore, a successful IPM plan addresses not only the pest or pathogen itself, but also the relevant aspects of the host crop and the environment (Carroll et al., 2015). IPM is key to sustainable production of organic day-neutral strawberries, which have a longer cycle of fruit production than June-bearing cultivars and are therefore at increased risk of harboring disease and insect pest problems (Burlakoti et al., 2014; Carroll et al., 2015).

For the high and low tunnel grower, an expanding assortment of tunnel designs and novel plastic covering materials fabricated for specific light absorption and transmission properties are now commercially available (Karlsson and Werner, 2011). These plastic films selectively block or absorb light in wavelengths in the infrared or ultraviolet (UV), or diffuse incoming direct beam solar radiation without inhibiting necessary transmission of photosynthetically active radiation (PAR). PAR is both visible light and the spectral range important for photosynthesis, 400-700 nm (Björn, 2015). The plastic films most commonly used in horticultural production transmit lower levels of UV light, allowing little or no transmission of UV-B (280-315 nm) and reduced transmission of UV-A (315-400 nm), but there are now also plastic films available that are completely opaque to UV (Krizek et al., 2005; Paul et al., 2005). These specialty plastics may aid in pest and disease control in a tunnel system (Baeza and López, 2012; Karlsson and Werner, 2011; Krizek et al., 2005; Paul et al., 2005).

With multiple options, plastic materials used in low tunnels may contribute in multiple ways to insect pest management. Since UV light has a significant impact on insect vision and flight activity, specialty plastics that block UV light can be used to disrupt the movement and activity of insect pests in tunnels. However, responses to

UV light are species-specific (Antignus et al., 1996; Dáder et al., 2015; Díaz et al., 2006; Paul et al., 2012; Raviv and Antignus, 2004). Therefore, utilization of specialty plastics for insect pest management in strawberries must be evaluated under the growing conditions and with exposure to pests specific to strawberry production. A study on the effects of UV light on the strawberry pest *T. urticae* have shown that in a greenhouse environment, UV radiation may actually deter the pest (Tanaka et al., 2016). Sakai and Osakabe (2010) postulate that *T. urticae* preferentially reside on the under side of leaves to avoid UV light, but it is not known whether or not changing UV light conditions would significantly affect *T. urticae* presence in a strawberry production system in the field. Specific responses of *L. lineolaris* and *D. suzukii* to changes in ultraviolet light are unknown.

Beyond directly influencing insect pest activity, tunnels with specific light transmission qualities may provide indirect pest management by creating more favorable conditions for biopesticides (Parikka and Tuovinen, 2014; Solomon et al., 2001; Wekesa et al., 2011). Biopesticides are based on one of three types of ingredients: plant-incorporated protectants, naturally occurring substances that control by non-toxic mechanisms, or microbial agents (EPA, 2016). They represent appealing alternatives to synthetic chemical pesticides as they pose less of a threat to human health and carry fewer environmental risks. Biopesticides are generally biodegradable and more specific to the target pest species (Gupta and Dikshit, 2010). Fruit growers must be careful to meet maximum residual level (MRL) restrictions on pesticides on harvested fruit, and many biopesticides do not have residue restrictions. They can also be included in a spray rotation with other pesticides over the course of a season to reduce the risk of pests developing resistance to one or more products (Fanning et al., 2017; Haviland and Beers, 2012). Additionally, some biopesticides are certified for use in organic production.

One of the more effective biopesticides commonly used in berry production is spinosad (Bruck et al., 2011; Fanning et al., 2017), a fermentation product of the soil bacterium actinomycete *Saccharopolyspora spinosa* (Mertz and Yao, 1990). Several studies have demonstrated its effectiveness for control of *T. urticae* and *D. suzukii* (Bruck et al., 2011; Fanning et al., 2017; Ismail et al., 2007; Pavlova et al., 2017; Van Leeuwen et al., 2005). Entomopathogenic (insect pathogen) fungal-based

microbial biopesticides may also be useful in managing some insect pest species. These fungi are able to infect an insect at all life stages by penetrating the cuticle and invading the body of the insect host, though nymphal stages may be more resilient to attack by the fungus if they molt before infection takes hold (Arthurs et al., 2013; Cory and Hoover, 2006; Dara et al., 2016; Zou et al., 2014). Spores of the fungi can be extracted and formulated into a sprayable product for crop application (Caldwell et al., 2013). *Beauveria bassiana*, a naturally occurring entomopathogenic fungus found in soils worldwide (Caldwell et al., 2013), is considered to have high insecticidal potential for the control of *L. lineolaris* (Portilla et al., 2017; Sabbahi et al., 2008). It has also caused mortality in *D. suzukii* under laboratory conditions (Cossentine et al., 2016). Another entomopathogenic fungus, *Isaria fumosorosea*, has not been extensively studied as a control agent in berry crop production systems but was shown to successfully infect *D. suzukii* in a laboratory environment (Cossentine et al., 2016).

Biopesticides offer promising pest-management alternatives to the broad-spectrum insecticides commonly used in conventional production, particularly for organic growers, but much room remains for improvement in their utilization (Fanning et al., 2017; Ismail et al., 2007; Solomon et al., 2001). A substantial portion of the data on biopesticide efficacy is based on laboratory trials (Bruck et al., 2011; Cossentine et al., 2016; Dara et al., 2017; Ismail et al., 2007; Pavlova et al., 2017; Portilla et al., 2017), and in many cases it is challenging to replicate lab efficacy of a product in the field. Because microbial biopesticides rely on living microbial agents or their byproducts as active ingredients instead of synthetically derived chemicals, the level of control by a biopesticide can depend heavily on environmental factors and the timing of sprays. Some of the same abiotic factors that affect disease spread in strawberries – humidity, UV light, and rainfall – can also substantially impact the efficacy of biopesticide treatments and their persistence in the environment (Arthurs et al., 2013; Caldwell et al., 2013; Cory and Hoover, 2006; Fanning et al., 2017; Ray and Hoy, 2014).

Numerous studies have demonstrated that sunlight exposure quickly degrades microbial agents and reduces their efficacy in the field by damaging spore viability and insecticidal activity for fungal conidia. Ultraviolet (UV) light in the range

of 300–400 nm disrupts normal metabolic processes, preventing the proper transcription of DNA (Behle et al., 2011; Cory and Hoover, 2006). Understanding how these factors influence persistence of a biopesticide in the environment over time is important to appropriately scheduled sprayings. According to Sabbahi et al. (2008) *B. bassiana* conidia can remain viable for control of *L. lineolaris* adults for 6 days in strawberries in the lab. Bruck et al. (2011) found that field application of most spinosyns (the family of compounds related to spinosad) provided residual control of *D. suzukii* for anywhere from 5 to 14 days. Greater persistence is desirable for effective control of many insect species, such as spider mites, since eggs may hatch 5–10 days after a pesticide treatment (Van Leeuwen et al., 2005). Finding methods to extend the residual activity of microbial biopesticides could improve their efficacy in the field and make them more competitive with synthetic chemical pesticides. Specialty tunnel films could play a significant role in this approach. Adjusting the microclimate or filtering light to increase the persistence of microbial biopesticides in the field is an important area to explore (Behle et al., 2011). In the context of strawberry production, a low tunnel system with specialty plastic film covers could be used to create an environment with higher humidity and lower UV radiation around plants in conjunction with use of microbial biopesticides for insect pest management.

Optimizing Light Conditions for Strawberries

A key consideration in the modification of light for pest management is understanding how these changes could produce a range of responses in plant health or fruit quality. Though multiple studies have demonstrated that reduced UV light has a positive effect on management of certain insect pests, other evidence indicates that exposure to UV light improves crop resilience in the face of environmental stressors. As such, blocking UV light could have unintended harmful consequences (Wargent et al., 2011). Certain wavelengths of UV light may help plants by way of photoreceptors that trigger critical defense mechanisms (Ballaré et al., 2012). Changing levels of light exposure may influence many aspects of crop morphology and chemistry (Ballaré et al., 2011; Ballaré et al., 2012). Soluble solids content (including sugars) may be affected by the amount and quality of light a plant receives (Perkins-Veazie, 1995). Tsormpatzidis et al. (2011) found that strawberries from plants grown under plastic films transmitting reduced levels of UV light were

softer and slower to develop color. Elfadley et al. (2012) found that restricting UV light reduced plant growth and biomass over time in lettuce plants. At the same time, restricting UV light can reduce the spread of diseases that affect strawberry fruit quality (Baeza and López, 2012; Karlsson and Werner, 2011; Krizek et al., 2005).

There is no one set of light conditions that will produce the best results in all circumstances for all crops. Plant, invertebrate, and microbe responses to UV radiation have been widely studied and documented, yet uncertainties remain, particularly regarding the interplay of multiple responses and the effects of varying environmental conditions on those responses (Ballaré et al., 2011; Paul et al., 2012; Raviv and Antignus, 2004).

Conclusion

Historically, strawberry production in the Upper Midwest has been limited and low-yielding. Demand for locally produced strawberries is not currently met in this region; however, the low tunnel protected cropping system is one method shown to improve growth and performance of higher-yielding day-neutral strawberry cultivars in northern parts of the country where growing conditions are more challenging. Beyond the normal benefits furnished by growing strawberries under shelter, low tunnel production could enhance disease or insect pest management capabilities with the use of specialty plastic films designed to modify temperature, humidity, or UV and visible light transmission. This would be particularly useful for organic growers who cannot manage pests with the same set of synthetic pesticides commonly used in conventional production. It is not fully understood how these specialty plastic films could affect the insect pests specific to strawberry systems, and furthermore, how other characteristics of strawberry fruit and aspects of production could be affected. Evaluating different tunnel coverings in organic strawberry production systems in the Upper Midwest could help growers select plastics that optimize light conditions for pest management while improving biopesticide efficacy and promoting high yields and fruit quality to meet growing demand.

Chapter 1: Influence of Low Tunnel Covering on Light, Microclimate, Fruit Yield, and Fruit Quality in an Upper Midwest Organic Strawberry Production System

Introduction

Strawberries are among the most popular fruits in the United States based on total crop value and fresh sales at grocery stores (Nielsen, 2015; Zillman, 2014; USDA NASS, 2017). Most strawberry acreage in the United States is non-organic, but demand for organic strawberries is on the rise (Daugaard, 1999; Gu et al., 2017; Hoover et al., 2014). The high value placed on local agriculture, organic production, and direct market sales (Tourte et al., 2016), has led to increasing demand around the country for locally produced fruits and vegetables (Howard and Allen, 2010; Jensen and Malter, 1995; Tourte et al., 2016). Currently, the U.S. strawberry industry is concentrated in California and Florida (USDA NASS, 2016), and growers who are able to supply local strawberries in other parts of the country, especially organic, have a competitive edge in the direct to consumer market (Kadir et al., 2006a; Petran et al., 2017).

Climate plays a major role in determining regional and site suitability for strawberry production (Rysin et al., 2015). Depending on the cultivar, strawberries can be sensitive to variables such as late spring frosts, low winter minimum temperatures, and short growing seasons (Carroll et al., 2015). There are two types of commercially produced strawberry cultivars: June-bearing and day-neutral (Darrow and Waldo, 1933; Gu et al., 2017). Nationally, commercial strawberry production favors the day-neutral cultivars for their longer season and higher yield potential compared to June-bearing cultivars, but historically, day-neutral cultivars have not performed well in northern regions of the U.S. such as the Upper Midwest (Darrow and Waldo, 1933; Petran et al., 2017). However, recent developments in protected agriculture have created opportunities for producing day-neutral strawberries as annuals in some regions of the U.S. where conditions have traditionally been considered unsuitable (Hoover et al., 2014; Petran et al., 2017; Solomon et al., 2001).

Growing strawberries under protection can have many benefits. Shielded from rain and hail, fruits sustain less damage under high tunnels than in open fields

(Jett, 2007). Berries are also cleaner with less surface moisture at harvest (Karlsson and Werner, 2011). Tunnels can increase the amount of time at which strawberry plants are held at optimal growing temperatures (Kadir et al. 2006a; Rowley et al., 2011). High tunnel strawberry production has also been shown to promote earlier flowering and fruiting when compared to open field production (Kadir et al., 2006a). The more diffuse light conditions under tunnels may result in better light penetration to lower leaves, thereby increasing photosynthesis (Baeza and López, 2012; Demchak, 2009). One of the most important benefits of tunnels is disease management due to the lack of moisture accumulating on leaves in a sheltered environment (Burlakoti et al., 2014; Daugaard, 1999; Demchak, 2009).

Similar to high tunnels but relatively unexplored as a strawberry protected cropping tool are low tunnels. In the low tunnel system, strawberries are grown on raised beds with plastic mulch. Steel hoops spaced evenly down the length of a bed support a plastic covering 0.6 meters off the ground (Demchak and Hanson, 2013; Gu et al., 2017; Hoashi-Erhardt et al., 2013; Kadir et al., 2006a; Lewers et al., 2017). Low tunnels offer some unique advantages over high tunnels. Long-term high tunnel growers have identified soil compaction and quality as an issue in their systems (Demchak and Hanson, 2013). Low tunnels, which are not permanent structures, can easily be moved to new fields annually, reducing the risk of soil compaction. This also gives growers more flexibility in adjusting the scale of production from year to year. Soil-borne diseases are problematic in strawberry production, and having the ability to move tunnels in order to rotate the planting area can improve the sustainability of strawberry production systems. This is especially true in organic systems where synthetic soil fumigants cannot be used for disease management (Rysin et al., 2015). Other problems growers have observed with high tunnels include building and maintenance costs (Lewers et al., 2017), difficulties with temperature management, and loss of tunnels in extreme weather (e.g. severe winds, excessive snow) (Demchak and Hanson, 2013). Low tunnel materials may be expensive initially, but the hoops can be re-used year after year, and actual tunnel construction is relatively simple. Temperature management is fairly easy as low tunnels do not require complex venting schemes – the sides can be opened or closed manually, and some tunnel plastics incorporate ventilation holes that run the

length of the plastic. Air circulation, coupled with protection from rain, ensures that foliage, flowers and fruit remain dry for longer periods of time which can reduce the duration and frequency of disease infection periods (Carroll et al., 2015; Karlsson and Werner, 2011).

For growers interested in low tunnels, new structure options including plastics designed for specific light absorption and transmission characteristics are now commercially available (Karlsson and Werner, 2011). In northern climates with short growing seasons, non-traditional plastic materials that operate as photo-selective barriers could improve crop performance or even aid in pest and disease control in a tunnel system (Baeza and López, 2012; Karlsson and Werner, 2011; Krizek et al., 2005; Paul et al., 2005). These plastic films selectively block or absorb wavelengths of light in the infrared or ultraviolet (UV), or diffuse incoming direct beam solar radiation without inhibiting necessary transmission of photosynthetically active radiation (PAR). PAR is both visible light and the spectral range important for photosynthesis, 400-700 nm (Björn, 2015). The standard films most commonly used in horticultural production transmit lower levels of UV light, allowing little or no transmission of UV-B (280-315 nm) and reduced transmission of UV-A (315-400), but there are now other films that are completely opaque to UV (Krizek et al., 2005; Paul et al., 2005).

Changing levels of light exposure can influence many aspects of crop morphology and chemistry (Ballaré et al., 2011; Ballaré et al., 2012). Soluble solids content (including sugars) may be affected by the amount and quality of light a plant receives (Perkins-Veazie, 1995). Some studies have shown that fruit color and plant growth may be negatively affected by restricting UV exposure (Elfadly et al., 2012; Tsormpatzidis et al., 2011). And there is evidence to suggest that exposure to UV light improves crop resilience in the face of environmental stressors by way of photoreceptors that trigger critical defense mechanisms (Ballaré et al., 2012; Wargent et al., 2011). At the same time, restricting UV light can reduce the spread of diseases that affect strawberry fruit quality (Baeza and López, 2012; Karlsson and Werner, 2011; Krizek et al., 2005). While it is generally understood that UV exposure can be harmful in some ways and helpful in other ways for plants, it is unknown how

changing levels of UV exposure could impact overall growth and performance of strawberry plants under low tunnels.

The objectives of this study were to evaluate the effects of UV-blocking and UV-transmitting plastics on the light and microclimate in low tunnel environments and on fruit yield and quality. We also sought to determine whether spectral qualities through these plastics changes within one growing season. The broader context of this study is about improving the availability and quality of strawberries and sustainable production in the Upper Midwest to help growers meet demand for local, organic strawberries.

Materials and Methods

Experimental design and maintenance

This research was conducted at the Minnesota Agricultural Experiment Station (MAES) in St. Paul, Minnesota (44.996° N, 93.185° W) on USDA certified organic land in 2016 and 2017. In both years of the experiment, organic-approved practices were followed. Dormant, bare root 'Albion' strawberry plants were purchased and shipped from Nourse Farms (Whately, MA) prior to site preparation. The plants were stored in a cooler at 3.3°C for 4-5 weeks until planting. Extra plants were potted in plastic pots 12.7 cm deep and placed in cold frames for later replacement of plants that died in the field within two weeks of the first planting. The potting soil was a mix of OMRI listed Seed Starter Mix (Purple Cow Organics, LLC, Middleton, WI) and OMRI listed Black Gold Potting Mix (Sun Gro Horticulture, Agawam, MA).

Soil characteristics of the planting sites were evaluated in pre-planting soil tests conducted by the University of Minnesota Soil Testing Laboratory (Saint Paul, MN) and are described in Table 1. Soil core samples were taken from a depth of 15-20 cm at multiple sites around the field for the tests each year. Prior to planting, fields were rotovated. Raised beds were made with a model 2121-D bed shaper, and plastic mulch and drip tape were laid with a model 2133 mulch layer (Buckeye Tractor Company, Columbus Grove, OH). Each raised bed had 1.0 mil thick white on black embossed plastic mulch (Berry Plastics, Ag Resource Inc, Detroit Lakes, MN) 1.2 m wide for two rows of plants staggered 30 cm apart within rows and 36 cm between rows. A 0.6 m walkway between raised beds was covered with 3 oz. (28.35

g), 91.44 m by 0.91 m black landscape fabric (Boulder Ridge Spunbound Landscape Fabric; Central Landscape Supply, St. Cloud, MN). Planting took place from May 17-18th in 2016 and on May 15th in 2017. We constructed tunnels from the TunnelFlex Retractable Low Tunnel System (Dubois Agrinovation, Saint-Remi, Quebec, Canada), with all materials from Dubois Agrinovation except for the experimental low tunnel plastics. Galvanized steel hoops 71 cm wide by 100 cm tall were placed every 1.82 meters down the length of each plot.

We used a completely randomized design with 3 low tunnel treatments: UVT (a standard, partially ultraviolet-transmitting plastic), UVB (an ultraviolet-blocking plastic), and open (no plastic covering) replicated 4 times per treatment for a total of 12 plots. Two Lumisol experimental plastics (Visqueen, Stevenston, UK) were used as the low tunnel coverings. These plastics were 7.9 mils thick and varied in absorbance/transmittance properties. One was designed to block most light in the ultraviolet A and B ranges (UV-Blocking/UVB) and one was designed to transmit low amounts of light in the ultraviolet A and B ranges (UV-Transmitting/UVT). Prior to this experiment, these plastics were in use for one year on high tunnels at Michigan State University in East Lansing, Michigan. Each plot contained 64 plants in two staggered rows 9.75 m long and 1.2 m wide. Each plot was divided into four sections of 16 plants each, and data were collected on the 8 inner plants of each sub-section. Thus for each plot, data were collected on 32 plants of the 64 plants; the other 32 plants were designated as buffer.

The low tunnel plastic coverings were held in place on top of the steel hoops with bungee cords and were spliced on the short ends with 3 m of 1.5 mil Clear Film (Dubois Agrinovation). This made it possible to tie the ends of each tunnel to steel anchors in the ground as the Dubois Clear Film was thinner and more flexible than the experimental plastics. Greenhouse Premium Repair Tape (FarmTek, Dyersville, IA) was used to attach the different plastics together. For most of the season, the sides of each tunnel were open to allow for airflow and prevent high temperature inside the tunnels. In October, as night temperatures began to drop below 4-5°C, the tunnel sides were closed to a height of about 15 cm off the ground.

Irrigation was turned on as needed up to once per week for as long as 2 hours at a time at a rate of 10 psi or 1.55 lbs · cm⁻². Through mid-September, at the

time of irrigation, OMRI listed Organic Fish and Seaweed Fertilizer (2N-3P-1K) (Neptune's Harvest Organic Fertilizer, Gloucester, MA) was delivered to the plants up to once per week at 5 lbs N/acre (80 mL) through a 2-gallon EZ-FLO fertilizer injector (EZ-FLO Injection Systems, Inc, DripWorks, Willits, CA) connected to the drip irrigation system. This rate was based on local recommendations (Hoover et al., 2014). Weeding was done by hand as needed. Flowers and stolons were removed from the young plants up until July 1 each year to promote vegetative growth.

Light and Microclimate

Data on temperature, relative humidity, and light intensity in the Photosynthetically Active Radiation (PAR) range were collected using S-LIA-M003 Photosynthetic Light (PAR) Smart Sensors, S-THB-M008 Temp/RH Sensors, and the HOBO RX3000 Remote Monitoring Station Data Logger (Onset Computer Corp, Bourne, MA). Temperature/humidity sensors were installed at planting and placed in the center of two plots of each treatment. One PAR sensor was placed in the center of one plot of each treatment. The PAR sensors had a measurement range of 0 to 2500 $\mu\text{mol}/\text{m}^2/\text{s}$ over wavelengths from 400 to 700 nm. Data logged every 30 minutes from the beginning of the season until after the last harvest. A "Light Scout" Ultraviolet (UV) Meter (Spectrum Technologies, Inc, Aurora, IL) was used to gather data on the intensity of light in the UV range reaching the plants. The device reported intensity of ultraviolet light in the wavelength range of 250-400 nm in units of $\mu\text{mol}/\text{m}^2/\text{s}$. This data was gathered weekly, within an hour of solar noon. At each sampling, the UV meter was placed in a cup to hold it upright and perpendicular to the ground and set in the center of each raised bed for a two minute recording period. During that time, the UV intensity reading was recorded every 30 seconds and averaged.

Degradation of Plastics

At monthly intervals, a 25 cm² sample of plastic was removed from the top of each of the eight plastic tunnels. The first set of samples was taken within the first week of tunnel installation in the field and the last set of samples was taken after the last harvest. In 2016, these dates were May 22, June 22, July 22, Aug. 19, Sept. 27, and Nov. 15. In 2017, these dates were May 30, June 28, July 27, Aug. 29, Oct. 4, and Oct. 31. Each plastic sample was rinsed under deionized water to remove soil

and debris. Samples were then air dried and analyzed for absorbance using a NanoDrop 2000c Spectrophotometer (Thermo Fisher Scientific, Grand Island, NY), which reported light absorbance through the plastic in AU (Absorbance Units) at each wavelength between 190 and 840 nm. Percent light transmittance through the plastics was then calculated using the Beer-Lambert Law: $\text{Absorbance} = 2 - \log_{10}(\% \text{Transmittance})$.

Yield and Grade

Harvesting took place in the mornings between 9AM and noon. In 2016, harvest began on July 21st (65 days after planting) and was done twice per week during peak production until September 23, when it was reduced to once per week. In 2017, harvest began on July 17th (63 days after planting) and was done twice per week until September 14, when it was reduced to once per week. Harvesting ceased after the first frost that killed a majority of the strawberry flowers, on November 8th in 2016 and on October 23rd in 2017. At each harvest, all fully ripe fruit was picked. Yield was recorded on a per plot basis as the total weight in grams of harvested fruit. The total number of living plants in the experimental units of each plot was also recorded for the purpose of calculating mean yield per plant. In a portion of the harvests, the total yield of fruit was sorted and weighed by grade. USDA standards were used to group fruit into “U.S. No. 1”, “U.S. No. 2”, and all else (in this study designated as low grade or unmarketable) (USDA AMS, n.d.). For the purposes of this experiment, U.S. No. 1 and U.S. No. 2 together constituted the “marketable quality” fruit category; however, not all fruit designated as low grade or unmarketable was discarded. In many cases this fruit was still salable for local, direct market.

Fruit Color and Soluble Solids Content

Fruit color was measured with a Chroma Meter CR-400 (Konica Minolta Sensing, Inc, Ramsey, NJ). Measurements were taken from a subsample of 4 ripe, marketable fruit per plot. In 2016, sampling dates were Aug. 29 and Sept. 16. In 2017, sampling dates were Aug. 17, 24, Sept. 7, 14, Oct. 9 and 23. Measurements were taken on the surface of each berry at the point of widest diameter and recorded in terms of the Munsell Color System. The Munsell Color system is based on a three-dimensional model, which assigns a color values for three different attributes: Hue, Value, and Chroma. Hue describes the color itself (e.g. red), value describes

the lightness or darkness of the color from 0 (black) to 10 (white), and chroma describes the saturation or brilliance of the color (Munsell, 2017).

Soluble solids content was measured with a digital handheld refractometer (Spectrum Technologies, Inc) and recorded in °Brix, where 1 degree Brix equates to 1 gram of dissolved solid content per 100 grams of solution, an approximation of sugar content. Measurements were taken from a subsample of 4 ripe, marketable fruit per plot. In 2016, °Brix sampling dates were Aug. 5, 12, 18, 29, and Sept. 13. In 2017, sampling dates were Aug. 10, 24, 31, Sept. 7, Oct. 9 and 23. In cases where there weren't 4 marketable grade berries available for measuring color or soluble solids, the next highest quality, lower grade berries were used.

Statistical Analyses

All analyses were performed with R statistical software version 3.3.3. A one-way Analysis of Variance (ANOVA) was conducted with each measured factor as a function of covering treatment (UVT, UVB, and open) to determine presence of significant treatment differences at $p < 0.05$. Square-root transformations were used to correct for non-normality in UV intensity data. Pairwise comparisons were conducted using Tukey's Honest Significant Difference post-hoc test at $p < 0.05$.

Results

Light and Microclimate

As expected, in both years of the experiment, UV intensity was significantly different across treatments (Table 2). Open plots experienced the highest mean UV intensities integrated over the range of 250-400 nm, followed by UVT plots, followed by UVB plots. Open and UVT plots experienced the greatest variation in mean UV intensity, while UVB plots experienced a lower and narrower range of UV intensity. Open plots experienced the highest maximum and mean daily PAR intensities integrated over the range of 400-700 nm compared to UVT and UVB plots, for which differences were not statistically significant (Tables 3, 4). This indicates that though the UVT and UVB treatments transmitted different levels of UV intensity, they did not transmit significantly different levels of PAR intensity. Figure 1 shows trends in maximum and mean daily PAR intensities under each treatment over time. The differences between open and UVT/UVB treatments in mean daily PAR intensities

were smaller than the differences between open and UVT/UVB treatments in maximum daily PAR intensities.

Significant differences in temperature were observed in 2016 across treatments but not in 2017. In 2016, UVT and UVB plots had the highest maximum daily temperatures, followed by open plots, but in 2017, this trend was not observed (Table 5). Mean maximum daily temperatures varied by less than 3°C across treatments in both years. Differences in maximum daily temperatures were more pronounced later in the season when tunnels were closed (Figure 2). Maximum daily relative humidity levels were significantly different across treatments in both years. In 2016, UVB plots had the highest maximum daily relative humidity levels, but in 2017, open plots had the highest maximum daily relative humidity levels (Table 6). In both years, mean values of maximum daily relative humidity differed by less than 3.5% across treatments. As with temperature, differences were more pronounced late in the season when tunnels were closed (Figure 3).

Degradation of Plastics

Percent transmittance of light measured through the UVB plastics at each wavelength from 280-400 nm was relatively consistent from month to month, changing little throughout the course of a season. In contrast, percent transmittance of light measured through the UVT plastics had more variability from month to month (Figures 4a-4d). Percent transmittance increased with increasing wavelength at similar rates each month, but overall levels of transmittance varied up to about 30% between sampling dates for any given wavelength.

Yield and Grade

Season total yield per plant was significantly different across treatments in 2016, but not in 2017 (Table 7). Open plots produced the lowest mean total yield per plant in both years. In 2016, UVT plots produced significantly higher mean total yield per plant than open plots, but not significantly higher yields than UVB plots. In 2017, UVB plots produced the highest mean total yield per plant. Yield accumulated at different rates over the course of each season in 2016 and 2017 (Figure 5). In 2017, yield accumulated quickly early on in the season while in 2016, the greatest gains in yield did not occur until about 120 days after planting. UVT/UVB plots produced significantly higher proportions of marketable yield compared to open plots in both

years (Table 8). In 2016, the difference in proportion marketable yield between UVT and UVB plots was also significant, with UVT plots producing a higher proportion of marketable yield than UVB plots. Differences in marketable yield across treatments were less pronounced in 2017 than in 2016, and overall, all treatments produced higher proportions of marketable yield in 2017 than in 2016. In 2017, Verticillium wilt infection was detected and confirmed in leaf tissue analyses conducted by the University of Minnesota's Plant Pathology lab (St. Paul, MN), affecting plants at random over the entire field area. Dead plants were removed, and yield data were corrected for missing plants by calculating average yield per living plant in each plot at each harvest.

Fruit Color and Soluble Solids Content

In 2016, no statistically significant differences in color hue ($p=0.554$), value ($p=0.404$), or chroma ($p=0.164$) were found (data not shown). In 2017, the fruit color value and chroma differed significantly by treatment (Table 9). Mean fruit color value and fruit color chroma were highest in UVB plots, followed by UVT plots, followed by open plots. Degrees brix did not differ significantly by treatment in either 2016 ($p=0.58$) or 2017 ($p=0.773$) (data not shown).

Discussion

PAR and UV intensities differed across treatments as expected, but it was somewhat surprising to find only small and mostly insignificant differences in mean daily temperature and humidity levels. This was probably due to the fact that the tunnel sides remained fully open for most of the season. Had the tunnel sides been lowered to some mid-point between closed and open, we may have observed higher temperatures and humidity levels under UVT and UVB plots compared to open plots. However, day-neutral strawberries are sensitive to extreme heat due to shallow root systems, and increasing temperatures could have been detrimental (Hoover et al., 2017; Kadir et al., 2006b). Strawberries are capable of flowering and producing fruit within a wide range of temperature conditions, but 29°C is considered the upper limit at which they will flower (Haifa, 2014; Hoover et al., 2017). This upper limit was reached in 2016 as the mean maximum daily temperature over the course of the season was 29.2°C in UVT plots and 28.8°C in UVB plots. Kadir et al. (2006b) have shown that temperatures above 30°C reduce photosynthetic rate in strawberries,

and so it is unlikely that adjusting tunnel sides to increase daily temperatures would have produced any benefits beyond those benefits already achieved by covering plots.

Results from the spectrophotometer analysis of UV transmittance through plastic coverings show that the UVB plastics maintained their structural integrity, blocking UV light throughout the entire growing season. On the other hand, differences in month to month levels of UV transmittance through UVT plastics suggest some degradation or changes in the material occurred over the course of each growing season. However, because the direction of change was inconsistent over time (i.e. UV transmittance increased some months and decreased in other months), it is possible that these differences are due only to measurement inaccuracies or to variations in the material at different sample locations. These plastics were in use on high tunnels for one growing season prior to use in this study, and some sections may have had more direct light exposure than other sections, causing uneven degradation of the material across sampling sites. Environmental factors such as solar radiation, temperature, agrochemical use, and humidity can alter the chemical composition of a plastic film, undermining its mechanical and optical properties over time (Dilara and Briassoulis, 2000). Comparing results from the spectrophotometer on UV transmittance to results from the UV meter on UV intensity, we speculate that most of the UV intensity experienced under both the UVT and UVB treatments falls in the UV-A (315-400) range.

There are many factors that may have contributed to the differences in fruit yield and marketability observed in this study. Yield may have peaked at different points in the season each year due to differences in weather conditions early on. In 2016, the month of June was drier and cooler, with 2.13 cm of rainfall and a mean temperature of 19.61°C, compared to 2017, with 10.74 cm of rainfall and a mean temperature of 21.89°C (NWS, 2017). Though the intensity of PAR was lower in UVT/UVB plots compared to open plots, fruit yield and marketability were still higher in UVT/UVB plots both years, indicating that the lower threshold for PAR intensity necessary for plant growth and functioning was still met by conditions in UVT/UVB plots. Leaf-level photosynthetic measurements taken on strawberry plants in both

open and covered production systems in Maryland showed that 90% of the light-saturated photosynthetic rate occurred at a PAR of 800 $\mu\text{mol}/\text{m}^2/\text{s}$ (Condori et al., 2017). In our study, the maximum daily PAR intensity far exceeded that level across all treatments. Furthermore, even if PAR intensity levels are lower under coverings, the plastic will diffuse the PAR, resulting in better light penetration through the leaf canopy and less shadowing of lower leaves by upper leaves (Baeza and López, 2012).

The fact that UVT plots produced higher yields and a higher proportion of marketable yield compared to UVB plots in 2016, but not compared to UVB plots in 2017 suggests that the differences in UV intensity between those two treatments was not significant enough to affect fruit yield or marketability. Nechet et al. (2015), similarly found that differences in UV intensity did not promote changes in strawberry fruit production or quality in studies in Brazil. However, increases in UV can increase sporulation for certain fungal diseases which can negatively impact both yield and quality (Nechet et al., 2015; West, 2000).

Sugar content in strawberries may be affected by the amount and quality of light a plant receives (Perkins-Veazie, 1995), but the different light environments created by the treatments in this study did not result in significant differences in fruit sugar content. This is in contrast to a study by Palmieri et al. (2017), which found increases in sugar content at lower levels of UV radiation. Overall, the Brix levels observed in our study were comparable to Brix levels reported by Kallio et al. (2000) who evaluated Brix in multiple strawberry cultivars in Finland under both organic and conventional practices.

The lack of significant differences found in fruit color in 2016 may have simply been due to the small sample size that year, when color measurements were taken on just two separate dates. In 2017, when color measurements were taken on six separate dates, fruit color chroma and value were significantly higher in fruit from UVB plots compared to fruit from open plots, but not compared to UVT plots. Visual attributes are perceived by consumers as among one of the strongest determinants of purchasing choice (Moser et al., 2011), but the color differences observed in our study, though statistically significant, were small and possibly indiscernable to the naked eye. Additionally, without further analyses, we do not know whether or not

these color differences reflected significant differences in nutritional value or other quality parameters. In at least one study, UV radiation was found to speed the rate of color development, which was also correlated with an increase in fruit anthocyanin, flavonoid, and phenolic contents at harvesting (Tsormpatsidis, 2011). Temperature may also play a role. Kadir et al. (2006b) found that holding strawberry plants at low temperatures (below 20°C) increased redness of fruits. However, this effect was not observed in all cultivars.

It's important to consider that the results obtained from any experiment manipulating natural light or microclimate conditions will vary depending on the ambient environmental conditions particular to that site. Presently, use of plastic coverings to selectively block UV light in protected cropping systems may be most relevant at higher altitudes or closer to the equator where ambient UV intensity is highest. However, in the future, large-scale shifts in environmental conditions due to climate changes that alter cloud cover and snow cover, a weakening ozone layer, and various land use intensifications may increase ambient UV intensity around the world (Ballaré et al., 2011; Gigahertz-Optik, Inc. 2008; Paul et al., 2012).

In conclusion, low tunnels can improve organic strawberry production systems in Minnesota by increasing yields and fruit quality, but there are no clear advantages or disadvantages to using a UV-blocking versus a UV-transmitting plastic covering. Both plastic types maintained their spectral properties over the course of a growing season. 'Albion' day-neutral strawberries produced higher yields and higher proportions of marketable quality fruit in UVT/UVB covered plots compared to in open control plots, but it is unclear whether or not the differences in UV transmission through the plastic coverings influenced these measured variables. This study included four replicates per treatment in each year; larger sample sizes might have revealed more consistent differences in fruit yield and quality across treatments. In this study, fruit color was minimally affected by the type of covering and fruit sugar content was not affected. We were able to assess fruit quality based on a few parameters, but to more fully assess the effects of light on the fruit, it would be useful to evaluate levels of secondary metabolites as well and to evaluate more cultivars. Differences in UV transmission through plastics might have variable effects on plant growth and production from year to year depending on weather conditions.

To better understand how PAR or UV transmission through plastic coverings affects strawberry plant growth or production, it would be useful to look beyond evaluating UV and PAR intensity as a whole to focus instead on the intensity at different wavelengths within the PAR and UV spectral ranges.

Chapter 1: Tables and Figures

Table 1. Soil characteristics of planting sites in 2016 and 2017. The two sites were roughly 100 meters apart in the same field area on USDA-certified organic land in St. Paul, Minnesota.

	Year	
	2016	2017
Soil Texture	Medium (silty loam)	
Soil Organic Matter	7.4%	5.7%
pH	7.2	6.8
Bray 1 P (ppm)	100+ (very high)	
K (ppm)	300+ (very high)	
Previous crop	Edible beans	

Table 2. UV intensity ($\mu\text{mol} \cdot \text{m}^{-2} \cdot \text{s}^{-1}$) measured at weekly intervals from planting date through the last harvest in twelve plots under three covering treatments in 2016 and 2017. Mean values are integrated over 280-400 nm. Analyses were performed on square-root transformed means, but untransformed means are reported. Letters denote statistically significant differences within years by Tukey's post-hoc test at $p < 0.05$. Research was carried out on certified organic land planted with 'Albion' strawberries in St. Paul, Minnesota.

Covering		UV intensity [mean \pm SE ($\mu\text{mol} \cdot \text{m}^{-2} \cdot \text{s}^{-1}$)]			
		Year		Year	
		2016		2017	
Open		89.1 \pm 7.8 a		101.8 \pm 4.2 a	
UVT		59.1 \pm 5.2 b		59.5 \pm 2.9 b	
UVB		4.8 \pm 0.3 c		8.2 \pm 0.5 c	
ANOVA	df	F	P	F	P
Covering	2	124.2	<2*10⁻¹⁶	413.9	<2*10⁻¹⁶

Table 3. Maximum daily PAR intensity ($\mu\text{mol} \cdot \text{m}^{-2} \cdot \text{s}^{-1}$) calculated from measurements collected every 30 minutes from planting date through the last harvest in three plots under three covering treatments. PAR values are integrated over 400-700 nm. Letters denote statistically significant differences within years by Tukey's post-hoc test at $p < 0.05$. Research was carried out on certified organic land planted with 'Albion' strawberries in St. Paul, Minnesota.

Covering		Maximum daily PAR intensity [Mean \pm SE ($\mu\text{mol} \cdot \text{m}^{-2} \cdot \text{s}^{-1}$)]			
		Year		Year	
		2016		2017	
Open		1669.0 \pm 42.2 a		1669.5 \pm 50.8 a	
UVT		1233.8 \pm 47.0 b		1272.1 \pm 42.7 b	
UVB		1249.3 \pm 37.2 b		1210.0 \pm 40.8 b	
ANOVA	df	F	P	F	P
Covering	2	34.07	1.6*10⁻¹⁴	30.7	5.34*10⁻¹³

Table 4. Mean daily PAR intensity ($\mu\text{mol} \cdot \text{m}^{-2} \cdot \text{s}^{-1}$) calculated from measurements collected every 30 minutes from planting date through the last harvest in three plots under three covering treatments. PAR values are integrated over 400-700 nm. Letters denote statistically significant differences within years by Tukey's post-hoc test at $p < 0.05$. Research was carried out on certified organic land planted with 'Albion' strawberries in St. Paul, Minnesota.

Mean daily PAR intensity [Mean \pm SE ($\mu\text{mol} \cdot \text{m}^{-2} \cdot \text{s}^{-1}$)]					
		Year			
Covering		2016		2017	
	Open	416.3 \pm 15.3 a		411.3 \pm 17.0 a	
	UVT	285.6 \pm 13.8 b		295.6 \pm 12.6 b	
	UVB	295.1 \pm 12.4 b		286.6 \pm 12.2 b	
ANOVA	df	F	P	F	P
Covering	2	27.57	4.93*10⁻¹²	24.27	1.38*10⁻¹⁰

Table 5. Maximum daily temperature ($^{\circ}\text{C}$) calculated from measurements collected every 30 minutes from planting date through the last harvest in six plots under three covering treatments in 2016 and 2017. Letters denote statistically significant differences within years by Tukey's post-hoc test at $p < 0.05$. Research was carried out on certified organic land planted with 'Albion' strawberries in St. Paul, Minnesota.

Maximum daily temperature [Mean \pm SE ($^{\circ}\text{C}$)]					
		Year			
Covering		2016		2017	
	Open	26.6 \pm 0.4 b		26.3 \pm 0.4	
	UVT	29.2 \pm 0.4 a		26.8 \pm 0.4	
	UVB	28.8 \pm 0.4 a		27.1 \pm 0.4	
ANOVA	df	F	P	F	P
Covering	2	13.55	1.57*10⁻⁶	1.119	0.327

Table 6. Maximum daily relative humidity (%) calculated from measurements collected every 30 minutes from planting date through the last harvest in six plots under three covering treatments. Letters denote statistically significant differences within years by Tukey's post-hoc test at $p < 0.05$. Research was carried out on certified organic land planted with 'Albion' strawberries in St. Paul, Minnesota.

Maximum daily relative humidity [Mean \pm SE (%)]					
		Year			
Covering		2016		2017	
	Open	95.5 \pm 0.3 b		95.9 \pm 0.3 a	
	UVT	95.8 \pm 0.2 ab		92.4 \pm 0.4 b	
	UVB	96.3 \pm 0.2 a		93.6 \pm 0.3 b	
ANOVA	df	F	P	F	P
Covering	2	2.825	0.0598	26.19	1.17*10⁻¹¹

Table 7. Season total strawberry fruit yield per plant (g) calculated from season total yield per plot divided by total number of living plants per plot in twelve plots under three covering treatments. Letters denote statistically significant differences within years by Tukey's post-hoc test at $p < 0.05$. Research was carried out on certified organic land planted with 'Albion' strawberries in St. Paul, Minnesota.

		Yield per plant [mean \pm SE (g)]			
		Year			
Covering		2016		2017	
	Open	519.9 \pm 31.3 b		557.1 \pm 27.6	
	UVT	668.6 \pm 25.4 a		604.1 \pm 33.8	
	UVB	570.1 \pm 36.6 ab		654.4 \pm 32.7	
ANOVA	df	F	P	F	P
Covering	2	5.789	0.00579	2.388	0.103

Table 8. Proportion marketable strawberry fruit yield in twelve plots under three covering treatments measured as sum of grade 1 and grade yield (g) divided by total yield (g), analyzed across eight sampling dates in 2016 and sixteen sampling dates in 2017. Letters denote statistically significant differences within years by Tukey's post-hoc test at $p < 0.05$. Research was carried out on certified organic land planted with 'Albion' strawberries in St. Paul, Minnesota.

		Proportion marketable yield [mean \pm SE (g)]			
		Year			
Covering		2016		2017	
	Open	0.42 \pm 0.02 c		0.67 \pm 0.02 b	
	UVT	0.71 \pm 0.01 a		0.78 \pm 0.02 a	
	UVB	0.55 \pm 0.02 b		0.78 \pm 0.01 a	
ANOVA	df	F	P	F	P
Covering	2	49.61	<2*10⁻¹⁶	12.62	4.14*10⁻⁶

Table 9. Strawberry fruit color value and chroma, as described by the Munsell scale, in twelve plots under three covering treatments analyzed across six sampling dates in 2017. Value can range from 1 (black) to 10 (white). Chroma can range from 0 (no saturation) to 12 (maximum saturation). Letters denote statistically significant differences by Tukey's post-hoc test at $p < 0.05$. Research was carried out on certified organic land planted with 'Albion' strawberries in St. Paul, Minnesota.

Covering		Color value [mean \pm SE]		Color chroma [mean \pm SE]	
	Open	3.2 \pm 0.03 b		7.5 \pm 0.08 b	
	UVT	3.3 \pm 0.03 ab		7.7 \pm 0.08 ab	
	UVB	3.4 \pm 0.05 a		7.8 \pm 0.09 a	
ANOVA	df	F	P	F	P
Covering	2	2.904	0.0565	4.332	0.014

Figure 1. Maximum and mean daily PAR intensities ($\mu\text{mol} \cdot \text{m}^{-2} \cdot \text{s}^{-1}$) measured in three plots under three covering treatments in 2016 and 2017. PAR values are integrated over 400-700 nm. Points indicate daily maximum and mean readings; lines displayed are local regression trend lines. In each graph, the upper set of lines and points are maximum daily PAR intensities and the lower set of lines and points are mean daily PAR intensities. Research was carried out on certified organic land planted with 'Albion' strawberries in St. Paul, Minnesota.

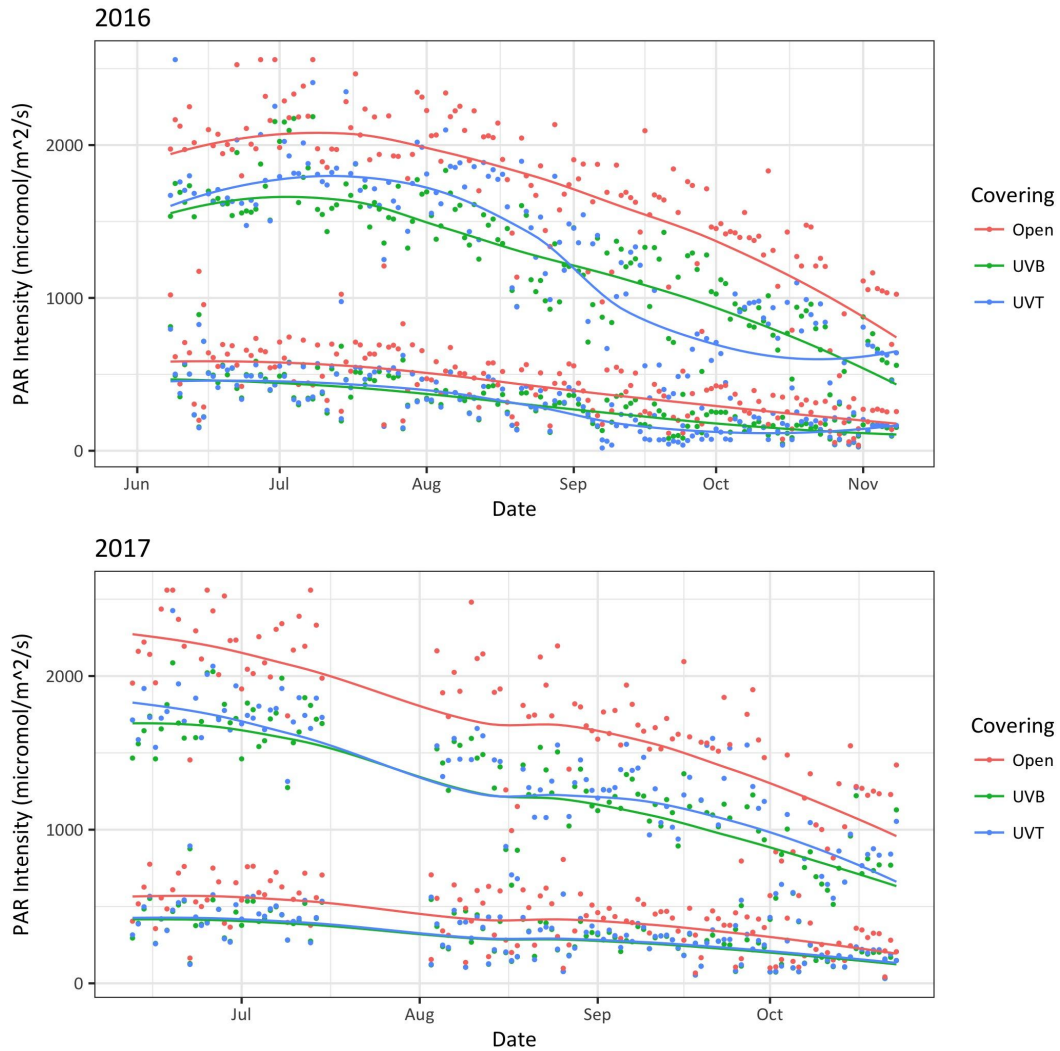


Figure 2. Maximum daily temperature (°C) in six plots under three covering treatments in 2016 and 2017. Points indicate daily maximum readings averaged between the 2 reps of each covering type; lines displayed are local regression trend lines. Research was carried out on certified organic land planted with 'Albion' strawberries in St. Paul, Minnesota.

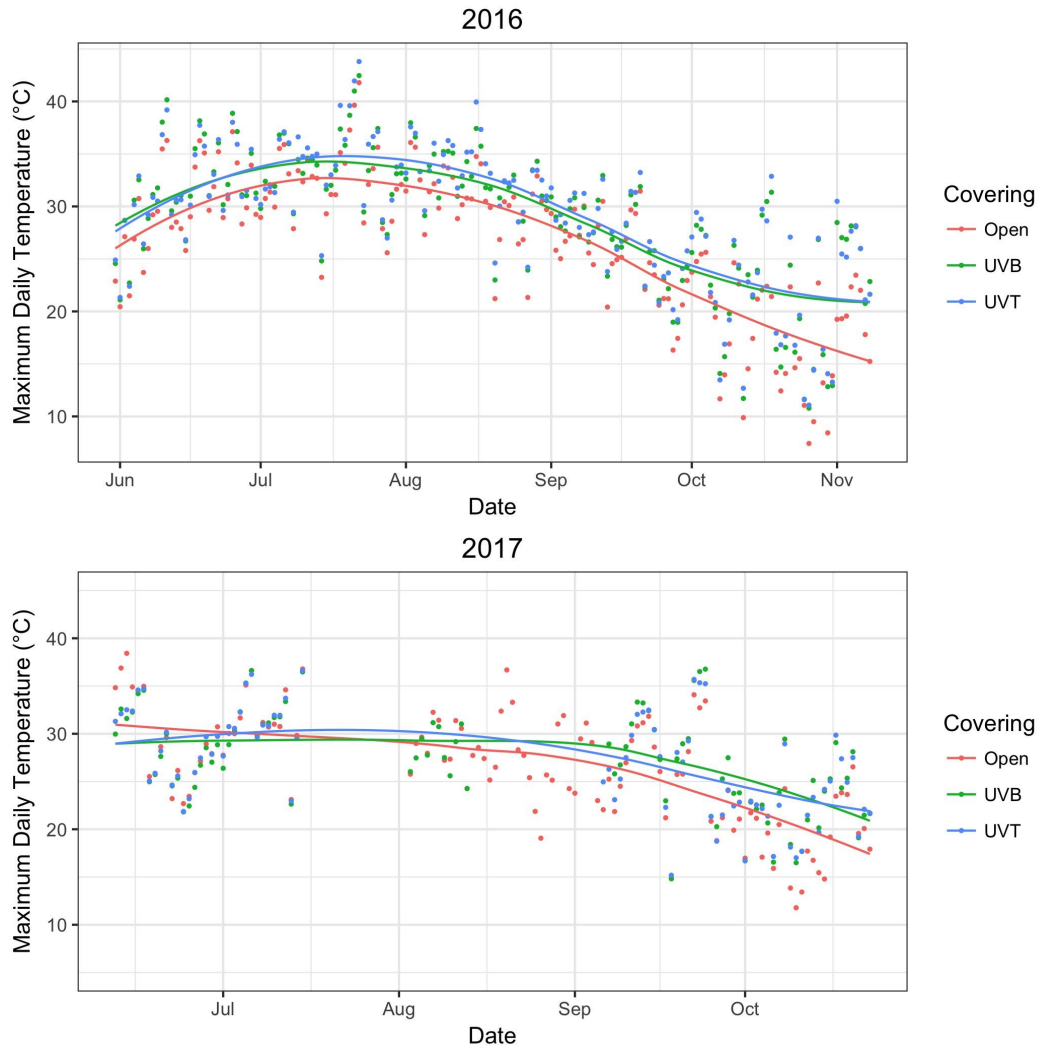
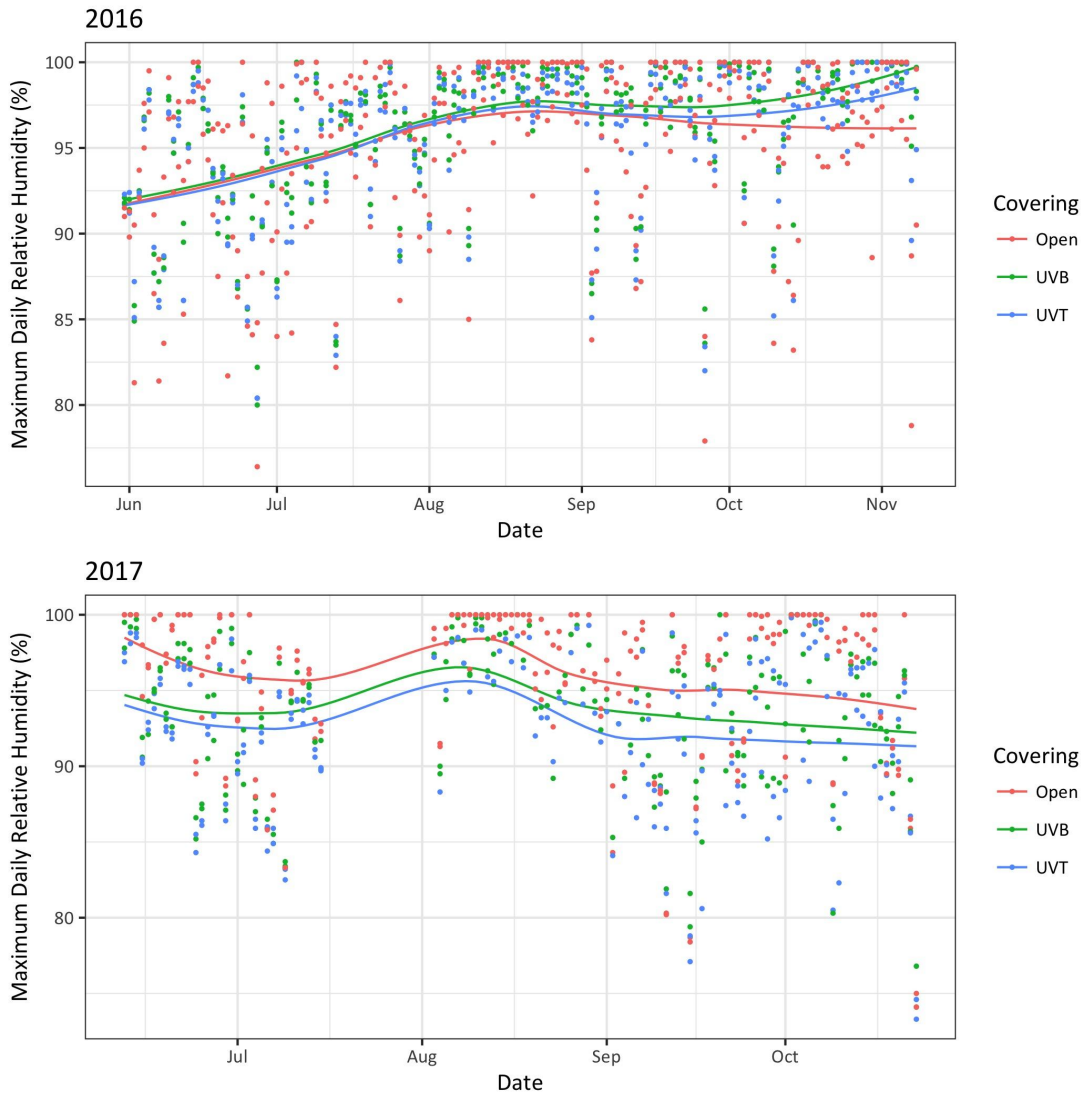
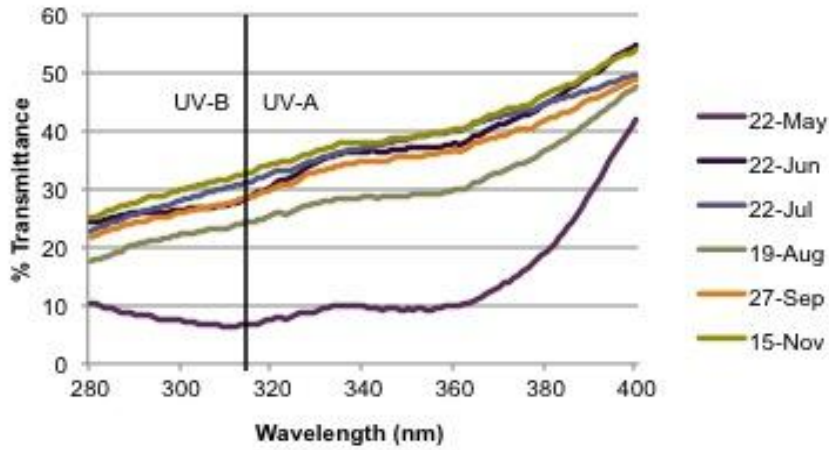


Figure 3. Maximum daily relative humidity (%) in six plots under three covering treatments in 2016 and 2017. Points indicate daily maximum readings averaged between the 2 reps of each covering type; lines displayed are local regression trend lines. Research was carried out on certified organic land planted with 'Albion' strawberries in St. Paul, Minnesota.

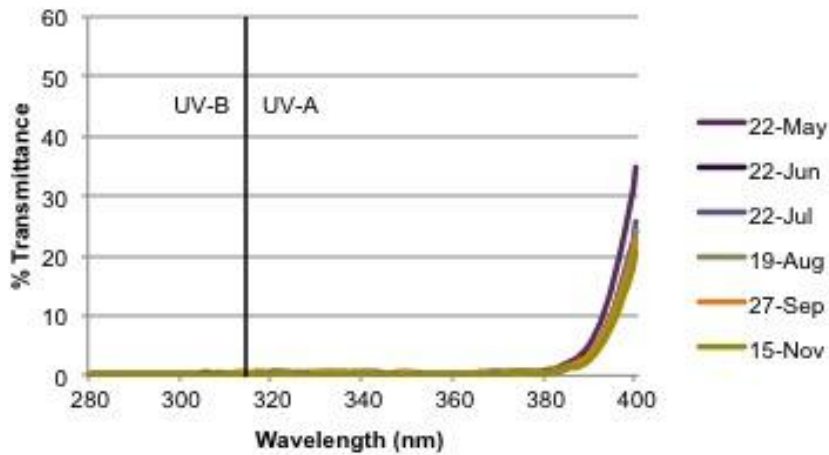


Figures 4a-4d. Percent UV transmittance in UV-B (280-315 nm) and UV-A (315-400 nm) spectral ranges through two types of plastic films used as two covering treatments in 2016 and 2017 at different sampling dates. Each graph represents results averaged at each wavelength across 4 reps of a treatment within years. Graphs for 2017 continue on the next page. Research was carried out on certified organic land planted with 'Albion' strawberries in St. Paul, Minnesota.

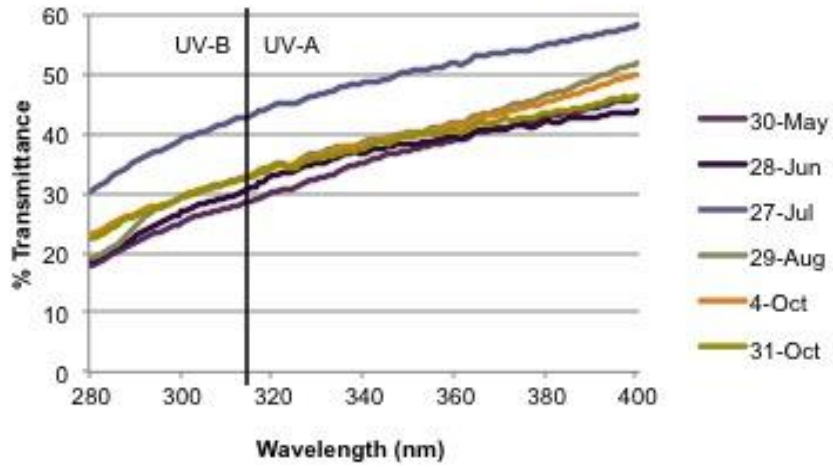
4a.
2016 UVT



4b.
2016 UVB



4c.
2017 UVT



4d.
2017 UVB

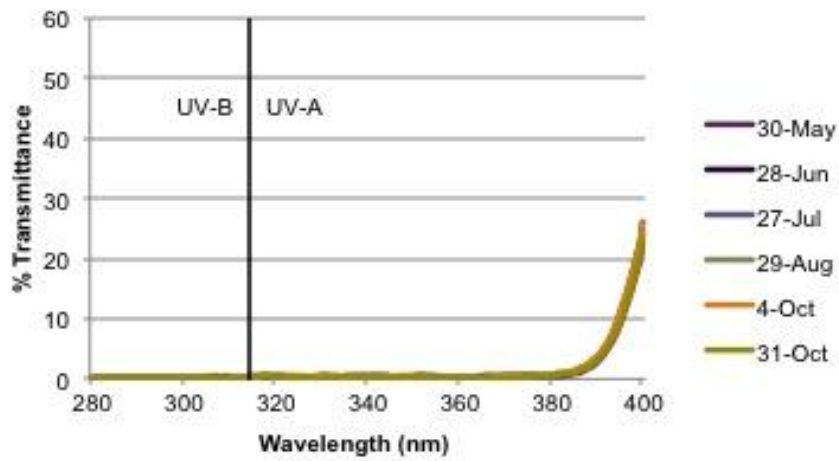
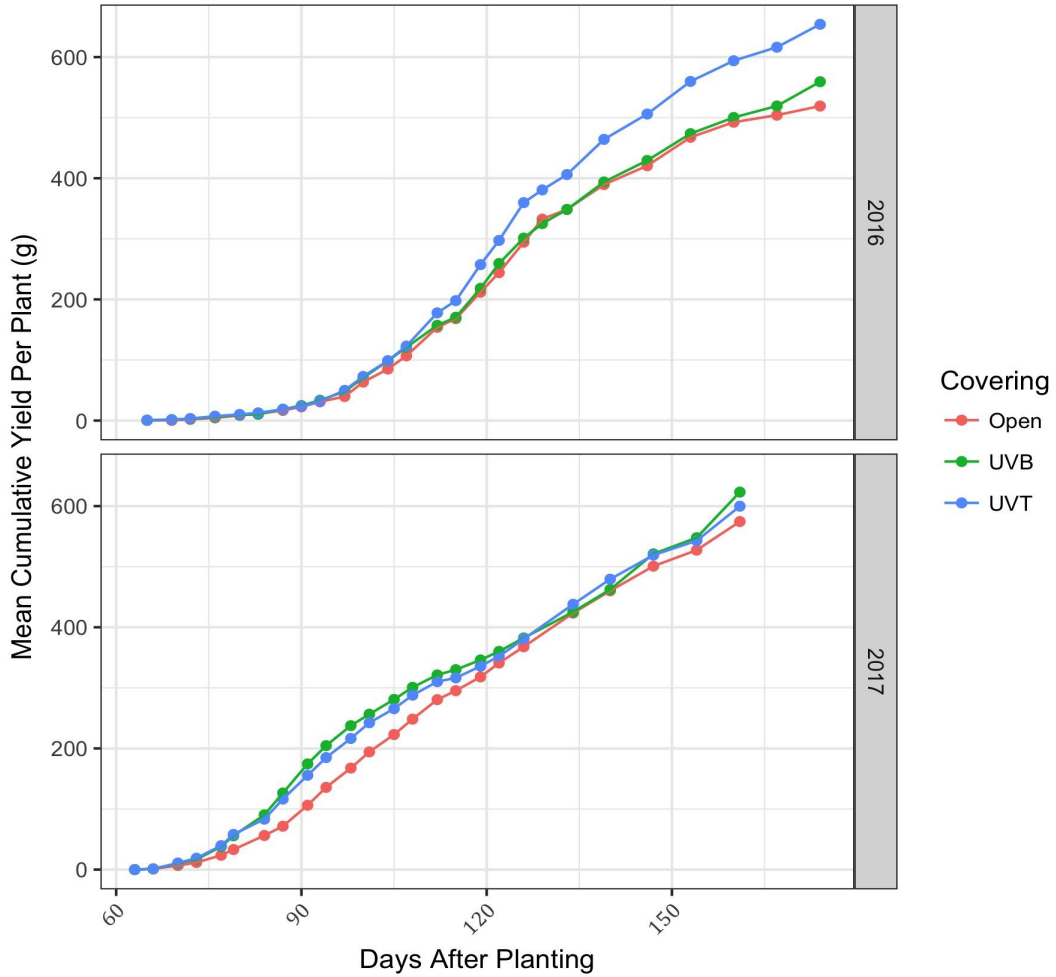


Figure 5. Mean cumulative strawberry fruit yield per plant (g) in twelve plots under three covering treatments in 2016 and 2017. Each point is the mean cumulative yield per plant at that point in time (days after planting), calculated from total yields per plot divided by total number of living plants per plot. Research was carried out on certified organic land planted with ‘Albion’ strawberries in St. Paul, Minnesota.



Chapter 2: Influence of Low Tunnel Covering Type on Insect Pest Presence and Biopesticide Efficacy in Organic, Day-Neutral Strawberries

Introduction

Among fruits, strawberries (*Fragaria ×ananassa* Duchesne) are one of the top-selling organic products in the U.S. (USDA NASS, 2015). Consumer concern over pesticide residues on conventionally grown fruits and vegetables has contributed to an increasing demand for organic products (Daugaard, 1999; Hoover et al., 2014). However, growing strawberries organically can be challenging as they are highly susceptible to insect pests and diseases (Andrade et al., 2016; Nes et al., 2017). Moreover, organic farmers must rely first on preventive and cultural control methods before resorting to use of a limited set of pest control substances certified for organic use (Caldwell et al., 2013).

The range of strawberry insect pests includes many generalist herbivores with alternate wild and cultivated hosts, making control or elimination difficult. The most important pests of strawberries globally are insects in the Miridae family (capsid bugs) (Solomon et al., 2001). Among them, *Lygus lineolaris* (tarnished plant bug) is a significant pest of strawberries in Minnesota (Wold and Hutchison, 2003a). These insects are piercing, sucking insects that feed on the achenes of developing fruits, rendering otherwise good fruit unmarketable by causing distinctive deformities referred to as “cat-faced” or “button” berries (Carroll et al., 2015; Day and Hoelmer, 2012; Solomon et al., 2001). Compared to June-bearing cultivars, day-neutral strawberry cultivars are particularly vulnerable to *L. lineolaris* due to their extended flowering and fruiting period later in the growing season when *L. lineolaris* is more prevalent (Carroll et al., 2015).

Tetranychus urticae (two-spotted spider mite) is another primary pest of strawberries in the U.S. and globally (Demchak and Hanson, 2013; Solomon et al., 2001). Mites begin to feed on the leaves of strawberry plants in early spring in the upper midwest. With heavy infestation, damage can decrease overall rates of photosynthesis and transpiration, leading to sparse new growth and reduced quality and quantity of fruit (Carroll et al., 2015; Livinali et al., 2014; Wold and Hutchison, 2003b).

Drosophila suzukii, commonly known as the spotted wing drosophila or SWD, is a more recent pest in strawberries, first detected in the U.S. in 2008 (Goodhue et al., 2011). It has since become one of the most serious pests in soft fruit, including raspberries, blueberries, grapes, blackberries, and cherries, as well as strawberries (Gong et al., 2016). Day-neutral strawberries are especially at risk from infestations due to crop phenology that overlaps with high *D. suzukii* population density (Demchak and Hanson, 2013; Tourte et al., 2016).

High and low tunnel protected agriculture systems are popular among growers for season extension and improving crop quality (Demchak, 2009; Jensen and Malter, 1995; USDA NRCS, n.d.), and it is possible that these systems could also be used to aid with insect pest management (Baeza and López, 2012; Krizek et al., 2005). An expanding assortment of tunnel structural designs and novel plastic covering materials fabricated for specific light absorption and transmission properties are now commercially available (Karlsson and Werner, 2011). These plastic films selectively block or absorb light in specific wavelengths in the infrared or ultraviolet (UV), or diffuse incoming direct beam solar radiation without inhibiting necessary transmission of photosynthetically active radiation (PAR). PAR is both visible light and the spectral range important for photosynthesis, 400-700 nm (Björn, 2015). The plastic films most commonly used in horticultural production transmit lower levels of UV light, allowing little or no transmission of UV-B (280-315 nm) and reduced transmission of UV-A (315-400 nm). But there are now also plastic films available that are completely opaque to UV (Krizek et al., 2005; Paul et al., 2005). UV light plays a significant role in insect vision and flight activity; blocking UV with a specialty plastic covering could therefore be used to disrupt movement and activity of insect pests under tunnels (Antignus et al., 1996; Dáder et al., 2015; Díaz et al., 2006; Paul et al., 2012).

Tunnel plastic coverings can also provide indirect pest management by creating more favorable conditions for biopesticides (Parikka and Tuovinen, 2014; Solomon et al., 2001). Biopesticides contain one of three types of ingredients: plant-incorporated protectants, naturally occurring substances that control pests by non-toxic mechanisms, or microbial agents (EPA, 2016). They represent appealing alternatives to synthetic chemical pesticides as they pose less of a threat to human

health and carry fewer environmental risks. Biopesticides are generally biodegradable and more specific to the target pest species (Gupta and Dikshit, 2010). Fruit growers must be careful to meet maximum residual level (MRL) restrictions on pesticides on harvested fruit, and many biopesticides do not have residue restrictions. Biopesticides can also be included in a spray rotation with other pesticides over the course of a season to reduce the risk of pests developing resistance to one or more products (Fanning et al., 2017; Haviland and Beers, 2012). Additionally, some biopesticides are certified for use in organic production.

One of the more effective biopesticides commonly used in berry production is spinosad (Bruck et al., 2011; Fanning et al., 2017), a fermentation product of the soil bacterium actinomycete *Saccharopolyspora spinosa* (Mertz and Yao, 1990). Several studies have demonstrated its effectiveness for control of *T. urticae* and *D. suzukii* (Bruck et al., 2011; Fanning et al., 2017; Ismail et al., 2007; Pavlova et al., 2017; Van Leeuwen et al., 2005). Entomopathogenic (insect pathogen) fungal-based microbial biopesticides may also effectively manage some insect pest species. These fungi are able to infect an insect at all life stages by penetrating the cuticle and invading the body of the insect host, though nymphal stages may be more resilient to attack by the fungus if they molt before infection takes hold (Arthurs et al., 2013; Cory and Hoover, 2006; Dara et al., 2016; Zou et al., 2014). Spores of the fungi can be extracted and formulated into a sprayable product for crop application (Caldwell et al., 2013). *Beauveria bassiana*, a naturally occurring entomopathogenic fungus found in soils worldwide (Caldwell et al., 2013), is considered to have high insecticidal potential for the control of *L. lineolaris* (Portilla et al., 2017; Sabbahi et al., 2008). It has also caused mortality in *D. suzukii* under laboratory conditions (Cossentine et al., 2016). Another entomopathogenic fungus, *Isaria fumosorosea*, has not been extensively studied as a control agent in berry crop production systems but was shown to successfully infect *D. suzukii* in a laboratory environment (Cossentine et al., 2016). In other crops, it was shown to significantly reduce populations of *Bemisia tabaci* (sweetpotato whitefly) in the laboratory (Zou et al., 2014) and populations of *Scirtothrips dorsalis* (chilli thrips) in a greenhouse environment (Arthurs et al., 2013). Spinosad, *B. bassiana*, and *I. fumosorosea* are the active

ingredients in the organic biopesticide products Entrust SC Naturalyte Insect Control, Mycotrol WPO, and PFR-97 20% WDG, respectively.

Microbial biopesticides rely on living microbial agents or their byproducts as the active ingredient rather than synthetically derived chemicals, as most conventional pesticides do. Because of this, certain abiotic factors such as light quality and intensity can influence their efficacy and persistence in the environment (Arthurs et al. 2013; Cory and Hoover, 2006; Ray and Hoy, 2014). Understanding the rate at which a biopesticide degrades in the environment is important to appropriately schedule sprayings. Greater persistence is desirable for effective control of many insect species as eggs could hatch days to weeks after a biopesticide treatment (Van Leeuwen et al., 2005). Use of specialty plastic coverings to filter light could theoretically be used to improve persistence of microbial biopesticides in the field (Behle et al., 2011). More information on these tools and methods could help low tunnel organic strawberry growers select plastics to facilitate pest management and biopesticide efficacy while optimizing production to meet growing demand. While there are effective synthetic pesticides available for management of certain strawberry insect pests for conventional growers, organic growers depend on the development of alternative strategies for strawberry insect pest management (Fernandes et al., 2012; Marques-Francovig et al., 2014).

The objectives of this study were to evaluate how low tunnel coverings that transmit different amounts of UV light influence the presence of *L. lineolaris* and *T. urticae* in an organically managed strawberry production system, as well as the effectiveness of the organic microbial-based biopesticides Entrust SC Naturalyte, Mycotrol WPO, and PFR-97 20% WDG for control of *D. suzukii* in semi-field bioassays.

Materials and Methods

Experimental design and maintenance

This research was conducted at the Minnesota Agricultural Experiment Station (MAES) in St. Paul, Minnesota (44.996° N, 93.185° W) on USDA certified organic land in 2016 and 2017. In both years of the experiment, organic-approved practices were followed. Dormant, bare root 'Albion' strawberry plants were purchased and shipped from Nourse Farms (Whately, MA) prior to field preparation.

The plants were stored in a cooler at 3.3°C for 4-5 weeks until planting. Extra plants were potted in plastic pots 12.7 cm deep and placed in cold frames for later replacement of plants that died in the field within two weeks of the first planting. The potting soil was a mix of OMRI listed Seed Starter Mix (Purple Cow Organics, LLC, Middleton, WI) and OMRI listed Black Gold Potting Mix (Sun Gro Horticulture, Agawam, MA).

Fields were rotovated prior to planting. Raised beds were made with a model 2121-D bed shaper, and plastic mulch and drip tape were laid with a model 2133 mulch layer (Buckeye Tractor Company, Columbus Grove, OH). Each raised bed had 1.0 mil thick white on black embossed plastic mulch (Berry Plastics, Ag Resource Inc, Detroit Lakes, MN) 1.2 m wide for two rows of plants staggered 30 cm apart within rows and 36 cm between rows. A 0.6 m walkway between raised beds was covered with 3 oz. (28.35 g), 91.44 m by 0.91 m black landscape fabric (Boulder Ridge Spunbound Landscape Fabric; Central Landscape Supply, St. Cloud, MN). Planting took place from May 17-18th in 2016 and on May 15th in 2017. We constructed tunnels from the TunnelFlex Retractable Low Tunnel System (Dubois Agrinovation, Saint-Remi, Quebec, Canada), with all materials from Dubois Agrinovation except for the experimental low tunnel plastics. Galvanized steel hoops 71 cm wide by 100 cm tall were placed every 1.82 meters down the length of each plot.

We used a randomized split-plot design, assigning low tunnel covering treatment to main plots and biopesticide treatment to split plots. Each of the 12 main plots was assigned 1 of 3 low tunnel covering treatments: UVT (a standard, partially ultraviolet-transmitting plastic), UVB (an ultraviolet-blocking plastic), or open (no plastic cover). Two Lumisol experimental plastics (Visqueen, Stevenston, UK) were used for the two covered treatments. These plastics were 7.9 mils thick and varied in UV absorbance/transmittance properties. One type was designed to block most light in the ultraviolet range (280-400 nm) (UV-Blocking/UVB) and one type was designed to transmit low amounts of light in the ultraviolet range (UV-Transmitting/UVT). Prior to this experiment, these plastics were in use for one year on high tunnels at Michigan State University in East Lansing, Michigan. Each main plot contained 64 plants evenly spaced across 9.75 m of length and 1.2 m of width. The low tunnel

plastic coverings were held in place on top of the steel hoops with bungee cords and were spliced on the short ends with 3 m of 1.5 mil Clear Film (Dubois Agrinovation). This made it possible to tie the ends of each tunnel to steel anchors in the ground as the Dubois Clear Film was thinner and more flexible than the experimental plastics. Greenhouse Premium Repair Tape (FarmTek, Dyersville, IA) was used to attach the different plastics together.

Main plots were split lengthwise into 4 equal sections, with each section assigned one of 4 biopesticide treatments: Mycotrol WPO (*Beauveria bassiana* Strain GHA, 22.0% AI, 4.41×10^{10} viable spores/g; BioWorks, Inc, Victor, NY) PFR-97 20% WDG (*Isaria fumosorosea* Apopka Strain 97, 20.0% AI, 1×10^9 CFU/g; Certis USA, LLC, Columbia, MD), Entrust SC Naturalyte Insect Control (Spinosad, 22.5% AI; Dow AgroSciences, LLC, Indianapolis, IN), or water. This made for a total of 48 covering:biopesticide replication units or split plots. Of the 16 plants in each split plot, data were only collected on the interior 8 plants to reduce possible interaction effects between biopesticides. Plants outside these experimental zones were designated as buffer.

Irrigation was turned on as needed up to once per week for as long as 2 hours at a time at 10 psi or $1.55 \text{ lbs} \cdot \text{cm}^{-2}$. Through mid-September, at the time of irrigation, OMRI listed Organic Fish and Seaweed Fertilizer (2N-3P-1K) (Neptune's Harvest Organic Fertilizer, Gloucester, MA) was delivered to the plants up to once per week at 5 lbs N/acre (80 mL) through a 2-gallon EZ-FLO fertilizer injector (EZ-FLO Injection Systems, Inc, DripWorks, Willits, CA) connected to the drip irrigation system. Weeding was done by hand, as needed. Flowers and stolons were removed from the young plants up until July 1 to promote crown growth and prevent early fruiting. For most of the season, the sides of each tunnel were left fully opened to allow for plenty of airflow and prevent overheating inside the tunnels. In October, as night temperatures began to drop below $4\text{-}5^\circ\text{C}$, the tunnel sides were closed to a height of about 15 cm above the ground.

Leaf Sampling and Bioassays

The bioassay design was modified from methods described in Van Timmeren and Isaacs (2013). Each biopesticide was sprayed in one split plot per main plot, as dictated by the experimental design. We used a single nozzle boom CO₂ backpack

sprayer (Bellspray, Inc, Opelousas, LA). Biopesticide treatments were done first thing in the morning or late in the afternoon, no less than 24 hours before the next scheduled harvest, with wind speeds below 7 mph, dry weather conditions, and no rain in the forecast for 48 hours. Two liters of water were run through the sprayer between applications of different biopesticides to clean the sprayer and avoid contamination of the next product. Application rates were calculated based on highest recommended levels of use for each product on strawberries in the field. These rates were as follows (rates are reported here in either units of spray volume or units of spray area based on how they are described on the pesticide labels). Mycotrol: 359 grams/hundred liters of spray volume, PFR: 2.24 kg/hectare of spray area, Entrust 140 grams/hectare of spray area. Between the July and September bioassay experiments, the application rate of Entrust was doubled in response to acquiring updated information on recommended application rates. Between the July and September experiments, the application rate of Mycotrol was halved in response to possible phytotoxicity observed on plants sprayed with the product. Total spray volume was 1.5 liters per product per treatment, determined as the amount required to achieve full leaf coverage across all split plots of a given treatment.

In 2016, biopesticide treatments were done on June 29 and on July 29. In 2017, biopesticide treatments were done on June 30, July 28, August 23, and September 11. Following the July and September treatments in 2017, leaf samples were collected to conduct bioassay experiments with *Drosophila suzukii* (spotted wing drosophila). Bioassays were not conducted after the June treatment because at that time, plants were still too small to withstand destructive sampling. Of July, August, and September, July and September were selected for bioassays as the two treatment dates with the greatest probable difference in ambient environmental conditions, being farthest apart in time. *Drosophila suzukii* was selected based on its relevance to berry producers as a significant invasive insect pest species.

For the bioassays, one leaf sample (three leaflets) was randomly collected from each split plot 0 days after treatment (2-4 hours after spraying, when leaves had completely dried), 1 day after treatment, and 3 days after treatment. Bioassay chambers were assembled immediately after collecting the leaf samples. Each chamber contained one leaf sample held in a flower pik with water, one cup with 5

mL of fly diet prepared according to Dalton et al. (2011), and 5 male and 5 female adult *D. suzukii*. The fly specimens provided for this experiment were housed in growth chambers at 25°C and 47% relative humidity on a 16:8 (light:dark) photoperiod.

Bioassay chambers were stored at 25°C in a windowless laboratory to slow any further light-induced degradation of the biopesticides on the leaves. Percent fly mortality in each chamber was recorded at several intervals between 1 and 7 days after exposure of the flies to the treated leaves. The purpose of this experimental design was to provide a way to evaluate not only the efficacy of the biopesticides themselves but also the efficacy of each biopesticide relative to time in the field under the three different covering treatments. Table 1 shows the sequence of steps involved with each bioassay experiment.

Insect Monitoring

Throughout both growing seasons, traps baited with Scentry lures (Scentry Biologicals, Inc, Billings, MT) were placed around the field site to monitor for *D. suzukii*. In 2016, traps were removed after the first captures of both male and female *D. suzukii* early in the season on June 30, prior to the first harvest. In 2017, traps were checked and replaced in the field at weekly intervals throughout the season. A floatation test to look for *D. suzukii* larvae was performed on a sub-sample of 4 harvested fruits from each split plot, once in September and once in October.

The two insect pests *Tetranychus urticae* (two-spotted spider mite) and *Lygus lineolaris* (tarnished plant bug) were monitored in the field, collecting data from a subsample of 4 plants within each 8-plant split plot. *Lygus lineolaris* were tallied individually as adults or nymphs. In 2016, *T. urticae* were counted as present or absent based on one mite per leaf per plant. In 2017, *L. lineolaris* were again tallied individually, but *T. urticae* presence was recorded as “none,” “low,” “medium,” or “high” to signify finding 0, 1-4, 5-19, or 20+ mites on one leaflet of one leaf. These ranges were selected based on the economic thresholds for control of *T. urticae* where “low” corresponds to an infestation level below thresholds at any time in the season, “medium” corresponds to an infestation level above threshold for early season but at or below threshold for later in the season (during harvest, strawberries are more tolerant to mite feeding), and “high” corresponds to infestation levels above

threshold for any time of the season (UC IPM, 2017). Monitoring in 2016 was done on July 1, July 6, July 29, August 1, August 4, August 11, August 24, and September 30. Monitoring in 2017 was done on June 14, June 20, June 27, July 6, July 13, July 21, July 26, August 4, August 9, August 25, August 29, September 12, and October 18. Monitoring took place between the hours of 10 AM and 2 PM when *L. lineolaris* and *T. urticae* seemed most active.

Statistical Analyses

All analyses were performed with R statistical software version 3.3.3. Two-way ANOVAs were performed on bioassay mortality data, with covering treatment as the main plot factor and biopesticide treatment as the split plot factor. Pairwise comparisons were conducted using Tukey's Honest Significant Difference post-hoc test at $p < 0.05$. An ANOVA was performed on a logistic regression model of the presence/absence *T. urticae* survey data from 2016 and a Chi-square test of independence was conducted on the *T. urticae* survey data from 2017. A Kruskal-Wallis rank sum test was conducted on the *L. lineolaris* survey data.

Results

Leaf Sampling and Bioassays

Following the July biopesticide treatment in the field and associated bioassay trials in the lab, *D. suzukii* mortality rates were significantly different across biopesticide treatments for the Day 0 and Day 1 trials, but not for the Day 3 trials (Table 2). A higher percentage of flies exposed to leaves sprayed with Entrust died compared to flies exposed leaves sprayed with any other biopesticide treatments. In the Day 0 trial, *D. suzukii* mortality rates were also significantly different between male and female flies. On average, 22.9% of males died after seven days of exposure compared to only 16.6% of females. Mortality rates were not significantly different among covering treatments, but there was a statistically significant interaction effect between covering and biopesticide treatments on the Day 1 trial (Table 3). In both the open and the UVB covering treatments, Entrust produced higher mortality rates than any of the other biopesticide treatments.

Following the September biopesticide treatment in the field and associated bioassay trials in the lab, *D. suzukii* mortality rates were again significantly different among biopesticide treatments for the Day 0 and Day 1 trials, but not the Day 3 trials

(Table 4). A higher percentage of flies exposed to leaves sprayed with Entrust died compared to flies exposed to any of the other biopesticide treatments. In contrast to the results from the July bioassay trials, *D. suzukii* mortality rates did not differ significantly between males and females. Percent mortality for total files (male plus female) at each of the sample intervals after exposure are summarized in Figures 1 and 2. By the Day 3 trials, percent mortality dropped significantly across all biopesticide treatments. In both the July and the September trials, only Entrust resulted in total mortality rates greater than 20%.

Insect Monitoring

In 2016, trap captures indicated male and female adult *D. suzukii* were present at the field site before the first harvest in July (data not shown). In 2017, traps captured male and female adults at each sampling date between July 5 and October 18, encompassing the full duration of the harvest season (Figure 3). The highest numbers of *D. suzukii* flies were found on July 12 and September 6. Larvae were observed in fruit at multiple harvests throughout the 2016 and 2017 seasons; however, no larvae were found in either of two floatation tests on sampled fruit in 2017.

In 2016, the probability of observing *T. urticae* was not significantly different across covering or biopesticide treatments (Table 5). Figures 4 and 5 show the predicted probability of observing *T. urticae* in field plots over time under each covering treatment and each biopesticide treatment. Though not statistically significant, the UVT treatment appears to have a slightly lower probability of *T. urticae* presence compared to the open and UVB treatments (Figure 4). The PFR biopesticide treatment appears to have a slightly higher probability of *T. urticae* presence compared to the other biopesticide treatments (Figure 5). In 2017, a similar trend was observed. Infestation levels of *T. urticae* did not differ significantly across covering treatments (Table 6), but they did differ significantly by biopesticide treatment (Table 7). Table 8 shows multiple pairwise comparisons of levels of *T. urticae* in plots receiving different biopesticide spray treatments, with the difference between Entrust and PFR most significant. Though not statistically significant, Figure 6 appears to show a distribution of infestation levels weighted more toward lower levels for the UVT treatment compared to the open and UVB treatments, as in 2016.

Figure 7 shows how the distribution of infestation levels was weighted closer to higher levels for PFR compared to the other biopesticide treatments, as in 2016.

In both 2016 and 2017, no statistically significant differences in numbers of *L. lineolaris* nymphs or adults were observed across covering or biopesticide treatments (Tables 9 and 10). Figures 8 and 9 show the mean number of total (nymphs plus adults) *L. lineolaris* observed across covering and biopesticide treatments over the course of the 2017 season. In 2016, the *L. lineolaris* population was very low overall, reaching a peak of a mean 0.3 individuals per four-plant replication unit on the sampling day with the highest total count of *L. lineolaris*. By contrast, in 2017, *L. lineolaris* peaked with a mean of 1.4 individuals per four-plant replication unit on the sampling day with the highest total count of *L. lineolaris*.

Discussion

Most strawberry acreage in the United States is non-organic, but demand for organic strawberries is rising (Daugaard, 1999; Gu et al., 2017; Hoover et al., 2014;). Strawberries are challenging to manage organically as they are highly susceptible to pests and diseases (Andrade et al., 2016; Nes et al., 2017). Although organically approved biopesticides are commercially available and listed for use in strawberry production, there is limited research on their efficacy against common strawberry insect pests. In general, information available to organic growers regarding crop and pest responses to specific organic environments and management practices is limited (Hoashi-Erhardt et al., 2013).

For the biopesticides used in this study, Entrust consistently provided the highest levels of control against *D. suzukii* in semi-field bioassays. However, a 30-40% population reduction in *D. suzukii*, as observed in the Day 0 and Day 1 trials in July and September from the Entrust treatment, would not necessarily translate into a 30-40% reduction in crop damage in a field infested with *D. suzukii*. Compared to water, Mycotrol and PFR caused some mortality of *D. suzukii* in the bioassays, but it was still too low for practical benefit. We generally observed higher mortality rates of male *D. suzukii* compared to females, though the differences between sexes were not always statistically significant. This is unsurprising as male *D. suzukii* are smaller than females, so at any application rate, male flies receive a higher dosage per unit of body mass than female flies.

The differences in mortality between the Day 0 and Day 3 trials with Entrust suggest that within 3 days of spraying the product in the field, the active ingredient (spinosad) is significantly dispersed or degraded. The rate of degradation likely varies with environmental conditions, but it is consistent with a study by Leach et al. (2017) that found spinosad residues fall to undetectable levels within 3 days of application in UV transmitting or uncovered environments. However in that study, spinosad resulted in higher levels of mortality of *D. suzukii* under UV-blocking treatments compared to UV-transmitting or open treatments. This was not the case in our study, where covering treatment did not have a significant impact on the levels of mortality caused by any biopesticide treatment.

The microbial agents in the Mycotrol and PFR treatments may not have degraded as quickly based on differences in mortality levels observed between the Day 0 and Day 3 trials within each bioassay experiment; however, the mortality levels were very low overall. Numerous studies have demonstrated that sunlight exposure quickly degrades microbial agents and reduces their efficacy in the field by damaging spore viability and insecticidal activity for fungal conidia. Ultraviolet (UV) light in the range of 300-400 nm disrupts normal metabolic processes (Behle et al., 2011; Cory and Hoover, 2006). One might therefore expect mycoinsecticides such as Mycotrol or PFR to work more effectively in an environment with lower UV exposure, but this did not manifest in our study, as fly mortality rates were not significantly higher under UVB treatments on any trial day. It is possible that higher mortality rates would have been observed beyond seven days of exposure. In a study exposing *D. suzukii* adults to surfaces treated with *B. bassiana* (the active ingredient in Mycotrol) and *I. fumosorosea* (the active ingredient in PFR), mortality rates did not exceed 50% until at least ten days of exposure to *B. bassiana* or twelve days of exposure to *I. fumosorosea* (Cossentine et al., 2016). Due to how quickly strawberry leaf samples decayed in the lab after removal from the field, we were not able to observe for *D. suzukii* mortality beyond seven days after setting up the bioassay chambers.

The biopesticides in our study provided no significant, observable control of *L. lineolaris* or *T. urticae*. Other evidence, however, suggests that Mycotrol and Entrust can provide some control of these insect pests. Ismail et al. (2007) found spinosad to

have substantial acaricidal and indirect ovicidal properties on *T. urticae* under laboratory conditions. Spraying sublethal concentrations of spinosad reduced the mites' fecundity compared to water-sprayed control groups, and when sprayed directly on newly deposited eggs, hatching rate and healthy development was significantly reduced compared to the control group. Van Leeuwen et al. (2005) found that spinosad could also be administered as a systemic insecticide for protection against *T. urticae*. In laboratory trials with *L. lineolaris*, adults and nymphs were effectively controlled by *B. bassiana* and the fungal conidia remained viable for control for 6 days on strawberries in the lab (Sabbahi et al., 2008).

Our study did not produce similar results, but the effectiveness of microbial biopesticides can depend heavily on environmental factors and the timing of sprays (Caldwell et al., 2013). Furthermore, we were not necessarily expecting to see any reductions in pest levels from biopesticide treatments in the field because we applied spray treatments only three to four times over the course of each growing season. They were primarily applied for the purpose of conducting the semi-field bioassays and not for controlling insects in the field. It was somewhat surprising to find any effect at all of the biopesticides on *T. urticae*, especially since the effect seemed to be that use of PFR may have increased the probability of observing *T. urticae* (2016) or the level of *T. urticae* infestation (2017). It is possible therefore that *I. fumosorosea* (the microbial agent in PFR) actually controls for predators of *T. urticae* rather than *T. urticae*. The fact that *T. urticae* presence was sometimes higher in plots treated with PFR compared to plots treated with water highlights the need to understand how this control product affects non-target species.

Covering treatments had no statistically significant effects on the presence of either *L. lineolaris* or *T. urticae* in the field. However, in both years, presence of *T. urticae* was slightly lower under the UVT treatment compared to both the UVB and open treatments. This is interesting considering evidence from a study by Tanaka et al. (2016), which found that UV light deters *T. urticae* in a greenhouse environment. Thus we might have expected to see higher levels of *T. urticae* under a UVB treatment. However, in our study the sides of each tunnel remained open for most of the season, allowing some UV light to enter.

In conclusion, covering type did not significantly affect insect pest presence or change pest control outcomes in an organically managed planting of the day-neutral strawberry 'Albion'. From an insect pest management standpoint, there was no clear advantage or disadvantage to either plastic covering compared to an open control. Overall, Entrust was the most effective biopesticide against *D. suzukii*, followed by Mycotrol and PFR, but the mortality rates achieved in the bioassays would not translate to adequate control of the pest in the field. There remains a great deal to be learned about how to effectively utilize microbial biopesticides in the field for control of strawberry insect pest species. Future research could look at adjusting application rates of these biopesticides to better suit a tunnel environment or developing new formulations that improve the longevity of the living, active ingredients. With new advancements in biopesticide products for use in organic strawberry production, it will be important to evaluate for any negative effects to beneficial insects, including pollinators or predatory insects.

Chapter 2: Tables and Figures

Table 1. Steps involved in semi-field bioassays. Each bioassay experiment is associated with one spray treatment of four biopesticides (Entrust SC, Mycotrol WPO, PFR-97, and Water) applied to split plots within each of twelve main plots in a low tunnel strawberry production system. Main plots have one of three tunnel covering treatments: UV-blocking, UV-transmitting, or open. The first trial was conducted with leaves removed the same day as the spray date (Day 0 trial), the second trial was conducted with leaves removed one day after the spray date (Day 1 trial), and the third trial was conducted with leaves removed three days after the spray date (Day 3 trial). Leaf samples were introduced to bioassay chambers with 10 adults *Drosophila suzukii* in the lab. Fly mortality was recorded at multiple intervals after exposure to the leaves in the bioassay chambers, with the final mortality recorded after 7 days of exposure. The field portion of the bioassays was carried out on certified organic land planted with 'Albion' strawberries in St. Paul, Minnesota.

Spray Treatments: July 28, 2017 and September 11, 2017

Days after spray	Day 0 Trial	Day 1 Trial	Day 3 Trial
0 (Spray Day)	Leaf Samples Taken		
1	Mortality Recorded	Leaf Samples Taken	
2		Mortality Recorded	
3	Mortality Recorded		Leaf Samples Taken
4		Mortality Recorded	Mortality Recorded
5	Mortality Recorded		
6		Mortality Recorded	Mortality Recorded
7	Mortality Recorded		
8		Mortality Recorded	Mortality Recorded
9			
10			Mortality Recorded

Table 2. Percent mortality of male and female *D. suzukii* after 7 days of exposure to leaves in forty-eight experimental units sprayed with four biopesticide treatments under three covering treatments on July 28, 2017. Letters denote statistically significant differences within columns and factors by Tukey's post-hoc test at $p < 0.05$. Biopesticide treatments were carried out on certified organic land planted with 'Albion' strawberries in St. Paul, Minnesota.

		Percent Mortality After 7 Days of Exposure [Mean \pm SE]					
Covering		Day 0 Trial		Day 1 Trial		Day 3 Trial	
	Open	16.3 \pm 4.0		13.8 \pm 3.3		8.1 \pm 3.1	
	UVT	20.6 \pm 5.4		19.4 \pm 5.1		8.1 \pm 3.2	
	UVB	22.6 \pm 4.4		16.3 \pm 5.3		5.0 \pm 2.8	
Biopesticide							
	Entrust	37.5 \pm 5.6 a		32.5 \pm 6.9 a		6.7 \pm 3.7	
	Mycotrol	16.5 \pm 5.1 b		13.3 \pm 5.2 b		10.8 \pm 4.3	
	PFR	16.7 \pm 5.1 b		14.2 \pm 4.7 b		9.2 \pm 3.8	
	Water	8.3 \pm 3.8 b		5.8 \pm 1.9 b		1.7 \pm 1.2	
Sex							
	Female	16.6 \pm 3.4		11.3 \pm 3.3 b		5.0 \pm 2.0	
	Male	22.9 \pm 4.1		21.7 \pm 4.1 a		9.2 \pm 2.9	
ANOVA	df	F	P	F	P	F	P
Covering	2	0.575	0.581885	0.474	0.62444	0.349	0.707
Biopesticide	3	6.090	0.00857	5.742	0.00128	1.284	0.285
Sex	1	1.562	0.214978	4.852	0.03038	1.395	0.241
Covering:Bio.	6	0.229	0.966132	2.451	0.03128	0.795	0.576

Table 3. Interaction mean comparisons of percent mortality of *D. suzukii* in the Day 1 bioassay trial from the July 28, 2017 spray treatment. Means \pm SE of all covering:biopesticide combinations are displayed. Letters denote statistically significant differences within covering types by Tukey's post-hoc test at $p < 0.05$. Biopesticide treatments were carried out on certified organic land planted with 'Albion' strawberries in St. Paul, Minnesota.

Biopesticide		Open		Covering UVT		UVB	
	Entrust	32.5 \pm 6.5 a		17.5 \pm 10.3		47.5 \pm 16.0 a	
	Mycotrol	10.0 \pm 7.6 b		27.5 \pm 12.5		2.5 \pm 2.5 b	
	PFR	7.5 \pm 3.7 b		27.5 \pm 11.9		7.5 \pm 5.3 b	
	Water	5.0 \pm 3.3 b		5.0 \pm 3.3		7.5 \pm 3.7 b	
ANOVA	df	F	P	F	P	F	P
Biopesticide	3	5.208	0.00552	1.099	0.366	5.792	0.00327

Table 4. Percent mortality of male and female *D. suzukii* after 7 days of exposure to leaves under sprayed with four biopesticide treatments under three covering treatments on September 11, 2017. Letters denote statistically significant differences within leaf sample days and factors by Tukey's post-hoc test at $p < 0.05$. Biopesticide treatments were carried out on certified organic land planted with 'Albion' strawberries in St. Paul, Minnesota.

		Percent Mortality After 7 Days of Exposure [Mean±SE]					
Covering		Day 0 Trial		Day 1 Trial		Day 3 Trial	
	Open	13.1 ± 4.5		13.1 ± 4.1		7.5 ± 2.5	
	UVT	10.6 ± 4.2		15 ± 4.2		6.7 ± 2.6	
	UVB	6.3 ± 2.6		8.8 ± 3.5		8.8 ± 3.0	
Biopesticide							
	Entrust	35.8 ± 6.1 a		38.3 ± 5.4 a		14.2 ± 4.1	
	Mycotrol	2.5 ± 1.8 b		5.8 ± 3.5 b		5.5 ± 2.3	
	PFR	0.8 ± 0.8 b		1.7 ± 1.2 b		5.8 ± 3.1	
	Water	0.8 ± 0.8 b		3.3 ± 1.6 b		5.0 ± 2.2	
Sex							
	Female	9.2 ± 3.3		10.8 ± 3.0		9.4 ± 2.6	
	Male	10.8 ± 3.0		13.8 ± 3.4		6.0 ± 1.7	
ANOVA	df	F	P	F	P	F	P
Covering	2	1.551	0.218	1.298	0.2785	0.155	0.857
Biopesticide	3	28.556	8.77*10⁻¹³	28.806	7.31*10⁻¹³	2.089	0.108
Sex	1	0.267	0.607	0.805	0.3721	1.235	0.270
Covering:Bio.	6	1.218	0.306	1.890	0.0922	0.983	0.442

Table 5. Analysis of deviance table for a logistic regression model of *T. urticae* presence measured on eight sampling dates in twelve field plots sprayed with four biopesticide treatments under three covering treatments in 2016. In the table, "residual" is abbreviated by "res," "deviance" is abbreviated by "dev," and "null" is abbreviated by "N." Research was carried out on certified organic land planted with 'Albion' strawberries in St. Paul, Minnesota.

Treatment	df	Dev.	Res. df	N. Res. df	Res. Dev.	N. Res. Dev.	P
Covering	2	2.2698	1524	1526	2000.4	2002.7	0.3215
Biopesticide	3	3.5897	1523	1526	1999.1	2002.7	0.3093

Table 6. Distribution of frequencies of “none,” “low,” “med,” and “high” levels of *T. urticae* infestation observed across thirteen sampling dates in twelve field plots under three covering treatments in 2017. Below each level of infestation, cells on the left report actual counts and cells on the right report the proportion represented by each covering treatment out of the total counts across all treatments for that level of infestation. Research was carried out on certified organic land planted with ‘Albion’ strawberries in St. Paul, Minnesota.

Covering	Levels of <i>T. urticae</i> Infestation							
	None		Low		Med		High	
	Observed Frequency	Proportion of Total	Obs.	Prop.	Obs.	Prop.	Obs.	Prop.
Open	606	0.324	117	0.371	58	0.354	30	0.319
UVB	623	0.334	99	0.314	64	0.390	36	0.383
UVT	639	0.342	99	0.314	42	0.256	28	0.298

Chi-square test: $\chi^2 = 8.646$, df = 6, P = 0.1945

Table 7. Distribution of frequencies of “none,” “low,” “med,” and “high” levels of *T. urticae* infestation observed across thirteen sampling dates in twelve field plots sprayed with four biopesticide treatments in 2017. Below each level of infestation, cells on the left report actual counts and cells on the right report the proportion represented by each biopesticide treatment out of the total counts across all treatments for that level of infestation. Research was carried out on certified organic land planted with ‘Albion’ strawberries in St. Paul, Minnesota.

Biopesticide	Levels of <i>T. urticae</i> Infestation							
	None		Low		Med		High	
	Observed Frequency	Proportion of Total	Obs.	Prop.	Obs.	Prop.	Obs.	Prop.
Entrust	500	0.268	75	0.238	31	0.189	15	0.160
Mycotrol	471	0.252	75	0.238	36	0.220	24	0.255
PFR	451	0.241	76	0.241	52	0.317	32	0.340
Water	446	0.239	89	0.282	45	0.274	23	0.245

Chi-square test: $\chi^2 = 17.793$, df = 9, P = **0.03765**

Table 8. Multiple pairwise comparisons of levels of *T. urticae* infestation observed across thirteen sampling dates in twelve strawberry field plots sprayed with four different biopesticide treatments in 2017. P-values are calculated by Pearson's chi-squared test and adjusted p-values are corrected for false discovery rate. Research was carried out on certified organic land planted with 'Albion' strawberries in St. Paul, Minnesota.

Treatments	Raw P-value	Adjusted P-value
Entrust vs. Mycotrol	0.3715	0.4088
Entrust vs. PFR	0.0030	0.0182
Entrust vs. Water	0.0406	0.1218
Mycotrol vs. PFR	0.2148	0.4088
Mycotrol vs. Water	0.4088	0.4088
PFR vs. Water	0.3951	0.4088

Table 9. Kruskal-Wallis rank sum test of numbers of *L. lineolaris* nymphs and adults observed across eight sampling dates in 2016 and thirteen sampling dates in 2017 in twelve field plots sprayed with four biopesticide treatments under three covering treatments. Research was carried out on certified organic land planted with 'Albion' strawberries in St. Paul, Minnesota.

		Year							
		2016				2017			
		χ^2		P		χ^2		P	
Chi-square	df	Nymph	Adult	Nymph	Adult	Nymph	Adult	Nymph	Adult
Covering	2	0.953	2.826	0.621	0.243	1.838	0.042	0.399	0.979
Biopesticide	3	3.338	3.752	0.342	0.290	2.897	3.700	0.408	0.296

Figure 1. Percent mortality of total (male plus female) *D. suzukii* (mean \pm SE) at different intervals after exposure to leaves sprayed with four biopesticide treatments on July 28, 2017. Each graph represents results from one bioassay trial day (day 0, day 1, or day 3). Letters denote statistically significant differences within days of exposure by Tukey's post-hoc test at $p < 0.05$.

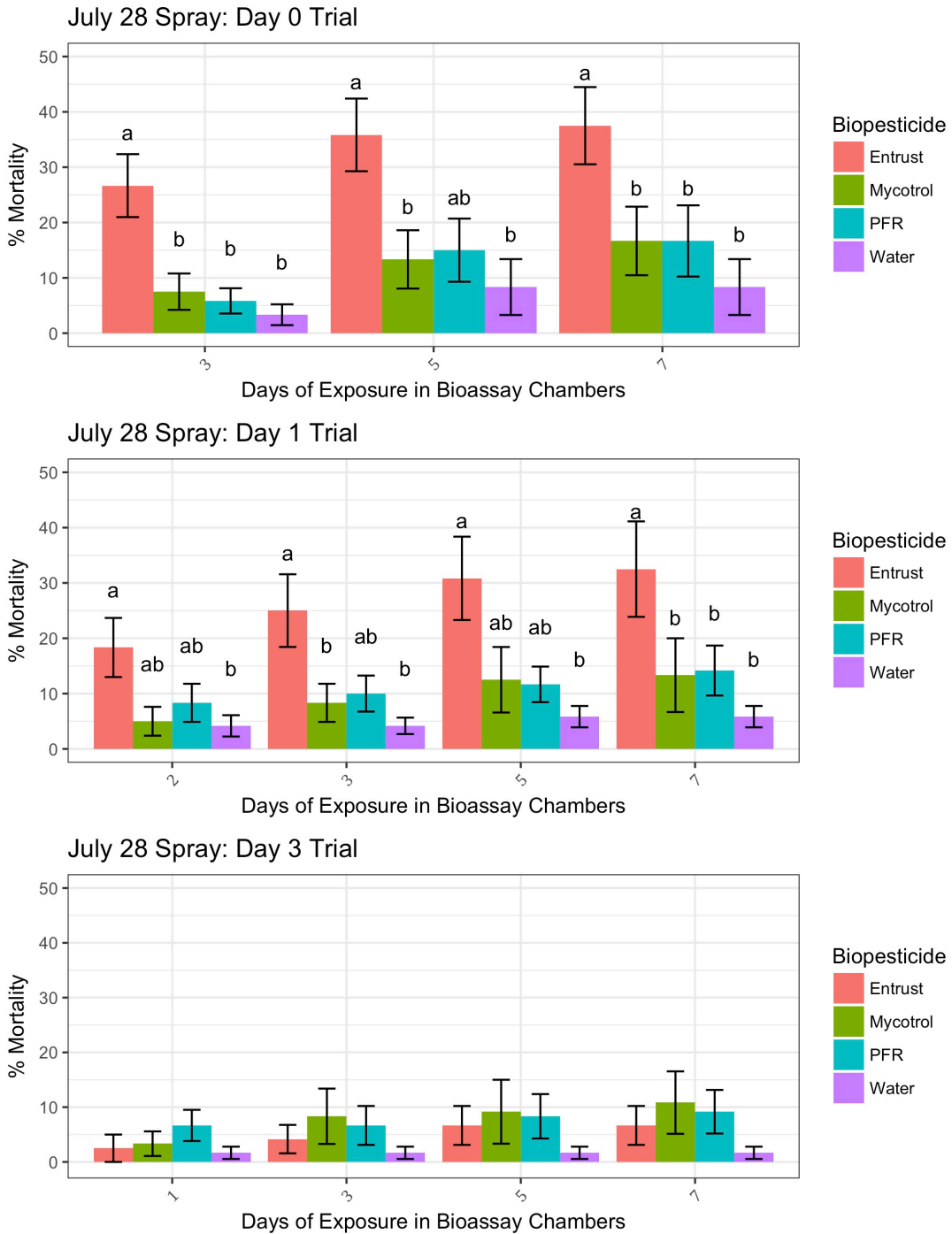


Figure 2. Percent mortality of total (male plus female) *D. sukii* (mean \pm SE) at different intervals after exposure to leaves sprayed with four biopesticide treatments on September 11, 2017. Each graph represents results from one bioassay trial day (day 0, day 1, or day 3). Letters denote statistically significant differences within days of exposure by Tukey's post-hoc test at $p < 0.05$.

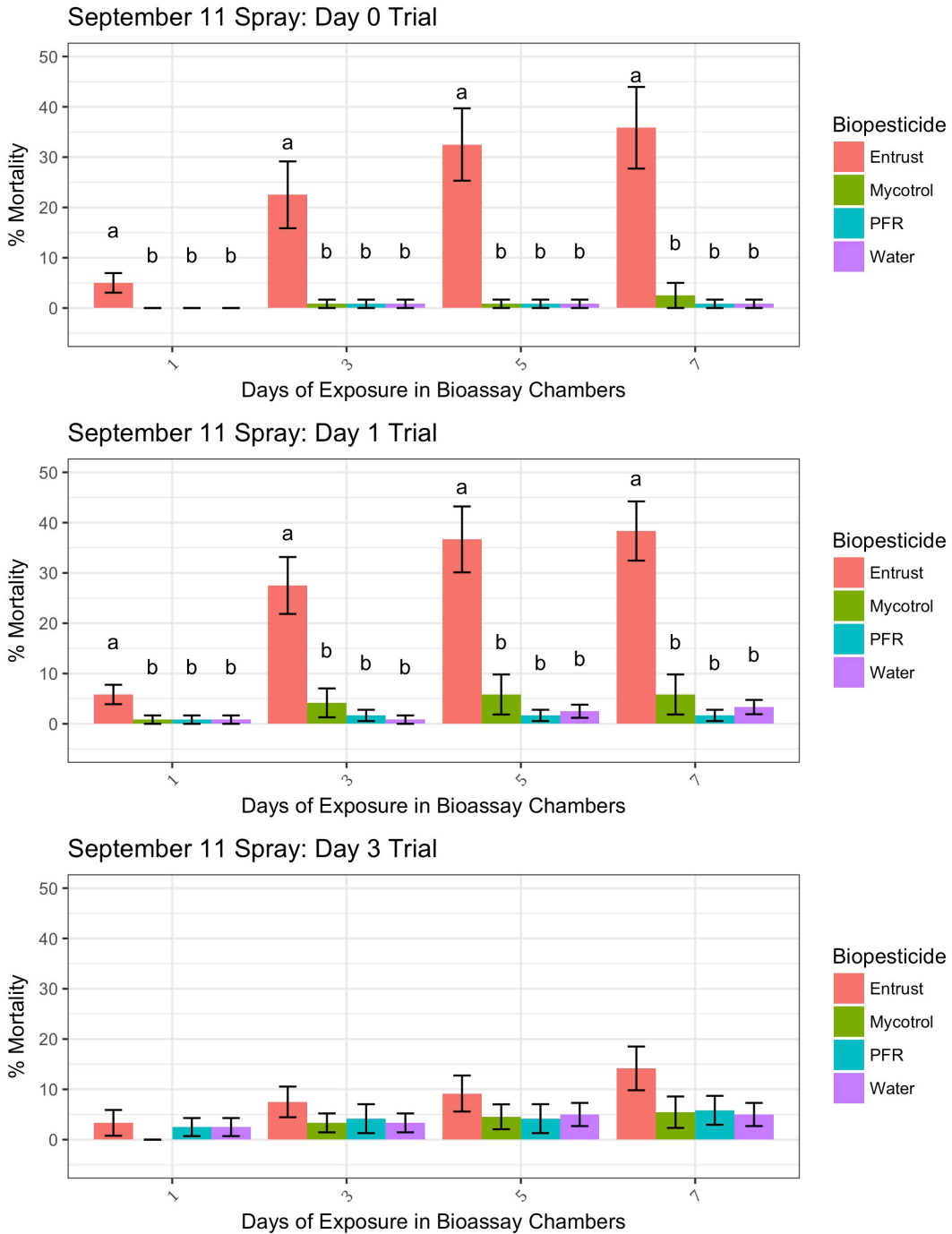


Figure 3. Captures of *D. suzukii* (spotted wing drosophila) (mean) in three traps spaced randomly around the field research site on eighteen sampling dates in 2017. Research was carried out on certified organic land planted with 'Albion' strawberries in St. Paul, Minnesota.

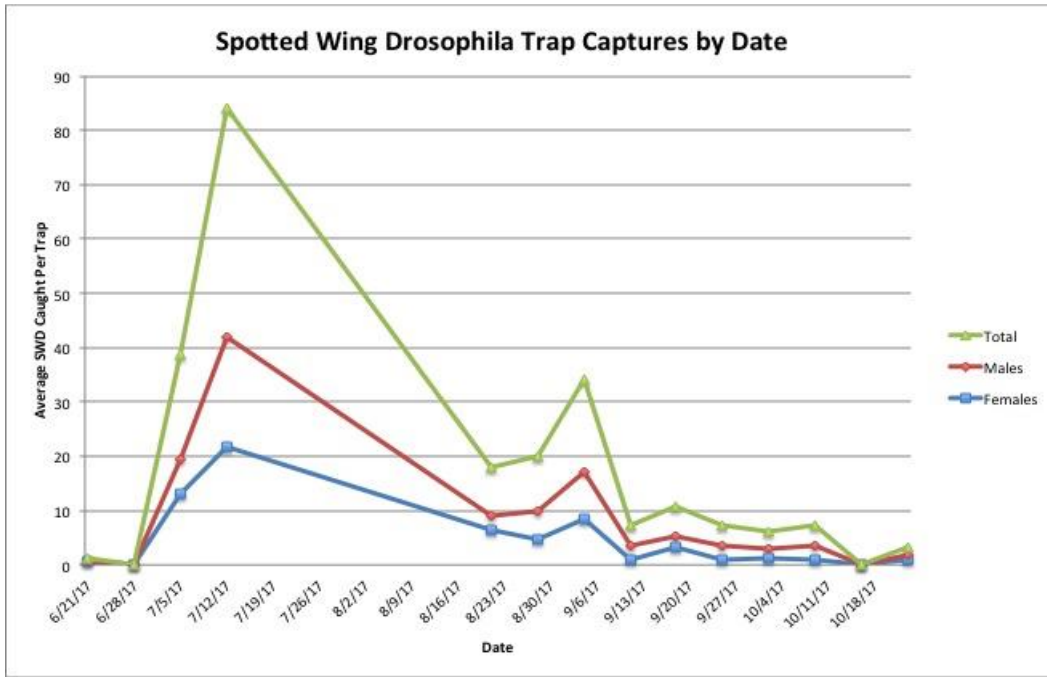


Figure 4. Predicted probability of observing *T. urticae* in field plots under three covering treatments in 2016 (based on a logistic regression model of data gathered over eight sampling dates). Research was carried out on certified organic land planted with 'Albion' strawberries in St. Paul, Minnesota.

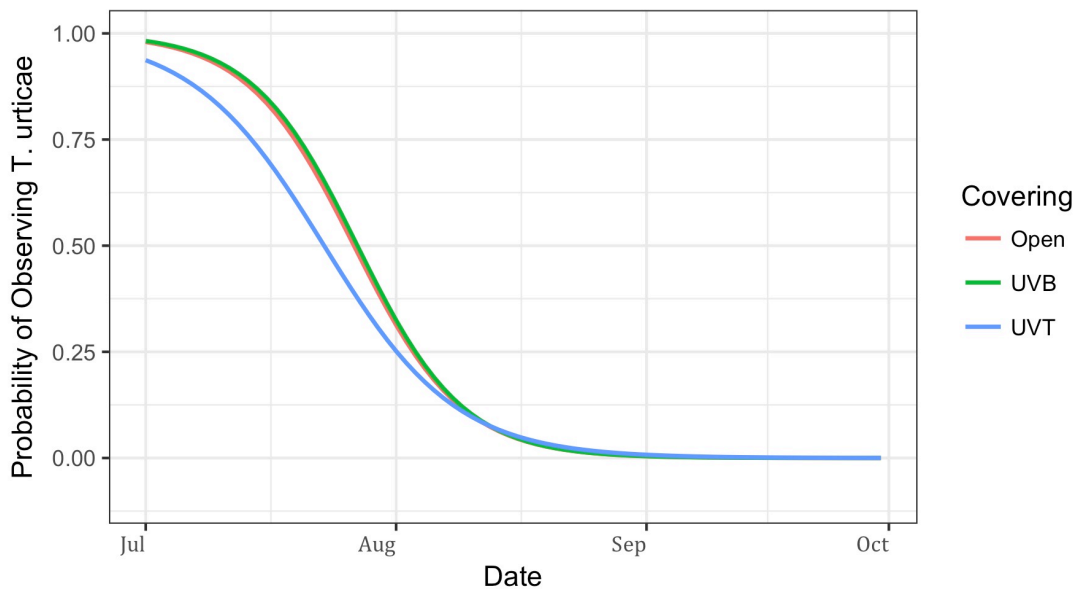


Figure 5. Predicted probability of observing *T. urticae* in field plots sprayed with four biopesticide treatments in 2016 (based on a logistic regression model of the data gathered over eight sampling dates). Research was carried out on certified organic land planted with ‘Albion’ strawberries in St. Paul, Minnesota.

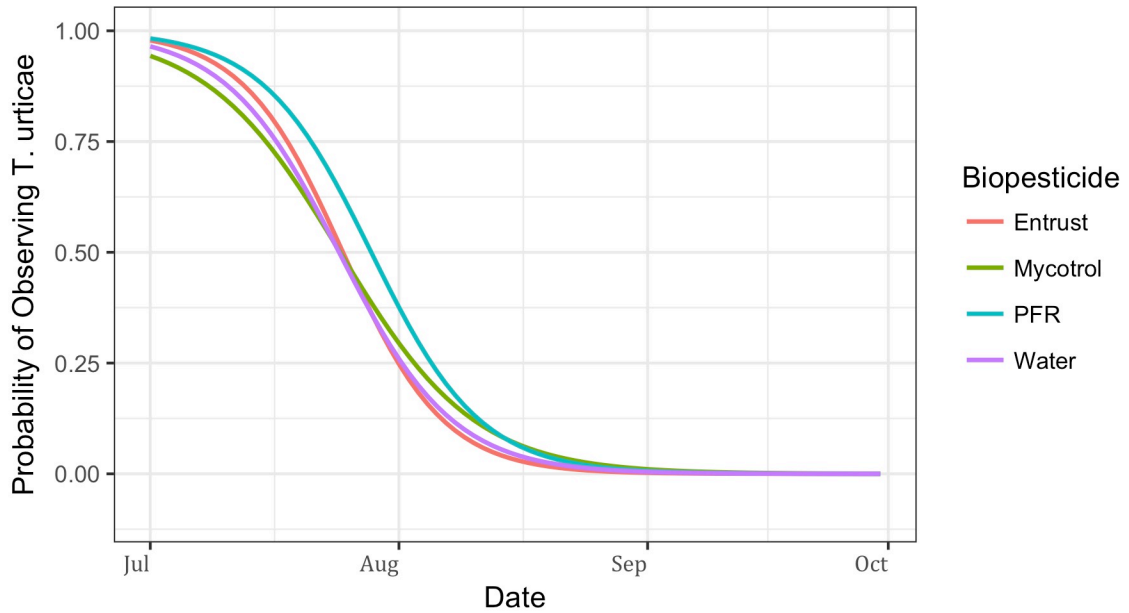


Figure 6. Observations of *T. urticae* in field plots under three covering treatments in 2017. Points indicate individual observations across thirteen sampling dates; local regression trend lines indicate changes in *T. urticae* infestation over time. Research was carried out on certified organic land planted with ‘Albion’ strawberries in St. Paul, Minnesota.

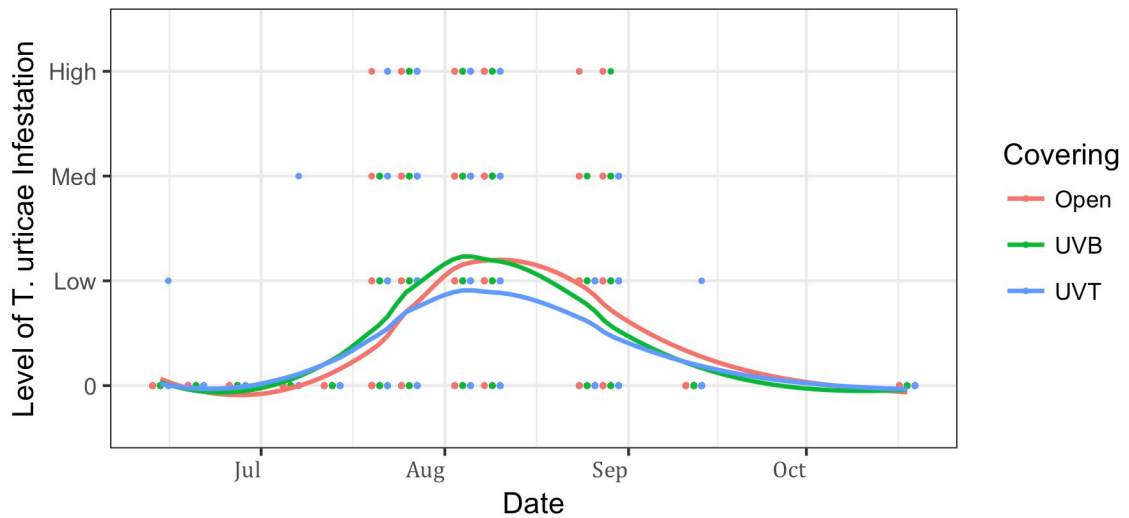


Figure 7. Observations of *T. urticae* in field plots sprayed with four biopesticide treatments in 2017. Points indicate individual observations across thirteen sampling dates; local regression trend lines indicate changes in *T. urticae* infestation over time. Research was carried out on certified organic land planted with ‘Albion’ strawberries in St. Paul, Minnesota.

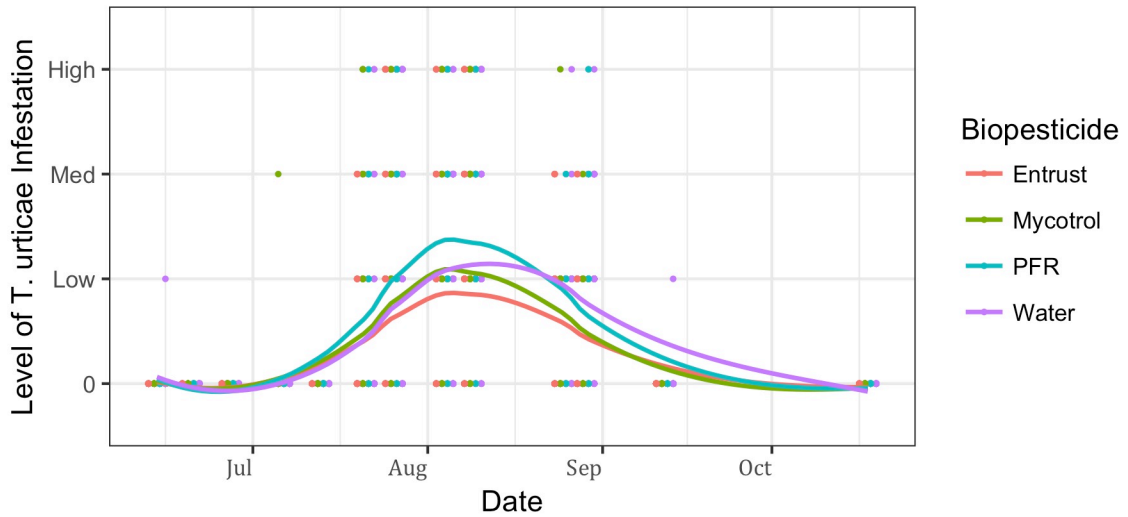


Figure 8. Total number of *L. lineolaris* (mean \pm SE) per 4-plant replication unit observed on each of thirteen sampling dates in field plots under three covering treatments in 2017. Research was carried out on certified organic land planted with ‘Albion’ strawberries in St. Paul, Minnesota.

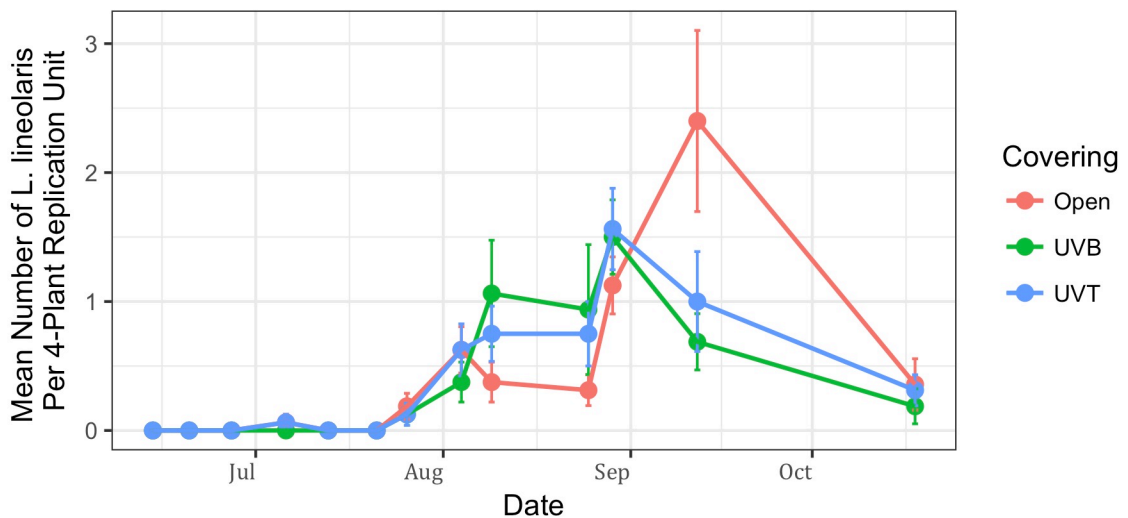
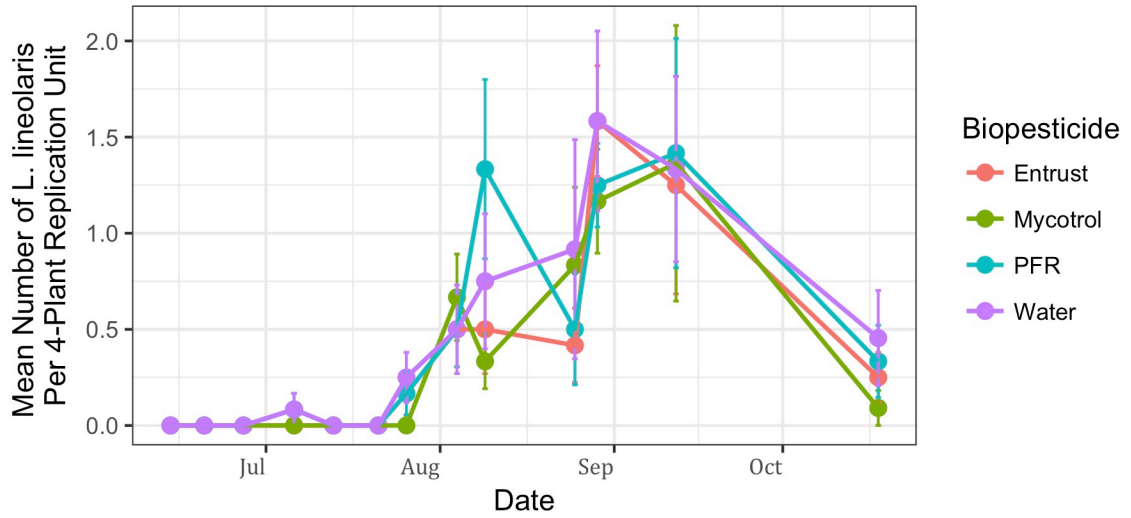


Figure 9. Total number of *L. lineolaris* (mean \pm SE) per 4-plant replication unit observed on each of thirteen sampling dates in field plots sprayed with four biopesticide treatments in 2017. Research was carried out on certified organic land planted with 'Albion' strawberries in St. Paul, Minnesota.



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Appendix A: Optimizing Protected Culture Environments for Berry Crops

The studies described in this thesis were conducted as part of a larger research project funded by the National Institute of Food and Agriculture, U.S. Department of Agriculture, Specialty Crops Research Initiative under award number 2014-51181-22380. The project includes researchers at Michigan State University, Pennsylvania State University, Cornell University, University of New Hampshire, University of Vermont, Rutgers University, USDA-ARS-Appalachian Fruit Research Station, USDA-Beltsville, and the Lancaster Environment Centre at Lancaster University. Additionally, an advisory board, grower organization partners, and industry collaborators have all contributed their expertise to the design and development of the project, with three focus areas related to growing berries in protective structures in the Northeast and Upper Midwest:

1. Identifying and addressing threats from pests and diseases, including threats to specialty crop pollinators.
2. Improving production efficiency, productivity, and profitability over the long term.
3. Testing new innovations and technology.

The long-term goals of this research are to improve the profitability of berry production in the Northeast and Upper Midwest by:

1. Identifying the most effective tunnel type and plastic for different berry crops and locations.
2. Determining if specialty plastics can reduce pesticide use by suppressing diseases and insect pests.
3. Minimizing the negative environmental impact of plastic use by increasing the recycling of tunnel plastics.

More information on this research initiative can be found at www.tunnelberries.org

Appendix B: Leaf Plating Experiment

In the 2016 field season, I tried an experiment to measure persistence of fungal biopesticides in the environment over time. Following a biopesticide treatment in the field, I collected leaf samples from each split plot the same day, one day after the treatment, and three days after the treatment. Immediately after collecting the samples, I pressed them individually onto selective growth media in plates and left them for 24 hours, covered. When I removed the leaf samples the following day, I scanned each one with WinFOLIA leaf analysis software (Regent Instruments, Inc., Québec, Canada) to measure leaf surface area.

In theory, if viable fungal spores were present on the surface of the leaves, they would transfer to the agar and germinate, allowing me to count colony-forming units (CFUs) and approximate the number of viable spores present on the leaves. The two biopesticides I sought to evaluate in this experiment, *Beauveria bassiana* and *Isaria fumosorosea*, are easily identifiable when they germinate. Unfortunately, a high level of contamination from other fungal and bacterial species obscured results, making it difficult to identify species and distinguish individual CFUs. Additionally, it was difficult to achieve full surface contact between leaf samples and the agar for the initial 24 hours because strawberry leaves are hairy and resist sticking to agar! Had it been easier to count CFUs, I would have used the measure of each leaf sample's surface area to calculate density of CFUs per unit of leaf area, comparing results between the two biopesticides and with water-sprayed leaves.

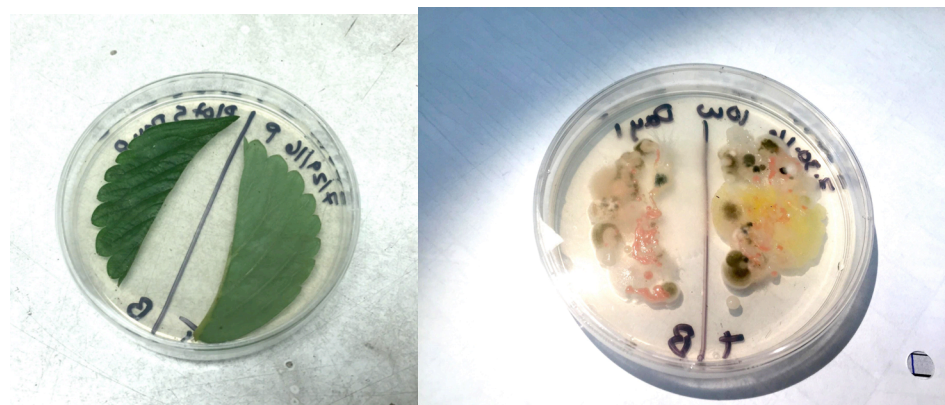


Figure 1. Leaf imprints. On the left, half a leaflet is pressed topside down in the agar, and half is pressed right side up. On the right, a mix of fungi and bacteria grow on a plate after five days of germination in a growth chamber. CFUs blend together.