INTEGRATED PEST MANAGEMENT FOR TWO RED RASPBERRY PESTS
UTILIZING UV-BLOCKING HIGH TUNNEL PLASTIC FILMS

A Thesis in
Horticulture
by
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Submitted in Partial Fulfillment
of the Requirements
for the Degree of

Master of Science

December 2018
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ABSTRACT

In the early 2000’s, red raspberry crops in the Northeast United States were increasingly produced in high tunnels. High tunnels have potential to decrease the amount of pesticides used to produce raspberries; fungicides can be unnecessary and many arthropod pests are successfully controlled through natural enemies in the tunnels. However, insecticides are still the primary tool that growers use to protect raspberry crops from pests like Japanese beetles and spotted wing drosophila (SWD). Using insecticides for SWD control is especially problematic because the short residual activity of treatments requires growers to make many applications over the season putting high selection pressure on SWD for resistance. Controlling SWD is so difficult and costly that adoption of high tunnels for red raspberry has slowed in Pennsylvania.

While Japanese beetles have only a single generation per year, and resistance is less likely, their control also often relies on very few chemicals. In both cases, sprays present risks to beneficials that are important for the ecosystem of the tunnel.

Many insect pests are reduced in greenhouses using UVA-blocking plastic films. We investigated the effects of plastics that transmit different amounts of ultraviolet light on Japanese beetles and SWD. Many insects are sensitive to light in the UVA range and use it for navigation, and high tunnel plastics that block varying amounts of UV radiation are increasingly available.

Combining deterrents and attractants in a “push pull” system has been a successful IPM strategy in many crops. Attracticidal spheres, red balls containing sugar and a toxicant, are effective for reducing SWD populations in trials of field-grown raspberries. Shortened harvest interval has decreased infestation in tunnels. We hypothesized that combining the “pushes” of UV-blocking plastic and daily harvest with the “pull” of the attracticidal spheres could create an effective non-spray control program for SWD in high tunnels.
We grew two primocane-bearing red raspberry cultivars, ‘Polka’ and ‘Josephine’, under six different covering treatments in 2016 and 2017. Five were plastics which blocked the ultraviolet range to varying degrees, and one treatment had no plastic covering. In 2016 Japanese beetles were counted and removed daily from the plants by hand. In 2017 beetles were removed by hand every 5 days. In both years SWD populations were monitored using apple cider vinegar traps which were collected on a weekly basis throughout the harvest season. Foliage temperature was measured in each tunnel twice in 2017 with an infrared thermometer. Spectral transmittance characteristics of the plastics were measured with a spectroradiometer in 2015 or 2016 and 2018. Mean beetle counts by date and for the whole season were compared for the plastics and cultivars. Japanese beetle numbers were significantly higher in the no plastic treatment than under all plastic treatments. The plastic that when new blocked >90% of the UV range in our measurements had significantly lower beetle populations while the plastics which blocked the least UV had significantly higher beetle counts. It appears that using a UV-blocking plastic can reduce Japanese beetle aggregation and feeding damage on raspberries. This could benefit growers by eliminating the cost of purchasing and applying insecticides, and also decrease the exposure risk for to non-target organisms.

Spotted wing drosophila trap numbers were significantly impacted by whether the raspberry plants were grown inside tunnels. Compared to the outside treatment, all plastics had lower numbers in 2016 and higher numbers in 2017, but plastics did not significantly differ. Given these results, it seems unlikely that spotted wing drosophila is affected by plastics with different transmittances. This was supported by bioassays conducted in laboratory and field cage settings in 2018 where there was no difference in foraging behavior between UV-blocking and UV-transmitting plastics, and a control treatment.
In 2018 we also grew ‘Josephine’ raspberries in tunnels under two covering treatments, a UV-blocking plastic and a UV-transmitting plastic. We tested combinations of these plastics, attracticidal spheres, and different harvest intervals. Fruit was harvested for two twelve-day periods during which we measured the marketable weight, total weight, and average fruit weight, and calculated the percentage of marketable fruit. Subsamples of fruit were submitted to saline floats to extract larvae and evaluate infestation. There were no significant differences between plastics in the amount or percentage of marketable yield, fruit weight, or infestation of marketable fruit. Daily harvest significantly increased total and marketable yield compared with a Monday, Wednesday, Friday harvest schedule. Daily harvest and spheres significantly decreased the infestation of marketable fruit. In fruit that was considered unmarketable, UV-transmitting plastics also significantly reduced infestation.

This suggests that a daily harvest schedule can increase marketable yields for growers and using attracticidal spheres can reduce infestation. Plastic seems unlikely to make a difference in the infestation of marketable fruit, and many growers already use UV-transmitting plastics. Overall, our research shows that combining a daily harvest interval with the application of attracticidal spheres can significantly reduce infestation and yield losses. Adoption of these control tactics would greatly reduce the risk to non-target and beneficial organisms in tunnels, the cost to growers in sprays, and the risk of resistance development by SWD.
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ACKNOWLEDGEMENTS

I thank Matthew Cooper, Sara Emigh, Dan Wisniewski, and Alys Tucker for their help on this project. Thanks to Nigel Paul and Jason Moore for providing the experimental plastics used in this experiment and Michael Glenn (retired, USDA) for conducting initial transmittance scans used in plastic cover selection. Thanks to Sharon Jones for help conducting bioassays. Thanks to Julie Cramer for constant support and generously editing my thesis.

This work is based upon research supported by the USDA National Institute of Food and Agriculture, Section 7311 of the Food, Conservation and Energy Act of 2008 (AREERA), Specialty Crops Research Initiative under Agreement 2014-51181-22380. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the author(s) and do not necessarily reflect the view of the U.S. Department of Agriculture.

This material is also based upon work supported by the National Institute of Food and Agriculture, U.S. Department of Agriculture, through the Northeast Sustainable Agriculture Research and Education program under subaward number GNE17-147-31064.
Chapter 1

Introduction

Red raspberry (*Rubus idaeus* subsp. *Idaeus*) is an important crop throughout the United States, where demand for conventional and organic fruit is growing (Hanson and Glick, 2013). Raspberry plants are perennial brambles in the genus *Rubus* and family *Rosaceae*. The fruit is a cup-shaped aggregate of drupelets that abscises from its receptacle during ripening and harvest. There are many differently colored raspberry species and hybrids with commercial value including red and yellow (*Rubus idaeus* subsp. *Idaeus*), black (*Rubus occidentalis*), and purple cultivars (hybrids of red and black raspberry) currently available. Of these species, red and black raspberries are the most widely grown for fresh market and processing (Jennings, 1988). Between 2007 and 2012, the most recently available censuses, the United States’ raspberry acreage grew more than 7%, and among berry crops was surpassed in growth only by blueberry and strawberry production (USDA, 2015). Much of this demand is driven by health-focused consumers looking for colorful, fresh fruit to supply phytonutrients and antioxidants (Harshman et al., 2014).

As consumer demand for raspberry fruit has grown, increased availability of primocane-fruiting cultivars has made raspberry production easier and more profitable for growers. Unlike traditional floricane-fruiting cultivars, primocane-fruiting raspberries produce canes that flower and set fruit in one season without requiring an overwintering period (Pritts, 2008). As a result, primocane-fruiting cultivars can produce fruit within the planting year. They also produce crops at novel times. While floricane-fruiting raspberries produce one crop at the beginning of the summer, primocane-fruiting raspberries set an additional fall crop with a much longer harvest period than the floricane crop. As the plants are still able to produce the floricane crop, depending upon management, they may produce two crops in a year, increasing yields. Additionally, primocane-fruiting raspberries do not require labor-intensive selective pruning and can instead be
mown to the ground if only a fall crop is desired (Pritts, 2008). The increasing adoption of
primocane-fruiting raspberries has enabled growers to grow berries with less labor, extend the
season so that berries are available throughout the year, and produce greater overall yields.

**High tunnel production for raspberry**

While Pennsylvania is among the top ten producing states of fruit, nuts, and berries, the
majority of fresh market raspberry production is in California (USDA, 2015; Hanson et al., 2013).
With ever-increasing transportation costs and demand for locally-produced fruit, there is potential
for raspberry production in the Northeast to grow (Weber, 2010). Horticultural production in
Pennsylvania and surrounding states tends to be on small and diversified farms which sell directly
to customers, often through farmers markets (Hanson et al., 2011). In this setting, consumers
frequently want fruit grown using low-spray or organic production practices, making these
practices economically desirable for the grower (Hanson and Glick, 2013).

Producing raspberry crops organically or conventionally is a challenge. The fruit is soft
and delicate and susceptible to gray mold (Harshman et al., 2014). The significant rainfall in
northeast states exacerbates this challenge and is likely to continue to increase with climate
change (EPA, 2016). Another challenge to raspberry production is that growers in areas with
short growing seasons and early fall frosts may not be able to take full advantage of the
production potential of primocane-fruiting cultivars.

Over the last twenty years high tunnel production has emerged as a solution to these
challenges (Demchak, 2009; Yao and Rosen, 2011). High tunnels are structures similar to
greenhouses, but less complex and not permanent. They generally consist of a metal or wooden
framework covered with a single layer of 4 to 6 mil plastic. Typically, crops are grown in the soil,
and irrigated, and there is no supplemental heating. They are generally vented with sides that can
roll up and down (Lamont, 2009). In the Northeast, high tunnels tend to be stand-alone structures
on small farms, while in California multi-bay tunnels covering many acres are used (Demchak, 2009). High tunnels are used worldwide for horticultural crop production as well, with Asia and Europe having higher adoption rates than the United States, although use in the U.S. is on the rise (Lamont, 2009; Chang et al., 2011). It is difficult to precisely gauge production and adoption of high tunnels, because the U.S. census does not distinguish between high tunnels and greenhouses, and because the language used to describe different protected culture structures internationally is not standardized (Carey et al., 2009; Lamont, 2009).

While reliable data on adoption rates are not available, based on anecdotal evidence it is clear that high tunnels have a tremendous impact on fruit and vegetable growers. The growing season can be extended and produce is generally of higher quality and requires reduced inputs for pest and pathogen control. In the case of raspberries and other brambles, high tunnels help ameliorate the challenge of heavy rainfall, with reduced wetness leading to fewer diseases including gray mold (Demchak, 2009; Hanson and Glick, 2013). Fruit are also larger and have longer shelf life, even without fungicides (Demchak, 2009). The season extension provided by high tunnels allows for much more of the fall primocane crop to be harvested, so growers in northern states can take full advantage of the opportunities these cultivars offer. In Rock Springs, Pennsylvania, the harvest season was extended by 50%, allowing yields in the first year of production that were equivalent to 2- to 3-year-old plantings in the field (Demchak, 2009).

Films for high tunnel production

In the past twenty years, a significant amount of research has gone into the cladding materials for greenhouses, and, more recently, the plastics available for high tunnel covering. Historically, interest was focused on maximizing photosynthetically active radiation (PAR) to enhance crop growth. Today, a more complete understanding of different ranges of light and different types of radiation is driving the development and adoption of new plastics (Giacomelli
and Roberts, 1993). Plastics can also partially or completely block certain regions of the spectrum, such as the infrared range, to moderate temperatures during hot days or reduce heat escape on cool nights (Karlsson and Werner, 2011). Plastics can diffuse light to greater or lesser extents, which scatters light and may enhance its distribution throughout the canopy (Johansen et al., 2011; Demchak, 2016).

Because light in the UV range (UV-A and UV-B) is responsible for shortening the life of plastic exposed to sunlight, plastics used for high tunnels and greenhouses contain chemical blockers and stabilizers to prevent degradation. As a result, some plastics prevent transmission of the light in the UV range while others allow only some. Whether or not a plastic transmits the UV range to the crop has traditionally been a byproduct of the technology used to extend the plastics durability (Demchak, 2016); however, this unintentional blocking of UV light has had important results. It has repeatedly been observed that aphid, thrips, and whitefly populations were lower in greenhouses covered with UV-blocking plastics (Antignus, 2000; Antignus et al., 2001; Doukas and Payne, 2007).

While a number of studies with plastics have characterized the transmittance properties of certain plastics that are commercially available, and many greenhouse studies investigated the effect of UV blocking plastics on insect pest populations, there are still many uncharacterized plastics available to growers (Krizek et al., 2005; Demchak, 2016), suggesting a current need to evaluate the characteristics of these plastics, including their impact on pests.

Further, whether or not UV light has a similar effect on insect pests in high tunnels, where open sides allow insects to move more freely than in greenhouses, remains a question (Krizek et al., 2005). When UV-blocking plastic which reduced insect pest populations in greenhouses was used on high tunnels it reduced the number of leaf miners, but not whiteflies and thrips. The lack of impact on these pests may have been due to unfiltered light and passive movement through the sides of the tunnels (Costa et al., 2003). However, there are many
variables which may affect populations of various species in high tunnels, such as the levels and range of UV light, the behavior of the pest, and the specific crop, so additional research is needed.

**Japanese beetle** (*Popillia japonica*) *(Coleoptera: Scarabaeidae)*

The ability of high tunnels to increase food production locally in regions with unfavorable growing conditions has had a significant positive environmental impact, even when considering a life-cycle analysis of the materials that go into a high tunnel (Chang et al., 2011). However, a challenge that remains to the overall sustainability of raspberry high tunnel production is the control of arthropod pests. Interestingly, researchers have observed that some pests which are devastating in field production can be reduced in high tunnels. One such pest is Japanese beetle (*Popillia japonica*) *(Coleoptera: Scarabaeidae)*.

Japanese beetle is a pest that came to the Eastern United States over 100 years ago. It quickly became established as a pest of field crops, ornamentals, and small fruit. Today Japanese beetle causes significant damage to these crops throughout the area east of the Mississippi River and has several populations outside of that range including a recent outbreak in Oregon (Davis, 1920; Potter and Held, 2002; Suits et al., 2017). The adult beetles skeletonize leaves and cause defoliation, and also cause direct damage by feeding on ripening fruit. While Japanese beetle is polyphagous, raspberry is a preferred small fruit host. (Isaacs et al., 2003).

Through most of its range Japanese beetle has one generation in a year. Adult beetles emerge over a 2-month period, peaking in July and August in Pennsylvania. Immediately upon emerging as adults from soil, virgin females mate and lay eggs. They then alternate between periods of feeding and oviposition, laying between 40 and 60 eggs over a 4- to 6-week lifespan (Fleming, 1972). These eggs, which the beetles preferentially lay into turf, hatch within 10-14 days. The larvae begin to feed on plant roots. The larvae go through two molts and most grubs have reached their third instar and are full sized by the winter (Potter and Held, 2002). Grubs
overwinter 5-15 cm (though sometimes deeper) under the soil surface. In the spring, as the soil warms, the beetles move up towards the surface. The grubs have a prepupal stage that lasts about 10 days and occurs around May, and then they pupate for 7-17 days. Following pupation, adults remain in the soil for several days to weeks to finish developing before emergence (Fleming, 1972).

While Japanese beetle has only one generation per season, and populations of adult beetles are very high in only July and August, the damage they incur typically requires some type of control. Insecticides are primarily used to control the adults surrounding and during peak adult emergence and mating. Even sprays with harsh chemistries require multiple applications over the season (Potter and Held, 2002; Suits et al., 2017). Some extension materials recommend controlling beetles in surrounding areas and in turf grass to reduce the adult pressure on berry crops (Isaacs et al., 2003), enlarging the area impacted. Utilizing entomopathogenic bacteria and nematodes has shown promise as well as challenges, including difficulties with culturing and application. The obligate nature of bacteria Btj and P. popilliae make them poor choices for high value crops (Stahly and Klein, 2002; Koppenhöffer et al., 2000). The heavy reliance on a narrow range of insecticides, and increased awareness of the impact of insecticides on pollinators and other non-target species suggest a need for integrated pest management (IPM) techniques for Japanese beetle management.

A key step in developing IPM strategies is understanding the biology and behavior of the target pest. Japanese beetle primarily locates its host plants via olfaction (Ahmad, 1982), aggregating in response to the volatile compounds that plants release as a result of herbivory (Loughrin et al., 1996). Although beetles navigate in response to odors, their activity is also influenced by light quantity and quality. Research involving wind tunnel bioassays and field observations found that the degree of Japanese beetle response to attractants was positively
related to light intensity and may also have been inhibited by certain ratios of light (Lacey et al., 1994; Heath et al., 2001).

As with earlier studies in greenhouses, the spectral transmittance of high tunnel films may have an impact on Japanese beetle presence. In an early demonstration planting, compared to plants grown outside of the tunnels, Japanese beetle populations were numerically lower on plants in tunnels (Demchak, 2009). In another study many herbivorous pests including Japanese beetle could be tolerated in high tunnels. The Japanese beetles were managed with hand removal (Hanson and Glick, 2013). The Japanese beetle is not closely related to the insect orders reported to be affected by UV-blocking plastics in the literature so far, but its observed flight activity suggests that spectral sensitivity plays a part in its response to attractants (Heath et al., 2001).

**Spotted Wing Drosophila, Drosophila suzukii (Matsumura) (Diptera: Drosophilidae)**

Spotted wing drosophila (SWD) is a recently arrived invasive pest of raspberries and other fruits, and its response to UV light has not been characterized. It is probably the most important insect pest of raspberries and presents the most significant challenge to both conventional and organic growers. In the last six years it has likely slowed the increase in high tunnel raspberry adoption in the northeast where many growers struggle with the spray schedules needed to control it.

SWD is native to southeast Asia, and it was first observed as a pest in cherries in Japan in 1916. In the 1980’s it was found in Hawaii where it was not a cause for significant concern. Then it appeared in California in 2008 infesting strawberries and bramble fruits and began to move across North America (Lee et al., 2011). Spread of the pest has likely been facilitated by climatic factors, the movement of infested fruit, an abundance of alternative hosts, and a lack of natural enemies to moderate their populations (Klick et al., 2015; Haye et al., 2016). SWD was first observed in Pennsylvania in 2011, with breeding populations established by 2012 (Freda and
Braverman, 2013). In Pennsylvania SWD appears in the first and second weeks of July and infests a range of small fruit and berry crops. Because of its late emergence in this region it is particularly damaging to late-season small fruits, especially blackberries and primocane-fruiting raspberries (Joshi et al., 2016).

SWD is particularly problematic for a number of reasons. First, females are equipped with an ovipositor with a pair of saw-like plates which cut into ripe and ripening fruit. (Lee et al., 2011). Unlike other fruit flies, which can only penetrate fruit which is already damaged or rotting, SWD females can lay eggs in marketable fruit. In the worst-case scenario consumers may encounter fruit with visible larvae. Even when eggs do not hatch before the fruit is used, oviposition wounds can reduce shelf life. Burrack et al. (2013) found that while SWD damages many soft fruits, including cherries, blackberries, and strawberries, in field production red raspberry is the preferred host. Raspberry had the lowest penetration resistance among the hosts sampled, which likely explains the preference. When SWD appeared in California it caused almost complete crop losses (Van Steenwyck and Bolda, 2015). An economic model suggests that crop losses in California without treatment could be 50% of the fruit and result in a 37% decrease in revenue for growers (Goodhue et al., 2011).

For many pests, understanding and predicting population stages and growth help in control. This is complicated for spotted wing drosophila, because so much about its development is highly variable depending on factors including temperature, relative humidity, nutrition source, and microbial interactions, and significant variation has been reported for development times (Tochen et al., 2014; Hamby et al., 2016). It is also hard to monitor population size with traps (Wiman et al., 2014).

Adult SWD are attracted to ripening fruit. Females pierce the skin and lay eggs under the wound, where larvae develop through three instars while feeding on the fruit (Asplen et al., 2015). Pupation nearly always occurs outside of the fruit, generally on the floor of the planting
Females go through a preoviposition phase before egg-laying. After this, females lay for their entire lifespan, which may result in the production of more than 400 eggs (Hamby et al., 2013). In some places they may have 13 generations a year (Asplen et al., 2015). These characteristics combine to make a pest that can exponentially multiply and reach very high populations very quickly. As the majority of the population is generally either in the larval or pupal stage, insecticide sprays can be ineffective (Woltz and Lee, 2017). Additionally, because of the number of generations, the risk of insecticide resistance is particularly high.

SWD appeared suddenly in the United States and spread very quickly, so developing approaches to control damage has been a challenge. Despite the aforementioned problems, most growers rely on insecticides, including organic insecticides, to control SWD. In response to SWD many growers switched from sprays based on the detection of pests to spraying on a calendar schedule, in some cases using an additional five to eight sprays per season (Van Timmerman and Isaacs, 2013). In California, seasonal treatment increased from 1.06 applications per hectare before infestation to 3.7 applications after (Van Steenwyck and Bolda, 2015). Some studies comparing conventional insecticide treatments to other practices have even found high levels of infestation following a standard spray schedule (van Timmerman and Isaacs, 2013; Rogers et al., 2016). For organic production where few products are available, efficacy is a challenge and growers risk exceeding maximum rates in a season (Hanson and Glick, 2013). Increased reliance on insecticides (including the development of new products) is potentially harmful to beneficials and natural enemies that control other pest species (Haye et al., 2016; Rogers et al., 2016). Secondary outbreaks of two-spotted spider mites, which are generally kept in check by predatory mites, were associated with these increased-spray programs (Hanson and Glick, 2013). In California increased insecticide use led to a 3.8 times increase in miticides for mite populations previously kept in check by beneficial arthropods. (Van Steenwyck and Bolda, 2015). Finally, because of the heavy reliance on a narrow range of products and the frequency of their
application, combined with the short generation time of SWD, the risk of insecticide resistance is high (Haye et al., 2016). For these reasons, complete dependence on insecticides is not an ideal solution. Integrating additional strategies into a SWD control program is important for efficacy, encouraging natural enemies, and postponing resistance.

**Development of IPM strategies for the control of spotted wing drosophila**

Developing IPM strategies in North America for spotted wing drosophila is challenging because much of the pest’s phenology and behavior was unknown outside of East Asia before its spread. Many different biological controls, including native North American parasites and predators have been investigated, but most only show moderate potential (Haye et al., 2016). Other possible tactics have included mass trapping, semiochemicals, and post-harvest treatment (Lee et al., 2011). None of these tactics have proved effective on their own. Meanwhile, SWD abundance in wild refuges keeps the population pressure high (Klick et al., 2016).

Several researchers have suggested that a push-pull system may be a successful control strategy. Push-pull describes an insect management strategy which combines taking steps to make the host crop less desirable and at the same time providing an attractant (another host or some sort of lure) (Cook et al., 2007). Klick et al. (2016) suggest that alternative host plants, while problematic because they create refuges, might be valuable for this purpose. Another approach is an attract and kill strategy using red spheres with a feeding stimulant and toxicant, like those used in orchards for apple maggot. A study exploring this technology found that the use of spheres significantly reduced SWD infestation compared to untreated plots, and also increased the effectiveness of insecticides on a conventional schedule (Rice et al., 2017a). Although push-pull strategies generally do not include pesticides, attracticidal spheres are likely to be highly target-specific, luring pests to one place where they are killed, rather than treating entire plantings with insecticides (Cook et al., 2007; Rice et al., 2017a).
There are a number of non-pesticide approaches to control SWD that may provide a good “push” in a push-pull system. Sanitation by removing excessive foliage and infested fruit is one strategy that has been suggested. SWD prefers cool, humid microclimates, and preventing such conditions is a step in limiting SWD populations (Hamby et al., 2016; Haye et al., 2016). Other cultural controls which show promise include exclusion netting and high tunnel production (Rogers et al., 2016). A discouraging effect from high tunnels is especially salient because many raspberry growers are already using them. When exclusion netting was compared to exclusion netting combined with a high tunnel, the high tunnel treatment had significantly lower counts. This was attributed to the high tunnel sustaining prolonged periods of temperatures above the development threshold for SWD, as well as a reduction in relative humidity (Rogers et al., 2016). In a study of conventionally treated high tunnel and field-grown raspberries, high tunnels also had lower SWD numbers, which was attributed to the extended efficacy of pesticides in the tunnel environment, as well as possible disruption of visual cues. The tunnels were covered with Luminance® plastic (Leach and Isaacs, 2018). This plastic diffuses radiation and is partially UV-A blocking (K. Demchak, unpublished data).

**Visual ecology of spotted wing drosophila**

While literature on the visual system of SWD is lacking, information about their foraging behavior may be informative. In addition, literature about the common fruit fly, *Drosophila melanogaster* may provide some understanding as well. Early research into the vision of *Drosophila melanogaster* showed that not only does this species have UV receptors, but that UV light is a vital aspect of stimulus for positive phototaxis (the movement towards a light source) (Schümperli, 1973). More recent work with *Drosophila melanogaster* shows that they are additionally sensitive to polarized light and that it serves as a behavioral stimulus as well, guiding flight (Wernet et al., 2012). We do not know whether polarized light and UV light serve as
stimuli for *Drosophila suzukii*. Further, should SWD be sensitive to these light qualities, research would be required to know whether light provides cues for navigation, host location, or reproduction. Sensitivity suggests that modifying spectral distribution might in some way disrupt SWD behavior, but this can only be theorized from what is currently known.

Studies of SWD behavior provide some information on how they might respond to light conditions. A review of studies of mating, foraging, and oviposition behavior at different points in a simulated day found a range of results. Overall, the majority of these studies found that these behaviors took place diurnally. For virgin females, the majority of activity occurred in the morning and evening, while mated females were most active during the afternoon. One unpublished study referenced by Hamby et al. (2016) found that female oviposition increased with increasing light intensity. However, these were laboratory experiments where light intensity and spectral composition may have been significantly different from natural conditions. Lab conditions tend to have much lower visible and UV radiation compared with natural conditions (Heath and Phelan, 2001).

Studies of SWD behavior in the field suggest that they might not depend on light for navigation. In contrast to the large amount of literature reporting Japanese beetle’s dependence on bright light for movement, the literature for SWD instead suggests that it may avoid sunlight in the field. A study of foraging preferences in red raspberry indicated that lower and more interior fruit were favored for oviposition (Rice et al., 2017b). While the authors theorized that this preference was related to lower temperatures and higher relative humidity in the low interior of the canopy, the results also suggest an avoidance of light, or indicate that it is not required for oviposition. Further, response to visual stimuli tended to have similar trends and occur in similar proportions in laboratory and semi-field experiments (Rice et al., 2016).

Because SWD is likely to be sensitive to UV-light and seems to require high light intensity for oviposition, but at the same time responds positively to visual stimuli in laboratory
settings and prefers to oviposit on low, interior fruit, it is not clear whether UV-blocking plastics might reduce infestations in high tunnels. It is important to know whether these plastics might affect SWD, and even a small effect may contribute to a push-pull system.

**Controlling SWD infestation using harvest interval**

An IPM technique that is likely easily integrated into high tunnel production is shortened harvest interval. A review of IPM strategies for SWD reported that harvesting every two days appeared to significantly reduce infestation in Swiss raspberry plantings compared to harvesting at longer intervals (Haye et al., 2016). Recent work in high tunnels found that shortening the harvest interval from 3 days to daily or every other day significantly reduced the infestation of raspberries. However, shortening the harvest interval did not eliminate infestation, and it was recommended that the shortened harvest interval be combined with pesticide application for greatest effect. It was also emphasized that daily harvest may be less economically feasible for growers both in terms of the labor to pick daily and the decrease in yields that comes from frequent harvest (Leach et al., 2017). While shortening the harvest interval is not a solution to SWD infestation on its own, it does provide another degree of control. It is a technique especially suited to tunnel production where it is possible to harvest in any weather and also maintain greater sanitation.

**Rationale and Significance**

Japanese beetles and spotted wing drosophila are two pests that significantly impact red raspberry production. While both are primarily controlled with insecticides, there are reasons why insecticide reliance is undesirable. These include a short residual activity, risks of developing resistance, risks of exceeding maximum seasonal allowances, impacts on non-target arthropods, and market preferences. It is crucial to find non-spray approaches to control these pests. A first
step is understanding how the technologies available to growers through UV-blocking high tunnel plastics impact these pests.

A second important step in controlling spotted wing drosophila is learning to integrate control tactics. High tunnel production is a system with new challenges and potentials and taking advantage of these is essential. The ability to do so will be furthered by investigating whether and how the plastic film impacts SWD. Additionally, investigating the use of attracticidal spheres, a technology that has proved extremely promising in field conditions, will be extremely informative for high tunnel production. Finally, it will be important to evaluate frequent harvest, which has been shown to be significant but not fully effective, when combined with other non-spray controls. Researching these approaches in combination will provide rich and valuable information for the development of future IPM approaches for SWD.

**Objectives**

The objectives of this study were 1) to evaluate the effect of UV-blocking high tunnel plastics on populations of Japanese beetle and spotted wing drosophila, 2) to investigate the effect of UV on SWD host-location behavior, and 3) to evaluate the degree of control provided by UV-blocking plastic in combination with the use of attract-and-kill technology and shortened harvest interval.
Chapter 2

USE OF UV-BLOCKING PLASTIC FOR THE CONTROL OF JAPANESE BEETLES IN PRIMOCANE-FRUITING RASPBERRIES GROWN IN HIGH TUNNELS

The purpose of this study was to evaluate the influence of plastics with different spectral transmittance on Japanese beetle presence on raspberries grown in high tunnels. While doing this, we also characterized several types of UV-blocking plastics currently available, including their transmittance properties in the UV-B, UV-A, and visible ranges.

Materials and Methods

High tunnels and experimental design. The two-year experiment was conducted at the Russell E. Larson Agricultural Research Center in Rock Springs, PA (latitude: 40-57’ 19” N, longitude 078-44’ 14” W) in 2016 and 2017. The study utilized 18 research-sized high tunnels (5.18 m by 10.67 m). The tunnels were manually vented with roll-up sides. They had fixed endwalls throughout this study which were covered with the same plastic as the tunnel tops and sides, with a door in the south end. They were arranged in six rows of three tunnels. In 2016 fifteen tunnels were used to evaluate five plastic coverings and the experimental design was a split-plot, with three replications. Plastic was considered the whole-plot and cultivar was the split-plot. Outside plants were placed in plots between the tunnels. In 2017 three additional tunnels were added to include three replicates of the six treatments (the no-plastic treatment plus five plastics) which were randomly reassigned to the three blocks. Two primocane-bearing red-raspberry cultivars,
‘Josephine’ and ‘Polka,’ were used. Bareroot plants (Nourse Farms, South Deerfield, MA) were grown in 11.36 L plastic nursery bags (Hydro-Gardens Inc., Colorado Springs, Co) in 2016, and repotted into 18.93 L bags in 2017. Plants were grown in a 2:1 peat:perlite medium and fertigated throughout the season with 20N-3.1P-16.6K general purpose fertilizer for alkaline water (Plant Marvel, Chicago Heights, IL), supplying a 100 ppm nitrogen constant feed. Each tunnel contained one row of each cultivar, with 12 plants per row; plants at the end of each row were considered guard plants and were not used for data collection. Plants were spaced on 0.3 m centers within row in 2016, and 0.45 m centers in 2017. Rows were spaced 2.6 m apart in both 2016 and 2017. Cultivars were randomly assigned to east and west sides of each tunnel. Plants were pruned and trellised to regulate canopy density and to facilitate harvest.

**Plastics.** Five plastics were used in this study. In 2015 percent transmittance of radiation for the plastics was determined with a StellarNet model EPP2000 (StellarNet, Inc., Tampa, FL) spectroradiometer calibrated to NIST sources at the USDA-ARS Appalachian Fruit Research lab in Kearneysville, WV. These measurements were used to validate the limited descriptions of transmittances from the plastic manufacturers, and to provide specifications when none existed. The plastics used in this experiment, along with abbreviations used to refer to them, included TuffLiteIV™ “TIV” (Berry Global, Inc, Evansville, IN), Ginegar SunSaver “GSS” (Ginegar Plastic Products LTD, Kibbutz Ginegar, Ginegar 30053, Israel), KoolLite Plus “KLP” (RKW Hyplast NV, Hoogstraten, Belgium), and two custom-manufactured experimental plastics, one UV-transparent “UVT” and one UV-opaque “UVO” (BPI-Visqueen, Stevenston, U.K.; currently available through Lightworks Poly, Lancashire, U.K.). The “no plastic” treatment was abbreviated to “NP.”
Field spectral transmittance. In 2018, spectral distributions within the tunnels were measured on cloudless days with an Apogee Model PS-300 spectroradiometer equipped with a cosine-corrected detector (Apogee Instruments, Logan UT). The sensor was deployed in the center of the tunnel, varying from the center by a maximum of 8 inches (20 cm) to avoid shadows from the tunnel structure, and at a height of 40 inches (1 m) above the ground. Light intensity (µmol/m²/sec) was measured within each tunnel, as well as between tunnels. The sensor was contained in a leveling fixture and thus was held level for each reading. The transmittance at each wavelength within the UV-B, UV-A, and visible light ranges were summed, and compared to outside light levels and were expressed as percent transmittance for each plastic within each of these ranges.

Temperature. Foliage temperatures were measured on 2 dates in 2017 with a Model MI-220 Infrared Radiometer Meter, with a narrow 18 degree field of view (Apogee Instruments, Logan, UT). The first measurement date was 28 June from 1030 HR to 1130 HR. On 21 Sept., temperature was measured both in the morning (1030-1130 HR) and in the afternoon (1435-1530 HR). The sun angle was such that radiation reaching foliage passed through the plastic. On both dates, foliage temperature was measured on both the east and west sides of each row.

Japanese beetle population monitoring. Japanese beetles on each row of plants were counted while being removed into resealable gallon plastic bags for later disposal. Beetles that flew away were also counted. The proportion of beetles that evaded capture varied between treatments and days, but the proportion was judged to not exceed 20% of the total beetles counted. Beetles were generally counted daily from 12 July until 30 Aug. 2016, for a total of 37 dates. On 18 and 19 July, beetles were counted and removed three times per day (0820 to 0910 HR, 1320 to 1410 HR, and 1520 to 1600HR) when it was noticed that beetle populations varied through the day. Counts
made within about an hour of solar noon were approximately 4 to 5 times higher than in the morning or late afternoon. Noon and afternoon counts may have been affected by counts taking place earlier in the day. Subsequent counts in 2016 were conducted near solar noon to maximize beetle removal while minimizing damage from feeding. Due to intense beetle-feeding damage on outside plants, the plants were moved into tunnels on 20 July, and data collection on those plants was terminated. Therefore, there were two sets of data in 2016. One contained all 6 treatments for the first 9 dates, whereas the later 28 dates did not contain the outside treatment. In 2017, counts began on 15 July and continued every 5 days, until 1 Sept., for a total of 11 dates. Beetles were counted between 1100 to 1700 HR on every date except 20 July when the count was conducted between 0900 and 1300.

Statistical analyses. Foliage temperature data were analyzed as a 2 x 6 factorial (2 sides of each row and 6 plastic treatments) by analysis of variance (ANOVA) with SAS’s GLIMMIX procedure (Littell et al., 2006), where block was considered a random effect, and row side and plastic were considered fixed effects. Cultivar and side of tunnel were confounded, so these variables were not analyzed for temperature effects. When interactions were significant, the levels of one factor were compared within each level of the other factor with the SLICEDIFF option in the LSMEANS statement.

Japanese beetle count data were also subjected to ANOVA with PROC GLIMMIX. When there were multiple counts made on a date (18 and 19 July), counts were summed to give a single count for that date. The two data sets in 2016 (the first 9 dates where there was the NP treatment and the final 28 dates without the NP treatment) were analyzed separately. When the cultivar x plastic interaction was significant, the SLICEDIFF option was used to compare cultivar means within each plastic, and means for plastics were compared within cultivars.
The 2017 daily Japanese beetle counts were analyzed by ANOVA as a 3-way factorial (2 cultivars x 6 plastics x 11 dates). In addition, cumulative beetle counts were calculated and data were analyzed as a 2 x 6 factorial with a repeated measures analysis with GLIMMIX.

Japanese beetle count data from 2016 and 2017 were subjected to regression with PROC MIXED and PROC REG. Percent transmittance of UVA, UVB, and visible light, were fitted using linear and quadratic models. MIXED was used specifying block as a random variable. REG was used to produce R-square values for models that were determined to be significant using MIXED.

Results and Discussion

Plastic characteristics.

Characteristics of the five plastics, grouped by manufacturer’s descriptions, are presented in Table 1.1. The NP treatment had 100% transmittance in every range, because there was nothing to filter wavelengths. TIV and UVT transmitted the highest percentages of UV-B, UV-A, and visible light. GSS and KLP transmitted medium amounts of UV-B, UV-A, and visible light, although KLP transmitted the lowest percentage of visible light among the plastics. UVO transmitted the least UV-B and UV-A (less than 7%) but transmitted similar levels of visible light compared with the other plastics (80.8% compared with 73.2-84.7%).

Table 1.1. The effect of 6 plastic treatments on transmittance of three ranges of solar radiation.

<table>
<thead>
<tr>
<th>Plastic</th>
<th>UV Characteristics</th>
<th>Diffusion</th>
<th>UVB</th>
<th>UVA</th>
<th>Visible</th>
</tr>
</thead>
<tbody>
<tr>
<td>NP</td>
<td>N/A</td>
<td>N/A</td>
<td>100</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>TIV</td>
<td>Non-blocking</td>
<td>Minimal</td>
<td>76</td>
<td>73</td>
<td>83</td>
</tr>
<tr>
<td>UVT</td>
<td>Non-blocking</td>
<td>Yes</td>
<td>79</td>
<td>74</td>
<td>83</td>
</tr>
<tr>
<td>GSS</td>
<td>Partial</td>
<td>Yes</td>
<td>66</td>
<td>57</td>
<td>84</td>
</tr>
<tr>
<td>KLP</td>
<td>Partial</td>
<td>Yes</td>
<td>53</td>
<td>45</td>
<td>73</td>
</tr>
<tr>
<td>UVO</td>
<td>Blocking</td>
<td>Yes</td>
<td>36</td>
<td>7</td>
<td>81</td>
</tr>
</tbody>
</table>
Temperature.

Plastic and side of the row had a significant impact on foliage temperatures. For each date/time measurement, plastic and side of the row were significant at the 5% level (Table 1.2). In the morning, temperatures were significantly warmer on the east side of the rows than the west, which was shaded. The interaction between plastic and side of the row was never significant. In each case, the plastic treatments were always warmer than the NP treatment (0.3-2.7 °C greater) but the difference was not always significant. Plants under UVT were consistently among the warmest in both the morning and the afternoon. TIV was among the warmest in the morning but was among the coolest in the afternoon. Differences between mean temperatures within any date and time were never greater than 3° C. The tunnels were partially vented when data were collected, and the plants were within 1.5 m of the open sides, which may explain the magnitude of difference between inside and outside temperatures. When Wien (2009) measured daytime air temperatures in 10 m-wide high tunnels covered with clear polyethylene plastic treated with IR-blocking material (Tufflite IV “IR”), temperature differences of 10 °C and greater were found between tunnels and outside temperatures. However, Wien noted that these differences were reduced through ventilation; the difference between outside and inside temperatures decreased as the size of ventilation opening increased. Our foliar temperature data support those of Wien (2009), as temperatures within tunnels were higher than outside the tunnels and temperature differences within the tunnels were minimal, but the five plastics had relatively small effects on temperature.
Table 1.2. The effect of 6 plastics on raspberry foliage temperature (°C) on three date/time combinations in 2017.

<table>
<thead>
<tr>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>NP</td>
<td>19.5a&lt;sup&gt;z&lt;/sup&gt;</td>
<td>20.3a</td>
<td>24.1a</td>
</tr>
<tr>
<td>GSS</td>
<td>20.3ab</td>
<td>22.1b</td>
<td>25.2bc</td>
</tr>
<tr>
<td>KLP</td>
<td>20.5abc</td>
<td>21.3b</td>
<td>25.0bc</td>
</tr>
<tr>
<td>UVO</td>
<td>20.9bc</td>
<td>21.7b</td>
<td>25.1bc</td>
</tr>
<tr>
<td>TIV</td>
<td>20.9bc</td>
<td>23.0c</td>
<td>24.4ab</td>
</tr>
<tr>
<td>UVT</td>
<td>21.5c</td>
<td>22.0b</td>
<td>25.7c</td>
</tr>
<tr>
<td>Row side</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>East</td>
<td>23.1a</td>
<td>23.3a</td>
<td>23.6a</td>
</tr>
<tr>
<td>West</td>
<td>18.1b</td>
<td>20.2b</td>
<td>26.2b</td>
</tr>
</tbody>
</table>

**ANOVA P-values**

<table>
<thead>
<tr>
<th></th>
<th>Plastic</th>
<th>Row Side</th>
<th>Interaction</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0.0002</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
<td></td>
</tr>
<tr>
<td></td>
<td>0.5271</td>
<td>0.2692</td>
<td>0.9976</td>
</tr>
</tbody>
</table>

<sup>z</sup> Lsmeans within columns followed by common letters do not differ at the 5% level of significance by Tukey’s test.

**Japanese beetle populations.**

In 2016 and 2017 Japanese beetle numbers were lower on raspberry plants grown inside high tunnels than on plants grown with no cover. Although in 2016 there were no significant differences between plastics on individual dates, plants under all plastics had fewer total beetles accumulated over the first 9 dates than plants with no cover (Table 1.3). For the first 9 collection dates, cumulative beetle counts were affected by plastic, cultivar, and the plastic x cultivar interaction (P<0.0001). Within ‘Josephine’, the NP treatment had significantly higher mean counts (182) than all plastics (<39), while the five plastics did not differ significantly. For ‘Polka’, NP had the highest mean counts (357) and within the plastics there were significant differences: TIV and UVT had higher counts than UVO. Within the NP treatment, ‘Polka’ had nearly twice as many beetles as ‘Josephine’.

When we analyzed the cumulative count from the entire season, plastic was significant (P=0.0041), but not cultivar (P=0.9814) or the cultivar x plastic interaction (P=0.677). TIV and UVT had more beetles than the other three plastics (Table 1.3).
Table 1.3. LSmeans for cumulative numbers of Japanese beetles collected from 12-plant rows of two raspberry cultivars grown in high tunnels with 6 covering treatments in 2016.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Cumulative 1st 9 dates</th>
<th>Cumulative Season Total</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>‘Josephine’</td>
<td>‘Polka’</td>
</tr>
<tr>
<td>NP</td>
<td>182a^2</td>
<td>* 357a</td>
</tr>
<tr>
<td>TIV</td>
<td>38b</td>
<td>64b</td>
</tr>
<tr>
<td>UVT</td>
<td>35b</td>
<td>57b</td>
</tr>
<tr>
<td>KLP</td>
<td>25b</td>
<td>37bc</td>
</tr>
<tr>
<td>GSS</td>
<td>17b</td>
<td>21bc</td>
</tr>
<tr>
<td>UVO</td>
<td>28b</td>
<td>8c</td>
</tr>
</tbody>
</table>

^2LSmeans within columns followed by common letters do not differ at the 5% level of significance during the first 9 dates, and for the season total, by slicediff in GLIMMIX. Asterisks indicate that cultivars within the plastic treatment differ at the 5% level.

The order and separation of means between the plastics did not change before and after the termination of the NP treatment. After 29 July (day of the year 210), there was a rapid increase in beetle counts (Fig. 1.1), but it did not coincide with the unavailability of the outside hosts, which suggests that it was part of a second wave of adult emergence, rather than increased pressure from beetles that would have otherwise been feeding on the NP plots. This suggests that the proximity of a susceptible host may not alter the effects of the plastics.

Although treatments influenced the cumulative Japanese beetle numbers, and results were similar for individual dates, treatments on any given date were not significantly different in 2016. Because Japanese beetles aggregate in response to feeding-induced volatiles (Potter and Held, 2002), and the frequency and timing of beetle removal can influence the attraction of additional beetles (Switzer and Cumming, 2014), we hypothesized that frequent beetle removal (in some cases multiple times a day) may have altered the process by which beetles locate hosts. Based on this reasoning, in 2017 we reduced our removal frequency to about every 4 days. In 2017 we had much higher counts of beetles overall, which may have been due to modifying the sampling method, or populations may have been generally higher in 2017.

In 2017 beetle counts were significantly affected by plastics and cultivars on individual dates, so we performed analyses on both daily counts as well as season totals. Cultivar was not significant (P=0.38), but plastic, cultivar x plastic, date, cultivar x date, date x plastic, and cultivar
x date x plastic were all significant (P<0.0001, <0.0001, <0.0001, 0.0095, <0.0001, and 0.0347, respectively). The NP treatment had the most dates where cultivars differed (4 out of the 11 dates), followed by TIV on two dates, and GSS on one date. For the NP treatment, populations on ‘Josephine’ were significantly higher than on ‘Polka’, but under TIV and GSS, ‘Polka’ had higher beetle counts. For all plastic treatments, cultivar differences were not significant on the last five dates. Similarly, differences between the plastics decreased over the season, although the NP treatment consistently had the highest counts. TIV and KLP tended to have the next highest counts, followed by UVT. GSS and UVO tended to have the lowest counts.

When the cumulative beetle counts for 2017 were analyzed, cultivar was not significant (P=0.38), while plastic and cultivar x plastic were significant (P<0.0001 and P=0.0035, respectively). For the NP treatment, ‘Josephine’ had about 36% more beetles than ‘Polka’ (Table 1.4). Among plastic covers, with the exception of UVO, ‘Polka’ generally had more beetles than ‘Josephine’, but the difference was significant for only TIV. Within cultivars, beetle counts were influenced somewhat differently by plastic treatments. ‘Josephine’ plants under KLP and TIV had more beetles than plants under GSS and UVO. ‘Polka’ plants under KLP and TIV had more beetles than the plants under the other three plastics and UVO had fewer beetles than all other plastics except GSS (Table 1.4).

Table 1.4. Cumulative numbers of Japanese beetles collected from 12-plant rows of two raspberry cultivars grown in high tunnels with 6 covering treatments in 2017.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>‘Josephine’</th>
<th>‘Polka’</th>
</tr>
</thead>
<tbody>
<tr>
<td>NP</td>
<td>897a²</td>
<td>658a</td>
</tr>
<tr>
<td>KLP</td>
<td>403b</td>
<td>439b</td>
</tr>
<tr>
<td>TIV</td>
<td>287b</td>
<td>429b</td>
</tr>
<tr>
<td>UVT</td>
<td>270bc</td>
<td>250c</td>
</tr>
<tr>
<td>GSS</td>
<td>138c</td>
<td>159cd</td>
</tr>
<tr>
<td>UVO</td>
<td>85c</td>
<td>35d</td>
</tr>
</tbody>
</table>

²Lsmeans within columns followed by common letters do not differ at the 5% level of significance by slicediff. Asterisks between cultivars indicate that the cultivars within that plastic treatment differ at the 5% level.
In 2016 and 2017, cumulative beetle counts followed similar patterns (Fig. 1.1). Beetle counts generally were related to UV-A light transmission characteristics measured via spectroradiometer in 2018. We consistently counted the fewest beetles under UVO, which transmitted less than 7% of UV-A light. GSS and KLP tended to have mid-range populations, and transmitted about 57% and 45%, respectively. The most beetles were counted under TIV and UVT, which transmitted between 72-74% of UV-A. However, in 2017 KLP had very high counts (among the highest). The LSmean for KLP was influenced by one tunnel in the first block with extremely high populations which persisted throughout the season. This tunnel was added to the experiment in 2017. It is possible that the tunnel was placed where more Japanese beetles were emerging, although this 2016 results suggesting that plastic effects are consistent regardless of proximity of hosts. It is also possible that KLP does not have a consistent impact on Japanese beetle.
Figure 1.1. Cumulative numbers of Japanese beetles counted on plants of two raspberry cultivars grown in high tunnels with six plastic treatments over time in 2016 and 2017.

2016:

2017:
Models were evaluated to determine the ability to predict season total Japanese beetle counts using the percent transmittance of UV-B, UV-A, and visible light. Visible light was not significant in the model, likely because all the plastics transmitted similar amounts of the visible range. UV-B, UV-A, and a quadratic term for UV-A explained a significant amount of the variation in 2016 ($R^2=0.64$, $p<0.0001$, $N=30$) (Fig. 1.2) and 2017 ($R^2=0.81$, $p<0.0001$, $N=36$) (Fig. 1.3). When cultivar was included in the model, it was not significant and it did not interact with other variables, therefore it was not included in the final model.

**Figure 1.2. The relationship between the total number of Japanese beetles per plot for two red raspberry cultivars and percent UVA transmittance in 2016. Regression model: Total Japanese beetles=503+$2.2(UV-A)+0.1(UV-A^2)-14(UV-B)$ ($R^2=0.64$, $p<0.0001$).**
Figure 1.3. The relationship between the total number of Japanese beetles per plot for two red raspberry cultivars and percent UVA transmittance in 2016. Regression model: Total Japanese beetles = 2644 + 28.2(UV-A) + 0.3(UV-A^2) - 78.4(UV-B) (R^2 = 0.81, p < 0.0001).

The same variables explain a significant amount of the variation in both years. One reason why the model has a higher R^2 value in 2017 than 2016 may be due to the presence of the outside treatment, which extended the range of values. Additionally, with the exception of KLP, there was less spread in the means of the different tunnels. In both years the beetle counts for KLP tended to fall above the range of predicted values, which further suggests that there might be a quality of this plastic that makes less discouraging to Japanese beetle.

Although UV-B was significant in the model, UV-A and UV-B transmittance by the plastics were highly related (R^2 = 0.99, p < 0.0001, N = 30) (Fig. 1.4). Plastics manufactured and sold as “UV-blocking” do not necessarily differentiate between the UV-B and UV-A ranges. However, these data show that as more UV-A is blocked, more UV-B is blocked, but decreases at a lower rate.
All plastics evaluated in this study significantly reduced the number of Japanese beetles on both cultivars, and differences between plastics were usually apparent. In addition to transmittance of UV-A radiation, there are a number of other ways that the plastics may alter the environment and influence Japanese beetle behavior. It is possible that diffusion, the scattering of solar radiation as it passes through the plastic, plays a role. By scattering the light, diffusing plastics may produce light environments that have less direct and intense light (Giacomelli and Roberts, 1993). TIV was the least diffusing plastic in our experiment. TIV and UVT transmitted very similar amounts of UV-B, UV-A, and visible light. In 2016 TIV had 9 to 17% more beetles than UVT (although it was not significant), and in 2017 TIV had significantly more (9 to 40%) beetles than UVT. Because Japanese beetle movement is encouraged by direct and bright light (Lacy et al., 1994; Heath et al., 2001), the increased proportion of diffused light might inhibit beetle activity.
The plastics also alter the amount of UV-B light entering the tunnels. Altering UV-B light in greenhouse environments impacts fungal growth and plant defense pathways (Raviv and Antignus, 2004). It is possible that blocking UV-B radiation may indirectly influence plant susceptibility to pests through the secondary production of defense compounds or volatiles. More research is required to determine whether a change in plant defenses might be responsible for reduced beetle populations or differences in cultivar susceptibility.

Temperature may also be a factor that influences beetle populations within the tunnels. Warm temperatures are conducive to beetle activity, with a minimum temperature of 27 °C for flight (Potter and Held, 2002), and activity increases with warmer temperatures (Heath et al., 2001). Our highest beetle numbers were in the NP treatment, which was the coolest, but had no physical barriers and provided the most intense light. Because the foliage temperatures only differed between plastics by 3°C overall, temperature seems unlikely to be a cause of the consistently observed differences between the plastics.

**Conclusions**

Considering the variables we measured that might influence Japanese beetle populations, the observed differences are most likely the result of beetles responding to the amounts of UV-A radiation under the plastics. These results support previous attempts to use UV-blocking plastic as an IPM tactic to reduce direct insect damage and virus transmission (Raviv and Antignus, 2004; Johansen et al., 2011). Our new findings regarding Japanese beetle have practical implications. Many growers are already using high tunnels for small fruit production in the Northeast and Midwest because of a wide range of benefits, including disease control and season extension (Demchak, 2009; Yao and Rosen, 2011; Hanson and Glick, 2013). Additionally, many utilize
organic farming practices, or are marketing directly to consumers (Carey et al. 2009), and are not able to use the conventional pesticides typically used for Japanese beetle control or may have a market that prefers fruit that is not treated with insecticides. For conventional growers insecticidal sprays are an additional cost and risk exposure for non-target and beneficial species. The use of a UV-blocking covering in raspberry production may help growers avoid sprays. Mean cumulative counts under UVO ranged from 34 (‘Josephine’) to 11 (‘Polka’) in 2016, and 85 (‘Josephine’) to 35 (‘Polka’) in 2017, representing a 75-94% reduction compared to non-UV-blocking plastic. This reduction may delay the time before a control is necessary, or even reduce control to one or two hand removals. We found that on many collection dates, beetles in UVO tunnels were so scarce that the raspberry canopies had to be scrutinized in order to find them.

While these results are extremely promising for raspberry growers, they may be useful for other crops as well. Protected culture has been increasingly used for cherry production and even other stone fruit (Lamont, 2009; Lang, 2009). Should UV-blocking plastics be used for other crops in high tunnels, research needs to address whether the effects of the plastics are consistent when covering larger areas. Increasing the size of a tunnel covered with a UV-blocking plastic did not decrease populations of thrips and whiteflies (Costa et al., 2003). However, these pests are typically controlled by UV-blocking plastics covering greenhouses rather than tunnels. Our research has demonstrated that Japanese beetles are affected by UV-blocking plastic, even without physical barriers to movement. Therefore, tunnels covering larger areas will likely have similar results, though research should be performed to test this hypothesis.

A drawback of the UV-blocking UVO plastic is that it is not commercially available in the U.S, although similar plastics are available. The GSS plastic, which is available for purchase, also reduced beetles significantly, in some cases similarly to UVO. This, or another blocker of UV-A light, might be a good alternative choice. There is a need for increased availability of UV-blocking plastics, and a need for increased objective testing and evaluation of the transmittance
properties of available plastics. The results of this study are promising for producers of raspberries and other crops susceptible to Japanese beetle, and the effects of blocking UV on other high tunnel crop pests should be studied.
Chapter 3

Spotted Wing Drosophila Foraging Behavior in UV-Positive and UV-Deficient Environments

The purpose of this study was to investigate the relationship between SWD foraging behavior and the composition of solar radiation in the environment. We approached this by monitoring adult SWD populations under six different plastic treatments which transmitted varying amounts of the UVA range. We also conducted foraging assays in laboratory and field cage bioassays. The amount of radiation in each of these experiments was characterized and used to compare results of monitoring and the bioassays.

Materials and Methods

Field-monitoring. The two-year experiment was conducted at the Russell E. Larson Agricultural Research Center in Rock Springs, PA (latitude: 40-57’ 19” N, longitude 078-44’ 14” W) in 2016 and 2017. This experiment utilized 18 research-sized high tunnels (5.18 m by 10.67 m). The tunnels were manually vented with roll-up sides. They had fixed endwalls throughout this study that were covered with the same plastic as the tunnel tops and sides, with a door in the south end. They are arranged in six rows of three tunnels. In 2016 fifteen tunnels were used to evaluate five plastic coverings, and the experimental design was a randomized complete block design with three replications. Outside plants were placed in plots between the tunnels. In 2017 three additional tunnels were added to include three replicates of the six treatments (the no-plastic
treatment plus five plastics), which were randomly reassigned to the three blocks. Two primocane-bearing red-raspberry cultivars, ‘Josephine’ and ‘Polka,’ were used. Bareroot plants (Nourse Farms, South Deerfield, MA) were grown in 11.36 L plastic nursery bags (Hydro-Gardens Inc., Colorado Springs, Co) in 2016, and repotted into 18.93 L bags in 2017. Plants were grown in a 2:1 peat:perlite medium and fertigated throughout the season with 20N-3.1P-16.6K general purpose fertilizer for alkaline water (Plant Marvel, Chicago Heights, IL), supplying a 100 ppm nitrogen constant feed. Each tunnel contained one row of each cultivar, with 12 plants per row; plants at the end of each row were considered guard plants and were not used for data collection. Plants were spaced on 0.3 m centers within row in 2016, and 0.45 m centers in 2017. Rows were spaced 2.6 m apart in both 2016 and 2017. Cultivars were randomly assigned to east and west sides of each tunnel. Plants were pruned and trellised to regulate canopy density and to facilitate harvest.

**Plastics.** Five plastics were used in this study and their light transmittance characteristics were previously described in the previous chapter (Cramer et al., 2019).

**Adult trap data.** Adult SWD were monitored with apple cider vinegar traps. Traps were made with plastic deli containers (0.95 l) (Plastic Packaging Corporation, Springfield, MA) with 3.6 mm holes drilled in the sides to allow SWD to enter and to exclude larger insects were suspended with wire from a center trellis line inside the raspberry foliage, to maximize humidity and darkness. One side of the container had no holes to facilitate emptying the contents. Traps were filled with approximately 250 ml of an apple cider vinegar and unscented dish soap solution. Every seven days trap contents were filtered through a mesh half-sphere and stored in vials with apple cider vinegar for later identification with a dissecting scope. Male and female SWD were
identified and counted, and numbers of other fruit flies and other insects were also counted and recorded. Traps were deployed from 1 Sept. to 16 Nov., 2016 and from 14 June to 31 Oct., 2017.

Statistical analysis. For each year, data were analyzed as a split-plot design by analysis of variance with SAS’s PROC GLIMMIX. Plastic was the whole-plot and cultivar was the split-plot. Block and block*plastic were considered random effects and plastic and cultivar were included in the model as fixed effects. The treatment structure was a 2 x 6 factorial (2 cultivars and 6 plastic treatments). Because no interactions were significant, lsmeans for main effects were compared with Tukey’s test.

Laboratory Bioassays. Behavioral assays were conducted at the USDA-ARS Appalachian Fruit Research lab in Kearneysville, WV. *Drosophila suzukii* from a laboratory colony reared at 25 ± 2° C, 50 ± 10% RH, and a 16:8 (L:D) photoperiod. The SWD were 7 to 14 days old, and sexually mature. The night before each experiment, 10 males and 10 females were sorted into separate 50 ml plastic drosophila vials (Fisher Scientific, Waltham, MA). Several drops of 20% sugar syrup with red dye were placed on the bottom of the cork as a food resource for the flies. They were stored at 20° C for 15-16 hrs before the experiment began the following morning. Vials were checked in the morning for mortality, and flies replaced as necessary.

Sentinel berries were produced by inserting open paper clips (#1 Skilcraft, Ira, MI) into the receptacle depression of clean red raspberries (Driscolls, Watsonville, CA), and secured with hot glue. Fruits were then covered with a thin coat of Tangle-Trap (The Tangle-Foot Company, Grand Rapids, MI) and were hung on wire rungs to avoid contact with other objects and stored in a refrigerator until use.

 Arenas consisted of 12 large bioassay containers (cubes with 18” (45.72 cm) sides), with an eye hook located in the back wall, 9” (22.86 cm) from each side, and 5” (12.7 cm) from the
top. Four 19x19” (48.26x 48.26 cm) sheets each of experimental “UVT” (UV-transparent) and “UVO” (UV-opaque) high tunnel film (BPI-Visqueen, Stevenston, U.K.; currently available through Lightworks Poly, Lancashire, U.K.), and pallet wrap, as a control, covered the tops of the containers. Arenas were randomly assigned to three shelving units, each of which had two shelves that could hold two arenas. Above each shelf were fluorescent light fixtures holding one bulb of Philips cool white F32T8/TL841 Alto II (Philips, Andover, MA) and one Repti-sun 10.0 UVB bulb (Zoo Med Laboratories, San Luis Obispo, CA). In the testing area, temperatures were maintained at 26 ± 2° C, and RH at 44-45%.

**Spectroradiographs of Light Environment.** UVB (280-315 nm), UVA (315-400 nm) and total visible light (400-700 nm) were measured with an Apogee Model PS-300 spectroradiometer equipped with a cosine-corrected detector (Apogee Instruments, Logan, UT). Light intensity (µmol/m²/sec) was measured in the center of each bioassay container, 50-54 cm from the bulbs, which were turned on. The sensor was placed on the floor of the container and leveled before measuring light under the control plastic (pallet wrap), and under each experimental plastic. The transmittance at each wavelength within the UV-B, UV-A, and visible light ranges was summed, and compared to the outside radiation light levels taken in the field cage experiment.

**Lightment Bioassays.** Flies and fruit were brought to room temperature before commencing the experiment. Fruits were hung from the eyehook in each arena. One vial each of male and female SWD was placed in each arena. The stoppers were removed to release flies and the plastic was sealed around the top of the arena to prevent escape. The light fixture then ran for 14 hours to simulate summer day-length. The following morning, after 20-24 hours, the number of flies of each sex at the release site, at the target (stuck to the berry), and elsewhere in the arena were
recorded. Vials were removed, arenas were wiped down with damp paper towels, arenas were re-ranidized on the shelves, and bioassays were repeated five times over a two-week period.

**Statistical Analysis.** Male and female SWD with positive host location were summed and divided by the total number of flies found in the arena to calculate the proportion of flies that successfully located the fruit. When the total number of flies in the arena were fewer than ten, the observation was deleted, as that indicated that flies were escaping, and these were not usable observations. Proportion data were analyzed as a randomized complete block design, with an analysis of variance using SAS’s GLIMMIX procedure. Rep was considered to be a block and was specified as a random effect and plastic was the fixed effect.

**Field Cage Bioassays.** Behavioral assays were performed at the USDA-ARS Appalachian fruit Research Lab in Kearneysville, WV with *Drosophila Suzuki* from the colony previously described. Fruits were also prepared and stored as previously described, but 15 male and 15 female SWD were sorted into each vial.

Two arenas consisting of 32-thread count mesh 1.8 m³ field cages were covered with the two high tunnel plastics (UVO and UVT) used for the alightment bioassays and one field cage was left uncovered as a control. A 60 cm gap was left at the bottom of the cages with plastic to allow air flow and prevent excessive heat buildup.

Single 2- to 4-year-old potted ‘Joan J’ raspberry plants (Nourse Nursery, Deerfield, MA) of similar height and foliage density were placed in the back-center of each field cage, inside a 101.6 cm metal tomato cage (W. Atlee Burpee and Company, Warminster, PA) with 6 tiers to provide locations for the sentinel berries to be hung. All ripe and ripening fruit were removed from the plants. Four sentinel berries were hung on each side of the cage on three tiers at about 50.8, 71.1, and 91.4 cm above ground. To simulate fruit in the interior of the plant, wire was strung bisecting
the center of the cage at each of these rail heights, and a fruit was hung in the center at each height. A total of 15 fruit were placed in each cage. Two 40-cm-tall plastic nursery pots were placed in the front corners of the cage to provide a release site for the spotted wing drosophila. Above each raspberry plant an EL-USB-2-PLUS datalogger (3DL Industries, Windham, NH) was hung within a 0.47 L drink cup, about 40.5 cm from the top of the field cage, and flush with the top of the raspberry plant. These were programmed to measure temperature and humidity once each minute; however, they failed to properly record data. A total of 60 flies were released into arenas by placing 1 vial each of male and female flies on both release sites and removing corks. After 22 to 23 hours, arenas were inspected. Numbers of dead and live adults of each sex at the release sites were recorded. Sticky berries were inspected and captures at each location were recorded. Locations were categorized as either being ‘interior’ (at the center of the plant) or ‘exterior’ (the four locations around the outside of the plant). This experiment was repeated three times over a one-week period.

*Spectroradiographs of Light Environment.* Light intensity (μmol/m²/sec) was measured under each treatment with an Apogee Model PS-300 spectroradiometer (Apogee Instruments, Logan, UT). Measurements were made on a cloudless day between 1100 and 1140 HR, under the three plastics, and with no covering. The sensor was placed in the center of the cage and leveled at a height of 1.0 m above the ground. The transmittance at each wavelength within the UV-B, UV-A, and visible light ranges was summed, and compared to outside light levels and expressed as percent transmittance for each plastic within each of these ranges.

*Statistical Analysis.* Numbers of flies on individual fruits were analyzed as a 3-way ANOVA (3 plastics x 3 heights x 2 locations) in a randomized complete block design using SAS’s GLIMMIX
procedure. Rep was the block and was specified as a random variable. Interactions were not significant, so Tukey’s HSD was used to compare means of significant treatments.

**Results and Discussion**

*Effects of cultivar on trap counts in high tunnels.*

In both 2016 and 2017 cultivar did not significantly affect the number of total SWD captured in traps (p= 0.56 and 0.51, respectively). Cultivar was also not significant for male SWD. For female SWD, cultivar was significant in 2016 (p=0.05) but not in 2017 (p=0.46). In 2016 there was a general trend of lower captures in the ‘Josephine’ plots under each plastic, and there was a significant difference in the number of female SWD captured in 2016 under TIV (Table 2.1). ‘Josephine’ had a lower mean total capture (39) than ‘Polka’ (80). This may have been the result of higher yields in ‘Polka’ (K. Demchak, personal communication), which could have attracted larger numbers of females for oviposition. Some literature suggests that fruit density may be correlated with the density of SWD (Rice et al., 2017a, Rice et al., 2017b), while this relationship did not exist in other studies (Burrack et al., 2013). Trap counts may also have been affected by plant architecture. ‘Josephine’ plants were taller and less dense than ‘Polka.’ The difference in growth habit may have made ‘Polka’ more favorable for SWD, although the trend was generally not significant.
Table 2.1 Female SWD trap captures in raspberry plants as influenced by cultivar and plastic treatments in high tunnels in 2016 and 2017.

<table>
<thead>
<tr>
<th>Plastic</th>
<th>‘Josephine’</th>
<th>‘Polka’</th>
<th>‘Josephine’</th>
<th>‘Polka’</th>
</tr>
</thead>
<tbody>
<tr>
<td>GSS</td>
<td>49b</td>
<td>73ab</td>
<td>963ab</td>
<td>1079a</td>
</tr>
<tr>
<td>KLP</td>
<td>48b</td>
<td>52b</td>
<td>959ab</td>
<td>609ab</td>
</tr>
<tr>
<td>NP</td>
<td>118a</td>
<td>111a</td>
<td>562b</td>
<td>407b</td>
</tr>
<tr>
<td>TIV</td>
<td>59b</td>
<td>79ab</td>
<td>925ab</td>
<td>902ab</td>
</tr>
<tr>
<td>UVO</td>
<td>35b</td>
<td>40b</td>
<td>1392a</td>
<td>998ab</td>
</tr>
<tr>
<td>UVT</td>
<td>39b</td>
<td>*</td>
<td>80ab</td>
<td>608b</td>
</tr>
</tbody>
</table>

*Means within columns followed by common letters do not differ at the 5% level of significance and asterisks between cultivars indicate that cultivars differed at the 5% within that plastic, by slicediff.

Effects of plastic film on trap counts in high tunnels.

Plastic treatment had a significant impact on the number of total SWD captured in traps in 2016 (p=0.0007) but not in 2017 (p=0.07). In 2016 and 2017 the differences between plastics were not significant at the 5% level. The NP treatment had significantly more SWD than all but TIV in 2016. UVO had more SWD than the NP treatment in 2017, and all other plastic treatments did not differ from the NP treatment (Table 2.2).

Table 2.2. The effect of six plastic treatments on total SWD trap captures in 2016 and 2017 in high tunnel raspberries.

<table>
<thead>
<tr>
<th>Plastic</th>
<th>Mean cumulative trap count</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>2016</td>
</tr>
<tr>
<td>NP</td>
<td>194a</td>
</tr>
<tr>
<td>TIV</td>
<td>114ab</td>
</tr>
<tr>
<td>GSS</td>
<td>96b</td>
</tr>
<tr>
<td>UVT</td>
<td>90b</td>
</tr>
<tr>
<td>KLP</td>
<td>74b</td>
</tr>
<tr>
<td>UVO</td>
<td>57b</td>
</tr>
</tbody>
</table>

*Lsmeans within columns followed by common letters do not differ at the 5% level of significance, by Tukey’s test.

Weather may have been an important factor in explaining why the NP treatment had higher counts than all the tunnels in 2016 but lower counts in 2017. During the experimental period temperatures were much higher in 2016 than in 2017. SWD have an optimum development temperature of 28°C, and it has been observed that tunnels may create unfavorable environments if outside temperatures are high (Rogers et al., 2016). High temperatures in the
tunnels in 2016 likely exceeded the optimum range. In 2017, with low temperatures and overcast weather through much of the season, the tunnels may have been more favorable to SWD.

SWD numbers under plastic treatments may have been affected by the confounding effects of cultivar and yield. Although yields for outside plants in 2016 were not included in the statistical analysis, in 2017 plants grown in tunnels had significantly (p<0.0001) higher yields than the NP plants. Compared to the plastics, NP yield for ‘Josephine was only 36.5% and ‘Polka’ was 23.9% (Demchak, K, unpublished data). In 2016 yields may have been more similar between the plants, because the plants were only one year old and had not accumulated the growth effects of high tunnel environment. Considering this difference in yield, NP plots may have been less attractive to SWD in 2016 due to a lack of fruit regardless of the light conditions in which they were growing. However, even if weather and yield were largely responsible for the differences in trap counts, it is unlikely that the UV-transmission of high tunnel plastics alone impacted SWD populations.

Plotting the total trap captures for each plot (plastic and cultivar combination) for 2016 vs. 2017 shows that the NP treatment was the only one with minimal variation, having consistently high captures in 2016 and consistently low captures in 2017 (Fig. 2.1).
Figure 2.1. Scatter plot for mean SWD trap count under 6 different high tunnel covering treatments in 2016 and 2017.

Effects of UV-light on SWD foraging in laboratory bioassays.

Figure 2.2. Mean proportion of alightments under three plastic treatments in laboratory bioassays.

Plastic did not significantly affect the proportion of SWD successfully locating hosts in these bioassays (p=0.47). It appears unlikely that the amount of UV in the bioassays in this experiment impacted foraging behavior (Fig. 2.2).

A possible reason for the lack of difference may have been the experimental conditions. While the arenas covered with UVO had lower UVA light than either the control or the UVT-covered arenas (Table
2.3), all arenas had much lower light levels than under natural sunlight (as measured in the field cage bioassay experiment).

Table 2.3. Photon flux in three ranges of the light spectrum in three treatments in laboratory conditions and in natural sunlight.

<table>
<thead>
<tr>
<th>Conditions</th>
<th>Photon flux (µmol/m²/sec)</th>
<th>Solar radiation transmitted (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>UVB (280-315 nm)</td>
<td>UVA (315-400 nm)</td>
</tr>
<tr>
<td>Sunlight</td>
<td>1.9</td>
<td>73.9</td>
</tr>
<tr>
<td>Control (Pallet wrap)</td>
<td>0.1</td>
<td>0.9</td>
</tr>
<tr>
<td>UVT</td>
<td>0.1</td>
<td>0.8</td>
</tr>
<tr>
<td>UVO</td>
<td>0.01</td>
<td>0.07</td>
</tr>
</tbody>
</table>

It is evident from the transmittance data that the light environment in the lab did not recreate the environment that SWD experiences when foraging outside. The low light levels and the altered ratios between wavelengths may have prevented the presence or absence of UV light from having any impact. It is clear, however, that unlike some insects for which laboratory conditions fail to elicit positive phototaxis, SWD can successfully forage. A study by Rice et al. (2016) on the impact of color and shape on SWD visual stimulus found similar levels and trends of response in laboratory and semi-field settings. Their data suggest that it is unlikely that the low levels of visible radiation and absence of UV radiation in laboratory settings would not alter SWD response to stimuli. This supports our results, as well as the conclusion that UV light does not impact SWD behavior.

Effects of UV-light on SWD foraging in field cages.

Plastic did not significantly affect the ability of SWD to locate target berries within field cages (p=0.67) Location and height were significant (p= 0.014 and 0.001, respectively), and there were no significant interactions. Fruit at the top of the plant had significantly more SWD than
fruit at the bottom of the plant, and neither differed from fruit at the middle height (Table 2.4).

Interior fruit had more SWD than exterior fruit.

<table>
<thead>
<tr>
<th>Fruit height</th>
<th>SWD/fruit</th>
<th>Fruit location</th>
<th>Mean SWD/fruit</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bottom</td>
<td>2.2a²</td>
<td>Exterior</td>
<td>2.1a</td>
</tr>
<tr>
<td>Middle</td>
<td>2.6ab</td>
<td>Interior</td>
<td>3.6b</td>
</tr>
<tr>
<td>Top</td>
<td>3.8b</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Values within columns followed by common lowercase letters do not differ at the 5% level of significance by Tukey’s test.

The interior location had the highest number of flies per fruit and was not affected by plastic. This is consistent with research showing that SWD tends to prefer interior fruit for oviposition. The interior of the plant is more likely to have the cool, humid environment which SWD prefers (Rice et al., 2017b). In our experiment the most SWD reached the highest fruit and the least reached the lowest fruit. This is not consistent with results of Rice et al. (2017b) where lower fruit had the most SWD in field cages and higher rates of oviposition in field trials.

As with the laboratory bioassays, the lack of difference between the light environments in the field cages could be responsible for the absence of a plastic effect. The field cage mesh alone drastically impacted light transmission. Although UVO almost completely blocked UVA light (3.07% transmission of solar radiation), UVT and the control had similar levels of transmission (30.23% and 38.44%, respectively) (Fig. 2.3, Table 2.5). Visible light could also impact foraging behavior. All treatments had similar transmission of visible light, ranging from 39% to 42%, all well below ambient levels.
Figure 2.3. Photon flux measured under three plastic coverings on field cages.

**UVO Field Cage**

**UVT Field Cage**
Table 2.5. Transmittance of three ranges of solar radiation as influenced by three plastic treatments on field cages.

<table>
<thead>
<tr>
<th>Plastic treatment</th>
<th>UVB (280-315 nm)</th>
<th>Solar radiation transmited (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>UV (315-400 nm)</td>
</tr>
<tr>
<td>UVO</td>
<td>23</td>
<td>3</td>
</tr>
<tr>
<td>UVT</td>
<td>43</td>
<td>30</td>
</tr>
<tr>
<td>Control (field cage only)</td>
<td>53</td>
<td>38</td>
</tr>
<tr>
<td>Sunlight</td>
<td>100</td>
<td>100</td>
</tr>
</tbody>
</table>

Although temperature data were not successfully recorded, it seems likely that temperature differences due to different degrees of ventilation between the treatments may be the most important environmental variable that could have impacted foraging behavior. While temperature may have influenced foraging, plastic treatment was not significant, and within the two locations where plastic affected SWD captures, the effects of plastic were not consistent. It is more likely that the significant interactions were due to low replications rather than plastic.

Conclusions

In this study, level of UV light in the environment did not significantly impact SWD foraging behavior. In 2016, UV-blocking plastics tended to deter SWD, similar to effects
found in research with other protected culture pests. However in 2017 trends were reversed. It is possible that monitoring with traps was not sufficiently precise to reflect actual differences in SWD populations. Previous research showed weak correlations between trap count, population, and larval infestation (Wiman et al., 2014).

The potential impact on trap captures of year-to-year yield differences of the cultivars under the different plastics is unclear. Both cultivars had significantly higher yields in tunnels than in the NP treatment, and this may have caused the higher trap captures in tunnels. A previous study showed poor correlation between yield and infestation rate (Burrack et al., 2013), while another suggests that fruit density may affect visual attraction (Rice et al., 2017b).

In our research it seems likely that differing weather conditions in the two years may have influenced differences in populations, as high tunnels in the first year had excessively high temperatures for SWD development. In some studies, SWD populations were low in high tunnels with high temperatures (Rogers et al., 2016).

The laboratory and field cage bioassays, although potentially flawed in their design, support the conclusion that UV light is unlikely to influence an individual SWD’s ability to forage successfully. This matches with previous studies of SWD response to visual stimuli, which found that there were positive responses in laboratory settings, and similar responses in laboratory and semi-field settings (Rice et al., 2016). Plastic treatment did not significantly affect the proportion or number of flies successfully locating sentinel fruits under our UVO, UVT, and control treatments.

While populations of a range of insects were low under UV-blocking greenhouse films in previous studies (Antignus, 2000; Antignus et al., 2001; Doukas and Payne, 2007), it appears that SWD populations were not affected by the plastics tested in the present study. However, further research to monitor SWD population trends under the different plastics over more years would
improve our understanding of the relationship between plastic, yearly weather fluctuations, and SWD populations. Additionally, monitoring fruit infestation levels would provide more valuable information.
Combining UV-Blocking Plastic, Attracticidal Spheres, and Harvest Interval for Non-Spray Control of Spotted Wing Drosophila

The purpose of this experiment was to evaluate three non-spray approaches for controlling SWD in high tunnel raspberries. Three variables were studied in combination in an attempt to increase the percentage of marketable yield or decrease SWD infestation.

Materials and Methods

High tunnels and experimental design. The experiment was performed at the Russell E. Larson Agricultural Research Center in Rock Springs, Pennsylvania, in 2018. The tunnel design and the plastics used to cover the high tunnels were previously described in detail (Cramer et al., 2018). Three tunnels were covered with one of two plastics varying in their ability to transmit UV light. Each tunnel was divided cross-wise into two halves by vertically suspending 0.35 mm x 0.35 mm ProtekNet™ insect netting (Nolt’s Produce Supplies, Leola, PA) from ceiling to floor and treatments of attracticidal spheres or no spheres were randomly assigned to each half. Each tunnel half contained two parallel plots of 8 raspberry plants of the primocane-bearing cultivar ‘Josephine.’ Bareroot plants (Nourse Farms, South Deerfield, MA) were started in 11.36 L plastic nursery bags (Hydro-Gardens Inc., Colorado Springs, Co) in 2016, and repotted into 18.93 L bags in 2017. Additional plants were planted into 18.93 L bags in 2017 to serve as guard plants and placed at the ends of each plot. Plants were grown in a 2:1 peat:perlite medium and fertigated throughout the season with 20-7-20 general purpose fertilizer for alkaline water (Plant Marvel,
Chicago Heights, IL), supplying a 100 ppm nitrogen constant feed. One row of the plants in each tunnel half were harvested daily and the other half was harvested three times per week. The experimental design was a split-split-plot, where plastic tunnel covers was the whole plot, attracticidal spheres was the split-plot, and harvest interval was the split-split-plot. Each combination was replicated three times.

**Plastics and spectral transmittance.** Two custom-manufactured experimental plastics were used in this study: one UV-transparent (UVT) and one UV-opaque (UVO) (BPI-Visqueen, Stevenston, U.K.; currently available through Lightworks Poly, Lancashire, U.K.). In 2015 percent transmittance of radiation for the plastics was determined with a StellarNet model EPP2000 (StellarNet, Inc., Tampa, FL) spectroradiometer calibrated to NIST sources at the USDA-ARS Appalachian Fruit Research lab in Kearneysville, WV. These measurements were used to provide specifications for future studies. In 2018, spectral distributions within the tunnels were measured on cloudless days with an Apogee Model PS-300 spectroradiometer equipped with a cosine-corrected detector (Apogee Instruments, Logan UT). The sensor was placed in the center of the tunnel, varying from the center by a maximum of 20 cm to avoid shadows from the tunnel structure, and at a height of 1.0 m above ground. Light intensity (µmol/m²/sec) was measured within each tunnel, as well as outside between tunnels. The sensor was contained in a leveling fixture and thus was held level parallel to the ground for each measurement. The transmittances at each wavelength within the UV-B, UV-A, and visible light ranges were summed and expressed as percent transmittance compared to ambient measurements between tunnels.

**Attracticidal spheres.** Spheres consisted of two parts: the flat-topped red plastic base (Great Lakes IPM, Vestaburg, MI), and sphere caps that were manufactured for this experiment at the USDA-ARS Appalachian Fruit Research lab in Kearneysville, WV, following the procedure outlined by
Rice et al. (2017a). Sphere caps contained the pesticide Delegate (Dow AgroSciences, Indianapolis, IN) at 1% by weight, which causes mortality upon feeding. One sphere was deployed for every two plants, for a total of four spheres per plot, or eight spheres in each tunnel half with the sphere treatment. Two spheres were hung from trellis wires on each side of the row. From 21 to 31 Aug. spheres were hung at about 0.85 m and from 17 to 28 Sept. they were hung at about 1.33 m. Spheres were misted with a spray bottle (L. G. Sourcing, N. Wilkesboro, NC) on low-humidity days when the spheres appeared visibly dry. Otherwise, on rainy or high-humidity days, spheres remained wet and did not need to be misted. Between the Aug. and Sept. data collection, spheres were removed, caps discarded, the plastic bases cleaned, and a new set of caps applied.

Harvest interval. Harvest interval was based on previous literature (Haye et al., 2016; Leach et al., 2017). Treatments were daily harvest and three times per week harvest (Mon., Weds., Fri.), the standard harvest interval at the research farm. All fully-colored fruit that easily detached from the receptacle were harvested by two assistants trained in this harvesting technique. Fruit was harvested at these intervals for two 12-day periods (21-31 Aug. and 17-28 Sept.).

Yield data. During harvest periods, fruit was sorted into marketable and marketable categories. Marketable fruit was firm and unblemished, while any fruit that had damage, was not properly formed, or was soft (indicating SWD infestation) was considered unmarketable. For each date, the weight (g) of marketable fruit and unmarketable fruit in each treatment, and mean fruit weight estimated from a 50-fruit subsample, were recorded for each plot (combination of plastic, sphere, and harvest interval).
Monitoring fruit infestation. The saline-float method outlined by Van Timmerman et al. (2017) was used to monitor fruit infestation. Samples of up to 50 marketable fruit were placed in resealable 3.8 L freezer bags. The fruit were gently crushed and 250 to 500 ml of salt solution was added to each sample (312.6 g non-iodized salt for 3.8 L water). The samples rested one hour before filtering through 5 mm hardware cloth placed in a plastic canning funnel (Nopro, Inc, Everett, WA) to catch fruit, and then through a reusable metal basket coffee filter (Medelco Inc, Bridgeport, CT) to catch SWD larvae and eggs. The coffee filter was then examined under a dissecting microscope, and the numbers of eggs, first-, second-, and third-instar larvae were counted. First- and second-instar larvae were not differentiated. Infestation rates were calculated by dividing the total number of larvae by the number of fruit in the sample. Eggs are not reliably separated from the fruit in this method (Van Timmerman et al., 2017), so egg data were not subjected to analysis.

From 24 to 28 Sept. unmarketable fruit were also tested for larval infestation following the same procedure outlined above, except that a maximum of 30 fruit was used after 24 Sept., because larger samples tended to drain extremely slowly.

Monitoring adult SWD. Adult SWD were monitored throughout the experiment with apple cider vinegar traps. SWD were first captured on 19 July although they may have been present before this. Traps consisted of 0.95 l plastic deli containers (Plastic Packaging Corporation, Springfield, MA) with 3.6 mm holes in the side to allow SWD to enter and exclude larger insects. One side of the container had no holes to facilitate emptying the contents. Approximately 250 ml of apple cider vinegar and unscented dish soap solution was added to each trap. Traps were suspended with wire from a center trellis line inside the raspberry canopy to maximize humidity and shade. Approximately every seven days trap contents were filtered through a mesh half-sphere and stored in labeled vials with apple cider vinegar for later identification. Contents were examined
under a dissecting scope and male and female SWD, as well as other fruit flies and other insects, were counted.

Statistical analyses. Trap data were analyzed as a split-split-plot design by analysis of variance with SAS’s PROC GLIMMIX. Plastic was the whole-plot, sphere treatment was the split-plot, and harvest interval the split-split-plot. The treatment structure was a $2 \times 2 \times 2 \times 6$ factorial (2 levels of each treatment and 6 collection dates).

Data for yield and SWD infestation from the daily harvest plots were combined to obtain the same number of observations for both harvest-interval treatments. The first date of each of the two-week data collection periods was not included in analysis, because there would have not yet been an effect from the harvest interval treatment. Data from each two-week harvest period were analyzed as separate experiments. These data were tested for equal variances with all combinations of harvest date, sphere treatment, harvest interval, and plastic treatment (4-way factorial) by requesting absolute residuals with SAS’s PROC GLM. Levene’s test was used to evaluate homogeneity of variances at the 1% level of significance. When variances were not equal, analysis of variance (ANOVA) was performed with PROC MIXED using a heterogeneous variance model. When interactions were not significant, the F-test from the ANOVA was used to test equality of main effect means. When interactions were significant, the slicediff option in the Lsmeans statement was used to test the hypothesis that the two levels of one factor within each level of the other factor were equal.
Results and Discussion

Monitoring adult SWD.

The number of SWD per trap varied significantly with date and the differences between the dates corresponded to the management of the tunnels (Fig. 3.1). SWD counts were very high when monitoring began on 23 Aug. following a period of lengthened harvest interval due to labor constraints. Attracticidal spheres were not deployed and fruit were not consistently harvested before the first experiment began. At the beginning of the experiment spheres were deployed and plots were thoroughly and frequently harvested. Corresponding with this change in management, trap counts dropped dramatically. The second population spike on 20 Sept. was towards the beginning of data collection for experiment 2, when raspberry yields were beginning to increase again after heavy pruning. The fluctuation in the amount of ripe fruit in the tunnels may have influenced SWD trap capture.

Figure 3.1. Mean SWD per trap over time during two data collection periods.
Plastic, harvest interval, sphere treatment, and date all significantly affected the number of SWD captured in vinegar traps (p=0.04, <0.0001, 0.03, and <0.0001, respectively). There were no significant interactions. Only sphere treatment and date significantly affected male SWD. Fewer total SWD were captured in daily harvest treatments compared to three times a week (Fig. 3.2). Ripe fruit in daily harvested plots remained on the plant for a shorter time and there were fewer fruit at any given time, which might have made these plots less attractive to female SWD. Traps from daily harvested plots had about 25% fewer SWD despite there being no barriers to movement between plots with different harvest intervals. While our data may suggest that fruit density significantly impacts adult populations, there was a poor correlation between fruit density and fruit infestation in a previous study (Burrack et al., 2013).

**Figure 3.2. Effect of harvest interval, plastic, and attracticidal spheres on total SWD per trap.** Number SWD per trap was significantly affected (P=0.05) by three all variables.

The difference in trap captures under the different plastics suggests that UVO plastic may be less favorable than UVT to SWD. Data from chapter 2 of this thesis show that it is unclear whether UV-blocking plastic affects SWD behavior, although many previous studies reported UV-deficient environments decrease various insect species. Traps in plots with spheres consistently had fewer male, female, and total SWD. The presence of spheres in a tunnel reduced mean trap counts more than plastic type or harvest interval.
While trap capture is not a precise measurement of SWD populations or fruit infestation, it does provide a general indication of populations levels within the crop canopy and may be useful for interpreting yield and infestation data.

*Effects of plastic, harvest interval, and sphere treatments on raspberry yields.*

The daily harvest treatment had significantly higher percentages of marketable fruit compared with harvesting three times per week in both experiments (p<0.0001). In Experiment 1 the mean proportion of marketable fruit for plots harvested three times a week was 49% and 72% for plots harvested daily (Table 3.1). In Experiment 2 the difference between the harvest interval treatments was smaller, but the averages were still significantly different (55% vs. 66%) (Table 3.2). For both experiments date was significant (p<0.0001) as well as the interaction of harvest interval x date (p=0.01 and <0.0001, respectively). For Experiment 1, the proportion of marketable fruit from daily harvest declined in a curvilinear manner over time from 79 to 59%, while it declined from 50 to 45% for the three-day harvest interval. For Experiment 2, the proportion of marketable fruit slightly increased for daily harvest, but generally fell for the three-day harvest interval (Appendix: Supplementary Figures, Fig. 4.1).
Table 3.1. Exp. 1 ANOVA results.
Main effects lsmeans

<table>
<thead>
<tr>
<th>Plastic</th>
<th>Plastic</th>
<th>Total Yield (g)</th>
<th>Marketable Yield (g)</th>
<th>%Marketable</th>
<th>Mean Fruit Weight (g)</th>
<th>Larvae/fruit</th>
</tr>
</thead>
<tbody>
<tr>
<td>UVO</td>
<td></td>
<td>279.2</td>
<td>163.5</td>
<td>60%</td>
<td>3.4</td>
<td>0.53</td>
</tr>
<tr>
<td>UVT</td>
<td></td>
<td>292.4</td>
<td>180.3</td>
<td>62%</td>
<td>3.4</td>
<td>0.57</td>
</tr>
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</table>

<table>
<thead>
<tr>
<th>Spheres</th>
<th>Spheres</th>
<th>Total Yield (g)</th>
<th>Marketable Yield (g)</th>
<th>%Marketable</th>
<th>Mean Fruit Weight (g)</th>
<th>Larvae/fruit</th>
</tr>
</thead>
<tbody>
<tr>
<td>Yes</td>
<td></td>
<td>278.5</td>
<td>165.0</td>
<td>60%</td>
<td>3.4</td>
<td>0.46</td>
</tr>
<tr>
<td>No</td>
<td></td>
<td>293.2</td>
<td>178.7</td>
<td>61%</td>
<td>3.3</td>
<td>0.64</td>
</tr>
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</table>

<table>
<thead>
<tr>
<th>Interval</th>
<th>Interval</th>
<th>Total Yield (g)</th>
<th>Marketable Yield (g)</th>
<th>%Marketable</th>
<th>Mean Fruit Weight (g)</th>
<th>Larvae/fruit</th>
</tr>
</thead>
<tbody>
<tr>
<td>Daily</td>
<td></td>
<td>286.8</td>
<td>206.0</td>
<td>72%</td>
<td>3.3</td>
<td>0.45</td>
</tr>
<tr>
<td>3xs</td>
<td></td>
<td>284.9</td>
<td>137.7</td>
<td>49%</td>
<td>3.5</td>
<td>0.65</td>
</tr>
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</table>

Interaction lsmeans

<table>
<thead>
<tr>
<th>Plast.</th>
<th>Int.</th>
<th>Sphere</th>
<th>Total Yield (g)</th>
<th>Marketable Yield (g)</th>
<th>%Marketable</th>
<th>Mean Fruit Weight (g)</th>
<th>Larvae/fruit</th>
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</thead>
<tbody>
<tr>
<td>UVO</td>
<td>3xs</td>
<td>No</td>
<td>285.5</td>
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<td>51%</td>
<td>3.5</td>
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<td>0.44</td>
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<tr>
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<td>Daily</td>
<td>Yes</td>
<td>279.7</td>
<td>200.6</td>
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<td>3.2</td>
<td>0.34</td>
</tr>
<tr>
<td>UVT</td>
<td>3xs</td>
<td>No</td>
<td>299.0</td>
<td>149.2</td>
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<td>3.6</td>
<td>0.75</td>
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<tr>
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<td>201.1</td>
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</table>

P-values from ANOVA

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<tr>
<th>Effect</th>
<th>Total Yield (g)</th>
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<th>%Marketable</th>
<th>Mean Fruit Weight (g)</th>
<th>Larvae/fruit</th>
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<tbody>
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<td>Plastic</td>
<td>0.314</td>
<td>0.047</td>
<td>0.149</td>
<td>0.889</td>
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<tr>
<td>Sphere</td>
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<td>0.348</td>
<td>0.053</td>
<td>0.002</td>
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<tr>
<td>Interval</td>
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<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
<td>0.0004</td>
</tr>
<tr>
<td>Plastic*sphere</td>
<td>0.018</td>
<td>0.060</td>
<td>0.117</td>
<td>0.643</td>
<td>0.264</td>
</tr>
<tr>
<td>Plastic*interval</td>
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<td>0.116</td>
<td>0.821</td>
<td>0.264</td>
<td>0.752</td>
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<td>Sphere*interval</td>
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<td>0.686</td>
<td>0.382</td>
<td>0.170</td>
<td>0.211</td>
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<tr>
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<td>0.106</td>
<td>0.064</td>
<td>0.371</td>
<td>0.653</td>
</tr>
<tr>
<td>Date</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
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Table 3.2: Exp. 2 ANOVA results.
Main effects lsmeans

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<th>Marketable Yield (g)</th>
<th>%Marketable</th>
<th>Mean Fruit Weight (g)</th>
<th>Larvae/fruit</th>
</tr>
</thead>
<tbody>
<tr>
<td>UVO</td>
<td>199.0</td>
<td>108.6</td>
<td>58%</td>
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<tr>
<td>UV T</td>
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<td>116.4</td>
<td>62%</td>
<td>3.3</td>
<td>0.31</td>
</tr>
<tr>
<td>Spheres</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>208.1</td>
<td>117.5</td>
<td>61%</td>
<td>3.3</td>
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<tr>
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<td>205.3</td>
<td>107.5</td>
<td>60%</td>
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<td>0.50</td>
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<tr>
<td>Interval</td>
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<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Daily</td>
<td>204.6</td>
<td>143.4</td>
<td>66%</td>
<td>3.3</td>
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</tr>
<tr>
<td>3xs</td>
<td>208.8</td>
<td>81.50</td>
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Interaction lsmeans

<table>
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<tr>
<th>Plast.</th>
<th>Int.</th>
<th>Spheres</th>
<th>Total Yield(g)</th>
<th>Marketable Yield (g)</th>
<th>%Marketable</th>
<th>Mean Fruit Weight (g)</th>
<th>Larvae/fruit</th>
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<tr>
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<td>76.60</td>
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<td>0.34</td>
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<tr>
<td>UVO</td>
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<td>198.4</td>
<td>144.4</td>
<td>65%</td>
<td>3.3</td>
<td>0.26</td>
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<tr>
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<td>No</td>
<td>234.2</td>
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<tr>
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<td>No</td>
<td>166.9</td>
<td>109.2</td>
<td>67%</td>
<td>3.2</td>
<td>0.34</td>
</tr>
<tr>
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<td>Yes</td>
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<td>94.50</td>
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<td>3.5</td>
<td>0.13</td>
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<tr>
<td>UVT</td>
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<td>Yes</td>
<td>231.8</td>
<td>168.9</td>
<td>67%</td>
<td>3.3</td>
<td>0.28</td>
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P-values from ANOVA

<table>
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<tr>
<th>Effect</th>
<th>Total Yield(g)</th>
<th>Marketable Yield (g)</th>
<th>% Marketable</th>
<th>Mean Fruit Weight (g)</th>
<th>Larvae/fruit</th>
</tr>
</thead>
<tbody>
<tr>
<td>Plastic</td>
<td>0.223</td>
<td>0.233</td>
<td>0.091</td>
<td>0.148</td>
<td>0.047</td>
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<td>Sphere</td>
<td>0.825</td>
<td>0.121</td>
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<tr>
<td>Interval</td>
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<td>&lt;0.0001</td>
<td>0.663</td>
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<tr>
<td>Plastic*sphere</td>
<td>0.049</td>
<td>0.002</td>
<td>0.550</td>
<td>0.046</td>
<td>0.687</td>
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<tr>
<td>Plastic*interval</td>
<td>0.041</td>
<td>0.012</td>
<td>0.601</td>
<td>0.009</td>
<td>0.041</td>
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<tr>
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<td>0.150</td>
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<tr>
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</tr>
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<td>0.050</td>
<td>&lt;0.0001</td>
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</table>
Total yields (marketable and unmarketable fruit for each plot) were higher for Experiment 1 than for the second experiment, possibly because plants were pruned at the conclusion of Experiment 1, removing some of the future crop. Low temperatures in Sept. also likely delayed fruit ripening in Experiment 2. In Experiment 1, the plastic x sphere interaction significantly affected total yield (p=0.02) (Table 3.1). In the second experiment, this was again significant with the addition of the plastic x interval interaction (p=0.04). None of the treatments were significant by themselves.

Although the plastic x sphere interaction was significant, yields under the two plastics for the two experiments tended to change similarly. For both plastics in both experiments, yield declined from the first to second harvest date, then increased again before declining (Appendix: Supplementary Figures, Fig. 4.2). The sphere*plastic effect was unlikely to be the result of the spheres affecting pollinators. Field trials with attracticidal spheres have found almost no impact on pollinators (T. Leskey, personal communication). Further, all of the aspects of plant growth that affect total yield (cane vigor, number of flowers, and number of druplets per fruit) would have been determined prior to the spheres being placed in the tunnels.

When total yield for plots in Experiment 1 were plotted against total yield for the same plots in Experiment 2 there was a general linear relationship (Fig. 3.3), indicating certain plots had high yields in both experiments. Since sphere and harvest interval treatments were re-randomized for each experiment, the sphere x plastic interaction may have resulted from confounding effects with high- and low-yielding plots.
Total marketable yield in Experiment 1 was significantly affected by plastic and interval, but not spheres (p= 0.047, <0.0001, and 0.06, respectively) (Table 3.1). In Experiment 2 harvest interval was significant (p<0.0001), as were the interactions of plastic*sphere, plastic*interval, and sphere*interval (Table 3.2). In both experiments marketable yield was always higher for plots harvested daily than for those harvested at three-day intervals (206 g versus 138 g in Experiment 1, and 143 g vs 82 in Experiment 2). These data contradict a previous report where daily harvested plots had lower yields compared to longer harvest intervals (Leach et al., 2017). The authors attributed higher yields to increased fruit size in less frequently harvested plots. Difference in grading fruit may explain this discrepancy. Although fruit from less frequently harvested plots may have been larger, it was also soft. We assumed that soft fruit was infested with SWD larvae and was therefore considered unmarketable.
Although UVT plastic had significantly higher marketable yield than UVO plastic in Experiment 1, there was no difference in Experiment 2. In a related high tunnel experiment designed to test five different plastics, raspberry yields for the entire season were affected by plastic, although the plastics used in this experiment were not significantly different from each other (K. Demchak, personal communication). In that experiment in 2017, the cultivar ‘Polka’ had higher yields under TIV compared to the other plastics tested.

Mean fruit weight was affected by interval in Experiment 1 (p<0.0001) (Table 3.1). In Experiment 1, plots harvested more frequently consistently had lower mean fruit weight, but the difference was only 0.2 g (3.5 g vs. 3.3 g). This difference may be significant for growers who are filling containers for wholesale but may not make as great of a difference for a smaller grower or a pick-your-own operation. Plots with attracticidal spheres had nearly significant higher fruit weight, but again the difference was likely not economically important. In Experiment 2 there were no main effect differences (Table 3.2). Similarly, the significant interactions between plastic and harvest interval and plastic and spheres represented very small differences in fruit weight. Therefore, any differences in fruit weight attributable to the variables tested in this study are probably not commercially important.

Effect of plastic, harvest interval, and sphere treatments on infestation of marketable fruit.

In the first experiment, the number of larvae per fruit that was considered marketable was significantly affected by sphere treatment and harvest interval (p=0.002 and 0.0004, respectively) (Table 3.1). In Experiment 2 plastic, sphere, and harvest interval were significant (p=0.047, <0.0001, and 0.029, respectively) (Table 3.2).

In Experiment 1, the presence of spheres reduced the mean number of larvae per marketable fruit from 0.65 larvae per fruit to 0.45. In Experiment 2, the means were 0.50 and 0.25 larvae per fruit. Infestation was greater in Experiment 1 generally, which was consistent with the
data on adult SWD numbers obtained from the vinegar traps. Temperatures were warmer in Aug.
favoring population growth. In the second experiment, cooler temperatures and lower populations
may have reduced infestation over all. In fact, on one date in the second experiment no larvae
were extracted from fruit from plots with spheres.

These results correspond with those of previous research in field raspberries, which
showed that attracticidal sphere treatments significantly reduced the number of larvae per fruit
compared with nontreated controls. In that study, fruit from nontreated plots had more than 4
larvae per fruit, whereas fruit from the plots with spheres had fewer than 2 per larvae per fruit
(Rice et al., 2017a). Although infestation levels were lower in our trial, possibly due to more
frequent harvest, the treatment effects were similar.

In previous experiments spheres were used in field settings and relied on rainfall and dew
to moisten the sugar and toxicant substances. Adult SWD feed on the liquid sugar and insecticide
coating on the sticky cap of the sphere (Rice et al., 2017a). We hypothesized that the dry
environment in tunnels might reduce the efficacy of the spheres. Our results suggest that misting
the spheres on dry days was sufficient to keep them activated and that this technology is
compatible with high tunnel production. We also found significant impacts with a sphere
deployment rate of one sphere for every two plants, although the plants were very large. We
suspect that increasing the density of spheres in high tunnels with very large plants would further
decrease infestation, though this assumption needs to be verified by further research.

The number of SWD larvae recovered from marketable fruit was influenced by
harvest frequency. In Experiment 1 the mean larvae per fruit was 0.64 and 0.46 for daily and
three-day harvest, respectively. In Experiment 2 the number of larvae per fruit was 0.44 and 0.31.
Our finding that harvest interval affects the number of larvae per fruit is supported by previous
research (Leach et al., 2017). The practice of daily harvest may not be justified by the small
differences in the numbers of larvae per fruit in our experiment, but daily harvest also increased
the proportion of marketable fruit and total marketable yields, which would likely make this practice economically desirable.

Infestation was significantly affected by date in both experiments (p<0.0001). For both experiments, and with both the sphere and harvest interval treatment, infestation decreased over the 5 dates analyzed. This suggests that the effects of the treatments increase over time. If the treatments were not removed and re-assigned during the two-week period between the experiments the means in Experiment 2 might have been lower. This may be important for growers who would be likely to use these treatments over a season.

In Experiment 2, the sphere x harvest interval interaction indicates that daily harvest decreased the number of SWD larvae per marketable fruit, in the absence of spheres, but not when combined with spheres (Fig. 3.4). This suggests that growers using spheres may not need to harvest as frequently to reduce the number of larvae per fruit, but the sizable increase in marketable yield still makes daily harvest an economically important practice. It is possible that this interactive effect has to do with the method of grading fruit. If all fruit considered marketable had very low larval infestation, then the differences between treatments would be more apparent in the proportion of marketable fruit. It is also important to note that in Experiment 1, there was not a significant interaction and the effects of sphere and interval appeared additive (Appendix: Supplementary Figures, Fig. 4.3).
These data show that spheres consistently and significantly decreased larval infestation in fruit considered marketable. Daily harvest also decreased infestation, although the differences were smaller. However, it seems evident that the rigorous grading of the fruit into marketable and unmarketable categories, based on fruit firmness, may have obscured some of the treatment differences by sorting out much of the fruit that was infested. For this reason, we examined fruit graded as unmarketable.

*Effect of plastic, harvest interval, and sphere treatments on infestation in unmarketable fruit.*

Plastic, sphere, and harvest interval all significantly affected SWD infestation of unmarketable fruit (Table 3.3). In addition to these simple effects, plastic*sphere, sphere*interval, and the three-way interaction were all significant.

**Table 3.3. Unmarketable Fruit ANOVA results.**

<table>
<thead>
<tr>
<th>Main effects lsmeans</th>
<th>Plastic</th>
<th>Larvae/fruit</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>UVO</td>
<td>0.50</td>
</tr>
<tr>
<td></td>
<td>UVT</td>
<td>0.27</td>
</tr>
<tr>
<td>Sphere</td>
<td>Yes</td>
<td>0.21</td>
</tr>
<tr>
<td></td>
<td>No</td>
<td>0.56</td>
</tr>
<tr>
<td>Interval</td>
<td>Daily</td>
<td>0.29</td>
</tr>
<tr>
<td></td>
<td>3xs</td>
<td>0.49</td>
</tr>
</tbody>
</table>

**Interaction Means**

<table>
<thead>
<tr>
<th>Plastic Interval Sphere</th>
<th>Larvae/fruit</th>
</tr>
</thead>
<tbody>
<tr>
<td>UVO 3xs No</td>
<td>1.06</td>
</tr>
<tr>
<td>UVO Daily No</td>
<td>0.43</td>
</tr>
<tr>
<td>UVO 3xs Yes</td>
<td>0.29</td>
</tr>
<tr>
<td>UVO Daily Yes</td>
<td>0.24</td>
</tr>
<tr>
<td>UVT 3xs No</td>
<td>0.42</td>
</tr>
<tr>
<td>UVT Daily No</td>
<td>0.33</td>
</tr>
<tr>
<td>UVT 3xs Yes</td>
<td>0.18</td>
</tr>
<tr>
<td>UVT Daily Yes</td>
<td>0.15</td>
</tr>
</tbody>
</table>

(Table continued on next page)
Table 3.3 continued.

<table>
<thead>
<tr>
<th>P-values from ANOVA</th>
<th>Larvae/fruit</th>
</tr>
</thead>
<tbody>
<tr>
<td>Plastic</td>
<td>0.002</td>
</tr>
<tr>
<td>Sphere</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Interval</td>
<td>0.006</td>
</tr>
<tr>
<td>Plastic*sphere</td>
<td>0.505</td>
</tr>
<tr>
<td>Plastic*interval</td>
<td>0.0432</td>
</tr>
<tr>
<td>Sphere*interval</td>
<td>0.0226</td>
</tr>
<tr>
<td>Plastic<em>sphere</em>interval</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Date</td>
<td>&lt;0.0001</td>
</tr>
</tbody>
</table>

The presence of spheres decreased the number of SWD larvae per fruit from 0.56 to 0.21. Results for marketable fruit in Experiment 2 were similar; however, unmarketable fruit were sampled on only the last three days of Experiment 2 when infestation rates were low. If data were collected for the whole period, means would likely have been higher. Despite this, it is clear that soft fruit was not necessarily infested.

In unmarketable fruit daily harvest reduced mean infestation from 0.49 to 0.29 larvae per fruit. Although infestation was higher for daily harvest plots on the second date, the difference was still significant.

Unmarketable fruit harvested from plants under UVT plastic had significantly fewer larvae per fruit (0.27) than fruit from plants grown under UVO plastic (0.50). These data suggest that UV-light could impact some aspect of SWD behavior. Unlike previous work with UV-blocking films for insect pests in greenhouses, in this case using a UV-transmitting plastic appeared to be more effective in decreasing larval infestation in fruit. The high infestation under UVO plastic also conflicts with the lower numbers of adult SWD trapped under UVO, though this may simply further demonstrate the lack of correlation between trap counts and infestation. These data are only from three days and have low replication. Other data from Experiments 1 and 2 do not support the conclusion that plastic influences larval infestation.

The data suggest that the efficacy of a given treatment may vary when combined with other treatments (Fig. 3.5). As with marketable fruit in Experiment 2, unmarketable fruit from
daily harvests had fewer SWD larvae per fruit in the absence of spheres, but in the presence of spheres the difference was very small. This suggests that when spheres are used, more of the soft fruit that would otherwise be culled could possibly be considered marketable regardless of harvest interval. UVO plastic had the highest levels of infestation when harvested at three-day intervals and in the absence of spheres. The shorter harvest interval and the presence of spheres both reduced larval infestation of unmarketable fruit, but treatment differences were very small for fruit harvested from plants under UVT plastic.

**Figure 3.5. Mean number of SWD larvae in unmarketable ‘Josephine’ raspberry fruit grown in high tunnel as influenced by spheres, interval, and plastic in experiment 2.**

Results from the two experiments were similar and suggest that when practices are combined fruit could be graded less rigorously. A mean infestation of 0.21 larvae per fruit, as occurred in the presence of spheres, indicates that two fruits in ten contained one larva. In markets where low levels of infestation are acceptable, it is possible that more soft fruit could be marketed as long as the fruit is not extremely soft or leaking.
Potential for combination of treatments.

Mean infestation from the eight combinations of treatments in our study show that it may be possible to combine non-spray control approaches to significantly reduce infestation without spraying insecticides. In Experiment 1, the combination of UVT plastic, daily harvest, and spheres had a mean infestation rate of 0.08, in contrast with the UVO, 3 times a week harvest, and no sphere treatment, which had 0.82 larvae per fruit. In Experiment 2 the difference was 0.28 versus 0.78. In unmarketable fruit, the difference was 0.15 versus 1.06. While there are many interactions, and treatments are not equally effective depending on the other treatments with which they are combined, it is clear that combining treatments has real potential for decreasing infestation without insecticide spray.

Conclusions

Our data indicate that multiple practices can be combined in high tunnels to increase yields of marketable raspberry fruit while decreasing rates of SWD infestation. These results may be economically beneficial for growers, for whom the importation of SWD has dramatically decreased marketable yields (Bolda et al., 2010). In this experiment, when plants were harvested daily rather than three days a week, yields increased between 50% and 75%, suggesting that losses from SWD could be ameliorated by shortened harvest interval. Previous research in which daily harvest decreased yield compared with less frequent harvest intervals (Leach et al., 2017) conflicts with our results, but discrepancies were likely due to differences in grading procedure. Growers who allow softer fruit to be included in marketable yields may find that daily harvest decreases yield because fruit has fewer days to grow. However, daily harvest will probably
increase yields of ripe firm fruit. Some growers already harvest daily, and it seems to be an increasingly common practice for maximizing marketable yields with or without pesticides. Sphere and plastic treatments did not influence total yield, although these treatments interacted with other treatments.

Spheres significantly reduced larval infestation of marketable and cull fruit. These findings are supported by previous research in field-grown raspberries (Rice et al., 2017a). In our second experiment the use of spheres reduced infestation from roughly 50% to 25%. Whether or not this is acceptable depends on the market, but it is notable that conventional spray programs have not always eliminated infestation (Van Timmerman and Isaacs, 2013). The fact that the spheres were effective inside tunnels suggests that they can be a valuable technology in high tunnel raspberry production, which had been a high percentage of production in the Northeast prior to the introduction of SWD (Demchak, 2009). Our high tunnel-grown plants were also quite large with dense canopies and deploying one sphere for every two plants was effective. More research is needed on deployment rates to determine if efficacy can be increased while maintaining economic viability.

Non-target organisms were impacted to some degree by the spheres. Although not quantified, adverse effects to non-target organisms by the spheres was likely far less than would be expected from pesticide applications. Even when used in attract-and-kill technology, insecticides are applied at far lower rates than conventional treatments (El-Sayed et al., 2009). Data that have not yet been published show minimal impacts on pollinators (T. Leskey, personal communication). A drawback to the spheres is that they are currently under development and obtaining registration for this technology may be slow. Additionally, Spinetoram (marketed as Delegate), the active ingredient for the spheres, is widely used for SWD control. Although considered a low-risk insecticide with reduced impact on non-target organisms, Delegate is not approved for organic use (Piñero and Byers, 2013). Rice et al. (2017a) tested different toxicants
for attracticidal spheres and all the organic pesticides tested had very low efficacy and residual activity against SWD. Therefore, development of effective spheres for organic growers is unlikely. Research would be beneficial to develop a variety of chemistries for the spheres and possibly to develop a system of temporal or spatial refuges to reduce the risk of resistance.

Plastics with different UV-transmittance properties did not significantly impact yields or infestation in marketable fruit, and these results support our previous research on the effects of plastics with different light transmission properties. Previous research does suggest that compared to field-grown raspberries, raspberries in high tunnels generally had lower SWD populations (Rogers et al., 2016), but it is likely that the difference was due to higher than optimum temperatures for SWD development, rather than plastics affecting visual cues for navigation in any way.

Finally, the patterns of mean trap captures, yields, and infestation over the course of the two experiments suggest that if growers use these treatments for entire seasons they may realize more consistent control of SWD than was found in this study. Our infestation data indicate that if spheres and shortened harvest intervals were used consistently or starting earlier in the season before SWD populations build, infestation might be even lower than it was here.

Our results show that high tunnel raspberry producers who harvest daily and follow rigorous sanitation practices are likely to increase their marketable yields dramatically. Further, when spheres become available they can be an effective technology in high tunnels and can be combined with practices such as daily harvest and sanitation to even eliminate the need for insecticidal sprays.


Figure 4.1. The proportion of marketable yield by harvest interval over time in experiments 1 and 2.
Figure 4.2. Effects of plastic and sphere on total yield over time in experiments 1 and 2.

Experiment 1:

Experiment 2:
Figure 4.3. Effects of sphere treatment and harvest interval on number of larvae per fruit in experiment 1.