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Chemical profile for possible host selection and to induce resistance against spotted wing drosophila in grape

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This project addresses a relatively new invasive pest, spotted wing drosophila (SWD), an insect that is having a dramatic impact on berry and vineyard crops in much of the U.S.

Spotted wing drosophila, *Drosophila suzukii* (Matsumura), is a congeneric relative of other vinegar or pomace flies (popularly called fruit flies). This species is native to eastern Asia. It was introduced into California in 2008. During 2009, it spread up the Pacific Coast through British Columbia. Late in 2009, it was found in Florida. Because of the speed with which it moved up the west coast, we established a trapping program in South Carolina, North Carolina and Virginia in 2010. At that time, SWD was detected in both Carolinas but not Virginia; however, it was found in all five trapping locations in Virginia in 2011 (Pfeiffer et al. 2011, Pfeiffer et al. 2012). It should now be considered generally distributed in the state (Fig. 1).

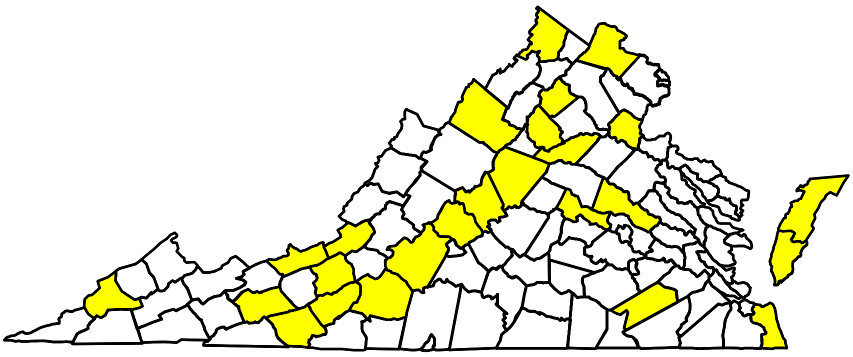

Fig. 1. Collection counties for spotted wing drosophila in Virginia, as of October 2013.

Unlike other *Drosophila* species, SWD attacks ripening fruit on the plant, not limited to overripe fruit material. SWD has a large, toothed ovipositor with which it cuts through healthy, intact fruit skin. Each female can lay 7-16 eggs per day, with an adult life span of up to to 9 weeks, averaging 350 eggs per female. There are about 13 generations per season. Larvae develop and feed in the fruit tissue, causing a premature softening with tissue breakdown. Infested clusters may become infected with sour rot organisms.

Rather than acting as passive victims in these interactions, plants respond to herbivory with the production of toxins and defensive proteins that target physiological processes in the insect. Herbivore-challenged plants also emit volatiles (Tholl et al. 2006) that attract insects and predators and bolster resistance to future threats. This highly dynamic form of immunity is initiated by the recognition of insect oral secretions and signals from injured plant cells. A detailed understanding of plant immunity to arthropod herbivores will provide new insights into basic mechanisms of chemical communication and plant-animal coevolution (Junker et al.

2013) and may also facilitate new approaches to crop protection and improvement (War et al. 2013). Plant attack by insect herbivores has been shown to increase plant defenses, generally referred to as induced plant resistance. Chemical defense strategies involve secondary metabolites (Hussain et al., 2014). Indirect induced defenses attract natural enemies, whereas direct induced defenses directly affect the performance and preference of the attacking herbivore (War et al. 2012). Secondary metabolites do not have any role in plant development but plays the major components of chemical defense strategies (Van et al. 2008) that regulate host plant utilization by insects. In nature, plants are constantly surrounded by herbivorous insects that negatively influence plant fitness. Chemical compounds that play a role in direct defense are produced and stored in tissues of the plant that are consumed by herbivores. This reaction results in the release of an array of toxic compounds such as isothiocyanates that reduce herbivore survival, growth, and development rate. In contrast to direct defense mechanisms, indirect defense mechanisms promote the effectiveness of the natural enemies (Thaler 2002) of herbivores e.g. through volatile secondary metabolites (War et al. 2013). Direct and indirect defense mechanisms can function additively against the herbivore. Induced resistance plays an important role in defense mechanism (Sharma et al. 2012) for pest management. Therefore, chemicals responsible for plant defense as well as for attraction against SWD have not been explored. Therefore, this study shall be conducted (i) to explore volatile compounds for SWD attraction to its host as well as for its natural enemies, and (ii) to study the chemical profile of infested grape fruits by SWD.

Fruit are mainly attacked during the ripening process. It is therefore critical to provide control of sensitive crops in the period shortly preceding harvest. It is important not merely to provide efficacy, but material must also be labeled with a short Preharvest Interval (PHI). With the introduction of SWD being very recent (mid-late summer 2011), we have not had the opportunity to perform control studies of this pest yet. Several likely materials are listed in the 2012 Virginia Tech Pest Management Guide (Pfeiffer et al. 2016). However, research needed to determine actually control provided in the field. Some likely pesticides for SWD were listed by Walsh et al. (2011). With the high number of generations and high reproductive capacity of SWD, there is high risk of insecticide resistance.

Control strategies for SWD now center almost exclusively on chemical control. This threatens to upset biological control systems for other pests. There is also a great risk of pesticide resistance because of the high reproductive rates of SWD. Sole reliance on insecticides is not sustainable; we must develop biological and cultural methods to supplement chemical control. There are currently exploration efforts in Asia to find natural enemies of SWD. But there is almost no information on natural enemies that are already here. It will be years before a suitable natural enemy can be found in Asia and approved for release.

Results under the specific objectives are as follows:

Identification of herbivore (SWD)-induced volatiles (HIV) in grape fruit
Determination of biochemical sources of resistance in grapes

Analysis is underway in cooperation with the Dept of Biochemistry. Some of the varietal differences noted are apparently due to other factors being investigated in a separate project.

Exogenous application of jasmonic acid and salicylic acid in vineyards.

Salicylic acid (SA) and jasmonic acid (JA) were applied in a winegrape vineyard with a history of SWD infestation. Applications were made using a CO_2-powered backpack sprayer.

1) Salicylic acid (SA) and jasmonic acid (JA) induce defensive response against insect pests. The pre application of SA & JA induced defensive response against SWD in two different wine grapes. In field study, plants presprayed with SA & JA exhibited highest defensive response relative to unsprayed plants. We quantified increased total phenol, tannin and flavonoid content in SA & JA treated fruits (Table 1) compared to untreated fruits. Among the two different genotypes, total tannin and flavonoid content was higher in Pinot Noir relative to Chardonnay. There were no differences in SWD infestation. Therefore, while compound often involved with host plant defensive response were produced in response to the applications, the response was insufficient to protect clusters from SWD infestation.

While infestation by SWD was not affected by application of SA or JA, there were nevertheless effects on the crop. Cluster weight (grams) was also highest in SA & JA sprayed plants relative to untreated vines.

2) We quantified total tannin, total phenol and total flavonoid content in six grape varieties to explore the possible resistance of wine grapes against SWD. We estimated highest amount of secondary metabolites in Petit Verdot and Petit Manseng relative to other varieties (Viognier, Cabernet Franc, Vidal, Pinotage). This is a complex situation that would bear further work. Our other SWD work has shown that Petit Manseng is less susceptible to SWD than other varieties; it has higher levelws of secondary metabolites (it also has thicker skin with higher penetration pressure, and different cluster structure). However, Petit Verdot is often noted to be susceptible. Viognier is more susceptible to infestation according to our other work, and here is reported to have lower levels of secondary metabolites.

Varieties	Treatments	µġ Quercetin equivalents/gram	µġ Tannic acid equivalents/gram	µġ Gallic acid equivalents/gram
Pinot Noir	SA	217.16	17.8	185.36
	JA	192.66	16.33	186.16
	Control	111	15.6	184.33
Chardonnay	SA	97.66	14.04	182.36
	JA	104.33	16.43	182.16
	Control	82.66	9.36	180.56

I. Quercetin used as standard for Total flavonoid estimation;
II. Tannic acid used as standard for Total Tannin estimation;
III. Gallic acid used as standard for Total phenol estimation;
IV. Data are on dry weight basis.

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