

1 Title: Insecticidal and oviposition deterrent effects of essential oils and their
2 constituents against the invasive pest *Drosophila suzukii* (Matsumura) (Diptera:
3 Drosophilidae)

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1 Abstract

2 Spotted wing drosophila (SWD), *Drosophila suzukii* (Matsumura), is an important
3 new invasive pest of stone and berry fruits in North America and current control
4 methods require frequent application of synthetic pesticides. This has created a
5 need for new and environmentally friendly biopesticides for the control of SWD.
6 This paper investigated the potential of nine essential oils from avocado (*Persea*
7 *americana* Mill.), neem (*Azadirachta indica* A. Juss), kukui nut (*Aleurites moluccana*
8 L.), macadamia nut (*Macadamia integrifolia* Maiden & Betcher), spike lavender
9 (*Lavandula latifolia* Vill.), Grosso lavandin leaf and flower (*Lavandula x intermedia*
10 cv 'Grosso'), and Provence lavandin leaf and flower (*Lavandula x intermedia* cv
11 'Provence') as well as three major monoterpene constituents of lavender essential
12 oils: 1,8-cineole, 3-carene and linalool for their ability to control SWD through
13 fumigation and contact toxicity assays as well as oviposition deterrent activity.
14 Linalool was found to be the most effective monoterpene in fumigation assays (EC50
15 1.85 μ L/L air) and spike lavender floral essential oil was found to be the most
16 effective whole oil (EC50 3.79 μ L oil/L air). In contact toxicity assays 1,8-cineole
17 (EC50 0.67%) was the most effective monoterpene while avocado (EC50 0.54 %)
18 and spike lavender (EC50 0.69%) oils were the most effective whole oils. No
19 significant oviposition deterrent activity was observed. This report indicates that *L.*
20 *latifolia* essential oil and commercially available avocado oil are strong lead
21 candidates for management of SWD. Furthermore the activity of *L. latifolia* essential
22 oil is likely due to the high content of 1,8-cineole and linalool. This is the first report
23 of significant insecticidal activity by these oils and compounds against SWD.

1 Keywords: spotted wing drosophila; lavender; avocado; biopesticide; terpene;
2 linalool

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4 Abbreviations

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6 GC-MS gas chromatography – mass spectrometry

7 EC50 half-maximal effective concentration

8 SWD spotted wing drosophila

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10 1. Introduction

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12 *Drosophila suzukii* (Matsumura) or spotted wing drosophila (SWD) is a vinegar fly
13 species native to Japan, and has recently become an invasive pest in North America.
14 Reported as early as the 1930s in Japan, first reports of SWD in North America came
15 from berry growers in California in 2008 (Hauser, 2011). SWD moved rapidly up the
16 west coast of North America and the first reports of SWD in British Columbia came
17 from a cherry (*Prunus avium* L.) orchard near Kelowna, British Columbia, Canada in
18 2009 (Acheampong and Thistlewood, 2011; Lee et al., 2011b; Thistlewood et al.,
19 2012). Since this first report, SWD has become a major invasive pest in British
20 Columbia and the Pacific Northwest (Washington and Oregon), and has spread as far
21 as Europe, colonizing diverse stone fruits and berry crops including blueberry
22 (*Vaccinium* spp.), raspberry (*Rubus* spp.), strawberry (*Fragaria* spp.), cherry (*Prunus*
23 spp.) and blackberry (*Rubus* spp.) (Lee et al., 2011b; 2011a). Estimates of economic

1 losses due to this pest range as high as \$511 million annually in the United States
2 alone with additional cost being generated due to the monitoring and control efforts
3 required to prevent primary infestations (Dreves, 2011; Goodhue et al., 2011; Lee et
4 al., 2011b;). Unlike related *Drosophila* species that oviposit only in decaying or
5 damaged fruit, SWD possess a unique serrated ovipositor that is capable of piercing
6 the skin of unripe and ripening fruit (Atallah et al., 2014).

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8 Current control methods for SWD management rely on frequent application of
9 traditional pesticides, as SWD has a relatively short life cycle with a full life cycle
10 from egg to mature adult taking only 21 days, and as such is able to go through
11 numerous life cycles in a single growing season with up to 13 life cycles being
12 achieved annually in Japan and 10 in North America (Beers and Van Steenwyk,
13 2011). Unfortunately, frequent application of insecticides such as neonicotinoids
14 and organophosphates are raising alarm due to their adverse effects on beneficial
15 insects, such as pollinators and predacious insects, and contamination of local
16 ecosystems and water sources (Regnault-Roger et al., 2012; Tan et al., 2014). As a
17 result of this, there is a growing interest in less ecologically damaging control
18 methods, and a strong push to develop new, organic and ecologically sustainable
19 control methods, which are still effective in controlling this economically
20 devastating pest.

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22 Essential oils are produced by many plant species and serve many important
23 commercial purposes including use in cooking, medicine and agriculture. Plants

1 produce these compounds for many purposes including: allelopathy, pollinator
2 attraction and plant defense (Mahmoud and Croteau, 2002). From an agricultural
3 perspective, their functions in insect attraction and plant defense are of greatest
4 interest. Some essential oil constituents, such as linalool, are also insect pheromones,
5 while many others both alone and as whole oils have shown strong activity as
6 repellents, feeding and oviposition deterrents, and insecticides for control of a range
7 of insect species (Peixoto et al., 2015; Mant et al., 2005; Moretti et al., 2014; Pinto et
8 al., 2015; Regnault-Roger et al., 2012; Rossi and Palacios, 2015; Tabata et al., 2015;
9 Yeom et al., 2015). One such commercial example is the impregnation of grafting
10 strips with lavender essential oil for the repellency of the red bud borer (*Resseliella*
11 *oculiperda* Rübs.), with 95% efficacy, while citronella oil has been effectively
12 employed at concentrations as low as 1 % to protect stored potato tubers from
13 infestation by the potato tuber moth (*Phthorimaea operculella* Zell.) (Sharaby et al. ,
14 2014; van Tol et al., 2007). Neem oil is one of the most commercially recognizable
15 oils that is now widely marketed due to the presence of the insecticidal, antifeedant
16 repellant properties of its active constituent, azadirachtan (Ahmad et al., 2014;
17 Benelli et al., 2014; Sokame et al., 2015; Togbé et al., 2014).

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19 To date, the effect of essential oils or their individual constituents for the control of
20 SWD has not been reported. The goals of this study are, therefore, to identify
21 commercial available essential oils with insecticidal or oviposition deterrent activity
22 that may then be exploited as part of a more ecologically sustainable integrated pest
23 management program for the control of SWD.

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2 2. Experimental Methods

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4 2.1 Insect rearing

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6 *Drosophila suzukii* adults were collected from sour cherry (*Prunus cerasus* L.), white
7 mulberry (*Morus alba* L.) and black mulberry (*Morus nigra* L.). Emerged adults of
8 mixed age were collected and maintained for all future generations in 25 x 95 mm
9 polypropylene vials (Applied Scientific, Fisher, Canada) filled with approximately 5
10 mL of yeast based *Drosophila* medium composed of 10% sucrose, 5% yeast, 1.8%
11 agar, 0.9% propionic acid, 0.8% potassium sodium tartrate, 0.19% methylparaben,
12 0.1% potassium phosphate monobasic, 0.09% o-phosphoric acid, 0.05% sodium
13 chloride, 0.05% magnesium chloride, 0.05% calcium chloride and 0.05% iron (III)
14 sulfate. The colony was maintained under a 16:8 hour light:dark photoperiod at
15 ambient temperature and ambient humidity. Adults were transferred to new food
16 twice weekly.

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18 2.1 Extraction and analysis of essential oil samples

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20 Essential oils from *Lavandula x intermedia* cv Grosso leaf and *Lavandula x*
21 *intermedia* cv Provence leaf and flower were obtained by distillation of respective
22 tissues. Briefly, samples were boiled in water for one hour and extracted into
23 pentane in a simultaneous steam distillation-solvent extraction system. Pentane was

then removed by drying under nitrogen gas. The composition of both leaf essential oils has not been previously reported and, thus, to identify the composition of these oils, dilutions of oil samples from triplicate distillations were made in pentane and samples were run on a Saturn Varian gas chromatography mass spectrometer (GC-MS) using a 30 m Varian VF-5 MS column, with the following method details: 1 µL of sample injected on column with a column flow rate of 1 mL/min and an injector temperature of 40 °C held for 0.1 min and then increased to 100 °C at 20 °C/min and held for 16.5 min; the column oven temperature was held at 40 °C for 5 min and was ramped to 170 °C at 6 °C/min, held for 4 min and then ramped to 230 °C at 30 °C/min and held for 5 min. Solvent delay for MS detection was 3.0 min and m/z values from 39-300 were monitored. Constituents were identified by comparison of mass spectra to the NIST Mass Spectral database v 5.0 and to the spectra of authentic standards, from which relative percent oil composition was determined.

2.2 Fumigation toxicity assays

To test the susceptibility of *D. suzukii* adults to the volatile compounds of the essential oils, thirty adult flies (four to seven day old) were chilled at 4°C and counted on ice then added to 500 mL canning jars along with a 35 mm petri plate containing approximately 2 mL of *Drosophila* medium and covered by organza mesh and allowed to recover for approximately 20 minutes. Oil was then directly applied to a 25 mm square of Whatman filter paper in a 35 mm petri plate affixed to the lid of the canning jar. The oil-impregnated filter paper was separated from the

1 remainder of the jar and protected from contact with flies with a square of mesh.
2 Eight concentration levels were tested: 0, 1, 2, 3, 4, 5, 7.5 and 15 μL oil/L air in
3 triplicate. Eleven oil samples and three monoterpenes were tested (Table 1).
4 Mortality was then recorded at 24 hours. Half-maximal effective concentration
5 (EC50) values at 24 hours were calculated by probit regression analysis of log-
6 transformed values using SPSS v22 (IBM, USA).

7 Table 1. Oils Tested

Oil/Constituent	Tissue Source	Manufacturer/Brand
Avocado	Fruit	Chosen Foods Inc. (San Diego, CA)
Cineole	Standard	Sigma-Aldrich (Canada)
delta-Carene	Standard	Sigma-Aldrich (Canada)
Linalool	Standard	Sigma-Aldrich (Canada)
Kukui nut	Nut	Essential Oils Trading Post (USA)
<i>Lavandula angustifolia</i>	Flower	Okanagan Lavender Herb Farm (Kelowna, BC)
<i>Lavandula latifolia</i> cv Medikus	Flower	F. P. I. Sales (Delta, BC)
<i>Lavandula x intermedia</i> cv Grosso	Flower	F. P. I. Sales (Delta, BC)
<i>Lavandula x intermedia</i> cv Grosso	Leaf	Distilled
<i>Lavandula x intermedia</i> cv Provence	Flower	Distilled
<i>Lavandula x intermedia</i> cv Provence	Leaf	Distilled
Macadamia nut	Nut	Life-flo (USA)
Neem	Kernel	Potter's Garden (USA)

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9 2.3 Contact toxicity assays

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11 To test the susceptibility of *D. suzukii* to direct exposure to essential oils and
12 individual monoterpenes, thirty adult flies (four to seven day old) were chilled at
13 4°C and counted on ice and transferred to oil exposure chambers created from 250
14 mL baby food jars (bottom diameter = 5.9 cm, opening diameter = 4.8 cm, height =
15 6.7 cm) lined with blotting paper to create an ~120 mL exposure chamber (6.7 cm x

4.8 cm). Oil was diluted to the 8 test concentrations: 0, 0.5, 1, 2.5, 5, 10, 20 and 40% oil in acetone and 0.5 mL of each solution was applied to flies in exposure chambers using a piston style manual pump sprayer. Flies were then removed from exposure chamber and transferred to clean vials with approximately 15 mL of fly media to prevent any further exposure to a treated surface and to prevent fumigation mortality. Mortality was recorded after 24 hours. Each treatment was repeated in triplicate. Oils and individual monoterpenes tested were the same as for fumigation toxicity assays (Table 1). Half-maximal effective concentration (EC₅₀ at 24 hours) values were calculated by probit regression analysis of log transformed values using SPSS v22 (IBM, USA)

2.4 Oviposition deterrent activity

The ability of essential oils and individual monoterpenes to prevent oviposition of *D. suzukii* in blueberries was determined by the following procedure (adapted from Beers et al. 2011). Cotton balls were dipped in sterile 5% sucrose solution and then placed in the bottom of *Drosophila* vials. Blueberries (store bought, organic) were then washed, allowed to dry and then dipped for 3s into 3 mL of oil solutions of 0, 1, 5, 10 or 20%, using acetone as a solvent. Blueberries were then allowed to dry on blotting paper and once dry were placed in tubes (1 berry/vial). All oil concentrations were performed with five replicates. Five female and three male *D. suzukii* adults (four to seven day old) were then added to each vial which was sealed with mesh using an elastic band. Vials were then kept in a controlled environment

chamber at 25°C, 16:8 hour light:dark photoperiod for 48 hours. Flies were then chilled and berries were removed from vials. The number of oviposition marks on each berry was counted under a dissecting microscope and recorded. Oils and individual monoterpenes tested were the same as for toxicity assays (Table 1).

3. Results & Discussion

To garner a broader scope of inference into the potential efficacy of essential oils for the control of SWD, a variety of essential oils were chosen. Lavender essential oils have been reported to possess repellent, insecticidal and behaviour disrupting effects against various species of insects (El-Sheikh et al., 2005; Rozman et al., 2007; Tabata et al., 2015; Yang et al., 2004). Similarly kukui nut (*Aleurites moluccana* L.) oil has been reported both in traditional Polynesian knowledge, and studies have reported insect repellent properties (Nakayama and Osbrink, 2010), while compounds have been isolated from avocado, which show activity against *Spodoptera exigua* (Hubner) (Rodriguez-Saona et al., 1997; 1998). Macadamia nut originated in a similar climate as kukui and avocado nut, and as it is a growing industry (Axel and Fairman 1992; Elevitch and Manner 2006; Rietow 2012); oils were tested for potential efficacy, though no previous reports exist examining its insecticidal activity. The activity of neem oil has been widely demonstrated and it was therefore included in these assays (Gahukar, 2014; Shafie and Basedow, 2003; Souza et al., 2015; Thacker et al., 2003).

1 As the composition of the leaf essential oil from the two intermedia cultivars used in
2 this study have not been previously reported, the essential oil profile of leaf oils as
3 well as in-laboratory distilled Provence floral oil was determined by GC-MS analysis
4 (Table 2). These results found that the oil from Grosso leaf was high in the
5 monoterpenes borneol (8-18 %), camphor (20-22 %) and 1,8-cineole (13-27 %) as
6 well as the sesquiterpene cadinol (21-29 %). The profile of Provence leaf essential
7 oil was similar but had much lower sesquiterpene content. The major constituents
8 in Provence essential oil were borneol, camphor and 1,8-cineole. The oil
9 composition for macadamia nut (*Macadamia integrifolia*), kukui nut (*Aleurites*
10 *moluccana*) and avocado (*Persea americana*) oils is readily available either in the
11 literature (Table 3) or on the packaging of the oil while the active constituent of
12 neem oil (*Azadirachta indica*) has been well defined as the limnoid terpene
13 derivative azadirachtin (Dreher and Davenport, 2013; Gahukar, 2014; Jaastad et al.,
14 2009; Nicoletti et al., 2012; Shafie and Basedow, 2003; Thacker et al., 2003;
15 Woronuk et al., 2010).

1 Table 2. Relative percent composition of lavender essential oils tested.

Essential oil constituent	<i>Lavandula</i> <i>angustifolia</i> ^a	<i>Lavandula</i> <i>latifolia</i> ^a	<i>Lavandula</i> x <i>intermedia</i> cv Grosso ^a	<i>Lavandula</i> x <i>intermedia</i> cv Grosso ^b	<i>Lavandula</i> x <i>intermedia</i> cv Provence ^b	<i>Lavandula</i> x <i>intermedia</i> cv Provence ^b
	Flower	Flower	Flower	Leaf	Leaf	Flower
Borneol	1.0-4.0	-	1.5-3	8-18	24.1	-
Cadinene	-	-	-	1-5	-	-
Cadinol	-	-	-	21 - 29	0.9	-
Camphor	trace	12-16	6-8	20 - 22	9.22	6-8
Carene	-	-	-	1-3	1.5	-
Caryophyllene	-	-	-	0-2	-	-
1,8-Cineole	trace	22-27	4-7	13-27	53.3	4-7
Linalool	25-38	27-41	24-35	-	trace	25-35
Linalyl acetate	25-45	trace	28-38	-	-	26-38
Ocimene	3-4	trace	0.5-1.5	-	trace	trace
Sabinene	-	-	-	0-2	trace	-
Terpinene	-	-	-	0-2	-	-
Terpineol	4-5	trace	1.5-5	-	-	trace

2 ^a(Woronuk et al., 2010)

3 ^bComposition determined by GC-MS analysis

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1 Table 3. Percent composition of kukui nut, avocado and macadamia nut oils.

Oil constituent	Percent composition		
	Kukui nut	Avocado	Macadamia Nut
<i>Fatty acids</i>			
Palmitic acid	0.051 - 8	12.4-18.80	1.4-11.6
Stearic acid	2.1-54.22	0.51-1.50	1.2-3.6
Oleic acid	15.5-47.98	54.3-60.61	37.6-74.7
Linoleic acid	19.96-43.7	0.11-14.7	1.0-13.7
Linolenic acid	2.5-28.1	0.62-0.73	0-3.0
Caprylic acid	0.01-0.019		
Capric acid	0.011-0.017		
Lauric acid	0.022-0.041		
Myristic acid	0.051	0.06-0.14	0-0.9
Palmitoleic acid		0.1-4.59	14.7-23.0
Elaidic acid		0.27-0.31	
Vaccenic acid		5.84-5.90	3.6-3.8
Eicosenoic acid		0.12	2.3-4.0
Eicosatrienoic acid		0.01	
Eicosapentanoic acid			0.28
Arachidonic acid		0.01	
Arachidic acid		0.36	1.96-2.8
Gadoleic acid		0.09	
Behenic acid		0.11	0.75
Lignoceric acid		0.05	0.28
Gondoic acid			2.62
Nervonic acid			0.19
Heptadecanoic acid			0.4-2.1
Heptadecenoic acid			0.9-3.3
Nonadecenoic			8.3
Margaric acid		0.34	
<i>Phytosterols</i>			
Campesterol		0.01692-0.01980	0.002-0.0118
Campestanol		0.0004-0.00046	
Stigmasterol		0.0007-0.00123	0.0021-0.0023
Lanosterol		0.00052-0.00064	
Sitosterol		0.23547-0.3081	0.1043-0.1393
Sitostanol		0.00197-0.00241	
Avenasterol		0.00215-0.01145	0.0151-0.0169
alpha-amyrin		0.00011-0.00015	
cycloartenol		0.01053-0.02163	
cycloeucalenol		0.00028-0.00033	
24-methylenecycloartnol		0.00112-0.00114	
citrostadienol		0.0072-0.01086	
<i>Other</i>			

Oil constituent	Percent composition		
	Kukui nut	Avocado	Macadamia Nut
Squalene		24.28-33.79	0.06244-0.020282
alpha-tocopherol		0.55-1.25	0-0.0008
gamma-tocopherol		0.0036-0.0040	0-0.0015
delta-tocopherol		0.0011-0.0017	0.0010-0.0012
cycloartenol acetate		3.15-5.76	
<i>Tocotrienols</i>			
delta-tocotrienol			0.000104-0.002006
gamma-tocotrienol			0.000394-0.005555
alpha-tocotrienol			0.00116-0.00549
<i>Other compounds identified but not quantified</i>			
<i>Furans</i>			
<i>avocadofurans</i>			
2-(pentadecyl)furan		present	
2-(heptadecyl)furan		present	
2-(1E-penta-decenyl)furan		present	
2-(8Z,11z-deptadecadienyl)furan		present	
<i>Fatty acid derivatives</i>			
Triolein		present	
Persin		present	
References	(Ako et al., 2005; Martin et al., 2010; Norulaini et al., 2004; Siddique et al., 2011)	(Madawala et al., 2012; Rodriguez-Saona et al., 1997; 1998; Rueda et al., 2014; Santos et al., 2013)	(Birch et al., 2009; Fard et al., 2003; Kornsteiner-Krenn et al., 2013; Wall, 2010; Madawala et al., 2012)
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1 As all monoterpenes have low molecular weights, and are easily volatilized, this
2 produces higher in-air concentrations than non-volatile compounds; therefore it
3 was not surprising that the oils containing high quantities of aromatic compounds,
4 including the monoterpenes, showed the greatest fumigant effect against SWD. *L.*
5 *latifolia* (spike lavender) showing the strongest effect of all oils tested (EC50 3.79 μ L
6 oil/L air), while the floral essential oil constituent linalool showed the greatest
7 activity of all samples tested (EC50 1.85 μ L/L air)(Table 4). The three oils with high
8 fatty acid content: kukui nut, macadamia nut and avocado oils, showed limited to no
9 activity in contact assays, nor did neem show any fumigation effect on SWD. All
10 lavender oils had some fumigant effect as did constituents carene, 1,8-cineole and
11 linalool. Linalool had the strongest fumigant effect of any oil or constituent followed
12 by spike lavender oil, Provence leaf and flower oils, Grosso flower and leaf oils,
13 carene and 1,8-cineole, respectively, though only linalool had a confidence interval
14 that did not overlap with other oils or constituents tested. Spike lavender oil is high
15 in linalool and 1,8-cineole, and the presence of linalool in particular, rather than the
16 less effective monoterpenes, could explain the increased activity of this oil as
17 compared with other oils. This may also be the reason for which Provence leaf and
18 flower oils performed better than Grosso leaf and flower oils, as Provence has a
19 higher linalool and 1,8-cineole content than Grosso oils.

- 1 Table 4. SWD susceptibility to oils measured as fumigation toxicity and reported as
2 half-maximal effective concentration (EC50).

Oil	Source	EC50 (uL oil/L air) (95% Confidence Interval)	df	χ^2	p-value ^a
Avocado	commercial	no activity	-	-	-
1,8-Cineole	standard	10.71 (8.44, 13.01)	19	181.633	<0.0001
delta-Carene	standard	9.47 (8.36, 10.58)	19	35.675	0.012
Linalool	standard	1.85 (1.48, 2.12)	19	27.813	<0.0001
Kukui nut	commercial	no activity	-	-	-
<i>Lavandula angustifolia</i>	flower	8.14 (7.65, 8.59)	19	82.737	<0.0001
<i>Lavandula latifolia</i>	flower	3.79 (3.28, 4.21)	19	181.633	<0.0001
<i>Lavandula x intermedia</i> cv Grosso	flower	8.06 (7.22, 8.87)	19	37.69	0.006
<i>Lavandula x intermedia</i> cv Grosso	leaf	8.22 (7.40, 9.01)	19	32.119	0.03
<i>Lavandula x intermedia</i> cv Provence	flower	6.71 (6.14, 7.26)	19	200.780	<0.0001
<i>Lavandula x intermedia</i> cv Provence	leaf	5.68 (5.22, 6.14)	19	157.882	<0.0001
Macadamia nut	commercial	no activity	-	-	-
Neem	commercial	no activity	-	-	-

3 ^a P < 0.05 indicates that the model fits better than a model with just an intercept.

4

5 Leaf essential oils were not tested in the contact bioassay due primarily to
6 difficulties in obtaining sufficiently large amounts. Due to this, and their poorer
7 activity in the fumigation toxicity assays, they were deemed poor candidates for
8 commercial control agents. The trend observed in the fumigation toxicity assays in
9 which volatile oils showed strongest activity was not observed in contact toxicity
10 assays, further confirming that the difference in activity is most likely due to the
11 ability of those oils to volatilize (Table 5). The commercially available avocado oil,
12 cineole, spike lavender flower, and Provence flower showed lowest ranges of

activities. The concentration of 0.54% for the EC50 of avocado oil is still greater than the reported active concentrations for commercially available control agents such as spinosad which is active at concentrations of 0.01% (Beers and Van Steenwyk, 2011). This effect may be ascribed to previously reported active avocado furans and the fatty acid derived persin as reported by Rodriguez-Saona et al. (1997, 1998), though further studies with individual active components would be required to prove such a correlation. The activities seen from 1,8-cineole (0.67 %) and spike lavender (0.69 %) essential oils had confidence intervals which were lower than all other compounds and oils tested did not overlap with the confidence intervals of other compounds. In combination with the potent activity of spike lavender essential oil in the fumigation assays, this suggests that spike lavender has potential for control of SWD in a field setting. It is also of interest to note that the presence of a different primary active constituent in *L. latifolia* oils, 1,8-cineole for contact assays and linalool for fumigation toxicity assays, indicates that these monoterpenes have different mechanisms for toxicity. Additionally, the combined use of the non-volatile avocado oil with the highly effective oil of *L. latifolia* could show an increased synergistic activity, as compared to each oil alone due to the presence of multiple active compounds, with potentially different mechanisms of action. Lesser contact toxicity was observed for Provence, macadamia nut and neem oils, though it is of interest to note, that to the authors' knowledge, this is the first report of an insecticidal effect from macadamia nut oil. No contact toxicity was observed for kukui nut oil. As current control methods for stone fruits, such as cherries, recommended by Agriculture Canada require the use of broad spectrum insecticides

1 such as diazanon or malathion, the use of these oils as part of an integrated pest
 2 management program could reduce the environmental impact of using exclusively
 3 synthetic pesticides while still maintaining effective control. Extensive field trials
 4 are required, however, before these products would be ready for commercial use.

5

6 Table 5. SWD susceptibility to oils measured as contact toxicity and reported as half-
 7 maximal effective concentration (EC50).

Oil	Source	EC50 (%) (95% Confidence Interval)	df	χ^2	p- value ^a
Avocado	commercial	0.54 (0.03, 1.40)	19	88.611	<0.0001
Cineole	standard	0.67 (0.13, 1.41)	19	98.886	<0.0001
delta-Carene	standard	2.38 (1.57, 3.43)	19	71.42	<0.0001
Linalool	standard	9.85 (6.56, 17.83)	19	230.864	<0.0001
Kukui nut	commercial	no activity	-	-	-
<i>Lavandula angustifolia</i>	flower	2.10 (1.55, 2.69)	19	70.191	<0.0001
<i>Lavandula latifolia</i>	flower	0.69 (0.29, 1.12)	19	78.542	<0.0001
<i>Lavandula x intermedia</i> cv Grosso	flower	7.21 (4.22, 12.58)	19	247.360	<0.0001
<i>Lavandula x intermedia</i> cv Provence	flower	1.23 (0.86, 1.64)	19	46.589	<0.0001
Macadamia nut	commercial	11.17 (7.67, 19.39)	19	114.110	<0.0001
Neem	commercial	14.83 (10.08, 21.49)	19	55.334	<0.0001

8 ^a P< 0.05 indicates that the model fits better than a model with just an intercept.

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1 Lastly the oviposition deterrent activity of these oils was tested in an assay adapted
2 from Beers et al. (2011). Though none of the oils showed powerful repellent
3 activities, some repellency is suggested based on the results from all four floral
4 essential oils tested as well as the commercial oils (data not shown). Though these
5 activities are not significant, in conjunction with the reported toxicities, this may
6 augment the efficacy of oils such as spike lavender oil. Similar repellent trends were
7 also observed for avocado, macadamia nut and neem oils. However due to extremely
8 high variability of response with some standard errors as high as 300%, the
9 methodology used may not be the most appropriate for testing these products.

10
11 The primary hindrance to the use of essential oils as pest control agents has been
12 their cost. This would, however, likely not be a significant factor limiting their use
13 for the control of SWD, as the levels at which these compounds are active are equal
14 to or lower than active concentrations reported in other species including domestic
15 and agricultural pests including *Blatella germanica* (L.) (EC50 0.28-0.3 mg/L), and
16 *Sitophilus zeamais* (Mots.) (EC50 41.4 µL/L – 16.2 µL/mL) (de Lira et al., 2015;
17 Peixoto et al., 2015; Yeom et al., 2015) . The volatility of essential oils, such as spike
18 lavender oil, means that they will not persist in the environment or on food crops,
19 minimizing the concern of pesticide residues for human consumption and their
20 impact on the surrounding environment, making them safer alternatives to the
21 existing pesticides. Less volatile oils such as avocado and macadamia nut oils have
22 been used extensively for cosmetic and food purposes and have as such been
23 demonstrated to have a very low risk; due to their wide commercial uses as

1 carrier oils, cooking oils, cosmetic oils and increasing as an alternative bio-fuel. This
2 widespread use also means their commercial availability is not as limited as that of
3 lavender essential oil, making them a lower cost alternative to lavender essential oil
4 (Ako et al., 1995; Aysu and Durak, 2015; Dreher and Davenport, 2013; Harris and
5 Macfarlane 1980; Yasir et al., 2010). In the case of spike lavender and avocado oils,
6 this balance between a low cost and a high cost active may support the use of a
7 combination of the two oils with a greater concentration of avocado oil with spike
8 lavender oil as an additive. The bio-friendly aspect of these oils may, therefore,
9 outweigh some of the cost concerns as the importance of sustainable and eco-
10 friendly agriculture practices are of growing importance to many consumers and
11 growers alike.

12 13 4. Conclusions

14
15 The experiments provide evidence that plant oils and in particular the commercially
16 available avocado oil and the essential oil from *L. latifolia* show potential as new
17 commercial agents in the control of the invasive pest *D. suzukii* as part of an
18 integrated pest management program. In particular, this is the first report of the use
19 of essential oils for the control of SWD as well as the first report of insecticidal
20 activity for macadamia nut oil. The activity demonstrated by *L. latifolia* has been
21 demonstrated to be attributable to linalool and 1,8-cineole, the two major
22 monoterpene constituents of the oil.

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2

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