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Research Article

Two volatiles from anthracnose-infected blueberries trigger electrophysiological and aversive behavioral responses in *Drosophila suzukii* (Diptera: Drosophilidae)

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Native to Southeast Asia, the spotted-wing drosophila (*Drosophila suzukii* Matsumura) is an economically important invasive pest of thin-skinned fruits such as raspberries, blueberries, and strawberries worldwide. To reduce the reliance on insecticides for managing this pest, alternative strategies like behavioral manipulation are needed. Previous studies have shown that *D. suzukii* adults avoid blueberry fruits infected with the fungal pathogen *Colletotrichum fioriniae* Marcelino & Gouli, which causes anthracnose fruit rot, leading to the identification of nine potential repellent compounds. In this study, we further investigated the two most potent of these compounds—ethyl butanoate and ethyl (*E*)-but-2-enoate—to assess their repellent properties on the antennal and behavioral responses of *D. suzukii*. Electroantennogram (EAG) assays revealed that both esters elicited similar dose-dependent responses in male and female *D. suzukii*, which were often stronger than those triggered by 2-pentylfuran, a known repellent of this species. Additionally, we examined the behavioral responses of adult *D. suzukii* to these three repellent compounds under semi-field and field conditions using outdoor cages containing potted and planted blueberry bushes, respectively. Results from the cage studies showed that all three tested compounds can significantly reduce *D. suzukii* oviposition and adult emergence from blueberry fruits, with ethyl (*E*)-but-2-enoate sometimes outperforming the other compounds. Our findings indicate that the esters ethyl butanoate and ethyl (*E*)-but-2-enoate, which are induced by *C. fioriniae*-infected blueberries, elicit dose-dependent effects on *D. suzukii* antennae and act as effective oviposition deterrents. This supports their potential as promising tools for managing this pest through behavioral strategies.

Key words: spotted-wing drosophila, invasive pest, volatiles, EAG, oviposition deterrents, semiochemicals

Introduction

The spotted-wing drosophila, *Drosophila suzukii* Matsumura (Diptera: Drosophilidae), is an invasive and highly destructive pest of soft-skinned fruits, including berries, cherries, and grapes (Walsh et al. 2011, Lee et al. 2011, Asplen et al. 2015, Tait et al. 2021). Females of this species have a serrated ovipositor, which allows them to penetrate the skins of ripening and ripe fruits for oviposition (Atallah et al. 2014). Currently, *D. suzukii* is primarily managed through frequent insecticide applications (Diepenbrock et al. 2016, Joshi et al. 2023), which can be costly for growers and may lead to the development of resistance (Gress and Zalom 2019, Disi and Sial 2021, Ganjisaffar et al. 2022, Tabuloc et al. 2024). An alternative method of control that may reduce insecticide use is behavioral manipulation using semiochemicals (Tait et al. 2021). Previous efforts to manage *D. suzukii* with semiochemicals have shown some success (Hampton et al. 2014, Rice et al. 2017, Wallingford et al. 2017, Wallingford et al. 2018, Cloonan et al. 2018), demonstrating the potential of this approach. However, discovering new repellent chemicals could further improve this strategy by increasing aversion, enhancing specificity to *D. suzukii*, or identifying more affordable compounds to produce.

Like many insects, *D. suzukii* interacts with microbes in its environment, and these interactions can potentially be exploited to control this pest. Adults of *D. suzukii* are known to be attracted to fermentation odors (Cha et al. 2012, Hamby and Becher 2016) and other microbial volatiles, such as those associated with the symbiotic yeast *Hanseniaspora uvarum* (Niehaus) Shehata, Mrak & Phaff (Hamby et al. 2012, Bueno et al. 2020, Kleman et al. 2022). Many of these fermentation and microbial odors, such as acetic acid, ethanol, methionol, acetoin, isoamyl acetate, and isobutyl acetate, have been

used in lure-baited traps for monitoring this pest in the field (Iglesias et al. 2014, Cha et al. 2017, Swoboda-Bhattari et al. 2017, Cha et al. 2018, Cloonan et al. 2019, Spitaler et al. 2022). However, not all microbial associations elicit positive responses. Pathogenic microbes can provide sources of repellent or oviposition deterrent compounds. For example, 1-octen-3-ol and geosmin, associated with contamination and spoilage of stored food products, respectively, are known repellents against *D. suzukii* (Wallingford et al. 2017). Another microbial volatile, 2-pentylfuran, isolated from fermenting wheat bread dough has also been identified as having oviposition deterrent properties (Cha et al. 2021). Among the three previously identified repellents for *D. suzukii*, 2-pentylfuran has shown the most promise due to its effectiveness under field conditions and environmental friendliness. Additionally, Cha et al. (2020) discovered that *D. suzukii* females were less likely to oviposit in raspberries infected with *Botrytis cinerea* Pers., the causative agent of gray mold disease; however, the volatiles responsible for this deterrence remain unidentified. Moreover, *B. cinerea*-infected raspberries significantly reduced larval survival and adult size in *D. suzukii* (Cha et al. 2020). These findings suggest that other plant diseases might similarly influence *D. suzukii* behavior and development.

Blueberries are susceptible to a disease known as anthracnose fruit rot, caused by species of the genus *Colletotrichum*, including *Colletotrichum fioriniae* Marcelino & Gouli (Damm et al. 2012, Pszczółkowska et al. 2016). *Colletotrichum fioriniae* is an ascomycete fungus that spreads to blooms and healthy berries via rain or wind dispersal. Symptoms of anthracnose in blueberries include the development of orange or salmon-colored droplets containing the fungus's conidia and the eventual collapse of the fruit, rendering it unmarketable (Miles and Schilder 2013). Early studies on the interaction

between anthracnose and *D. suzukii* have shown that, given a choice, sexually mature females are three times more likely to select healthy blueberry fruits over anthracnose-infected ones (Urbaneja-Bernat et al. 2020). Additionally, *D. suzukii* oviposited fewer eggs in infected berries, and anthracnose infection reduced adult emergence. More recent research by Rering et al. (2023) identified two esters—ethyl butanoate and ethyl (*E*)-but-2-enoate—from anthracnose-infected blueberries as being equally or more repellent than known *D. suzukii* repellents such as 1-octen-3-ol, geosmin, and 2-pentylfuran. Given these promising results, the present study aims to further investigate whether *D. suzukii* antennae can detect these esters and how these compounds influence the fly's oviposition behavior under semi-field and field cage conditions.

In this study, we evaluated the antennal responses of *D. suzukii* to ethyl butanoate and ethyl (*E*)-but-2-enoate using electroantennography and assessed their oviposition-deterrent activity in outdoor cages. We hypothesized that *D. suzukii* can detect these compounds via their antennae and that they function as oviposition deterrents. These findings could contribute to the development of new compounds for the behavioral manipulation of this pest, offering alternative integrated pest management strategies for *D. suzukii*.

Materials and methods

Insect rearing

The *D. suzukii* colony used for experiments was established in 2013 and maintained on a standard artificial diet (Jaramillo et al. 2015) at the Rutgers P.E. Marucci Center (Chatsworth, NJ). The colony was kept under controlled conditions at 22 ± 2 °C, 55 ± 5%

relative humidity (RH), and a 16:8 h L:D cycle. To maintain genetic diversity, wild flies were introduced into the colony every 2–3 years. The flies used in the experiments were 5–10 days old, ensuring they were sexually mature (Revadi et al. 2015).

Synthetic Chemicals

Ethyl butanoate (99%, CAS No. 105-54-4), ethyl (*E*)-but-2-enoate (99%, CAS No. 623-70-1), and 2-pentylfuran (≥98%, CAS No. 3777-69-3) were purchased from Sigma-Aldrich (St. Louis, MO, USA).

Electroantennogram experiments

Electroantennogram (EAG) assays were conducted to determine the antennal response of sexually mature male and female *D. suzukii* to ethyl butanoate, ethyl (*E*)-but-2-enoate, and 2-pentylfuran. The antennal responses of male and female *D. suzukii* to each compound were tested at five doses (0.001 mg, 0.01 mg, 0.1 mg, 1 mg, and 10 mg) diluted in n-hexane. The stimulus cartridge preparations, antennal preparations, and EAG apparatus used were similar to those described in Cloonan et al. (2019). Stimulus applicators consisted of a 14.5-cm-long glass Pasteur pipette containing 20 µl of each volatile dose (or n-hexane control) pipetted onto a 6 × 0.5-cm strip of filter paper. The applicators containing the impregnated filter paper were placed under the fume hood for 2 min to allow the n-hexane to evaporate. For the recording and base electrodes, a silver wire was inserted into a drawn capillary tube filled with phosphate-buffered saline (NaCl, 4 g; NA_2HPO_4, 0.57 g; KH_2PO_4, 0.1 g; KCL, 0.1 g in 500 ml distilled water). To attach the base electrode to the fly, the fly's abdomen was removed, and the sharp tip of the

saline-filled capillary tube was pulled directly into the thoracic cavity. Once the fly preparation was mounted, the recording electrode was carefully moved toward the antenna using a micromanipulator until the antenna touched the pool of saline solution on the recording electrode. Antennal preparations were exposed to a constant stream of charcoal-filtered and humidified air at a rate of 1.5 L/min.

The EAG apparatus consisted of an IDAC-02 interface board for data acquisition and used Syntech software (Syntech Ltd., Hilversum, The Netherlands) for recording, storing, and quantifying EAG responses. Antennal preparations were primed with 1 mg of acetoin, a known attractant and antennally active compound (Cha et al. 2012, Cloonan et al. 2019), to ensure that the antennae were prepared correctly and responsive (positive control). Then, the antennae were exposed to an n-hexane control, followed by exposure to increasing doses of one of the volatiles. Each antenna was exposed to four rounds of each dose of a single compound before being discarded. Six antennae were tested daily: three from males and three from females. Test and control compounds were applied at 10-s intervals at a pulse rate of 0.5 s, with a 1-min interval between each stimulus. Maximum amplitudes of depolarizations were measured (in millivolts) for each compound with the response from the n-hexane-only controls subtracted from the other doses to normalize the antennal response. In total, each dose of each compound was replicated 10 times for each sex.

Semi-field cage experiments

Semi-field cage experiments were conducted over a seven-week period, from 20 June until 9 August of 2023 at the Rutgers P.E. Marucci Research Center (mean ± SE

168 temperature: 23.5 ± 0.27°C; RH: 78.8 ± 0.9%) to evaluate the oviposition deterrent
169 effects of ethyl butanoate, ethyl (*E*)-but-2-enoate, and 2-pentylfuran against *D. suzukii*.
170 These experiments are considered semi-field because they took place in an isolated open
171 field surrounded by woods with potted plants inside outdoor cages. The cages were
172 constructed using polyvinyl chloride (PVC) pipes (3.8 cm diameter) to construct 1.8 m ×
173 1.8 m × 1.8 m frames. Screen tents (*Lumite*® screen portable field cages; BioQuip, CA,
174 USA) were placed over the PVC frames, and large nails were used to secure the tents to
175 the ground (Fig. 1A). Cages were spaced 10 m apart.
176 To obtain blueberries for the experiment, prior to the start of the experiment,
177 field-grown highbush blueberry (*Vaccinium corymbosum* L. var. 'Bluecrop') clusters
178 were bagged early in the season with cloth bags when they were still green to prevent
179 infestation from resident *D. suzukii* populations for use in the experiment. In addition, 80
180 3–4-year-old potted blueberry bushes were stripped of their berries and used for the
181 experiment.
182 On the day of testing, two bushes were placed in each cage (Fig. 1B). Each bush
183 in the tent was surrounded by a metal tomato cage. The bagged blueberry fruit clusters in
184 the field were clipped and brought inside the cages. Clusters of 10 berries were created
185 and placed in water picks. Five berry clusters were distributed randomly and evenly on
186 each tomato cage at different heights using green twist ties (Fig. 1B). The sachets used
187 for the treatments were prepared according to the methods described by Gale et al.
188 (2024). They were constructed from 8 cm strips of polyethylene tubing (5.1 cm width, 2
189 MIL thickness; ULINE, Pleasant Prairie, WI, USA) and polyester felt (Grainger, Lake
190 Forest, IL, USA). One end of the polyethylene tubing was sealed with an impulse sealer.

A 5 cm strip of felt was placed inside the sachet's open end, and a 2.5 mL aliquot of each treatment (neat compound) was pipetted onto the felt. The open end was then sealed shut with the impulse sealer, entirely sealing the saturated felt. A hole was punched at the top of each sachet, away from the treatment area to avoid damaging the sealed section, and green twist ties were used to hang the sachets in the center of a bush (Fig 1B). Before loading the sachets, the weights of the empty sachets were taken. The sachets were weighed again directly after loading with the compounds and then a final time after the 24-hour test period to measure the emission rates of each volatile.

In each cage, one of the bushes contained a sachet with the test volatile, while the other contained a blank sachet (control). Sachets were hung in the bushes 30 minutes prior to the start of experiments. Choice tests included: 1) control versus control; 2) control versus ethyl butanoate; 4) control versus ethyl (*E*)-but-2-enoate; and 4) control versus 2-pentylfuran. Fifty flies (1:1 male: female) were released in each cage at 18:00 hrs. After 24 hours, the berry clusters were collected and placed in 118 mL plastic cups. Berries were inspected under a dissecting microscope (AmScope SM-1) for the number of eggs laid and were then incubated in 236.6 mL (8-oz) deli containers lined with two cotton pads on a laboratory bench at 22 ± 2°C and 55 ± 5% relative humidity for two weeks to monitor for adult emergence. The study was replicated 10 times for each choice combination ($N = 4$ choice combinations × 2 bushes each × 10 replicates = total of 80 bushes; $N = 50$ flies × 4 choice combinations × 10 replicates = total of 2,000 flies).

Field cage experiments

213 Field cage experiments were conducted over a two separate periods, from 24–27 June
214 (mean ± SE temperature: 25.1 ± 0.9°C; RH: 76.1 ± 5.9%) and 8–11 July (27.6 ± 0.1°C;
215 RH: 80.3 ± 2.1%) of 2024, at the Rutgers P.E. Marucci Research Center using highbush
216 blueberries (*V. corymbosum* var. 'Bluecrop'). The studies were carried out in 5.5 m long
217 × 2.7 m tall cages, constructed with a PVC pipe frame covered with No-See-Um mesh
218 (Quest Outfitters Inc., Sarasota, FL, USA) (Fig. 1C). Cages were placed in separate rows
219 within a blueberry field, with bushes spaced approx. 0.76 m apart within the rows and
220 3.05 m between rows. Each row contained two cages at least 9 m in distance. To avoid
221 interference between treatments, alternate cages were used during testing, ensuring that
222 no adjacent cages contained sachets simultaneously.
223 Two, 3-m rebar poles were used to reinforce the mesh on the long sides of each
224 cage as well as 10 cm long garden staples to hold mesh flush to the ground, while the
225 shorter ends were secured with clips to allow access to the cage. Each cage contained five
226 blueberry bushes approx. 1.5 m in height. Sachets containing ethyl butanoate, ethyl (*E*)-
227 but-2-enoate, or 2-pentylfuran were prepared as described before. To test the effects of
228 the repellents on the treated (focal) bush and in neighboring bushes at various distances,
229 two sachets of the same compound were hung from one of the end bushes in each cage
230 (Fig. 1D). There were four treatments: 1) ethyl butanoate, 2) ethyl (*E*)-but-2-enoate, 3) 2-
231 pentylfuran, and 4) control (no repellent). Each treatment was replicated four times, with
232 each cage assigned to a single treatment, resulting in a total of 16 cages.
233 Twenty-four hours after deploying the sachets, 120 flies (approximately 60 males
234 and 60 females) were released into each cage at 18:00 hrs (N = 120 flies × 4 treatments ×
235 4 replicates = 1,920 total flies). To ensure even distribution within the cages, 30 flies

were released at four equidistant points between the bushes. A pre-sampling of the berries, conducted before the flies were released, confirmed the absence of any infestation. Berries were then collected 1-, 2-, and 3-days post-treatment. From every other bush (starting at the focal bush) in the cage, 50 berries from the top half and 50 from the bottom half were collected into two separate 236.6 mL deli containers lined with two cotton pads. From each berry sample, a random subsample of 10 berries were examined and egg counts recorded before being returned to the original container. The blueberries were incubated on a light bench in the laboratory for up to two weeks, as previously described, and adult emergence was recorded. The sachets were weighed before deployment and 3 days after deployment to measure the emission rates of each compound.

Statistical analysis

To analyze the effect of dose of the three repellents on *D. suzukii* antennal responses, a generalized linear model (GLM) was used with a Poisson distribution and a log link function in SPSS Statistics 23.0 (IBM Corp, Armonk, NY, USA). The model included 'Treatment' (ethyl butanoate, ethyl (*E*)-but-2-enoate, and 2-pentylfuran), 'Dose', 'Sex', and their interactions as independent variables. This analysis, when significant, was followed by post hoc Bonferroni tests ($\alpha = 0.05$) to determine individual differences among groups. Prior to analysis, EAG data were normalized relative to the response to the n-hexane control.

Semi-field cage data were analyzed using paired *t*-tests to determine differences between the number of eggs laid and adults that emerged from treated berries compared

to control berries, and survival data were analyzed using two-way analysis of variance (ANOVA) (R statistical software version 4.1.1; R Development Core Team, Vienna, Austria). In addition, a deterrence index (DI) was calculated for each treatment as follows:

$$DI = \frac{(n_{control} - n_{volatile})}{n_{total}}$$

Where $n_{control}$, $n_{volatile}$, and n_{total} are the number of eggs laid in the control fruits, treatment fruits, and total number of eggs laid in the control and treatment fruits, respectively. The DI values were compared among treatments using ANOVA (R statistical software). Before the analysis, data were checked for normality and equal variance using an Anderson-Darling test and Levene's test, respectively.

Field cage data for both oviposition and adult emergence were non-normal, so non-parametric tests were applied using R statistical software. The effects of 'Treatment' (untreated control, ethyl butanoate, ethyl (*E*)-but-2-enoate, and 2-pentylfuran) across 'Bush Position' (focal, center, end) and 'Day After Treatment' (1, 2, 3 DAT) were assessed using the Kruskal-Wallis Test. When significant Treatment effects were found, post-hoc separation was performed using Dunn's Test. Initial analyses showed no significant differences in oviposition or adult emergence based on location on the bush (top vs. bottom); therefore, the data were averaged and analyzed at the bush level.

Finally, the emission rates of compounds in the semi-field and field cage studies were calculated by subtracting the final weight of the sachets from the initial weight and dividing by the time interval (24 h or 3 days). These rates were then compared across repellent treatments using ANOVA, with Tukey pairwise comparisons conducted when significant differences were found.

Results

Electroantennogram experiments

Both male and female *D. suzukii* exhibited dose-dependent antennal responses to ethyl butanoate, ethyl (*E*)-but-2-enoate, and 2-pentylfuran (Table 1; Fig. 2). The strength of the EAG responses varied among treatments, with ethyl butanoate and ethyl (*E*)-but-2-enoate eliciting stronger antennal responses than 2-pentylfuran (Table 1; Fig. 2). There was no significant effect of sex on the antennal responses to these compounds, nor was there a significant interaction between treatment and sex (Table 1), indicating that the antennal responses of male and female *D. suzukii* to ethyl butanoate, ethyl (*E*)-but-2-enoate, and 2-pentylfuran were similar. Although the EAG responses increased with rising doses of the compounds, the antennae of *D. suzukii* responded more strongly to ethyl butanoate and ethyl (*E*)-but-2-enoate than to 2-pentylfuran at higher doses (Fig. 2), as indicated by the significant treatment-by-dose interaction (Table 1).

Semi-field cage experiments

Semi-field cage assays using potted blueberry plants were used to test the efficacy of ethyl butanoate, ethyl (*E*)-but-2-enoate, and 2-pentylfuran on *D. suzukii* oviposition. There were no differences in the emission rates of the three compounds ($F = 1.66$; df = 2,12; $P = 0.231$; mean emission rates (± SE) were 70.4 ± 14.2 mg/h for ethyl butanoate, 54.9 ± 8.9 mg/h for ethyl (*E*)-but-2-enoate, and 82.3 ± 7.6 mg/h for 2-pentylfuran). *Drosophila suzukii* consistently laid fewer eggs in berries paired with the treatments in comparison to control berries (Fig. 3A). Flies laid 54% fewer eggs in ethyl butanoate-

305 treated berries (t = 2.29, P = 0 .047), 75% fewer berries in ethyl (*E*)-but-2-enoate treated
306 berries (t = 3.76, P = 0.005), and 67% fewer eggs in 2-pentylfuran treated berries (Fig.
307 3A).

308 When comparing the emergence of adult progeny (Fig. 3B), more flies emerged
309 from the control berries compared to the berries treated with the two anthracnose-
310 associated compounds, with 59% fewer flies emerging from ethyl butanoate treated
311 berries (t = 4.07, P = 0.003) and 78% fewer flies emerging from ethyl (*E*)-but-2-enoate
312 treated berries (t = 2.81, P = 0.02). The difference in adult emergence between untreated
313 berries and 2-pentylfuran treated berries was nonsignificant (t = 2.06, P = 0.069).

314 The percentage of eggs that survived to adulthood ranged from 57% (±14%) to
315 83% (±8%) across all treatments (Fig. 3C). A two-way ANOVA showed no significant
316 differences in offspring survival among the treatments or between treatments and controls
317 (all P values > 0.05), indicating that the treatments only affected *D. suzukii* oviposition
318 behavior which resulted in reduced adult emergence.

319 After calculating the DI based on the number of eggs laid within each cage, ethyl
320 butanoate, ethyl (*E*)-but-2-enoate, and 2-pentylfuran all performed similarly and were
321 significantly different from the control (F = 4.85; df = 4,36; P = 0.005) (Fig. 4).

322

323 ## Field cage experiments

324 Field cage assays using cultivated blueberry bushes tested the efficacy of ethyl butanoate,
325 ethyl (*E*)-but-2-enoate, and 2-pentylfuran on *D. suzukii* oviposition. In these trials, the
326 emission rates differed significantly among the three tested compounds (F = 16.29; df =
327 2,27; P < 0.001). The emission rates for ethyl butanoate (mean ± SE = 697.97 ± 7.96

328 mg/day) and ethyl (*E*)-but-2-enoate (679.23 ± 17.73 mg/day) were significantly higher
329 than those for 2-pentylfuran (559.73 ± 23.54 mg/day).
330 Egg counts were higher in the control than all the treatment groups regardless of
331 bush position or DAT (Fig. 5A). Among the three repellent treatments, the response
332 varied depending on the bush position and DAT. At 1 DAT, all repellent treatments
333 reduced egg counts in the focal bush compared to the control ($\chi^2 = 222.01$; df = 3; $P <$
334 0.001), but there were no differences among them; 2-pentylfuran was lower at the central
335 bush ($\chi^2 = 206.70$; df = 3, $P < 0.001$); and 2-pentylfuran and ethyl (*E*)-but-2-enoate had
336 the fewest eggs at the end bush ($\chi^2 = 143.43$, df = 3, $P < 0.001$) (Fig. 5A). At 2 DAT, the
337 treatments again demonstrated similar oviposition repellency at the focal bush ($\chi^2 =$
338 174.37; df = 3; $P < 0.001$), but ethyl (*E*)-but-2-enoate had the lowest egg counts at both
339 the center ($\chi^2 = 151.81$; df = 3; $P < 0.001$) and the end bush ($\chi^2 = 148.01$; df = 3; $P <$
340 0.001) (Fig. 5A). By 3 DAT, ethyl (*E*)-but-2-enoate had the lowest egg count at all three
341 bushes sampled (focal: $\chi^2 = 91.26$; df = 3; $P < 0.001$; center: $\chi^2 = 91.31$; df = 3; $P <$
342 0.001; end: $\chi^2 = 85.12$; df = 3; $P < 0.001$) (Fig. 5A).
343 The emergence of adult progeny was also higher in the untreated control cages
344 than any of the repellent treatments (Fig. 5B). At 1 DAT and 2 DAT, there were
345 differences in adult emergence between the control and all the repellent treatments at the
346 focal bush (1 DAT: $\chi^2 = 25.01$; df = 3; $P < 0.001$; 2 DAT: $\chi^2 = 25.48$; df = 3; $P < 0.001$)
347 but no differences among them; ethyl (*E*)-but-2-enoate had the fewest adults emerge at
348 the center (1 DAT: $\chi^2 = 25.48$; df = 3; $P < 0.001$; 2 DAT: $\chi^2 = 23.20$; df = 3; $P < 0.001$)
349 and end bush (1 DAT: $\chi^2 = 19.56$; df = 3; $P < 0.001$; 2 DAT: $\chi^2 = 14.94$; df = 3; $P <$
350 0.001) (Fig. 5B). At 3 DAT, 2-pentylfuran and ethyl (*E*)-but-2-enoate had the lowest

adult emergence at the focal ($\chi^2 = 19.20$; df = 3; $P < 0.001$) and center bush ($\chi^2 = 15.72$; df = 3; $P < 0.001$) but, at the end bush, only ethyl (*E*)-but-2-enoate was significantly different compared to the other repellents ($\chi^2 = 15.35$; df = 3; $P < 0.001$) (Fig. 5B).

Discussion

This study demonstrated that: 1) both male and female antennae of *D. suzukii* can detect ethyl butanoate, ethyl (*E*)-but-2-enoate, and 2-pentylfuran in a dose-dependent manner; and 2) these repellent compounds reduce *D. suzukii* oviposition and adult emergence in blueberry fruits under semi-field and field cage conditions.

After anthracnose-infected blueberries were found to repel *D. suzukii* (Urbaneja-Bernat et al. 2020), Rering et al. (2023) screened 14 volatiles emitted at higher levels in infected berries compared to healthy ones for their repellent activity against this pest in laboratory studies. They found that nine of these volatiles had repellent properties. Among them, two esters—ethyl butanoate and ethyl (*E*)-but-2-enoate—showed the strongest repellent effects, and as demonstrated in this study, these compounds also trigger strong dose-dependent antennal responses in adult *D. suzukii* and act as oviposition deterrents under semi-field and field cage conditions. These compounds are naturally present in the headspace of blueberries and are mainly associated with fruit ripening (Beaulieu et al. 2014, Farneti et al. 2017). Since anthracnose infections cause rapid ripening and collapse of the fruit (Miles and Schilder 2013), simultaneously the emission rate of these volatiles increases (Rering et al. 2023). Given that *D. suzukii* are typically attracted to ripening or ripe fruits for oviposition rather than overripe ones (Lee et al. 2011, Keesey et al. 2015), the observed oviposition deterrent effects in this study

374 may indicate that the flies perceive the fruits as beginning to rot, thus discouraging them
375 from laying eggs.

376 Ethyl butanoate has been previously identified as an antennally active compound
377 in *D. suzukii* (Cloonan et al. 2019, Urbaneja-Bernat et al. 2021) and shown to reduce
378 attraction to lures (Cha et al. 2012). However, its role as an oviposition deterrent under
379 field conditions had not been confirmed until this study. Ethyl (*E*)-but-2-enoate, while
380 structurally similar to ethyl butanoate, has also not been previously identified as an
381 oviposition deterrent. Both compounds elicited similar dose-dependent responses in EAG
382 assays for both male and female *D. suzukii*. However, when compared to the known
383 repellent 2-pentylfuran, these esters showed comparable or stronger antennal detection
384 efficacy, especially at higher doses. A similar trend was observed in the semi-field cage
385 studies, where ethyl butanoate, ethyl (*E*)-but-2-enoate, and 2-pentylfuran significantly
386 reduced *D. suzukii* oviposition in treated berries compared to the control. In the field cage
387 studies, ethyl (*E*)-but-2-enoate tended to outperform both ethyl butanoate and 2-
388 pentylfuran, particularly at longer distances from the focal plant and after at least three
389 days of deployment, demonstrating greater oviposition deterrent activity.

390 Urbaneja-Bernat et al. (2020) demonstrated that only female *D. suzukii* were
391 repelled or deterred from ovipositing by volatiles from anthracnose-infected fruits, likely
392 because they are searching for suitable oviposition sites. Anthracnose infection likely
393 reduces the quality of fruits for *D. suzukii* offspring development (Urbaneja-Bernat et al.
394 2020), showing a positive relationship between female oviposition preference and
395 offspring performance. However, when examining the antennal response of *D. suzukii* to
396 ethyl butanoate and ethyl (*E*)-but-2-enoate, both males and females showed similar

397 responses, indicating that both sexes can detect these compounds. Although males may
398 be less behaviorally responsive to anthracnose-infected fruits than females, both male and
399 female *D. suzukii* were found to be repelled by these compounds in laboratory assays
400 (Rering et al. 2023). The role of these volatiles in influencing male behaviors remains
401 unclear.

402 Ethyl butanoate and ethyl (*E*)-but-2-enoate have demonstrated equal or superior
403 performance compared to other known *D. suzukii* repellents, such as 2-pentylfuran,
404 geosmin, and 1-octen-3-ol (Rering et al. 2023; this study), showing promise as effective
405 repellents and oviposition deterrents against this pest. Future research should explore
406 whether ethyl butanoate and ethyl (*E*)-but-2-enoate synergize with other *D. suzukii*
407 repellents or with repellent compounds identified in anthracnose-infected blueberries
408 (Rering et al. 2023). Combining these compounds could help maintain their repellent
409 efficacy in the field, especially since ethyl butanoate and ethyl (*E*)-but-2-enoate are more
410 volatile than other known *D. suzukii* repellents. Further research is also needed to identify
411 optimal deployment methods for these compounds. In the field cage study, ethyl
412 butanoate and ethyl (*E*)-but-2-enoate exhibited higher emission rates than 2-pentylfuran,
413 with minimal amounts remaining in the sachets after 3 days. Employing slow-release
414 technologies, such as the inert matrix SPLAT® (Specialized Pheromone and Lure
415 Application Technology) (Wallingford et al. 2016a), aerosol diffusers (Stockton et al.
416 2021), and nanoencapsulation (de Oliveira et al. 2018), could help sustain adequate
417 emission rates of these compounds in the field.

418 Previously, repellents against *D. suzukii* have shown some success in reducing
419 infestations in raspberries in both greenhouse and field studies (Wallingford et al. 2016a,

Wallingford et al. 2016b, Stockton et al. 2021). However, repellents alone are typically insufficient to fully eradicate *D. suzukii* infestations, which is critical in crops like blueberries where there is zero tolerance for infested fruit (Rodriguez-Saona et al. 2019). Nonetheless, repellent or oviposition deterrent compounds can be valuable tools when used in combination with other behavioral manipulation methods. For example, they could be paired with attract-and-kill devices to develop push-pull systems for *D. suzukii*. Push-pull systems work by using a repellent or oviposition deterrent to "push" the pest away from the target crop, while the "pull" component attracts pests to a kill device (Cook et al. 2007). Push-pull systems using 1-octen-3-ol (Wallingford et al. 2018) or methyl benzoate (Gale et al. 2024) as the push component, combined with an attract-and-kill device as the pull component, have already shown some success in managing *D. suzukii* in raspberries and blueberries.

In conclusion, the current study provides additional evidence that the esters ethyl butanoate and ethyl (*E*)-but-2-enoate, derived from pathogen-infected fruit, could serve as promising oviposition deterrents for *D. suzukii*. While previous studies have shown that pathogen infections and isolated compounds can repel *D. suzukii* and deter oviposition (Urbaneja-Bernat et al. 2020, Cha et al. 2021, Rering et al. 2023), the physiological and behavioral effects of specific compounds remained largely unexplored. In this study, ethyl butanoate and ethyl (*E*)-but-2-enoate elicited dose-dependent antennal responses in *D. suzukii* and significantly reduced oviposition and adult emergence in semi-field and field trials, performing comparably to, or sometimes even better than, the known *D. suzukii* repellent, 2-pentylfuran. Further research is necessary to evaluate the spatial and temporal efficacy of these compounds, as well as optimal deployment

methods, under more realistic field conditions with natural levels of pest pressure. Additionally, their repellent effects on other pests, as well as their compatibility with other *D. suzukii* management tactics such as biological control, need to be explored. Nevertheless, the strong antennal responses and oviposition deterrent effects observed suggest that these compounds could serve as valuable tools for managing *D. suzukii* behavior in the field.

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460 ## References

461 Asplen MK, Anfora G, Biondi A, et al. 2015. Invasion biology of spotted wing
462 Drosophila (*Drosophila suzukii*): a global perspective and future priorities. J. Pest
463 Sci. 88:469–494. https://doi.org/10.1007/s10340-015-0681-z

464 Atallah J, Teixeira L, Salazar R, et al. 2014. The making of a pest: the evolution of a
465 fruit-penetrating ovipositor in *Drosophila suzukii* and related species. Proc. R
466 Soc. B 281(1781):20132840. https://doi.org/10.1098/rspb.2013.2840

467 Beaulieu JC, Stein-Chisholm RE, Boykin DL. 2014. Qualitative analysis of volatiles in
468 rabbiteye blueberry cultivars at various maturities using rapid solid-phase
469 microextraction. J. Amer. Soc. Hort. Sci. 139(2):167–177.
470 https://doi.org/10.21273/JASHS.139.2.167

471 Bueno E, Martin KR, Raguso RA, et al. 2020. Response of wild spotted-wing drosophila
472 (*Drosophila suzukii*) to microbial volatiles. J. Chem. Ecol. 46(8):688–698.
473 https://doi.org/10.1007/s10886-019-01139-4

474 Cha DH, Adams T, Rogg H, et al. 2012. Identification and field evaluation of
475 fermentation volatiles from wine and vinegar that mediate attraction of spotted-
476 wing drosophila, *Drosophila suzukii*. J. Chem. Ecol. 38(11):1419–1431.
477 https://doi.org/10.1007/s10886-012-0196-5

478 Cha DH, Hesler SP, Brind'Amour G, et al. 2020. Behavioral evidence for contextual
479 olfactory-mediated avoidance of the ubiquitous phytopathogen *Botrytis cinerea*
480 by *Drosophila suzukii*. Insect Sci. 27(4):771–779. https://doi.org/10.1111/1744-
481 7917.12691

Cha DH, Landolt PJ, Adams TB. 2017. Effect of chemical ratios of a microbial-based feeding attractant on trap catch of *Drosophila suzukii* (Diptera: Drosophilidae). Environ. Entomol. 46(4):907–915. https://doi.org/10.1093/ee/nvx079

Cha DH, Loeb GM, Linn CE, et al. 2018. A multiple-choice bioassay approach for rapid screening of key attractant volatiles. Environ. Entomol. 47(4):946–950. https://doi.org/10.1093/ee/nvy054

Cha DH, Roh GH, Hesler SP, et al. 2021. 2-Pentylfuran: a novel repellent of *Drosophila suzukii*. Pest Manag. Sci. 77(4):1757–1764. https://doi.org/10.1002/ps.6196

Cloonan KR, Abraham J, Angeli S, et al. 2018. Advances in the chemical ecology of the spotted wing drosophila (*Drosophila suzukii*) and its applications. J. Chem. Ecol. 44(10):922–939. https://doi.org/10.1007/s10886-018-1000-y

Cloonan KR, Hernández-Cumplido J, De Sousa ALV, et al. 2019. Laboratory and field evaluation of host-related foraging odor-cue combinations to attract *Drosophila suzukii* (Diptera: Drosophilidae). J. Econ. Entomol. 112(6):2850–2860. https://doi.org/10.1093/jee/toz224

Cook SM, Khan ZR, Pickett JA. 2007. The use of push-pull strategies in integrated pest management. Annu. Rev. Entomol. 52:375–400. https://doi.org/10.1146/annurev.ento.52.110405.091407

Damm U, Cannon PF, Woudenberg JHC, et al. 2012. The *Colletotrichum acutatum* species complex. Stud. Mycol. 73:37–113. https://doi.org/10.3114/sim0010

de Oliveira JL, Campos EVR, Pereira AES, et al. 2018. Geraniol encapsulated in chitosan/gum arabic nanoparticles: a promising system for pest management in

504 sustainable agriculture. J. Agric. Food Chem. 66(21):5325–5334.
505 https://doi.org/10.1021/acs.jafc.8b00331
506 Diepenbrock LM, Rosensteel DO, Hardin JA, et al. 2016. Season-long programs for
507 control of *Drosophila suzukii* in southeastern U.S. blueberries. Crop Prot. 81:76–
508 84. https://doi.org/10.1016/j.cropro.2015.12.012
509 Disi JW, Sial AA. 2021. Laboratory selection and assessment of resistance risk in
510 *Drosophila suzukii* (Diptera: Drosophilidae) to spinosad and malathion. Insects
511 12(9):794. https://doi.org/10.3390/insects12090794
512 Farneti B, Khomenko I, Grisenti M, et al. 2017. Exploring blueberry aroma complexity
513 by chromatographic and direct-injection spectrometric techniques. Front. Plant
514 Sci. 8:617. https://doi.org/10.3389/fpls.2017.00617
515 Gale CC, Ferguson B, Rodriguez-Saona C, et al. 2024. Evaluation of a push-pull strategy
516 for spotted-wing drosophila management in highbush blueberry. Insects 15(1):47.
517 https://doi.org/10.3390/insects15010047
518 Ganjisaffar F, Demkovich MR, Chiu JC, et al. 2022. Characterization of field-derived
519 *Drosophila suzukii* (Diptera: Drosophilidae) resistance to pyrethroids in
520 California berry production. J Econ Entomol. 115(5):1676–1684.
521 https://doi.org/10.1093/jee/toac118
522 Gress BE, Zalom FG. 2019. Identification and risk assessment of spinosad resistance in a
523 California population of *Drosophila suzukii*. Pest Manag. Sci. 75(5):1270–1276.
524 https://doi.org/10.1002/ps.5240

525 Hamby KA, Becher PG. 2016. Current knowledge of interactions between *Drosophila*
526 *suzukii* and microbes, and their potential utility for pest management. J. Pest Sci.
527 89:621–630. https://doi.org/10.1007/s10340-016-0768-1
528 Hamby KA, Hernández A, Boundy-Mills K, et al. 2012. Associations of yeasts with
529 spotted-wing drosophila (*Drosophila suzukii*; Diptera: Drosophilidae) in cherries
530 and raspberries. Appl. Environ. Microbiol. 78(14):4869–4873.
531 https://doi.org/10.1128/AEM.00841-12
532 Hampton E, Koski C, Barsoian O, et al. 2014. Use of early ripening cultivars to avoid
533 infestation and mass trapping to manage *Drosophila suzukii* (Diptera:
534 Drosophilidae) in *Vaccinium corymbosum* (Ericales: Ericaceae). J. Econ.
535 Entomol. 107(5):1849–1857. https://doi.org/10.1603/EC14232
536 Iglesias LE, Nyoike TW, Liburd OE. 2014. Effect of trap design, bait type, and age on
537 captures of *Drosophila suzukii* (Diptera: Drosophilidae) in berry crops. J. Econ.
538 Entomol. 107(4):1508–1518. https://doi.org/10.1603/EC13538
539 Jaramillo SL, Mehlferber E, Moore PJ. 2015. Life-history trade-offs under different
540 larval diets in *Drosophila suzukii* (Diptera: Drosophilidae). Physiol. Entomol.
541 40(1):2–9. https://doi.org/10.1111/phen.12082
542 Joshi NK, Demchak K, Biddinger D. 2023. Seasonal population of *Drosophila suzukii*
543 (Diptera: Drosophilidae) and pesticide use pattern after its invasion in caneberry
544 crops in Pennsylvania (USA). Arthropoda 1(1):25–34.
545 https://doi.org/10.3390/arthropoda1010005

546 Keesey IW, Knaden M, Hansson BS. 2015. Olfactory specialization in *Drosophila*
547 *suzukii* supports an ecological shift in host preference from rotten to fresh fruit. J.
548 Chem. Ecol. 41(2):121–128. https://doi.org/10.1007/s10886-015-0544-3
549 Kleman I, Rehermann G, Kwadha CA, et al. 2022. *Hanseniaspora uvarum* attracts
550 *Drosophila suzukii* (Diptera: Drosophilidae) with high specificity. J. Econ.
551 Entomol. 115(4):999–1007. https://doi.org/10.1093/jee/toac029
552 Lee JC, Bruck DJ, Curry H, et al. 2011. The susceptibility of small fruits and cherries to
553 the spotted-wing drosophila, *Drosophila suzukii*. Pest Manag. Sci. 67(11):1358–
554 1367. https://doi.org/10.1002/ps.2225
555 Miles TD, Schilder AC. 2013. Host defenses associated with fruit infection by
556 *Colletotrichum* species with an emphasis on anthracnose of blueberries. Plant
557 Health Prog. 14(1). https://doi.org/10.1094/PHP-2013-1125-01-RV
558 Pszczółkowska A, Okorski A, Paukszto Ł, et al. 2016. First report of anthracnose disease
559 caused by *Colletotrichum fioriniae* on blueberry in western Poland. Plant Dis.
560 100(10):2167. https://doi.org/10.1094/PDIS-04-16-0425-PDN
561 Rering CC, Quadrel A, Urbaneja-Bernat P, et al. 2023. Blueberries infected with the
562 fungal pathogen *Colletotrichum fioriniae* release odors that repel *Drosophila*
563 *suzukii*. Pest Manag. Sci. 79(12):4906–4920. https://doi.org/10.1002/ps.7692
564 Revadi S, Lebreton S, Witzgall P, et al. 2015. Sexual behavior of *Drosophila suzukii*.
565 Insects 6(1):183–196. https://doi.org/10.3390/insects6010183
566 Rice KB, Short BD, Leskey TC. 2017. Development of an attract-and-kill strategy for
567 *Drosophila suzukii* (Diptera: Drosophilidae): evaluation of attracticidal spheres

under laboratory and field conditions. J. Econ. Entomol. 110(2):535–542. https://doi.org/10.1093/jee/tow319

Rodriguez-Saona C, Vincent C, Isaacs R. 2019. Blueberry IPM: Past successes and future challenges. Annu. Rev. Entomol. 64:95–114. https://doi.org/10.1146/annurev-ento-011118-112147

Spitaler U, Cossu CS, Delle Donne L, et al. 2022. Field and greenhouse application of an attract-and-kill formulation based on the yeast *Hanseniaspora uvarum* and the insecticide spinosad to control *Drosophila suzukii* in grapes. Pest Manag. Sci. 78(3):1287–1295. https://doi.org/10.1002/ps.6748

Stockton DG, Wallingford AK, Cha DH, et al. 2021. Automated aerosol puffers effectively deliver 1-octen-3-ol, an oviposition antagonist useful against spotted-wing drosophila. Pest Manag. Sci. 77(1):389–396. https://doi.org/10.1002/ps.6028

Swoboda-Bhattarai KA, McPhie DR, Burrack HJ. 2017. Reproductive status of *Drosophila suzukii* (Diptera: Drosophilidae) females influences attraction to fermentation-based baits and ripe fruits. J. Econ. Entomol. 110(4):1648–1652. https://doi.org/10.1093/jee/tox150

Tabuloc CA, Carlson CR, Ganjisaffar F, et al. 2024. Transcriptome analysis of *Drosophila suzukii* reveals molecular mechanisms conferring pyrethroid and spinosad resistance. Sci. Rep. 14:19867. https://doi.org/10.1038/s41598-024-70037-x

Tait G, Mermer S, Stockton D, et al. 2021. *Drosophila suzukii* (Diptera: Drosophilidae): a decade of research towards a sustainable integrated pest management program. J. Econ. Entomol. 114(5):1950–1974. https://doi.org/10.1093/jee/toab158

591 Urbaneja-Bernat P, Cloonan K, Zhang A, et al. 2021. Fruit volatiles mediate differential
592 attraction of *Drosophila suzukii* to wild and cultivated blueberries. J. Pest Sci.
593 94(4):1249–1263. https://doi.org/10.1007/s10340-021-01332-z
594 Urbaneja-Bernat P, Waller T, Rodriguez-Saona C. 2020. Repellent, oviposition-deterrent,
595 and insecticidal activity of the fungal pathogen *Colletotrichum fioriniae* on
596 *Drosophila suzukii* (Diptera: Drosophilidae) in highbush blueberries. Sci. Rep.
597 10:14467. https://doi.org/10.1038/s41598-020-71341-y
598 Wallingford AK, Cha DH, Linn CE, et al. 2017. Robust manipulations of pest insect
599 behavior using repellents and practical application for integrated pest
600 management. Env. Entomol. 46(5):1041–1050. https://doi.org/10.1093/ee/nvx125
601 Wallingford AK, Cha DH, Loeb GM. 2018. Evaluating a push–pull strategy for
602 management of *Drosophila suzukii* Matsumura in red raspberry. Pest Manag. Sci.
603 74(1):120–125. https://doi.org/10.1002/ps.4666
604 Wallingford AK, Connelly HL, Dore Brind'Amour G, et al. 2016a. Field evaluation of an
605 oviposition deterrent for management of spotted-wing drosophila, *Drosophila*
606 *suzukii*, and potential nontarget effects. J. Econ. Entomol. 109(4):1779–1784.
607 https://doi.org/10.1093/jee/tow116
608 Wallingford AK, Hesler SP, Cha DH, et al. 2016b. Behavioral response of spotted-wing
609 drosophila, *Drosophila suzukii* Matsumura, to aversive odors and a potential
610 oviposition deterrent in the field. Pest Manag. Sci. 72(4):701–706.
611 https://doi.org/10.1002/ps.4040
612 Walsh DB, Bolda MP, Goodhue RE, et al. 2011. *Drosophila suzukii* (Diptera:
613 Drosophilidae): invasive pest of ripening soft fruit expanding its geographic range

614 and damage potential. J. Integr. Pest Manag. 2(1):G1–G7.

615 https://doi.org/10.1603/IPM10010

Table 1. Results of a generalized linear model (GLM) for the effects of 'Treatment' (ethyl butanoate, ethyl (*E*)-but-2-enoate, and 2-pentylfuran), 'Dose', 'Sex', and their interactions on the electroantennogram (EAG) responses of *Drosophila suzukii*.

Source of Variation	**Wald χ^2**	**df**	***P*[a]**
(Intercept)	1578.21	1	**< 0.001**
Treatment	91.72	2	**< 0.001**
Dose	477.87	4	**< 0.001**
Sex	0.05	1	0.817
Treatment × Dose	67.78	8	**< 0.001**
Treatment × Sex	5.21	2	0.074
Dose × Sex	1.89	4	0.754
Treatment × Dose × Sex	1.16	8	0.997
[a] Significant P values are indicated in bold.			

Figure captions

Fig. 1. Cage setups used in semi-field (A and B) and field (C and D) trials. In semi-field trials, each cage consisted of two potted blueberry bushes. In field trials, each cage contained five cultivated blueberry bushes. One of the bushes (focal bush) within the cage contained a polyethylene sachet with 2.5 mL of the repellent treatments: ethyl butanoate, ethyl (*E*)-but-2-enoate, or 2-pentylfuran (B, D). Control cages did not contain any repellent treatments.

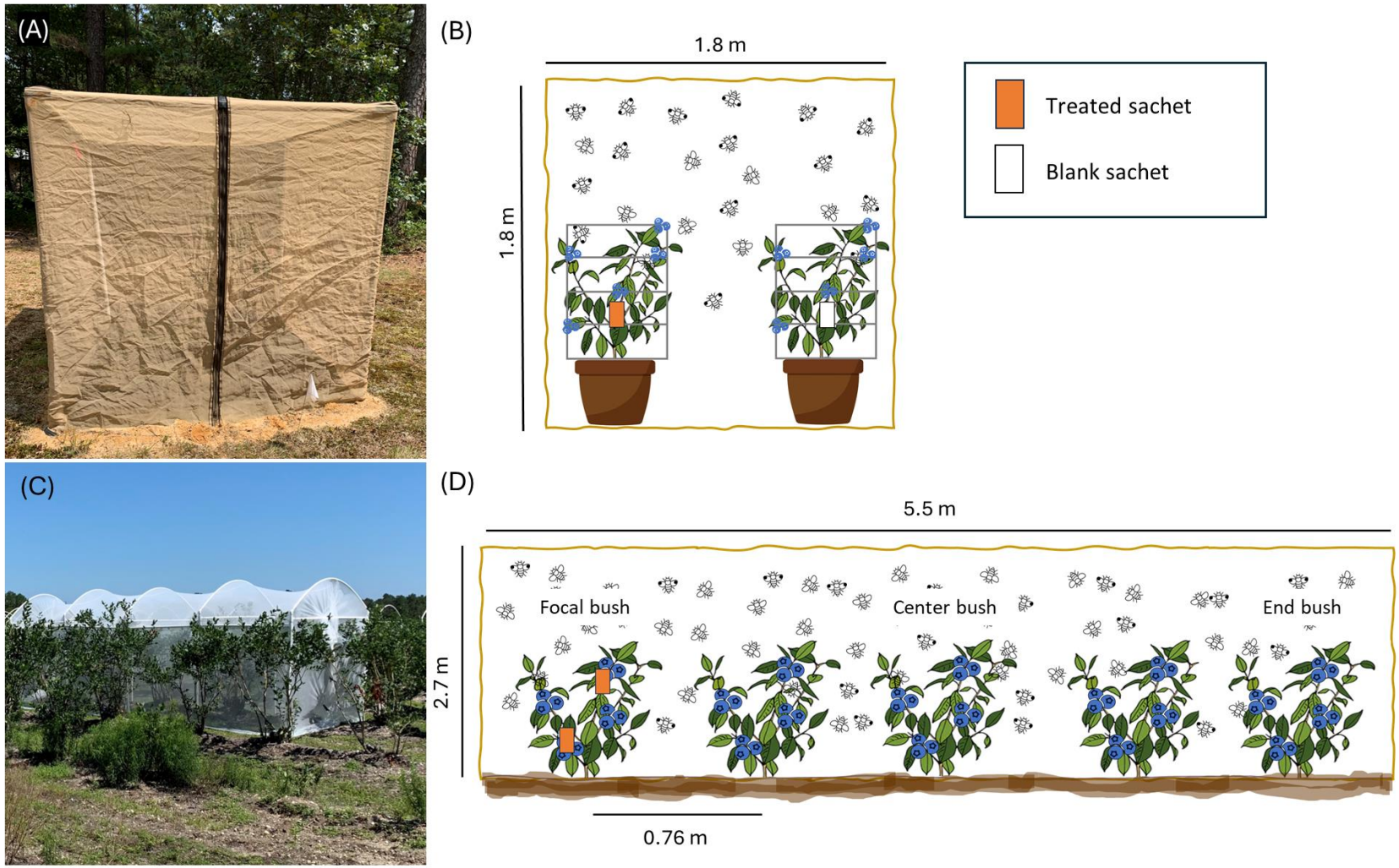
Fig. 2. Electroantennogram (EAG) response curves of male (solid lines) and female (dashed lines) *Drosophila suzukii* antennae to ethyl butanoate (A), ethyl (*E*)-but-2-enoate (B), and 2-pentylfuran (C). EAG amplitudes are presented as antennal depolarizations (mV ± SE) normalized relative to the response to the n-hexane control. Different letters indicate significant differences among doses. $N = 10$.

Fig. 3. Effects of ethyl butanoate, ethyl (*E*)-but-2-enoate, and 2-pentyfuran on *Drosophila suzukii* oviposition (A), adult emergence (B), and survival from eggs to adults (C) in semi-field cage studies. An asterisk indicates significant differences between the control (white bars) and the treated (gray bars) berries within each cage. n.s. = no significant differences between the control and treatment. $N = 10$.

Fig. 4. Effects of ethyl butanoate, ethyl (*E*)-but-2-enoate, and 2-pentyfuran on the deterrence index of *Drosophila suzukii* in semi-field cage studies. The deterrence index was calculated as (number of eggs per berry in the control – number of eggs per berry in

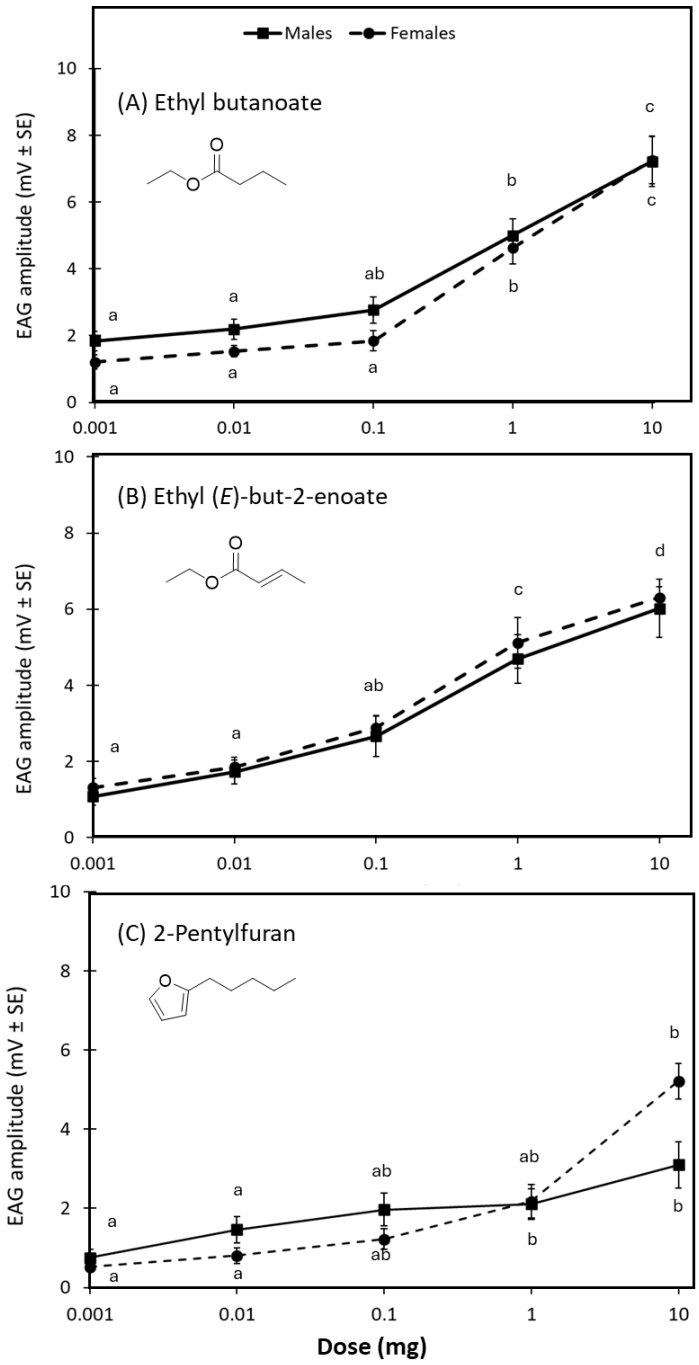
643 the treatment)/total number of eggs per berry. Different letters indicate significant
644 differences among treatments. $N = 10$.
645
646 **Fig. 5.** Effects of an untreated control, ethyl butanoate, ethyl (*E*)-but-2-enoate, and 2-
647 pentyfuran on oviposition (A) and adult emergence (B) of *Drosophila suzukii* in field
648 cages. Egg counts represent the mean number of eggs from a 10-berry subsample, while
649 adult emergence refers to the average number of *D. suzukii* adults emerging from a 50-
650 berry sample. $N = 4$.

651 Fig. 1


(A)
(B)
1.8 m
1.8 m
Treated sachet
Blank sachet
(C)
(D)
5.5 m
Focal bush
Center bush
End bush
2.7 m
0.76 m

652

653 Fig. 2



654

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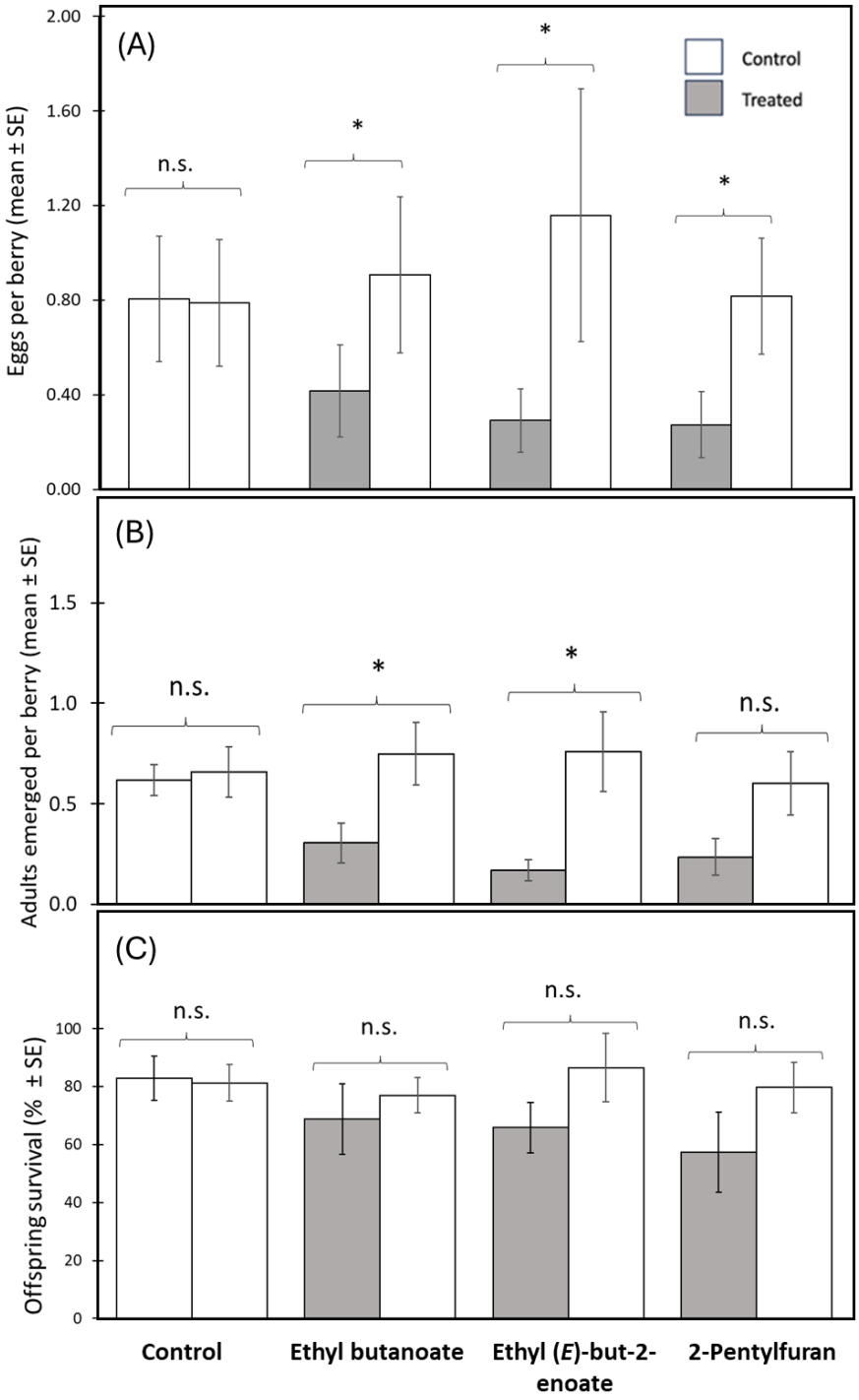
656 Fig. 3



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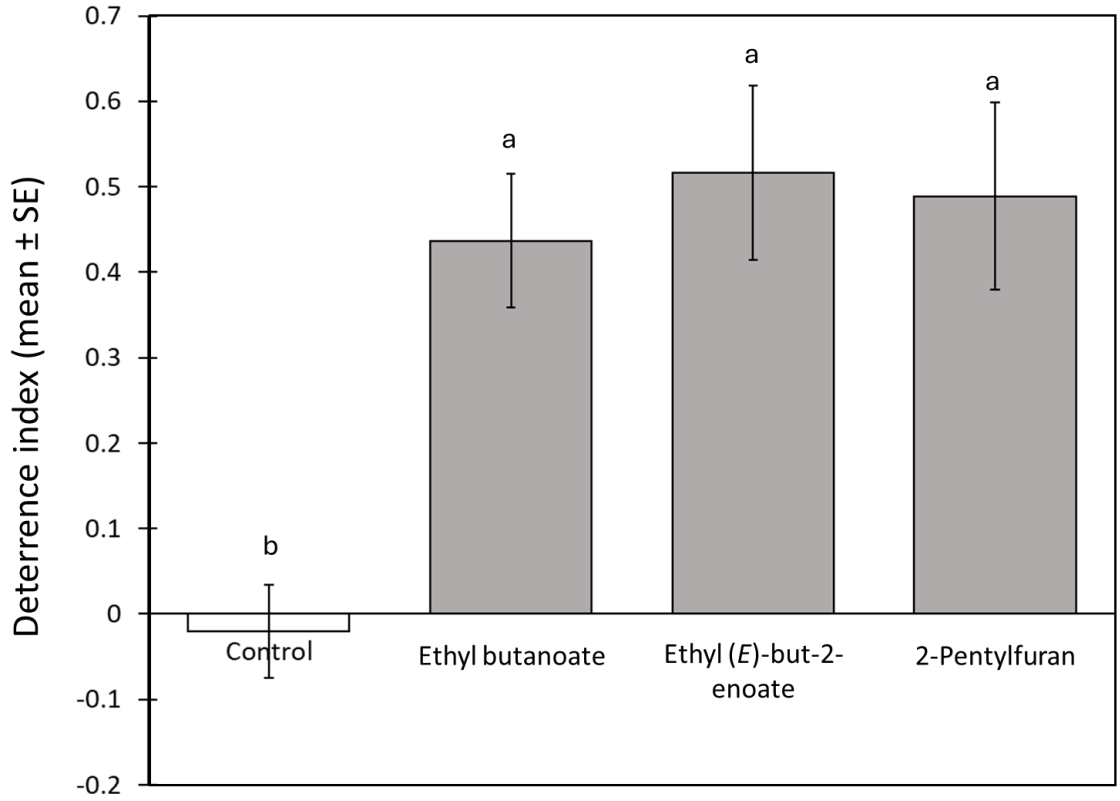
659 Fig. 4



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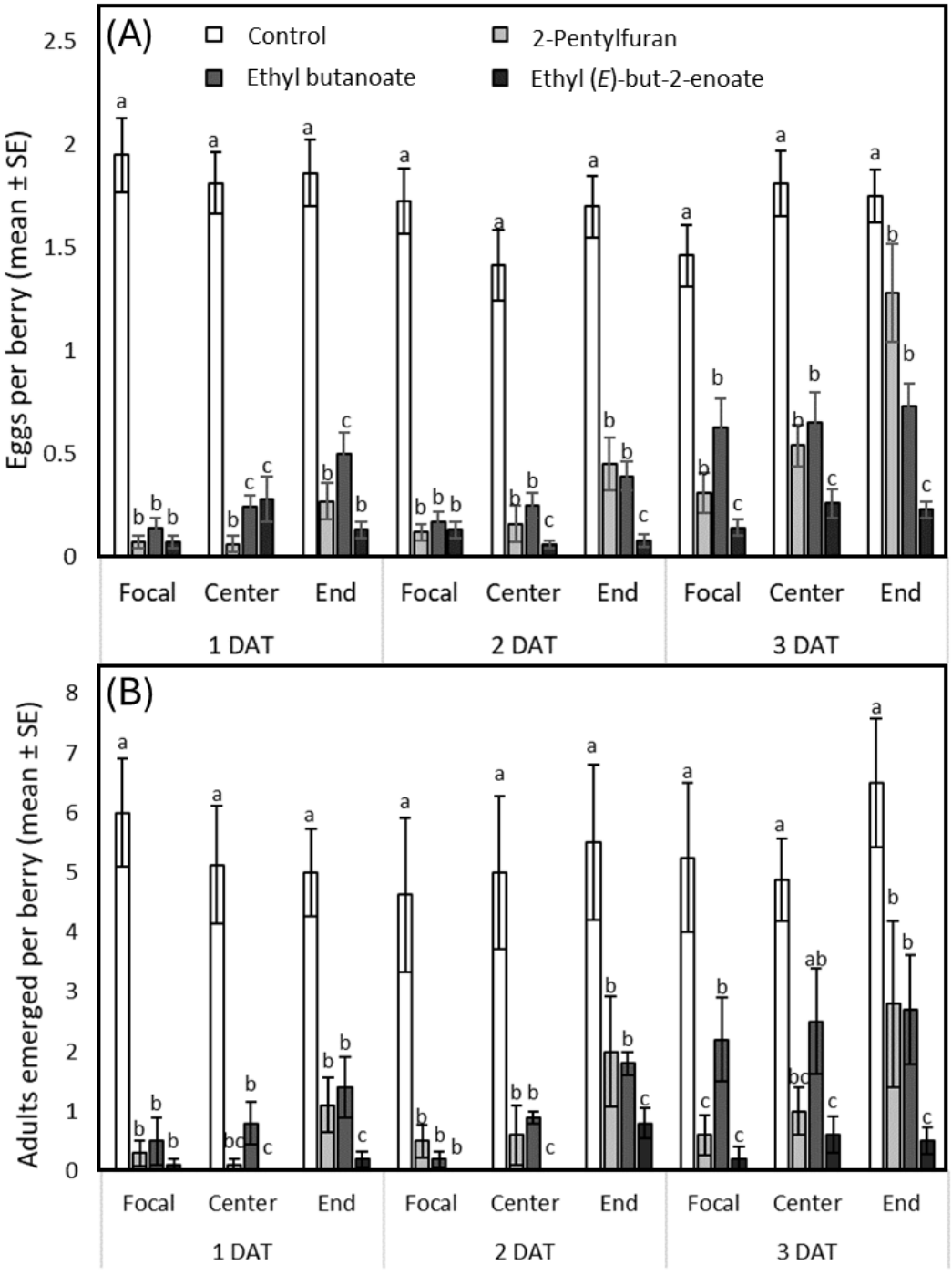
662 Fig. 5


(A)
Control
2-Pentylfuran
Ethyl butanoate
Ethyl (E)-but-2-enoate
Eggs per berry (mean ± SE)
2.5
2
1.5
1
0.5
0
Focal
Center
End
1 DAT
2 DAT
3 DAT
(B)
Adults emerged per berry (mean ± SE)
8
7
6
5
4
3
2
1
0

663