

**Potential of the European earwig (*Forficula auricularia*) as a biocontrol agent of the soft and stone fruit pest *Drosophila suzukii***

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## Abstract

BACKGROUND: The unintentional introduction of *Drosophila suzukii* (Matsumura) from Asia has caused global economic losses in soft and stone fruit industries. Pesticide use can have unintended negative impacts on natural enemies, disrupting attempts to incorporate integrated pest management (IPM) programmes. Generalist predators could potentially act as biocontrol agents of *D. suzukii*. In this context, the predatory capabilities of the European earwig (*Forficula auricularia*) were investigated.

RESULTS: In semi-field conditions, *F. auricularia* were effective at reducing the reproductive rate of *D. suzukii* in more densely populated enclosures. In controlled laboratory conditions, significant negative effects of earwigs were observed for both low (3 breeding pairs) and high (6 breeding pairs) *D. suzukii* densities. Both semi-field and laboratory experiments revealed that *F. auricularia* predation on adult *D. suzukii* could not account for the subsequent reductions in population density.

CONCLUSIONS: Reductions in both larval and adult offspring in the presence of earwigs indicate an impact on *D. suzukii* via predation prior to metamorphosis or disruption of oviposition. Although *F. auricularia* may predate *D. suzukii* populations, its capacity to act as a biocontrol agent may be limited. However, results suggest that *F. auricularia* may be a more effective biocontrol agent earlier in the growing season.

Keywords: beneficial, cherry, Integrated Pest Management, natural enemy, predation, Spotted Wing Drosophila

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## 1. Introduction

The spotted-wing Drosophila (SWD), *Drosophila suzukii* Matsumura, has proven to be highly invasive since its initial detection in new territories across the USA and Europe.<sup>1, 2, 3, 4, 5</sup> The success of SWD predominantly stems from their ability to oviposit in healthy, ripening fruit rather than over-ripe or rotting fruit.<sup>6,7</sup> Subsequently, through fruit imports, this has led to considerable economic losses in soft and stone fruit industries across Asia, the Americas and Europe since 2008.<sup>8, 9, 10, 11</sup> The cherry industry has been one of the most severely impacted due to this pest, accredited to its early ripening period and the lack of alternative hosts during this time.<sup>9, 11, 12</sup> Early oviposition also ensures SWD are often the only *Drosophila* species in foreign territories developing so early in the growing season, thus creating a situation where larval interspecific competition is reduced.<sup>13</sup> The presence of *D. suzukii* in a crop results in economic losses not only via fruit damage, but also via additional costs to fruit growers that are unavoidable to ensure a commercially viable harvest. The deployment and maintenance of monitoring traps, higher labour costs due to increased hygiene requirements and the up-front

expense of netting orchards are some of the examples Swizz cherry growers are incurring to manage SWD.<sup>14,15</sup>

In favourable climatic conditions, up to 13 generations of SWD can develop on fruit in a single growing season.<sup>7</sup> Moreover, mature females can lay in excess of 25 eggs per day.<sup>16</sup> Such short generation times and high reproductive rates not only lead to high pest pressure, resulting in greater economic damage, but could result in the development of insecticide resistance.<sup>1, 14, 17</sup> Although effective neonicotinoid, pyrethroid, spinosyn and organophosphate chemical controls are currently available for SWD management, resistance to the latter three chemical classes has been observed in *D. melanogaster*, engendering uncertainty regarding their future efficacy.<sup>15, 18, 19, 20</sup> In laboratory studies, malathion resistance has been demonstrated in SWD, underscoring the importance of IPM for long-term control.<sup>21</sup>

Parasitic wasps are the primary natural-enemy of SWD in Japan.<sup>22</sup> Host-specificity, common in parasitic wasps, may explain why parasitoids in recently colonized regions are adapted to target native species which develop far later into the growing season than SWD.<sup>23</sup> Therefore, early oviposition may afford SWD a reduced threat from parasitic wasps. European parasitoids have had difficulty switching hosts to SWD since their introduction and have been unable to suppress SWD populations.<sup>24</sup> Natural parasitism rates on *D. melanogaster* and *D. simulans* can reach 90% by *Leptopilina* parasitoids in southern France, revealing the magnitude of the benefit conferred to SWD through avoidance of parasitic wasps in foreign regions.<sup>25</sup> Additionally, some studies have shown that SWD larvae produce large numbers of hemocytes.<sup>26</sup> Hemocyte cells play a key role in 'encapsulation', the primary *Drosophila* immune response.

Encapsulation is effective at killing developing parasitoids, furthering the argument that native parasitoids may not be effective in suppressing SWD populations.<sup>27, 26</sup>

The limited potential of native parasitoids to control SWD populations in regions where they have been introduced necessitates investigation into predators as biocontrol agents. Considerable research has been undertaken in this area using a variety of species targeting SWD at larval, pupal, and adult life-stages. However, many studies utilize controlled laboratory conditions, where SWD are fed directly to predators.<sup>28, 29, 30</sup> Although significant predation was observed, these conditions do not accurately represent the complex ecosystem structure of a fruit orchard, where such predators are often ineffective.<sup>31</sup>

The role of earwigs, particularly the European earwig (*Forficula auricularia* Linnaeus), has largely been neglected when considering the potential of predators as biocontrol agents of SWD. Being well established in North America and native to Europe and Asia,<sup>32</sup> *F. auricularia* could be an effective biocontrol agent across much of the newly established SWD territory and play a contributory role in SWD control. Although omnivorous, *F. auricularia* have been documented as accomplished predators of aphids in both northern European apple orchards and Mediterranean clementine orchards, demonstrating their versatility across ecosystems and crops.<sup>33, 34</sup> Recently, a newly-developed DNA analysis technique revealed that 48.7% of *F. auricularia* collected from the wild had recently consumed SWD, more than twice as prolific as spiders and heteropterans.<sup>35</sup> These studies strongly suggest *F. auricularia* should be considered as an effective biocontrol agent within cherry orchards, with considerable potential to be

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implanted to defend crops against SWD. However, due to their omnivorous nature, *F. auricularia* can feed as generalist predators or by creating small holes in soft and stone fruit, meaning that in certain cases they have been unsuccessful in reducing overall damage to fruit crops.<sup>36, 37</sup> To fully establish the true efficacy of *F. auricularia* as a biocontrol agent of SWD, we investigated the capability of *F. auricularia* as a predator of SWD in cherry in both laboratory and semi-field experiments.

## 2. Materials and methods

### 2.1 Field trials

The field site was located at NIAB EMR in Kent, England (51°17'37" N, 0°26'46" E). Field trials were conducted in an unsprayed cherry orchard (with cvs, Penny and Sweetheart). *D. suzukii* cultures used for the experiment were established at NIAB EMR from an Italian strain collected in 2013. Cultures were held at 25°C in a 16h:8h light/dark cycle and fed on a standard cornmeal diet (100% dH<sub>2</sub>O, 1% Fisher agar, 9% table sugar, 9% precooked ground maize, 2% baker's yeast, 0.2% methylparaben, dissolved in 10 ml 70% ethanol). Undamaged cherries were harvested from the aforementioned cherry orchard and used as the breeding substrate within the field trials. Ripe cherries, a random mix of both cultivars, were picked 24 hours before each assay. The field experiment consisted of two tests: the first was completed without earwigs, the second with earwigs. This allowed for a comparison to be made between reproduction rates of *D. suzukii* populations living with or without the threat of a potential predator.

## *2.2 First field trial without earwigs*

Cherries were placed in Perspex boxes (230 x 125 x 85 mm) lined with paper towel to absorb excess moisture. A cigarette filter (Swan, UK) was placed in a Sterlin 8 ml plastic tube filled with water and covered using Parafilm (Bemis NA, USA) to create a water-tight seal. Forceps were used to pull the filter through the Parafilm seal to create a 'wick', providing water for SWD adults during the experimental period. The water source was taped to the inside of each box, and 8 cherries were also placed inside. A cold table was used to immobilise flies so individual flies could be sexed; breeding pairs were placed into boxes at densities of 0 (control), 1, 3, 7 and 10 pairs, creating 5 treatment densities in total. Control samples were used to ensure fruit was free from prior inoculation of SWD eggs, which could disrupt results. Each treatment density was replicated 6 times, creating a set of 30 boxes. A ventilated lid was taped onto each box to prevent flies escaping. Before boxes were moved to the field all SWD individuals were mobile and fully active.

The experiment was deployed in the cherry orchard on 20/07/17. Boxes were hung in every other tree, in red delta traps to protect against rain and direct sunlight, 3 trees from the perimeter of the orchard. Treatments were randomly distributed and each tree held one of each treatment. A data logger was placed in a delta trap in trees at either end of the trial to record temperature and humidity.

Boxes were removed from the orchard on 27/07/17. Of the 8 cherries in each box, 4 were removed and placed into small plastic bags (~100 x 70 mm flattened area) for larval



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extraction. A sugar solution of 70 g sugar in 1L distilled water was poured into each bag until just covering the cherries. Cherries were then gently broken open using fingers and rested for 10 minutes, broken apart further, and left for a further 10 minutes. Larvae floated to the surface and were then counted and recorded. To record the number of first-generation offspring naturally emerging from cherries, remaining adult SWD were removed from boxes which were then resealed containing the final 4 cherries. Boxes were maintained for 14 days at 24.5°C in a 16h:8h light/dark cycle for 14 days. Afterwards, boxes were frozen for 3 hours and newly emerged adults counted.

### *2.3 Second field trial with earwigs*

For the second experiment earwigs, *F. auricularia*, were introduced into arenas with SWD adults. The earwigs were collected by tap sampling from blackcurrant bushes at NIAB EMR. Earwigs were collected the morning of deployment in the field and were provisioned with water until being transferred to the assay.

The experimental procedure remained the same as for the first experiment, except that one earwig was added to each box after the SWD adults. Male and female earwigs were added alternately so that treatment densities had an equal number of males and females. Earwigs were not added until all flies had become fully active following immobilisation on the cold table. Boxes for this experiment were placed in the orchard on 11/08/17. To maintain the same randomisation, each box was placed in the same delta trap as its respective equivalent in the first trial (same treatment density and replicate number). The data loggers were placed in the

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same positions as the previous trial. Boxes were removed from the orchard on 18/08/17. Unlike the first trial, in the earwig trial the number of dead or alive SWD adults remaining was recorded prior to incubation. This was to give an indication of the maximal level of inferred predation on SWD adults by earwigs as indicated by the reduction in SWD flies relative to the original number placed into each box.

#### *2.4 Insectary trials*

The second trial took place during August, when temperatures were higher than during the first trial in July. To investigate the impact of climatic differences between the two field experiments, a laboratory experiment, set up at the University of Southampton, was designed where trials with and without earwigs were run simultaneously. SWD cultures used were established on a cornmeal diet (for 42.5 L of food: 39 L water, 675 g yeast, 390 g soy flour, 2850 g yellow cornmeal, 225 g agar, 3 L light corn syrup, 188 ml propionic acid). Earwigs and cherries (cvs Penny and Sweetheart) were collected from NIAB EMR and additional cherries (Sweetheart) from a local cherry producer in Sittingbourne, Kent.

The same experimental set-up used in the field trials was repeated in the insectary, however, SWD were anaesthetised with CO<sub>2</sub> injected into culture tubes to immobilise flies. Treatment densities of flies were either 0 (control), 3 males+3 females or 6 males+6 females in each arena. There were 6 replicates of each treatment. These treatments either had earwigs added or no earwigs were introduced. Hence there were 18 boxes with earwigs and 18 boxes without earwigs. Boxes both with and without earwigs were placed in an incubator set to a June

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cycle (11-21°C temperature cycle, 16h:8h light/dark cycle, peak temperature at 13:00, lowest at 05:00) on 03/10/17 for 7 days. A June cycle was selected as early summer is the optimal season for *F. auricularia* activity, with daytime temperatures between 20-25°C.<sup>32, 38, 39</sup> Upon removal, the same method of sugar extraction for 4 of the 8 cherries was executed as previously outlined. The remaining 4 cherries were placed in controlled chambers in the insectary at 23°C in a 12h:12h light/dark cycle. After 14 days, boxes were removed and frozen for 3 hours to enable counting of SWD adults.

### *2.5 Statistical analyses*

Field and insectary trials were analysed as two separate experiments. To analyse the effects earwigs had on reproductive rate in each SWD treatment density, a two-tailed Mann-Whitney U-test was used to compare the number of larvae produced per parental pair at each SWD treatment density in the presence of an earwig with the number of larvae produced per parental pair at the corresponding treatment density in earwig-free boxes. The same test was used to compare the overall impact of earwigs on larval production in the field or the insectary. SWD reproductive success in the presence versus the absence of earwigs was determined across SWD densities by conducting the Mann-Whitney test on the full set of normalized larval counts. Emerged adult counts per parental SWD pair were analysed in an identical way.

## **3. Results**

The mean number of larvae or subsequently emerged adult SWD was lower in the presence of earwigs for almost all treatments across both the laboratory and field experiments

(e.g. Figure 1). In field trials, a significant reduction in larvae per parental pair was observed when ten SWD pairs were present ( $U = 2, P = 0.009$ ). Moreover, when pooling across treatment densities earwigs were successful in significantly reducing the number of SWD larvae per parental SWD pair ( $U = 111, P < 0.0005$ ), on average reducing by 41.45%. In the presence of earwigs, SWD adult emergence was reduced in treatments of both seven and ten SWD pairs ( $U = 0, P = 0.002$  and  $U = 0, P = 0.002$ , respectively). The treatment of three SWD pairs in the field condition represents the only occasion in which the average number of emerged adults was higher – though not significantly so – in boxes containing earwigs as opposed to those that were earwig-free. Regardless, the effect of earwigs on reducing adult emergence was significant when pooling all treatment levels ( $U = 112.5, P < 0.0005$ ). It must be mentioned that all earwigs were alive and active at the end of each assay, so no treatments were exposed to earwigs for longer than others through earwig mortality.

Figure 1c illustrates that in the insectary, earwig presence not only led to reductions in larval numbers in the higher treatment density of six SWD breeding pairs, but also in the lower treatment density of three pairs ( $U = 4.5, P = 0.026$  and  $U = 3, P = 0.015$ , respectively). This resulted in a highly significant pooled effect ( $U = 17, P = 0.001$ ). Similarly, the effect of earwig presence on natural SWD adult emergence in the laboratory was significant at treatment densities of three and six SWD pairs ( $U = 3, P = 0.015$  and  $U = 1, P = 0.004$ , respectively). Figure 1d shows the extent of the negative effect of earwig presence. The pooled effect of earwigs on natural emergence was again significant ( $U = 10, P < 0.0005$ ). No larvae or emerged adults were found in any of the control boxes containing 0 SWD pairs (Figure 1).

The presence of an earwig resulted in the reduction of larval and adult offspring emergence by an average of 45.65%, in comparison to those without earwigs. To ascertain the level of adult predation by *F. auricularia* and whether a reduction in offspring was the result of a reduction in parent SWD, the percentage of SWD recovered from each box, both alive and dead (i.e. not eaten by earwigs), was recorded (Table 1). If predation on adult SWD was accountable for SWD population decline, a low recovery rate would be expected. However, on average 82.37% of parent SWD were recovered, after incubation with earwigs, even in a confined space after 1 week. This suggests that parent predation was not the sole factor in reducing offspring numbers.

#### 4. Discussion

##### 4.1. *F. auricularia* biocontrol of SWD

Across all trials, the mean percentage of recovered SWD in the presence of earwigs was 82.37%, suggesting limited predation by *F. auricularia* on adult flies within these assays. If high levels of adult predation were occurring, a lower recovery rate would have been expected. Although previous work, using DNA extraction techniques, have revealed that *F. auricularia* are active in predation of SWD, it is likely that they consume more eggs, larvae and pupa than adults.<sup>35</sup> Additionally, high levels of larval predation have been observed in other earwig species and a variety of different prey insects.<sup>28, 29, 40, 41</sup> Limited levels of SWD adult predation contrasted

with significant corresponding reductions in SWD larval and emerged adult densities imply that adult predation accounted for little of the population suppression even though SWD were caged with the earwigs within this assay. Hence, *F. auricularia* may more easily predate immature stages of SWD, which are not capable of flight and so easier to target.

Alternative mechanisms may also have accounted for reductions in SWD population density. It seems plausible that the presence of earwigs in relatively close proximity to SWD adults may have produced sensory cues or be directly disruptive to *Drosophila* courtship and oviposition. Previous work has outlined olfactory avoidance of *Leptopilina* wasps by *D. melanogaster*.<sup>42</sup> Additionally, the presence of butyl atranilate (BA) significantly reduced SWD oviposition in blueberries.<sup>43</sup> Although, further study would be required to establish the presence of olfactory cues between earwigs and SWD which may limit reproductive rate.

#### *4.2 Predation differences in the field and insectary*

The insectary trial was designed to verify whether the field predation results could be reproduced in an artificial 'June' temperature gradient and photocycle in the laboratory. The presence of earwigs suppressed SWD larval and adult emergence both in the field and in the insectary. However, the reductions in SWD reproduction were more readily detectable and significant at lower SWD population densities in the laboratory. These differences in predation could be explained by the increased variability under field conditions or by one or more environmental differences between the field conditions in late July/early August and the simulated 'June' conditions in the laboratory.

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One environmental difference of particular interest is the daily temperature cycle. In August, temperatures during the day were regularly between 30-35°C; far higher than the 21°C daytime temperature of the June conditions in the laboratory. *F. auricularia* are known to be most active in temperatures between 20-24°C, akin to those of early summer, with large fluctuations in temperature and high maximum temperatures discouraging activity.<sup>32, 38, 39</sup> Thus, it seems possible that predation may have been higher in the insectary trials as temperatures were optimal for earwig activity. Additionally, optimum temperatures for oviposition are 19-25°C and *D. sukuzii* is also likely to exhibit behavioural rhythms in locomotor activity and reproductive success that are sensitive to environmental temperature.<sup>44, 45, 46</sup> Therefore, follow-up studies are required to more extensively pursue the possible impact of seasonal environmental changes on earwig predation of SWD.

#### 4.3 Potential for application of *F. auricularia* in orchards

In field trials, *F. auricularia* had no significant effect on SWD reproductive rate at the lower population densities of one or three breeding SWD pairs, which may be suggestive of a limited biocontrol impact in orchards.

Additionally, in the confines of the experimental set-up SWD eggs were laid in a more clumped distribution than would occur naturally. In natural settings, SWD generally lay eggs in a random distribution with one egg per clutch,<sup>47</sup> resulting in a greater reduction in proximity of earwigs to SWD in the field. The resultant increase in energy costs to hunt more sparsely

distributed SWD would likely reduce earwig predation.<sup>48</sup> Alternatively, even though SWD are more likely to be sparsely distributed in a natural setting, nocturnal foraging activity of earwigs could still result in frequent encounters with SWD life stages as evidenced the Wolf et al. (2018) paper.

Ecological conditions in boxes are very different to natural settings. In sealed boxes, earwigs only had two food sources available: SWD and cherries. Yet, *F. auricularia* are known to be highly omnivorous in the wild, eating a variety of plants and other arthropods.<sup>49, 50</sup> Therefore, predation of SWD in natural settings is likely to be lower than witnessed in sealed boxes because SWD will likely contribute only a small part to the varied diet of *F. auricularia*.

The higher predation by earwigs in the insectary, when a cooler temperature regime was applied, suggests that it may be worthwhile exploring the impact of environmental temperature profiles on earwig predation of SWD. It is possible that *F. auricularia* could be a more efficient biocontrol agent of SWD at the start of the growing season before temperatures increase. Coincidentally, at earlier times of the year SWD are often the only fruit fly species in newly occupied regions which are actively laying eggs in ripening fruit.<sup>13</sup>

#### *4.4 Methodology limitations and improvements*

In the present study, cherries had been detached from plants. However, these fruits are no longer economically viable and SWD preferentially oviposit in attached fruit.<sup>51, 52, 53</sup> Therefore, it is recommended that future work focuses on attached fruit.



Similar studies using predators such as that by Gabarra *et al.* (2014)<sup>28</sup> starved predators for 24 hours prior to use in feeding trials to ensure each individual was similarly hungry. This procedural detail was not included as part of the current methodology but could be incorporated in future studies. In addition, we focused on adult earwigs. It is possible that growing larval earwigs are more voracious and could be more effective than their adult counterparts in preying on SWD. Moreover, no work has looked at how the daily activity cycles of earwigs and SWD interact, i.e. earwigs are night active and adult SWD are, generally, day active.

While the convenient sugar extraction technique for larval extraction used here was predictive of infestation and subsequent emergence, it could be further refined to minimise the level of human error when recording larvae. Some eggs were inevitably laid within 72 hours of sugar extraction, and thus may have still been eggs or only first instars at the time of testing, both of which >1mm and undetectable by the human eye in the cherry pulp.<sup>16</sup> To negate this problem, the technique outlined by Van Timmeren *et al* (2017)<sup>54</sup> is recommended.

## 5. Conclusions

*F. auricularia* significantly reduced the density of SWD larval and emerged adult populations. In the field, these reductions were confined to conditions with higher SWD densities, illustrating the limited effectiveness of *F. auricularia* as a biocontrol agent. The mean recovery rate of SWD from boxes that contained earwigs was 82.37%, suggesting low adult predation by *F. auricularia*. Conditions in orchards are very different to the design used in this study where both pest and predator were housed in the same box: SWD are in lower densities

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in less clumped distributions, and they contribute only a small part to the varied diet of *F. auricularia*. For these reasons it is concluded that the potential of *F. auricularia* as a biocontrol agent of SWD has its limits and that further study on the interaction between earwigs and SWD across additional environmental conditions is warranted to more fully establish the potential use of *F. auricularia* in SWD integrated pest management.

## 6. References

1. Asplen M, Anfora G, Biondi A, Choi D, Chu D, Daane K et al. Invasion biology of spotted wing *Drosophila* (*Drosophila suzukii*): a global perspective and future priorities. *Journal of Pest Science*. 2015;88(3):469-494.
2. Cini A, Anfora G, Escudero-Colomar L, Grassi A, Santosuosso U, Seljak G et al. Tracking the invasion of the alien fruit pest *Drosophila suzukii* in Europe. *Journal of Pest Science*. 2014;87(4):559-566.
3. Cini A, Ioriatti C, Anfora G. A review of the invasion of *Drosophila suzukii* in Europe and a draft research agenda for integrated pest management. *Bulletin of Insectology*. 2012;65(1):149-160.
4. Langille A, Arteca E, Ryan G, Emiljanowicz L, Newman J. North American invasion of Spotted-Wing *Drosophila* (*Drosophila suzukii*): A mechanistic model of population dynamics. *Ecological Modelling*. 2016;336:70-81.

5. Lee J, Bruck D, Dreves A, Ioriatti C, Vogt H, Baufeld P. In Focus: Spotted wing drosophila, *Drosophila suzukii*, across perspectives. *Pest Management Science*. 2011;67(11):1349-1351.
6. Kanzawa T. Studies on *Drosophila suzukii*. *Review of Applied Entomology*. 1939;29:662.
7. Tochen S, Dalton D, Wiman N, Hamm C, Shearer P, Walton V. Temperature-Related Development and Population Parameters for *Drosophila suzukii* (Diptera: Drosophilidae) on Cherry and Blueberry. *Environmental Entomology*. 2014;43(2):501-510.
8. Toda M. Vertical Microdistribution of Drosophilidae (Diptera) within various Forests in Hokkaido. : I. Natural Broad-Leaved Forest. *Japanese Journal of Ecology*. 1977;27(3).
9. Hauser M, Gaimari S, Damus M. *Drosophila suzukii* new to North America. *Fly Times*. 2009;43:12-15.
10. Kaneshiro K. *Drosophila* (Sophophora) *suzukii* (Matsumura). *Proceedings of the Hawaiian Entomology Society*. 1983;24(179).
11. Walsh D, Bolda M, Goodhue R, Dreves A, Lee J, Bruck D et al. *Drosophila suzukii* (Diptera: Drosophilidae): Invasive Pest of Ripening Soft Fruit Expanding its Geographic Range and Damage Potential. *Journal of Integrated Pest Management*. 2011;2(1):G1-G7.
12. Tonina L, Grassi A, Caruso S, Mori N, Gottardello A, Anfora G et al. Comparison of attractants for monitoring *Drosophila suzukii* in sweet cherry orchards in Italy. *Journal of Applied Entomology*. 2017;142(1-2):18-25.

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13. Atallah J, Teixeira L, Salazar R, Zaragoza G, Kopp A. The making of a pest: the evolution of a fruit-penetrating ovipositor in *Drosophila suzukii* and related species. *Proceedings of the Royal Society B: Biological Sciences*. 2014;281(1781):20132840-20132840.
  14. Mazzi D, Bravin E, Meraner M, Finger R, Kuske S. Economic Impact of the Introduction and Establishment of *Drosophila suzukii* on Sweet Cherry Production in Switzerland. *Insects*. 2017;8(1):18.
  15. Beers E, Van Steenwyk R, Shearer P, Coates W, Grant J. Developing *Drosophila suzukii* management programs for sweet cherry in the western United States. *Pest Management Science*. 2011;67(11):1386-1395.
  16. Kinjo H, Kunimi Y, Nakai M. Effects of temperature on the reproduction and development of *Drosophila suzukii* (Diptera: Drosophilidae). *Applied Entomology and Zoology*. 2014;49(2):297-304.
  17. Van Timmeren S, Mota-Sanchez D, Wise J, Isaacs R. Baseline susceptibility of spotted wing *Drosophila* (*Drosophila suzukii*) to four key insecticide classes. *Pest Management Science*. 2017;74(1):78-87.
  18. Pittendrigh B, Reenan R, French-Constant R, Ganetzky B. Point mutations in the *Drosophila* sodium channel gene *para* associated with resistance to DDT and pyrethroid insecticides. *Molecular and General Genetics MGG*. 1997;256(6):602-610.

- Accepted Article
19. Perry T, McKenzie J, Batterham P. A  $\Delta\alpha6\Delta\alpha6$  knockout strain of *Drosophila melanogaster* confers a high level of resistance to spinosad. *Insect Biochemistry and Molecular Biology*. 2007;37(2):184-188.
  20. Feyereisen R. Molecular biology of insecticide resistance. *Toxicology Letters*. 1995;82-83:83-90.
  21. Smirle M, Zurowski C, Ayyanath M, Scott I, MacKenzie K. Laboratory studies of insecticide efficacy and resistance in *Drosophila suzukii* (Matsumura) (Diptera: Drosophilidae) populations from British Columbia, Canada. *Pest Management Science*. 2016;73(1):130-137.
  22. Nomano F, Mitsui H, Kimura M. Capacity of Japanese *Asobara* species (Hymenoptera; Braconidae) to parasitize a fruit pest *Drosophila suzukii* (Diptera; Drosophilidae). *Journal of Applied Entomology*. 2014;139(1-2):105-113.
  23. Quicke D. *Parasitic Wasps*. London: Chapman & Hall; 1997.
  24. Chabert S, Allemand R, Poyet M, Eslin P, Gibert P. Ability of European parasitoids (Hymenoptera) to control a new invasive Asiatic pest, *Drosophila suzukii*. *Biological Control*. 2012;63(1):40-47.
  25. Fleury F, Ris N, Allemand R, Fouillet P, Carton Y, Boulétreau M. Ecological and Genetic Interactions in *Drosophila*–parasitoids Communities: A Case Study with *D. Melanogaster*, *D. Simulans* and their Common *Leptopilina* Parasitoids in South-eastern France. *Genetica*. 2004;120(1-3):181-194.

26. Kacsoh B, Schlenke T. High Hemocyte Load Is Associated with Increased Resistance against Parasitoids in *Drosophila suzukii*, a Relative of *D. melanogaster*. PLoS ONE. 2012;7(4):e34721.
27. Strand M, Pech L. Immunological Basis for Compatibility in Parasitoid-Host Relationships. Annual Review of Entomology. 1995;40(1):31-56.
28. Gabarra R, Riudavets J, Rodríguez G, Pujade-Villar J, Arnó J. Prospects for the biological control of *Drosophila suzukii*. BioControl. 2014;60(3):331-339.
29. Ballman E, Collins J, Drummond F. Pupation Behavior and Predation on *Drosophila suzukii* (Diptera: Drosophilidae) Pupae in Maine Wild Blueberry Fields. Journal of Economic Entomology. 2017;110(6):2308-2317.
30. Cuthbertson A, Blackburn L, Audsley N. Efficacy of Commercially Available Invertebrate Predators against *Drosophila suzukii*. Insects. 2014;5(4):952-960.
31. Woltz J, Donahue K, Bruck D, Lee J. Efficacy of commercially available predators, nematodes and fungal entomopathogens for augmentative control of *Drosophila suzukii*. Journal of Applied Entomology. 2015;139(10):759-770.
32. Crumb S, Eide P, Bonn A. The European Earwig. Technical Bulletin United States Department of Agriculture Washington. 1941;766:1-76.
33. Mueller T, Blommers L, Mols P. Earwig (*Forficula auricularia*) predation on the woolly apple aphid, *Eriosoma lanigerum*. Entomologia Experimentalis et Applicata. 1988;47(2):145-152.

- Accepted Article
34. Piñol J, Espadaler X, Cañellas N, Pérez N. Effects of the concurrent exclusion of ants and earwigs on aphid abundance in an organic citrus grove. *BioControl*. 2008;54(4):515-527.
  35. Wolf S, Zeisler C, Sint D, Romeis J, Traugott M, Collatz J. A simple and cost-effective molecular method to track predation on *Drosophila suzukii* in the field. *Journal of Pest Science*. 2018;91(2):927-935.
  36. Hilton R, VanBuskirk P, Westigard P. Control of Secondary Pests in a Selective Pest Management Program. *Acta Horticulturae*. 1998;(475):479-486.
  37. Saladini M, Asteggiano L, Pansa M, Giordani L, Serre L, Vittone G et al. Glue barriers reduce earwig damage on apricots in north-western Italy. *International Journal of Pest Management*. 2016;62(3):214-221.
  38. Capinera J. European Earwig *Forficula auricularia* Linnaeus (Insecta: Dermaptera: Forficulidae). Department of Entomology and Nematology, UF/IFAS Extension. 2016;.
  39. Burnip G, Daly J, Hackett J, Suckling D. European Earwig Phenology and Effect of Understorey Management on Population Estimation. *New Zealand Plant Protection*. 2002;55:390-395.
  40. Renkema J, Telfer Z, Garipey T, Hallett R. *Dalotia coriaria* as a predator of *Drosophila suzukii*: Functional responses, reduced fruit infestation and molecular diagnostics. *Biological Control*. 2015;89:1-10.
  41. Woltz J, Lee J. Pupation behavior and larval and pupal biocontrol of *Drosophila suzukii* in the field. *Biological Control*. 2017;110:62-69.

- Accepted Article
42. Ebrahim S, Dweck H, Stökl J, Hofferberth J, Trona F, Weniger K et al. *Drosophila* Avoids Parasitoids by Sensing Their Semiochemicals via a Dedicated Olfactory Circuit. *PLOS Biology*. 2015;13(12):e1002318.
  43. Krause Pham C, Ray A. Conservation of Olfactory Avoidance in *Drosophila* Species and Identification of Repellents for *Drosophila suzukii*. *Scientific Reports*. 2015;5(1).
  44. Zerulla F, Augel C, Zebitz C. Oviposition activity of *Drosophila suzukii* as mediated by ambient and fruit temperature. *PLOS ONE*. 2017;12(11):e0187682.
  45. Ryan G, Emiljanowicz L, Wilkinson F, Kornya M, Newman J. Thermal Tolerances of the Spotted-Wing *Drosophila* *Drosophila suzukii* (Diptera: Drosophilidae). *Journal of Economic Entomology*. 2016;109(2):746-752.
  46. Shaw B, Fountain M, Wijnen H. Recording and reproducing the diurnal oviposition rhythms of wild populations of the soft- and stone- fruit pest *Drosophila suzukii*. *PLOS ONE*. 2018;13(10):e0199406.
  47. Mitsui H, Takahashi K, Kimura M. Spatial distributions and clutch sizes of *Drosophila* species ovipositing on cherry fruits of different stages. *Population Ecology*. 2006;48(3):233-237.
  48. Pyke G, Pulliam H, Charnov E. Optimal Foraging: A Selective Review of Theory and Tests. *The Quarterly Review of Biology*. 1977;52(2):137-154.
  49. Capinera J. *Handbook of vegetable pests*. San Diego, California: Academic Press; 2001.



50. Behura B. Life-history of the European Earwig, *Forficula auricularia*. *Nature*. 1950;166(4210):74-74.
51. Stemberger T. Survey of hanging and fallen cherry fruit use by spotted wing drosophila, *Drosophila suzukii* (Matsumura, 1931) (Diptera: Drosophilidae), and other Drosophilidae species. *Pan-Pacific Entomologist*. 2015;91(4):347-31.
52. Bal H, Adams C, Grieshop M. Evaluation of Off-season Potential Breeding Sources for Spotted Wing Drosophila (*Drosophila suzukii* Matsumura) in Michigan. *Journal of Economic Entomology*. 2017;110(6):2466-2470.
53. Matsuura A, Mitsui H, Kimura M. A preliminary study on distributions and oviposition sites of *Drosophila suzukii* (Diptera: Drosophilidae) and its parasitoids on wild cherry tree in Tokyo, central Japan. *Applied Entomology and Zoology*. 2017;53(1):47-53.
54. Van Timmeren S, Diepenbrock L, Bertone M, Burrack H, Isaacs R. A Filter Method for Improved Monitoring of *Drosophila suzukii* (Diptera: Drosophilidae) Larvae in Fruit. *Journal of Integrated Pest Management*. 2017;8(1).

## 7. Tables

Table 1. Results from the Mann-Whitney U analysis describing the impact of earwig presence on the number of offspring per parental SWD pair for each treatment density as well as the pooled analysis across all treatment densities. The % difference illustrates the percentage reduction in the normalised number of larvae or emerged adults for trials with earwigs compared

to those without. The mean percentage of recovered flies, both alive and dead, at the end of the earwig trials are also given below.

	<b>Mann-Whitney U result</b>	<b>% Difference (2.dp.)</b>	<b>Recovered Flies (%)</b>
<b>Field Larval Density</b>			
<b>0 (control)</b>	-	-	-
<b>1</b>	U = 6, P = n.s	-62.50	91.67
<b>3</b>	U = 11, P = n.s	-22.22	83.33
<b>7</b>	U = 9, P = n.s	-30.30	76.19
<b>10</b>	U = 2, P = 0.009	-50.79	76.33
<b>Across treatments</b>	U = 111, P < 0.0005	-41.45	82.38
<b>Field Adult Density</b>			
<b>0 (control)</b>	-	-	-
<b>1</b>	U = 7.5, P = n.s	-62.50	91.67
<b>3</b>	U = 20,	+8.89	83.33

		P = n.s		
	7	U = 0,	-80.10	76.19
		P = 0.002		
	10	U = 0,	-76.54	76.33
		P = 0.002		
	<b>Across treatments</b>	U = 112.5,	-52.56	82.38
		P < 0.0005		
<hr/>				
	<b>Insectary larval</b>			
	<b>density</b>			
<hr/>				
	<b>0 (control)</b>	-	-	-
	3	U = 3,	-51.85	77.78
		P = 0.015		
	6	U = 4.5,	-28.95	88.89
		P = 0.026		
	<b>Across treatments</b>	U = 17,	-40.40	83.33
		P = 0.001		
<hr/>				
	<b>Insectary adult</b>			
	<b>density</b>			
<hr/>				
	<b>0 (control)</b>	-	-	-
	3	U = 3,	-40.88	77.78
		P = 0.015		
	6	U = 1,	-50.00	88.89

	P = 0.004		
Across treatments	U = 10,	-45.44	83.33
	P < 0.0005		

## 8. Figure Legends

Figure 1. Mean ( $\pm$  SEM) of (a) field larval counts from floatation extraction, (b) field natural adult emergence counts, (c) insectary larval counts from floatation extraction, (d) insectary natural adult emergence counts for each treatment density of SWD. For each treatment density both counts with and without earwigs have been depicted together to illustrate the effect of the presence of earwigs on SWD reproductive rate. C = control (no SWD).

