

Article

Field-Based Spatiotemporal Dynamics, Ovarian Maturation and Laboratory Oviposition Behavior of *Drosophila suzukii* in Peach: Key Insights for Integrated Pest Management

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Abstract

Drosophila suzukii is a key invasive pest, and infestation in peach orchards can lead to significant economic losses. This study monitored the spatial distribution and reproductive biology of *D. suzukii* in central Italy to inform integrated pest management (IPM) strategies. In the surveyed orchard, the pest exhibited multiple generations, with captures highest along mixed-species-orchard edges, highlighting these margins as potential hotspots for targeted mass trapping. Seasonal dissections of females revealed delayed ovarian development during winter, while maturation progressed during fruit ripening and post-harvest periods. This result provides relevant information on the likely timing of oviposition, useful for informing pest management. A laboratory oviposition trial on nectarines revealed a clear preference for healthy, mechanically damaged fruits, whereas fungal infection reduced the attractiveness. This suggests that field sanitation, especially the timely removal of damaged or fallen fruits, could reduce pest presence and inoculum for the following season. Overall, these findings offer practical insights to support sustainable IPM approaches against *D. suzukii* in peach production systems.

Keywords: Spotted Wing *Drosophila*; spatial distribution; sex ratio; ovarian maturation; oviposition



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1. Introduction

From 2008, *Drosophila suzukii* Matsumura, 1931 (Diptera: Drosophilidae), commonly known as Spotted Wing *Drosophila* (SWD), has rapidly spread throughout Europe and North America [1,2]. More recently, it has established in South America [2,3], and its distribution is expected to expand into previously unsuitable areas under climate change [4,5]. This expansion is facilitated by the species' adaptation to new cultivated and wild hosts [6] and by its remarkable phenotypic plasticity, which allows survival even in colder climates [7]. In recent years, this invasive species has caused substantial economic losses to fruit production, including direct yield losses and increased production costs associated with specific control strategies [2,8]. The damage is mainly linked to its high fecundity and ability to oviposit in healthy fruit, with damages resulting from larval feeding and oviposition punctures also facilitate fungal infections and secondary pest attacks [9]. Damaged

fruits are unmarketable, and chemical control strategies must comply with strict residue limits for export markets [6,10], complicating management under integrated and organic production systems. As a result, a rapidly growing body of literature has focused on the management of SWD [11–15].

Drosophila suzukii is an extremely polyphagous species, infesting a wide range of cultivated, ornamental, and wild host plants [16–18]. Fruit firmness is often negatively correlated with oviposition and larval development [19–23], but host suitability varies considerably among species and cultivars. Peaches (*Prunus persica* (L.) Batsch) [Rosaceae] have generally been considered a less suitable host compared to other soft fruits [24–26]. Nevertheless, certain conditions, such as the presence of damaged fruits, can increase the attractiveness of peaches to SWD females [3], and the risks posed by *D. suzukii* to peach production may be underestimated [6]. The issue may be particularly relevant in organic peach orchards, where chemical interventions are limited. Diversified hedgerows may provide valuable support for pest management by promoting biodiversity and facilitating the control of harmful arthropods [27]. However, the use of mixed hedgerows has been reported as contrasting in pests control [28], and their influence on *D. suzukii* has never been investigated in peach orchards. In these agroecosystems, the presence of preferred hosts within the hedgerows might contribute to a higher pest occurrence in the field margins.

Moreover, a detailed understanding of *D. suzukii* reproductive biology is essential for targeted pest management. Ovarian maturation has been studied in Northwestern Europe and Northern Italy [29–32] but no studies are available for Central Italy. Information from multiple localities is essential for the development of area-specific IPM strategies, as oogenesis is highly temperature-dependent and tends to increase with higher average monthly temperatures [30]. Under Mediterranean climatic conditions, the period suitable for adult mating and oviposition may be prolonged compared to cooler climates, potentially resulting in greater population build-up and increased pest pressure. Consequently, local population dynamics must be considered to optimize management strategies.

Our study addresses the current gaps regarding the management of Spotted Wing *Drosophila* on organic peach orchards of central Italy, with the aim to provide valuable biological information for the development of effective and sustainable Integrated Pest Management (IPM). We conducted field monitoring of *D. suzukii* in an organic peach orchard in the Marche region (central Italy). The objectives of this work were to: (i) investigate the spatial distribution and abundance of *D. suzukii* relative to mixed-species-orchard edges, (ii) describe seasonal patterns of female ovarian development to estimate periods of highest oviposition risk, and (iii) evaluate oviposition preferences under controlled laboratory conditions to inform field sanitation practices.

2. Materials and Methods

2.1. Site Description

The monitoring and collection of adult insects was conducted in an organic peach orchard located in the Marche region, central Italy (Montedinove, 42°59′13.26″ N; 13°33′16.94″ E, 211 m a.s.l.). Rows were oriented north–south and no pesticide applications were applied throughout the experiments. In the northern edge, the orchard was bordered by a mixed hedgerow composed of *Rubus ulmifolius* Schott. [Rosaceae], *Sambucus nigra* L. [Adoxaceae], *Robinia pseudoacacia* L. [Fabaceae], and *Hedera helix* L. [Araliaceae], whereas the southern border was characterized by a cane thicket and scattered *S. nigra* plants. Due to terrain topography, the southern edge appears wetter than the northern edge.

The site included four consecutive central rows corresponding to a 20 × 140 m plot.

2.2. Ethics Statement

All procedures involving insects were performed in accordance with Italian and European legislation on experimental animals. According to Italian Legislative Decree 26/2014, which implements EU Directive 2010/63/EU, invertebrates other than cephalopods are not considered laboratory animals for regulatory purposes. Therefore, no specific authorization was required for the collection, rearing, and experimental use of *Drosophila suzukii* in this study.

2.3. *Drosophila suzukii* Monitoring

Monitoring was performed biweekly during the 2020–2021 season using red chromotropic traps (Droso Trap[®]—Biobest, Westerlo, Belgium). Each trap consisted of a plastic container with 21 lateral holes (8 mm diameter) and a lid with a hook for hanging. A total of 18 traps were deployed along a north–south transect. Two traps were placed every 20 m along the transect at 1.5 m above the ground. For analytical purposes, trap positions were expressed relative to each edge: 0, 20, 40, 60, and 80 m for the northern edge, and 0, 20, 40, and 60 m for the southern edge.

Traps were baited with 250 mL of a solution composed by 75% apple vinegar, 25% red wine, and 20 g/L of cane sugar [33,34]. The solution was replaced at each sampling event. Captured *Drosophila* spp. were preserved in 70% ethanol and stored at 4 °C until *D. suzukii* identification [35] and counting.

2.4. Assessment of Reproductive Status

For each sampling date, up to 100 *D. suzukii* females were collected and dissected under a stereomicroscope to determine their reproductive status. When more than 100 females were captured, subsampling per trap was homogenized using the following formula:

$$X = \frac{n_{trap}}{n_{date}} \times 100$$

where

- X: number of females dissected per trap;
- n_{trap} : number of females captured in the given trap;
- n_{date} : total number of females captured across all traps on that date.

Females were classified into five ovarian maturation stages following Grassi et al. (2017) [30]:

1. No ovaries;
2. Immature ovaries;
3. Ripening eggs in ovarioles;
4. Mature eggs in ovarioles;
5. Mature eggs outside the ovaries (abdominal).

2.5. Laboratory Evaluation of Oviposition Preferences

Oviposition assays were conducted under controlled laboratory conditions (25 °C, 70% RH, 16:8 L:D) at Marche Polytechnic University (Ancona, Italy). Groups of four mated females (4–5 days old) from laboratory colony were placed in ventilated 500 mL glass bakerys with hydrophilic cotton and a nectarine fruit. Four treatments were tested, with 30 replicates per treatment:

- T1: Intact, healthy fruit;
- T2: Cut, healthy fruit (2 × 1 cm superficial incision);
- T3: Intact, inoculated fruit (*Monilinia fructicola* isolated from the University collection);
- T4: Cut, inoculated fruit.

After 24 h, fruits were removed and dissected to determine the number of eggs per fruit, which were counted under a stereomicroscope (Leica Microsystems Model EZ4W, Heerbrugg, Swiss).

2.6. Statistical Analysis

Statistical analyses were conducted using R (version 4.3.1; R Core Team, 2025). Generalized linear models (GLMs) and generalized linear mixed models (GLMMs) were fitted with the glmmTMB package.

Count data (adult or eggs) were modeled using negative binomial distribution to account for overdispersion. Model assumptions were checked using the DHARMA package based on simulated residuals ($n = 250$), assessing dispersion, uniformity, and outliers via graphical inspection and formal tests. Significance of fixed effects, including interactions, was assessed using Type II Wald χ^2 test. Estimated marginal means (EMMs) were computed using the emmeans package and are reported on the response scale. Post hoc pairwise comparisons were adjusted using Tukey's method.

To investigate seasonal trends in total adult captures, we modeled trap-level counts using period as fixed effect and trap ID as random effect.

To analyze the seasonal variation in the sex-specific abundance, we modeled male and female counts with a GLMM including the interaction Sex $\times$ Period (15th Feb–14th May, 15th May–14th Aug, 15th Aug–14th Nov, 15th Nov–14th Feb) with trap ID as random effect.

To assess the influence of edge proximity and edge position (north vs. south edge) on adult abundance, we modeled trap counts as a function of sex, distance from field edge, and their interaction, using edge position as an additional fixed effect and trap as a random intercept.

To evaluate temporal differences in female reproductive status, we modeled the ovary maturation state, period and their interaction as fixed effects, with date accounted for repeated sampling.

GLMs were used to assess whether oviposition was influenced by treatment (T1, T2, T3, T4), by fruit incision (cut vs. intact) and by *Monilia* infection (*Monilia*: healthy vs. inoculated). Each fruit was treated as an independent observational unit.

Significance threshold was set at $p = 0.05$.

3. Results

3.1. Seasonal Dynamics of *Drosophila suzukii*

During the sampling period, a total of 5870 *D. suzukii* individuals were captured. Trap captures occurred year-round, with a major seasonal peak between mid-May and mid-August (mean ≈ 64 individuals per trap), corresponding to the peach ripening period. The lowest catches were recorded in mid-August–mid-November. Analyses confirmed a significant effect of season on adult abundance (Type II Wald χ^2 test = 77.57, $df = 3$, $p < 0.0001$). Pairwise comparisons (Tukey-adjusted) revealed that all periods were significantly different ($p < 0.01$), except mid-February 2020–mid-May 2020 vs. mid-November 2020–mid-February 2021 (Figure 1).

Analyses on the sex-specific abundance showed a significant effect of period (Type II Wald χ^2 test = 139.49, $df = 3$, $p < 0.001$) and a significant interaction between sex and period (Type II Wald χ^2 test = 28.35, $df = 3$, $p < 0.001$). When averaged across periods, females were nearly twice as abundant as males (ratio = 1.89, $p < 0.0001$). However, pairwise comparisons within periods showed that this difference was only significant from mid-February to mid-May, when female abundance was about seven times higher than male abundance (ratio = 7.18, $p < 0.0001$). In all other periods (mid-Aug–mid-Nov, mid-May–mid-Aug, and mid-Nov–mid-Feb), no significant differences between sexes were detected (Figure 2).

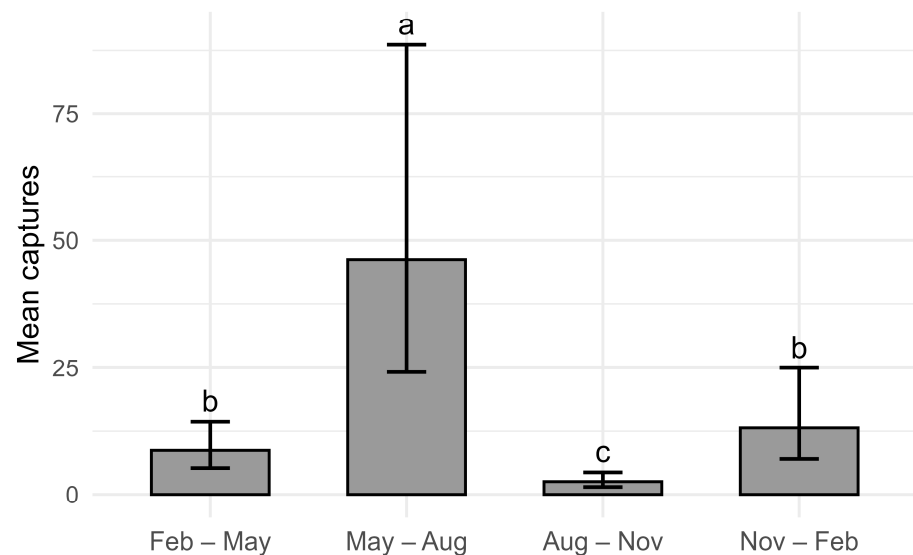


Figure 1. Seasonal variation in *Drosophila suzukii* adults captured in an organic peach orchard in Montedinove (Marche region, Italy) from mid-February 2020 to mid-February 2021. Bars represent predicted marginal means ($\pm 95\%$ confidence intervals). Letters above bars indicate statistically significant differences between periods (Tukey-adjusted comparisons, $p < 0.05$).

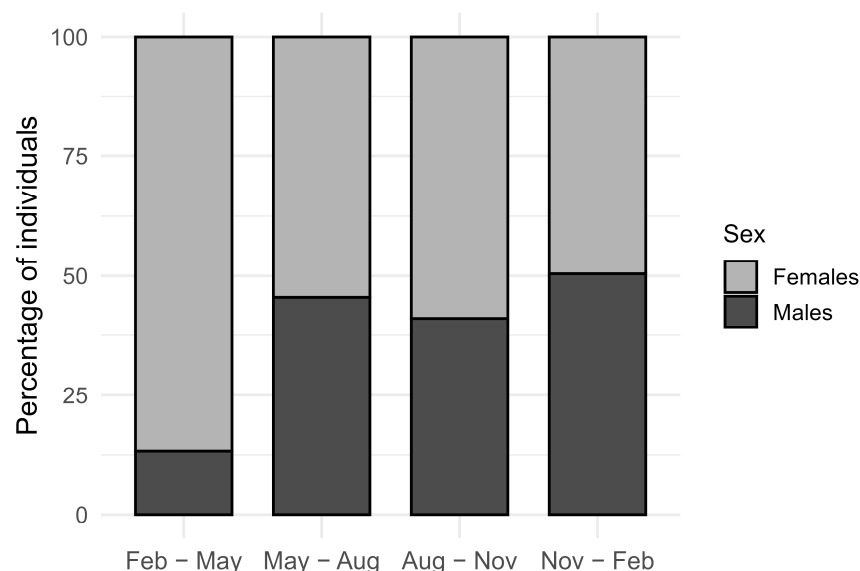


Figure 2. Seasonal variation in the sex ratio of *Drosophila suzukii* adults captured in an organic peach orchard from mid-February 2020 to mid-February 2021. Data represent mean percentage of males and females across four seasonal periods (mid-February–mid-May, mid-May–mid-August, mid-August–mid-November, mid-November–mid-February).

Model diagnostics indicated no evidence of overdispersion, uniform residuals, and only a minimal number of non-influential outliers.

3.2. Spatial Distribution Within the Orchard

Results showed a significant effect of distance from the field edge (Type II Wald χ^2 test = 95.59, $df = 4$, $p < 0.001$) and edge position (Type II Wald χ^2 test = 4.46, $df = 1$, $p < 0.05$) on *D. suzukii* abundance. The interaction of sex and distance was not significant, indicating that males and females exhibited similar spatial distribution patterns. A significantly higher number of individuals was captured along the southern edge compared with the northern edge ($p < 0.05$). Pairwise comparisons showed that edge traps captured significantly more

flies than traps placed at the crop interior ($p < 0.0001$), whereas differences among non-edge traps were not significant (Figure 3).

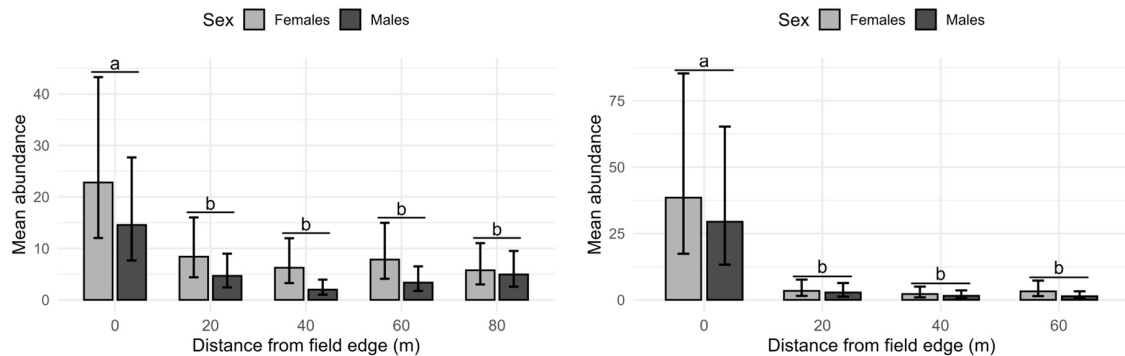


Figure 3. Spatial distribution of *Drosophila suzukii* adults captured in an organic peach orchard as a function of distance from the field edge and trap position. The northern edge is showed on the left, and the southern edge on the right. Bars represent predicted marginal means ($\pm 95\%$ confidence intervals). For each graph, letters above bars indicate statistically significant differences among different edge distances (Tukey-adjusted comparisons, $p < 0.001$).

3.3. *Drosophila suzukii* Reproductive Stages in the Field

A total of 1091 *D. suzukii* females were dissected and classified according to ovarian maturation stage. Sexually mature females were present throughout the year, but during the colder months (mid-November–mid-February) the majority of captured females belonged to classes 1–2, indicating a predominance of immature individuals. The highest proportion of females carrying eggs was recorded between mid-February and mid-May, whereas females with mature eggs outside the ovarioles peaked between mid-August and mid-November (Figure 4). Results showed no statistically significant differences in the number of individuals in each maturation state across sampling periods ($p > 0.05$).

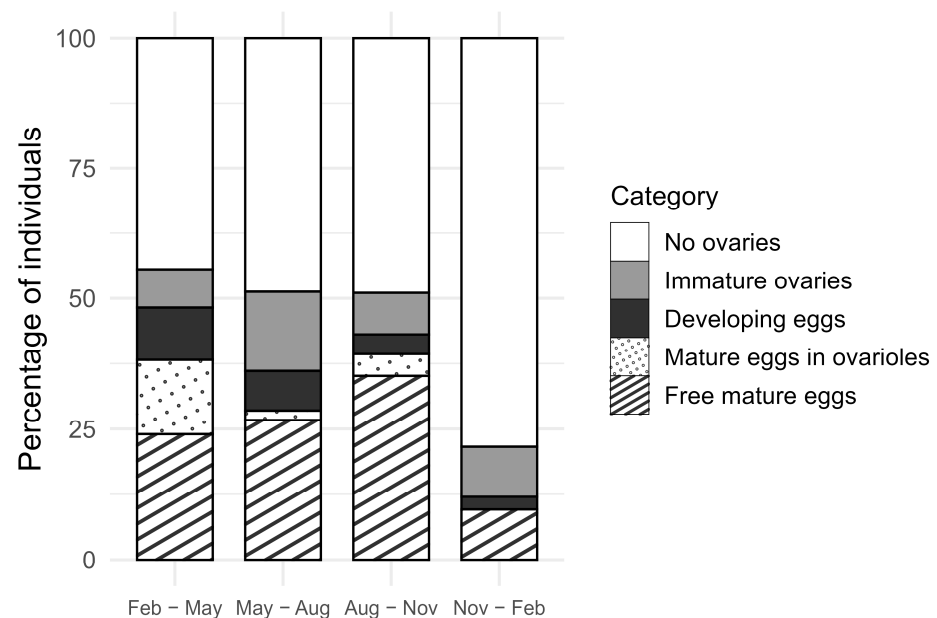


Figure 4. Seasonal distribution of *Drosophila suzukii* females across the five ovarian maturation classes (Class 1: no ovaries; Class 2: immature ovaries; Class 3: ripening eggs; Class 4: mature eggs in ovarioles; Class 5: mature eggs outside ovarioles) from mid-May 2020 to February 2021. Data are expressed as percentages relative to the total number of dissected females for each sampling period.

3.4. *Drosophila suzukii* Reproductive Performance

The total number of eggs laid by *D. suzukii* differed significantly among treatments (Type II Wald χ^2 test = 14.76, df = 3, $p < 0.005$). Pairwise comparisons indicated that T2 (cut, healthy peach) had a significantly higher mean number of eggs compared with T1 (intact, healthy peach, $p < 0.05$) and T3 (intact, inoculated peach, $p < 0.005$). No significant differences were observed between T4 (cut, inoculated peach) and the other treatments ($p > 0.05$) (Figure 5).

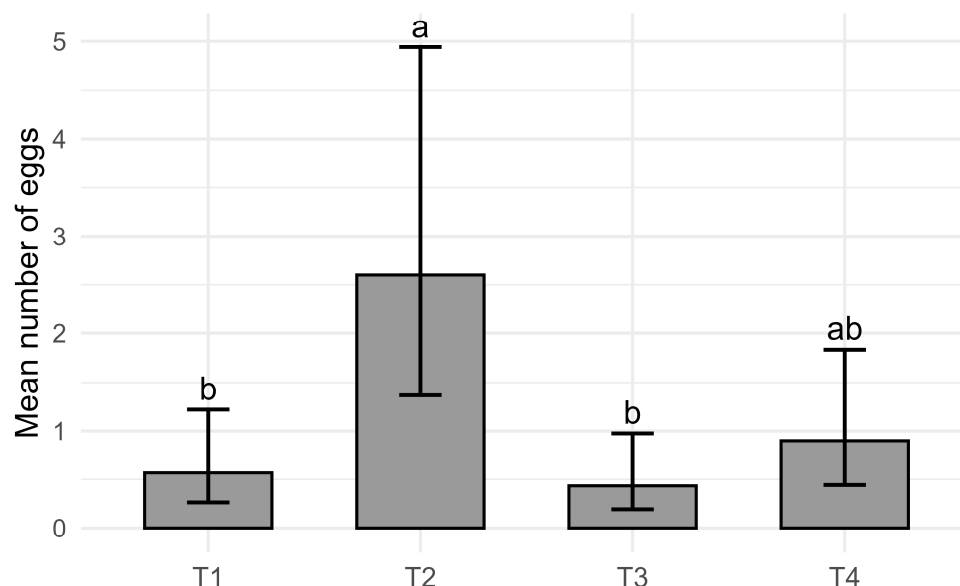


Figure 5. Mean number of eggs recorded on peaches under four treatments in laboratory assays: T1—intact, healthy; T2—cut, healthy; T3—intact, inoculated with *Monilinia fructicola*; and T4—cut, inoculated. Bars represent predicted marginal means ($\pm 95\%$ confidence intervals). Letters above bars indicate statistically significant differences between periods (Tukey-adjusted comparisons, $p < 0.05$).

When factors were analyzed independently, both fruit incision and inoculation status had significant effects on oviposition (Figure 6). Cut peaches received significantly more eggs than intact peaches (Type II Wald χ^2 test = 10.55, df = 1, $p < 0.005$), with predicted means of 1.75 and 0.5 eggs, respectively. Conversely, inoculated fruits received fewer eggs than healthy ones ($\chi^2 = 4.88$, df = 1, $p < 0.05$), with predicted mean of 0.67 and 1.58 eggs, respectively.

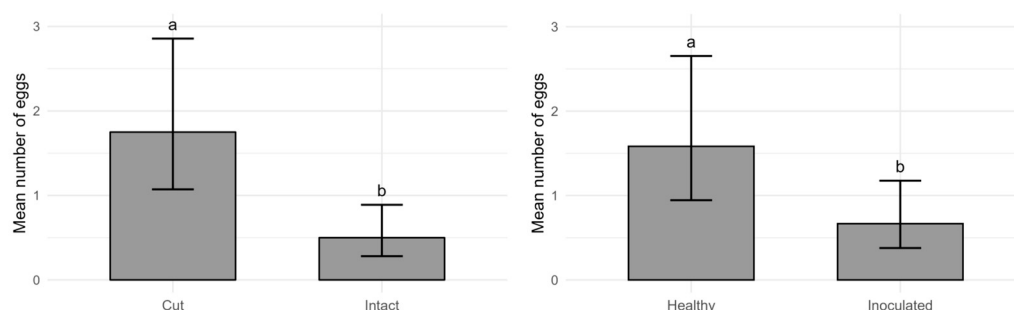


Figure 6. Mean number of eggs according to (left) fruit incision (cut vs. intact) and (right) *Monilia* infection status (inoculated vs. healthy). Bars represent predicted marginal means ($\pm 95\%$ confidence intervals). Letters above bars indicate statistically significant differences between periods (Tukey-adjusted comparisons, $p < 0.05$).

4. Discussion

During the monitoring activities, a relatively high abundance of *D. suzukii* was recorded, indicating a potential concern for the management of this pest in organic peach orchards.

Our data showed low captures of SWD in spring (mid-February to mid-May), followed by a peak coinciding with the presence of ripe peaches between mid-May and mid-August, highlighting this period as the highest-risk window for fruit damage. Mature peaches are likely more attractive because their softer texture facilitates oviposition and provides a suitable environment for larval development. This interpretation is consistent with other studies reporting a negative correlation between fruit firmness and SWD oviposition success [19–22]. The high adult population presence observed during fruit ripening identifies this as a critical period for intensive field monitoring.

Sex ratio analysis revealed a relatively stable male-to-female proportion throughout the year, with a significantly lower abundance of males observed from mid-February to mid-May. A similar pattern during spring months was observed by Rossi-Stacconi et al. (2016) [29]. However, while their study consistently reported a higher abundance of females during winter, our data showed a more balanced number of males and females from mid-November to mid-February. Further research might be useful to evaluate the attractiveness of the bait to males and females throughout the year [29].

Significantly higher captures were observed along orchard edges, particularly the southern margin, suggesting that mixed hedgerows may act as reservoirs for SWD populations. This pattern may be partly explained by the presence of palatable host plants, such as *S. nigra* [18], and potentially more favorable microclimatic conditions, such as higher humidity linked to terrain topography of the southern margin [36,37]. This result agrees with other studies indicating that *D. suzukii* is attracted to wild vegetation near crop fields [28]. However, the literature on field edge diversification shows variable effects on pest populations, with some studies reporting increased abundance and others showing neutral or suppressive effects [38–41].

From an IPM perspective, these findings support the use of orchards borders as strategic zones for intensive monitoring and localized control measures such as mass trapping, attract-and-kill, or push-and-pull tactics. Nevertheless, because they can also serve as overwintering refuges [17,42], hedgerows should be managed carefully to balance biodiversity benefits with the risk of sustaining pest populations.

Ovarian maturation states did not differ significantly across the different sampling periods. Considering the period mid-November–mid-February, the predominance of females with immature ovaries in our samples suggests that control actions during winter (e.g., mass trapping) could help suppress the founder population before fruits become available, potentially reducing infestation pressure in the following season [29].

The timing of ovarian maturation has been investigated by several studies [29–32], who reported that oogenesis may increase with rising mean temperatures, leading to a seasonal synchronization of oviposition capacity. Our results seem consistent with these findings and provide additional insights for the development of IPM programs in central Italy.

For instance, in our study, females without visible ovaries represent the majority of the population during the coldest months (up to 81% on December 30), confirming that ovarian development is largely arrested in winter. Conversely, females carrying free eggs became progressively more frequent from late winter through early summer, reaching their highest proportion between mid-August and mid-November. This pattern indicates that the majority of females are physiologically ready to oviposit when ripe fruit becomes available, representing the most vulnerable stage for crop damage.

These observations are also relevant to understand the overwintering biology of the pest: the predominance of females with immature ovaries in winter suggests a form

of reproductive diapause, as previously described by Rossi Stacconi et al. (2016) [29] and Grassi et al. (2017) [30]. This strategy allows females to resume oogenesis when environmental conditions become favorable, ensuring early season infestations [31]. Our data also align with studies reporting high male survival under cold climatic conditions [43], even though the literature shows conflicting results regarding sex-specific overwintering success [37].

From an IPM perspective, these findings highlight the importance of early-season monitoring and control efforts, particularly in winter, before mated females are ready to initiate the next generation [29,31]. Implementing population suppression tools, such as mass trapping or attract-and-kill systems before fruit ripening could significantly reduce oviposition pressure during the critical period of fruit susceptibility.

Our results provide preliminary information on the spatiotemporal distribution and ovarian maturation dynamics of *D. suzukii* in a peach orchard. Longer-term studies conducted across multiple sites are recommended to further validate the findings reported here.

The results from the laboratory oviposition assay confirmed that *D. suzukii* readily lay eggs on nectarines, showing a clear preference for healthy fruits, whereas fungal infection significantly reduced oviposition rates. This result agrees with Sato et al. (2021) [44], who suggested that microbial growth can discourage oviposition in SWD. We also observed a strong preference for mechanically damaged fruits over intact ones, consistent with Stewart et al. (2014) [24] and Andreazza et al. (2017) [3], who reported increased attractiveness of injured peaches. This may be related to the release of attractive volatiles of damaged tissue, as observed in other fruit pest systems (e.g., [45]), and to the easier access provided by cut surfaces.

During summer, it is common to find peaches damaged by insects, birds, or weather events such as hail [3]. These injuries may act as attractants for oviposition, and the increasing occurrence of extreme weather events could potentially enhance fruit damage, thereby increasing the risk of infestation.

From IPM perspective, timely removal of damaged and fallen fruits is a key sanitation measure to limit oviposition opportunities and suppress the local population [13]. This practice is particularly important during the post-harvest period, when a high proportion of females carrying mature eggs is present, potentially representing a reservoir for the next generation. Combining sanitation with edge-targeted mass trapping or attract-and-kill strategies may enhance control efficacy in organic systems.

5. Conclusions

The Marche region is becoming an increasingly important area for fruit production, highlighting the need for evidence-based strategies to manage key orchard pests. This study provides new insights into the spatiotemporal dynamics, reproductive biology, and oviposition behavior of *Drosophila suzukii* in organic peach orchards.

We demonstrated that SWD populations in our peach orchard are more abundant along mixed-species-orchard edges, identifying these areas as a hotspot for monitoring. The dynamics of female ovarian maturation confirmed that the period of eggs development coincides with the presence of peaches in the field, indicating a high risk of damage. Laboratory assays further revealed that females preferentially oviposit on healthy and mechanically damaged fruits, suggesting that injured peaches may serve as strong attractants.

From an IPM perspective, our results highlight the need to integrate multiple tactics for sustainable population control. Specifically, mixed hedgerows could be used as strategic sites for winter interventions such as mass trapping, attract-and-kill, or push-pull systems to reduce the initial founder population. Field sanitation, including the timely removal of damaged or fallen fruit, should be prioritized to minimize oviposition sites. The combi-

nation of these practices offers a feasible approach for reducing SWD pressure in organic peach production systems while minimizing reliance on chemical control.

Further studies across multiple orchards and seasons would help to support and extend these results.

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Conflicts of Interest: The authors declare no conflicts of interest.

Abbreviations

The following abbreviations are used in this manuscript:

SWD Spotted Wing Drosophila
IPM Integrated Pest Management

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