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# *Drosophila suzukii* preferentially lays eggs on spherical surfaces with a smaller radius

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*Drosophila suzukii* is an agricultural pest that predominantly harms small fruits, having a serrated ovipositor that is able to pierce the skin of ripening fruits. Its oviposition preference has been studied from various aspects including chemical and physical properties of oviposition substrates. However, its preference for certain shapes or sizes of substrates has not been explored. In this study, we tested the oviposition preference of *D. suzukii* for artificial oviposition substrates with different surface curvatures using 27 strains recently established from wild populations collected in Japan. We found that *D. suzukii* laid more eggs on a surface with smaller radii (4.8 and 5.7 mm) compared with larger radii (7.7 and 9.6 mm). We also found that the most preferred radius differed among strains. Notably, the preference was independent of the volume of substrates, suggesting that *D. suzukii* uses the surface curvature as a cue for its oviposition site selection. These results provide an additional explanation for why *D. suzukii* preferentially uses small fruits as its oviposition sites.

*Drosophila suzukii*, the spotted wing Drosophila, is an invasive agricultural pest expanding its range around the world<sup>1,2</sup>. *D. suzukii* has a serrated ovipositor that is able to penetrate the skin of ripening fruits and preferentially damages small fruits<sup>3,4</sup>. *D. suzukii* uses various cues in its oviposition site selection, including chemical cues (odorants, tastants, and acidity) and physical cues (colour, firmness, and texture)<sup>4–23</sup>. Among them, firmness of the fruit skin has been considered to play an important role in limiting the host range of *D. suzukii* to small fruits<sup>2</sup>. In agreement with the morphological characteristic in its ovipositor, *D. suzukii* could lay eggs on relatively harder artificial substrates compared with other Drosophila species<sup>16</sup>. In contrast, the much harder skin of large fruits has been thought to prevent oviposition even by *D. suzukii*, limiting its hosts to small fruits<sup>1–3</sup>. Although firmness of the fruit skin explains why *D. suzukii* cannot harm large fruits, it does not exclude the possibility that *D. suzukii* prefers small fruits for other reasons. In fact, its oviposition preference for certain shapes or sizes of substrates has not been tested experimentally.

In preliminary experiments using artificial oviposition substrates, we accidentally found that *D. suzukii* laid more eggs on spherical surfaces than on flat surfaces. In this study, we examined whether *D. suzukii* prefers a certain surface curvature, using multiple strains collected in Japan. We also tried to disentangle the effects of substrate size and surface curvature controlling the volume of substrates independently from the curvature.

## Material and methods

***D. suzukii* strains.** *D. suzukii* strains were established from wild populations collected at various locations in Japan during 2019–2021 (Table 1). In most cases, single pairs that emerged from collected host fruits were used to establish the strains to minimize the influence of possible genetic drift during the laboratory culture by reducing the initial genetic variation within a strain. Some strains were established using other methods (trap collection as a source, and a single mated female or multiple pairs as an origin). The strains were cultured in the same way as for *D. melanogaster*. Newly eclosed adults were transferred into vials containing standard Drosophila culture medium made of corn meal and glucose and maintained at 20 °C with the 16L:8D light cycle until the next generation emerged. In this condition, one generation took 3 weeks (17 generations/year). Individuals used for experiments were reared at 25 °C from the larval stage.

**Four-choice assay.** Oviposition substrate was made of 2% agar solution (Seakem® LE Agarose, Lonza, Basel, Switzerland) cast in silicone moulds for UV resin-crafting hobbies, which have hemispherical holes of various sizes. Oviposition substrates with different surface curvatures of the same volume were prepared by pouring 200 µl of agar solution into each hole and allowed to solidify (shapes of the oviposition substrates are

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