

Figure S1. Scanning electron microscope images of ovipositors and pocket-shaped structures. Lateral (A)(C) and posterior (B)(D) views of ovipositors (OV) and pocket-shaped structures of the post-anal cuticle at the dorsal base regions of ovipositors (circled in blue) in *D. sukii* (WT3)(A)(B) and *D. subpulchrella* (H243)(C)(D) females. The samples were fixed in ice-cold ethanol, substituted with t-butanol, sublimation dried, and observed under a JEOL JSM-6510 scanning electron microscope after being coated with gold. Scale bar is 100 μm.

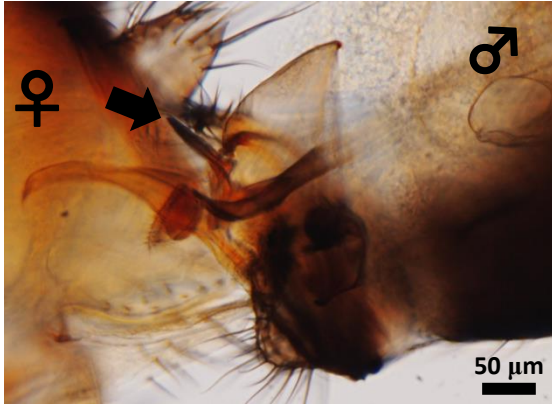
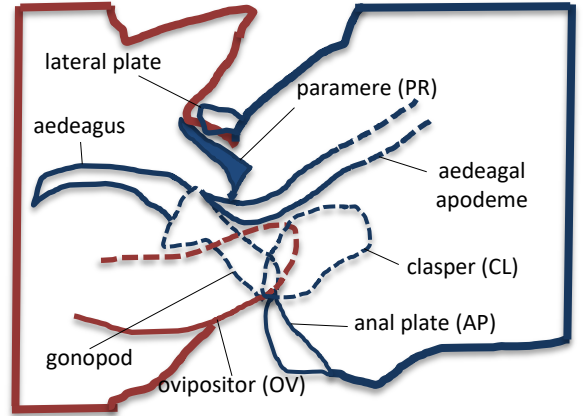
A**A'**

Figure S2. Paramere contacts female during copulation in *D. biarmipes* (MYS118). (A) Genitalia coupling of a copulating pair of *D. biarmipes*, which was flash frozen in chilled ethanol at 13 min from initiation. Observed after being cleared. Arrow indicates the paramere. (A') Coloured map of (A). Red and blue colours indicate female and male genital structures, respectively.

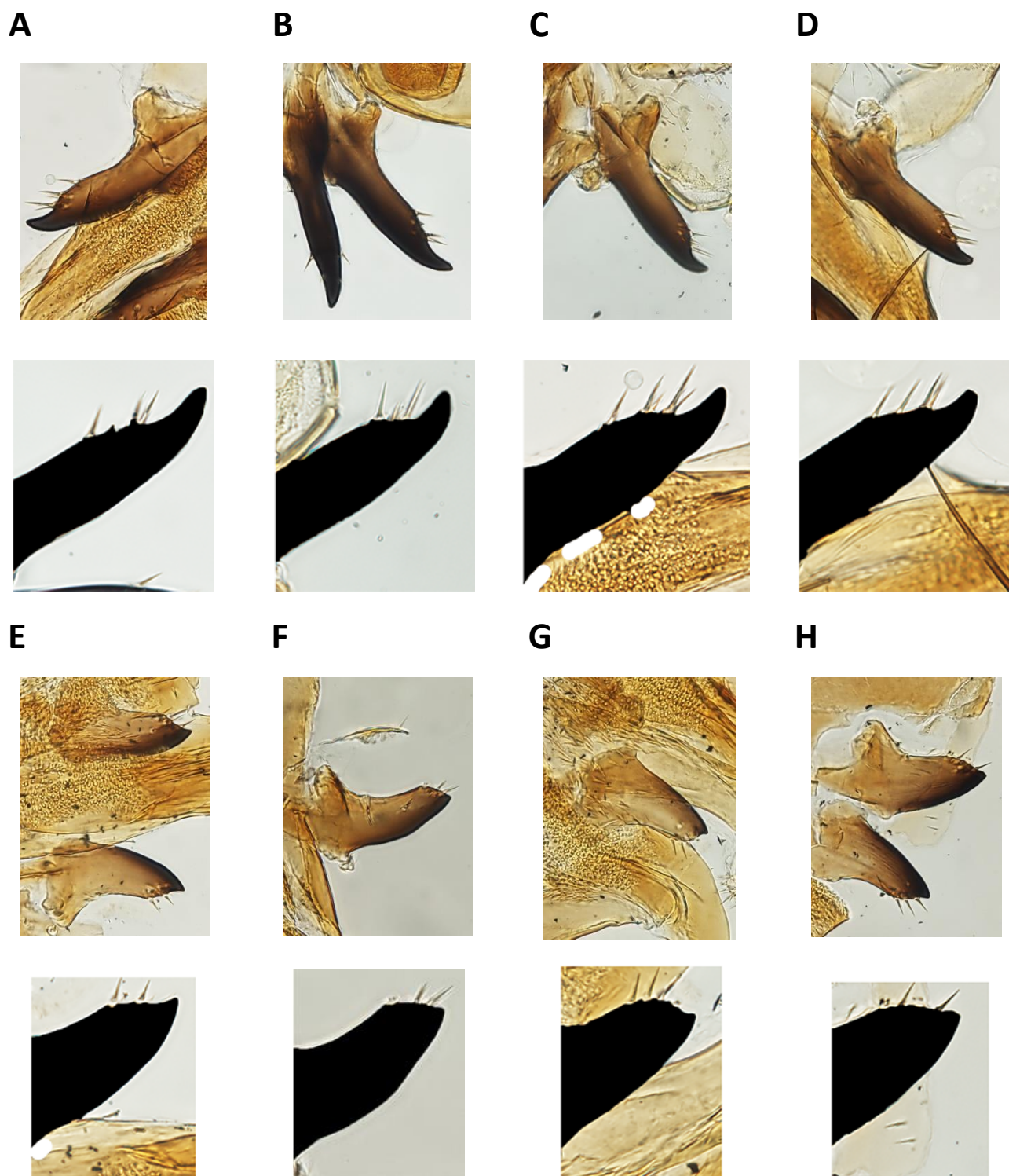


Figure S3. Variation in paramere shape. (A)–(H) Original photograph ($\times 64$)(upper panel) and the extracted shape (lower panel) are shown for each sample corresponding to A–H in figure 4c. (A) *D. subpulchrella* NAR9-04. (B) *D. subpulchrella* NAR12-02. (C) *D. subpulchrella* NAR1-02. (D) *D. subpulchrella* NAR14-04. (E) *D. suzukii* TMUS5-04. (F) *D. suzukii* TMUS21-05. (G) *D. suzukii* TMUS8-03. (H) *D. suzukii* TMUS5-01.

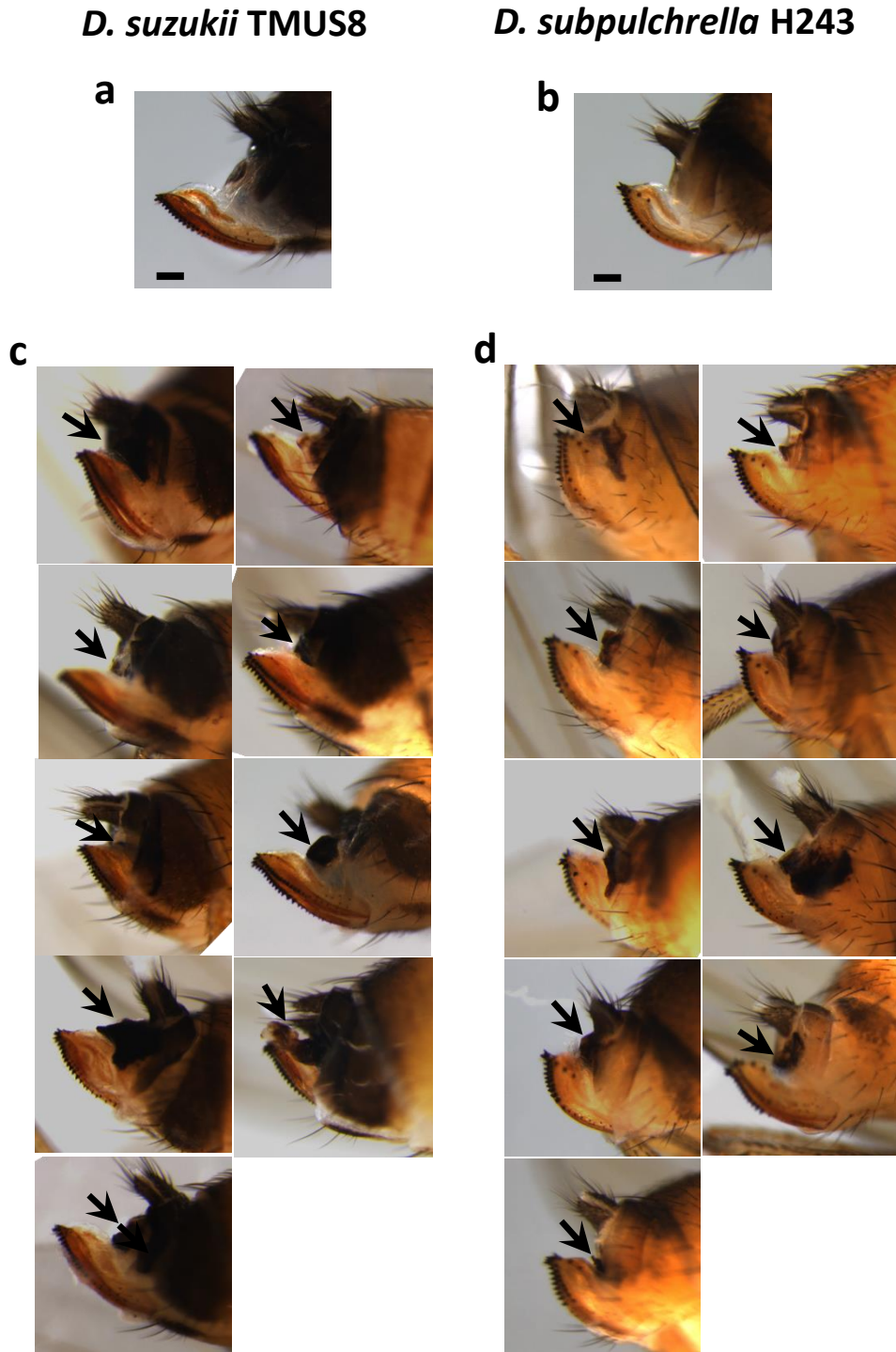


Figure S4. Surgical wound induced on the pocket-shaped structure of the post-anal cuticle to disrupt paramere contact during copulation. (a) Intact post-anal cuticle of female *D. suzukii* (TMUS8). (b) Intact post-anal cuticle of *D. subpulchrella* (H243) female. (c) Nine females of *D. suzukii* (TMUS8) copulated within 3 h of video recording with wound induced on the pocket-shaped structure indicated by arrows. (d) Nine females of *D. subpulchrella* (H243) copulated within 3 h of video recording with wound induced on the pocket-shaped structure indicated by arrows.