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The genus *Leucophenga* (Diptera, Drosophilidae), part III: the *interrupta* species group from the Oriental region, with morphological and molecular evidence

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Abstract

A new species group, the *interrupta* group, is established within the genus *Leucophenga* based on two known and three new species, all of which are endemic to the Oriental region: *L. interrupta* Duda, 1924, *L. neointerrupta* Fartyal & Toda, 2005, *L. bifurcata* sp. nov., *L. quadrifurcata* sp. nov. and *L. retifoliacea* sp. nov. A key to the five species of the *interrupta* group is provided. Sixteen mtDNA *COI* sequences of the five species are analyzed; the molecular data are used as complementary evidence for the species boundaries defined by the morphological data.

Key words: DNA barcoding, *Leucophenga interrupta* species group, new species, Oriental region

Introduction

Leucophenga interrupta Duda, 1924 and *L. neointerrupta* Fartyal & Toda, 2005 have been reported from the Oriental region and assigned to the *L. ornata* species group (Okada 1990; Fartyal *et al.* 2005; Bächli 2013); however, they distinctly differ from the other species of the *ornata* group in having the wing usually clear on r–m and dm–cu crossveins and with broad, dark longitudinal stripe along anterodistal portion, and the aedeagus with strong processes basolaterally.

In the present study, three new species from Yunnan, China are described; they are morphologically similar to *L. interrupta* and *L. neointerrupta* in the wing color pattern and/or the aedeagus with basolateral processes. Thus, a new species group, namely the *interrupta* group, is established here, based on two known and three new species. One Afrotropical species, *Leucophenga denigrata* Bächli, 1971, shares the wing with broad, dark longitudinal stripe along anterodistal portion (fig. 38f in Bächli 1971), but its male terminalia has not been described. We provisionally refrain from assigning this Afrotropical species to the *interrupta* group to avoid further confusion. We analyze sixteen *COI* barcode sequences of the two known and three new species in order to evaluate morphological delimitation for these species.

Material and methods

Material and morphological terminology. All specimens examined were collected by sweeping on tussocks and tree trunks along streams in forest. The type specimens are deposited in Department of Entomology, South China Agricultural University, Guangzhou, China (SCAU). We followed Zhang & Toda (1992) and Chen & Toda (2001) for the definitions of measurements, indices and abbreviations, and Chen & Toda (1994) for the interpretation of the homology on the phallic organs in the genus *Leucophenga*.

Phylogenetic analyses. DNA extraction, gene amplification, sequencing and sequence alignment were made by the same methods as in Huang *et al.* (2013). A NJ (neighbor-joining) tree was constructed in MEGA 5 with K-2P distances, and a Bayesian tree was constructed in MrBayes 3.1.2 (Huelsenbeck & Ronquist 2001; Ronquist &

Huelsenbeck 2003). A substitution model of GTR with gamma-distributed rate variation among sites (+G) was selected with AIC criterion in jModelTest 0.1.1 (Guindon & Gascuel 2003; Posada 2008). In total, 3,000,000 generations were executed in Bayesian analyses, and trees were sampled every 100 generations, and the initial 25% of the samples were discarded as burn-in. All compatible groups were shown in the consensus tree. In both NJ and Bayesian analyses, two *Leucophenga* species, *L. abbreviata* and *L. angusta*, were used as out-group taxa (Table 1).

Results

The NJ and Bayesian trees are shown in Fig. 1 (A and B, respectively). In the phylogeny of the *ornata* group, the five focal species formed a monophyletic group, which was strongly supported in both phylogenetic trees (Bootstraps, BP = 97, in NJ tree; Posterior probability, PP = 1.0, in Bayesian tree). In the clade of this group, the monophyly of *L. interrupta* and *L. retifoliacea* were robustly supported in both phylogenetic trees, however, *L. neointerrupta* was paraphyletic with *L. quadrifurcata* in the Bayesian tree. The status of *L. bifurcata* was not stable; it was placed as the earliest diverged species in the NJ tree but as the sister species to *L. interrupta* in the Bayesian tree.

Morphological study

Leucophenga interrupta species group

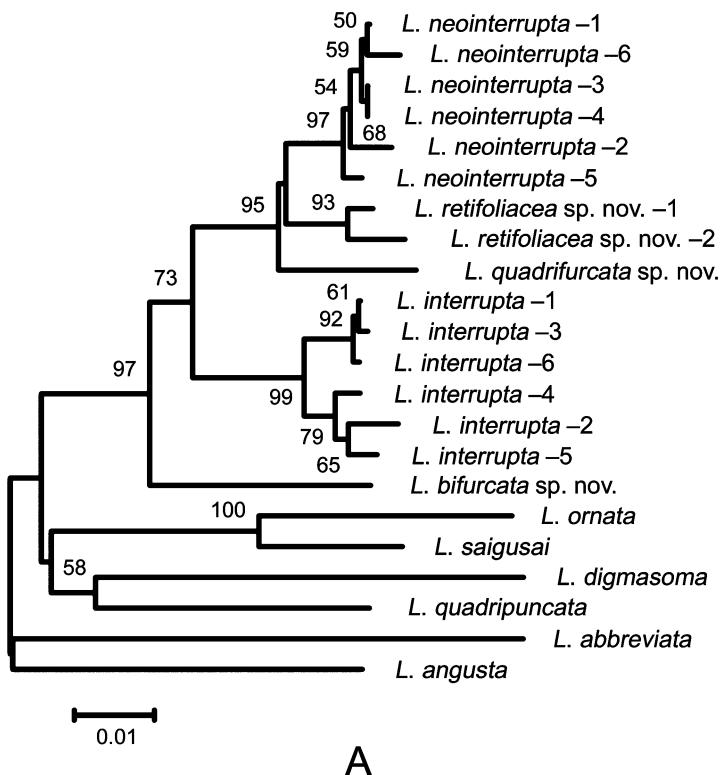
Diagnosis. Wing usually clear on r-m and dm-cu crossveins, with broad, dark longitudinal stripe along anterodistal portion (Figs. 2A, D, G, J, M); paramere large, usually with wrinkles (Figs. 4D, 5–8C); aedeagus with sclerotized processes basolaterally (bp in Figs. 4E, 5–8D).

Description. Male and female: Eyes red to brownish red. Ocellar triangle yellow to yellowish brown; ocellar (oc s) and postocellar setae (poc s) developed (Fig. 4A). Frons narrow, nearly parallel, with a few minute setulae medially. All orbital setae large; proclinate (Orb_1) and anterior reclinate orbital setae (Orb_2) very close together, separated by distance less than 1/2 of that between anterior reclinate and posterior reclinate (Orb_3) (Fig. 4A). Arista long plumose. Face mostly yellow; facial carina undeveloped. Palpus mostly brownish yellow, slightly larger distally in female than in male. Vibrissa prominent; subvibrissal setae small. Gena and postgena narrow. Scutum mostly brownish yellow, slightly silvery pollinose, lacking patch or stripe. Postpronotal lobe with 1–3 long seta(e) and a few of shorter setae. Acrostichal setulae in 10–14 irregular rows. Prescutellar setae large. Scutellum paler around apical scutellar setae but not at tip. Pleura lacking longitudinal stripe above. Katepisternal mostly brownish yellow, with 2 long and some small setae. Meron mostly brownish yellow. Subscutellum swollen. Basal medial-cubital crossvein absent. Costal vein between R_{2+3} and R_{4+5} distally with 5–9 peg-like spinules on ventral surface; R_{2+3} slightly curved; R_{4+5} and M_1 nearly parallel distally. Halter mostly yellow. Legs mostly yellowish. Abdominal tergites: first entirely yellow; second mostly yellow, with a pair of small dark patches anterolaterally; rest variable in color and pattern (Fig. 3). Male terminalia: Epandrium usually pubescent, with several setae around ventral corner to posterior margin and developed apodeme along anterior margin. Surstyli usually broad and flat, with pubescence and several setae on outer and inner surfaces. Cercus separated from epandrium, pubescent and setigerous. Hypandrium (gonopod in Bächli *et al.* 2004) anteriorly fused to aedeagal apodeme, laterally broad, lacking paramedian setae. Gonopods (dorsal arch in Bächli *et al.* 2004) fused with each other, forming slightly triangular plate, anteroventrally with curved, median rod. Paramere (outer paraphysis in Bächli *et al.* 2004) contiguous to arm of aedeagal apodeme basally, glabrous, usually with a few sensilla distally. Aedeagal basal bridges contiguous to median rod of gonopods.

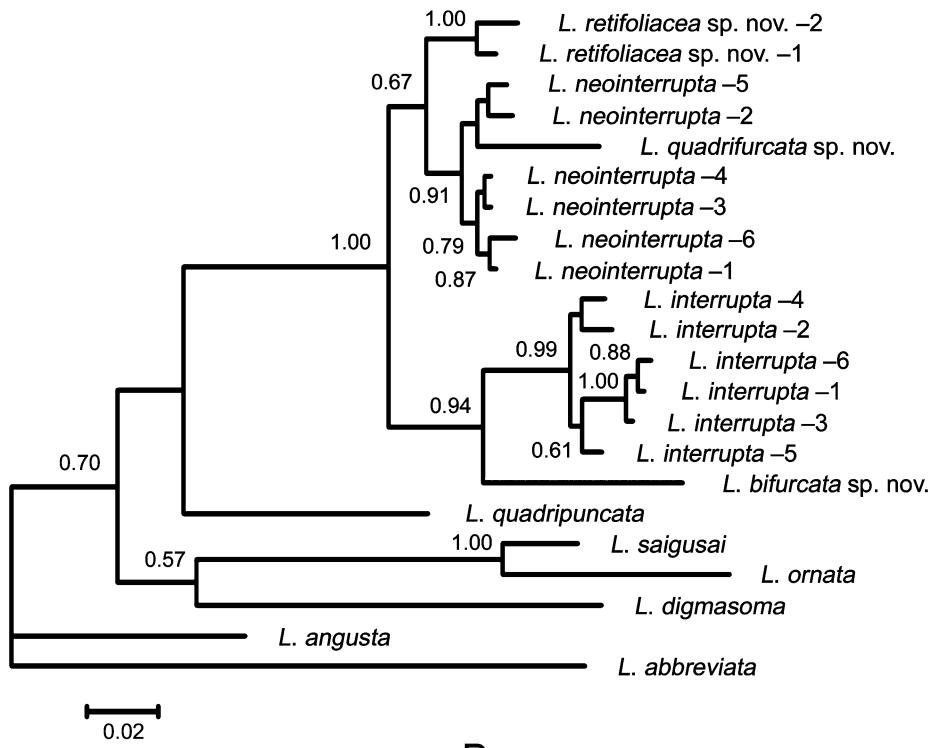
In the following descriptions of individual species, only characters that depart from the above universal characters are described for brevity.

TABLE 1. Details of the samples using in the DNA and accession numbers of the *COI* sequences (* Sequences determined in Li *et al.* 2013; Su *et al.* 2013; Huang *et al.* 2013).

Species groups	Species	Sex	Localities	BOLD Process ID	GenBank accession numbers
<i>abbreviata</i>	<i>L. abbreviata</i> (de Meijere, 1911)	♂	Dinghushan, Zhaoqing, Guangdong, China	/	HM636455*
<i>mutabilis</i>	<i>L. angusta</i> Okada, 1956	♂	Minamiohsawa, Hachijoji, Tokyo, Japan	/	HQ842780*
<i>ornata</i>	<i>L. ornata</i> Wheeler, 1959	♂	Tomochi, Kumamoto, Kyushu, Japan	BDOR001-12*	JX887657*
	<i>L. saigusai</i> Okada, 1968	♂	Maoniuhé, Danba, Sichuan, China	BDOR004-12*	JX887678*
	<i>L. digmasoma</i> Lin & Wheeler, 1972	♂	Jianfengling, Ledong, Hainan, China	BDORD001-13	KC861387
	<i>L. quadripunctata</i> (de Meijere, 1908)	♂	Minamiohsawa, Hachijoji, Tokyo, Japan	BDORD002-13	KC861400
<i>interrupta</i>	<i>L. interrupta</i> Duda, 1924-1	♀	Lugu, Nantou, Taiwan, China	BDORD003-13	KC861390
	<i>L. interrupta</i> Duda, 1924-2	♂	Guanshan, Taidong, Taiwan, China	BDORD004-13	KC861391
	<i>L. interrupta</i> Duda, 1924-3	♂	Guanshan, Taidong, Taiwan, China	BDORD005-13	KC861392
	<i>L. interrupta</i> Duda, 1924-4	♀	Jianfengling, Ledong, Hainan, China	BDORD006-13	KC861389
	<i>L. interrupta</i> Duda, 1924-5	♂	Wangtianshu, Mengla, Yunnan, China	BDORD007-13	KC861388
	<i>L. interrupta</i> Duda, 1924-6	♂	Sagarmatha, Siraha, Nepal	BDORD008-13	KC861393
	<i>L. neointerrupta</i> Fartyal & Toda, 2005-1	♀	Mangshan, Yizhang, Hunan, China	BDORD009-13	KC861394
	<i>L. neointerrupta</i> Fartyal & Toda, 2005-2	♀	Conghua, Guangzhou, Guangdong, China	BDORD010-13	KC861399
	<i>L. neointerrupta</i> Fartyal & Toda, 2005-3	♂	Dinghushan, Zhaoqing, Guangdong, China	BDORD011-13	KC861398
	<i>L. neointerrupta</i> Fartyal & Toda, 2005-4	♀	Hesong, Menghai, Yunnan, China	BDORD012-13	KC861397
	<i>L. neointerrupta</i> Fartyal & Toda, 2005-5	♂	Wangtianshu, Mengla, Yunnan, China	BDORD013-13	KC861396
	<i>L. neointerrupta</i> Fartyal & Toda, 2005-6	♂	Muyiji, Ximeng, Yunnan, China	BDORD014-13	KC861395
	<i>L. bifurcata</i> sp. nov.	♂	Menglun, Mengla, Yunnan, China	BDORD015-13	KC861386
	<i>L. quadrifurcata</i> sp. nov.	♂	Muyiji, Ximeng, Yunnan, China	BDORD016-13	KC861401
	<i>L. reitfoliaeas</i> sp. nov. -1	♂	Hesong, Menghai, Yunnan, China	BDORD017-13	KC861402
	<i>L. retifoliacea</i> sp. nov. -2	♀	Hesong, Menghai, Yunnan, China	BDORD018-13	KC958573



A



B

FIGURE 1. The phylogenetic trees are used in this study. A. Neighbor-joining (NJ) tree; B. Bayesian Inference (BI) tree. The numbers around the nodes of NJ tree are bootstrap values (BP), which re-sampling 1,000 replicates; the numbers around the nodes of BI tree are posterior probabilities (PP). BP values lower than 50 and PP values lower than 0.5 are not shown in the trees. The bar indicates the estimated number of substitutions per site.

***Leucophenga interrupta* Duda, 1924**

(Figs. 2A, 2B, 2C, 3A, 3B, 4)

Leucophenga interrupta Duda, 1924: 187; Okada, 1956: 36; Okada, 1990: 684.

Diagnosis. Aedeagus slightly acute apically, with a pair of strong, acute processes subbasally (pr in Fig. 4E); aedeagal, basolateral process slender, with several serrated processes distally (Fig. 4E).

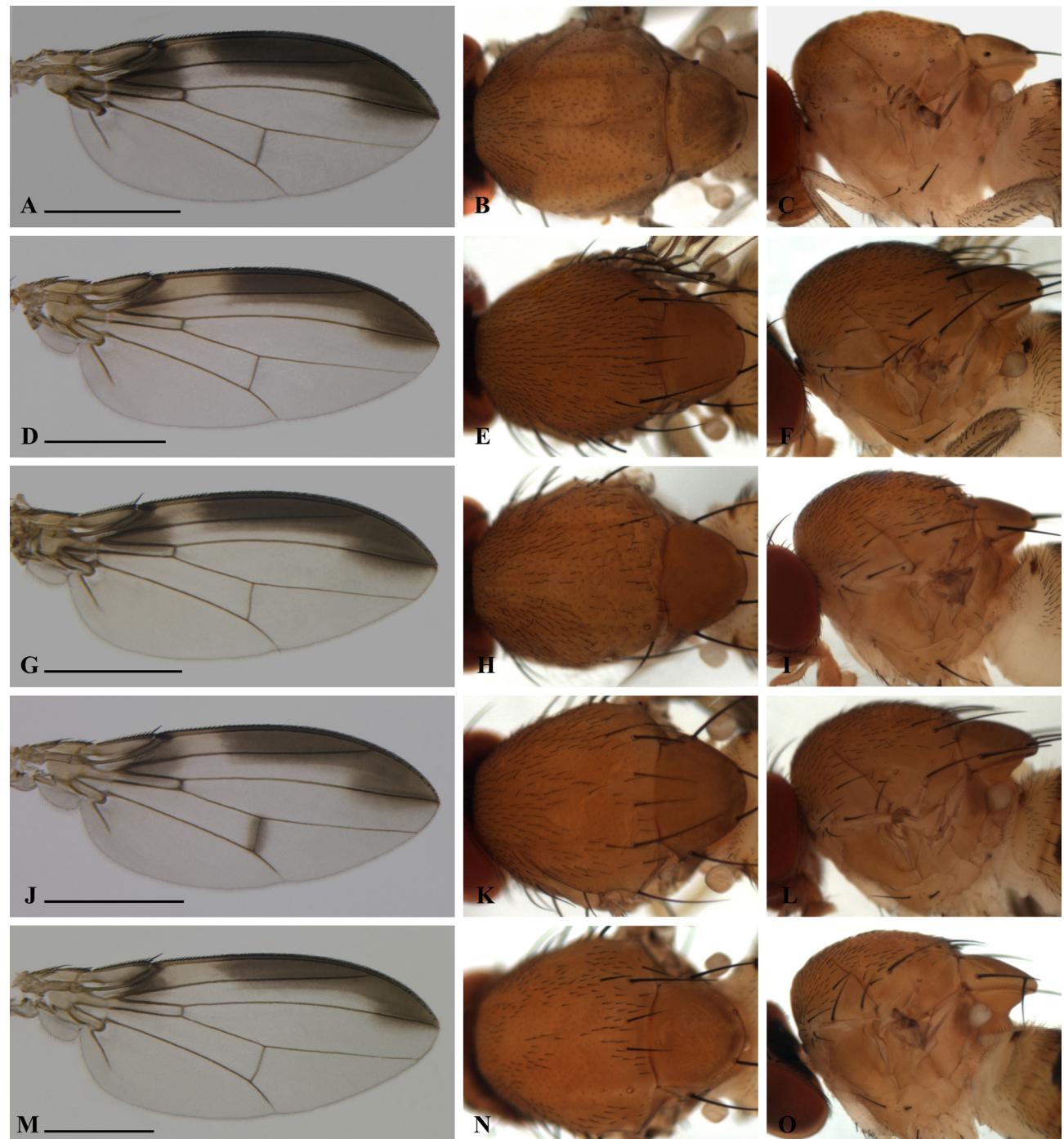


FIGURE 2. Wing, mesonotum, scutellum and pleura of male: A, B, C. *Leucophenga interrupta* Duda, 1924; D, E, F. *Leucophenga neointerrupta* Fartyal & Toda, 2005; G, H, I. *Leucophenga bifurcata* sp. nov.; J, K, L. *Leucophenga quadrifurcata* sp. nov.; M, N, O. *Leucophenga retifoliacea* sp. nov.

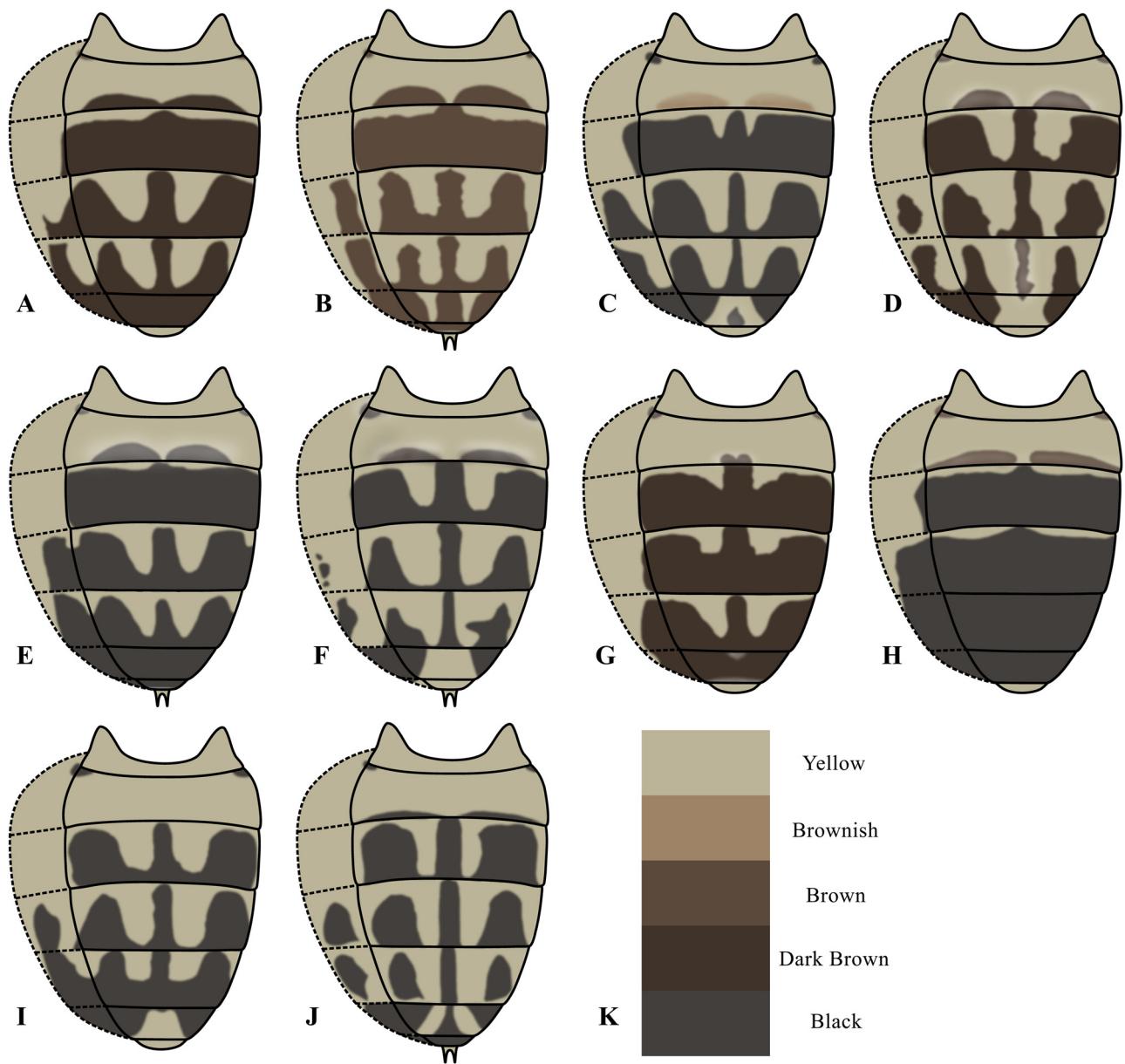


FIGURE 3. Patterns of abdominal tergites of male and female: A, B. *Leucophenga interrupta*; C, D, E, F. *L. neointerrupta*; G. *L. bifurcata* sp. nov.; H. *L. quadrifurcata* sp. nov.; I, J. *L. retifoliacea* sp. nov.; K. Color legend.

Description. Ocellar triangle yellowish brown, with 3–4 setae above ocellar setae (Fig. 4A). Frons brownish yellow to yellowish brown. Pedicel brownish yellow; first flagellomere yellow. Clypeus brownish yellow medially and brown laterally. Postpronotal lobe brownish yellow (Fig. 2C). Scutellum brownish yellow (Fig. 2B). Wing (Fig. 2A): Costal vein between R_{2+3} and R_{4+5} distally with 5–7 peg-like spinules on ventral surface. Halter brownish laterally. Abdominal tergites brown to dark brown (Fig. 3A, B); second mostly yellow, but brown posteriorly; third with a pair of yellow patches submedially on anterior margin, yellow laterally; fourth and fifth each with 2 pairs of yellow patches submedially on anterior margin, yellow laterally; sixth dark brown in male, with a pair of yellow patches submedially in female. Male terminalia: Epandrium with ca. 11 setae near posterior margin and ventral corner per side (Fig. 4B). Paramere nearly rectangular, with distinct wrinkles and 4 sensilla (Fig. 4D). Aedeagus bifurcated from base, slightly reticulated ventrally (Fig. 4E).

Measurements. BL = 2.73–3.53 mm in ♂, 3.20 mm in ♀ (range in 4♂ and 1♀), THL = 1.30–1.80 mm in ♂, 1.40 mm in ♀, WL = 2.17–3.03 mm in ♂, 2.33 mm in ♀, WW = 1.00–1.27 mm in ♂, 1.00 mm in ♀, arb = 8–10/4–5, avd = 0.80–1.07, adf = 1.88–2.57, flw = 1.73–2.43, FW/HW = 0.35–0.40, ch/o = 0.03–0.04, prorb = 0.65–0.80, rcorb = 0.61–0.91, vb = 0.36–0.45, dcl = 0.43–0.54, presctl = 0.53–0.58, sctl = 1.14–1.33, sterno = 0.64–0.92,

orbito = 1.82–2.25, dcp = 0.19–0.28, sctlp = 0.97–1.13, C = 1.93–2.43, 4c = 1.05–1.33, 4v = 1.67–2.03, 5x = 0.75–0.94, ac = 2.21–2.86, M = 0.38–0.53, C3F = 0.92–0.98.

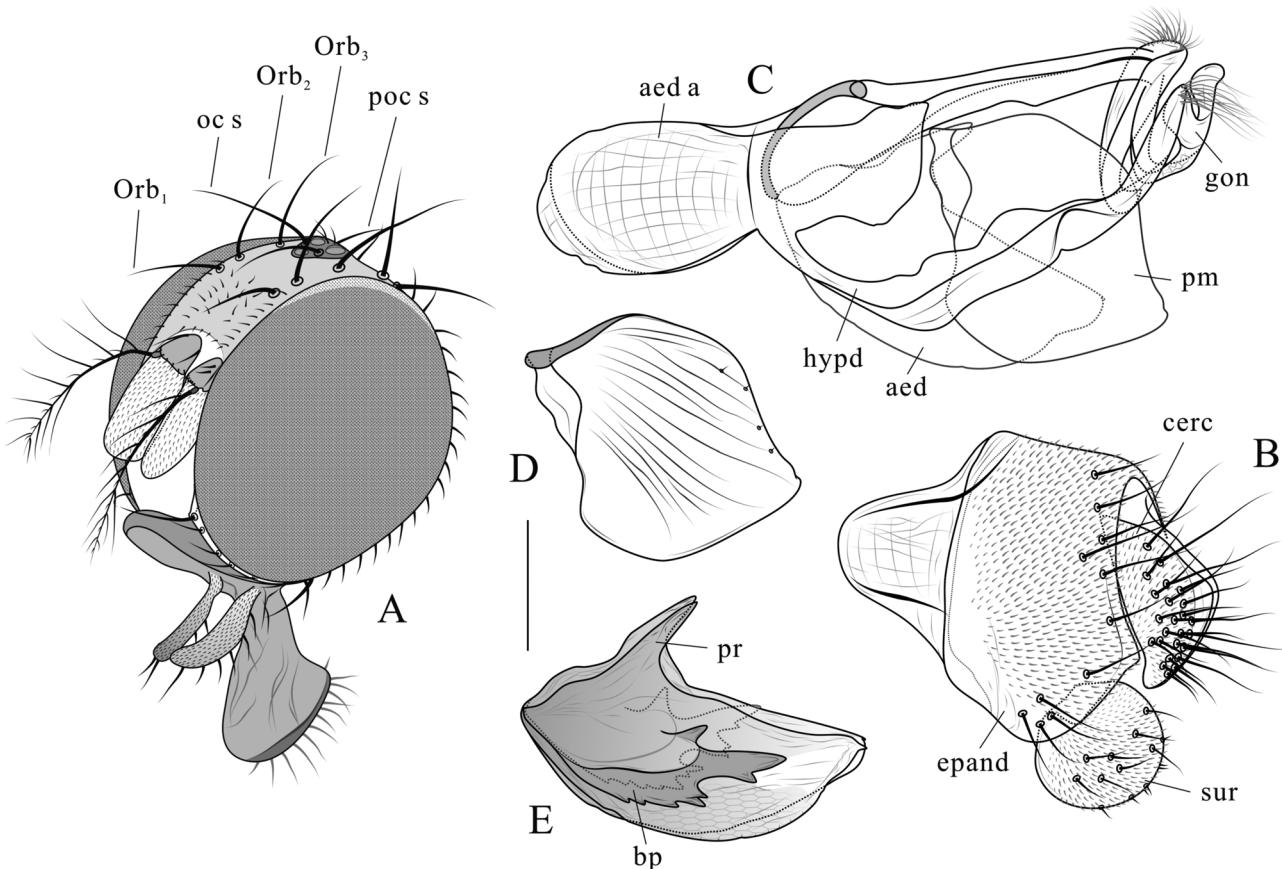


FIGURE 4. *Leucophenga interrupta* Duda, 1924, male head and terminalia in lateral view. A. head; B. epandrium (epand), surstyli (sur) and cercus (cerc); C. hypandrium (hypd), paramere (pm), gonopods (gon), aedeagus (aed) and aedeagal apodeme (aed a); D. paramere; E. aedeagus. Scale bars = 0.1 mm.

Specimens examined. CHINA: 2♀ (SCAU, Nos 122947, 48), Lugu, Nantou, Taiwan, 27°44'N, 120°47'E, 350m, 18.x.2012, ex tussocks, HW Chen; 1♂ (SCAU, No. 122900), Guanshan, Taidong, Taiwan, 23°10'N, 121°03'E, 770m, 29.x.2012, ex tree trunk, HW Chen; 1♂ (SCAU, No. 122901), Zhiben, Taidong, Taiwan, 23°10'N, 121°03'E, 340m, 30.x.2012, ex tussock, HW Chen; 1♀ (SCAU, No. 122902), Jianfengling, Ledong, Hainan, 18°41'N, 108°52'E, 750m, 18.iv.2008, ex tussock, JJ Gao; 2♂ (SCAU, Nos 122903, 04), Wangtianshu, Mengla, Yunnan, 21°28'N, 101°38'E, 580m, 23.iv.2007, ex tree trunks, HW Chen. NEPAL: 2♂ (SCAU, Nos. 122905, 06), Sagarmatha, Siraha, 27°59'N, 86°40'E, 19.x.2011, ex tussocks, XS Chen.

Distribution. China (Taiwan, Hainan, Yunnan), Japan (Kyushu, Ryukyu Is.), Nepal (Siraha).

Leucophenga neointerrupta Fartyal & Toda, 2005

(Figs. 2D, 2E, 2F, 3C, 3D, 3E, 3F, 5)

Leucophenga neointerrupta Fartyal & Toda in Fartyal *et al.* 2005: 412.

Diagnosis. Parameres slightly triangular, sclerotized dorsally, with weak wrinkles and 3 serrations subapically (Fig. 5C); aedeagus with 5 serrations in regular row distally; aedeagal, basolateral process short, tripartite distally (Fig. 5D).

Measurements. BL = 3.20–3.70 mm in ♂, 2.90–3.73 mm in ♀ (range in 5♂ and 5♀), THL = 1.53–1.90 mm in ♂, 1.37–1.93 mm in ♀, WL = 2.60–3.07 mm in ♂, 2.20–3.27 mm in ♀, WW = 1.13–1.37 mm in ♂, 0.98–1.40 mm in ♀, arb = 7–11/3–5, avd = 0.81–1.00, adf = 1.79–2.40, flw = 1.78–2.13, FW/HW = 0.35–0.39, ch/o = 0.04–0.05,

$\text{prorb} = 0.54\text{--}0.82$, $\text{rcorb} = 0.59\text{--}0.73$, $\text{vb} = 0.33\text{--}0.43$, $\text{dcl} = 0.39\text{--}0.48$, $\text{presctl} = 0.49\text{--}0.60$, $\text{sctl} = 1.06\text{--}1.53$, $\text{sterno} = 0.68\text{--}0.82$, $\text{orbito} = 1.82\text{--}2.10$, $\text{dcp} = 0.20\text{--}0.29$, $\text{sctlp} = 1.09\text{--}1.29$, $C = 2.03\text{--}2.44$, $4c = 1.07\text{--}1.39$, $4v = 1.73\text{--}2.10$, $5x = 0.76\text{--}0.91$, $\text{ac} = 2.33\text{--}2.94$, $M = 0.41\text{--}0.50$, $\text{C3F} = 0.88\text{--}1.00$.

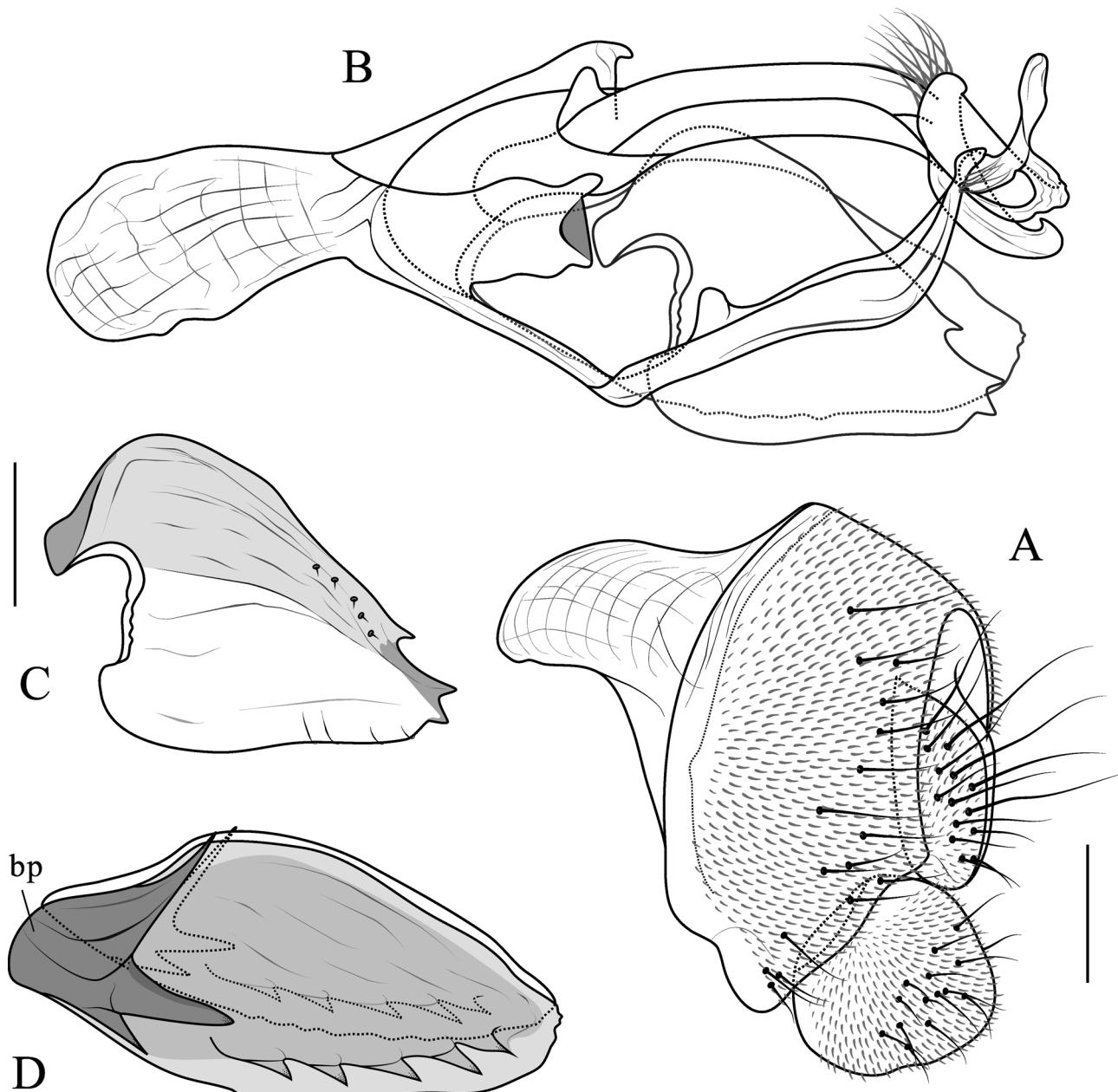


FIGURE 5. *Leucophenga neointerrupta* Fartyal & Toda, 2005, male terminalia in lateral view: A. epandrium, surstyli and cercus; B. hypandrium, paramere, gonopods, aedeagus and aedeagal apodeme; C. paramere; D. aedeagus. Scale bars = 0.1 mm.

Material examined. CHINA: 1♀ (SCAU, No. 122907), Mangshan, Yizhang, Hunan, 24°55'N, 112°45'E, 1200m, 3.x.2004, ex tussock, MF Xu; 3♀ (SCAU, Nos 122908–10), Conghua, Guangzhou, Guangdong, 23°26'N, 113°30'E, 200m, 24.v.2011, ex tussocks, SJ Yan; 1♂, 1♀ (SCAU, Nos 122911, 12), Dinghushan, Zhaoqing, Guangdong, 25°00'N, 112°34'E, 570m, 5.iv.2005, ex tree trunks, HL Cao; 4♂, 6♀ (SCAU, Nos 122913–122922), Wangtianshu, Mengla, Yunnan, 580m, 23.iv.2007, ex tree trunks, HW Chen, JJ Gao; 1♀ (SCAU, No. 122924), Hesong, Menghai, Yunnan, 21°50'N, 100°06'E, 1900m, 4.iv.2011, ex tussock, JM Lu; 1♂, 2♀ (SCAU, Nos 122925–27), Muyiji, Ximeng, Yunnan, 22°37'N, 99°36'E, 1100m, 4.iv.2011, ex tussocks, YR Su.

Distribution. China (Hunan, Guangdong, Yunnan), India (Uttaranchal, Uttar Pradesh, West Bengal).

Leucophenga bifurcata sp. nov.

(Figs. 2G, 2H, 2I, 3G, 6)

Diagnosis. Paramere curved cucullately, subbasally with several serrations on inner margin (Fig. 6C); aedeagal basolateral process slender, longer than aedeagus, slightly curved submedially, slightly bifurcated apically (Fig. 6D).

Description. Ocellar triangle yellow, with 4 setae above ocellar setae. Frons yellow. Pedicel yellowish brown; first flagellomere yellow. Clypeus brown. Postpronotal lobe yellow (Fig. 2I). Scutellum brownish yellow (Fig. 2H). Wing (Fig. 2G): Costal vein between R_{2+3} and R_{4+5} distally with 5–7 peg-like spinules on ventral surface. Abdominal tergites dark brown (Fig. 3G); second mostly yellow; third to fifth each with a pair of yellow patches submedially on anterior margin, yellow laterally; sixth with a small yellow patch medially on anterior margin, yellow laterally. Male terminalia: Epandrium with ca. 15 setae near posterior margin and ventral corner per side (Fig. 6A). Paramere with several sensilla subbasally and submedially (Fig. 6C). Hypandrium with parameres and gonopods (Fig. 6B). Aedeagus with aedeagal apodeme (Fig. 6D).

Measurements. BL = 3.73 mm in holotype, THL = 1.73 mm, WL = 2.73 mm, WW = 1.22 mm, arb = 9/5, avd = 0.94, adf = 2.12, flw = 1.94, FW/HW = 0.37, ch/o = 0.03, prob = 0.70, rcorb = 0.78, vb = 0.56, dcl = damaged, presctl = damaged, sctl = 1.17, sterno = damaged, orbito = 1.80, dcp = 0.26, sctlp = 1.03, C = 2.49, 4c = 1.08, 4v = 1.90, 5x = 0.90, ac = 2.26, M = 0.45, C3F = 0.95.

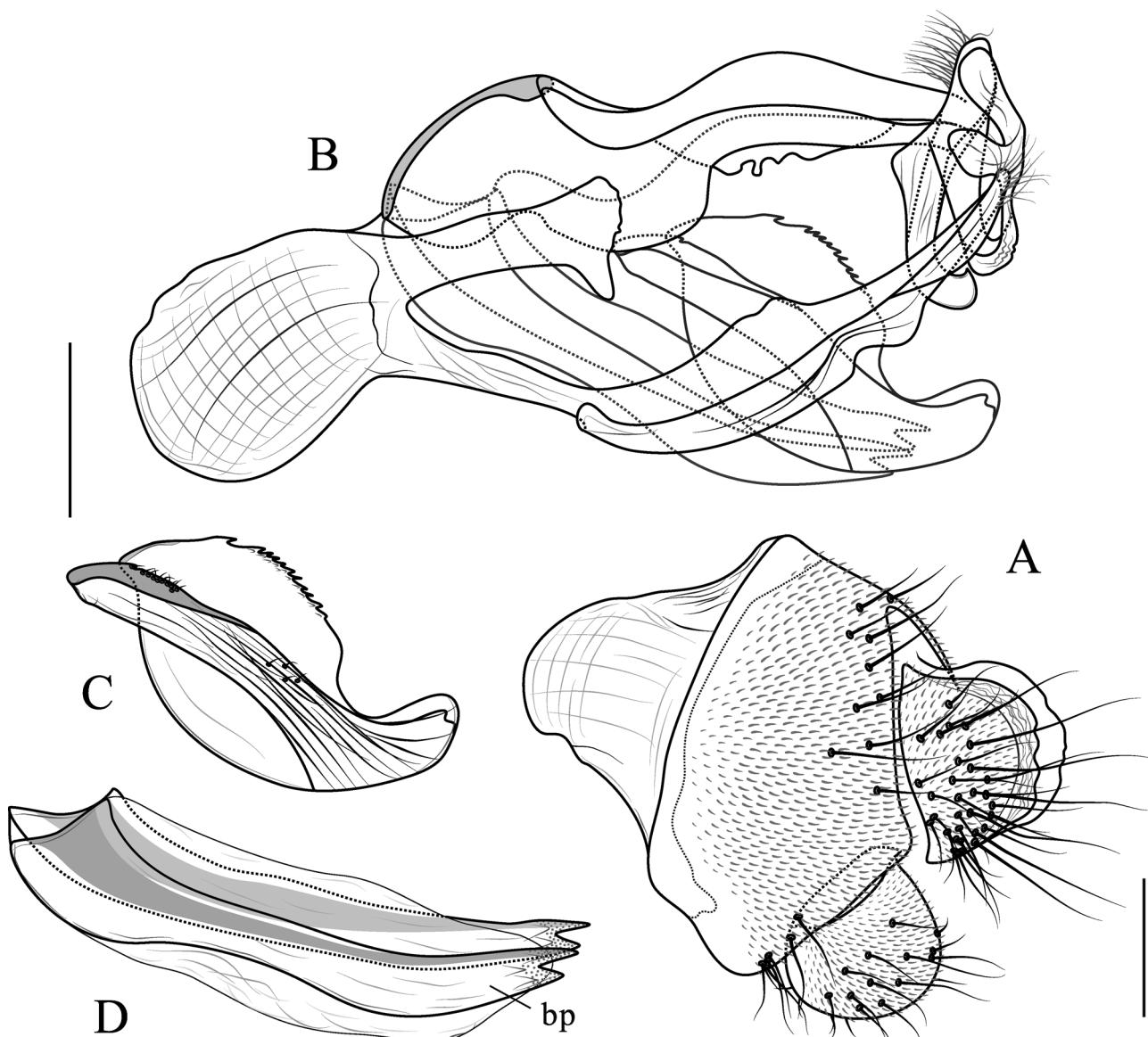


FIGURE 6. *Leucophenga bifurcata* sp. nov., male terminalia in lateral view: A. epandrium, surstyli and cercus; B. hypandrium, paramere, gonopods, aedeagus and aedeagal apodeme; C. paramere; D. aedeagus. Scale bars = 0.1 mm.

Type specimens. Holotype ♂ (SCAU, No. 122928), CHINA: Menglun, Mengla, Yunnan, 21°41'N, 101°25'E, 580m, 24.iv.2007, ex tree trunk, HW Chen.

Etymology. A combination of the Latin word “*bi-*” (= two) + “*furca*” (= furcate), referring to the aedeagal basolateral process bifurcated apically.

Distribution. China (Yunnan).

***Leucophenga quadrifurcata* sp. nov.**

(Figs. 2J, 2K, 2L, 3H, 7)

Diagnosis. This species differs from the other species of the *interrupta* group in having the following characters: wing clouded on r-m and dm-cu crossveins (Fig. 2J); aedeagus not bifurcated; aedeagal basolateral process slender basally, quadrifurcated distally (Fig. 7D).

Description. Ocellar triangle yellowish brown to dark brown, with 3–4 setae above ocellar setae. Frons brownish yellow to yellowish brown. Pedicel yellowish brown; first flagellomere yellow. Clypeus brown. Scutum yellowish brown (Fig. 2K). Postpronotal lobe brownish yellow (Fig. 2L). Scutellum yellowish brown (Fig. 2K). Wing (Fig. 2J): Costal vein between R_{2+3} and R_{4+5} distally with 6–8 peg-like spinules on ventral surface. Halter mostly brownish. Abdominal tergites black (Fig. 3H); second mostly yellow, but brown posteriorly; third and fourth each with a pair of narrow, yellow patches submedially on anterior margin, yellow laterally; fifth to sixth mostly black. Male terminalia: Epandrium with ca. 10 setae near posterior margin and ventral corner per side (Fig. 7A). Paramere nearly rectangular distally, with several weak wrinkles, 2 sensilla and pubescence subdorsally (Fig. 7C).

Measurements. BL = 2.93 mm in holotype (range in 1♂ and 2♀ paratypes: 3.60 mm in ♂, 3.50–3.73 mm in ♀), THL = 1.53 mm (1.77 mm in ♂, 1.70–1.93 mm in ♀), WL = 2.60 mm (3.00 mm in ♂, 3.10–3.27 mm in ♀), WW = 1.20 mm (1.33 mm in ♂, 1.24–1.40 mm in ♀), arb = 5/3 (7–8/3–4), avd = 0.91 (0.81–0.90), adf = 1.94 (2.06–2.13), flw = 1.56 (2.25), FW/HW = 0.38 (0.34–0.37), ch/o = 0.03 (0.03–0.04), prorb = 0.56 (0.63–0.63), rcorb = 0.72 (0.65–0.75), vb = 0.29 (0.39–0.46), dcl = 0.39 (0.45–0.55), presctl = 0.61 (0.48–0.64), sctl = 1.29 (1.03–1.13), sterno = 0.75 (0.78–0.86), orbito = 1.89 (2.00), dcp = 0.21 (0.19–0.20), sctlp = 1.22 (1.11–1.13), C = 2.35 (2.69–2.71), 4c = 1.08 (0.93–0.95), 4v = 1.81 (1.68–1.82), 5x = 0.75 (0.82–1.00), ac = 2.67 (2.10–2.18), M = 0.41 (0.36–0.39), C3F = 0.85 (0.88).

Type specimens. Holotype ♂ (SCAU, No. 121883), CHINA: Muyiji, Ximeng, Yunnan, 1100m, 2.iv.2011, ex tussock, SJ Yan. Paratypes: 1♂, 2♀ (SCAU, Nos 123296–98), CHINA: Yixiang, Puer, Yunnan, 27°12'N, 100°03'E, 1400m, 29.iii.2013, ex tussocks, JJ Gao.

Etymology. A combination of the Latin word “*quadrus*” (= four) + “*foliaceus*” (= folium), referring to the aedeagal basolateral process quadrifurcated distally.

Distribution. China (Yunnan).

***Leucophenga retifoliacea* sp. nov.**

(Figs. 2M, 2N, 2O, 3I, 3J, 8)

Diagnosis. This species can be distinguished from the other species of the *interrupta* group by having the following characters: aedeagus reticulated basally; aedeagal basolateral process broadened, distally with several serrations on dorsal margin (Fig. 8D).

Description. Ocellar triangle yellowish brown, with 2–6 setae above ocellar setae. Frons yellowish brown. Pedicel yellowish brown; first flagellomere yellow. Clypeus yellowish brown. Postpronotal lobe yellow (Fig. 2O). Acrostichal setulae in 11–13 irregular rows (Fig. 2N). Scutellum yellowish brown (Fig. 2N). Wing (Fig. 2M): Costal vein between R_{2+3} and R_{4+5} distally with 6–7 peg-like spinules on ventral surface. Halter slightly brownish apically. Abdominal tergites black (Fig. 3I, J); second mostly yellow; third with a pair of yellow patches submedially on anterior margin, yellow laterally; fourth to fifth each with 2 pairs of yellow patches submedially on anterior margin, yellow laterally; sixth with yellow patch posteriorly in holotype, with a pair of yellow patches submedially in one female paratype. Male terminalia: Epandrium with ca. 12 setae near posterior margin and ventral corner per side (Fig. 8A). Paramere expended and slightly oblong, with distinct wrinkles, lacking sensillum (Fig. 8C).

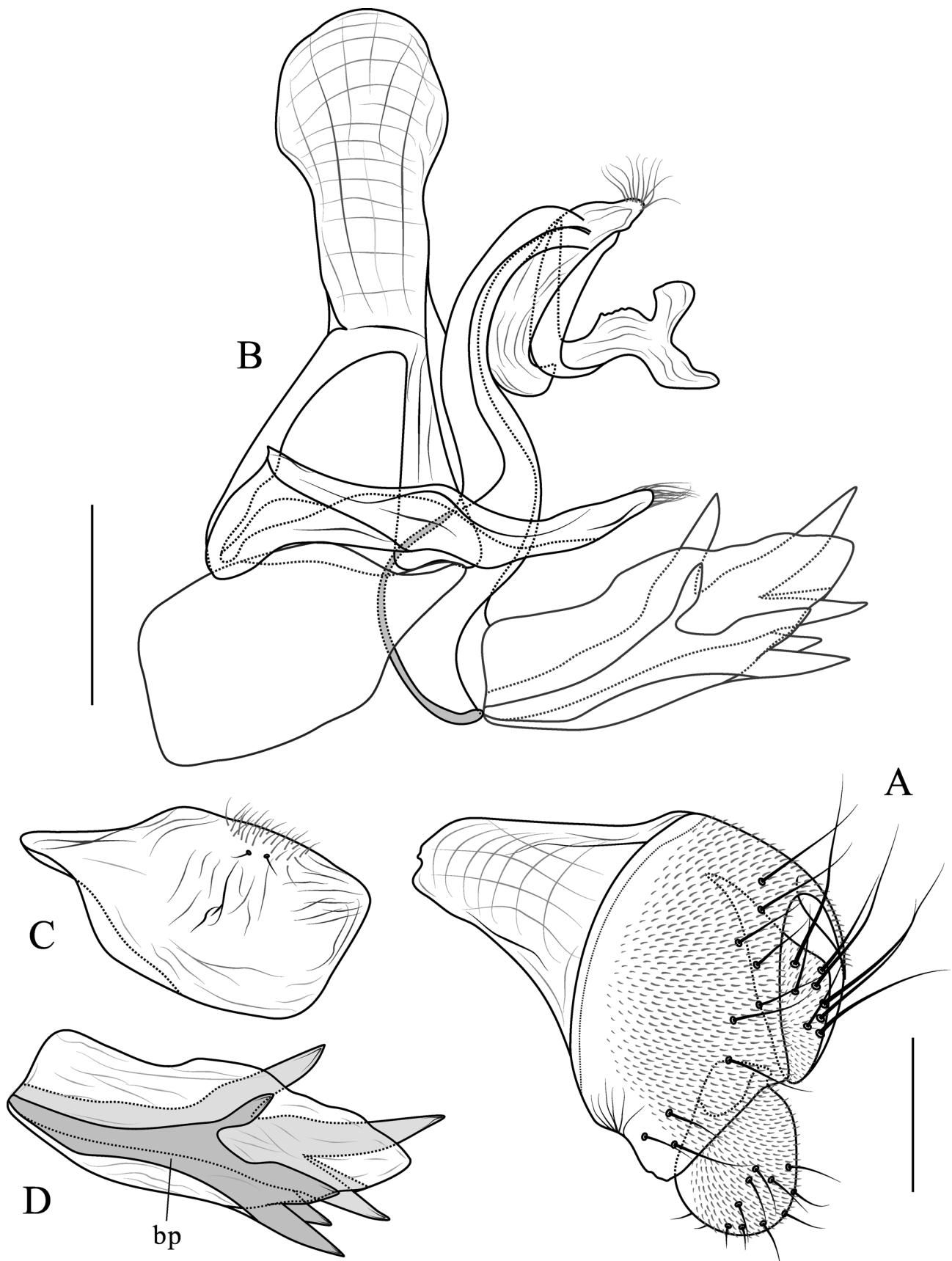


FIGURE 7. *Leucophenga quadrifurcata* sp. nov., male terminalia in lateral view: A. epandrium, surstyli and cercus; B. hypandrium, paramere, gonopods, aedeagus and aedeagal apodeme; C. paramere; D. aedeagus. Scale bars = 0.1 mm.

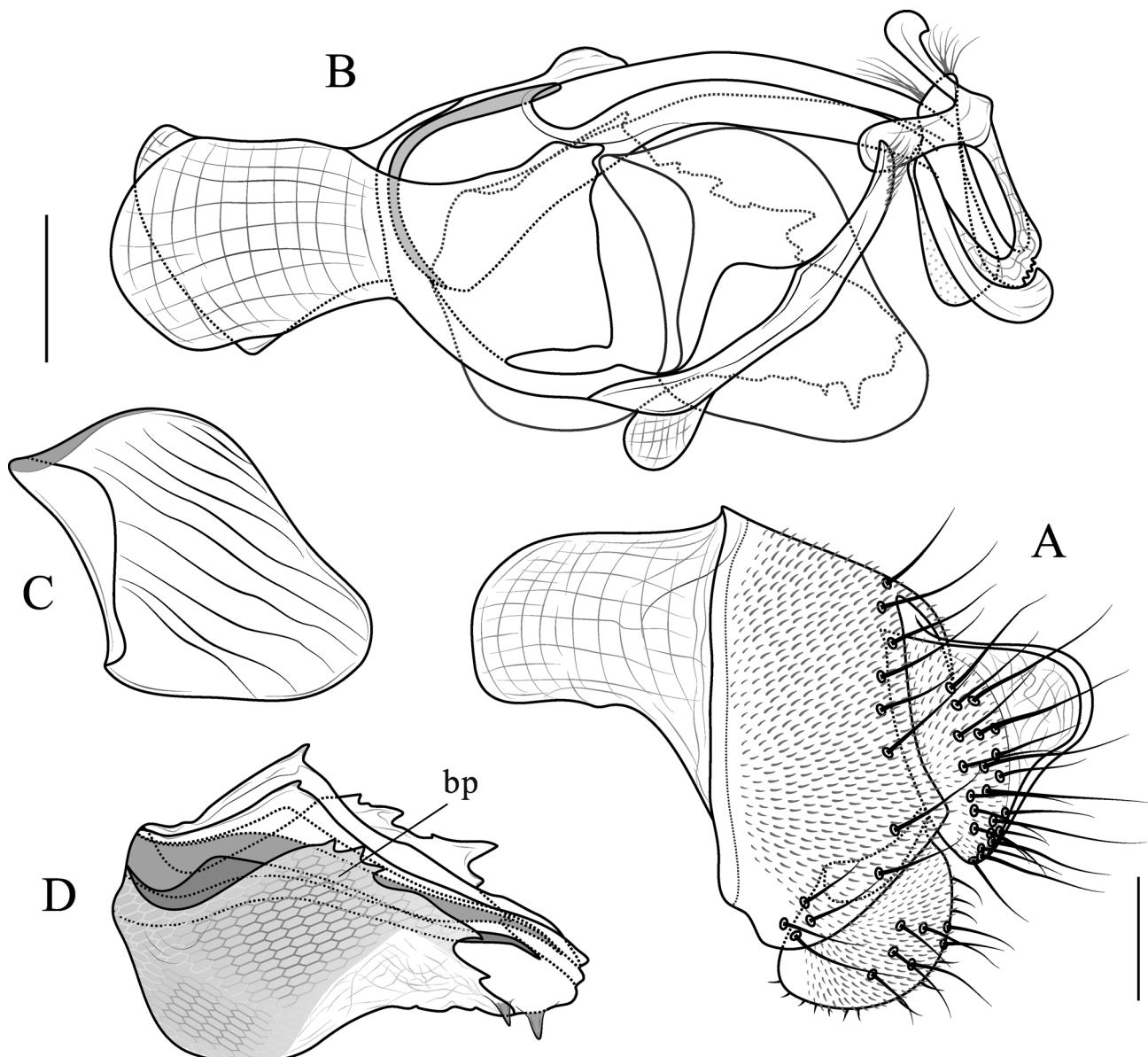


FIGURE 8. *Leucophenga retifoliacea* sp. nov., male terminalia in lateral view: A. epandrium, surstyli and cercus; B. hypandrium, paramere, gonopods, aedeagus and aedeagal apodeme; C. paramere; D. aedeagus. Scale bars = 0.1 mm.

Measurements. BL = 3.67 mm in holotype (range in 1♂ and 1♀ paratypes: 3.60 mm in ♂, 3.73 mm in ♀), THL = 1.93 mm (1.90 mm in ♂, 1.77 mm in ♀), WL = 3.30 mm (2.70 mm in ♂, 3.00 mm in ♀), WW = 1.37 mm (1.17 mm in ♂, 1.47 mm in ♀), arb = 10/4 (8–9/3–4), avd = 0.89 (0.81–0.90), adf = 2.12 (1.88–2.31), flw = 2.00 (2.00–2.13), FW/HW = 0.34 (0.31–0.39), ch/o = 0.03 (0.03–0.06), prorb = 1.27 (0.84), rcorb = 1.03 (0.68), vb = 0.38 (0.43–0.45), dcl = 0.38 (0.38–0.44), presctl = 0.56 (0.47–0.52), sctl = 1.21 (1.15–1.28), sterno = 0.79 (0.69–0.71), orbito = 2.18 (2.20–2.22), dcp = 0.22 (0.22–0.24), sctlp = 1.24 (1.18–1.43), C = 1.89 (2.20–2.41), 4c = 1.20 (1.16–1.23), 4v = 2.07 (1.85–2.03), 5x = 0.90 (0.76–1.00), ac = 2.94 (2.32–2.72), M = 0.41 (0.42–0.50), C3F = 0.75 (0.89–0.98).

Type specimens. Holotype ♂ (SCAU, No. 121884), CHINA: Hesong, Menghai, Yunnan, 1900m, 9.iv.2011, ex tree trunk, ZF Shao. Paratypes: CHINA: 1♀ (SCAU, No. 121885), same data as holotype; 1♀ (SCAU, No. 121886), Menglun, Mengla, Yunnan, 580m, 22.iv.2007, ex tree trunk, HW Chen; 1♂ (SCAU, No. 121887), Yixiang, Puer, Yunnan, 1400m, 13.v.2012, JJ Gao.

Etymology. A combination of the Latin word “*rete*” (= reticulated) + “*foliaceus*” (= folium), referring to the aedeagus reticulated.

Distribution. China (Yunnan).

Key to species of the *interrupta* group

1. Wing with broad, dark longitudinal stripe along anterodistal portion (Fig. 2A, D, G, J, M); paramere with wrinkles (Figs. 4D, 5C, 6C, 7C, 8C); aedeagus with sclerotized processes basolaterally (Figs. 4E, 5D, 6D, 7D, 8D) *interrupta* group ... 2
- Wing without broad, dark longitudinal stripe along anterodistal portion; paramere lacking wrinkles; aedeagus without processes basolaterally other *Leucophenga* species
2. Aedeagus slightly acute apically, with a pair of strong and acute processes subbasally (Fig. 4E) *L. interrupta*
- Aedeagus neither acute apically nor acute processes subbasally 3
3. Parameres with 3 serrated processes subapically (Fig. 53C); aedeagus with 1 regular row (ca. 5) of serrated processes distally, the basolateral process shorter than half of aedeagal length (Fig. 53D) *L. neointerrupta*
- Parameres lacking serrated processes subapically (Figs. 6C, 7C, 8C); aedeagus lacking regular row of serrated processes distally, the basolateral process nearly as long as aedeagus (Figs. 6D, 7D, 8D) 4
4. Aedeagal basolateral process broadened, distally with several serrated processes on dorsal portion (Fig. 8D)
..... *L. retifoliacea*
- Aedeagal basolateral process slender, distally lacking several serrated processes (Figs. 6D, 7D) 5
5. Paramere curved to cucullate, subbasally with several serrated processes on inner margin (Fig. 6C); aedeagus not reticulated, the basal process slender, longer than aedeagus, slightly curved submedially, and slightly bifurcate apically, lacking serrated processes (Fig. 6D) *L. bifurcata*
- Paramere expended and slightly oblong, lacking serrated processes and sensillum (Fig. 7C); aedeagus reticulated basally, the basolateral process broadened, distally with several serrated processes on dorsal portion (Fig. 7D) *L. quadrifurcata*

Discussion

The *ornata* group was established based on their distinct character on the wing, i.e. the presence of dark patch(es) (Bächli 1971), and this classification has been followed in subsequent studies (Okada 1990; Fartyal *et al.* 2005). Majority of this group has pubescence on the distal portion of aedeagus (Okada 1968, 1990; Bächli 1971; Chen & Aotsuka 2003), but there are exceptions. Recently, we classified 15 new species lack pubescence on aedeagus, and they formed a high supported monophyletic group (Huang *et al.* 2013).

Within the species lack of pubescence on aedeagus, *L. interrupta* and *L. neointerrupta* are distinctive in having the wings clear on r-m and dm-cu crossveins, with broad, dark longitudinal stripe along anterodistal portion, and the aedeagus with strong processes basolaterally. These distinctness imply that *L. interrupta* and *L. neointerrupta* are probably compose a new species group (Huang *et al.* 2013). In present study, we described three new species (*L. retifoliacea*, *L. bifurcata* and *L. quadrifurcata*), all of them shared the similar aedeagus structures with *L. interrupta* and *L. neointerrupta*. However, the wings of *L. quadrifurcata* are clouded not clear on r-m and dm-cu crossveins. Herein, as the priority of aedeagus structure in the species delimitation of flies, we proposed a new species group, the *interrupta* group, including with the above mentioned five species. Furthermore, to investigate the monophyly of the *interrupta* group, we employed four representative species of the *ornata* group in the molecular phylogenetic analyses, two of them have pubescence on the distal portion of aedeagus (*L. digmasoma* and *L. quadripuncata*), the other two species are the pubescence lacking ones (*L. ornata* and *L. saigusai*). The results of the phylogenetic analyses were robustly support the monophyly of the *interrupta* group, but the relationships among the *interrupta* species were not resolved. Moreover, the single sample of *L. quadrifurcata* was clustered with samples of *L. neointerrupta* in the Bayesian tree, which challenged the monophyly of the two species. As lacking the molecular information of other geographical samples of *L. quadrifurcata*, the real relationship between these two species are not unambiguous.

In the further study, more species and molecular evidence should be involved into the analyses, which will draw a more robust conclusion on the establishment of the *interrupta* group, and also provide an opportunity to uncover the phylogenetic relationship within these *interrupta* species thoroughly.

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