

Review of the *Lispe tentaculata*-group (Diptera: Muscidae) in China, with one new synonym

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ABSTRACT

The Chinese fauna of *Lispe tentaculata*-group is reviewed. A new synonym for *L. sericipalpis* Stein, 1904 is established: *L. fanjingshanensis* Wei, 2006 n. syn. Taxonomic status of the suspected species *L. alpinicola* Wu & Fan, 1981 is discussed. The keys to males and females of all five species within the group in China are established. Morphological details of both sexes are illustrated using light microscopy photographs, scanning electronic microscopy images and line drawings, of which male and female terminalia are summarized as novel characteristics for diagnosis. The group is divided into two subgroups (*L. tentaculata*-subgroup and *L. orientalis*-subgroup) based on two characteristics (first fore tarsomere and phallus). To fill the gap of molecular research in *Lispe*, we construct a phylogenetic tree within the genus focusing on *L. tentaculata*-group, which verifies the division of two subgroups.

RÉSUMÉ

Révision du groupe de *Lispe tentaculata* (Diptera: Muscidae) de Chine, avec établissement d'un nouveau synonyme.

Les espèces chinoises du groupe de *Lispe tentaculata* (Muscidae) sont révisées. Un nouveau synonyme pour *L. sericipalpis* Stein, 1904 est établi: *L. fanjingshanensis* Wei, 2006 n. syn. Le statut taxonomique de l'espèce supposée *L. alpinicola* We & Fan, 1981 est discuté. Des clés sont établies pour les mâles et les femelles des cinq espèces du groupe présentes en Chine. Les détails morphologiques des deux sexes sont illustrés avec des photographies en microscopie optique, des images en microscopie électronique à balayage, et des dessins au trait; notamment, les terminalia mâles et femelles sont présentés comme de nouveaux caractères de diagnose. Le groupe est divisé en deux sous-groupes (les sous groupes *L. tentaculata* et *L. orientalis*) sur la base de deux caractères (premier tarsomère antérieur et phallus). Pour faire avancer les recherches moléculaires sur *Lispe*, nous avons reconstruit un arbre phylogénétique partiel du genre en nous focalisant sur le groupe *L. tentaculata*: cet arbre confirme la séparation en deux sous-groupes.

KEY WORDS
Muscidae,
Lispe tentaculata-group,
review,
new subgroups,
phylogenetic relationships,
China,
new synonym.

MOTS CLÉS
Muscidae,
groupe *Lispe tentaculata*,
révision,
sous-groupes nouveaux,
relations phylogénétiques,
Chine,
synonymie nouvelle.

INTRODUCTION

Numerous dipteran taxonomists have contributed their efforts to *Lispe* in the past, by which they built up the fundamental taxonomy system of this genus (Kowarz 1892; Stein 1897; Becker 1903, 1904; Aldrich 1913; Malloch 1922; Snyder 1954; Hennig 1960; Pont 1977, 1986; Lopes 1992). In China, systematic investigations on this genus were mainly conducted by Wu (1940), Fan (1965, 1992), Xue & Chao (1998) and Xue & Zhang (2005). Most recently, Vikhrev (2014, 2015) has took comprehensive taxonomic notes on the worldwide fauna of nine species-groups and two species-complexes.

Concerning the *Lispe tentaculata*-group, it was initially proposed by Synder (1954), consisting of three species in the Nearctic Region: *L. patellata* Aldrich, 1913; *L. sociabilis* Loew, 1861 and *L. tentaculata* (DeGeer, 1776). In the study of Palearctic diptera, Hennig (1960) redefined this species-group and expanded it with four additional species: *L. consanguinea* Loew, 1858; *L. draperi* Séguy, 1933; *L. orientalis* Wiedemann, 1824 and *L. quaerens* Villeneuve, 1936. Pont (1986) considered *L. draperi* as a synonym for *L. tentaculata* and Vikhrev (2011) re-evaluated it as a valid species. Meanwhile, Vikhrev (2011) synonymized *L. quaerens* under *L. sericipalpis* Stein, 1904. Besides, two more species was added to this group after Hennig (1960): *L. alpinicola* from Lhasa, Xizang, China by Zhong, Wu & Fan (1981) and *L. emdeni* from Jaipur, Rajasthan, India by Vikhrev (2012). The validity of *L. alpinicola* is doubted by Vikhrev (2011, 2014) but we hold a different view and consider it an independent species (see the remarks below). In sum, we reviewed the five species of *L. tentaculata*-group in China.

Well-defined as it is, *L. tentaculata*-group remains to be treated more explicitly and carefully since considerable morphological differences still exist between the species within this group.

Though work on *Lispe* morphology is quite abundant, rare molecular research is available for us to explore the true relationship within this genus. As mitochondrial DNA sequences (12S, 16S, COI and Cytb) and nuclear genome sequences (28S, Efla and a fragment of the carbamoly phosphate synthetase (CPS) region of the CAD gene) are becoming increasingly useful to reconstruct the relationships within the Muscoidea, especially the Muscidae (Kutty *et al.*, 2008). Here we present the phylogeny of four species from *L. tentaculata*-group and species from other *Lispe* groups based on the seven sequences mentioned above.

MATERIAL AND METHODS

TAXONOMY AND MORPHOLOGY

All specimens studied are deposited in MBFU except that the type specimens of *L. alpinicola* Zhong, Wu & Fan and *L. tienmuensis* Fan are deposited in SEMCAS. Some specimens of *L. tentaculata*-group are stored in MNHN, which are listed beneath.

Distribution data in China is taken from Fan (1992) with additional records of collection. For distribution information around the world, it refers to Vikhrev (2014). Well-acknowledged synonyms and quotations are omitted here as they are available in Hennig (1960), Pont (1977, 1986) and Vikhrev (2014).

Light microscopy photographs were taken with a Canon 550D camera mounted on an Olympus SZX16 stereomicroscope except that photos of *L. tienmuensis* with low quality were taken years ago with poor equipments. Scanning electronic Microscopy (SEM) photographs were taken with a HITACHI S34Q scanning electron microscope (Hitachi Corp., Tokyo, Japan) at the Microscopy Core Facility, Biological Technology Center, Beijing Forestry University.

Morphological terminology follows McAlpine (1981).

Molecular phylogeny

Eight *Lispe* species were selected, including four species from *L. tentaculata*-group (*L. consanguinea*, *L. orientalis*, *L. sericipalpis* and *L. tentaculata*) which represent most characters of this species group. As *Limnophora simulans* Stein, 1913 and *Coenosia semifumosa* Stein, 1914 are close to *Lispe* (according to Kutty *et al.* 2014), they are selected to root the tree.

Pinned specimens are used for molecular experiment in this paper (for collecting date and location of the specimens, see Table 3). DNA extraction for all species is carried out utilizing phenol-chloroform extraction as described in Zhang *et al.* (2016), except that genomic DNA of *L. apicalis* Mik, 1869 is extracted using QIAGEN DNeasy Blood and Tissue Kit following the manual. Seven genes are amplified including four mitochondrial genes (12S, 16S, COI and Cytb) and three nuclear genes (28S, Efla and CAD) using primers (Table 2). The reaction system of PCR (polymerase chain reaction) follows Zhang *et al.* (2016) and the PCR procedure for amplifications are shown in Kutty *et al.* (2010). PCR products are sequenced bidirectionally by BGI in the same way as Zhang *et al.* (2016).

All sequences are edited and assembled with SeqMan (DNASTar, Steve ShearDown, 1998-2001 version, DNASTAR Inc., USA), then aligned using MAFFT (version 7, Katoh *et al.* 2013). SequenceMatrix (v1.7.9, Meier *et al.* 2006) is employed to concatenate individual alignments. And PartitionFinder (v1.1.1, Lanfear *et al.* 2012) is used to evaluate the best partition: schemes = greedy, branch lengths = unlinked, model selection = BIC. The best partitioning scheme for dataset and the respective best models are determined (Table 1). Phylogenetic analysis is performed using maximum likelihood (ML) methods and Bayesian as described in Zhang *et al.* (2016).

ABBREVIATIONS

Leg chaetotaxy

<i>a</i>	anterior seta(e);
<i>d</i>	dorsal seta(e);
<i>p</i>	posterior seta(e);
<i>v</i>	ventral seta(e);
<i>ad</i>	anterodorsal seta(e);
<i>av</i>	anteroventral seta(e);
<i>pd</i>	posterodorsal seta(e);
<i>pv</i>	posteroventral seta(e).

TABLE 1. — Information of species used for phylogenetic analyses and GenBank Accession Numbers. **n.a.**, no data; *, new sequences generated for this study.

Species	Species collecting date	Collecting location (province of China)	COI	12S	16S	28S	Cytb	Ef1a	CAD
<i>Coenosia semifumosa</i> Stein, 1914	n.a.	n.a.	KJ510613	KJ476265	KJ476301	KJ476336	KJ510547	KJ510586	n.a.
<i>Limnophora simulans</i> Stein, 1913	n.a.	n.a.	KJ510626	KJ476278	KJ476313	KJ476348	KJ510559	n.a.	n.a.
<i>Lispe apicalis</i> Mik, 1869	2014	Xinjiang	KX856023*	KX856037*	KX856045*	KX856040*	KX856017*	KX856031*	KX856022*
<i>Lispe consanguinea</i> Loew, 1858	2013	Xinjiang	KX856024*	KX856035*	KX856048*	n.a.	KX856016*	n.a.	KX856021*
<i>Lispe leucospila</i> Hennig, 1960	n.a.	n.a.	KP161694	n.a.	JN226652	n.a.	n.a.	n.a.	KP161827
<i>Lispe neimongola</i> Tian & Ma, 2000	2009	Xinjiang	KX856028*	KX856034*	KX856046*	KX856041*	KX856014*	KX856030*	KX856020*
<i>Lispe nivalis</i> Wiedemann, 1830	n.a.	n.a.	KJ510628	n.a.	KJ476315	KJ476350	KJ510561	n.a.	n.a.
<i>Lispe orientalis</i> Wiedemann, 1824	2013	Hubei	KX856026*	KX856036*	KX856043*	n.a.	KX856013*	n.a.	n.a.
<i>Lispe sericipalpis</i> Stein, 1904	2013	Hubei	KX856025*	KX856033*	KX856047*	KX856042*	KX856012*	KX856029*	KX856019*
<i>Lispe tentaculata</i> (DeGeer, 1776)	2013	Liaoning	KX856027*	KX856038*	KX856044*	KX856039*	KX856015*	KX856032*	KX856018*

TABLE 2. — Primers sequences used for PCR amplification and sequencing.

Gene	Primer Sequence	References
12S	AAA CTA GGA TTA GAT ACC CTA TTA CCC TGA TAC ACA AGG TA	Kutty <i>et al.</i> 2014
16S	CGC CTG TTT AAC AAA AAC AT TGA ACT CAG ATC ATG TAA GAA A	
28S	AGC GGA GGA AAA GAA AC GCT ATC CTG AGG GAA ACT TCG G	
COI	TAC AAT TTA TCG CCT AAA CTT CAG CC CCC GGT AAA ATT AAA ATA TAA ACT TC CCA CAT TTA TTT TGA TTT TTT GG TCC AAT GCA CTA ATC TGC CAT ATTA GGT CAA CAA ATC ATA AAG ATA TTG G TAA ACT TCA GGG TGA CCA AAA AAT CA	Folmer <i>et al.</i> 1994
Cytb	TAT GTT TTA CCT TGA GGA CAA ATA TC AAA TTC TAT CTT ATG TTT TCA AAA C	Kutty <i>et al.</i> 2014
Ef1a	CAG GAA ACA GCT ATG ACC GCT GAG CGY GAR CGT GGT ATC AC ACA GCV ACK GTY TGY CTC ATR TC	
CAD	GGD GTN ACN GCN TGY TTY GAR CC TTN GGN AGY TGN CCN CCC AT	Moulton & Wiegmann 2003

Anatomy

ap appendage;
cer cerci;
ep epiproct;
hp hypoproct;
ster sternite;
ter tergite.

Institutions

SEMCAS Shanghai Entomological Museum, Chinese Academy of Science, Shanghai;

MBFU
 MNHN

Museum of Beijing Forestry University, Beijing;
 Muséum national d'Histoire naturelle, Paris.

Molecular phylogeny

12S 12S ribosomal RNA gene;
 16S 16S ribosomal RNA gene;
 COI cytochrome oxidase subunit I gene;
 Cytb cytochrome b gene;
 28S 28S ribosomal RNA gene;
 Ef1a Elongation factor 1 alpha.

TABLE 3. — Best partitioning scheme for the dataset and the respective models.

Partitions	Best Model
12S, 16S	GTR+G
28S, CAD, Efla	HKY+G
COI, Cytb	GTR+I

SYSTEMATIC ACCOUNT

Lispe tentaculata-group

Lispe tentaculata-group Snyder, 1954: 6. — Hennig 1960: 408. — Xue & Zhang 2005: 122. — Vikhrev 2011: 59; 2014: 153.

DIAGNOSIS. — The *Lispe tentaculata*-group is distinguished from other species-group of this genus by the following characters (modified from Snyder 1954, Hennig 1960 and Vikhrev 2011, 2014): arista plumose, palpi abruptly expanded in apical half; meron setulose above hind coxa and the following leg chaetotaxy: fore tibia mostly without *p* setae (only present in *L. alpinicola* and some aberrant *L. tentaculata*); mid tibia mostly with 1 *p* (3 *p* in *L. alpinicola*), hind tibia with 1 weak *pd* and 1 strong *ad*.

Herein, we update the diagnosis with more detailed genital characters of both sexes (some male genital characters have been mentioned by Snyder 1954) and bring up the distinctive characteristics of two subgroups:

Male: fifth sternite with a median, anteriorly directed process in addition to the short and apically semi-membranous lateral processes; cercal plate with an inverted U- to V-shaped incision on each side (Fig. 1); gonopod degenerated and fused with hypandrium into a projection (Figs 6; 7); phallus non-membranous.

Female: abdominal tergites 6 and 7 sometimes divided medially (*L. consanguinea* and *L. tentaculata*), abdominal sternite 6 sometimes degraded into 2 small semi-transparent setulose sclerites (*L. consanguinea* and *L. tentaculata*), abdominal tergite 7 fused with sternite 7, abdominal tergite 8 partly fused with sternite 8, epiproct divided medially into 2 setulose sclerites, cerci with appendage (Figs 3; 4).

— *L. tentaculata*-subgroup: first fore tarsomere modified (except *L. sociabilis*); phallus straight.

— *L. orientalis*-subgroup: first fore tarsomere regular; phallus with a vertically bended diatal part.

— Chinese Species:

— *L. tentaculata*-subgroup: *L. alpinicola* Zhong, Wu & Fan, 1981; *L. consanguinea* Loew, 1858 and *L. tentaculata* (DeGeer, 1776).

— *L. orientalis*-subgroup: *L. orientalis* Wiedemann, 1824 and *L. sericipalpis* Stein, 1904.

KEY TO MALES OF *LISPE TENTACULATA*-GROUP IN CHINA (characters of *L. alpinicola* adapted from Zhong *et al.* 1981)

1. First fore tarsomere with a long finger-like protuberance, end of which blunt and with short black thorns 2
— First fore tarsomere without finger-like protuberance 4
2. Mid and hind tibiae dark gray, first hind tarsi slightly sunken in the middle of ventral surface, slightly swollen in basal, with dense black brush-like setae on ventral 3
— Mid and hind tibiae mostly or entirely yellowish-brown, first hind tarsomere straight and tenuous, not sunken in the middle of ventral surface and not swollen basally, without black brush-like setae on ventral surface
..... *Lispe consanguinea* Loew, 1858
3. Palpi yellow, apex broad and round; fore tibia without median *p*, mid tibia with 1 *pd*, length of finger-like protuberance equal to ½ of second fore tarsomere; only abdominal tergite 3 with dark stripe
..... *Lispe tentaculata* (DeGeer, 1776)
— Palpi black, apex slightly narrow; fore tibia with 1 median *p*, mid tibia with 3 *pd*, length of finger-like protuberance longer than ½ of second fore tarsomere; abdominal tergites 3-5 with dark stripes
..... *Lispe alpinicola* Zhong, Wu & Fan, 1981
4. Palpi brownish yellow; hind femur with complete *av* and *pv* rows *Lispe orientalis* Wiedemann, 1824
— Palpi blackish brown; hind femur without complete *av* and *pv* rows *Lispe sericipalpis* Stein, 1904

KEY TO FEMALES OF *LISPE TENTACULATA*-GROUP IN CHINA (characters of *L. alpinicola* adapted from Zhong *et al.* 1981)

1. Palpi dark brown; hind femur only with 4 or 5 fine *av* on apical half *Lispe sericipalpis* Stein, 1904
— Palpi yellow 2
2. Hind femur with complete *av* and *pv* row *Lispe orientalis* Wiedemann, 1824
— Hind femur without *pv* row, without *av* or at most on apical ⅔ with 1 or 2 *av* 3
3. Postesutural part of scutum without velutinous patch; mid and hind tibiae yellowish-brown; hind femur without *av* on apical ⅔ *Lispe consanguinea* Loew, 1858
— Postsutural part of scutum with yellowish-brown to blackish brown velutinous patch between the 2 *dc* rows; mid hind tibia dark grey; hind femur with 1 or 2 long *av* on apical ⅔ 4
4. Posterior part of prescutal sulcus with dark or light yellowish-brown velutinous patches
..... *Lispe tentaculata* (DeGeer, 1776)
— Posterior part of prescutal sulcus with brown to black brown velutinous patches
..... *Lispe alpinicola* Zhong, Wu & Fan, 1981



FIG. 1. — Male terminalia of *Lispe tentaculata*-group, ventral view: **A**, *L. consanguinea* Loew, 1858; **B**, *L. orientalis* Wiedemann, 1824; **C**, *L. sericipalpis* Stein, 1904; **D**, *L. tentaculata* (De Geer, 1776). Scale bars: 0.25 mm.

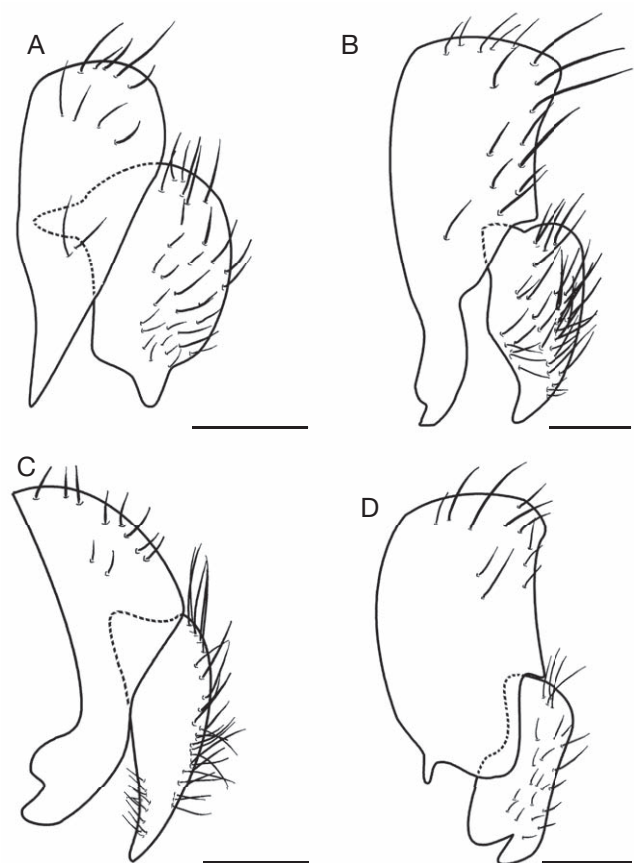


FIG. 2. — Male terminalia of *Lispe tentaculata*-group, lateral view: **A**, *L. consanguinea* Loew, 1858; **B**, *L. orientalis* Wiedemann, 1824; **C**, *L. sericipalpis* Stein, 1904; **D**, *L. tentaculata* (De Geer, 1776). Scale bars: 0.25 mm.

CATALOGUE OF *LISPE TENTACULATA*-GROUP IN CHINA

Lispe alpinicola Zhong, Wu & Fan, 1981 (Fig. 11A)

Lispe alpinicola Zhong, Wu & Fan, 1981: 245. — Fan 1992: 368. — Xue & Chao 1998: 994. — Xue & Zhang 2005: 123. — Vikhrev 2011: 63.

DISTRIBUTION IN CHINA. — Xizang (type locality: Lhasa).

REMARKS

Vikhrev (2011) doubted that this species might be a synonym for *L. tentaculata* and later Vikhrev (2014) proposed this synonym for the reason that the presence of additional *p* on fore and mid tarsi of *L. alpinicola* also presents in several specimens of *L. tentaculata* in the collection of Zoological Museum of the Moscow University. However, no other characters were compared and discussed between these two species in his works. In order to clarify the status of *L. alpinicola*, we tried to find the types in SEMCAS but failed, which is likely to be lost. Without examining the types, we can't draw a convincing conclusion about its validity. Here we present the original text (Zhong *et al.* 1981, in Chinese and with an English abstract, not cited in Vikhrev's works): "This species is closely related to the Holarctic species *Lispe tentaculata*, from which it differs in having the palpi black, the fore tibia with a median seta on posterior surface, the process of the fore metatarsus rather long, the mid tibia with 3 *pd* etc." Also, in Zhong, Wu & Fan's

remarks, females are distinguished from the color of velutinous patches at postesutural part of scutum. According to the original description, we separate these two species in the keys above.

Another evidence confirms our opinion about the validity of *L. alpinicola* is its difference in male phallus compared with other related species. Their phallus images are placed together for comparison (Fig. 11) especially concentrated on: 1) relative length of aedeagal apodeme and hypandrium; 2) relative length of paramere and phallus; and 3) relative size of submedian membranous part which only exists in *L. alpinicola*.

Lispe consanguinea Loew, 1858 (Figs 1A; 2A; 3A, C, E; 5A, B, C)

Lispe consanguinea Loew, 1858: 8.

MATERIAL EXAMINED. — 3 ♂ and 1 ♀, Liaoning: Dalian, Xiajiahe, 7.VIII.2009, Coll. R. Bi; 1 ♂, Xinjiang: Qiakuertu, 27.VIII.2009, Coll. D. Zhang; 1 ♂ and 1 ♀, Shandong: Yantai, 1.VII.2009, Coll. Grade 2008 of Biology; 2 ♀, Shandong: Weifang, 16.VII.2012, Coll. X. H. Liu; 3 ♂ and 3 ♀, Yunnan: Baoshan, 11.VII.2013, Coll. J. Na; 3 ♂ and 3 ♀, Xinjiang: Tekesi, 22.VIII.2013, Coll. M. Zhang; 11 ♂ and 6 ♀, Liaoning: Benxi, 26.VIII.2013, Coll. Y. T. Guan. MNHN: 1 ♂, Shandong: Weifang, 16.VIII.2009, Coll. D. Zhang; 1 ♂, Xinjiang: Kalamaili, 14.VIII.2009, Coll. D. Zhang.

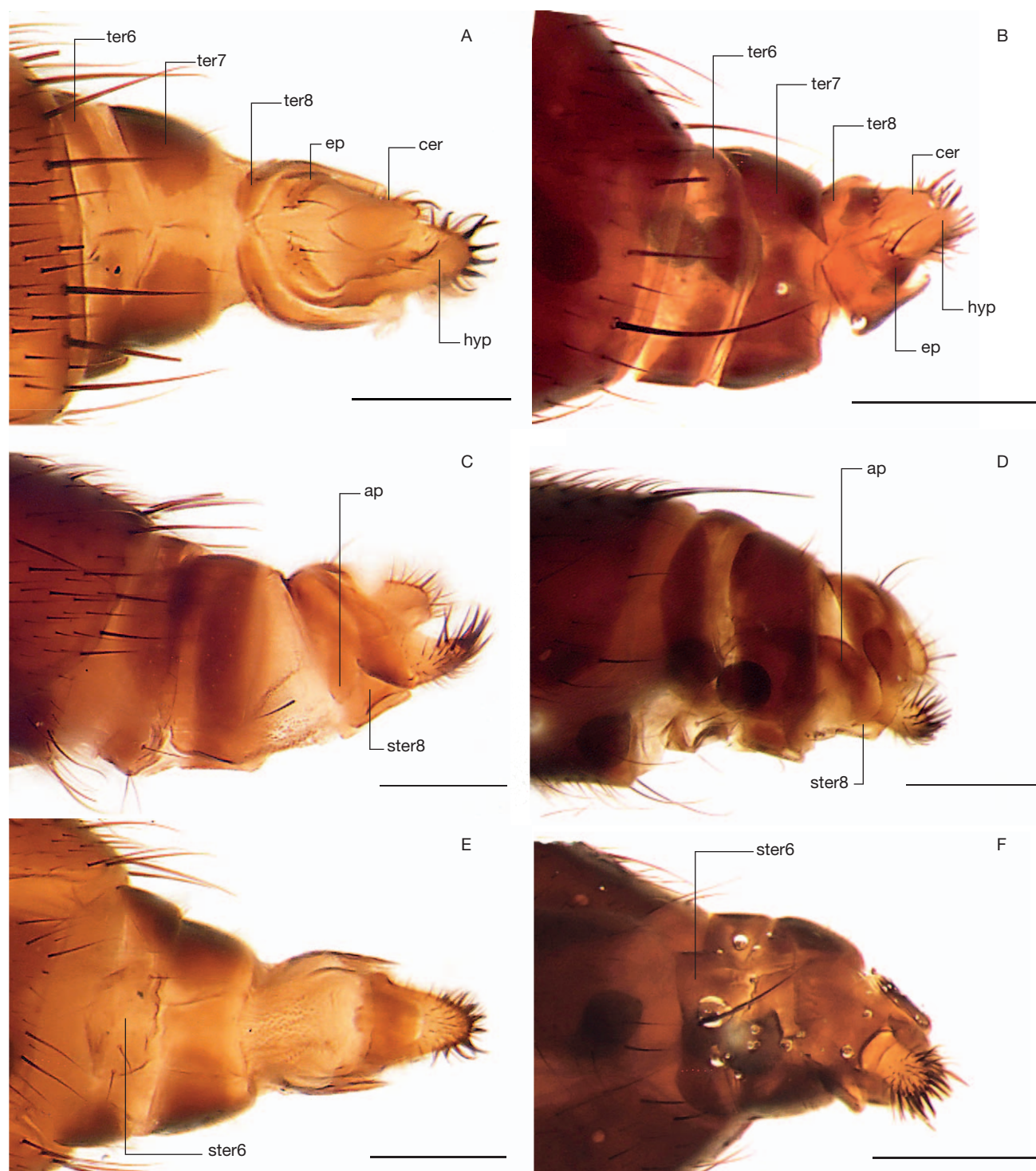


FIG. 3. — Female abdomen and terminalia of *Lispe consanguinea* Loew, 1858 (A, C, E) and *L. orientalis* Wiedemann, 1824 (B, D, F). A, B, dorsal view; C, D, lateral view; E, F, ventral view. Scale bars: 0.50 mm. Abbreviations: see Material and methods.

DISTRIBUTION IN CHINA. — Beijing, Gansu, Hebei, Heilongjiang, Jilin, Liaoning, Neimenggu, Shaanxi, Shandong, Shanxi, Xinjiang, Yunnan.

REMARKS

The finger-like protuberances on fore tarsi of *L. consanguinea* and *L. tentaculata* look identical by eyes. However, SEM images

of the structure reveal interspecific difference concerning the blunt end of the protuberances (Fig. 5): *L. consanguinea* possesses less thorns (Fig. 5C) while *L. tentaculata* has thicker and bended thorns (Fig. 5F). More to mention, all these thorns are bifurcate except the middle one in both species (Fig. 5C, F).

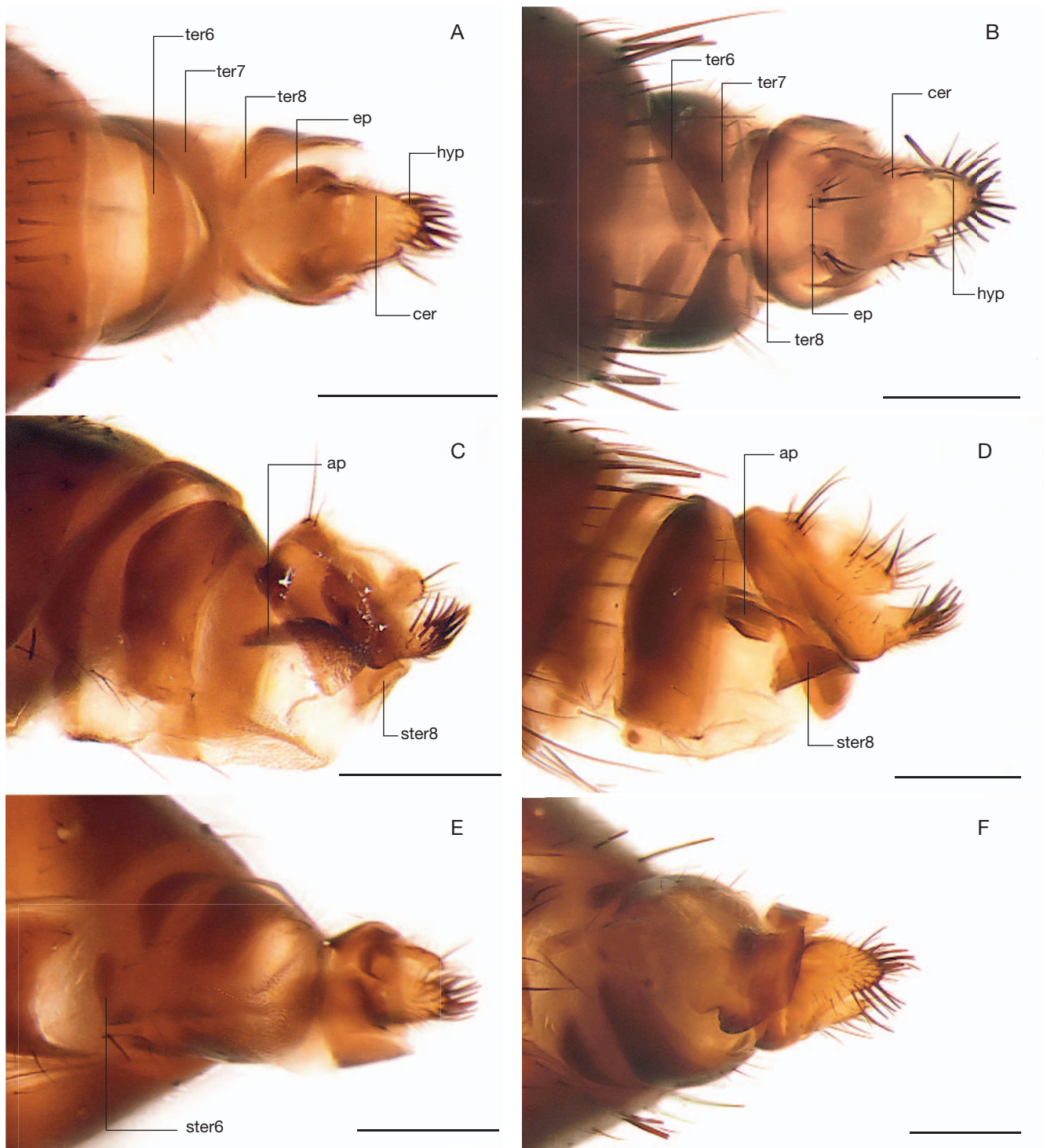


FIG. 4. — Female abdomen and terminalia of *Lispe sericpalpis* Stein, 1904 (A, C, E) and *L. tentaculata* (De Geer, 1776) (B, D, F). A, B, dorsal views; C, D, lateral views; E, F, ventral view. Scale bars: 0.50 mm. Abbreviations: see Material and methods.

Lispe orientalis Wiedemann, 1824
(Figs 1B; 2B; 3B, D, F; 6A, C, E; 7A, B)

Lispe orientalis Wiedemann, 1824: 51.

MATERIAL EXAMINED. — 1 ♂, Beijing: Mt. Song, 31.V.2009, Coll. by D. Zhang; 9 ♂ and 2 ♀, Beijing: Mt. Song, 1.VI.2009, Coll. D. Zhang; 5 ♂ and 3 ♀, Beijing: The Old Summer Palace, 10.V.2009, Coll. D.P. Liu, R. Bi and J.H. Chen; 11 ♂ and 5 ♀, Yunnan: Ba-

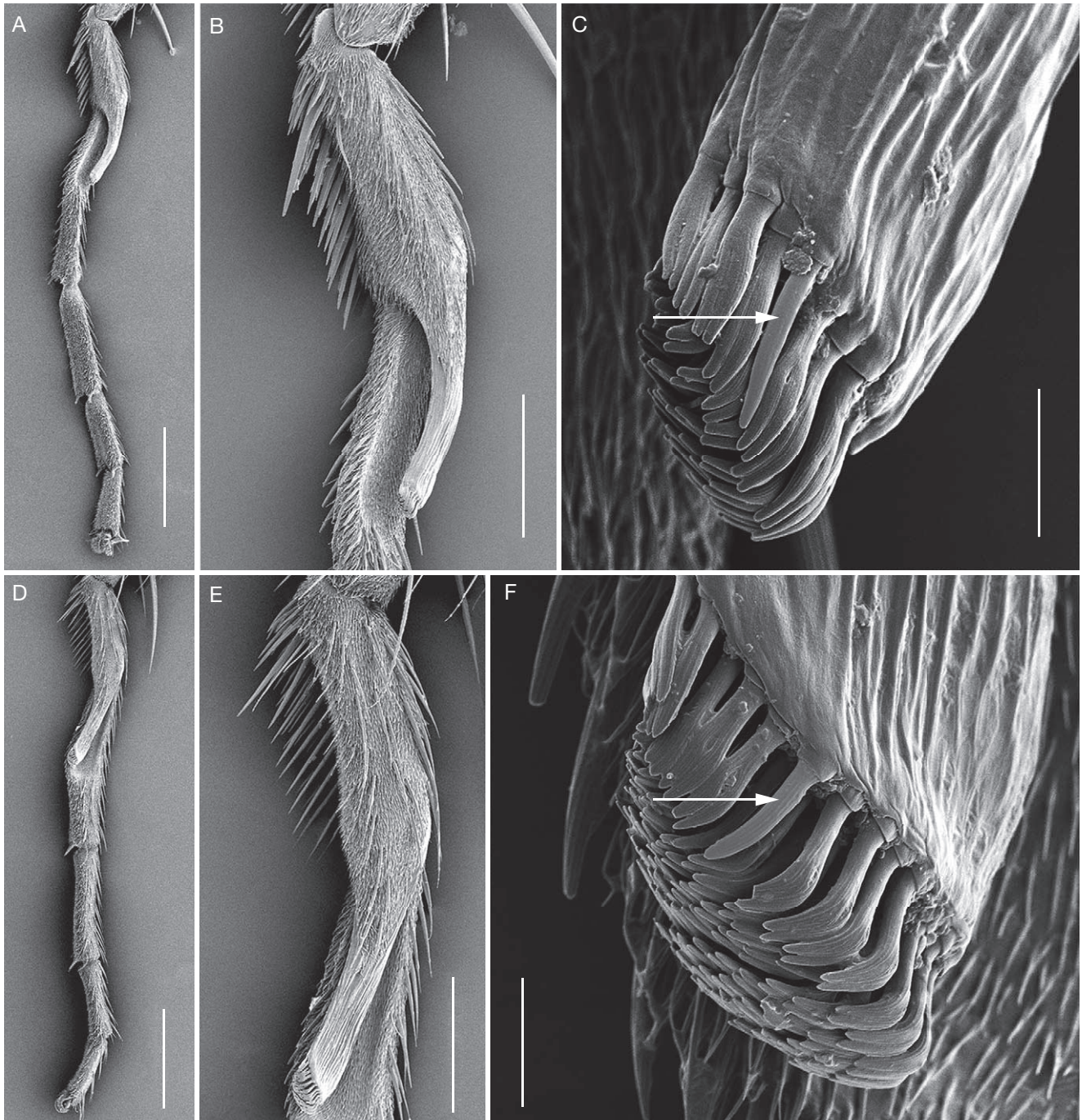


FIG. 5. — SEM images of finger-like protuberance on first tarsi of: *Lispe consanguinea* Loew, 1858 (A-C) and *L. tentaculata* (De Geer, 1776) (D-F), ventral views: A, D, fore tarsi; B, E, finger-like protuberances; C, F, blunt apex of the protuberance. Scale bars: A, D, 400 µm; B, E, 200 µm; C, F, 20 µm. Arrows: the only unbifurcate middle thorn on the blunt end of the finger-like protuberance.

oshan, 14.VII.2013, Coll. J. Na; 10 ♂ and 5 ♀, Hunan: Yichang, 7-22.VII.2013, Coll. M. Zhang.

DISTRIBUTION IN CHINA. — Anhui, Beijing, Fujian, Gansu, Guangdong, Guangxi, Guizhou, Hainan, Hebei, Hubei, Hunan, Jilin, Jiangsu, Liaoning, Shanghai, Shaanxi, Sichuan, Taiwan, Zhejiang, Yunnan.

Lispe sericipalpis Stein, 1904
(Figs 1C; 2C; 4A, C, E; 6B, D, F; 7C, D; 8-10)

Lispe sericipalpis Stein, 1904: 110.

Lispe quaerens Villeneuve, 1936: 157.

Lispe tienmuensis Fan, 1974: 94. — Pont 1986: 191.

Lispe fanjingshanensis Wei, 2006: 511 (n. syn.)

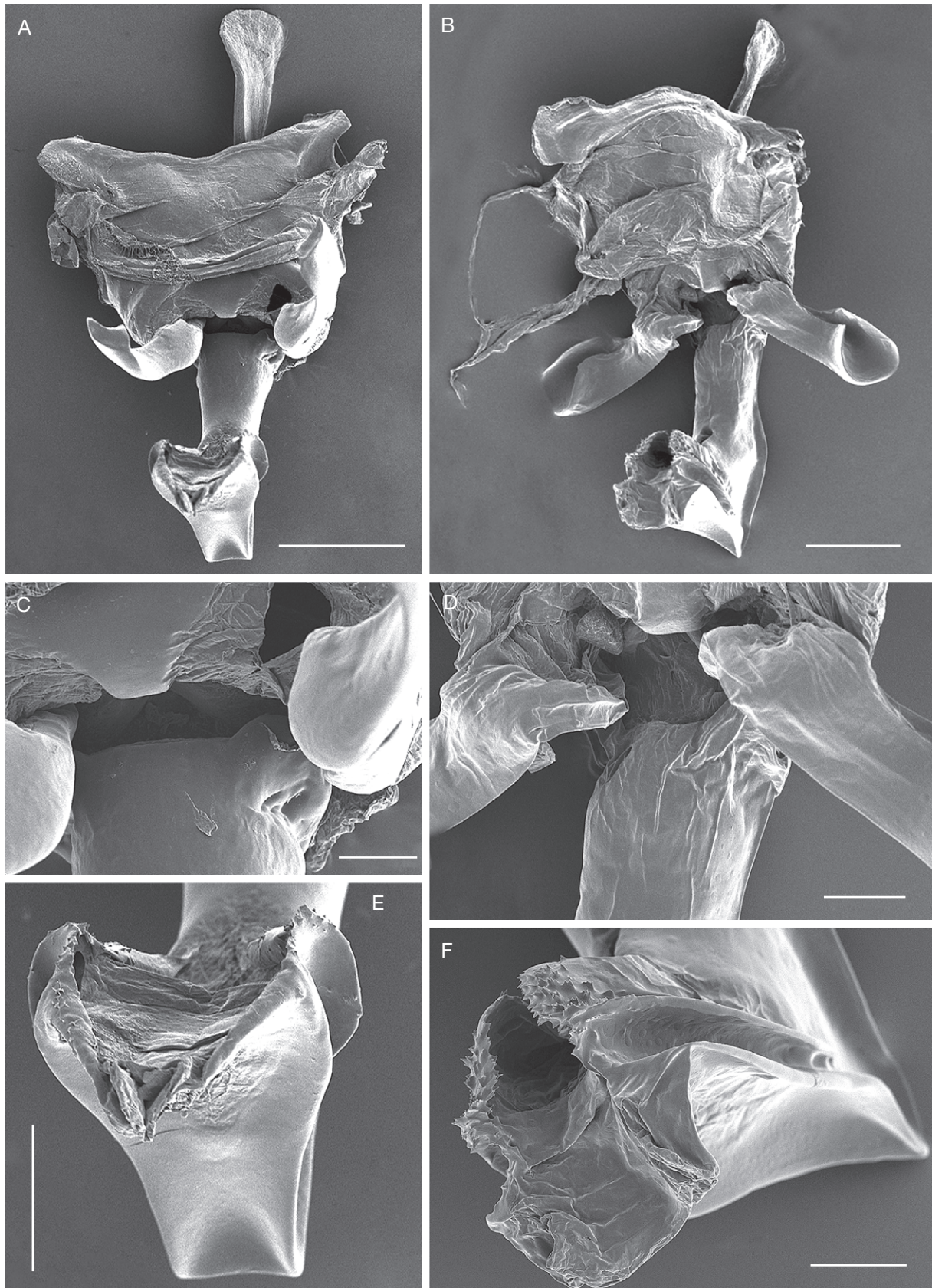


FIG. 6. — SEM images of male genitalia of *Lispe orientalis* Wiedemann, 1824 (**A**, **C**, **E**) and *L. sericipalpis* Stein, 1904 (**B**, **D**, **F**): **A**, **B**, genitalia, anterior views; **C**, **D**, origins of parameres; **E**, **F**, distal part of phalli. Scale bars: A, 200 μ m; B, 120 μ m; C, D, 40 μ m; E, 80 μ m.

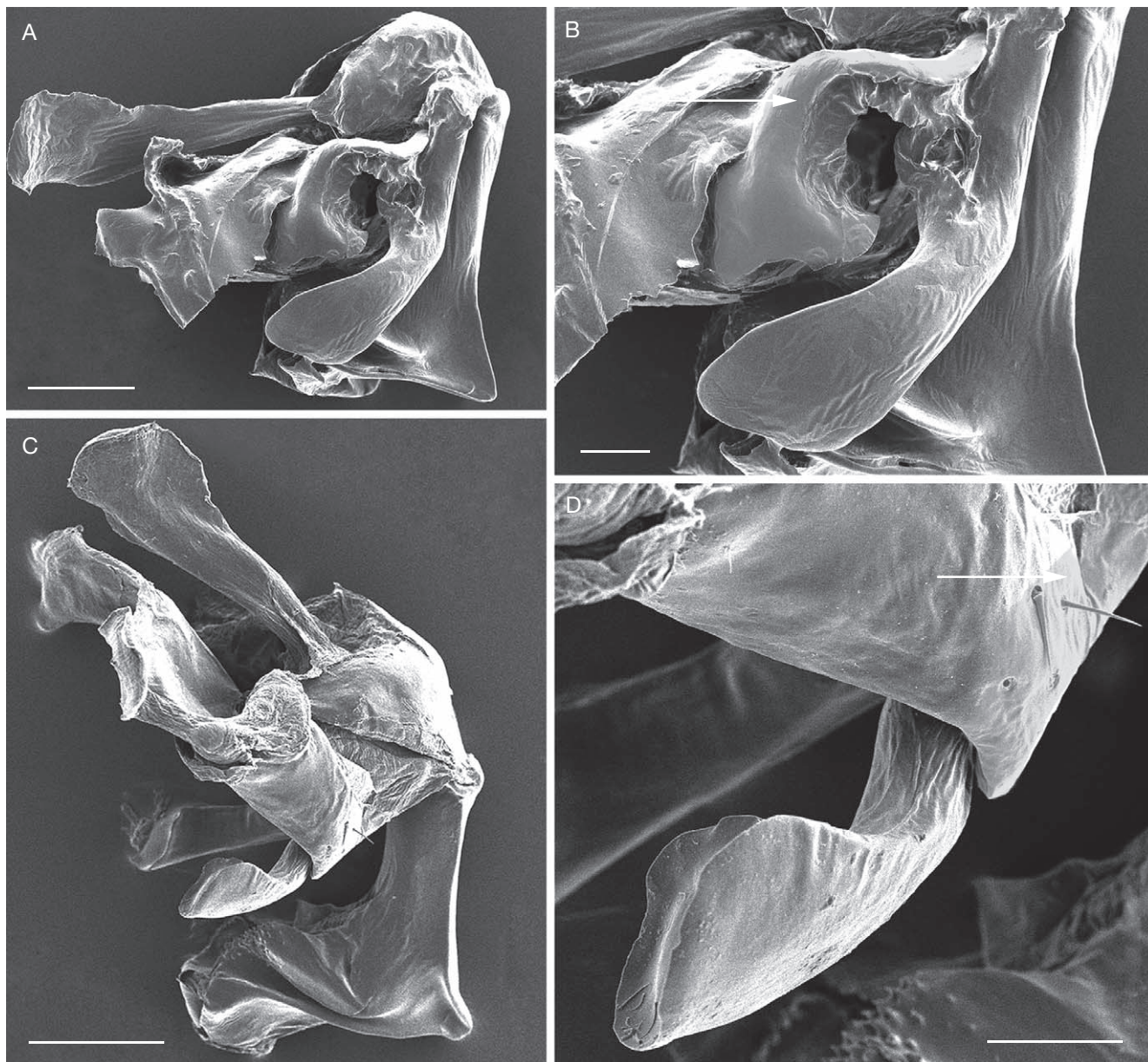


FIG. 7. — SEM images of male genitalia of *Lispe orientalis* Wiedemann, 1824 (A, B) and *L. sericipalpis* Stein, 1904 (C, D): A, C, genitalia, lateral views; B, D, degenerated gonopods fused with hypandrium, lateral view (arrows). Scale bars: A, C, 120 μ m; B, D, 40 μ m.

TYPE MATERIAL. — Holotype (of *Lispe tienmuensis* Fan), in SEMCAS, σ , labeled (1) Mt. Tianmu/ Chinese Academy of Science (CAS) [printed, in Chinese]; (2) 1954.IX.11, Coll. Fan & Shen [printed except IX.11 handwritten, in Chinese]; (3) holotype [printed]; (4) type/ *Lispe tienmuensis* Fan 1963, V [first line printed, second handwritten]; (5) *Lispe quaerens* Vill., 1936/ Fan, 1974.IV.22 [handwritten, in Chinese]; allotype (of *Lispe tienmuensis* Fan), in SEMCAS, 1 σ , labeled (1) Tienmushan/ July.28.1937 [printed, Tienmushan= Mt. Tianmu]; (2) allotype/ *Lispe tienmuensis* Fan, 1963.V. [first line printed, second handwritten]; (3) allotype [printed].

OTHER MATERIAL EXAMINED. — 6 σ and 3 σ , Beijing: Mt. Song, 1-5.VI.2009, Coll. Grade 2007 of Biology; 7 σ and 6 σ , Sichuan: Meigu County, VIII.2010. Coll. L.P. Ma and H.Q. Huang; 3 σ and 2 σ , Shandong: Yantai, 1.VII.2011, Coll. Grade 2008 of Biology; 2 σ and 2 σ , Beijing: Mt. Song, 18.VI.2013, Coll. 7 σ and 7 σ , Yunnan: Baoshan, 14.VII.2013, Coll. J. Na; 82 σ and 67 σ ,

Hubei: Yichang, Mt. Dalaoling, 22.VII.2013, Coll. D. Zhang and M. Zhang; 5 σ and 10 σ , Jiangxi: Ji'an, Suichuan County, 12.X.2014, Coll. M. Zhang and C. Wang. MNHN: 2 σ and 1 σ , Sichuan: Dafengding, VIII.2010, Coll. H. Q. Huang, C. W. Gu and L. P. Ma.

DISTRIBUTION IN CHINA. — Hubei, Gansu, Guangdong, Guangxi, Guizhou, Jilin, Liaoning, Shanxi, Shaanxi, Sichuan, Yunnan, Zhejiang.

REMARKS

Fan (1974) recorded *Lispe tienmuensis* (Figs 9, 10) (though the specimens were labeled as type material in 1963, it was not until 1974 when the work was published) and noted, "Closely related to *L. orientalis* Wd. but with narrowed parafacialia, poorly haired mid-femur in both sexes, narrowly pointed anal



FIG. 8. — Male of *Lispe tienmuensis* Fan, 1974 (holotype, deposited in SEMCAS) (*L. sericipalpis* Stein, 1904): **A**, habitus, lateral view; **B**, labels. Scale bar: 1 mm.



FIG. 9. — Female of *Lispe tienmuensis* Fan, 1974 (allotype, deposited in SEMCAS) (*L. sericipalpis* Stein, 1904): **A**, habitus, lateral view **B**, labels. Scale bar: 1 mm.

cerci in male”. But after re-examining the type specimens, he corrected *L. tienmuensis* as a synonym for *L. quaerens* and published this synonym in his book, *Key to the Common Flies of China* (Fan 1992).

Wei (in Li & Jin 2006) reported *L. fanjingshanensis* from Mt. Fanjing, Tongren, Guizhou. He commented that it is closely related to *L. quaerens* but differs in its greyish blue abdominal pollinosity and developed distiphallus which is bended at a vertical angle (Fig. 10B). After going through original description (focus on leg chaetotaxy especially hind legs: femur with complete *ad* row and incomplete *av* row in basal half, tibia with a developed median *ad*) and illustrations of *L. fanjingshanensis*, we propose that it is conspecific to *L. sericipalpis*.

Lispe tentaculata DeGeer, 1776 (Figs 1D; 2D; 4B, D, F; 5D, E, F; 11C, D)

Lispe tentaculata DeGeer, 1776: 86.

MATERIAL EXAMINED. — 3 ♂, Beijing: Mt. Song, 31.V.2009, Coll. D. Zhang; 1 ♀, Beijing: Mt. Song, 1.VI.2009, Coll. D. Zhang; 47 ♂ and 35 ♀, Xinjiang: Kalamaili Nature Reserve, 11–14.VIII.2009, Coll. D. Zhang; 6 ♂ and 7 ♀, Xinjiang: Tekesi, 24.VIII.2013, Coll. M. Zhang; 85 ♂ and 62 ♀, Xinjiang: Kalamaili Nature Reserve, 18.V–23.VI. 2014, Coll. M. Zhang.

DISTRIBUTION IN CHINA. — Beijing, Hebei, Heilongjiang, Jilin, Liaoning, Shandong, Shanxi, Xinjiang.

REMARKS

Hennig’s drawing (1960: 408, fig. 131, with a bended distal part) of the phallus of *L. tentaculata* is not the same as in our dissection (Fig. 11D, specimen from Xinjiang). In our dissection, the phallus of *L. tentaculata* is straight and without a bended distal part, identical to Snyder’s drawing (1954: 15, fig. 53) of the phallus of *L. sociabilis*, about which he stated “might serve equally well for *patellata* and Nearctic specimens of *tentaculata*” (Snyder 1954: 38). Snyder’s statement was confirmed by Vikhrev (2014: 154). These images are placed in one plate for comparison. We speculate that Hennig’s drawing was based on either an aberrant *L. tentaculata* or an underlying species that curiously have both modified first fore tarsomere and bended phallus, an intermediate taxon between *L. tentaculata*-subgroup and *L. orientalis*-subgroup (more information in Discussion below).

MOLECULAR PHYLOGENY

Analysis of maximum likelihood and Bayesian inference recovered largely congruent results and most of the species has a high leaf stability value (Fig. 12). The monophyly of *Lispe* is highly supported (maximum likelihood bootstrap: 98, Bayesian posterior probability: 1), which was already suggested by Kutty *et al.* (2014). The *L. tentaculata*-group is confirmed to be monophyletic here as well (maximum likelihood bootstrap: 87, Bayesian posterior probability: 1).

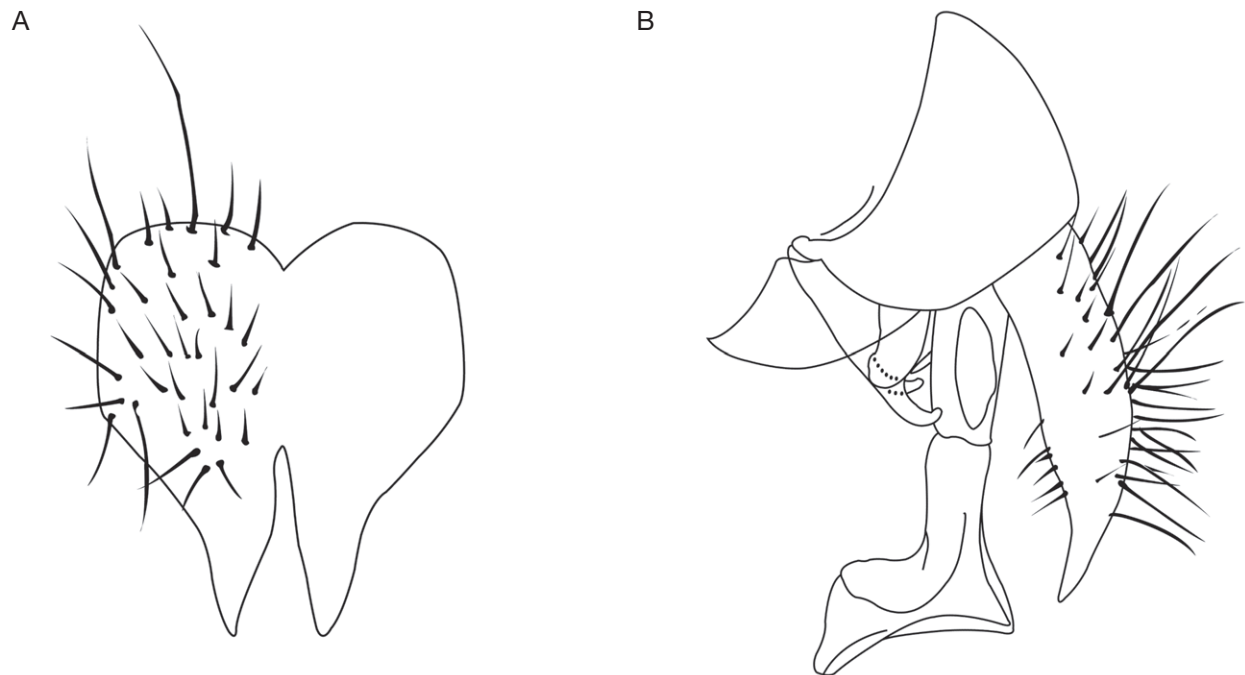


FIG. 10. — Male terminalia of *Lispe fanjingshanensis* Wei, 2006 (adapted from Li & Jin 2006) (*L. sericipalpis* Stein, 1904): **A**, cerci, ventral view; **B**, epandrium, surstyli, cerci and phallus, lateral view.

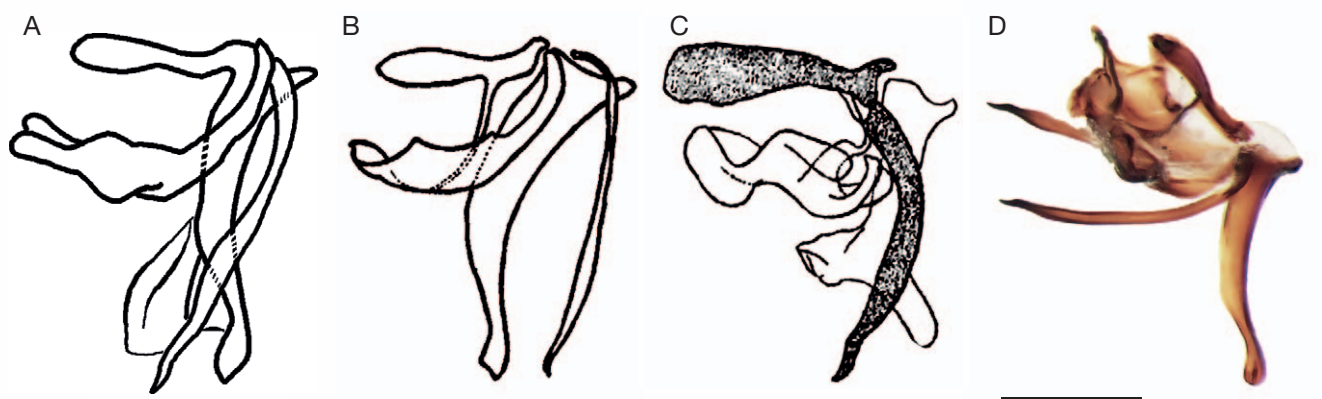


FIG. 11. — Male phalli of *Lispe tentaculata*-group, lateral view: **A**, *L. alpinicola* Zhong, Wu & Fan, 1981 (adapted from Zhong, Wu & Fan 1981); **B**, *L. sociabilis* Loew, 1862 (adapted from Snyder 1954); **C**, *L. tentaculata* (De Geer, 1776) (adapted from Hennig 1960); **D**, *L. tentaculata* (De Geer, 1776) (specimen from Xinjiang, China). Scale bar: 0.50 mm.

Based on two morphological features (first fore tarsomere and phallus), the *L. tentaculata*-group could be separated as two subgroups (*L. tentaculata*-subgroup with modified first fore tarsomere and straight phallus while *L. orientalis*-subgroup is distinguished with regular first fore tarsomere and bended phallus). The tree topologies from the different analyses, ML and Bayesian, are highly corroborated that the group is split into two clades (maximum likelihood bootstrap: 100, Bayesian posterior probability: 1). Therefore, the proposal of two subgroups within *L. tentaculata*-group is verified.

DISCUSSION

Our analyses of this group both in morphology and phylogeny reveal the relationships between four species within the *L. tentaculata*-group. Other holarctic species from the group all fall strictly into the definitions of two subgroups except the Nearctic *L. sociabilis*, which has straight phallus but unmodified first fore tarsomere. Another case with characters that contradict with the diagnoses of two subgroups is Hennig's drawing of *L. tentaculata* (1960: 408, fig. 131). We can not

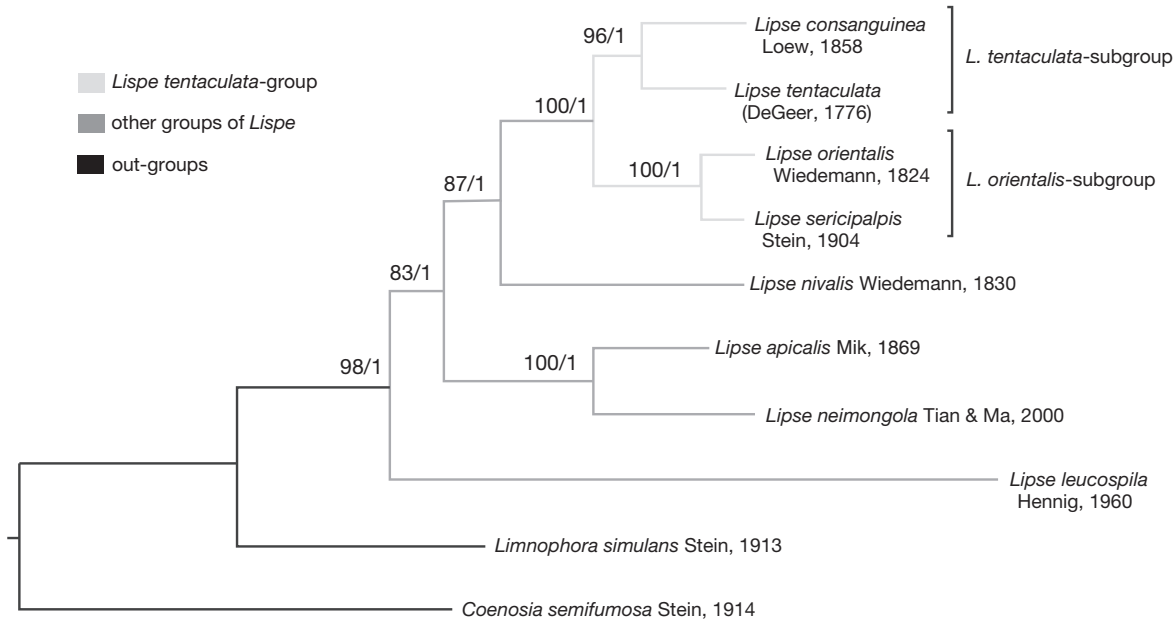


FIG. 12. — Phylogeny of the *Lispe tentaculata*-group, inferred from seven mitochondrial and nuclear genes. Above node left: bootstrap support for maximum likelihood; above node right: posterior probabilities for Bayesian.

be certain about whether Hennig's illustration was derived from an abnormal *L. tentaculata* or an independent species that has both modified first fore tarsomere and bended phallus. However, these two cases are clear evidences of the existence of intermediate taxa between *L. tentaculata*-subgroup and *L. orientalis*-subgroups. More species should be involved in both morphological and phylogenetic research to explore the diversification patterns within this species-group.

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