

Urticaceae-feeders from the family Tischeriidae: descriptions of two new species and new genus *Paratischeria* gen. nov.

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We describe a new genus *Paratischeria* Diškus & Stonis, gen. nov. and present the first observations of Urticaceae-feeding Tischeriidae species in South America and two new Urticaceae-feeding species (*Paratischeria fasciata* sp. nov. and *P. ferruginea* Diškus & Stonis, sp. nov. from the Andes of Bolivia and Ecuador). Together with the African *P. urticicolella* (Ghesquière) (comb. nov.) from Congo, they are attributed to the newly designated *ferruginea* species group.

Keywords: The Andes, leaf-mines, new genus, new species, *Paratischeria*, *Phenax* Wedd., South America, Tischeriidae, Urticaceae

INTRODUCTION

The Tischeriidae represents the relatively small family of Lepidoptera. The most recent detailed family review was provided by Diškus, Puplesis, 2003. Among the most distinctive diagnostic features of the family is the long length of male antennal sensillae (Fig. 67); females possess much shorter sensillae, therefore, female antennae appear bare in ventral view. Among other characters that also have diagnostic importance for family recognition are the following: (1) frontal tuft (Fig. 26) projecting over triangular (or trapezoid) face smoothly covered with scales; (2) sensillae trichodea with

strongly recurved bases (Fig. 67); (3) strongly enlarged 3rd antennal segment (Fig. 67); (4) strongly narrowed phallus (Figs. 8, 33, 45), usually bifurcated or with spines at apex; (5) dark, short, strongly thickened, stout peg setae on female ovipositor (usually visible even without dissection) (Fig. 29); (6) specific rod-like or plate-like projections of modified 8th and 9th sternites in female genitalia collectively referred as prela (Fig. 31).

Literature on Tischeriidae is still rather scarce (for the history of research on the family investigation and a literature review, see Puplesis, Diškus, 2003). Since then several papers have been published, notably: Landry, Roque-Albelo, 2004; Puplesis et al., 2004; Mey, 2004, 2010; Puplesis, Diškus, 2005; Diškus, Stonis, 2006, 2012, 2015;

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Stonis, Diškus, 2007, 2008; Lees, Stonis, 2007; Stonis et al., 2008, 2014, 2016; Hua, Cai, 2009; van Nieukerken, 2010; Navickaitė et al., 2011; Diškus et al., 2014; Kobayashi et al., 2016.

Tischeriidae are trophically associated with plants belonging to rosid and asterid I core eudicot angiosperm families Fagaceae (Fagales), Rosaceae, Rhamnaceae (Rosales), Malvaceae (Tilioideae, Sterculioideae, Malvales), and Asteraceae (Asterales). A few representatives of Tischeriidae have also been recorded on plants from the Combretaceae (Myrtales), Euphorbiaceae (Malpighiales), Ericaceae, Symplocaceae (Ericales), Anacardiaceae (Sapindales), Theaceae (Ericales), Hypericaceae (Malpighiales), Betulaceae (Fagales), and Apocynaceae (Gentianales) (Diškus, Puplesis, 2003a, 2003b; Kobayashi et al., 2016). In temperate regions the family is strongly associated with plants from the Rosaceae and Fagaceae, in the Americas also with Malvaceae and Asteraceae. However, only a single record of the Urticaceae-feeding species was previously known from equatorial Africa (Puplesis, Diškus, 2005), and no records from the Americas.

In this paper, we review our recent findings in South America together with the previous record of the Urticaceae-feeding species from Congo, equatorial Africa (Fig. 1). We illustrate the African species and describe two new Tischeriidae species from the Andes: one trophically associated with *Phenax hirtus*, other feeding on unidentified Urticaceae plant but not *Phenax*.

Host plant Urticaceae is a medium-sized family comprising ca 2,500 species and 50 genera of woody shrubs, succulent herbs, trees and vines that are found on all of the world's continents with the exception of Antarctica. A short characterization of Urticaceae will be provided in our other paper on the Urticaceae-feeding Nepticulidae (Stonis et al., submitted).

MATERIALS AND METHODS

Descriptions of new species are based on the material deposited in the collection of the Zoological Museum, Natural History Museum of Denmark in Copenhagen, Denmark.

Collecting methods and protocols for species identification and description are outlined in Puplesis, Diškus (2003) and Diškus, Stonis (2012). After maceration of the abdomen in 10% KOH and subsequent cleaning, male genital capsules were removed from the abdomen and mounted ventral side uppermost. The phallus was removed and mounted in Euparal separately but on the same genitalia slide. Abdominal pelts and female genitalia were stained with Chlorazol Black (Direct Black 38/Azo Black) (for a detailed method's description see Stonis et al., 2014).

Permanent slides were photographed and studied using a Leica DM2500 microscope and a Leica DFC420 digital camera. The descriptive terminology of morphological structures follows Puplesis, Diškus (2003), except for the term "aedeagus" that is referred here as "phallus", and the term "cilia" that is referred here as "fringe".

For DNA extraction we used specimens which were stored in 96% ethanol. Total genomic DNA was extracted from head or thorax using a Nucleospin Tissue Kit (Machery-Nagel, Düren, Germany) according to the protocol of the manufacturer.

Amplification of a 577-bp fragment of the mtDNA region was performed using mitochondrial cytochrome c oxidase subunit I gene (COI): LCO1490 (5'-GGT CAA CAA ATC ATA AAG ATA TTG G-3') and HCO2198 (5'-TAA ACT TCA GGG TGA CCA AAA AAT CA-3') (Folmer et al., 1994; Herbert et al., 2003a, 2003b). PCR were performed in 25 µl volumes including: 2 µl DNA, 2 µl of each primer (MBI Fermentas, Lithuania), 0.5 µl of Amplitaq DNA polymerase (5U/µl), 2.5 µl 25 mM MgCl₂, 2.5 µl 10X Buffer (Fermentas) and 1 µl 10 mM dNTP (Fermentas) and water.

PCR were carried out in an Eppendorf Mastercycler gradient, 5331 (Germany). Cycling parameters were an initial denaturation step at 94 °C for 2 min, followed by 94 °C for 30 s, 50 °C for 45 s, and 72 °C for 1 min. This cycle was repeated 35 times, followed by 4 min of extension at 72 °C. PCR products were visualized on 1.5% agarose gel stained with ethidium bromide and UV light photographs of the gels with DNA bands were taken using "Herolab"

transluminator (Germany). The positive bands of the expected size range were excised from the gel. DNA was extracted using the GeneJet Gel Extraction kit (Fermentas, Lithuania). Amplicons were then sequenced in both directions on an automated sequencer, ABI 3130xl (Applied Biosystems, USA).

Electropherograms were checked by eye for poor base calls and sequence quality and then sequences were edited and aligned using the ClustalW (Thomson et al., 1994) algorithm in MEGA v.7.0.21 (Kumar et al. 2015). Phylogenetic relationships among control region haplotypes were reconstructed using the neighbour-joining (NJ) method (Saitou, Nei, 1987). NJ analysis was performed using the Kimura-2

paratemeter correction model (Kimura, 1980) by bootstrapping with 1000 replicates.

Partial fragment of Cytochrome oxidase I (COI) gene were amplified in 21 Tischeriidae specimens. Amplified fragment produced a 577-bp sequence which corresponded to COI gene sequence position 2239–2944.

The partial nucleotide sequences of the Tischeriidae species COI gene were added to the GenBank database. Sequences accession numbers are presented in Table.

Institutional abbreviations used in the text of the current paper: MRAC – Musée Royal de l’Afrique Centrale, Tervuren, Belgium; ZMUC – Zoological Museum, University of Copenhagen, Denmark.

Table 1. GenBank accession numbers of examined specimens of Tischeriidae

Examined specimens	Accession number GenBank
<i>Coptotriche marginea</i> (1) (Haworth, 1828)	HM244382
<i>Coptotriche marginea</i> (2) (Haworth, 1828)	HM244379
<i>Coptotriche marginea</i> (3) (Haworth, 1828)	HM244380
<i>Coptotriche marginea</i> (4) (Haworth, 1828)	HM244381
<i>Tischeria ekebladella</i> (1) (Bjerkander, 1795)	HM244384
<i>Tischeria ekebladella</i> (2) (Bjerkander, 1795)	HM244385
<i>Tischeria ekebladella</i> (3) (Bjerkander, 1795)	HM244383
<i>Tischeria dodonea</i> (1) Stainton, 1858	HM244386
<i>Tischeria dodonea</i> (3) Stainton, 1858	HM244387
<i>Astrotischeria</i> sp. 4848	HM244389
“ <i>Astrotischeria</i> ” sp. 4877, <i>Paratischeria</i> sp.	HM244393
“ <i>Astrotischeria</i> ” sp. 4894, <i>Paratischeria ferruginea</i> Diškus & Stonis, sp. nov.	HM244395
“ <i>Astrotischeria</i> ” sp. 4909.1, <i>Paratischeria neotropicana</i> (Diškus & Stonis, 2015)	HM244391
“ <i>Astrotischeria</i> ” sp. 4909.2, <i>Paratischeria neotropicana</i> (Diškus & Stonis, 2015)	HM244392
<i>Astrotischeria</i> sp. 4910	HM244390
“ <i>Astrotischeria</i> ” sp. 4936, <i>Paratischeria ferruginea</i> Diškus & Stonis, sp. nov.	HM244394
“ <i>Astrotischeria</i> ” sp. 4937, <i>Paratischeria neotropicana</i> (Diškus & Stonis, 2015)	HM244396
“ <i>Astrotischeria</i> ” sp. 4939, <i>Paratischeria</i> sp.	HM244398
<i>Astrotischeria</i> sp. 4944.2	HM244397
<i>Astrotischeria</i> sp. 4957.1	HM244399
<i>Astrotischeria</i> sp. 49572	HM244400

RESULTS

Taxonomy of the Urticaceae-feeding Tischeriidae

Three currently known Urticaceae-feeding Tischeriidae, including one Afrotropical and two Neotropical species, together with some American Malvaceae and Asteraceae-feeding species, belong to a new genus, *Paratischeria* Diškus & Stonis, gen. nov., which is described below.

Genus *Paratischeria* Diškus & Stonis, gen. nov.

Type species: *Paratischeria ferruginea* Diškus & Stonis, sp. nov.

Diagnosis. In the male genitalia, the new genus differs from *Astrotischeria* in the undivided, usually narrow valva (divided in *Astrotischeria*), long undivided uncus (usually short and divided in *Astrotischeria*), and the strongly though variously developed anellus (Figs. 48–53); from *Coptotriche* it differs in the slender valva, well developed anellus with lateral papillae and setae (Figs. 48–53), also in the absence of transtilla, absence of spines on the diaphragma, larger vinculum, and differently shaped phallus (not tulip-shaped as in *Coptotriche*); from *Tischeria* it differs in the absence of juxta, presence of strongly developed anellus (absent in *Tischeria*), and usually larger vinculum. Also see Figs. 73, 74.

Head (Figs. 54–67). Scape with a pecten. 3rd antennal segment greatly enlarged.

Wing venation (Figs. 68–70). Greatly reduced, similar to *Astrotischeria* (Fig. 71). In the forewing, vein Sc almost reaching middle of wing; R with five short radial branches terminating on costa; radial cell reduced; M with two short branches distally and with base absent; vein Cu single, very long and straight; 2A well-developed, but simple; in hindwing, Sc very short; R, M and Cu expressed only in distal half (if not entirely reduced), vein A single or represented by two short veins.

Abdomen (Fig. 72). Male with a short anal tuft of piliform scales.

Male genitalia. Uncus with long lateral lobes (Fig. 38). Socii not thickened, similar to those in *Astrotischeria*. Tegumen large, with

strongly sclerotized anterior processes; posteriorly not extending into a lobed pseuduncus. Diaphragma smooth or with membranous wringles but without spines. Valva relatively slender, undivided. Transtilla absent. Anellus always well-developed, slightly thickened laterally and with three or four pairs of papillae and setae, sometimes additionally with a dorsal sclerite (pseudotranstilla) (Figs. 48–53). Juxta absent. Vinculum moderately large to very large, anteriorly either broadly rounded or gradually narrowed. Phallus long and distinctly bifurcate in apical half or apical third (Figs. 33, 45).

Female genitalia. Ovipositor lobes large (Figs. 29, 31); the second pair of lobes very small. Anterior and posterior apophyses supplemented with three pairs of shorter rod-like and plate-like sclerites (prela) (Fig. 31). Vestibulum membranous or hardened but antrum absent. Corpus bursae slender, as long as anterior apophyses or longer, usually without pectination; signa absent. Ductus spermathecae membranous, slender, with a few to many convolutions.

Biology. Larvae produce blotch-like mines (Figs. 11–19) in leaves of plants from Malvaceae, Urticaceae, and Asteraceae.

Distribution. Known from the New World (the Americas) and equatorial Africa. There are many species from Central and South America which have already been dissected, identified but still undescribed (Stonis et al., in prep).

Etymology. The name combines *Tischeria* (a name of the type genus of the family) and Greek prefix *para* (at or to one side of, side by side, beyond).

The *ferruginea* species group

Designated here for the first time for the *Paratischeria* species feeding on Urticaceae and possessing a dorsal sclerite (pseudotranstilla) in the male genitalia.

Diagnostics: forewing dark brown to brown (not sparsely speckled as is usual in other *Paratischeria* or *Astrotischeria*), with spot(s) (Figs. 5, 26), fascia (Fig. 5), or without spots or fascia, uniform. In male genitalia, phallus deeply divided in distal half (Figs. 7, 8, 32–34,

45); valva moderately slender (but wider than in many other *Paratischeria* species) or strongly widened in basal half (Fig. 36); basal process of valva long (Figs. 6, 39, 44); uncus with two long (Fig. 38) to very long lobes (Fig. 46); anellus with a dorsal sclerite (pseudotranstilla) (Figs. 42–44); vinculum large but smaller than in most of other *Paratischeria*, triangular (Fig. 35) or distally broadly rounded (Figs. 6, 44).

Currently the group comprises three equatorial or subequatorial species (two from the Andes of Ecuador and Bolivia), one from Africa (Congo); all species are trophically associated with plants from Urticaceae (*Phenax* and *Fleurya*).

***Paratischeria fasciata* Diškus & Stonis, sp. nov.**

Type material. Holotype: ♂, BOLIVIA: Nor Yungas Province, Coroico, 16°11'39"S,

67°43'21"W, elevation 1880 m, mining larva on unidentified Urticaceae plant 25.iv.2014, ex pupa v.2014, A. Diškus, genitalia slide no. AD868♂ (ZMUC). Paratype: 1 ♂, same label data as holotype (ZMUC).

Diagnosis. Externally, the new species differs from all other known tischeriids by a distinctive fascia of the forewing. In male genitalia, a specific shape of phallus and dorsally developed anellus distinguish *Paratischeria fasciata* sp. nov. from all other Tischeriidae species; the Urticaceae host plant, shared with three other related species, also makes this taxon distinctive.

Male (Figs. 4, 5). Forewing length 3.0–3.2 mm; wingspan 6.5–6.8 mm. Head: face smoothly scaled, palpi and face grey-brown or olive brown, very glossy and with some golden shine and purple iridescence; frontal tuft grey-brown to olive brown, smooth, not erected

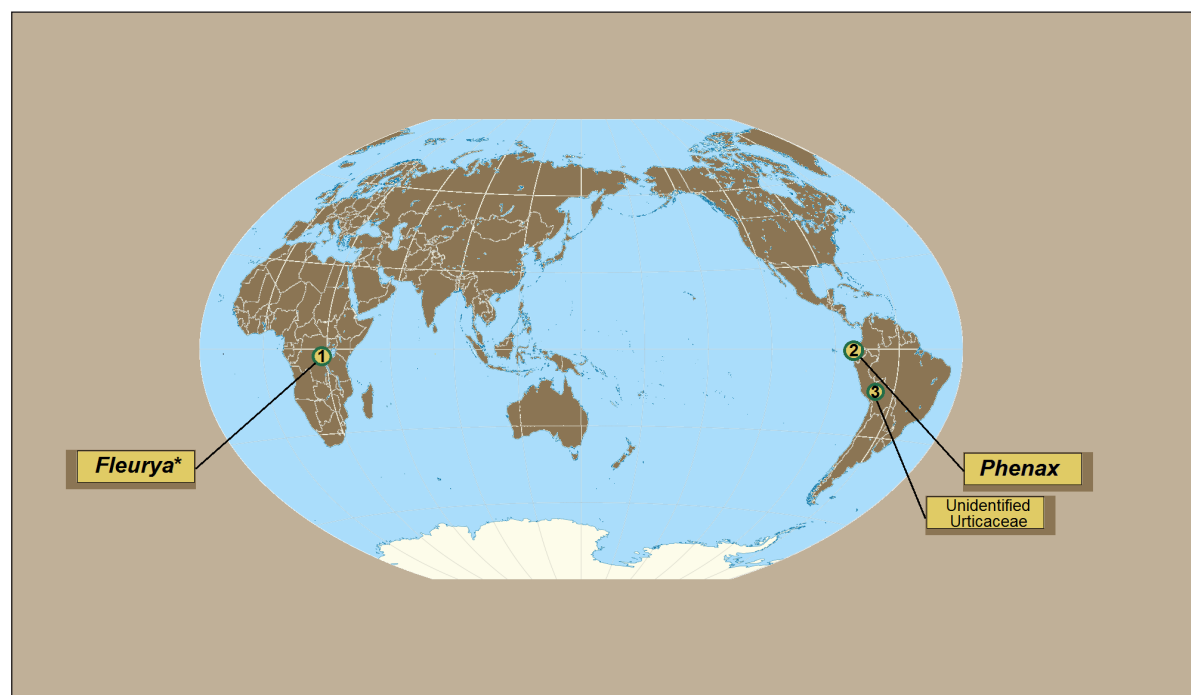


Fig. 1. Currently known records of Tischeriidae trophically associated with the genera of Urticaceae: 1 – *Paratischeria urticicolella* (Ghesquière, 1940) on *Fleurya podocarpa* Wedd. from Congo (see Puplesis, Diškus, 2005); 2 – *P. ferruginea* Diškus & Stonis, sp. nov. on *Phenax hirtus* (Sw.) Wedd. from Pichincha Province, Ecuador; 3 – *P. fasciata* Diškus & Stonis, sp. nov. on unidentified Urticaceae plant, Coroico, Bolivia; * – earlier, by Ghesquière, 1940, the host plant was reported as *Laportea podocarpa* Wedd., however, the genus *Laportea* has been split and the host plant is attributed now to *Fleurya*. *Fleurya podocarpa* Wedd. is placed as a synonym of *ovalifolia*, however, such combination of *ovalifolia* with *Fleurya* has not been published.

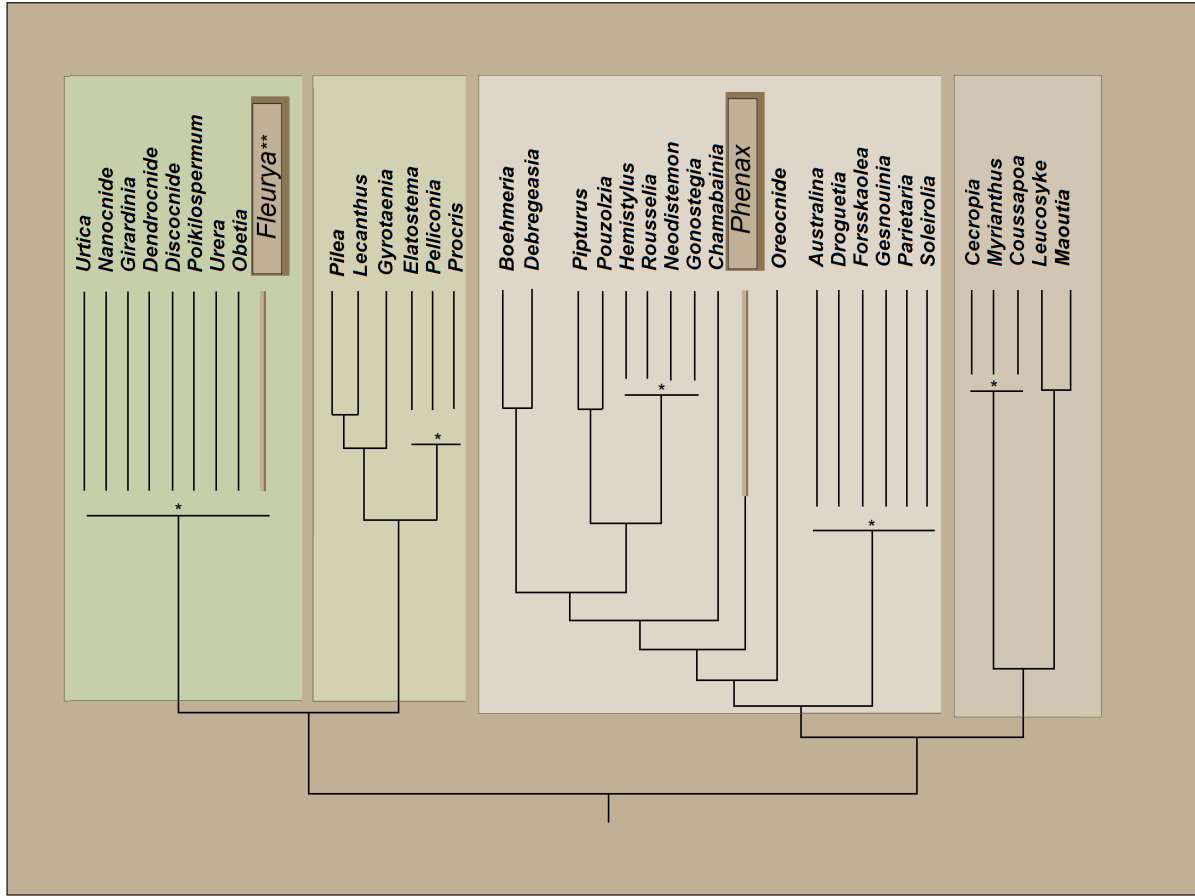


Fig. 2. Urticaceae clades engaged by leaf-mining Tischeriidae (simplified phylogeny after Wu et al., 2013. Note: this is not an ultrametric tree)

* – for detailed relationships of the clades, see Wu et al., 2013

** – originally, the host plant was reported as belonging to the genus *Laportea* (see Ghesquière, 1940), however, the genus *Laportea* has been split and the host plant is attributed now to genus *Fleurya*.

but overlapping over face; antenna slightly longer than half the length of forewing, grey-brown, golden glossy, with long hair-like sensillae trichodea sticking out. Thorax and tegula black-brown with strong golden gloss and some purple iridescence. Forewing black-brown with some golden gloss and purple and blue iridescence; median fascia and basal spot yellow orange, very distinctive; fringe dark brown with some golden gloss; underside of forewing brown-black, without spots except small irregular scaleless patch at the base. Hindwing dark brown on upper side, brown-black on underside, without androconia or spots except small irregular scaleless patch at the base on underside; its fringe blackish

brown. Legs grey, glossy. Abdomen black-brown on upper side, dark grey, glossy on underside; genital segments dark grey-brown to grey, distally white; anal tufts short, blackish brown.

Female. Unknown.

Male genitalia (Figs. 6–10). Capsule about 455 µm long. Uncus with two long lateral lobes. Valva about 350 µm long, not divided, without processes (except the basal one); transtilla absent. Anellus thickened laterally and with setae (Figs. 6, 51), dorsally with a specific plate-like sclerite (pseudotranstilla) (Figs. 6, 43). Vinculum rather short, rounded distally. Phallus (Figs. 7, 8) 455 µm long, with three pairs of large carinae.

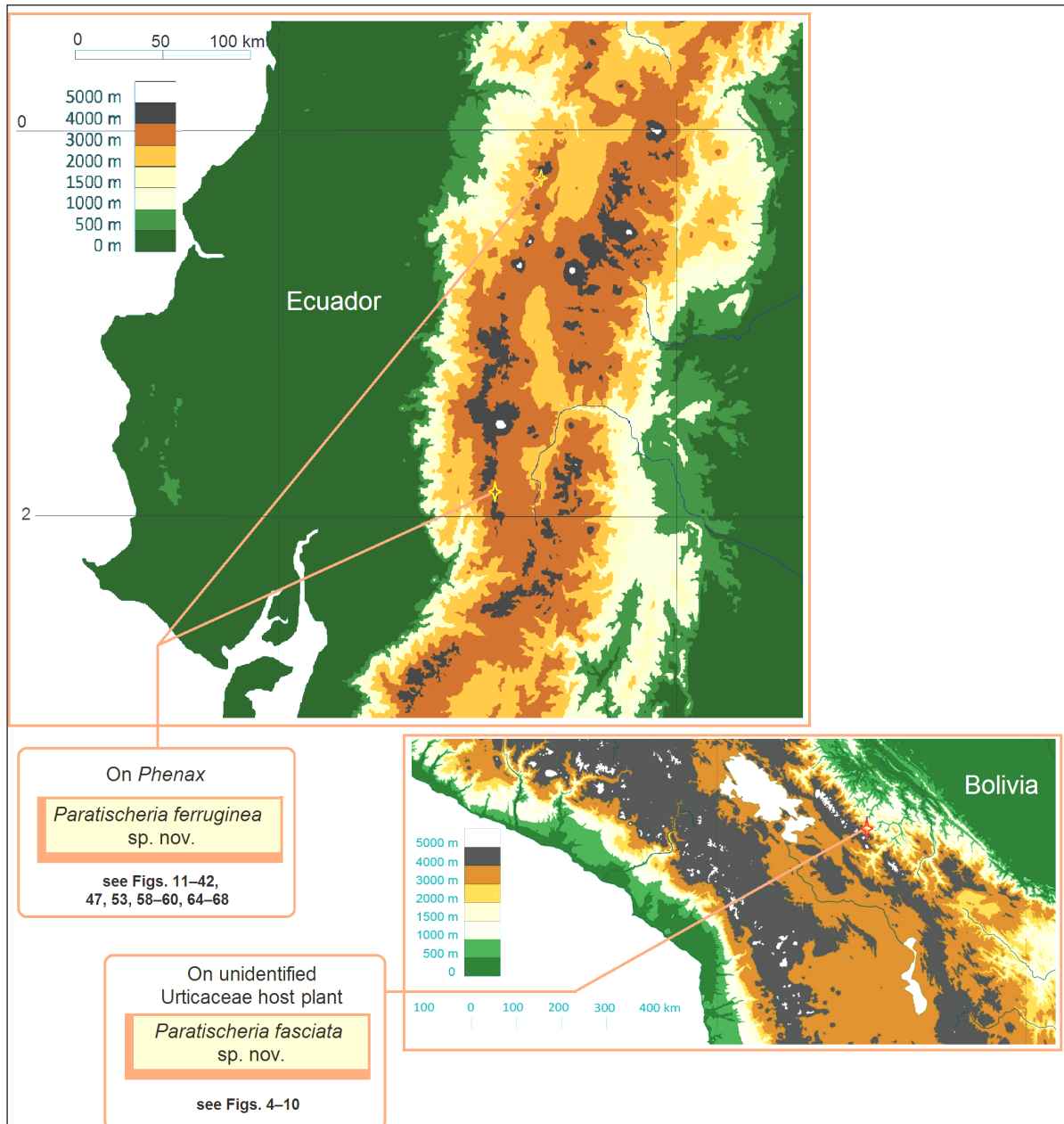


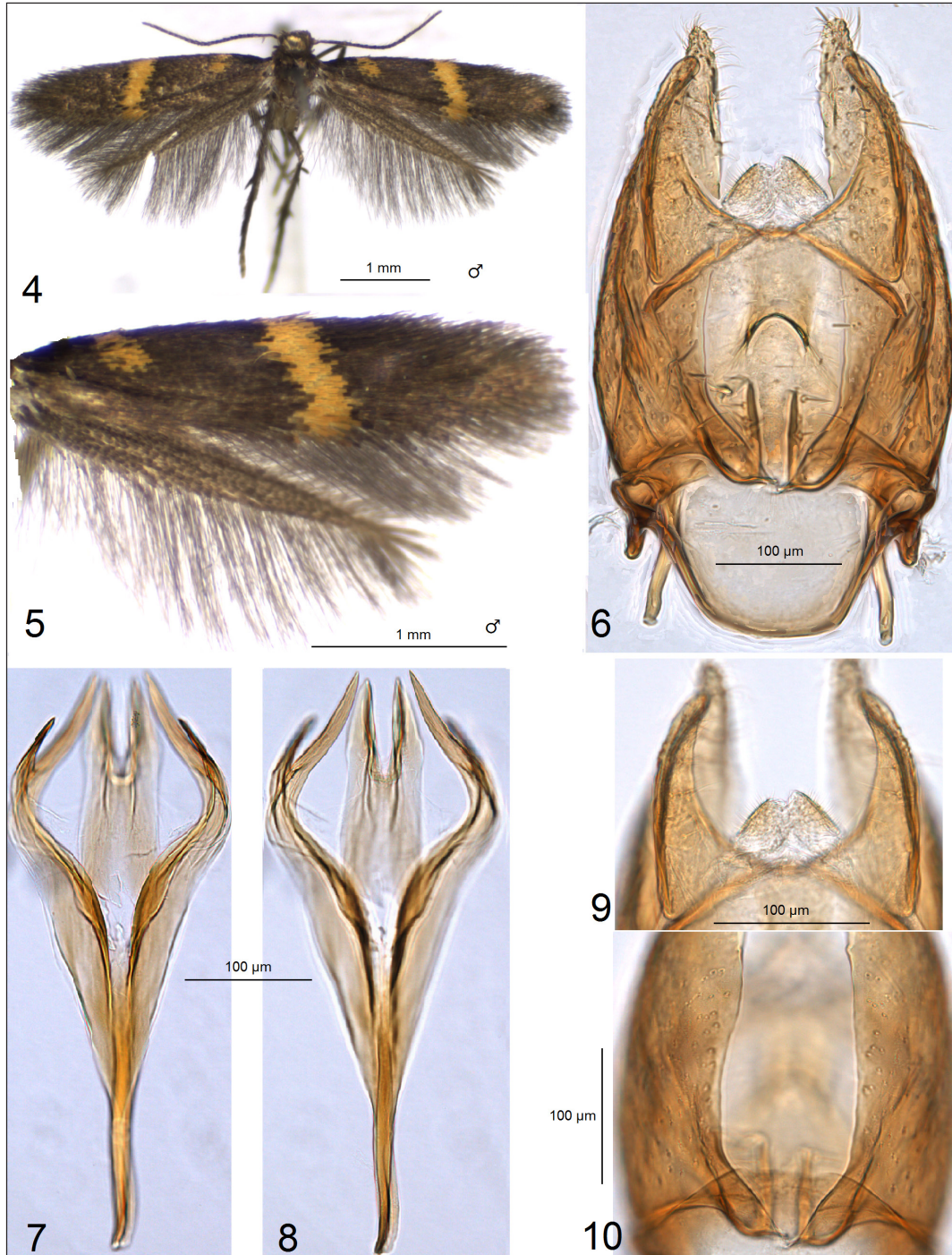
Fig. 3. Current records of Urticaceae-feeding Tischeriidae from South America

Bionomics. Host plant belongs to Urticaceae (species and genus unidentified; host-plant sample neither preserved nor documented). Larvae mine in leaves in April. A leaf-mine similar to those illustrated in Figs. 11–19). Exit slit on the upper side of the leaf. According to the “Formula of Determining of Abundance and Occurrence of Leaf-Miners” (see Diškus, Stonis 2012: 52–54), *Paratischeria fasciata* is a rare species:

sparse mining of the new species was observed in a single locality of the Bolivian Andes.

Distribution (Figs. 1, 3). This species occurs on the eastern slopes of the Bolivian Andes in tropical montane forests at altitudes ca. 1880 m.

Etymology. The species name is derived from the Latin *fascia/fasciata* (a band, fascia/with fascia) in reference to the distinctive fascia on the forewing, so unusual among the Tischeriidae.

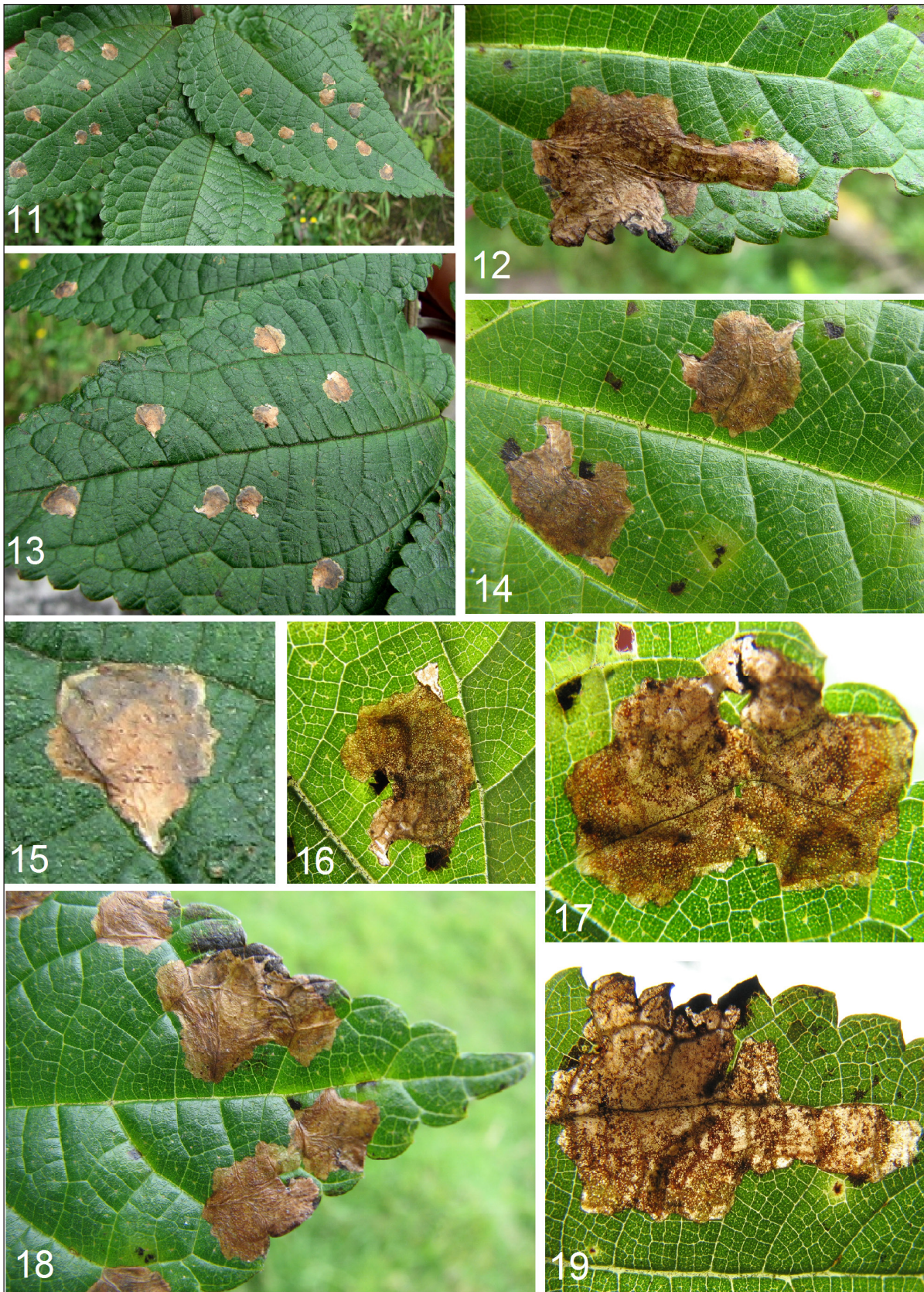


Figs. 4–10. *Paratischeria fasciata* Diškus & Stonis, sp. nov.: 4, 5 – male adult, holotype; 6, genitalia slide no. AD868, capsule with phallus removed; 7, 8 – same, phallus; 9 – same, uncus and socii; 10 – same, basal part of valvae (ZMUC)

***Paratischeria ferruginea* Diškus & Stonis, sp. nov.**

Type material. Holotype: ♂, ECUADOR: Pichincha Province, 11 km NW Alóag, 0°26'45"S, 78°37'34"W, elevation 3090 m,

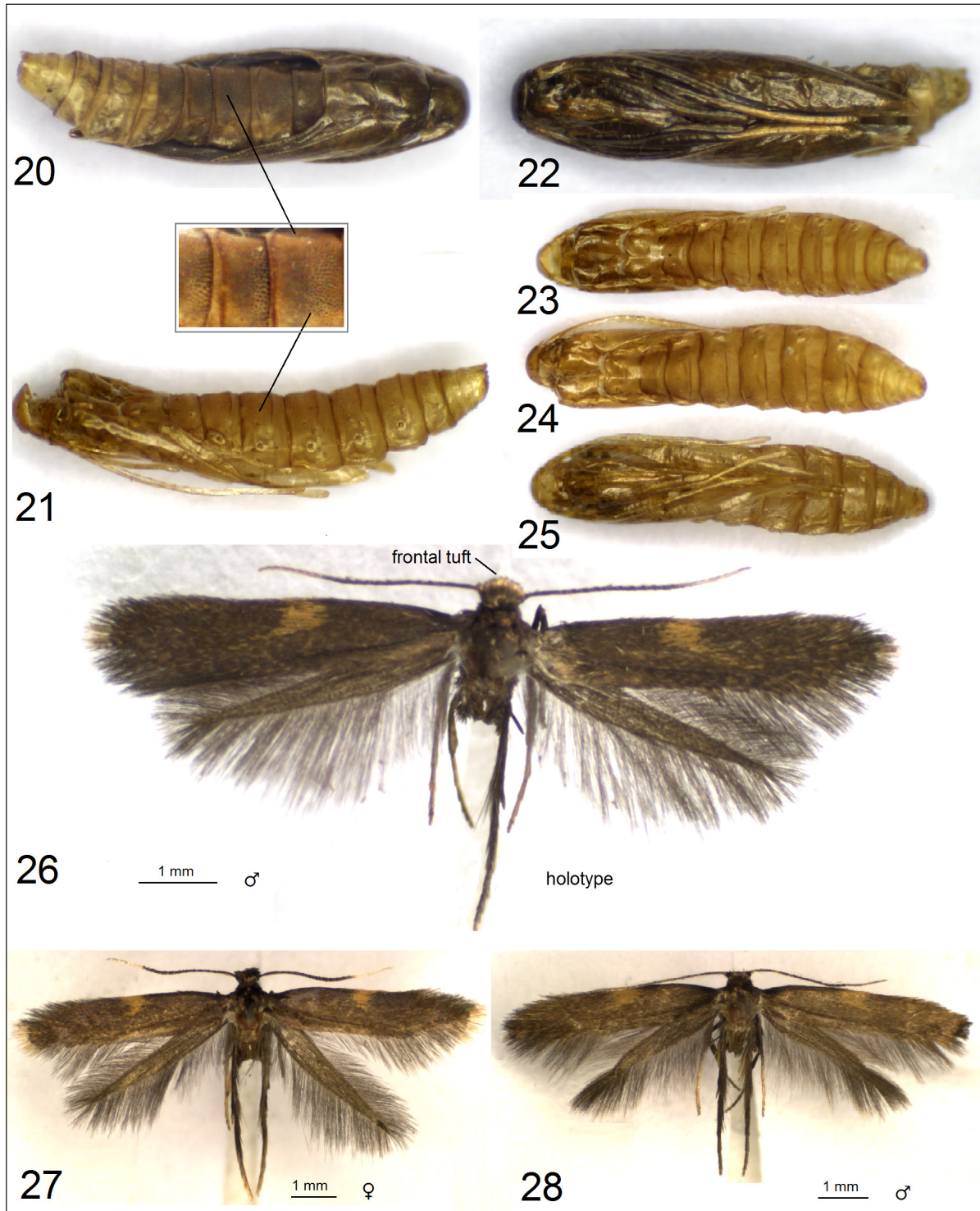
mining larvae on *Phenax hirtus* (Urticaceae) 20.xi.2007, field card no. 4936, A. Diškus, genitalia slide no. AD775♂ (ZMUC). Paratypes: 4 ♂ (including a genitalia and head slide of one adult in the pupal skin, with no pinned



Figs. 11–19. Leaf-mines of *Paratischeria ferruginea* Diškus & Stonis, sp. nov. on *Phenax hirtus*, Ecuador, Pichincha Province, 11 km NW Alóag, 0°26'45"S, 78°37'34"W, elevation 3090 m

specimen preserved), 5 ♀, same label data as holotype, slide nos. AD839♂ (genitalia), AD866♂ (genitalia), AD865 (head), AD863 (forewing venation), AD867♂ (genitalia and

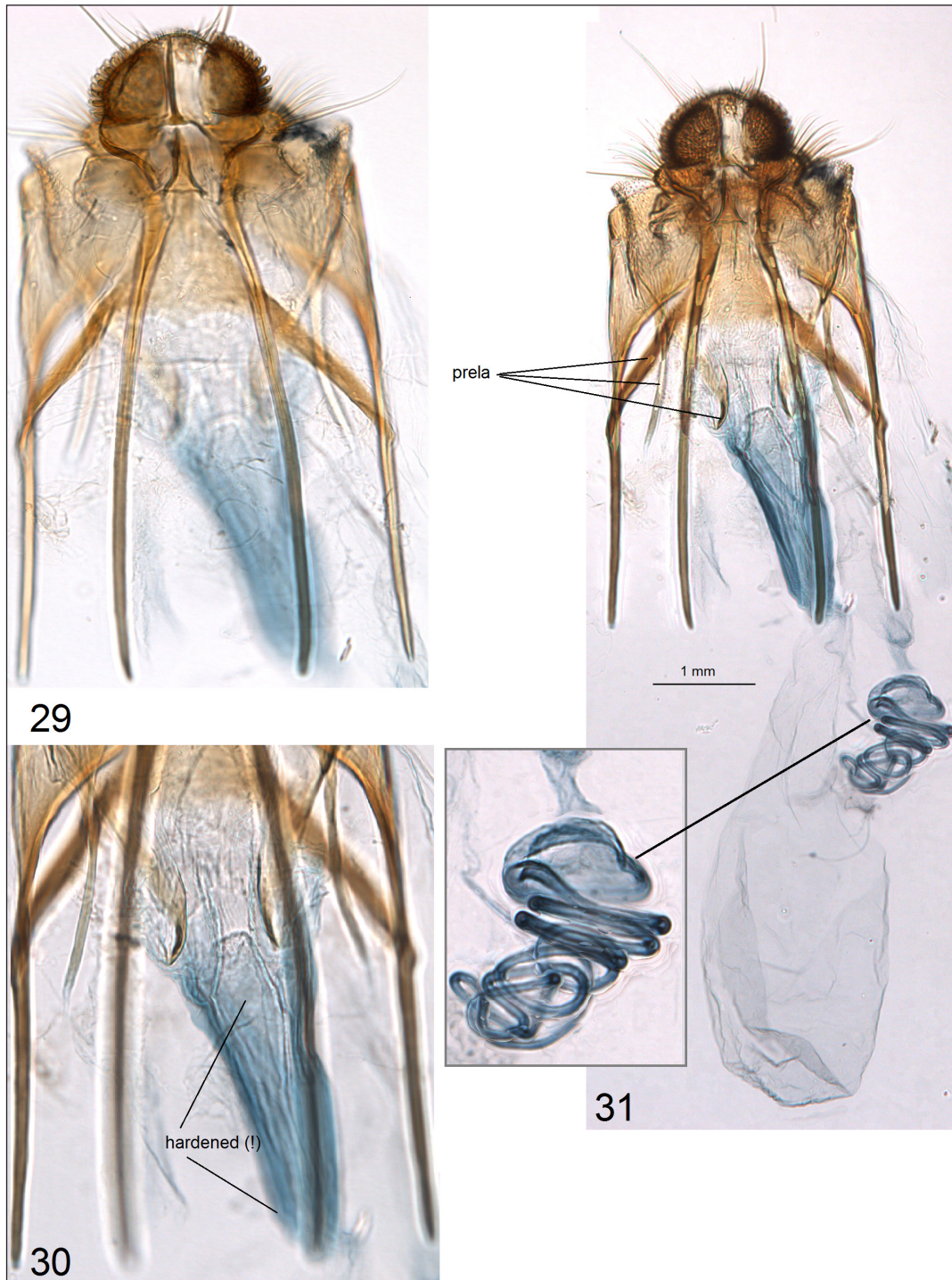
head; both from adult in the pupal skin with no pinned specimen preserved), AD840♀ (genitalia) (ZMUC); 1 ♂, 1 ♀, same locality as holotype, mining larvae on *Phenax hirtus*



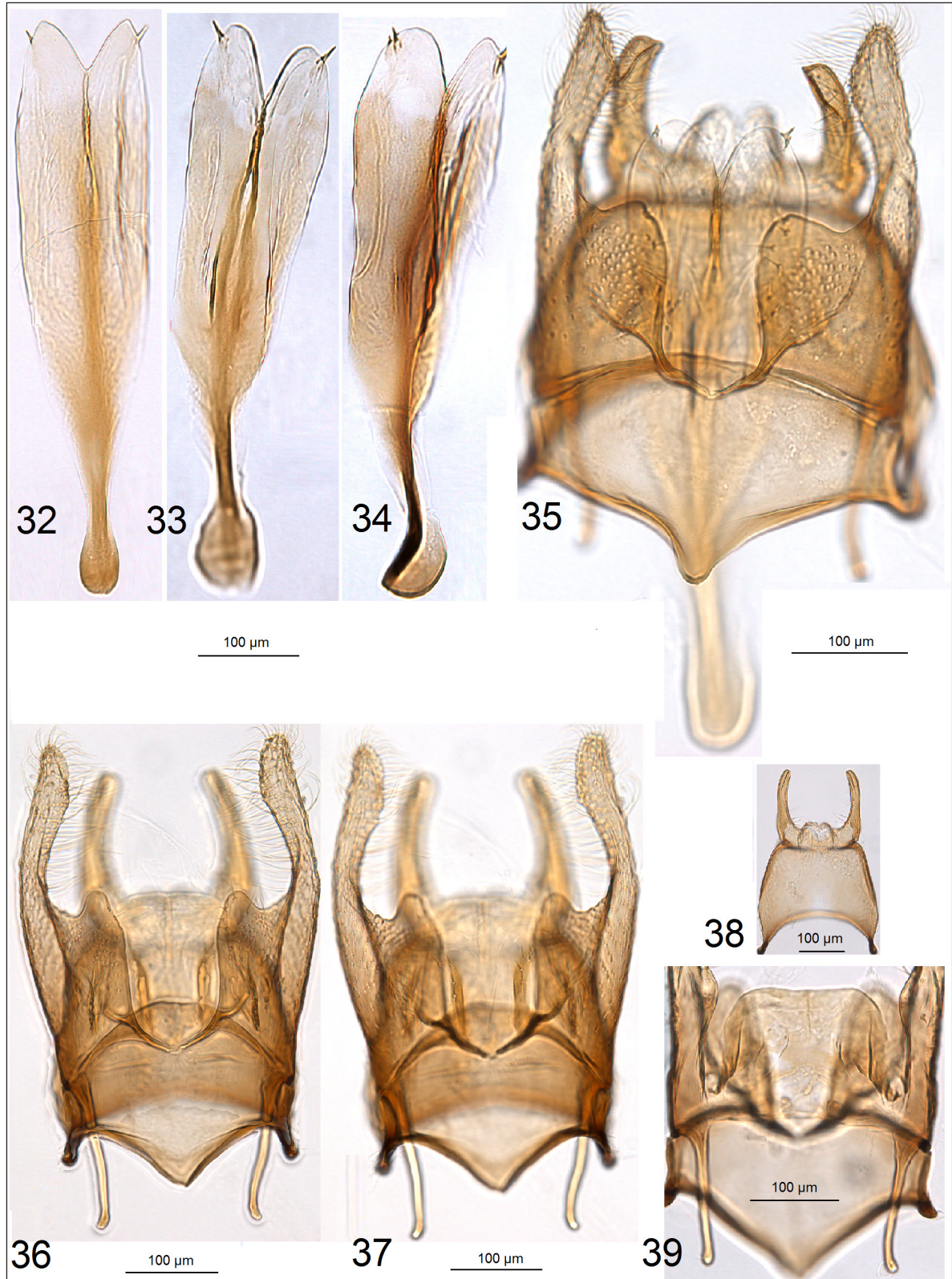
Figs. 20–28. *Paratischeria ferruginea* Diškus & Stonis, sp. nov.: 20, 22 – pupae; 21, 23–25 – pupal exuviae; 26 – adult male, holotype; 27 – adult female, paratype AD840; 28 – adult male, paratype AD841 (ZMUC)

24.ii.2007, field card no. 4894, Diškus and Stonis, slide no. AD841♂ (dissected genitalia) (ZMUC); 1 ♂, 1 ♀, Loja Province, 5 km SW Saraguro, Washapamba Forest Reserve,

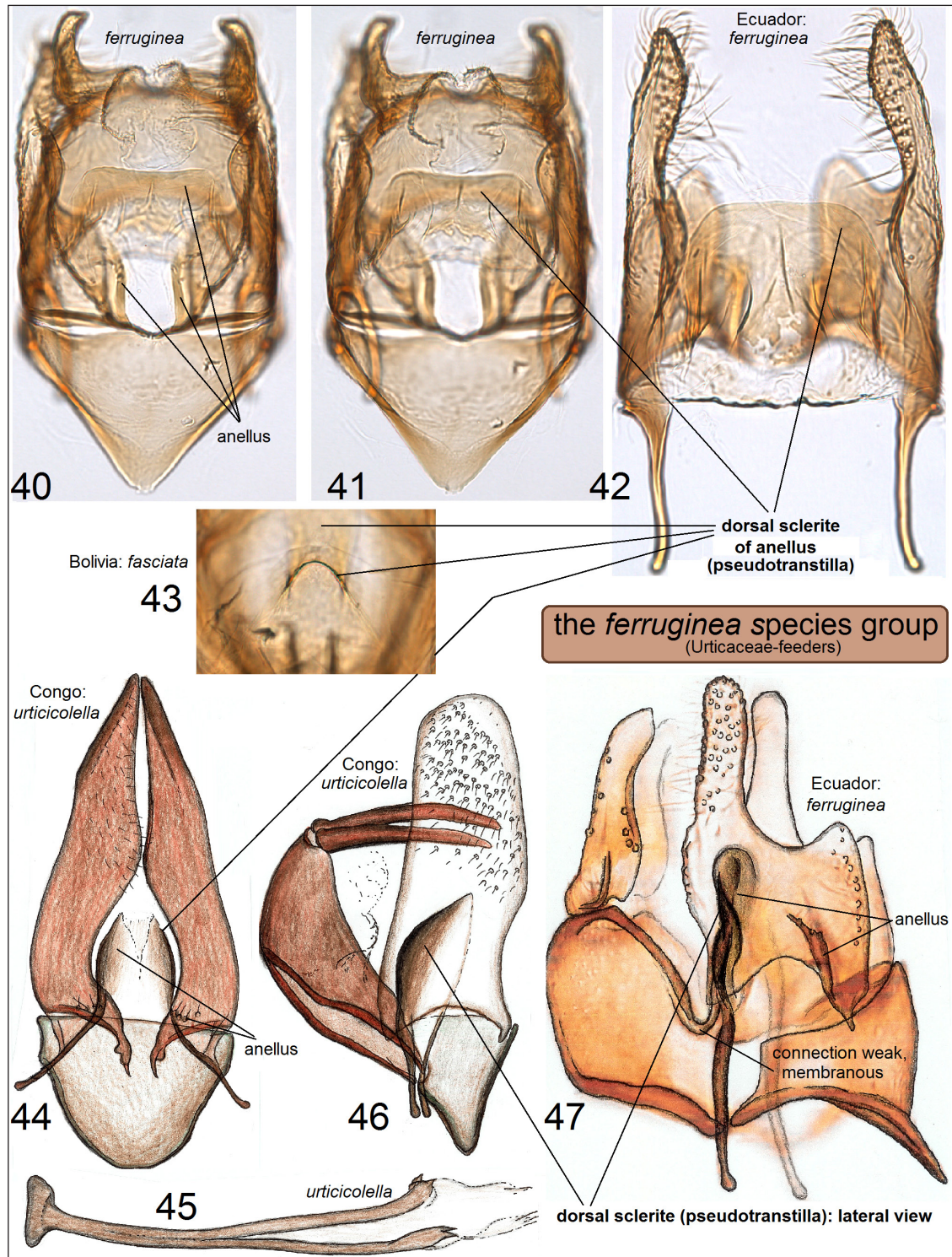
3°39'43"S, 79°16'9"W, elevation 2940 m, mining larvae on *Phenax* sp. (probably *Ph. hirtus* or closely related species) 25.i.2017, field card no. 5233, A. Diškus (ZMUC).



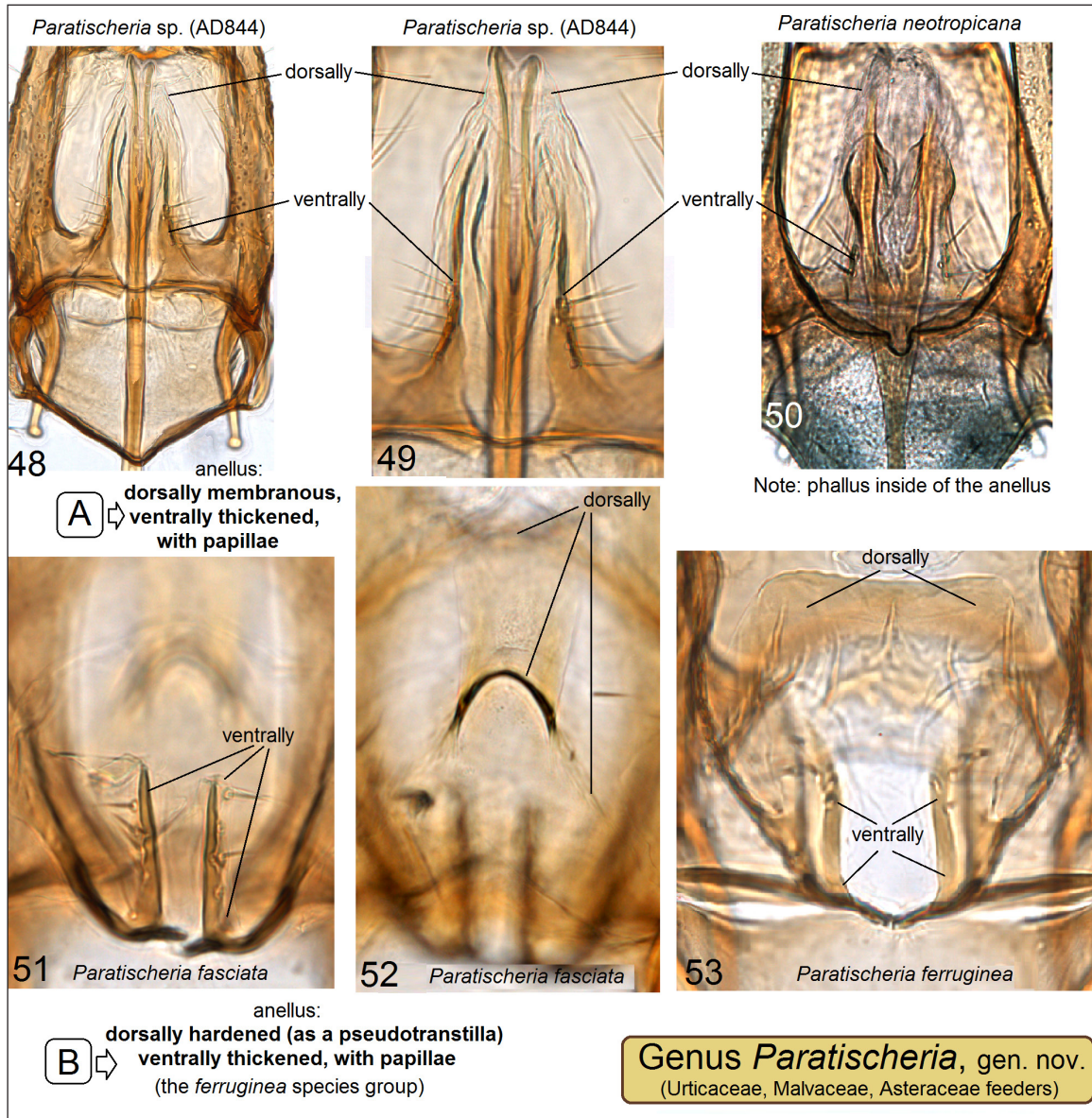
Figs. 29–31. Female genitalia of *Paratischeria ferruginea* Diškus & Stonis, sp. nov., paratype, genitalia slide no. AD840: 29 – apophyses and ovipositor; 30 – vestibulum; 31 – general view of genitalia (ZMUC)



Figs. 32–39. Male genitalia of *Paratischeria ferruginea* Diškus & Stonis, sp. nov.: 32, phallus, holotype, genitalia slide AD775; 33 – same, paratype, genitalia slide no. AD867; 34 – same, paratype, genitalia slide no. AD841; 35 – general view, AD866; 36–37 – capsule, holotype, AD775; 38 – uncus and tegumen, paratype, AD839; 39 – dorsal sclerite of anellus (pseudotranstilla), paratype, AD839 (ZMUC)



Figs. 40–47. Male genitalia of the *ferruginea* group: 40, 41 – capsule of *Paratischeria ferruginea* Diškus & Stonis, sp. nov., paratype, genitalia slide no. AD867; 42, same, genitalia slide no. AD841; 43 – dorsal sclerite of *P. fasciata* Diškus & Stonis, sp. nov., holotype, genitalia slide no. AD868 (ZMUC); 44 – ventral view of capsule of *P. urticolella* (Ghesquière, 1940) (comb. nov.), holotype, genitalia slide no. AD091 (MRAC); 45 – same, phallus; 46 – same, lateral view of capsule, slide no. AD091 (MRAC); 47 – lateral view of capsule of *P. ferruginea* Diškus & Stonis, sp. nov., paratype, genitalia slide no. AD841 (ZMUC)

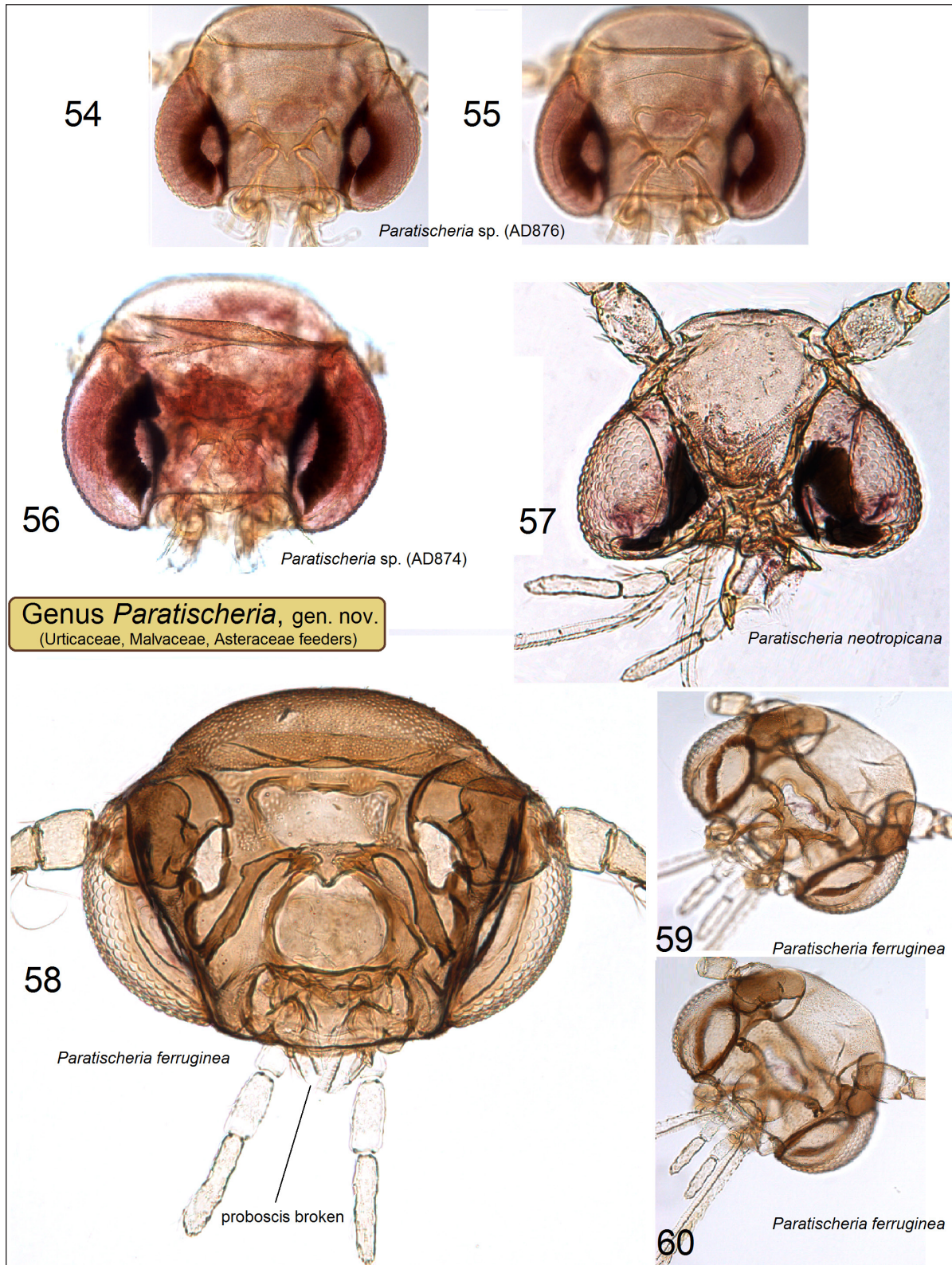


Figs. 48–53. Anellus of *Paratischeria* Diškus & Stonis, gen. nov.: 48, 49 – *P.* sp., genitalia slide AD844; 50 – *P. neotropicana* (Diškus & Stonis, 2015), comb. nov., genitalia slide AD861; 51, 52 – *P. fasciata* Diškus & Stonis, sp. nov., genitalia slide AD868; 53 – *P. ferruginea* Diškus & Stonis, sp. nov., genitalia slide AD867 (ZMUC)

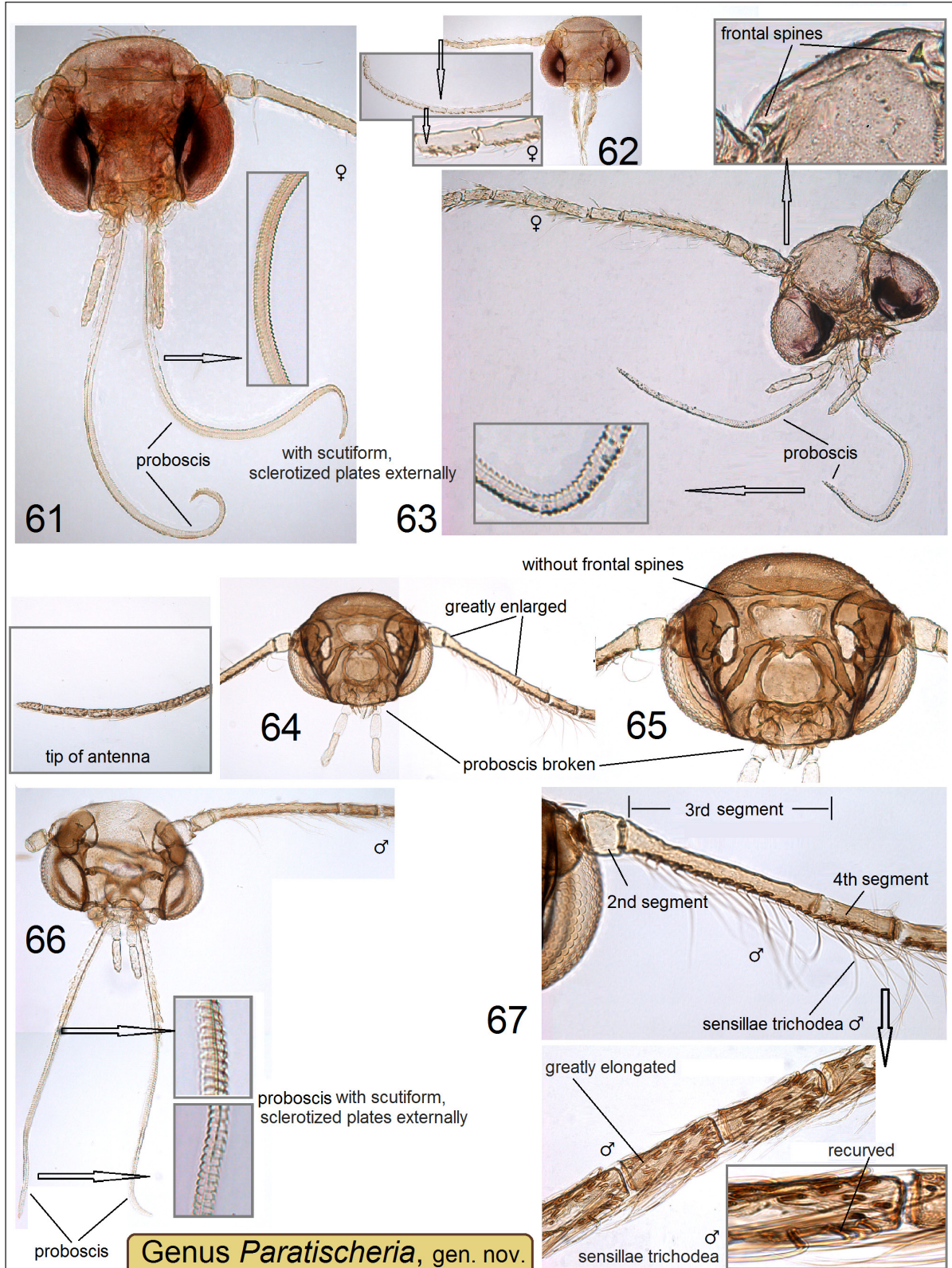
Diagnosis. Externally, the new species differs from all other known tischeriids by a unique pattern of forewing with one distinctive ochre-beige spot. In male genitalia, a specific shape of bilobed valva and dorsally developed anellus distinguish *Paratischeria ferruginea* sp. nov. from all other Tischeriidae species; the host plant *Phenax hirtus* also makes this species distinctive.

Male (Figs. 26, 28). Forewing length 4.3–5.0 mm; wingspan 9.1–10.7 mm. Head: face

dark grey-brown with golden gloss and purple iridescence; palpi and proboscis ochre; frontal tuft grey-brown with golden gloss and little or strong purple iridescence; collar grey-brown with golden gloss and sometimes with strong purple iridescence; antenna significantly longer than half the length of forewing, grey-brown with some golden gloss on upper side, paler on underside, with rather indistinctive hair-like sensillae trichodea. Thorax and tegula



Figs. 54–60. Descaled head of *Paratischeria* Diškus & Stonis, gen. nov.: 54, 55 – *P.* sp., slide no. AD876♀ [5116]; 56 – *P.* sp., slide no. AD874♀; 57 – *P. neotropicana* (Diškus & Stonis, 2015), comb. nov., slide no. AD861; 58 – *P. ferruginea* Diškus & Stonis, sp. nov., paratype, slide no. AD865; 59, 60, same, paratype, dissected from adult in pupal skin, slide no. AD867 (ZMUC)



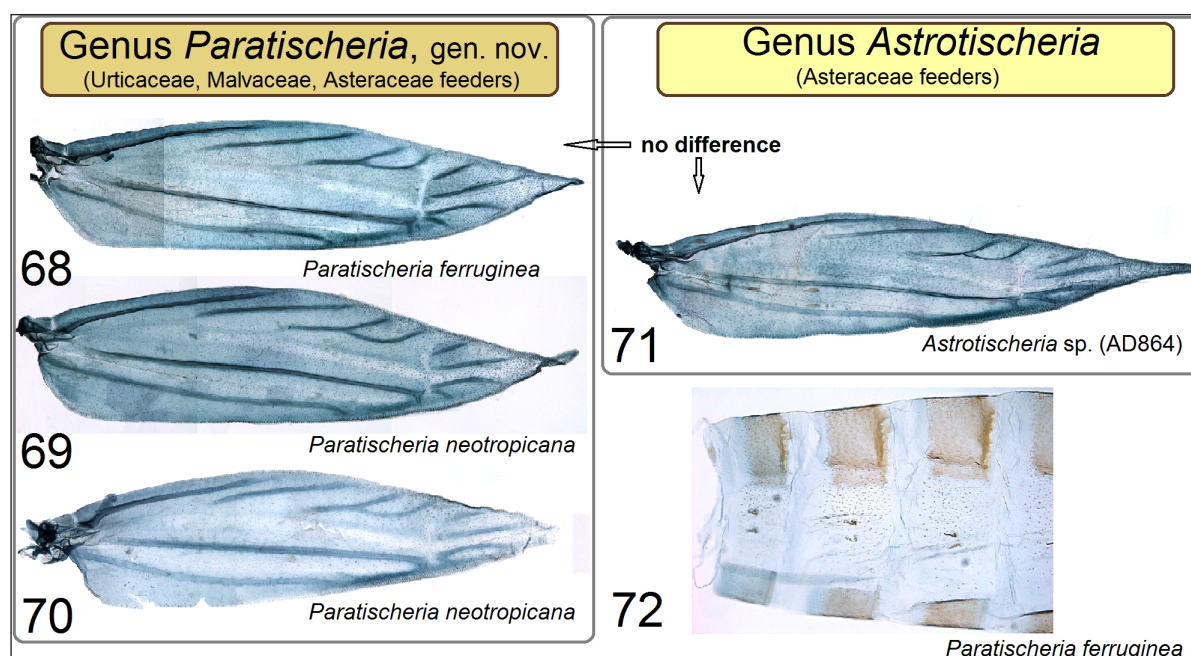
Figs. 61–67. Descaled head of *Paratischeria* Diškus & Stonis, gen. nov.: 61 – *P.* sp., slide no. AD874♀ [5109]; 62 – *P.* sp., slide no. AD876♀ [5116]; 63 – *P. neotropicana* (Diškus & Stonis, 2015), comb. nov., slide no. AD861; 64, 65 – *P. ferruginea* Diškus & Stonis, sp. nov., paratype, slide no. AD865; 66, same, paratype, slide no. AD867; 67 – same, paratype, slide no. AD865 (ZMUC)

dark grey-brown to black-brown, with golden gloss and strong purple iridescence. Forewing dark grey-brown with some purple and blue iridescence; median spot along costal margin, ochre beige (ferruginous), usually large and very distinctive, occasionally small and indistinctive; fringe grey-brown with some golden gloss (particularly prominent at costa), distinctly ochre at the very apex of forewing; underside of forewing dark grey-brown, without spots except small, irregular scaleless patch at the base. Hindwing glossy, grey-brown to olive grey on the upper side, grey-brown on underside, without androconia or spots except very small irregular scaleless patch at the base on underside; fringe grey-brown. Legs glossy, grey-brown, except femur of middle legs which distinctly ochre beige (ferruginous); tibia of hindlegs with a large tuft of ochre yellow pili-form scales. Abdomen dark brown with some golden gloss on the upper side, bright yellow ochre to orange yellow and glossy on underside; genital segments dark grey-brown; anal tufts indistinctive.

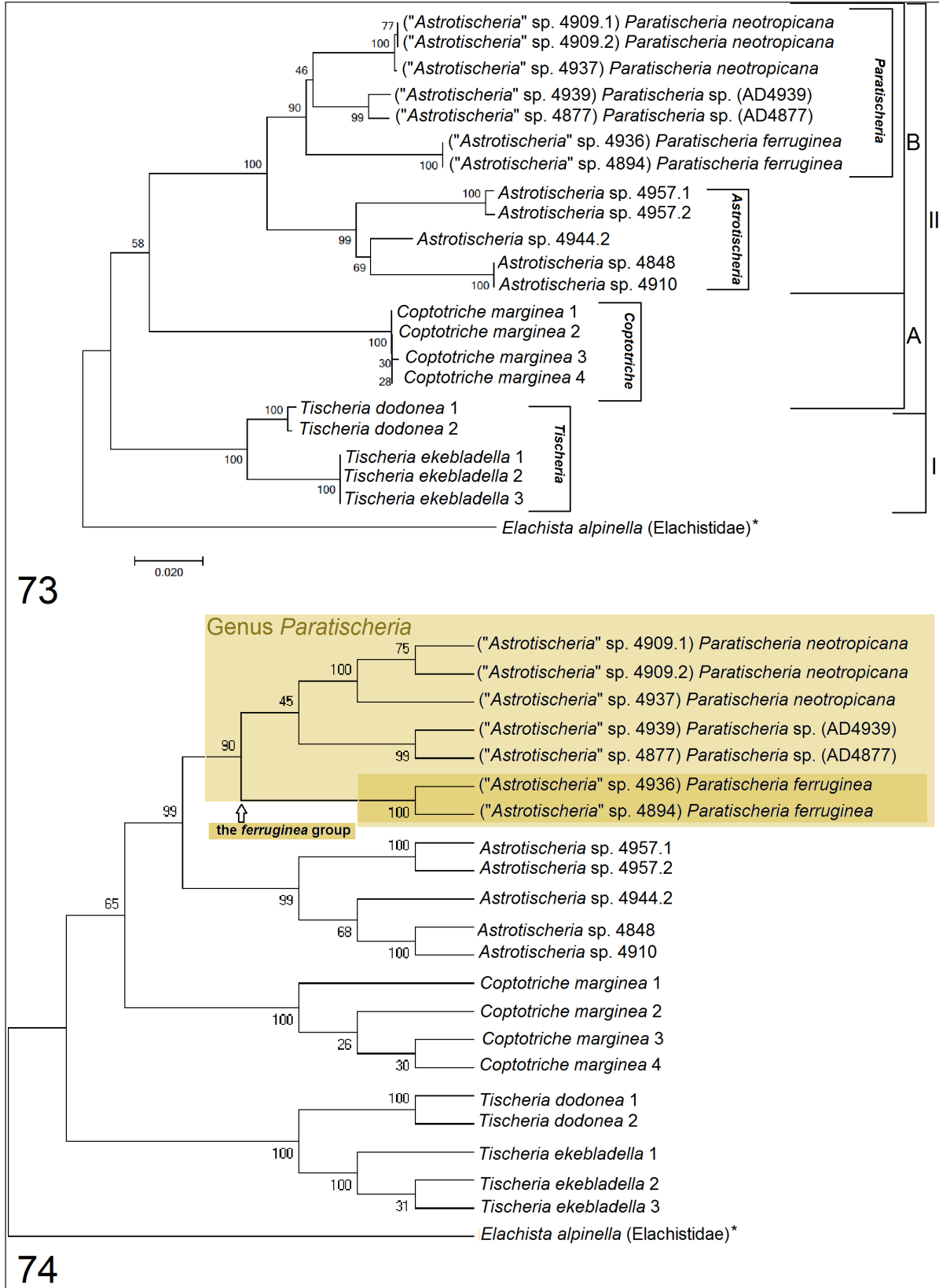
Female (Fig. 27). Forewing length 4.0–4.5 mm; wingspan 8.7–9.7 mm. Antenna shorter, sensillae trichodea indistinctive, 8–12 distal segments greyish white. Legs dark grey on the upper side, mostly ochre beige on underside. Abdomen dark brown on the upper side, bright yellow ochre yellow and glossy on underside. Otherwise as in male.

Male genitalia (Figs. 32–42, 47, 53). Capsule about 420–470 μm long, 305 μm wide. Uncus with two long lateral lobes. Valva about 320–375 μm long, slender in distal half, with wide lobe basally; transtilla absent. Anellus slightly thickened and with setae laterally (Fig. 40), dorsally with a specific plate-like sclerite (pseudotranstilla) (Figs. 41, 42, 47). Vinculum triangular. Phallus (Figs. 32–34) 500–570 μm long, distally 140–150 μm wide, distinctly bilobed; each lobe with a tiny spine.

Female genitalia (Figs. 29–31). Ovipositor clothed in short, stout and darker, modified setae which we refer to as ‘peg setae’ (Fig. 29). Second pair of lobes, lateral and anterior to the ovipositor lobes, are three times smaller, bearing



Figs. 68–72. Forewing venation and abdominal pelt: 68, forewing venation, *Paratischeria ferruginea* Diškus & Stonis, sp. nov., slide no. AD863; 69 – same, *P. neotropicana* (Diškus & Stonis, 2015), comb. nov., slide no. AD862; 70 – same, slide no. AD861; 71 – same, *Astrotischeria* sp., slide no. AD864; 72 – fragment of abdominal pelt, lateral view, *P. ferruginea* Diškus & Stonis, sp. nov., slide AD841 (ZMUC)



Figs. 73, 74. Molecular phylogeny of Tischeriidae: 73 – neighbour-joining tree for specimens of Tischeriidae with *Elachista alpinella* (GU248257.1) as outgroup; bootstrap values are shown above; 74 – same, other presentation

long slender setae. Anterior and posterior apophyses very long and slender (Fig. 29). Additionally there are three rod-like and plate-like projections (of possibly modified 8th and 9th sternites) collectively referred to as the prela described by Braun (1972) (Figs. 30, 31). Ductus bursae considerably narrower than corpus bursae, without spines but with a hardened plate (Fig. 30). Corpus bursae membranous, relatively very small (500 µm long, 200 µm wide) (Fig. 31), without spines or signum. Ductus spermatahaecae with many large coils (Fig. 31).

Bionomics (Figs. 11–25). Host plant: *Phenax hirtus* (Sw.) Wedd. (Urticaceae) (Fig. 11). Larvae mine in late January–February and October–November. Leaf-mine as a blotch (at the early stage triangular or distally widely rounded, later very irregular), without frass. Larva pale greenish grey, with pale grey-brown intestine. Pupation inside of the leaf-mine without cocoon; pupa brown (Figs. 20–25). Exit slit on the upper side of the leaf. Adults known from February, March, and December. According to the “Formula of Determining of Abundance and Occurrence of Leaf-miners” (see Diškus & Stonis 2012: 52–54), *P. ferruginea* is not a rare species: limited in distribution, but abundant mining in the locality where it has been recorded.

Distribution (Figs. 1, 3). This species occurs in the Ecuadorian Andes in tropical montane and cloud forests at altitudes about 2940–3100 m.

Etymology. The species name is derived from the Latin *ferruginea* (brown, yellowish brown) in reference to the distinctive yellow beige spot of the forewing and same ferruginous abdomen on underside.

DISCUSSION

Within the family Tischeriidae, analysis of morphological characters indicates at least four main lineages of a generic rank (Stonis et al., in prep.).

During our molecular studies, partial fragment of the COI gene were amplified in 21 specimens of Tischeriidae resulting in a 577-bp fragment. The sequences have 147 polymorphic nucleotides. 145 nucleotides were found to

be parsimony-informative characters. All four lineages indicated by morphological studies were well supported by our molecular data; see the cladogram (NJ tree) in Figs. 73, 74. The NJ tree of Tischeriidae (Fig. 74) has two big clusters: I and II. The cluster I includes species from the genus *Tischeria*: European *T. dodonaea* and *T. ekebladella*. Among these two species, 24 polymorphic nucleotides were detected.

Cluster II is formed of two groups: the first group (A) is represented by *Coptotriche* (European *Coptotriche marginea*) and the second group (B) by the species of South American *Astrotischeria* and *Paratischeria*. The COI gene sequences of investigated specimens from cluster B indicated 52 polymorphic and 525 conservative nucleotides.

There is a difference between the cladogram published by Diškus, Puplesis (2003b) and our NJ tree (Fig. 74). Previously two clades, *Tischeria* and *Astrotischeria*, were shown as sibling (Diškus, Puplesis, 2003); in our NJ tree, the sibling group is *Coptotriche* and *Astrotischeria* + *Paratischeria*; other major cluster presents the genus *Tischeria*.

Our study has identified several distinct groups which are supported by the COI gene region (it also has shown that this particular gene is suitable for the reconstruction of the Tischeriidae phylogeny). However, more molecular data and further studies are necessary to confirm the detected clades in our preliminary cladogram.

The conducted specific molecular research provides a theoretical and practical framework for a further use of DNA molecular markers in taxonomy and phylogenetic studies of Tischeriidae.

ACKNOWLEDGEMENTS

For helpful and frequent discussions on various host plants (not only those which are listed in the current paper) we thank Nixon Cumbicus Torres (Universidad Técnica Particular de Loja, Loja, Ecuador), Arvind Singh (Banaras Hindu University, Varanasi, India); Narayanan Nair Mohanan (Jawaharlal Nehru Tropical Botanic

Garden and Research Institute, India), José Luis Fernández-Alonso (Universidad de Salamanca, Spain), Theodor C. H. Cole (Universität Heidelberg, Germany), Maximilian Weigend (University of Bonn, Germany), and Franz Starlinger (Federal Research and Training Centre for Forests, Natural Hazards and Landscape, Vienna, Austria).

Further we would like to thank Andrius Reimekis (Nature Research Centre, Lithuania) for providing us with photographs of genitalia and adults of Tischeriidae.

Received 3 January 2017

Accepted 30 March 2017

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**URTICACEAE AUGALUS MINUOJANTYS
TISCHERIIDAE: DVI NAUJOS MOKSLUI
RŪŠYS IR NAUJA PARATISCHERIA GENTIS**

Santrauka

Šiame straipsnyje pirmą kartą publikuojami duomenys apie Urticaceae šeimos augalus minuojančius Tischeriidae Pietų Amerikoje. Aprašoma nauja *Paratischeria* gentis ir dvi naujos mokslui rūšys: *Paratischeria fasciata* (aptikta Bolivijoje) ir *P. ferruginea* (aptikta Ekvadore). Šios dvi rūšys kartu su pusiaujo Afrikoje paplitusia *P. urticicolella* (Ghesquière) (comb. nov.) pirmą kartą išskiriamos į atskirą *Paratischeria* genties rūšių grupę, kuriai būdingas pakitęs patino *anellus* ir dilgėlinių augalų (*Phenax*, *Fleurya*) minavimas.

Raktažodžiai: Andai, nauja gentis, nauja rūšis, *Paratischeria*, Pietų Amerika, *Phenax* Wedd., Tischeriidae, Urticaceae