



## New leaf-mining Nepticulidae (Lepidoptera): potential pests of aromatic Lamiaceae plants from equatorial Andes

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### Abstract

Members of the Lamiaceae, or mint family, are used worldwide for medicinal, culinary and/or magical-religious purposes, as well as in pesticides and as ornamental plants. Very little is known about nepticulids, or pygmy moths, as leaf miners of Lamiaceae, but they may be an important component of South American diversity and potential pests of economically-important species of the mint family. In this paper, four new species of leaf-mining Nepticulidae are described from the equatorial Andes of Ecuador: *S. mentholica* Diškus & Stonis, **sp. nov.**, *Stigmella aromatica* Diškus & Stonis, **sp. nov.**, *S. odora* Diškus & Stonis, **sp. nov.**, feeding on *Minthostachys mollis* (Benth.) Griseb., and *S. tomentosella* Diškus & Stonis, **sp. nov.**, feeding on *Clinopodium tomentosum* (Kunth) Govaerts. It is hypothesized that host-plant distribution ranges can provide clues to potential distribution ranges of these newly discovered, trophically specialized leaf miners. The leaf mines, adults, and the genitalia of the new species are illustrated with photographs.

**Key words:** the Andes, *Clinopodium axillare*, *Clinopodium tomentosum*, Ecuador, *Minthostachys mollis*, Mentheae, new species, pygmy moths, the *singularia* group, *Stigmella*

### Introduction

Some species of Nepticulidae, or pygmy moths, have been considered pests of wild and cultivated plants (e.g., Payne *et al.* 1973, Hyerdahl & Dutcher 1985, Kuznetsov & Puplesis 1994, Alford 2012, Remeikis 2017, Stonis *et al.* 2019b). Pygmy moth larvae are plant miners, usually leaf miners, living under the epidermis and producing slender, gallery-like or blotch-like leaf mines, or a combination of an early, slender gallery in early instars that expands to an irregular blotch later in its development (for details of Nepticulidae biology, see the reviews by Johansson *et al.* (1990), Puplesis (1994), and Remeikis *et al.* (2016)). Although nepticulid adults are among the smallest lepidopterans on the planet and many of them are less than 4 mm in wingspan (Stonis *et al.* 2021), a leaf mine produced by a tiny, pygmy moth larva can be relatively large on a single leaf, conspicuous, and easy to spot. It can also cause significant damage to a leaf, particularly to smaller leaves, because the larva consumes much of the photosynthetic tissues. Usually, leaf mines are not very abundant, but upon a significant population increase or explosion, damage to the entire host plant may be severe. For example, in Far East Russia Puplesis (1985) reported about 400–500 leaf mines of *Ectoedemia picturata* Puplesis, 1985 on a single compound leaf of cultivated *Rosa rugosa*, which is the source of edible rose hips high in vitamin C (Dimitrov *et al.* 1980). Larval abundance was so high that full development of the larvae was impossible, and foliage of observed plants lost their green color, or photosynthetic parenchyma, and became brown. Additionally, based upon our field observations, at least in the case of some Palaearctic

species of *Fomoria* Beirne, mines can also cause premature abscission of leaves, and defoliation of trees can cause a reduction in yield of a fruit product (e.g., Hyerdahl & Dutcher 1985, Alford 2012).

It was hypothesized (Remeikis *et al.* 2016) that Lamiaceae feeders may be a significant component of the Neotropical fauna of leaf-mining Nepticulidae. However, until now, only three Lamiaceae-feeding Nepticulidae species have been described from South America: the central Andean *Stigmella clinopodiella* Diškus & Stonis, 2016, feeding on *Clinopodium axillare* (Rusby) Harley in Bolivia (Stonis *et al.* 2016), the northern Andean *S. lamiacifoliae* Remeikis & Stonis, 2017, feeding on *Salvia palifolia* Kunth in Colombia, and *S. scutellariae* Remeikis & Stonis, 2017, feeding on *Scutellaria volubilis* Kunth in southern Ecuador (Stonis *et al.* 2017b). No other published evidence of this Lamiaceae-feeding diversity of nepticulids has been provided until now.

The discovery of Nepticulidae on Lamiaceae host plants seems important both for theoretical and applied reasons. In all continents and cultures, humans use various species of Lamiaceae plants for medicines (Floríndez 2012) or condiments for food. Lamiaceae are also widely used as ornamental plants and they are especially appreciated for their aroma; sometimes they are used for religious purposes (Fernández-Alonso & Rivera-Díaz 2006). Many Lamiaceae species are cultivated in Andean indigenous medical gardens, as well as in our orchards and gardens, both in temperate and tropical zones. This is especially true for species in the following Lamiaceae genera: *Ocimum* L., *Origanum* L., *Mentha* L., *Marrubium* L., *Plectranthus* L. Her., *Thymus* L., *Satureja* L., *Clinopodium* L., *Minthostachys* (Benth.) Spach., *Melissa* L., *Salvia* L., *Scutellaria* L., *Lepechinia* Willd., *Hyptis* Poir. (García Barrija 1975, Fernández-Alonso & Rivera-Díaz 2006, Morales 2010). Although the production of essential oils from Lamiaceae and their varied use in industry and medicine is well documented (El-Gazzar & Watson 1970, Rehm & Espig 1991), there is new recent information on different bioactive molecules from native, Andean Lamiaceae as a result of ethnobotanical documentation and prospecting in recent decades (Pérez *et al.* 1998, Stashenko 2009, Bernal *et al.* 2011). The genera *Minthostachys*, *Clinopodium*, *Salvia*, *Hyptis*, *Scutellaria* and *Lepechinia*, which are native to the Andean region, are the most commonly used as medicinals (Fernández-Alonso & Rivera-Díaz 2006, Huamantupa *et al.* 2011, Puentes *et al.*, 2020). There are several published works in the last decades specifically dealing with essential oils of the *Minthostachys* and *Clinopodium*, and their usage in traditional medicine in South America (e.g., Schmidt-Lebuhn 2008, Vera Salto 2015). Recent research has also analyzed mucilages, lectins, terpenes, and alkaloids in genera such as *Salvia*, *Hyptis*, *Lepechinia* and *Scutellaria* (Fernández-Alonso *et al.* 2009, Oliveira *et al.* 2013).

In this paper we document newly discovered feeders and potential pests of the Andean aromatic Lamiaceae and describe four new species of leaf-mining *Stigmella* Schrank: *S. mentholica* Diškus & Stonis, **sp. nov.**, *S. aromatica* Diškus & Stonis, **sp. nov.**, and *S. odora* Diškus & Stonis, **sp. nov.**, feeding on *Minthostachys*, and *S. tomentosella* Diškus & Stonis, **sp. nov.**, feeding on *Clinopodium*. For comparison, we also provide photographs of leaf mines and genitalia of the related, but previously described, *S. clinopodiella* which also feeds on *Clinopodium*.

## Material and methods

Descriptions of new species are based on material collected in Ecuador by J. R. Stonis, formerly R. Puplesis, thanks to a collecting permit from the Instituto Ecuatoriano Forestal y de Areas Naturales (INEFAN, Ecuador, 2000) and a research project in cooperation with Professor Giovanni Onore, former professor at the Pontifical Catholic University of Ecuador, Quito, Ecuador. In addition, type material of the earlier described Central Andean species, *Stigmella clinopodiella*, was available for our study from the collection of ZMUC.

The identification of the host plants was undertaken from photographic images by José L. Fernández-Alonso, a specialist of Lamiaceae and author of the current paper.

Methods and protocols for species identification and description are outlined in Puplesis (1994), Puplesis & Robinson (2000), Puplesis & Diškus (2003), Diškus & Stonis (2012), and Stonis *et al.* (2014, 2018). 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 often removed and mounted alongside the genital armature. Abdominal pelts were stained with Chlorazol Black (Direct Black 38/Azo Black) and mounted in Euparal.

Permanent slides were photographed and studied using a Leica DM2500 microscope and Leica DFC420 digital camera. The descriptive terminology of morphological structures follows Johansson *et al.* (1990) and Puplesis & Robinson (2000) except for the term “aedeagus”, which is referred here as “phallus” and the term “cilia”, which is referred here as “fringe”.

Abbreviation for specimen depository: USNM—National Museum of Natural History (NMNH), formerly the U.S. National Museum of Natural History, Washington D.C.; ZMUC—Zoological Museum, Natural History Museum of Denmark, Copenhagen, Denmark.

Abbreviation for herbariums: MA—Herbarium of the Royal Botanical Garden, Madrid, Spain; MO—the Missouri Botanical Garden Herbarium.

## Descriptions of new species

### *Stigmella mentholica* Diškus & Stonis, sp. nov.

(Figs 3, 25, 26, 31, 32–40, 63, 64, 68)

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**Type material.** Holotype: ♂, ECUADOR: SW of Quito, western slopes of the equatorial Andes, Via Aloag to Sto. Domingo, 0°26'46"S, 78°37'39"W, elevation ca. 3100 m, mining larva on *Minthostachys mollis* (Kunth) Griseb. (Lamiaceae), 24.ii.2000, ex pupa iii.2000, leg. R. Puplesis, genitalia slide no. AD697 (USNM). Paratypes: 2 ♂, 2 ♀, same label data as holotype, genitalia slides nos AD688♂, AD1033♂, AD1026♀ (USNM).

**Diagnosis.** The new species belongs to the *Stigmella singularia* species group, designated and characterized by Stonis *et al.* (2017a). Externally, males of *S. mentholica* sp. nov. differ from the most similar *S. aromatica* sp. nov. in the slender, subapical fascia of the forewing and indistinctive patch of silvery or golden shiny scales on the apex. In the male genitalia, *S. mentholica* differs from all other known Neotropical *Stigmella*, including other species of the *S. singularia* group, by the combination of a distinctly truncate uncus and very long caudal processes of the gnathos (Fig. 34). Only two other species, *S. aromatica* and *S. odora* sp. nov., feed on the same host plant *Minthostachys mollis*, but there are significant differences in the morphology of the leaf mine: in *S. mentholica*, the leaf mine is a wide, relatively short gallery (Fig. 3), while the leaf mine of *S. aromatica* is a slender and long gallery, almost fully filled with frass (Fig. 10), and the leaf mine of *S. odora* is a combination of a very slender gallery and an irregular blotch (Fig. 6).

**Male** (Figs 25, 26, 31). Forewing length 2.6–3.0 mm; wingspan 5.8–6.7 mm (n = 3). Head: palpi cream; frontal tuft dark orange; scape golden cream; collar golden cream but at certain angle may look pale grey, metallic glossy; antenna distinctly longer than half the length of forewing; flagellum with 36 segments, dark grey to brown-grey or pale grey, with golden gloss and some purple iridescence. Thorax and tegula golden brown. Forewing pale golden brown, strongly shining, at base with intense purple iridescence; fascia subapical, slender, comprised of silvery or golden shiny scales; apex of forewing brown-purple, with indistinctive patch of silvery or golden shiny scales; fringe grey; underside of forewing black-brown, without spots or androconia, except for slender scaleless spot at base. Hindwing pale grey to dark brown depending from angle of view, without androconia; fringe grey. Legs glossy grey cream; on upper side, covered with grey to brown-black scales with purple iridescence. Abdomen blackish grey to grey-brown on upper side and underside; genital segments pale brown; anal tufts short but distinctive, dark grey to fuscous.

**Female.** Smaller than male; forewing length 2.3–2.4 mm; wingspan 5.2–5.4 mm (n = 2). Palpi and scape snow white. Frontal tuft pale yellow. Fascia of forewing post median-subapical, glossy white, distinctive. Abdomen fuscous to grey-brown on upper side, ochreous grey to grey-brown on underside; genital segments cream-ochre on underside, ovipositor pointed. Otherwise as in male.

**Male genitalia** (Figs 32–40). Capsule longer (215–220 µm) than wide (195–200 µm). Vinculum with short, pointed lateral lobes. Uncus truncate caudally (Figs 33, 34). Gnathos with two very long, close-set caudal processes and a slender plate (Fig. 34). Valva (Fig. 38) 210–215 µm long, 75–80 µm wide, with slender apical process; trans-tilla with sublateral processes (Figs 35, 36). Juxta membranous, indistinctive. Phallus (Figs 37, 39, 40) 270–335 µm long, 85–90 µm wide; vesica with about 15–16 large spine-like cornuti and some minuscule cornuti (Fig. 37).

**Female genitalia** (Figs 63, 64). Total length 1070–1080 µm. Anterior apophyses gradually narrowing distally; posterior apophyses very slender, longer than anterior ones, distally bent (Fig. 63). Vestibulum without sclerites. Corpus bursae with a strongly folded distal part and oval-shaped basal part with many indistinctive pectinations. Accessory sac very small, indistinctive; ductus spermathecae without coils, but with a small, tube-like vesicle.

**Bionomics** (Figs 1–3). Host plant is *Minthostachys mollis* (Benth.) Griseb., Lamiaceae: Mentheae (Figs 1, 2).

Larva pale green with brown-green intestine and pale brown head; feeds in February and probably in late January (note that in late February some leaf mines are already vacant). Prefer open, sunny areas. Leaf mine (Fig. 3) is a wide, sinuous, gradually widening gallery (easy to spot because the generally whitish appearance); at the beginning, with an interrupted line of brown-black frass, further on, with a wide margin, unfilled with the frass. Cocoon brown. Adults probably fly in March (indoors, emerged in March).

**Distribution** (Fig. 68). This species is known from a single locality in Ecuador, on the western slopes of the equatorial Andes, at elevation of ca. 3100 m, but the host plant has a much wider distribution in the northern and central Andes (see Discussion).

**Etymology.** The species name is derived from *menthol*, one of the principal components of essential oil of its host plant, *Minthostachys mollis*, in reference to the strong mint smell of its leaf mines.

### ***Stigmella aromatica* Diškus & Stonis, sp. nov.**

(Figs 9–11, 22–24, 28–30, 41–47, 65, 66, 68)

lsid:zoobank.org:act:42F003AD-AAE4-4A52-AD3B-E2DE745B8E19

**Type material.** Holotype: ♂, ECUADOR: SW of Quito, western slopes of the equatorial Andes, Via Aloag to Sto. Domingo, 0°26'46"S, 78°37'39"W, elevation ca. 3100 m, mining larva on *Minthostachys mollis* (Benth.) Griseb. (Lamiaceae), 24.ii.2000, ex pupa iii.2000, leg. R. Puplesis, genitalia slide no. AD1022 (USNM). Paratypes: 9 ♂, 6 ♀, same label data as holotype, genitalia slides nos AD815♂, AD819♂ (from adult in pupal skin, pinned adult unavailable), AD1021♂, AD1032♂, AD1027♀, AD1031♀ (USNM).

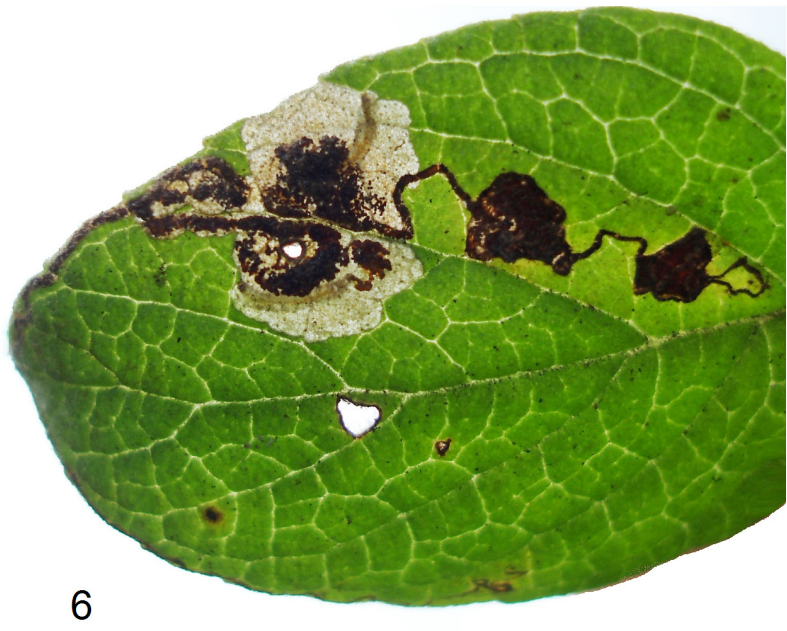
**Diagnosis.** The new species belongs to the *Stigmella singularia* species group, designated and characterized by Stonis *et al.* (2017a). Externally, males of *S. aromatica* **sp. nov.** differ from the most similar *S. mentholica* **sp. nov.** in the wide, median fascia of the forewing and distinctive apical fascia. In the male genitalia, *S. aromatica* differs from all other known Neotropical *Stigmella*, including species of the *S. singularia* group, by the combination of an uncus with two unique caudal lobes (Fig. 41), a large gnathos with two very close-set caudal processes (Fig. 43), a bifid juxta (Fig. 44), and a set of large, spine like cornuti (Figs 45–47). The host plant, *Minthostachys mollis*, also make this species distinctive among other *Stigmella* species, except for *S. mentholica* and *S. odora* **sp. nov.** However, *S. aromatica* differs from both species in the morphology of the leaf mine: the leaf mine of *S. aromatica* is a long, slender, sinuous gallery almost entirely filled with frass (Fig. 10), while the leaf mine of *S. mentholica* is a relatively short and wide gallery (Fig. 3), and the leaf mine of *S. odora* is combined of a very slender gallery and irregular blotch (Fig. 6).

**Male** (Figs 22–24, 30). Forewing length 2.0–2.5 mm; wingspan 4.5–5.6 mm (n = 5). Head: palpi golden cream; frontal tuft large, dark orange; scape golden cream; collar golden cream but at certain angle may look metallic grey; antenna about 2/3 of the length of forewing; flagellum with 33–37 segments, brown-grey, golden glossy. Thorax and tegula concolorous with the forewing base. Forewing pale golden brown, strongly shining, with some purple iridescence along costa; postmedian fascia wide, comprised of silvery or golden shiny scales; apex of forewing brown with strong purple iridescence, and with a distinctive apical fascia of silvery or golden shiny scales (occasionally apical fascia is indistinctive, see Figs 28, 29); fringe grey, glossy, distally whitish; underside of forewing dark grey-brown, without spots or androconia, except for a slender, scaleless spot at base. Hindwing grey to dark grey, without androconia; fringe grey. Legs glossy cream to grey; on upper side, covered with dark grey-brown scales. Abdomen blackish grey with purple iridescence on upper side, brown-grey on underside; genital segments pale brown; anal tufts short but distinctive, grey.

**Female.** Smaller than male; forewing length 2.1–2.3 mm; wingspan 4.7–5.2 mm (n = 5). Antenna shorter than in male, only slightly longer than half the length of forewing. Abdomen grey-brown on upper side and underside; genital segments pale brown; anal tufts short but distinctive, blackish grey to grey; ovipositor pointed. Otherwise as in male.

**Male genitalia** (Figs 41–47). Capsule longer (290–300 µm) than wide (175–210 µm). Vinculum with short, pointed lateral lobes. Uncus with unique caudal lobes (Fig. 41). Gnathos large, with two very close-set caudal processes (Fig. 43). Valva (Fig. 44) 200 µm long, with pointed, partially divided apical process (Fig. 44); transtilla without sublateral processes (Fig. 42). Juxta triangular, distally split (Fig. 44). Phallus (Figs 45–47) 210–330 µm long, 90–115 µm wide; vesica with about numerous large spine-like cornuti and a lateral set of small cornuti (Fig. 45).





**FIGURES 1–6.** Bionomics of new *Stigmella* species feeding on Lamiaceae plants. 1, 2, *Minthostachys mollis* (Benth.) Griseb var. *mollis*, a host plant; 3, leaf mine of *S. mentholica* Diškus & Stonis, **sp. nov.** on *Minthostachys mollis*, Ecuador, SW of Quito, Via Aloag to Sto. Domingo, ca. 3100 m; 4–6, leaf mines of *S. odora* Diškus & Stonis, **sp. nov.** on *M. mollis*, Ecuador, Baños, slopes of Tungurahua, ca. 1860 m





FIGURES 7–11. Bionomics of *Stigmella aromatica* Diškus & Stonis, **sp. nov.** 7, 8, host plant *Minthostachys mollis* (Benth.) Griseb var. *mollis*, Lamiaceae; 9–11, leaf mines, Ecuador, SW of Quito, Via Aloag to Sto. Domingo, ca. 3100 m





**FIGURES 12–17.** Bionomics of *Stigmella tomentosella* Diškus & Stonis, **sp. nov.** 12, 13, host plant *Clinopodium tomentosum* (Kunth) Govaerts, Lamiaceae; 14–17, leaf mines, Ecuador, Baños, slopes of Tungurahua, ca.1860 m

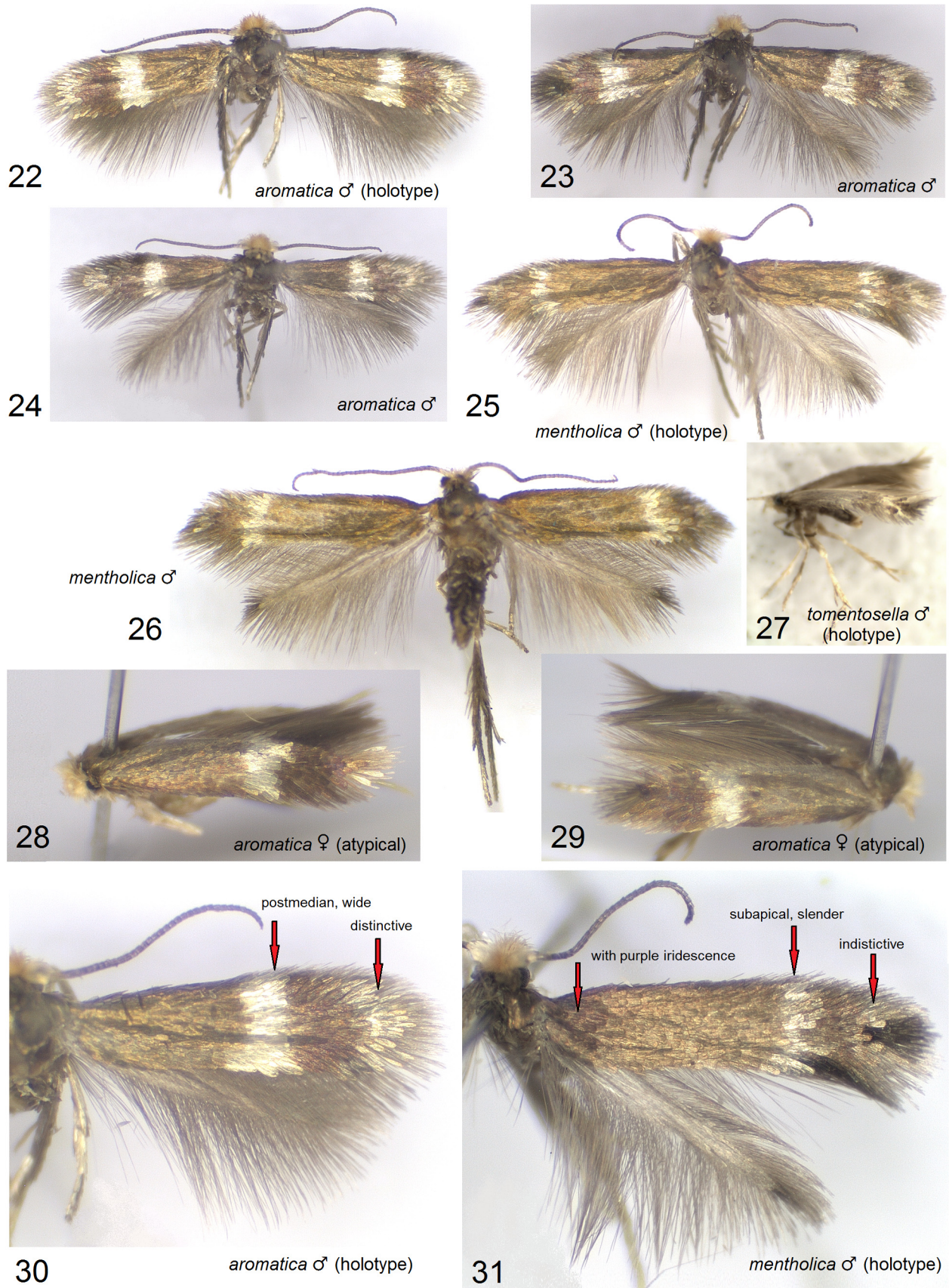




**FIGURES 18–21.** Bionomics of *Stigmella clinopodiella* Diškus & Stonis, 2016. 18, host plant *Clinopodium axillare* (Rusby) Harley, Lamiaceae: Menthae; 19–21, leaf mines, Bolivia, Copacabana (southern shore of Lake Titicaca), ca. 3930 m.

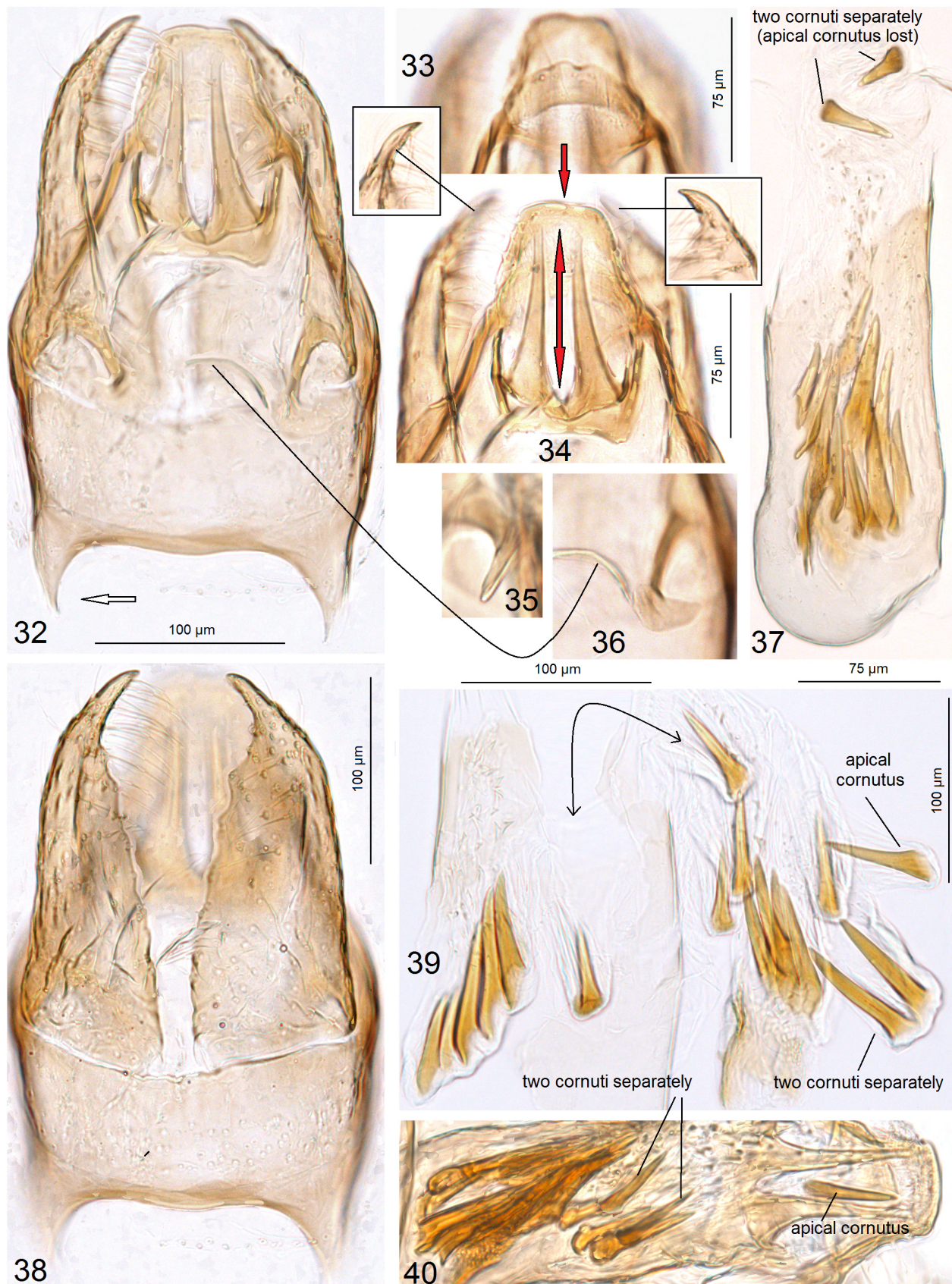
**Note.** Figure 18 most likely represents *Clinopodium axillare* subsp. *uniflorum* (Rusby ex Briq.) J.R.I. Wood. The only difference of note is that the locality of our sample is somewhat higher in elevation (3930 m) than those so far known for this species (3000–3300 m) in herbarium material and the literature (Wood 2011). However, there are frequent intermediate forms and possible hybrids which are difficult to assign with precision to a taxon (Schmidt-Lebuhn 2008)





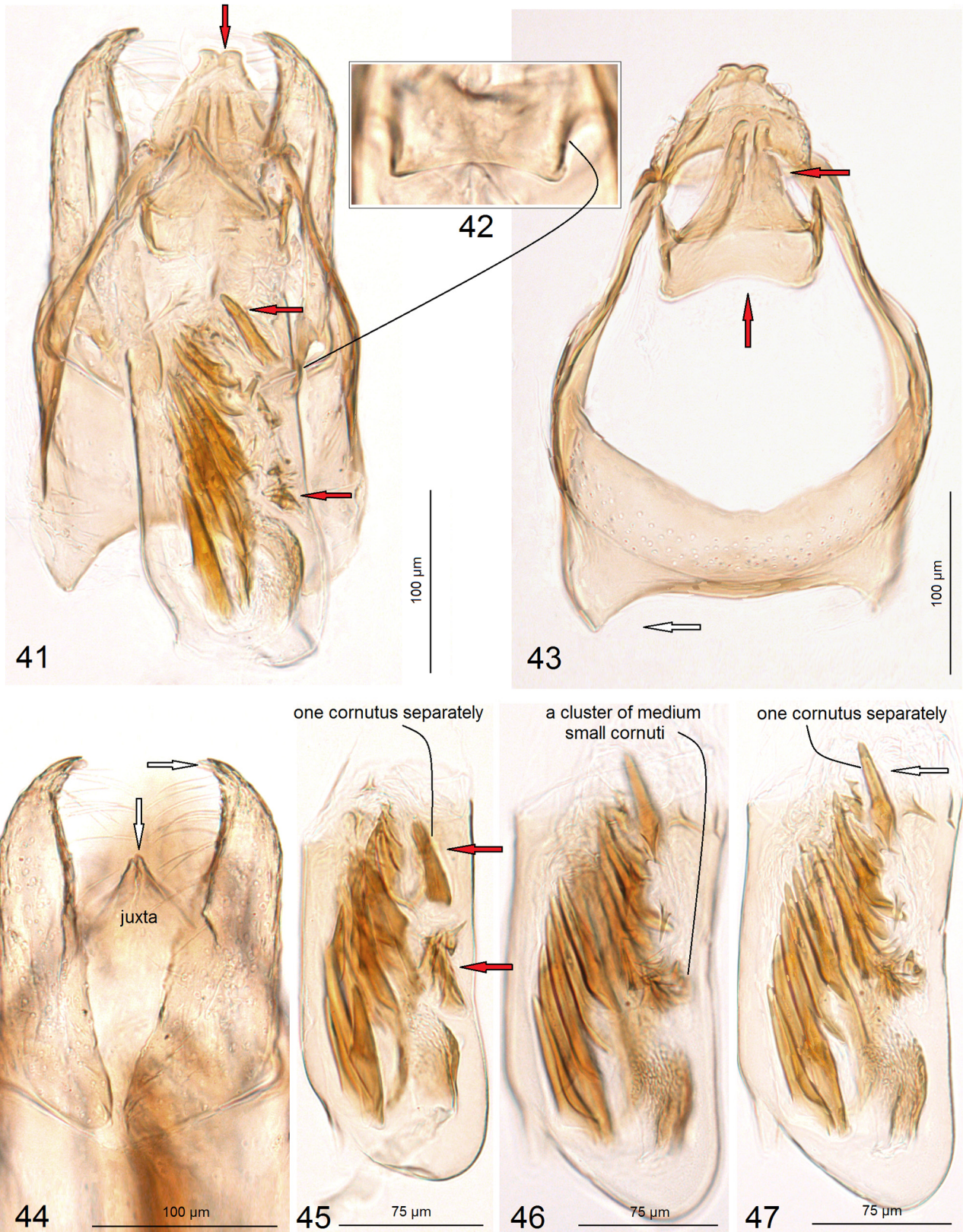
FIGURES 22–31. Adults of new *Stigmella* species. 22–24, *S. aromatica* Diškus & Stonis, **sp. nov.**; 25, 26, *S. mentholica* Diškus & Stonis, **sp. nov.**; 27, *S. tomentosella* Diškus & Stonis, **sp. nov.**; 28–30, enlarged right side of *S. aromatica*, **sp. nov.**; 31, enlarged right side of *S. mentholica*, **sp. nov.** (USNM)





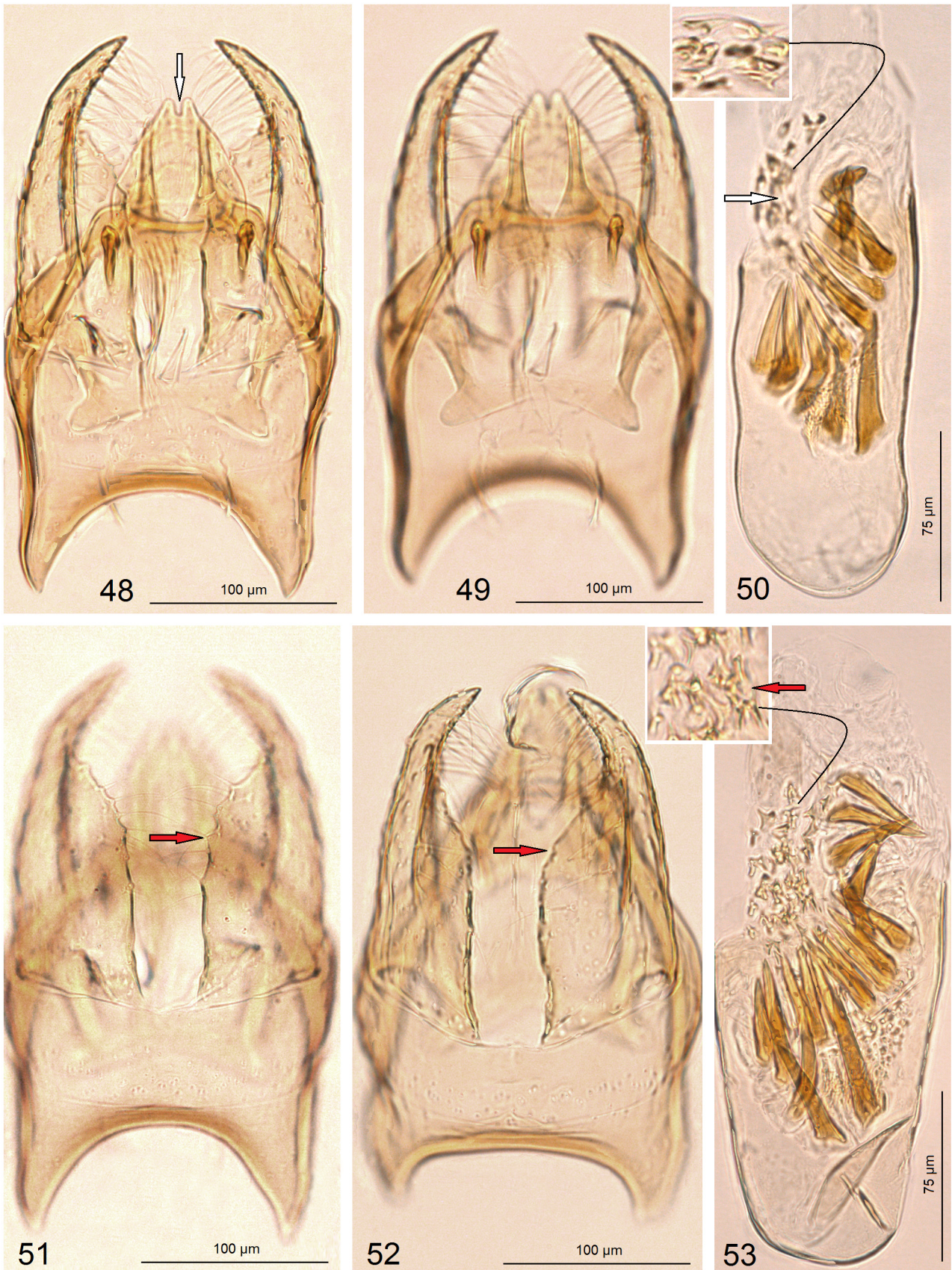
**FIGURES 32–40.** Male genitalia of *Stigmella mentholic* Diškus & Stonis, **sp. nov.** 32, slide AD697, holotype, capsule with phallus removed; 33, same, tegumen; 34, same, uncus and gnathos; 35, slide AD1033, paratype, left sublateral process of transtilla; 36, slide AD697, holotype, right sublateral process of transtilla; 37, slide AD688, paratype, phallus; 38, slide AD697, holotype, valvae; 39, same, cornuti in torn phallus; 40, slide AD1033, paratype, phallus inside genital capsule (USNM)





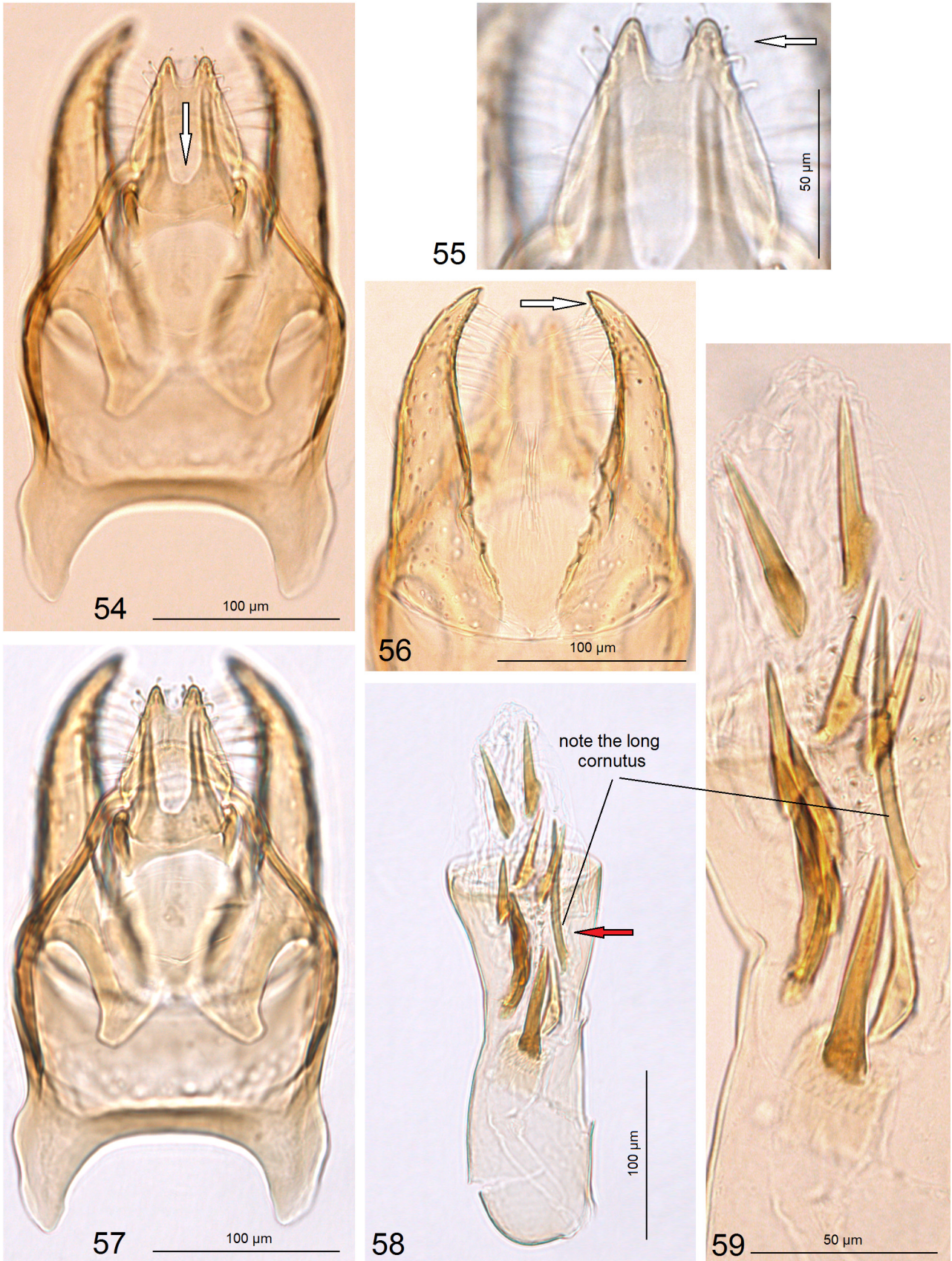
**FIGURES 41–47.** Male genitalia of *Stigmella aromatica* Diškus & Stonis, **sp. nov.** 41, slide AD1022, holotype; 42, slide AD1032, paratype, transtilla; 43, slide AD1021, paratype, capsule with valvae and phallus removed; 44, slide AD1022, holotype, valvae and juxta; 45, slide AD815, paratype, phallus; 46, 47, slide AD1021, paratype, phallus (USNM)





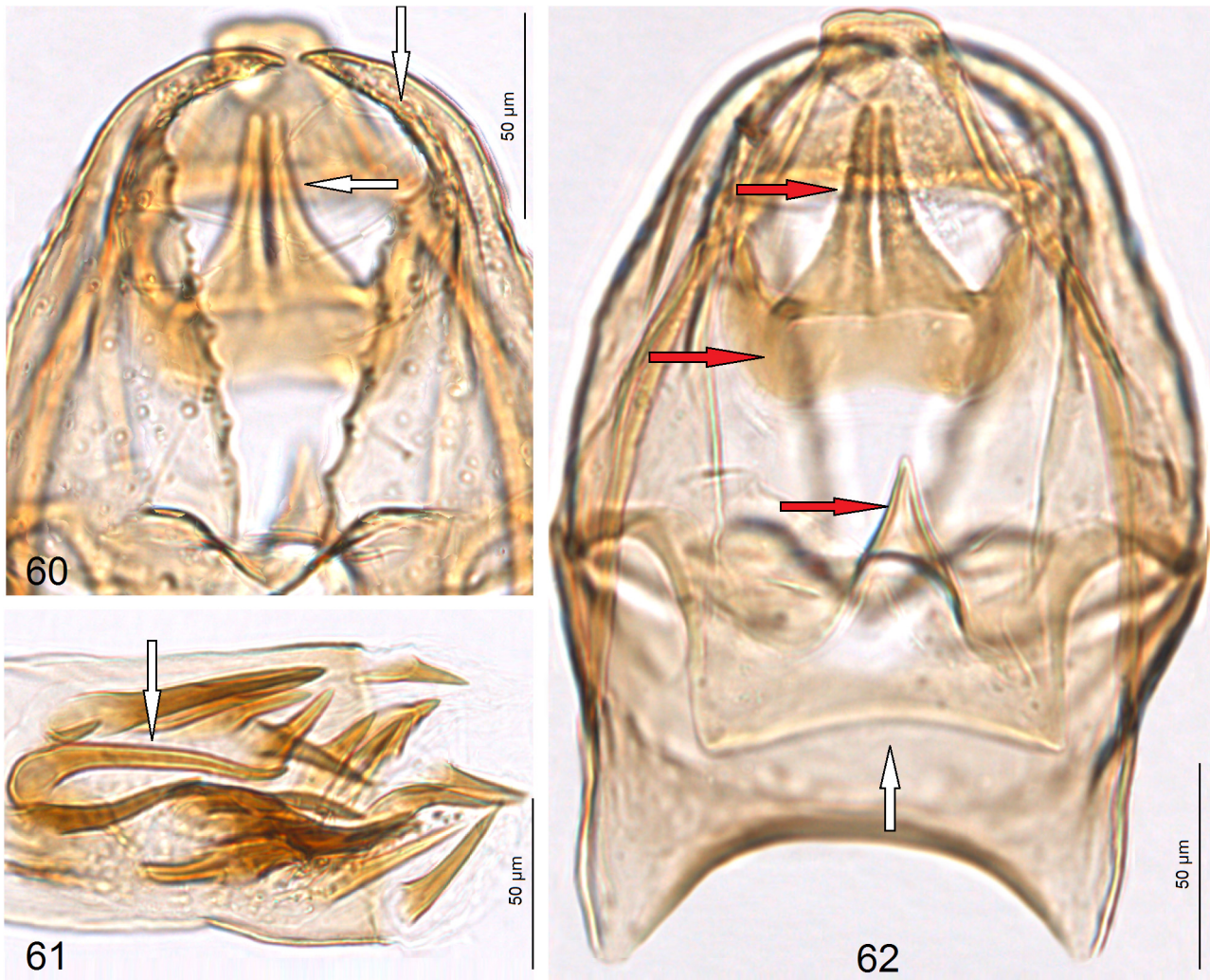
**FIGURES 48–53.** Male genitalia of *Stigmella odora* Diškus & Stonis, **sp. nov.** 48, slide AD685, paratype, capsule with phallus removed; 49, same, focused on gnathos and transtilla; 50, same, phallus; 51, same, capsule, focused on valvae and vinculum; 52, slide AD821, holotype, capsule with phallus removed; 53, same, phallus (USNM)





**FIGURES 54–59.** Male genitalia of *Stigmella tomentosella* Diškus & Stonis, **sp. nov.** 54, slide AD796, holotype, capsule with phallus removed; 55, same, uncus; 56, same, valvae; 57, same, capsule, focused on gnathos and transtilla; 58, 59, same, phallus (USNM)





**FIGURES 60–62.** Male genitalia of *Stigmella clinopodiella* Diškus & Stonis, 2016 (after Stonis *et al.* 2016, modified). 60, slide AD646, holotype, valvae; 61, same, cornuti; 62, same, capsule with phallus removed (ZMUC).

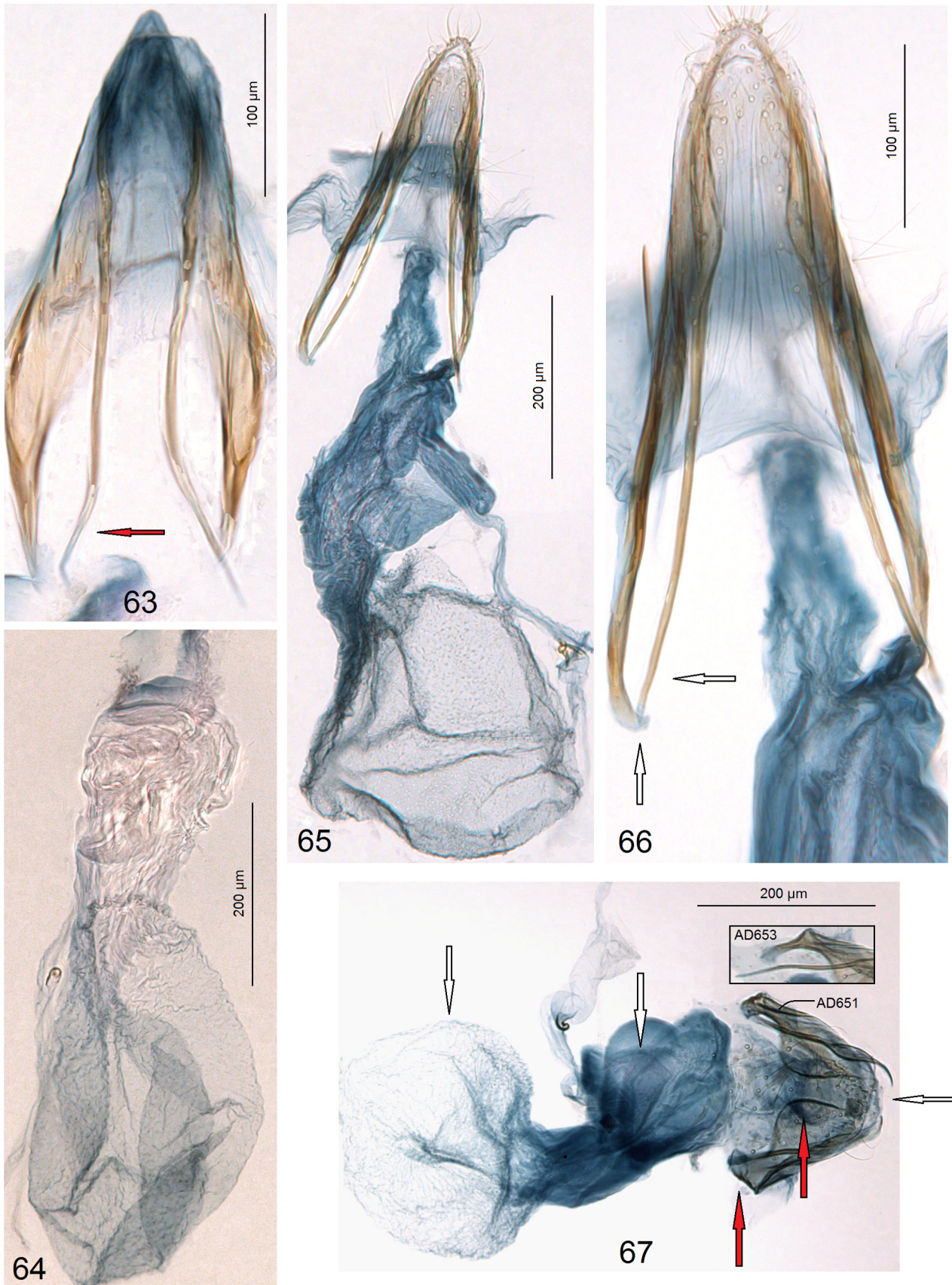
**Female genitalia** (Figs 65, 66). Total length 995–1000 µm. Anterior apophyses gradually narrowing and bent distally; posterior apophyses slender, approximately as long as anterior ones (Fig. 66). Vestibulum without sclerites. Corpus bursae with a strongly folded distal part and round or oval-shaped basal part with many distinctive pectinations. Accessory sac small; ductus spermathecae without coils, but with a small, tube-like vesicle.

**Bionomics** (Figs 7–11). Host plant is *Minthostachys mollis* (Kunth) Griseb., Lamiaceae: Mentheae (Figs 7, 8). Larva yellow with indistinctive, yellowish brown intestine and pale brown head; feeds in February and probably in late January (note that in late February most of the leaf mines are already vacant). Prefer to occur in shady places. Leaf mine (Figs 9–11) is a long, slender, sinuous gallery almost entirely filled with black frass (Fig. 10); in old, vacant leaf mines, frass may turn brown or black (Fig. 9). Cocoon brown. Adults probably fly in March (indoors, emerged in March).

**Distribution** (Fig. 68). This species is known from a single locality in Ecuador, on the western slopes of the equatorial Andes, at elevation of ca. 3100 m, but the host plant has a much wider distribution in the northern and central Andes (see Discussion).

**Etymology.** The species name is derived from Latin *aromaticus* (aromatic), due to the minty aroma of essential oil of the host plant, *Minthostachys mollis*, and its leaf mines.





FIGURES 63–67. Female genitalia of *Stigmella* species. 63, 64, *S. mentholica* Diškus & Stonis, **sp. nov.** genitalia slide no. AD1026 (USNM); 65, 66, *S. aromatica* Diškus & Stonis, **sp. nov.**, paratype, genitalia slide no. AD1027 (USNM); 67, *S. clinopodiella* Diškus & Stonis, 2016, paratype, genitalia slide no. AD651, with apophyses of AD653 (ZMUC)



***Stigmella odora* Diškus & Stonis, sp. nov.**

(Figs 4–6, 48–53, 68)

Isid:zoobank.org:act:5BEE514E-FA26-4B23-A414-BC1F8FDB12F

**Type material.** Holotype: ♂, ECUADOR: SE of Ambato, Baños de Agua Santa, on the slopes of the Tungurahua volcano, 1°24'13"S, 78°25'43"W, elevation ca. 2080 m, mining larva on *Minthostachys mollis* (Benth.) Griseb. (Lamiaceae), 10.ii.2000, leg. R. Puplesis, genitalia slide no. AD821♂ (pinned adult is lost) (USNM). Paratypes: 2♂, same label data as holotype, genitalia slides nos AD685♂ (pinned adult is lost), AD806♂ (from adult in pupal skin, pinned adult unavailable) (USNM).

**Diagnosis.** The new species belongs to the *Stigmella singularia* species group, designated and characterized by Stonis *et al.* (2017a). In the male genitalia, *S. odora* sp. nov. differs from the similar *S. mentholica* sp. nov. and *S. aromatica* sp. nov. by the unique uncus (see Fig. 48), very slender plate of the gnathos, presence of numerous small cornuti (Fig. 50), and the wide inner lobe and large apical process of the valva (Figs 51, 52). The host plant, *Minthostachys mollis*, makes this species distinctive among *Stigmella* species, except for *S. mentholica* and *S. aromatica*. However, *S. odora* differs from both species in the morphology of the leaf mine: the leaf mine of *S. odora* is a combination of a very slender gallery and an irregular blotch (Fig. 6), while the leaf mine of *S. aromatica* is a slender gallery almost entirely filled with frass (Fig. 10), and the leaf mine of *S. mentholica* is a relatively short and wide gallery (Fig. 3).

**Male.** External characters are unknown (see Remarks).

**Female.** Unknown.

**Male genitalia** (Figs 48–53). Capsule longer (260–280 µm) than wide (175–180 µm). Vinculum with short or moderately long, triangular lateral lobes. Uncus gradually narrowing towards apex, with characteristic caudal lobes (Fig. 48). Median plant of gnathos very slender, with two slender caudal processes (Fig. 48). Valva (Figs 51, 52) 170–190 µm long, with a large apical process and wide inner lobe (Fig. 52); transtilla with short sublateral processes (Fig. 49). Juxta membranous, indistinctive. Phallus (Figs 50, 53) 275–280 µm long, 80–100 µm wide; vesica with a set of numerous large spine-like cornuti and many small, triangular cornuti (Fig. 53).

**Bionomics** (Figs 1, 2, 4–6). Host plant is *Minthostachys mollis* (Benth.) Griseb., Lamiaceae: Mentheae (Figs 1, 2). Larva pale, grey yellowish green with indistinctive, ochre-brown intestine and dark brown head; feeds in February. Leaf mine (Figs 4–6) is combined of a long, very slender gallery and a large, irregular blotch; the slender gallery is heavily contorted in the initial part and entirely or almost entirely filled with black-brown frass (Fig. 6); the blotch-like part of the leaf mine is with medially scattered black-brown frass (Figs 4, 5). Adults fly in February and March.

**Distribution** (Fig. 68). This species is known from a single locality in equatorial Andes, on the slopes of the Tungurahua volcano (Ecuador: Baños) at the elevation of ca. 2100 m, but the host plant has a much wider distribution in the northern and central Andes (see Discussion).

**Etymology.** The species name is derived from Latin *odoros* (odorous, scented), in reference to the minty-scented host plant and leaf mines of *Stigmella odora*.

**Remarks.** The new species is described based on the genitalia of three male specimens and the leaf mines only. The pinned adult of the holotype was lost, and the two paratypes are from adults in pupal skins, and therefore pinned specimens are unavailable. Although external characters of the species remain unknown, the new species possesses unique male genitalia and a particularly unique morphology of the leaf mine. We describe this species for the record and hope that the adults will be discovered in the future.

***Stigmella tomentosella* Diškus & Stonis, sp. nov.**

(Figs 12–17, 27, 54–59, 69)

Isid:zoobank.org:act:D19FFC7B-9E35-4997-88CA-65AD65907308

**Type material.** Holotype: ♂, ECUADOR: SE of Ambato, Baños de Agua Santa, on the slopes of the Tungurahua volcano, 1°24'5"S, 78°25'45"W, elevation 1980 m, mining larva on *Clinopodium tomentosum* (Kunth) Govaerts (Lamiaceae), 9.ii.2000, leg. R. Puplesis, genitalia slide no. AD796 (adult documented but lost) (USNM).

**Diagnosis.** The new species belongs to the *Stigmella singularia* species group, designated and characterized by



Stonis *et al.* (2017a). In the male genitalia, *S. tomentosella* **sp. nov.** differs from the similar *S. odora* **sp. nov.** and other species of the group by the unique, long and very slender cornutus, as well as the combination of a slender valva, large median element of the gnathos, and a transtilla with short sublateral processes. The host plant *Clinopodium tomentosum* makes this species distinctive among *Stigmella* species, except for the Bolivian *S. clinopodiella* Diškus & Stonis, 2016 that feeds on the related host plant, *Clinopodium axillare* (Rusby) Harley (Fig. 18) (See Remarks below). However, *S. tomentosella* differs in the morphology of the leaf mine: the leaf mine of the new species is slender, almost entirely filled with brown-black frass (Fig. 15); the leaf mine of *S. clinopodiella* is a combination of a long, very slender gallery and an elongated blotch with wide margin unfilled with frass (Figs 19–21).

**Male** (Fig. 27). Forewing length 2.1 mm; wingspan 4.6 mm (n = 1). Head: palpi brownish cream; frontal tuft dark orange; scape and collar brownish cream; antenna (flagellum) brownish grey. Thorax and tegula concolorous with the forewing base. Forewing densely speckled with brown and dark brown scales; fascia ill-defined, subapical, comprised of brownish white scales; fringe pale brown; underside of forewing brown, without spots or androconia. Hindwing brownish grey, without androconia; fringe grey. Legs brownish cream; on upper side, covered with black-grey scales. Abdomen blackish grey on upper side, brownish cream on underside; genital segments cream; anal tufts distinctive, cream, half the width of terminal segment of the abdomen.

**Female.** Unknown.

**Male genitalia** (Figs 54–59). Capsule longer (290 µm) than wide (170 µm). Vinculum with distinctive, thickened, slightly angular lateral lobes (Fig. 57). Uncus gradually narrowing towards apex, with characteristic caudal lobes (Fig. 55). Gnathos with a moderately large median plate and two slender caudal processes (Fig. 54). Valva (Fig. 56) 180 µm long, slender, gradually narrowing to an apical process (Fig. 56); transtilla with short sublateral processes (Fig. 57). Juxta membranous, indistinctive. Phallus (Figs 58, 59) about 295 µm long, 55 µm wide in the middle and 90 µm wide at apical part; vesica with a set of about ten large spine-like cornuti; one of them is very slender and long (Fig. 59).

**Bionomics** (Figs 12–17, 69). Host plant is *Clinopodium tomentosum* (Kunth) Govaerts, Lamiaceae: Mentheae (Figs 12, 13). Larva pale, brownish yellow with an indistinctive intestine and pale brown head; feeds in February and in January (note that in early February most of leaf mines are already vacant). Leaf mine (Figs 14–17) is a gradually widening gallery, almost entirely filled with black-brown frass (Fig. 15). Cocoon beige to ochre-beige, 1.9–2.2 mm long, 1.1–1.4 mm wide (n = 4). Adults fly in February and probably in early March.

**Distribution** (Fig. 69). This species is known from a single locality in equatorial Andes, on the slopes of the Tungurahua volcano (Ecuador: Baños) at elevation of ca. 2000 m, but the host plant has a wider distribution in the equatorial Andes (see Discussion).

**Etymology.** The species is named after the host plant, *Clinopodium tomentosum*.

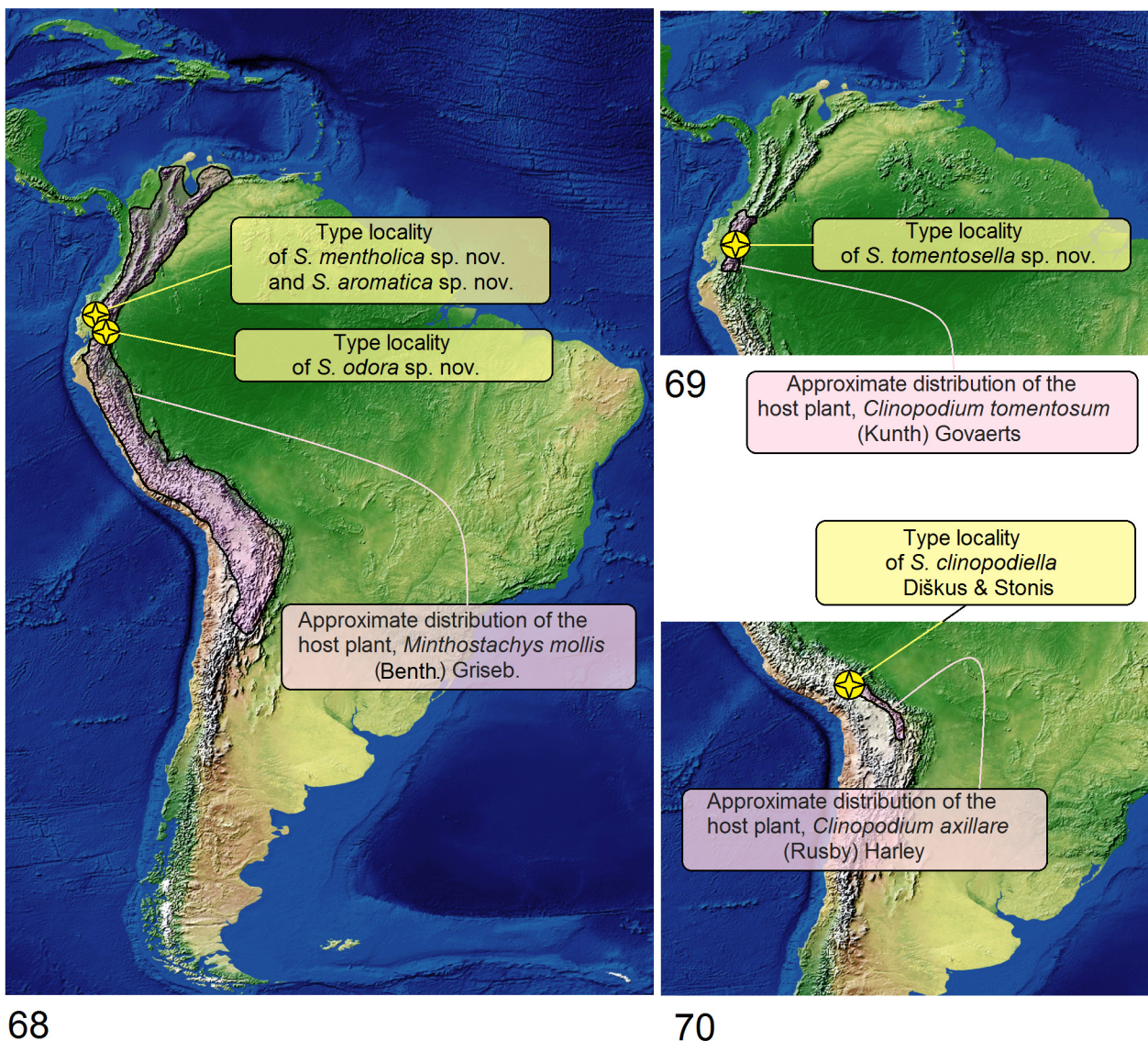
**Remarks.** *Stigmella clinopodiella* Diškus & Stonis, discovered feeding on a congeneric host plant to this species, was described from Bolivia (Fig. 70), and illustrated by Stonis *et al.* 2016: 79–82. The species also belongs to the *Stigmella singularia* species group. In the male genitalia (Figs 60–62), *S. clinopodiella* differs from all other species of the group in the combination of a large gnathos with two very close-set caudal processes, truncate uncus, large apical process of the valva, triangular juxta and long cornuti of the phallus. In the female genitalia, *S. clinopodiella* is characterized by the angular anterior apophyses (Fig. 67) and the rounded basal part of the bursa copulatrix.

## Discussion

It is well-known that leaf-mining larvae of Nepticulidae show great selectivity in their food choice: they are predominantly monophagous, and to a lesser degree, oligophagous. Therefore, although the distribution of these phytophagous insects may be dependent on many other factors than host plant alone, we recently hypothesized that the distribution of host plants can suggest much broader ranges for the host-specific leaf miners (Stonis *et al.* 2019a, 2020). In accordance with this, we provide (Figs 68–70) potential distribution ranges of all five species treated in this paper.

The leaf-mining nepticulids newly described here are potential pests of two, beneficial plant genera of the Andean aromatic Lamiaceae: *Minthostachys* and *Clinopodium*. Plants of South American *Minthostachys* species are commonly used throughout the Andes as medicinals to treat ailments of respiratory and digestive systems, as a

condiment, pest repellent, and preservative for tuber, and other fresh foods (Schmidt-Lebuhn 2008). *Minthostachys mollis* is known in the region as “muña”, “pennyroyal”, “oregano”, and “typo”. There are several studies on composition of its oils and usage of *M. mollis* in the northern Andes (Alkire *et al.* 1994, Ojeda *et al.* 2004, Zapata *et al.* 2009).



**FIGURES 68–70.** Predicted host-specific *Stigmella* distribution from host plant distribution. 68; approximate distribution of *Minthostachys mollis* (Benth.) Griseb., including var. *mollis*, and varieties *hybrida* Schmidt-Leb. in Colombia and Venezuela, and *mandoniana* (Briq.) Schmidt-Leb. in Perú and Bolivia, based on a review by Schmidt-Lebuhn (2008) and data from the herbarium of MA from Colombia, Ecuador, and Venezuela; 69, approximate distribution of *Clinopodium tomentosum* (Kunth) Govaerts, based on the collection by Epling & Játiva (1964), Harley (1999), and data from the herbariums of MA and MO (Fernández-Alonso & Morales 2013); 70, approximate distribution of *Clinopodium axillare* (Rusby) Harley, including subspecies *C. axillare* subsp. *axillare* and *C. axillare* subsp. *uniflorum* (Rusby ex Briq.) J.R.I. Wood, based on Epling & Játiva (1964), Wood (2011), Harley *et al.* (2014), and data from the herbariums of MA and MO.

The same can be said about the medicinal species of *Clinopodium* in the Andes, with several known traditional uses for different ailments and purposes (e.g., oral antiseptic, anti-inflammatory, gastrointestinal conditions, and insecticides). There are published studies dealing with the essential oils of some species of *Clinopodium* and their biological activity (notably, Solís Quispe *et al.* 2018). *Clinopodium tomentosum* is known by the vernacular names of “tiglán”, “santa maría”, and “pumín”, and is widely distributed in Ecuador. Several traditional uses have been documented, generally using infusions of its flowers, leaves, and stems (Vera Saltos *et al.* 2014, Vera Saltos 2015, Moncayo Miño 2020). *Clinopodium axillare* (Rusby) Harley, an endemic species from the highlands of Bolivia, has



several traditional medicinal uses, which have been verified by studies of the composition of their essential oils and properties (Vila *et al.* 1996, Senatore *et al.* 1998, López 2019).

We hope that this publication will stimulate further studies of the leaf-mining pests of the Andean aromatic Lamiaceae and will contribute to a more detailed account of the diversity of the Neotropical Nepticulidae.

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