



New gall midges (Diptera: Cecidomyiidae) from Papua New Guinea

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<http://zoobank.org/urn:lsid:zoobank.org:pub:43C64F1E-2972-4C30-B4FA-02D4B64AD9E0>

Abstract Two new species of gall midges that feed on trees in Papua New Guinea are described. The larvae of *Schizomyia novoguineensis* Kolesik sp. nov. transform the flowers of *Macaranga aleuritoides* (Euphorbiaceae) into spherical galls preventing sexual reproduction of the host tree. The larvae of *Rhopalomyia psychotriae* Kolesik sp. nov. induce pustulate leaf galls on *Psychotria ramuensis* (Rubiaceae). Descriptions of adults and immature stages, and DNA sequences of the cytochrome oxidase unit I mitochondrial gene segment, are given for each of the new species.

Key words barcoding, COI, insect taxonomy, primary tropical forest, secondary tropical forest.

INTRODUCTION

The gall midge (Diptera: Cecidomyiidae) fauna of Papua New Guinea (PNG) is almost entirely unknown. Only three named species have been recorded thus far – predators of mealybugs (Hemiptera: Pseudococcidae) *Diadiplosis duni* (Harris) and *D. smithi* Felt, and the mango leaf-feeding *Procontarinia pustulata* Kolesik (Gagné & Jaschhof 2014). Additionally, descriptions and illustrations of the galls of six unnamed gall midges collected in the western part of New Guinea were included by Docters van Leeuwen-Reijnvaan and Docters van Leeuwen in their 1926 book *The Zooecidia of the Netherlands East Indies*. These were leaf gall #20725 on *Antiaris toxicaria* (Moraceae), leaf gall #20682 on *Bulbophyllum macrobulbum* (Orchidaceae), leaf petiole gall #21186 on *Dimorphantera anchorifera* (Ericaceae), leaf gall #20662 on *Tapeinochilos ananassae* (Zingiberaceae), leaf gall #20968 on *Macaranga aleuritoides* (Euphorbiaceae) and the flower gall #20967 on *M. aleuritoides* caused by *Schizomyia novoguineensis* sp. nov., a species described and named here.

The new species were collected as part of a project assessing the host specificity and species richness of gall-forming insects in lowland tropical rainforests of Papua New Guinea (Butterill & Novotny in prep.). The study is a continuation of research focused on the ecology of various other herbivorous insect guilds (Novotny *et al.* 2010). Fieldwork was carried out at sites in the proximity of the New Guinea Binatang Research Center (BRC), Madang, a base for investigating the ecology and evo-

lution of insect–plant interactions in tropical rainforests of Papua New Guinea. The centre trains and employs native Papua New Guineans who work as para-ecologists helping to describe the great insect diversity of their country and assisting with ground-breaking ecological research projects.

Macaranga aleuritoides is a tree endemic to New Guinea and adjacent islands (Global Biodiversity Information Facility 2013). In PNG, it grows in primary and secondary forests to a height of up to 26 m, and its timber is used by local people for construction of houses and huts (Weiblen 2013).

Psychotria ramuensis (Rubiaceae) is a tree endemic to New Guinea (Global Biodiversity Information Facility 2013). In PNG, it grows in primary and secondary lowland rainforest to a height of up to 5 m (Weiblen & Molem 2013).

The two new insect species belong to large Cecidomyiidae genera with worldwide distributions (Gagné & Jaschhof 2014). *Rhopalomyia psychotriae* sp. nov. is the first species of *Rhopalomyia* known to feed on a plant from the family Rubiaceae. *Schizomyia novoguineensis* sp. nov. is the first species of this genus known to occur in the Australian and Oceanian regions.

MATERIAL AND METHODS

Insects

Galls were sampled in secondary and primary lowland rainforest near the villages of Baitabag, Mis and Ohu (5°08′ – 14′ S, 145°41′ – 47′ E, 0–200 m above sea level) in Madang Province between August 2010 and March 2011. Harvested galls were sorted according to gall morphology and host plant.

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Fig. 1. *Macaranga aleuritoides*. Left, healthy fruit with leaf in background. Image courtesy of GD Weiblen. Right, gall of *Schizomyia novoguineensis*. Fruit and gall are about 20 mm in diameter.

Some galls of the same morphotype were dissected to obtain larvae, and some were kept in clear plastic bags pegged to a washing line to rear adults, which were stored with larvae in 95% ethanol. Gall vouchers are currently stored at BRC. Insect type specimens were macerated in 20% KOH (larvae were perforated laterally with a thin needle to speed up maceration), washed in 20% acetic acid, followed by 70% and 99% ethanol, cleared in HistoClear and permanently mounted in Canada balsam on glass slides under round glass coverslips 10 mm in diameter. Whole larvae and pupal skins were mounted dorsoventrally. Adults were dissected into four pieces with the particular body parts mounted separately: wings, head frontally, thorax laterally, abdomen dorsoventrally or for some of female specimens laterally. Length measurements were made with a microscope imaging system. Drawings were made with the aid of a drawing tube. Types will be deposited in the National Agricultural Insect Collection at the National Agricultural Research Institute, Port Moresby, Papua New Guinea (NAIC), and the South Australian Museum, Adelaide (SAMA).

DNA extraction, amplification and sequencing

DNA was individually extracted from single legs of adults and whole larvae. Five adults and three larvae were used for *Schizomyia novoguineensis* sp. nov., and three adults and three larvae for *Rhopalomyia psychotriae* sp. nov. Extractions and sequencing were carried out at the Laboratories of Analytical Biology, Smithsonian National Museum of Natural History, Washington DC, USA. DNA was extracted with the Autogen robot using a phenol-chloroform extraction method, and the cytochrome oxidase subunit I mitochondrial gene (COI) gene fragment amplified using the LCO1490/HCO2198 primers (Folmer *et al.* 1994). Sequences were lodged in GenBank and compared with those of congeners using BLASTN programme optimised for dissimilar sequences ('Discontiguous Megablast').

TAXONOMY

Genus *Schizomyia* Kieffer

Schizomyia Kieffer 1889: 183

Citation list after 1889 in Gagné and Jaschhof (2014)

Type species: *Schizomyia galiorum* Kieffer 1889: 184

Schizomyia is a catch-all genus of the subtribe Schizomyiina for species with needle-like ovipositors and four-segmented palpi (Gagné & Jaschhof 2014). Papillae of the larval terminal segment vary between species ranging from four setose pairs to a single corniform pair. There are differences between species in the ovipositor, with some having a pair of large dorso-basal lobes, which are absent in others. On the other hand, several Schizomyiina genera fit the current scope of *Schizomyia* and could be included here – see Tokuda *et al.* (2005) and Gagné and Jaschhof (2014).

Schizomyia novoguineensis Kolesik sp. nov. (Figs 1–3)

<http://zoobank.org/urn:lsid:zoobank.org:act:F995DE5B-9D7F-4B03-99C5-F4BCA3409C0F>

Types. *Madang Province, Papua New Guinea.* Holotype male, Mis village, Madang Province (5°11'24'S, 145°45'4"E), emerged xi.2010, ex flower gall on *Macaranga aleuritoides* F.Muell. (Euphorbiaceae), collected xi.2010, P. Butterill (gall #059), NAIC. Paratypes: 2 males, 3 females, 2 larvae (NAIC); 2 males, 2 females, 2 larvae, 1 pupal skin (SAMA 29-003001 to 29-003007), collected and emerged with holotype.

DNA. COI sequence was analysed for 3 males, 2 females and 3 larvae revealing identical sequences (GenBank accession numbers KJ202119-20, KJ202122-25, KJ202129, KJ202129, 658 bp).

Description. *Male.* (Fig. 2a–i). Wing length 1.8 mm (1.7–1.8, $n = 5$), width 0.7 (0.6–0.7), length/width ratio 2.6. Colour of abdomen in live specimens not noted.

Head. Palpus four-segmented; segment 1 short, segments 2–4 long, progressively slightly longer; palpiger not present; labella hemispherical; frons with 8–12 setae; eye bridge 9–10 ocelli long, post-vertical peak minute. Antenna: scape as wide as long, pedicel slightly longer than wide; flagellomeres not complete in available specimens (note: *Schizomyia* species have 12 flagellomeres), each consisting of cylindrical node and short neck, first and second flagellomeres not fused, nodes cylindrical, 4× longer than wide at mid-length, with irregularly looped circumfila comprising two long transverse and two short longitudinal bands; necks about 1/10 node length.

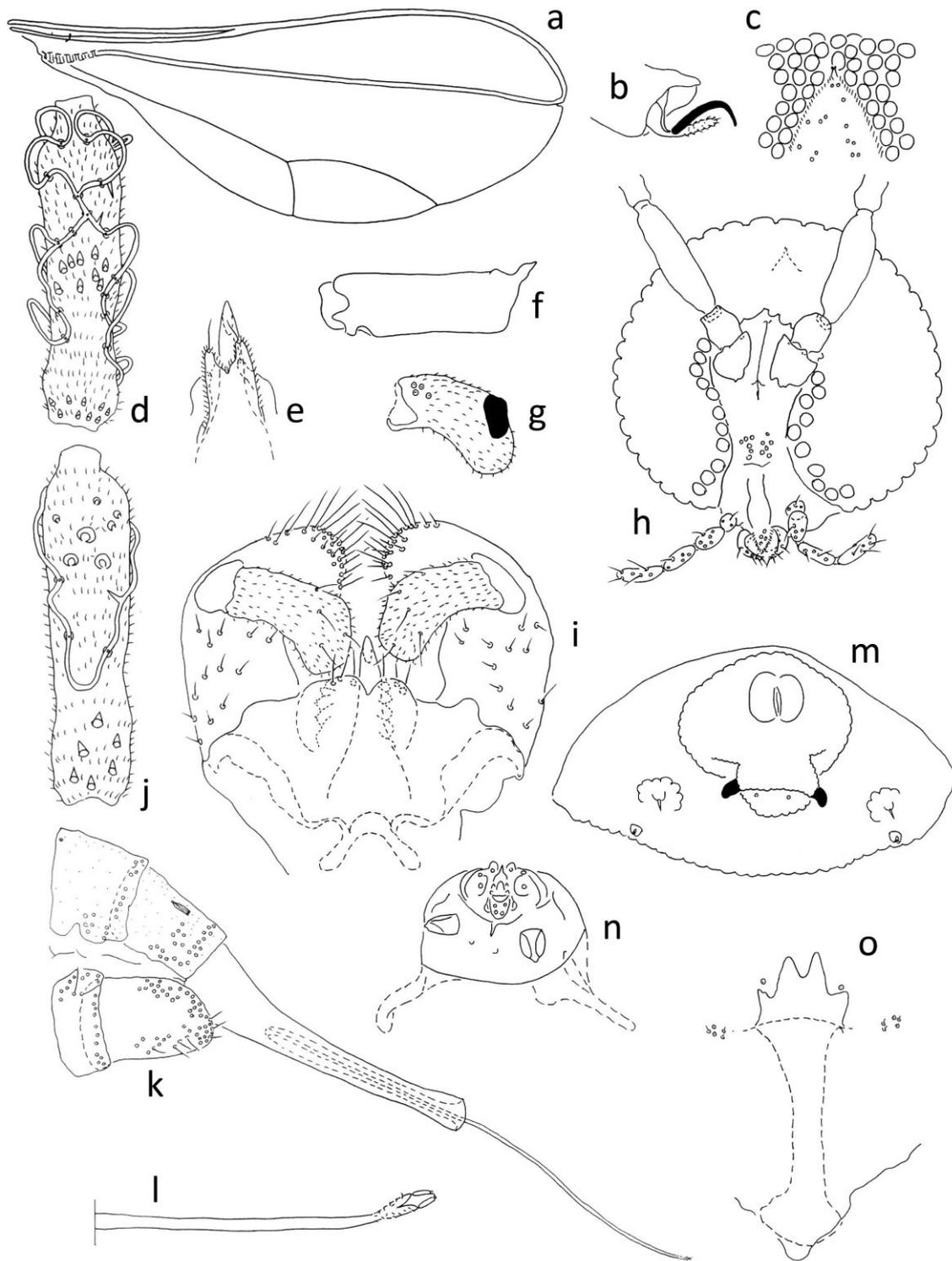


Fig. 2. *Schizomyia novoguineensis*. (a–i) male, (j–l) female, (m–o) larva. (a) Wing, (b) tarsal claw with empodium, (c) eye bridge with minute post-vertical peak, (d) sixth antennal flagellomere, (e) part of terminalia: aeadeagus, hypoproct, mesobasal lobes, (f) first tarsal segment, (g) gonostyle in ventral view, (h) head in frontal view, (i) terminalia in dorsal view with hypoproct and mesobasal lobes omitted, (j) sixth antennal flagellomere, (k) end of abdomen in lateral view, (l) end of ovipositor, (m) last two segments in caudal view (upper part of figure shows ventral, lower part dorsal sides of segments), (n) head in dorsal view, (o) sternal spatula with adjacent papillae.

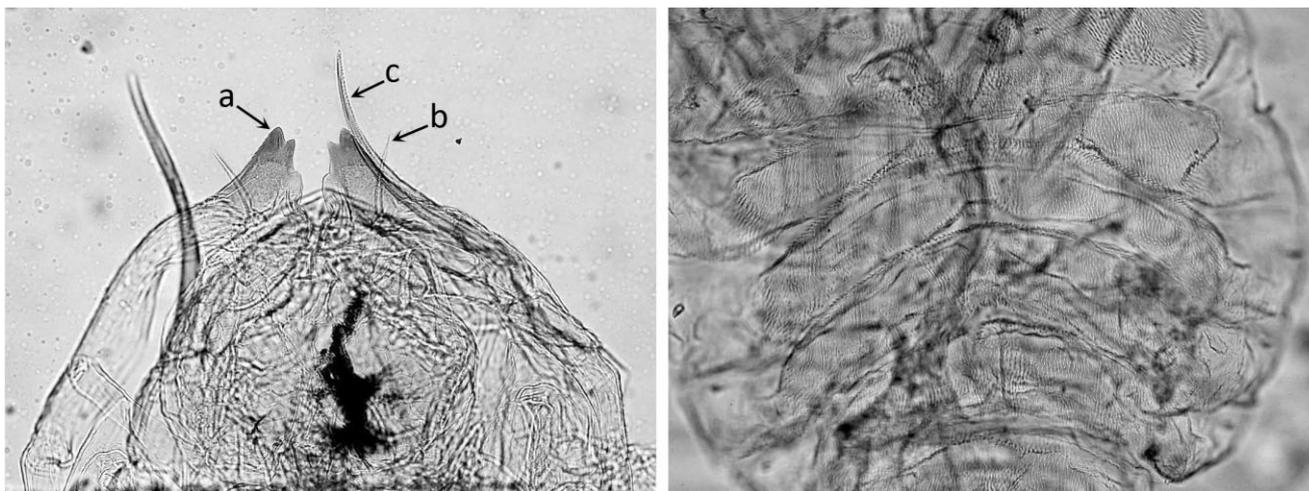


Fig. 3. Pupa of *Schizomyia novoguineensis*. Left, upper part of body with antennal horns (a), post-vertical setae (b) and prothoracic spiracles (c). Right, abdominal segments in dorsal view.

Wing. C with break at juncture with R_5 , R_5 reaching wing apex, R_5 not visible. Tarsal claws simple, bent at apical third, empodium shorter than tarsal claws, pulvilli minute. Legs with a robust, straight, ventro-distal spine on the first tarsomere.

Abdomen. Sclerites with pair of trichoid setae anteriorly, covered with scales and setae. Setation: sternites 2–8 with posterior row as long as sternite width and short anterior row; tergite 1 with single row of setae, tergites 2–7 with posterior row and small lateral group.

Terminalia. Gonocoxite with ventro-apical lobe bearing long setae at apex, gonocoxal apodemes separated, diverging anteriorly; gonostyle twice longer than wide in dorsal view, slightly bent at basal third, uniformly setulose on both sides, ventrally with apical tooth in shape of solid plate covering half width of gonostyle and with small group of aetose papillae basally, dorsally with few long setae apically; aedeagus tapering evenly, pointy apically, longer than cerci and hypoproct; hypoproct slightly longer than cerci, with incision reaching one-fourth height, single seta apically; cerci hemispherical in dorsal view, with lateral edges warped ventrally and bearing row of evenly spaced setae, few setae apically; mediobasal lobes shorter than cerci, narrow, bearing single seta apically.

Female. (Fig. 2j–l). Wing length 2.1 mm (2.0–2.1, $n = 5$), width 0.8 (0.7–0.8), length/width ratio 2.7 (2.6–2.8). Eye bridge 8–9 ocelli long. Flagellomeres not complete in available specimens, neck shorter than in male. Sternites and tergites with posterior row and lateral groups of setae. Terminalia: ovipositor needle-like, long, protractile, without basal lobes; cerci fused, small, setulose basally, aetose. Otherwise as in male.

Pupa. (Fig. 3). Around 1.5 mm long ($n = 4$). Antennal horns robust, bifid. Cephalic pair of setae strong, reaching apex of antennal horns. Prothoracic spiracle extremely long, considerably overreaching antennal horns. Frons smooth, without setae. Second to eighth abdominal segments dorsally each with field of numerous simple small spines.

Larva. (Fig. 2 m–o). Length 1.5 mm ($n = 1$). Yellow in colour with clearly visible sternal spatula. Integument covered with crescent-shaped plates and field of transverse rows of spiculae on anterior half of ventral side of each thoracic and first to seventh abdominal segments. Head: head capsule hemispherical; antennae slightly longer than wide at base; posterolateral apodemes as long as head capsule. Sternal spatula with robust shaft, four anterior lobes with two inner ones longer than outer; on either side with one sternal aetose papilla, two groups of lateral papillae with inner group consisting of two setulose papillae, outer group of one setose and two aetose. Terminal segment with terminal protuberance bearing one pair of robust, recurved, corniform and one pair of aetose papillae, anus ventral.

Etymology. The new species is named after the island of New Guinea where it occurs.

Remarks. *Schizomyia novoguineensis* is the first *Schizomyia* described from Australia and Oceania. Its only congener feeding on a host plant from the family Euphorbiaceae is *S. macarangae* Nayar (1953), which induces a hairy globular gall on the leaf of *Macaranga indica* Wight in India (see Mani, 2000, for gall description). The new species differs from *S. macarangae* by the absence of a dorsal lobe at the base of the ovipositor and the larval spatula having four anterior lobes as opposed to two. There is 86% identity in COI (using discontinuous megablast) between the eight identical sequences of the new species and the single published sequence of *Schizomyia galiorum* Kieffer, the only other congener with DNA data (Tokuda *et al.* 2005).

Biology and geographical distribution. The new species induces an irregularly globular gall on the flowers of *Macaranga aleuritoides* (Fig. 1). The gall is about 20 mm in diameter, containing a centrally placed larval chamber in a brittle casing occupied by a single larva. Several flowers within an inflorescence can be affected, making up a complex



Fig. 4. Galls of *Rhopalomyia psychotriae* on *Psychotria ramuensis*. Left: galls on lower side of leaf showing slit face (note circle of necrotised trichomes). Middle: galls on upper side of leaf showing hemispherical face (two galls in upper part of image) and slit face (gall in right bottom corner, note circle of fresh trichomes). Right: gall on leaf stalk. Gall diameter is about 5 mm.

gall that contains several larval chambers. No seeds are produced in galled flowers. Pupation takes place within the gall. Galls of the new species were collected at the villages of Mis and Ohu (5°11'24"S, 145°45'4"E and 5°13'52"S, 145°40'48"E, respectively) but were uncommon. Previously, galls of a similar appearance were collected from the same host plant species by K. Gjellerup at modern-day Jayapura (2°32'S, 140°43'E) in the Indonesian province of Papua on the island of New Guinea in August 1910 (Docters van Leeuwen-Reijnvaan & Docters van Leeuwen 1926, p. 309: gall #20967), and we conclude that they were induced by the new species described here. Most *Schizomyia* spp. pupate in the soil, but the new species pupates in the gall as does *S. macarangae* from *Macaranga indica*. The Indian species begins pupation once the galled leaf falls to the ground and is exposed to high humidity (Nayar 1953).

Genus *Rhopalomyia* Rübsaamen

Rhopalomyia Rübsaamen 1892: 370

Citation list after 1892 in Gagné and Jaschhof (2014)

Type species: *Oligotrophus tanaceticola* Karsch 1879: 27 (des. Kieffer 1896, 89)

Rhopalomyia is a large, worldwide genus of the tribe Oligotrophini with an undivided eighth female abdominal tergite and a completely setulose gonostyle. Maxillary palpus has one to three segments except for Australian *R. goodeniae* Kolesik (1996), which has three to four segments. The new species fits the genus in all characters except in the gonostyle, which is setulose entirely ventrally but only to 3/4 dorsally.

Rhopalomyia psychotriae Kolesik sp. nov. (Figs 4,5)

<http://zoobank.org/urn:lsid:zoobank.org:act:BD66BB2A-DD7E-466B-801B-6E5D98B989E9>

Types. *Madang Province, Papua New Guinea.* Holotype male, Baitabag village, Madang Province (5°8'35"S,

145°46'34"E), emerged x.2010, ex leaf gall on *Psychotria ramuensis* Sohmer (Rubiaceae), collected x.2010, P. Butterill (gall #089), NAIC. Paratypes: 2 males, 3 females, 2 larvae (NAIC); 2 males, 2 females, 1 larva (SAMA 29-003008 to 29-003012), collected and emerged with holotype.

DNA. COI sequence was analysed for 2 males, 1 female and 3 larvae revealing identical sequences (GenBank accession numbers KJ202118, KJ202121, KJ202126-8, KJ202130, 658 bp).

Description. Male. (Fig. 5a–e). Wing length 2.0 mm (1.8–2.1, $n = 5$), width 0.8 (0.8–0.9), length/width ratio 2.4 (2.3–2.5). Colour of abdomen bright orange.

Head. Palpus two-segmented, second segment slightly larger; palpiger absent; labella prolonged; short stout setae present on palpal segments and labella. Eye bridge 9–10 ocelli long. Antenna: scape and pedicel as wide as long; flagellomeres 15 in number, each consisting of node and neck, first and second flagellomeres fused dorsally, two apicalmost fused in some specimens; nodes cylindrical, 1.5× longer than wide, with closely appressed circumfila comprising one transverse and two longitudinal bands; necks about 1/3 node length.

Wing. C with break at juncture with R_5 , R_5 reaching C slightly anteriorly to wing apex, R_5 not visible. Tarsal claws bent at basal third, bearing two teeth of unequal size, empodium shorter than tarsal claws, pulvilli minute.

Abdomen. Sclerites rectangular, with pair of sensory setae anteriorly, covered with scales and setae. Sternites 2–8 with posterior and medial bands of setae. Tergites 1–7 with posterior row of setae. Terminalia: gonocoxite cylindrical; gonostyle tapered evenly towards apex, slightly curved at mid-length, setose, covered with setulae to about 3/4 length dorsally and entirely ventrally, bearing distal comb-like claw apically; aedeagus robust, trapezoid in dorsal view, longer than cerci and hypoproct; cerci wide, hemispherical in dorsal view,

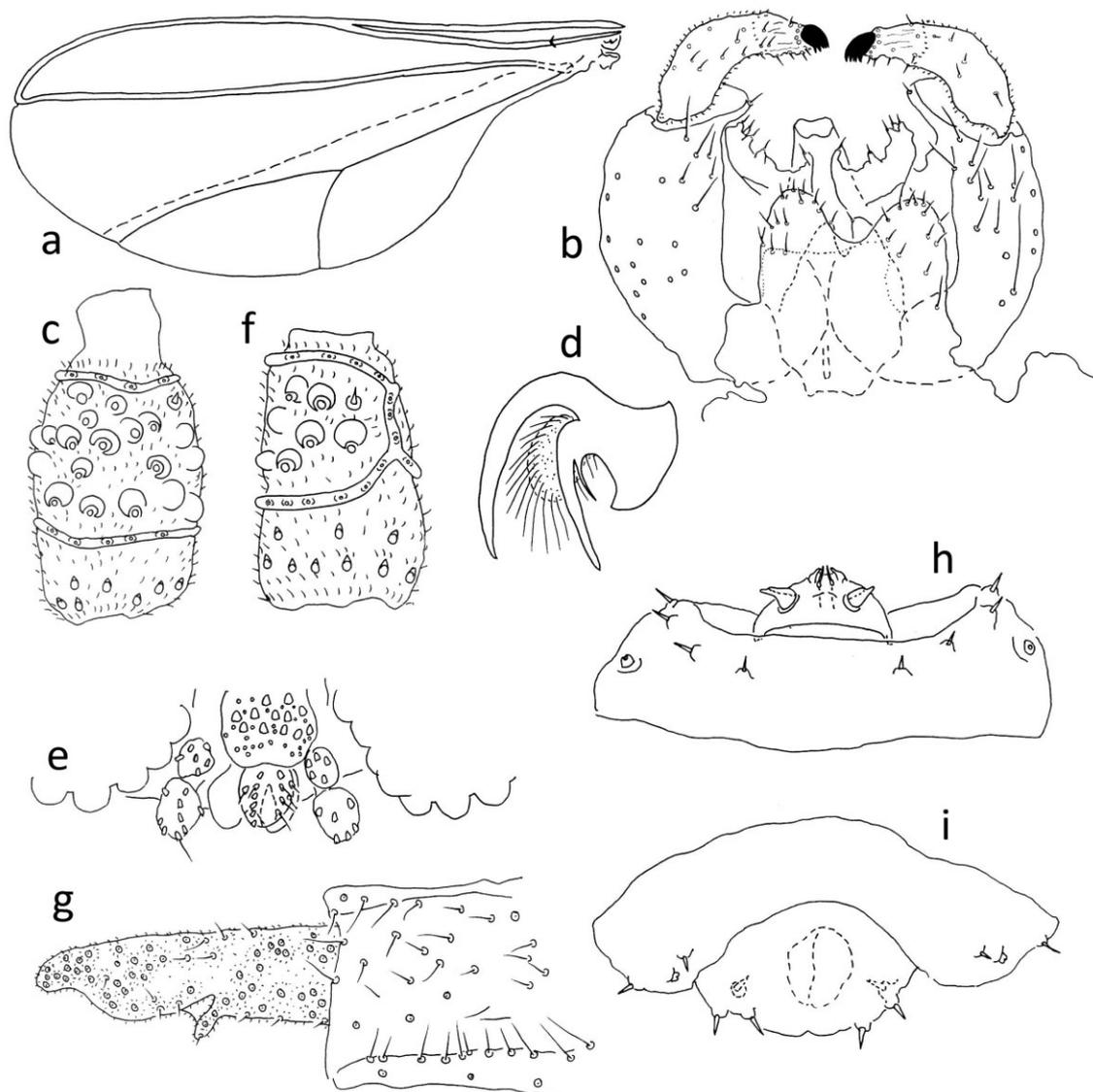


Fig. 5. *Rhopalomyia psychotriae*. (a–e) male, (f,g) female, (h,i) larva. (a) Wing, (b) terminalia in dorsal view, (c) sixth antennal flagellomere, (d) tarsal claw with empodium, (e) mouth with palpi, (f) sixth antennal flagellomere, (g) end of ovipositor in lateral view, (h) head and first thoracic segment in dorsal view, (i) last two segments in dorsal view.

longer than hypoproct, with several setae, covered with regular patches of microtrichia; hypoproct wide, incision U-shaped with depth about 1/3 hypoproct length, with one seta on each lobe, uniformly covered with microtrichia; mediobasal lobes wide, with five setose papillae on each side, loosely sheathing aedeagus.

Female. (Fig. 5f,g). Wing length 2.4 mm (2.4–2.5, $n = 5$), width 0.9 (0.9–1.0), length/width ratio 2.6 (2.5–2.7). Eye bridge 6–9 ocelli long. Flagellomeres not entire in available specimens; barrel-shaped, slightly wider at base, 1.6 \times longer than wide at base; first and second fused dorsally, necks on remaining flagellomeres very short. Terminalia: ovipositor protractile; fused cerci large, evenly covered with microtrichia and setae, no thick sensory hairs present; hypoproct 1/4–1/3 length of cerci, with pair of setae apically. Otherwise as in male.

Pupa. Unknown.

Larva. (Fig. 5h,i). Around 1.5 mm long ($n = 3$). Colour not noted. Integument smooth except for field of transverse rows of spiculae on anterior half of ventral side of each thoracic and first to seventh abdominal segments. Head: capsule hemispherical, antennae slightly longer than wide at base, cone-shaped; posterolateral apodemes absent. Spatula absent. Terminal segment with three pairs of setose papillae, anus ventral. Setae, except for lateral papillae, long and robust.

Etymology. The new species is named after the genus of its host plant.

Remarks. Of the nearly 300 described *Rhopalomyia* species, the majority have hosts from the family Asteraceae, with only

Table 1 Morphological differences between Australian and Oceanian *Rhopalomyia* spp.

	Palpal segments	Tarsal claws	Dorsal setation of gonostyle	Larval spatula	Larval terminal papillae	Posterolateral apodemes of larval head	Pupal dorsal spines	Pupal antennal horns
<i>R. goodeniae</i> Kolesik 1996	3–4	Simple	Entire	Bi-lobed	3–4 pairs	Long	Simple	Absent
<i>R. lawrenciae</i> Kolesik 1998	3	Toothed	Entire	Absent	4 pairs	Short	Absent	Bifid
<i>R. psychotriae</i> sp. nov.	2	Bi-toothed	3/4	Absent	3 pairs	Absent	Simple	Simple

eight species feeding on plants from other families, including *R. goodeniae* Kolesik on *Goodenia lunata* J. Black (Goodeniaceae) and *R. lawrenciae* Kolesik on *Lawrencia squamata* Nees (Malvaceae) in Australia (Gagné & Jaschhof 2014). The new species is the first *Rhopalomyia* known to feed on Rubiaceae. It differs from its two Australasian and Oceanian congeners (see descriptions in Kolesik 1996, 1998, DNA sequence not available) in several male, pupal and larval characters (Table 1). Of the *Rhopalomyia* spp. with published COI sequence, *R. pomum* (Felt) feeding on *Artemisia tridentata* Nutt. in North America (sequenced by Beckenbach & Joy 2009) was found to be the nearest, with 84% similarity followed closely by others (see GenBank for sequences).

Biology. The gall occurs on the blade, veins and stalk of the leaf of *Psychotria ramuensis* (Fig. 4). It has the shape of a truncated cone, circular to ovoid at the base, about 5 mm in diameter at the base and 5 mm in height, at the top with a slit surrounded by a circle of trichomes that necrotise at later stage of gall development. The slit has a various degree of openness and runs along the longer axis without reaching the outer limit of the gall. The galls can occur on either side of the leaf blade, with a simple hemispherical appearance on the opposite side. The gall can be easily split in half along the slit, revealing a single U-shaped gall chamber in a brittle casing containing a single larva. Pupation takes place within the gall. At the end of the larval development, a large necrotic area appears at the top of the slit face from which a pupa protrudes and the adult emerges. We collected galls of the new species at the villages of Baitabag and Mis, close to Madang (5°8'35"S, 145°46'34"E and 5°11'24"S, 145°45'4"E, respectively) where the gall was common.

ACKNOWLEDGEMENTS

This study was supported by the European Union Center of Excellence for the global study of the function and biodiversity of forest ecosystems (CZ.1.07/2.3.00/20.0064), the Grant Agency of the Czech Republic (13-10486S), the US National Science Foundation (Division of Environmental Biology 0515678), the Christensen Fund (US), and the UK Darwin Initiative for the Survival of Species (14/054). We thank the para-ecologists from the New Guinea Research Binatang Center and assistants from Ohu, Mis and Baitabag villages, especially Thomas Pius, Ismael Kiatik, Martin Keltim, Brus Isua, Isaac Kutil and Nataniel Mataton, for technical assis-

tance. We thank Amy Driskell and Scott Miller at the Laboratories of Analytical Biology, Smithsonian National Museum of Natural History, Washington DC, USA, for DNA barcoding, George D. Weiblen (University of Minnesota, St. Paul, Minnesota) for permission to use the image of the *Macaranga aleuritoides* fruit, and Raymond J. Gagné (Systematic Entomology Laboratory, USDA, Washington DC) for commenting on an early draft of the manuscript.

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Accepted for publication 13 March 2014.
Version of record published on 21 April 2014