

RESEARCH PAPER

Larval morphology of *Yateberosus*, a New Caledonian endemic subgenus of *Laccobius* (Coleoptera: Hydrophilidae), with notes on ‘*Berosus*-like’ larvae in Hydrophiloidea

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Abstract. The morphology and head chaetotaxy of the second and third instar larvae of *Laccobius* (*Yateberosus*) sp. are described based on specimens collected in New Caledonia. The larvae agree with those of other subgenera of *Laccobius* Erichson, 1837 in most morphological characters including the morphology of head and mouthparts and the head chaetotaxy, which undoubtedly supports its assignment to *Laccobius* (*Yateberosus*). It differs from other *Laccobius* in the closed spiracular system, reduced spiracular atrium and long abdominal tracheal gills, in which they resemble the larvae of *Berosus* Leach, 1817. We demonstrate that the ‘*Berosus*-like’ larval morphology evolved at least four times independently in Hydrophiloidea, and briefly discuss the possible reasons for it.

Key words. Coleoptera, Hydrophilidae, Hydrophilinae, Laccobiini, larval morphology, parallel evolution, New Caledonia, Pacific Region

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Introduction

In 1958, an expedition of the Osaka Museum of Natural History (Osaka, Japan) to New Caledonia gained two interesting *Berosus*-like hydrophilid adult specimens, which were later studied by SATŌ (1966) and described as a new genus, *Yateberosus* Satō, 1966, considered close to *Berosus* Leach, 1817 (genus of almost world-wide distribution represented by three species in New Caledonia; KOMAREK 2010) and *Hemiosus* Sharp, 1882 (occurring in the Neotropical and southernmost Nearctic regions; HANSEN 1999, SHORT & FIKÁČEK 2011). The presence of an unknown *Berosus*-like genus was independently confirmed by BERTRAND (1968) who briefly described a larva collected in 1965, being evidently unaware of SATŌ’s (1966) description of *Yateberosus*. Interestingly, BERTRAND (1968) mentioned that the larva was in some characters closer to larvae of the genus *Laccobius* Erichson, 1837. Nearly two decades later, GENTILI (1980)

studied a short series of *Yateberosus* from the Bishop Museum (Honolulu, USA), including the first males known for that taxon, and found that the adult morphology of these specimens agreed in nearly all important characters with representatives of *Laccobius*. He hence made *Yateberosus* a subgenus of *Laccobius*, a status it retains until today. The close relationships between *Yateberosus* and *Laccobius* were confirmed by molecular analyses by SHORT & FIKÁČEK (2013); an internal position of *Yateberosus* in the *Laccobius* clade was revealed by TOUSSAINT & SHORT (2018). Based on a large amount of material collected during recent expeditions to New Caledonia, GENTILI (2010) described two additional species raising the total number of *Yateberosus* species to five. *Laccobius* (*Yateberosus*) remains the only larger aquatic hydrophiloid clade endemic to New Caledonia.

The transfer of *Yateberosus* to the tribe Laccobiini posed a question about the identity of the larva of the



'Berosini genus' described by BERTRAND (1968). Recently, we got the opportunity to examine larvae corresponding to BERTRAND's (1968) description, collected by Christine Pöllabauer (ERBIO, Noumea, New Caledonia) in 2005. Results of our studies, confirming that this larva belongs to *Laccobius (Yateberosus)*, are summarized in this contribution.

Material and methods

Some of the examined larvae (3 specimens) were decapitated and both head and body were cleared using 10% KOH solution and subsequently examined in temporary slides with glycerine. After examination, the dissected specimens were transferred through 96% alcohol and isopropanol to permanent Euparal slides using H-S Slides (Kanto Rika Co., Ltd., Japan) (SHIRAYAMA et al. 1993). The remaining specimens were left without any treatment in the alcohol collection. Observations and dissections were carried out using an Olympus SZ61 stereoscopic microscope and an Olympus BX41 compound light microscope. Illustrations were made with the aid of a drawing tube attached to the aforementioned compound microscope. Photographs were taken with a Canon EOS

550D digital camera equipped with a Canon MP-E 65 mm macro lens.

The morphological terminology generally follows ARCHANGELSKY (1997) and MINOSHIMA & HAYASHI (2011) with the exceptions of the antennal segments, for which we follow BEUTEL (1999). For the chaetotaxy of the larval head we refer to FIKÁČEK et al. (2008) and BYTTEBIER & TORRES (2009). For the detailed account of the morphology of the larvae of *Berosus* and *Laccobius*, with which we compare the *Yateberosus* larva, see MINOSHIMA & HAYASHI (2015) and MINOSHIMA et al. (2017). The classification used follows SHORT & FIKÁČEK (2013).

The following abbreviations are used: AN – antenna; FR – frontale; gAN – group of antennal sensilla; gAPP – group of sensilla on inner appendage of maxilla; gFR – group of sensilla on frontale; gLA – group of sensilla on labium; gMX – group of sensilla on maxilla; LA – labium; MN – mandible; MX – maxilla; PA – parietale; SE – sensorium.

The specimens studied are deposited in the following collections:

- KMNH Kitakyushu Museum of Natural History and Human History, Kitakyushu, Japan;
- NHMW Naturhistorisches Museum Wien, Austria;
- NMPC National Museum, Prague, Czech Republic.



Fig. 1. Third instar larva of *Laccobius (Yateberosus)* sp. in dorsal (A), lateral (B), and ventral (C) view.

Results

Laccobius (Yateberosus) sp.

Material examined. 3 larvae of second instar, 5 larvae of third instar (NMPC, KMNH): "NEW CALEDONIA / Grande Terre (N-Prov.) / Koniambo / 05.01.05 (CONF-015) / leg. C. Pöllabauer / Confiance bassin / 479.822 mE / 7.673.494 mN / 45 m a.s.l.". All larvae were collected in the ultramafic gravel of riffles in an unshaded part of the middle reach of Confiance River, 21°2'26"S / 164°48'10"E (Koniambo Mountain, Koné Municipality, North Province, New Caledonia).

Additional material available. 9 larvae (NHMW): same label data as above. These larvae were not examined in detail in the present study.

General morphology. Third instar. Colour. Head and sclerotized body parts yellowish to reddish brown (reddish

brown colour probably stems from very high content of iron oxide in the habitat). Membranous parts yellowish to white (Figs 1A–C).

Head (Figs 1–2). Head superprognathous, slightly longer than wide, subquadrate, widest in anterior third, slightly narrowing posteriad. Frontal lines absent. Surface of head capsule smooth. Each side of head with a group of six stemmata, two anterodorsal ones larger than remaining ones. Clypeolabrum asymmetrical. Nasale asymmetrical, without distinct teeth, projecting anteriorly, truncate anteriorly. Epistomal lobes large, strongly asymmetrical, projecting much further than nasale, left lobe projecting slightly further than right one. Gular sulcus reduced, restricted to

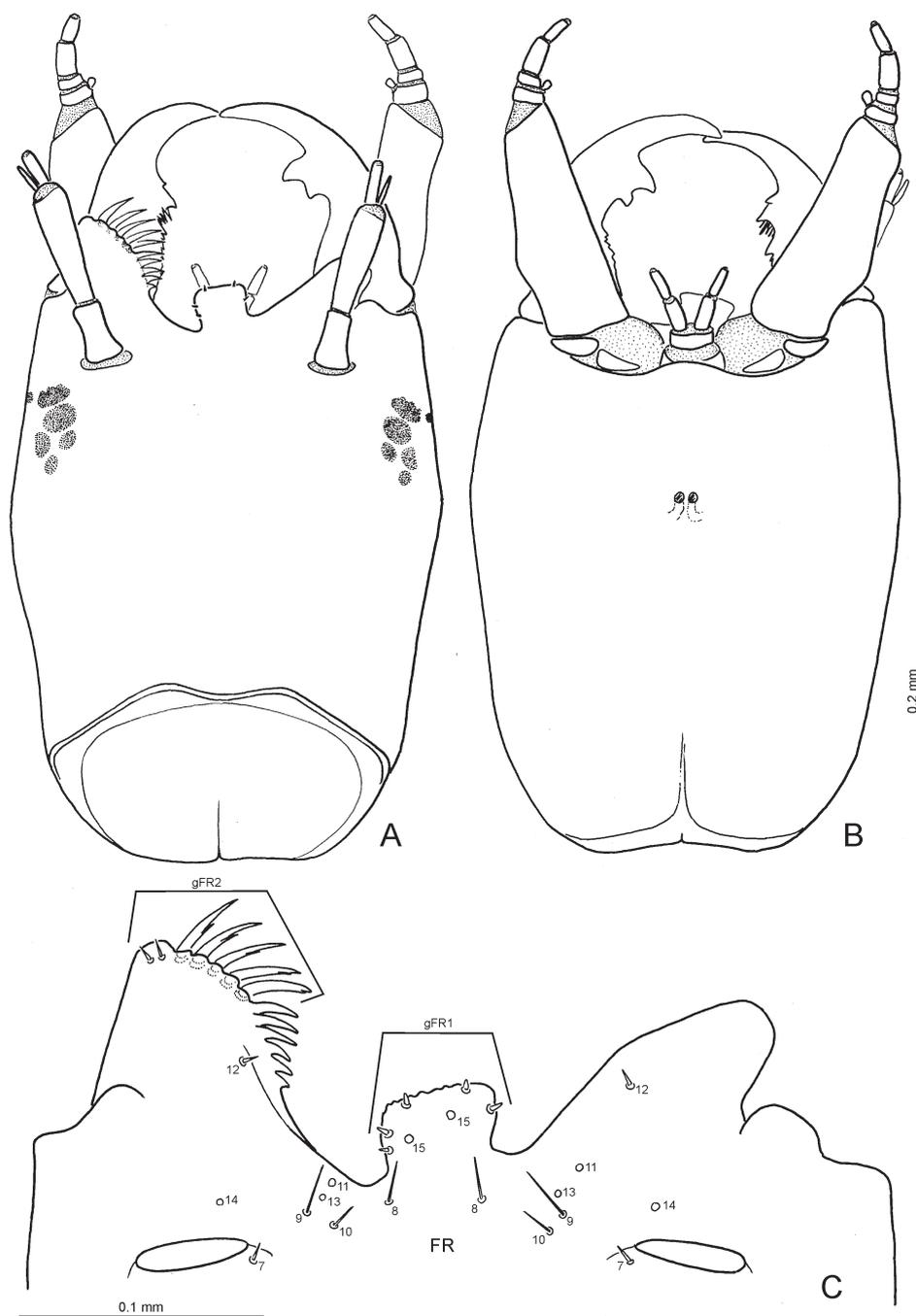


Fig. 2. Head morphology of the third instar larva of *Laccobius (Yateberosus) sp.* A – head in dorsal view; B – head in ventral view; C – detail of clypeolabrum in dorsal view. Chaetotaxy omitted in A–B.

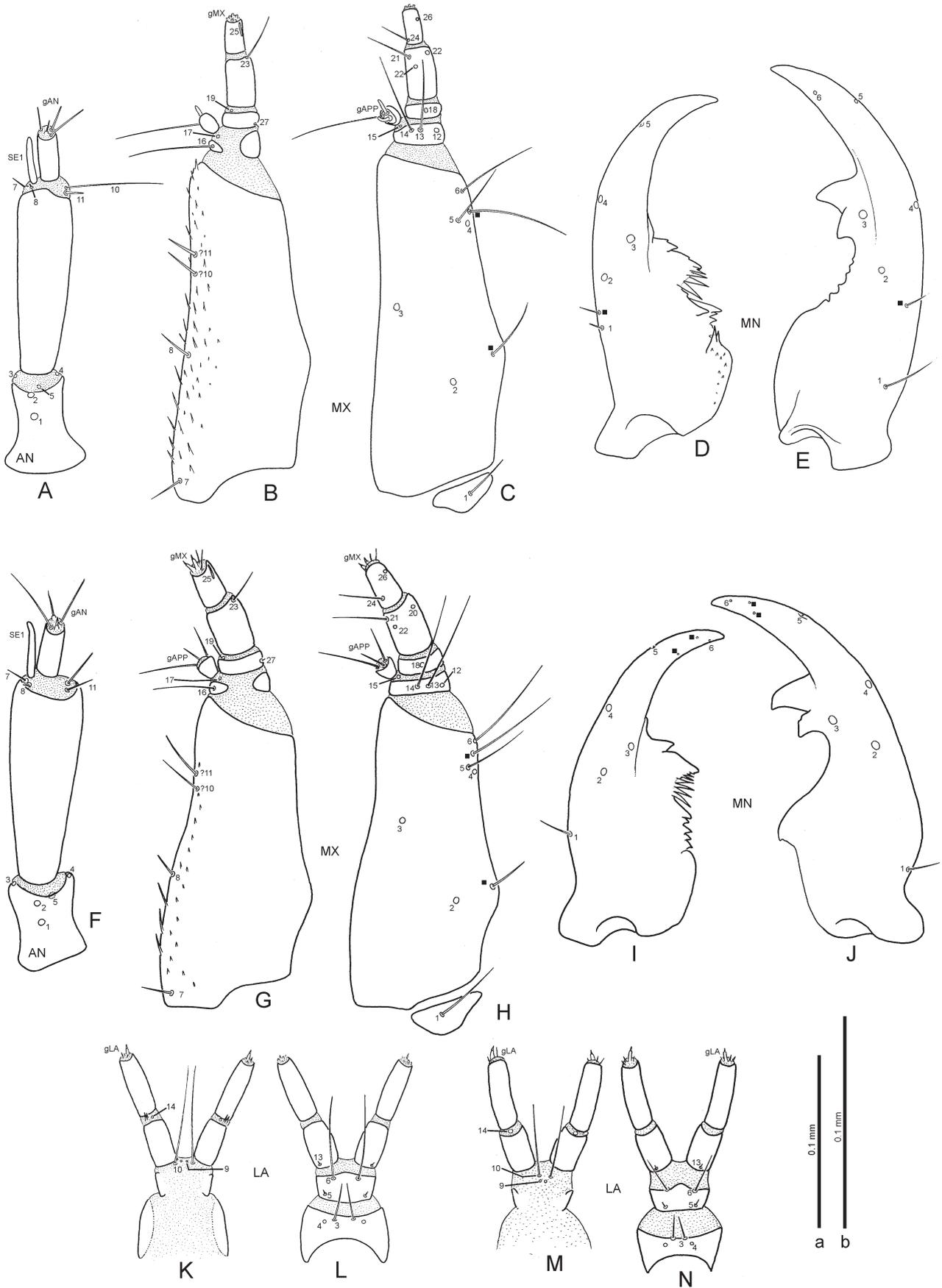


Fig. 3. Head appendages of the larvae of *Laccobius (Yateberosus)* sp. A–E, K–L – third instar; F–J, M–N – second instar. A, F – left antenna in dorsal view; B–C, G–H – right maxilla (B, G – dorsal view, C, H – ventral view); D–E, I–J – mandibles in dorsal view (D, I – left one, E, J – right one); K–N – labium (K, M – dorsal view, L, N – ventral view). Scale bars: a – third instar larva; b – second instar larva.

posterior quarter of head length. Posterior tentorial pits small, closely aggregated. Ventral anteromedian portion of parietale completely fused with submentum, submental sulcus absent. Pair of cervical sclerites present, but small and widely separated from each other.

Antenna (Fig. 3A) 3-segmented, slender, situated on dorsolateral surface of head capsule. Antennomere I ca. 0.5× as long as antennomere II. Antennomere II longer than antennomeres II and III combined, slightly widening distally. Antennomere III shortest, ca. 0.5× as long as antennomere I.

Mandibles (Figs 3D–E) asymmetrical. Right mandible with two inner teeth closely aggregated, distal one slightly larger than basal one; inner face basally of retinacular teeth denticulate. Left mandible with two inner teeth; distal tooth much smaller than basal one, with small denticle on anterior face; basal tooth large, bearing a comb of flat cuticular projections on posterior face; inner face basally with numerous small cuticular tooth-like projections, basal part with two strong spines projecting distad.

Maxilla (Figs 3B–C) 6-segmented (including cardo), distinctly longer than antenna. Cardo small, subtriangular. Stipes longest, ca. 2.5× as long as palpomeres 1–4 combined; inner face with numerous cuticular spine-like projections, inner distal part without large spine-like projection. Maxillary palpus 4-segmented; palpomere 1 short but widest, incompletely sclerotized dorsally; palpomere 2 shortest, slightly shorter than palpomere 1; palpomere 3 longest, ca. as long as remaining palpomeres combined; palpomere 4 narrowest, slightly shorter than palpomere 3. Inner appendage of maxilla rather large, well sclerotized except ventroapically.

Labium (Figs 2B, 3K–L) largely reduced. Submentum fused with ventral anteromesal portion of head capsule. Mentum wider than long, retracted below anterior margin of head capsule, sclerotized ventrally, largely membranous dorsally. Prementum slightly narrower than mentum, sclerotized ventrally, weakly sclerotized to membranous dorsally, without cuticular spines. Ligula absent. Labial

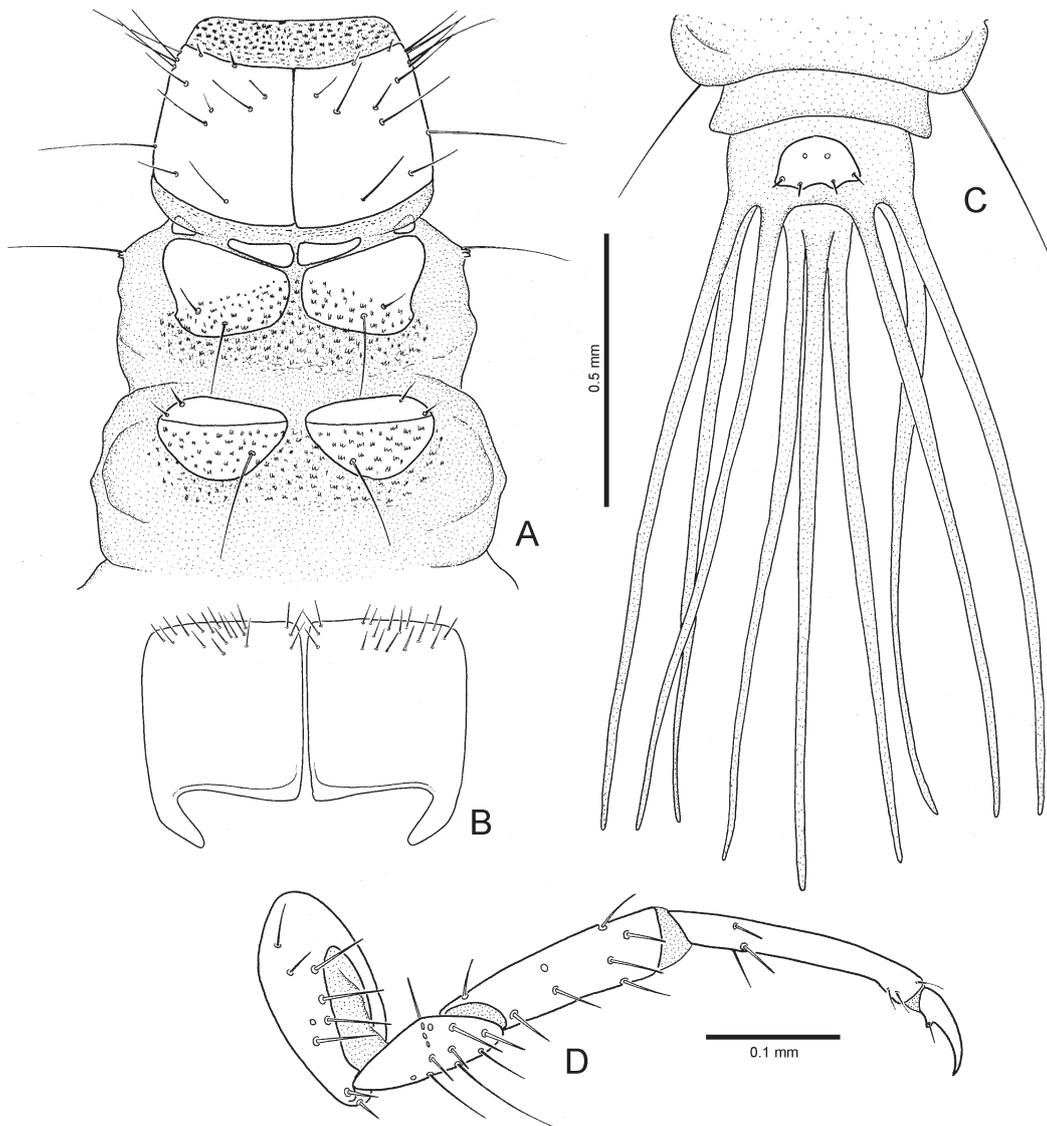


Fig. 4. Thorax and abdomen of the third instar larva of *Laccobius (Yateberosus)* sp. A – thorax in dorsal view; B – prosternum; C – abdominal apex; D – mesothoracic leg in anterior view.

palpi 2-segmented, well sclerotized, slightly longer than mentum and prementum combined. Few spine-like cuticular projections present on dorsal face of intersegmental membrane between palpomeres 1 and 2.

Thorax (Figs 1A–C, 4A–B). Prothorax slightly wider than head capsule. Proscutum formed by large plate subdivided by fine sagittal line; anterolateral corners with numerous long trichoid setae, dorsal surface with sparsely distributed, moderately long setae. Prosternum subquadrate, completely divided into two halves by rather wide median gap; anterior portion with numerous short setae. Mesonotum with large subtrapezoid sclerites of mesoscutum divided mesally by wide gap, each with two setae in posterior half; anterior of each scutal sclerite with one narrow subtriangular prescutal sclerite mesally and a small one sublaterally. Metanotum with one pair of subtriangular sclerites widely separated mesally, prescutal sclerites absent. Posterior portions of meso- and metanotum with area covered with asperities on surface of sclerites as well as membranous parts. Ventral parts of meso- and metathorax not sclerotized.

Legs (Figs 1A–C, 4D) 5-segmented, long and slender, distinctly visible in dorsal view; prothoracic ones closer to each other than meso- and metathoracic ones. Coxa transverse; trochanter elongate, ca. half as long as femur and as tibiotarsus; pretarsal claw with strong basal tooth. Chaetotaxy consisting of few pores and numerous moderately long setae, swimming setae absent. All three pairs of similar shape, prothoracic ones slightly shorter than meso- and metathoracic legs.

Abdomen (Figs 1A–C, 4C) 10-segmented, almost parallel-sided in anterior half, slightly narrowing posteriorly in posterior half. Surface with minute cuticular asperities, usually with attached fine dirt. Segment 1 not subdivided into anterior and posterior portion in dorsal view, subdivided into two folds in lateral view, without any sclerites. Segments 2–7 similar to each other, each subdivided into short anterior and longer posterior portion in dorsal view, posterior portion subdivided into two folds in lateral view; dorsal and ventral surface without any sclerites or areas with hooked cuticular projections. Chaetotaxy of abdominal segments not examined in detail. Segment 8 narrower than previous segments, subdivided into short anterior and longer posterior portion, posterior portion with small semicircular dorsal sclerite; posterior margin of sclerite with four blunt projections, each with a short seta; posterolateral portion of segment 8 with three long projections (tracheal gills) on each side. Segment 9 small, with three terminal long projections (tracheal gills). Segment 10 reduced, indistinct. Spiracles absent. Spiracular atrium not developed; styli, procerci and acrocerci absent.

Second instar. Similar to third instar, more weakly sclerotized than third instar.

Head (Figs 5A–B) slightly shorter, subquadrate, nearly parallel-sided laterally. Frontal sulci well developed, widely separated from each other at posterior margin of head capsule; closest to each other in posterior third, diverging both anteriorly and posteriorly; anteriorly reaching outer margin of antennal fossa and continuing to anterolateral

margin of head capsule; coronal sulcus absent. Gular sulcus weakly developed in posterior half. Posterior tentorial pits distinct, narrowly but distinctly separated from each other.

Antenna (Fig. 3F) stouter than in third instar, antennomere II more distinctly widened apically.

Mandibles (Figs 3I–J) shorter and stouter, inner face of right mandible without toothlets basally of basal retinacular teeth; armature of spine-like projections of basal retinacular teeth and inner basal face of left mandible less complex than in third instar, distally directed basal spines absent.

Maxilla (Figs 3G–H) relatively shorter and stouter, cuticular projection on inner face of stipes less numerous and generally shorter than in third instar.

Labium (Figs 3M–N) with mentum and prementum relatively narrower than in third instar, labial palps relatively longer than in third instar, without spine-like cuticular projections on dorsal face of intersegmental membrane between palpomeres 1 and 2.

Chaetotaxy of head. Second instar. Frontale altogether with 42 sensilla (Fig. 5B). Central part with three pairs of sensilla diverging posteriorly; FR1 rather long seta close to frontal line; FR2 pore-like, situated anteromesally of FR1, closer to FR3 than to FR1; minute seta FR3 situated anteromesally of FR2. Three setae (FR5–7) and one pore (FR4) situated posteriorly of antennal fossa; FR6 moderately long, situated close to frontal line, FR5 moderately long, situated anteromesally of FR6, FR7 minute seta at mesal margin of antennal fossa, FR4 mesally of FR7. Three setae and three pores situated anteriorly of each antennal fossa with three setae and three pores; moderately long setae FR9–10 close to each other, situated anteromesally of antennal fossa; pores FR11 and FR13 close to each other anteriorly of FR10, FR11 closer to anterior margin of head capsule on left side than on right side; pore FR14 situated slightly anteriorly of antennal fossa; short seta FR12 on inner basal portion of each epistomal lobe. Nasale (as in Fig. 2C) with five stout short spine-like setae on anterior margin (gFR1), median portion of nasal projection with asymmetrically situated pair of pores (FR15) and pair of moderately short setae (FR8) posterolaterally of FR15; ventral setae of nasale not found. Right epistomal lobe bare, lacking sensilla; left epistomal lobe with five stout long bifid setae with bifurcations between midlength and near apex (Fig. 2C) and two tiny trichoid setae in apical portion (gFR2), basal inner portion with long cuticular projections.

Parietale with 31 sensilla each (Figs 5A–B). Posterior portion of dorsal surface with oblique longitudinal group of five sensilla (PA1–5), setae PA1–2 and PA4–5 small, ca. equidistant from each other, PA3 pore-like, situated between PA2 and PA4. PA6 pore-like, situated on membranous part posterolaterally of frontale. Seta PA7 long, closer to frontal line, seta PA12 long, more lateral, situated ca. at midlength between stemmata and posterior margin of head capsule. Region around stemmata with four setae and one pore; long setae PA8–9 and one secondary, moderately long seta close to frontal line, pore PA10 within posterior group of stemmata; short seta PA11 in gap between anterior and posterior lateral stemmata. Lateral portion anteriorly with row of three long setae (PA20–22), pore PA19 not

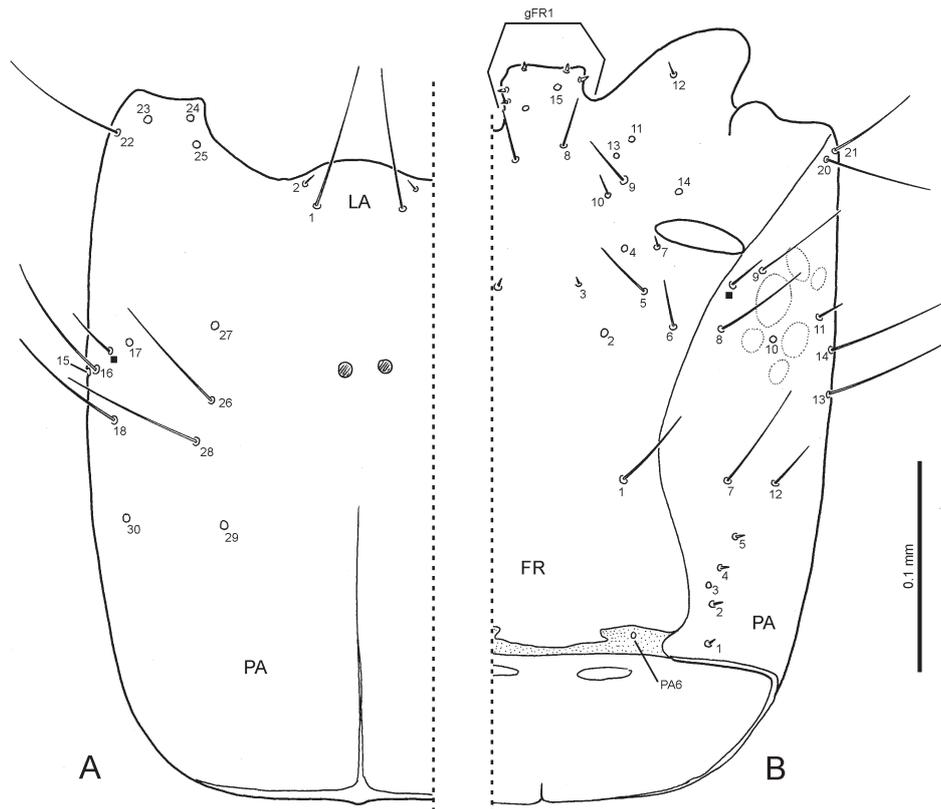


Fig. 5. Chaetotaxy of the head capsule of the second instar larva of *Laccobius* (*Yateberosus*) sp. A – ventral view; B – dorsal view.

found; midlength of head capsule laterally with four long setae (PA13–14 more dorsally, PA16 and PA18 more ventrally), one pore (PA15) between PA14 and PA16, and one moderately long secondary seta and pore PA17 ventrally of PA16. Ventral portion at mandibular articulation with three pores (PA23–25); central portion of ventral surface with four more or less equidistant sensilla (from anterior to posterior one): pore PA27, long setae PA26 and PA28, and pore PA29. Pore PA30 situated laterally of PA29.

Antenna (Fig. 3F). Antennomere I with five pore-like sensilla, two (AN1–2) situated in distal portion of dorsal surface, three (PA3–5) situated on distal margin of antennomere, PA3–4 on dorsal surface, PA5 on ventral surface. Antennomere II with five distally situated sensilla, AN6 and AN9 not found; long seta AN10 and short seta AN11 situated on inner face; sensorium (SE1), short seta AN7 and minute seta AN8 on outer face; sensorium long and slender, as long as antennomere III. Antennomere III with group of apical sensilla (gAN).

Mandibles (Figs 3I–J) with eight sensilla each. Outer face with one moderately long seta basally (MN1). Midlength with triangular group of three pores, MN4 lateral, MN2 sublateral and situated proximally of MN4, and MN3 situated near base of basal retinacular tooth. Apical portion with four tiny pore-like sensilla (MN5–6 and two secondary ones).

Maxilla (Figs 3G–H). Cardo with one moderately long seta (MX1). Stipes ventrally with two pores (MX2–3) situated in basal third and half, respectively; outer face with three long setae (MX5–6 and one secondary seta) and one pore (MX4) distally and one long secondary seta in basal

third. Inner face with moderately long seta basally (MX7) and three moderately long setae more distally, one ca. at midlength (MX8) and two closely aggregated ca. in distal third (likely representing MX10–11). Maxillary palpomere 1 ventrally with two long setae (MX13–14) and one pore (MX12), inner face with one long seta (MX16). Apical portion of inner appendage with group of sensilla (gAPP), membranous area basally of appendage with one pore dorsally (MX17) and one ventrally (MX15). Palpomere 2 with one pore ventrally (MX18) and one minute seta on outer face (MX27). Palpomere 3 with one distal long seta (MX21) and one subdistal pore (MX22) on inner face, and with one distal long seta (MX23) and pore (MX20) on outer face. Palpomere 4 with long seta (MX24) basally on inner face, and one digitiform sensillum (MX25) and one pore (MX26) subdistally on outer face; apical portion with group of minute sensilla (gMX).

Labium (Figs 3M–N, 5A). Submentum with two pairs of setae, LA1 long, LA2 small, situated anterolaterally of LA1. Mentum with pair of small setae (LA3) ventrally on distal margin of sclerite and pair of pores (LA4) posterolaterally of LA3. Prementum ventrally with pair of moderately long setae (LA6) on distal margin of sclerite and pair of short setae (LA5) close to basal margin; dorsal face with long setae LA10 and pair of pores slightly posteriorly of LA9. Labial palpus with one minute seta (LA13) at ventral base of palpomere 1 and one pore (LA14) on dorsal face of membranous area between palpomeres 1 and 2; apical portion of palpomere 2 with group of tiny sensilla (gLA).

Third instar larva. Head chaetotaxy identical to that of the second instar, except for the following characters.

Frontale close to posterior margin of head capsule, with pair of short setae (position of these setae corresponds to pores interpreted in the second instar as parietal sensilla (PA6) situated on membrane posterior to the *frontale*, and these setae are hence possibly homologous to that sensillum).

Parietale with two pores situated dorsally of seta PA20 (one probably corresponding to PA19 not found in the second instar, the other being obviously secondary).

Mandibles (Figs 3D–E) each with one secondary short seta on outer face, situated slightly distally of MN1.

Discussion

The generic identity of the examined larvae. Although the examined larvae resemble the larvae of *Berosus* (see e.g. ARCHANGELSKY 1994, 1997, 1999, 2002a, 2008; FERNANDEZ & CAMPOS 2002; WATTS 2002; MINOSHIMA & HAYASHI 2015; DELER-HERNÁNDEZ & FIKÁČEK 2016) at first glance based on the absence of the spiracular atrium and presence of long tracheal gills on the abdomen, they may be distinguished from *Berosus* by the characters listed below (see also Fig. 6). At the same time, the examined larvae share all these characters with the larvae of the *Laccobius* subgenera *Dimorpholaccobius* Zaitzev, 1938, *Hydroxenus* Wollaston, 1867, and *Laccobius* s. str. (PERKINS 1972, ARCHANGELSKY 1997, WATTS 2002, MINOSHIMA et al. 2017):

– Clypeolabrum with large epistomal lobes on both sides; the right one without sensilla (Fig. 2C). This character-

istic form of the clypeolabrum is shared by all known larvae of *Laccobius* and *Oocyclus* Sharp, 1882, and is likely a synapomorphy of the *Laccobius*-group of the tribe Laccobiini. In contrast, the right epistomal lobe is absent in most known larvae of *Berosus* (Fig. 6D) except in the Cuban endemic *B. chevrolati* Zaitzev, 1908 (DELER-HERNÁNDEZ & FIKÁČEK 2016).

- Submentum fused to ventral anterior margin of the *parietale* (Figs 2B, 5A). In *Berosus* (and most other hydrophilids), the submentum is distinctly divided from the *parietale* by submental sulci (Fig. 6E, SMS).
- The first antennomere lacks the inner distal projection (Figs 3A,F). The presence of this projection is a unique synapomorphy of the *Berosini* and is hence present also in all *Berosus* (ARCHANGELSKY 2008).
- The second antennomere is the longest, longer than the first and third combined (Figs 3A,F). The second antennomere is the longest one in all known larvae of *Laccobius* (as well as in *Oocyclus*, i.e. the other member of the *Laccobius* clade with known larvae: ARCHANGELSKY 1997). In contrast, in all known larvae of *Berosus* and *Hemiosus*, the first antennomere is the longest (see the references above and ARCHANGELSKY 2000, 2002b).
- Pore PA6 is situated in the membranous area posteriorly of *frontale* (Fig. 5B). The unique position of this pore is shared with *Laccobius* subgenera *Dimorpholaccobius* and *Laccobius* s. str. (Fig. 6C) (not examined for *Hydroxenus*), whereas the pore is situated normally in the *parietale* in *Berosus* (Fig. 6F).

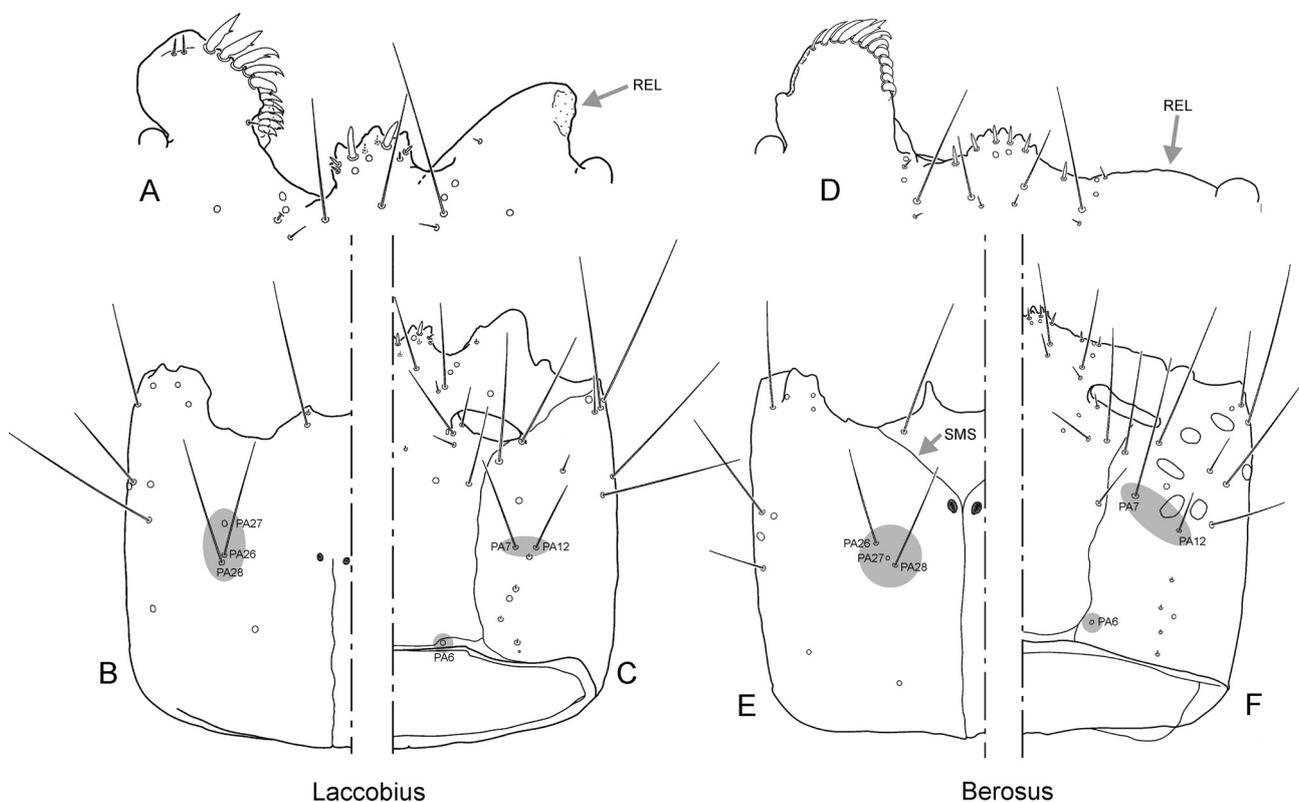


Fig. 6. Comparison of head morphology and chaetotaxy of *Laccobius* (s.str.) (A–C) and *Berosus* (D–F). A, C – clypeolabrum of the third larval instar; B–C, E–F – head chaetotaxy of the first larval instar (right half – dorsal view; left half – ventral view). Abbreviations: REL – right epistomal lobe; SMS – submental sulcus. Adapted from MINOSHIMA & HAYASHI (2015) and MINOSHIMA et al. (2017).

- Seta PA7 is situated as far from posterior margin of head capsule as PA12 (Fig. 5B). The same character state is found in the *Laccobius* subgenera *Dimorpholaccobius* and *Laccobius* s. str. (Fig. 6C) (not examined for *Hydroxenus*), whereas PA7 is situated distinctly more anteriorly than PA12 in *Berosus* (Fig. 6F).
- Pore PA27 is found far anterior of setae PA26 and PA28 (Fig. 5A). In most other hydrophilids including *Berosus*, pore PA27 is situated between setae PA26 and PA28 (Fig. 6E). The state observed in the examined larvae is only present in the *Laccobius* subgenera *Dimorpholaccobius* and *Laccobius* s. str. (Fig. 6B) (not examined for *Hydroxenus*), and the Coelostomatini genus *Coelostoma* Brullé, 1835 (FIKÁČEK et al. 2008).
- Mesonotum with large transverse scutal sclerites and two pairs of prescutal sclerites anteriorly of it; metanotum with a pair of sclerites (shared with *Laccobius* s. str., see MINOSHIMA et al. (2017)). *Berosus* is characterized by large triangular sclerites on mesonotum, and absence of sclerites on metanotum (e.g. ARCHANGELSKY 1997, Fig. 23).

All characters undoubtedly exclude the assignment of the studied larvae to the Berosini, and moreover clearly indicate that the larvae belong to the *Laccobius*-group of the tribe Laccobiini (SHORT & FIKÁČEK 2013). As the species of *Laccobius* (*Yateberosus*) are the only representatives of this clade in New Caledonia (KOMAREK 2010), and since two adults of the subgenus were collected together with these larvae, we may reliably assign them to *Laccobius* (*Yateberosus*).

Identification of instars of the examined larvae. In the material examined by us, there are two size categories of larvae. The larger ones bear secondary sensilla on mandibles, maxilla and parietale, and also lack the distinct frontal lines. This indicates that they represent the third instar. The smaller larvae, with well-developed frontal sulci, may represent either the first or the second instar. As they still bear some secondary sensilla on the parietale and maxilla (Figs 5B, 6C) that are not found in the first instar larva of the *Laccobius* subgenera *Dimorpholaccobius* and *Laccobius* s. str. (MINOSHIMA et al. 2017), we suppose that they belong to the second larval instar.

The specific identity of the examined larvae. Two adult specimens of *Laccobius* (*Yateberosus*) were collected together with the 17 larvae listed above (both are deposited in NHMW). These were tentatively identified as *Laccobius* cf. *wewalkai* Gentili, 2010.

In 2016, the third author visited Taléa River (20°59'29.0"S 164°45'25.0"E, ca. 55 m a.s.l.), which also flows from Koniambo Mountain and closely resembles Confiance River in its physical and chemical characteristics. Two species of *Laccobius* (*Yateberosus*) were collected there: *Laccobius* cf. *wewalkai* (6 spec.) and *L.* cf. *maculatus* Satô, 1966 (1 spec.) (all deposited in NHMW). It can be expected that these two species also occur in Confiance River. An exact identification can be provided only after a thorough taxonomic revision of *Laccobius* (*Yateberosus*) based also on molecular data. Hence, a species-level identification of the larval material is impossible at the moment.

Parallel evolution of the *Berosus*-like morphology on the Hydrophiloidea. Apneustic larvae (i.e. larvae with a closed spiracular system) bearing long abdominal tracheal gills are currently known in two clades within the Hydrophilidae: the genus *Berosus* and the subgenus *Yateberosus* of *Laccobius*. Interestingly, both these clades are also unusual within the Hydrophilidae due to their larval head morphology – the left epistomal lobe is always very large and bears a series of large spines and/or setae, the left mandible bears a complex set of flat spine-like projections, and the labium is extremely reduced and nearly completely concealed by the clypeolabrum in dorsal view. The same head morphology combined with a reduced spiracular atrium (but without developed abdominal tracheal gills) is also present in the Western Australian endemic *Hybograllius hartmeyeri* (Régimbart, 1908) of the tribe Hydrobiusini (WATTS 2002, ARCHANGELSKY 2008). Even more interestingly, the same combination of the unusual head characters is found in the genus *Epimetopus* Lacordaire, 1854 of the hydrophiloid family Epimetopidae, of which some species are also known to bear long projections at the end of the abdomen (FIKÁČEK et al. 2011). As all four mentioned clades are not closely related to each other (one belongs to the Epimetopidae, the remaining ones represent three different tribes of the hydrophilid subfamily Hydrophilinae), the unusual morphology of the head and abdomen had clearly developed multiple times within Hydrophiloidea. Moreover, abdominal tracheal gills were recently reported for the Tibetan *Helophorus* (*Lihelophorus*) *yangae* Angus, Fikáček & Jia, 2016 (Hydrophiloidea: Helophoridae) (ANGUS et al. 2016). Although the abdominal spiracles are open, the mandibles lack the dorsal groove and the clypeolabrum is not largely reduced in this species, it still resembles the remaining aforementioned lineages in the large epistomal lobes and the mandible which possesses numerous asperities; it may thus represent the fifth independent hydrophiloid lineage exhibiting the same morphological trend.

ARCHANGELSKY (2008) discussed the ‘*Berosus*-like’ morphology of the Berosini genera *Berosus* and *Hemiosus* in detail, assuming that the modification of the morphology of the head and abdomen was related to the shift to a benthic life style. Both are necessary to allow the larva to stay permanently submerged at the bottom, as larvae without any modification have to reach the water surface both for breathing (to draw the supply of the atmospheric air into their spiracles) and for digesting the prey (by holding the prey above the water surface in order to minimize the dilution of digestive fluids during the pre-oral digestion). Tracheal gills of larvae with closed spiracular system help them absorb oxygen dissolved in water and they hence do not need to reach the water level to draw the atmospheric air in. Explaining the modifications of the head seems more difficult. ARCHANGELSKY (2008) supposes it is primarily an adaptation for digesting the prey while submerged. The reduction of the labium is possible since it is likely not used to manipulate the prey above water, and the complex left mandible bears a dorsal groove which forms a canal to ingest the partially digested food to the pre-oral cavity

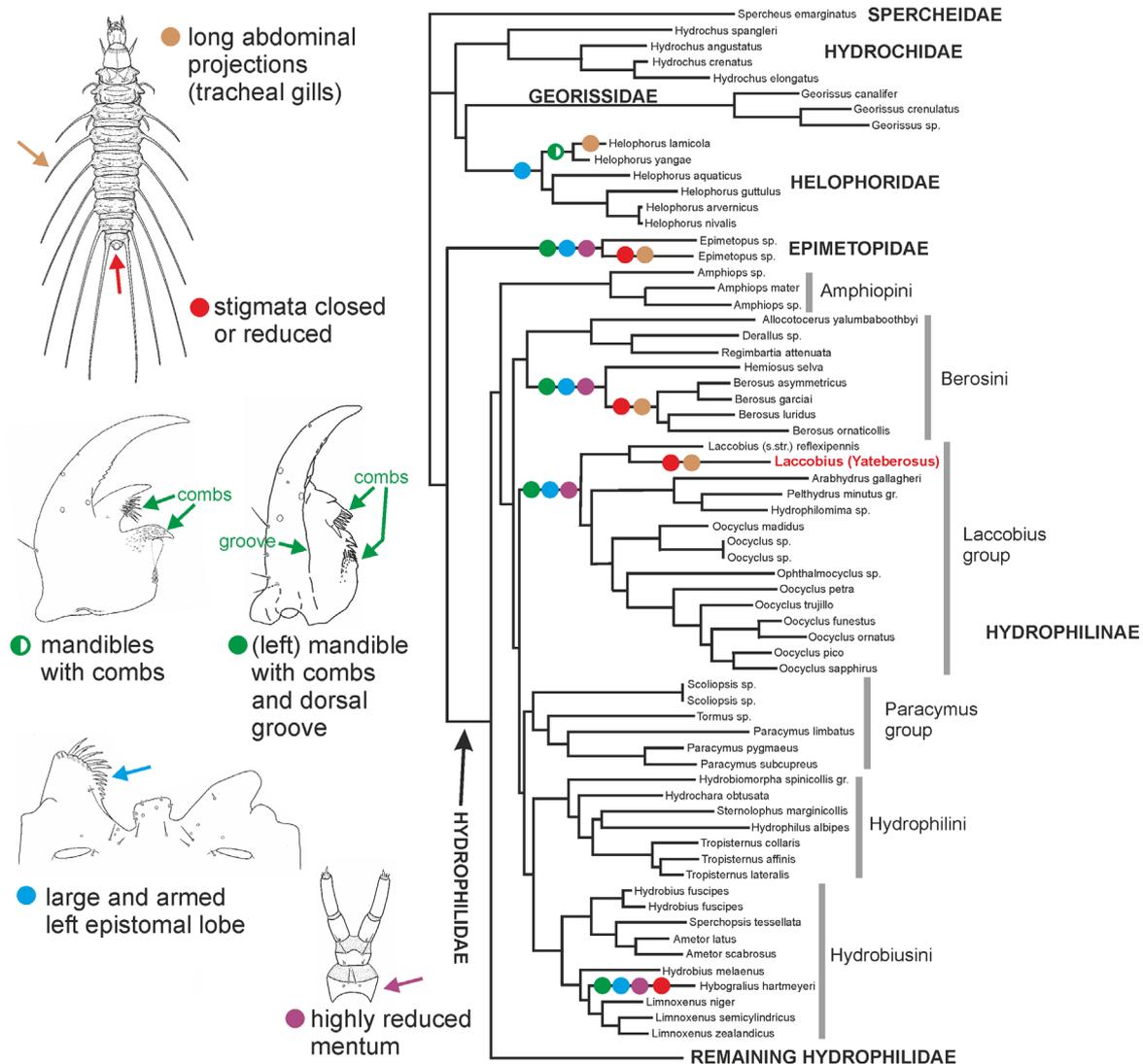


Fig. 7. Mapping of the characters of 'Berosus-like' morphology of the head and abdomen on the tree by SHORT & FIKÁČEK (2013), with *Helophorus (Lihelophorus)* species included based on ANGUS et al. (2016), and gross topology within the *Laccobius* group adapted according to TOUSSAINT & SHORT (2018). Characters are mapped using colour circles.

when pressed against the large left epistomal lobe (CHEARY 1971, ARCHANGELSKY 2008). The function of the combs of cuticular projections and stout setae on inner faces of left mandible and left epistomal lobe remains unexplained. When all these characters are mapped on the phylogenetic tree by SHORT & FIKÁČEK (2013) (Fig. 7), it is evident that the transformation of the usual hydrophiloid larva to the *Berosus*-like one always takes place in two steps: the head morphology changes first, allowing the change of the breathing strategy in some (usually not all) taxa within the clade with modified head morphology.

The parallel evolution usually implies that the same functional morphology is reached by different ways in each clade. The detailed studies on morphology of *Berosus*, *Laccobius (Yateberosus)* and *Epimetopus* along with available information about *Hybograllius* Régimbart, 1908 (WATTS 2002, ARCHANGELSKY 2008) reveal that this is also the case for the *Berosus*-like morphology within the Hydrophiloidea. The spine armature of the left epistomal lobe is partly formed by strong cuticular projections and only partly by setae in

Yateberosus and likely also in *Hybograllius*, whereas it is completely formed by setae in *Berosus* and *Epimetopus*. In *Berosus*, *Laccobius*-group of the Laccobiini, and *Hybograllius*, this structure seems to have developed *de novo* as the related taxa only bear low epistomal lobes with few simple setae. This is also supported by the different appearance of the right epistomal lobe (not developed in most *Berosus*, large but devoid of setae in the *Laccobius*-group, large with many trichoid setae in *Hybograllius*). In contrast, families of the helophorid lineage of the Hydrophiloidea all bear large epistomal lobes with strong setae, and the left epistomal lobe of *Epimetopus* and *Helophorus (Lihelophorus) yangae* is identical to the right one and in fact not modified from the state found in the Georissidae and the remaining Helophoridae (e.g. ARCHANGELSKY 1997, FIKÁČEK et al. 2012, ANGUS et al. 2016). The spinose armature of the left mandible is developed as a comb of simple projections in *Berosus*, *Laccobius* and *Hybograllius*, whereas the projections are complex and brush-like in *Epimetopus* (FIKÁČEK et al. 2011) and as a simple set of densely arranged asperities

in *Helophorus yangae*. The way of reduction of the spiracular atrium looks similar in *Berosus* and *Yateberosus*: the complex structures of abdominal segments 8–9 (urogomphi, cerci and spiracular openings) are completely reduced. In *Berosini*, an intermediate state between the usual hydrophilid morphology of the spiracular atrium (see e.g. CLARKSON et al. 2014, Figs 13–14) and apneustic *Berosus* exists – the larva of *Hemiosus* has the spiracles still preserved (but cribriform, not annular as in all other hydrophilid larvae, and possibly non-functional) but urogomphi and cerci are already reduced (ARCHANGELSKY 2000, 2008). In the helophorid lineage, the spiracular atrium is not developed. The larvae of *Epimetopus mendeli* Fikáček, Barclay & Perkins, 2011 bear largely reduced abdominal spiracles which may be non-functional, and the urogomphi are reduced in size but still present in this larva (FIKÁČEK et al. 2011). In *Helophorus yangae*, the spiracular atrium is absent, abdominal spiracles are open, and urogomphi are large and multisegmented (ANGUS et al. 2016). The origin of the tracheal gills is also different in each clade. In *Berosus* they lack any sensilla and are developed as lateral portions of abdominal segments 1–7 (one pair per segment, sometimes reduced on some segments), in *Yateberosus* multiple projections lacking sensilla are present on abdominal segments 8–9. In *Epimetopus mendeli* a single pair of projections bearing sensilla is present on abdominal segments 8–9, and in *Helophorus yangae* abdominal segments 1–8 each bear a pair of gills, and each gill bears a lateral abdominal sclerite with three short sensilla on its top. Larvae of *Hemiosus* and *Hybograllius* lack tracheal gills even though their spiracular atrium is reduced.

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