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Morphology of the Immature Stages of *Hydrochara libera* (Sharp) (Coleoptera, Hydrophilidae)

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Abstract Morphology of the immature stages of *Hydrochara libera* (Sharp, 1884) is described based on Japanese specimens. We described egg-case, all larval instars, and pupa of the species, and compared them with those of *H. affinis* (Sharp, 1873), another Japanese species. The mast of the egg-case of *H. libera* is longer and narrower than that of *H. affinis*. Larva of *H. libera* is distinguishable from *H. affinis* by the following combination of characters: 1) inner face of antennomere 1 with a few tooth-like cuticular projections in the first instar; 2) nasale slightly asymmetrical, weakly (first instar) to moderately (third instar) rounded; 3) lateral projections on the abdominal segments proportionally shorter than in *H. affinis* (all instars). Pupae of both species may be distinguished by the number of styli on the anterior face of the pronotum: four pairs of long styli and five short styli are present in *H affinis*, whereas two to three pairs of long styli and six to seven short styli in *H. libera*.
Key words: Hydrophilinae, Hydrophilini, aquatic beetles, egg-case, larva, pupa, morphology, chaetotaxy, Japan.

Introduction

The genus *Hydrochara* Berthold, 1827 of giant water scavenger beetles tribe Hydrophilini (Hydrophilinae) is represented by 23 large-sized species from Eurasian, African and North American continents (Smetana, 1980; Hansen, 1999; Short & Fikáček, 2011). The genus is well known taxonomically: Smetana (1980) revised world species, three species were newly described from Palaearctic and Nearctic Regions subsequently (Hilsenhoff & Tracy, 1982; Bameul, 1996; İncekara *et al*., 2009); one of them was later transferred to the genus *Brownephilus* Mouchamps, 1959 by Darilmaz, *et al*. (2010). On the other hand, the phylogenetic position of the genus is unclear; *Hydrochara* appeared as a paraphyletic group in the recent phylogenetic study by Short (2010).

Two species of the genus *Hydrochara* have been recorded from Japan: *H. affinis* (Sharp, 1873) and *H. libera* (Sharp, 1884). Morphology of the immature stages of the former species have been described including their larval chaetotaxy (Hosoi, 1947; Morioka, 1955; Hayashi, 2009; Minoshima & Hayashi, 2011), but those of *H. libera* have been hitherto unknown. The latter species is distributed in the Far East of the Palaearctic (Hansen, 1999). In Japan, it has been recorded from Hokkaido to Kyushu (Yoshitomi *et al*., 2001). We have recently succeeded in rearing the species from adults and an egg-case collected in the field. In this study, we describe the morphology of the immature stages of *H. libera*.

Materials and Methods

General methods follow MINOSHIMA and HAYASHI (2011). The examined specimens are deposited in the authors' collections.

Adults were kept in a plastic aquarium (16 cm length, 27 cm width, 14 cm height) filled with 8 cm of dechlorinated water and included aquatic plants from the collecting site or those available commercially. Egg cases were transferred to plastic cases (12 cm in diameter, 6 cm height) filled with 4 cm of dechlorinated water and with a plastic net as support for hatched larvae. A few days after hatching, larvae were transferred to ice trays with 10–20 mm of dechlorinated water individually; each tray was with a lid, otherwise larvae would escape from the tray. We used frozen Chironomidae larvae (Sanmi Co., Ltd. and Aso Tropical Fish Co., Ltd., Japan) to feed the first and second instar larvae, two sizes of fly maggots (presumably *Phormia* sp.) to feed second and third instars, both types of prey were commercially available. Larvae were fed manually with tweezers twice daily. The *Hydrochara* larvae were sometimes not able to feed on the maggots because they were not able to break their cuticle. Therefore, we broke the cuticle of maggots with tweezers before offering them. For pupation, larvae were transferred to terraria (12 cm in diameter, 6 cm height) filled with 4–5 cm of commercial soil (peat moss).

Specimens were fixed with 80% ethanol or boiling water; the colouration of the larvae and pupae may depend on the way of fixation. Larvae were preserved in screw-cap vials with 80% ethanol or were mounted on HS-slides (Higgins-Shirayama slide: SHIRAYAMA *et al.*, 1993) (Kanto Rika Co., Ltd., Japan) with Euparal (Chroma-Gesellschaft, Schmid & Co., Germany). Egg-cases and pupae were preserved in screw-cap vials with 80% ethanol. Observation and dissection were carried out using an Olympus SZX12 stereomicroscope and a Zeiss Axiophot compound light microscope; illustrations were made with the aid of a drawing tube attached to a Nikon SMZ-800 stereomicroscope and an Olympus BX40 compound light microscope. Line drawings were prepared using the software Paint Tool SAI (Systemax Inc., Japan) and Photoshop CS5 (Adobe Systems Inc., USA). Photographs of immature stages were taken with a Nikon DS-Fi1 digital microscope camera attached to a Nikon SMZ-800 microscope or directly by a Ricoh CX4 digital camera; composite images were created using the focus stacking software CombineZP (HADLEY, 2010). The photographs were retouched with Photoshop CS5 in needed cases.

Morphological terminology generally follows ARCHANGELSKY (1997) and MINOSHIMA and HAYASHI (2011) with the exception of the antennal segments, for which we followed BEUTEL (1999) and FIKÁČEK *et al.* (2008), *i.e.*, we chose the neutral term 'antennomere'; for the chaetotaxy of larval head we referred to FIKÁČEK *et al.* (2008) and BYTTEBIER and TORRES (2009).

Abbreviations. AN: antenna; E: egg-case; 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; L1, L2, L3: first, second and third larval instars; LA: labium; MN: mandible; MX: maxilla; P: pupa; PA: parietale; SE: sensorium.

Results

Genus ***Hydrochara*** BERTHOLD, 1827

Hydrochara libera (SHARP, 1884)

Material examined. Japan: 1 L2, 1 L3, Hongô, Memambetsu-chô, Hokkaido, 27–VI–2000, Y.

KAMITE leg.; 1 E, 10 L1, 8 L2, 7 L3, 4 P (larvae and pupae were reared from the egg-case collected in the field; rearing number: R12), Echigo-numa pond, Ebetsubuto, Ebetsu-shi, Hokkaido, alt. ca. 10 m, N 43°6′, E 141°36′, 7–VI–2011 (date of the egg case collected in the field), 12–VI–2011 (date of hatch), Y. MINOSHIMA leg. & rearing; 1 L1, Bibi-gawa R., Chitose-shi, Hokkaido, 29–VII–2011, M. HAYASHI leg.; 2 E, 18 L1, 11 L2, 6 L3, 3 P (larvae and pupae were reared from the egg cases laid by identified female; rearing number: R15 and R16), Hanzôgane, Nagaoka-shi, Niigata Pref., 13 to 14–VI–2011 (date of the adults collected in the field), Y. IWATA leg., rearing by Y. MINOSHIMA.

Diagnosis. The egg-case and larva of *Hydrochara libera* are distinguishable from another Japanese species *H. affinis* by the following combination of characters. Egg-case: mast longer and narrower than that of *H. affinis* (Fig. 9A–B) (*e.g.*, MORIMOTO & HAYASHI, 1986; YOSHIMURA, 1992). Larva (see also the key): inner face of antennomere 1 with a few tooth-like cuticular projections (first instar) (Fig. 2A–B); clypeolabrum slightly asymmetrical (Figs. 1C–D, 3A, 5C); nasale slightly asymmetrical, weakly (first instar) to moderately (third instar) rounded (Figs. 1C–D, 3A, 5C); lateral projections on abdominal segments shorter than those of *H. affinis* (Figs. 3B–C, 5A) (*e.g.*, MINOSHIMA & HAYASHI, 2011). Additionally, the larvae of *H. libera* may be distinguishable from all known larvae of *Hydrochara* by the short abdominal lateral projections (RICHMOND, 1920; HOSOI, 1947; MATTA, 1982; ARCHANGELSKY, 1997; MINOSHIMA & HAYASHI, 2011) (see Remarks). The morphology of the pupa of *H. affinis* was described by HOSOI (1947) which presented different number of styli on the anterior face of pronotum (four pairs of long styli and five short styli) from *H. libera* (two to three pairs of long styli and six to seven short styli; Fig. 7). In this study, however, the pupae of *H. libera* showed intraspecific variation of the number of the pronotal styli. HOSOI (1947) did not mention about the variation of styli, and therefore, an additional study of *H. affinis* is therefore necessary to confirm the above differences as reliably diagnostic.

Description of egg-case (Fig. 9A–B). Constructed on the water surface, free-floating, composed by semi-globular basal part and horn-shaped mast. White in colour, except for mast that white to brownish. Basal part covered with dead and/or fresh plants, with truncated cap closing the case. Mast above the cap closing the case, long and narrow, narrowed towards apically.

Description of larvae. *General morphology. Third instar larva*. Body rather thick, widest between abdominal segments 2–4 (Fig. 5A).

Colour. Head and sclerotised parts yellowish brown. Colouration of membranous parts may depend on the way of fixation: greyish to pale yellowish milky white in the specimens fixed with boiling water, dark greyish white in the specimens fixed with ethanol, greyish white in living individuals (Fig. 9C).

Head (Fig. 5B–C). Head capsule subtrapezial, widest anteriorly. Frontal lines nearly V-shaped, fused at base of head capsule, coronal line very short or absent. Surface of head capsule smooth. Six stemmata on each anterolateral portion of head capsule. Clypeolabrum slightly asymmetrical (Fig. 5C). Nasale slightly asymmetrical, rounded, moderately projected anteriorly. Epistomal lobes slightly asymmetrical, rounded.

Antenna (Fig. 6A–B) 3-segmented, long, slender. Antennomere 1 much longer than pedicel and flagellum combined; inner face of antennomere 1 smooth. Antennomere 2 with outer membranous area completely surrounded by sclerite, situated at apical part of sclerite. Antennomere 3 about as long as pedicel.

Mandibles (Fig. 6C–D) slightly asymmetrical, with two inner teeth, distal one large, basal one small; inner edge of distal inner tooth slightly serrated.

Maxilla (Fig. 6E–F) 6-segmented (including cardo), slightly longer than antenna. Cardo small, irregularly shaped. Stipes the longest, longer than palpomeres 1–4 combined; inner face bearing small

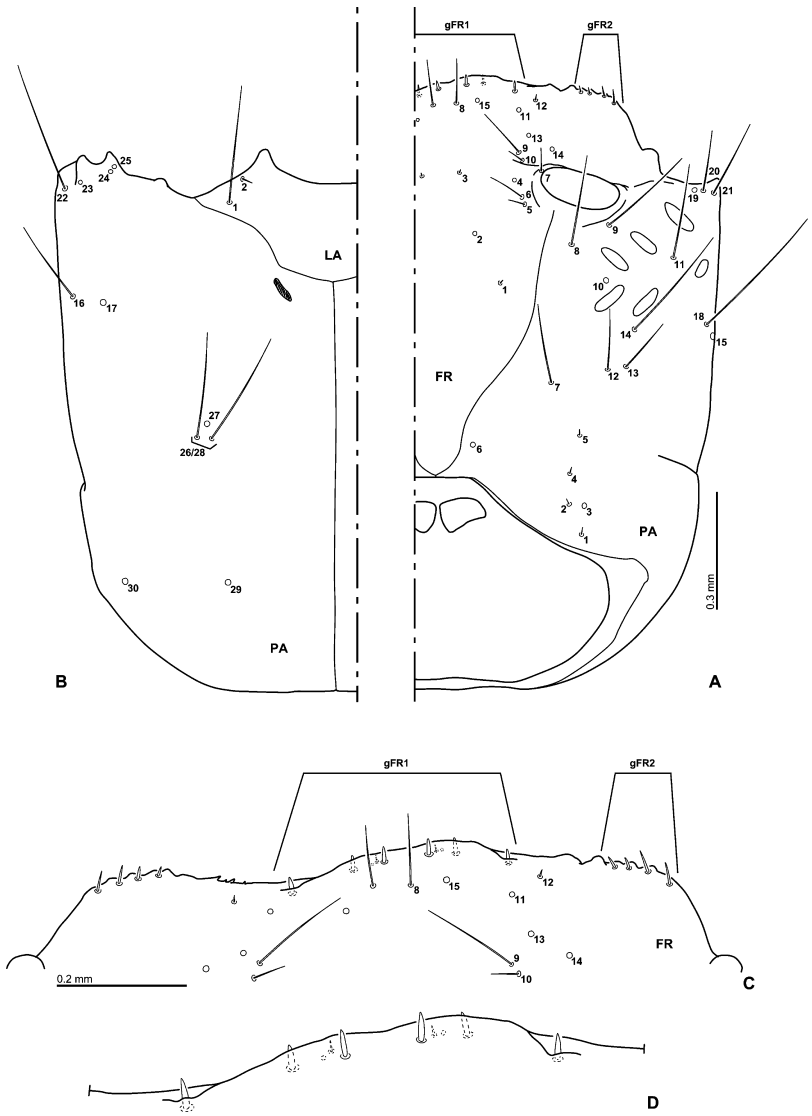

Fig. 1. Head capsule of *Hydrochara libera* (SHARP, 1884), first instar larva. —— A–B, Head capsule, dorsal (A) and ventral (B) view; C, detail of anterior margin of head capsule, dorsal view; D, detail of nasale, dorsal view.

spine-like cuticular projections between two basal-most setae; a small cuticular projection subapically on inner face of dorsal surface of sclerite. Maxillary palpus 4-segmented, palpomere 1 slightly longer than palpomere 3, palpomeres 1 and 3 each the longest, palpomere 2 the shortest, palpomere 4 longer than palpomere 2; palpomere 1 the widest, completely cylindrically sclerotised; palpomere 4 the narrowest; inner process sclerotised.

Labium (Fig. 6G–H) well developed. Submentum fused to head capsule, large, subpentagonal, wider than mentum (*e.g.*, Fig. 1B). Mentum subtrapezoidal in dorsal view, distinctly wider than prementum, about as long as wide, anterior corners strongly projected anteriorly; dorsal surface bearing densely arranged, strong but small cuticular projections on basal two-fifths, median part of the portion

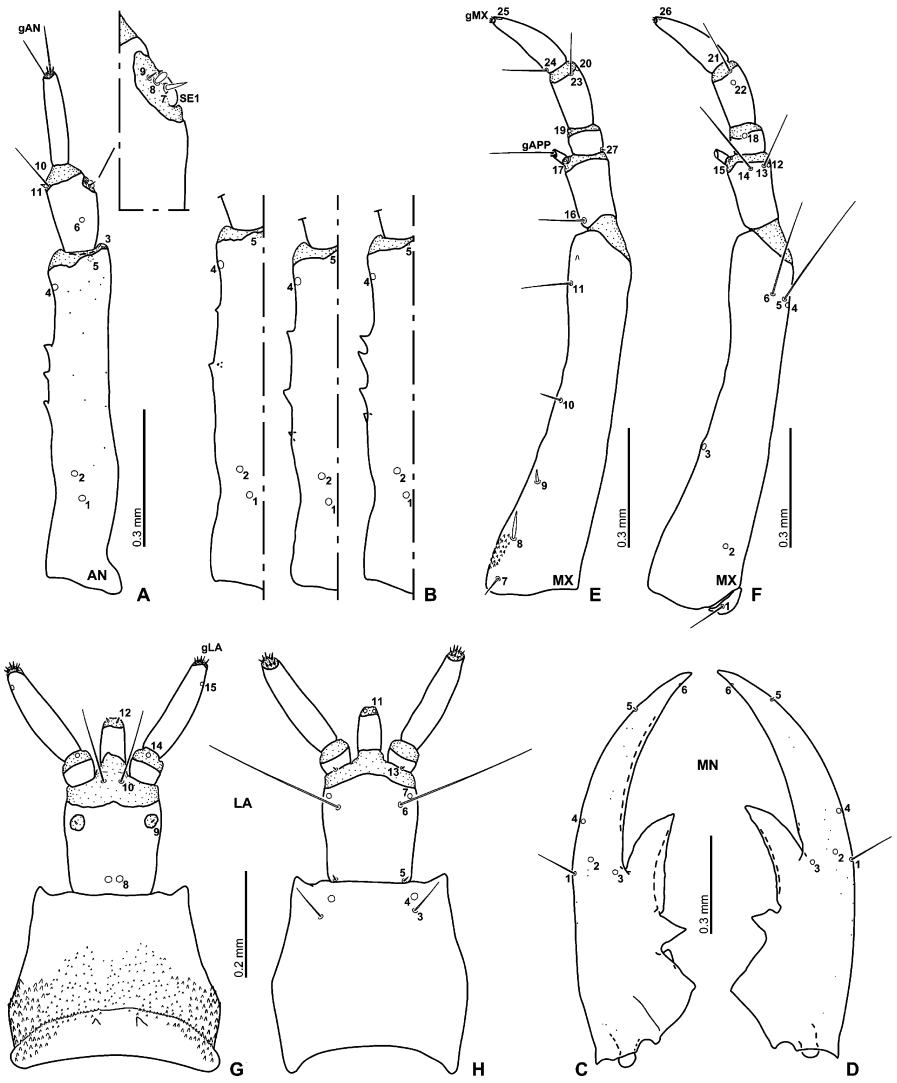

Fig. 2. Head appendages of *Hydrochara libera* (SHARP, 1884), first instar larva. —— A, Antenna, dorsal view; B, intraspecific variation of inner face of the first antennomere; C–D, mandible, dorsal view; E–F, maxilla, dorsal (E) and ventral (F) view G–H, labium, dorsal (G) and ventral (H) view.

with sparsely arranged ones; lateral part of dorsal surface with several cuticular teeth, basal part with two large cuticular teeth. Prementum subrectangular, longer than wide; dorsal surface of sclerite with one pair of small membranous areas completely surrounded by sclerite, situated on lateroanterior portion. Ligula stout, distinctly shorter than labial palpi, longer than labial palpomere 1, largely sclerotised, apical membranous area slightly bifid. Labial palpi moderately long, palpomere 1 small, palpomere 2 longer than ligula; outer face of palpomere 2 weakly convex.

Thorax (Fig. 5A). Prothorax wider than head capsule. Proscutum formed by one large plate subdivided by fine sagittal line, anterior part rather weakly sclerotised; whole sclerite bearing densely arranged setae of variable length. Prosternum subpentagonal, with long but incomplete sagittal line.

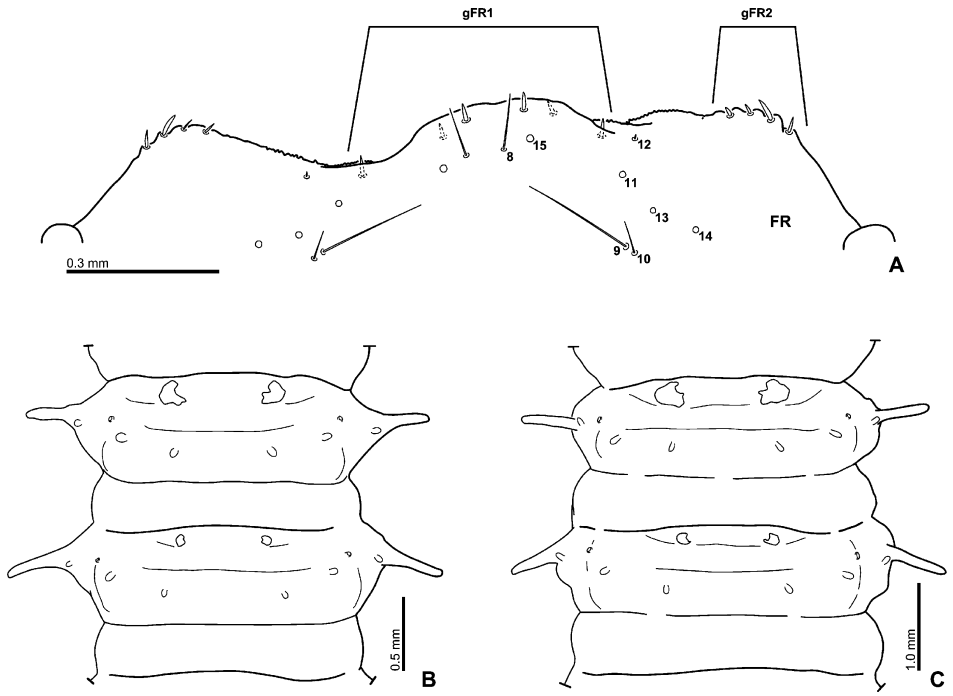

Fig. 3. *Hydrochara libera* (SHARP, 1884). —— A, Detail of anterior margin of head capsule of second instar larva, dorsal view: B–C, the first and second abdominal segments, first (B) and second (C) instar, dorsal view (setae and pubescence are omitted).

Mesonotum with three sclerites on each side; anterior two pairs small, narrow, one pair present medially, remaining pair laterally; posterior pair large, subtriangular, subdivided by transverse ridge, anterior part larger than posterior part, inner margin of anterior portion comes into contact with each other medially. Metanotum with one large, irregularly subtriangular sclerite each; anterior part lager than posterior part, subdivided by transverse ridge, comes into contact with each other medially. Legs long, visible in dorsal view, 5-segmented, bearing numerous long swimming hairs; all three pairs similar in shape.

Abdomen (Fig. 5A, D). Abdomen 10-segmented, tapering posteriad, covered with fine, cuticular pubescence and sparsely arranged short setae on membranous parts (*e.g.*, Fig. 8B); segments 1 to 7 similar in shape and size; lateral sides of each segment with one rather short but distinct setiferous projection (Fig. 5A). Segment 1 with one pair of small dorsal sclerites and three pairs of setiferous tubercles dorsally (*e.g.*, Fig. 3B–C); dorsal sclerites situated anteromedially; two setiferous tubercles on posterior portion behind sclerites, each bearing very long setae, remaining one setiferous tubercle on lateral part of the segment, close to lateral projection; these tubercles look like partially sclerotised. Segments 2 to 7 similar to segment 1, but dorsal sclerites smaller than those on first segment.

Spiracular atrium (Fig. 5D). Segment 8 with oval dorsal plate; median part of posterior edge of segment 8 with four strong projections, median two larger than lateral ones; procercus incompletely sclerotised, with one long and two short setae. Segment 9 trilobed, partially sclerotised; each lateral lobe with short, incompletely sclerotised acrocercus; urogomphi short, one-segmented; prostyli very long.

Second instar larva (Figs. 3A, C, 4, 8B). Closely similar to third instar larva.

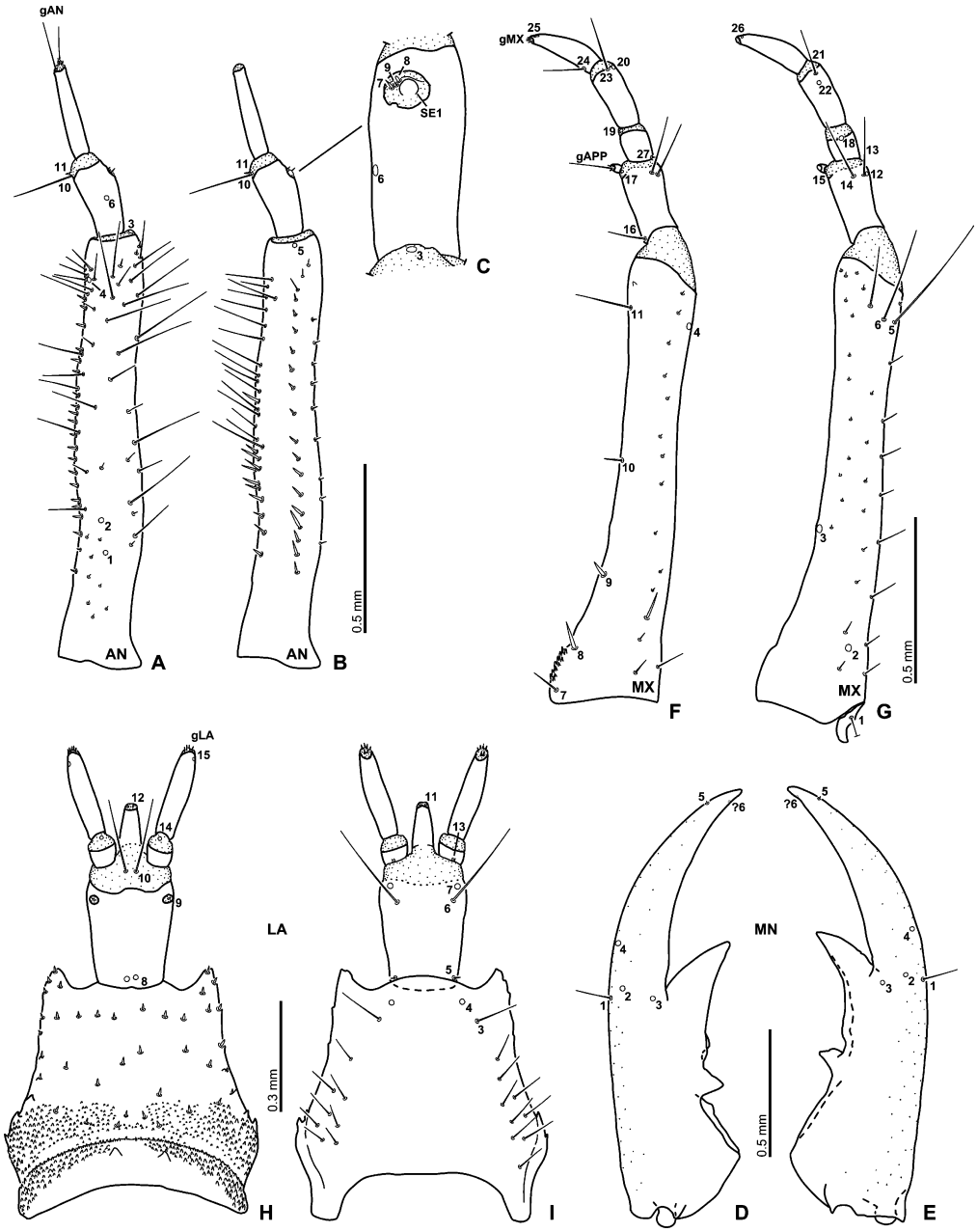

Fig. 4. Head appendages of *Hydrochara libera* (SHARP, 1884), second instar larva. —— A–B, Antenna, dorsal (A) and ventral (B) view; C, the second antennomere, lateral view; D–E, mandible, dorsal view; F–G, maxilla, dorsal (F) and ventral (G) view; H–I, labium, dorsal (H) and ventral (I) view.

First instar larva (Figs. 1–2, 3B, 8A). Similar to second and third instar larvae.

Head. Nasale weakly projecting anteriorly (Fig. 1C–D).

Antenna (Fig. 2A–B) proportionally stouter than that of second and third instar larvae, and anten-

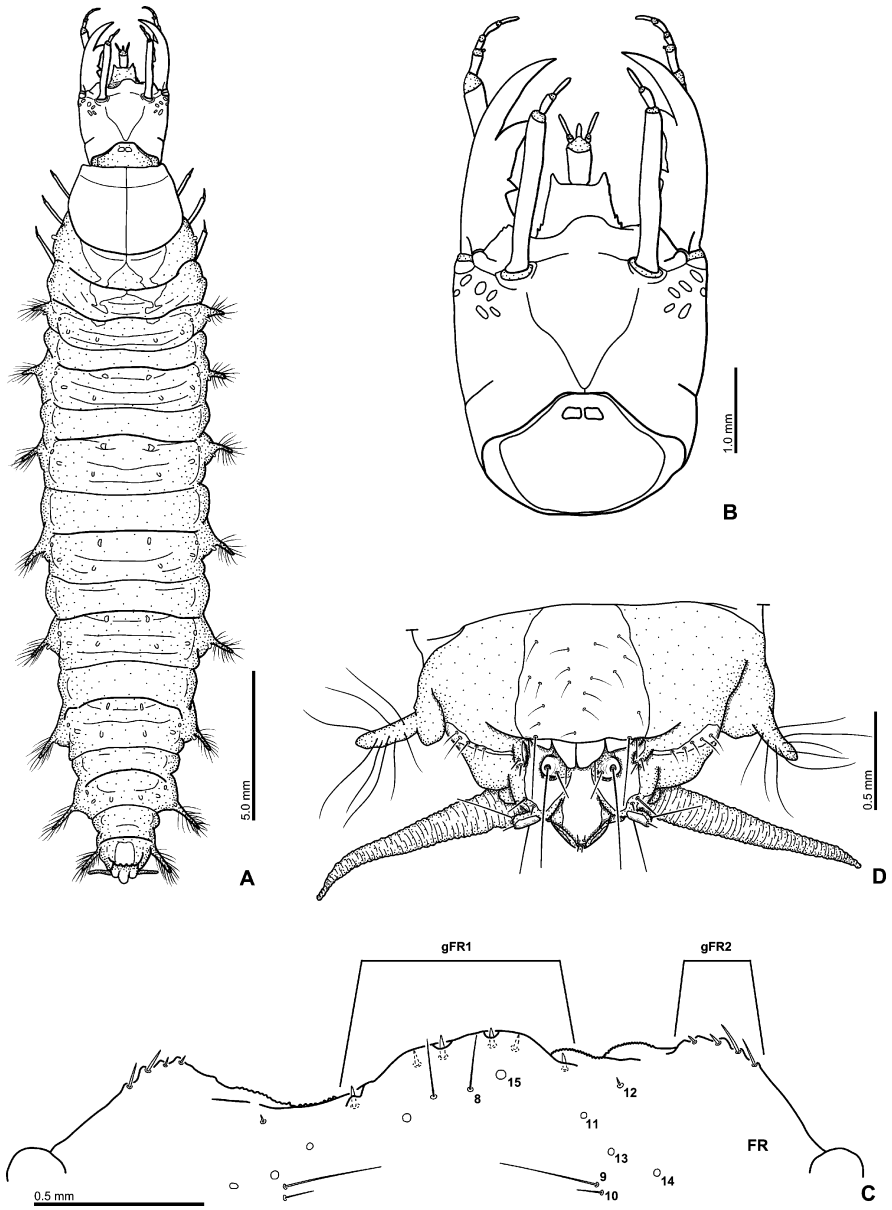

Fig. 5. *Hydrochara libera* (SHARP, 1884), third instar larva, dorsal view. —— A, Habitus; B, head; C, detail of anterior margin of head capsule; D, spiracular atrium.

nomere 1 proportionally shorter than that of second instar larva. Inner face of antennomere 1 with about two to four small but distinct tooth-like cuticular projections.

Maxilla (Fig. 2E–F) proportionally stouter than that of second and third instar larva.

Labium (Fig. 2G–H). Anterior corners of mentum weakly projected anteriorly. Prementum and labial palpi proportionally stouter than these of second and third instar larvae.

Thorax. Prothorax as wide as or slightly narrower than head capsule.

Abdomen. Lateral projections proportionally longer than those of second and third instars (Fig. 3B).

Primary chaetotaxy of head (first instar larva). Frontale (Fig. 1A, C–D) altogether with 48 sensilla. Central part with three pairs of sensilla (FR1–3) divergent posteriad; FR1 short seta, rather close to frontal line; FR2 pore-like, situated more anteriorly and more mesally than FR1, at midlength between FR1 and FR3; FR3 short and rather stout seta, situated more anteriorly and slightly more mesally than FR2. Three rather short setae (FR5–7) and one pore-like sensillum (FR4) close to antennal socket; FR4 located mesally to antennal socket, FR5–6 situated posteromesally to antennal socket; FR5 behind FR6, FR6 between FR4 and FR5; FR7 close to inner margin of antennal socket. Nasale with a group of six equidistant, stout and short setae (gFR1) and with four additional sensilla (one pair of short setae and one pair of pore-like sensilla) medially; the second stout seta from each side on ventral surface of nasale (Fig. 1D). Each epistomal lobe with a group of four rather stout setae (gFR2); lateral two longer than mesal ones. Rather long seta FR8 and pore-like sensillum FR15 placed posteriorly to median part of nasale; FR8 mesally to FR15. Two setae (FR9–10) and two pore-like sensilla (FR13–14) situated anteromesally to antennal socket; FR10 shorter than FR9, behind FR9; FR13 situated anteriorly and slightly laterally to FR9; FR13 laterally to remaining three sensilla. One pore-like sensillum FR11 and one short seta FR12 situated posterolaterally to each lateral-most seta of gFR1; FR12 anterolaterally to FR11.

Parietale (Fig. 1A–B) with 30 sensilla each. Dorsal surface with a group of five sensilla (PA1–5) forming irregularly longitudinal row at midwidth in posterior part of parietale; PA1–2 and 4–5 short setae, PA3 pore-like. PA6 pore-like, located posteromesally close to joint of coronal and frontal lines, more distant from posterior margin of head than PA1. PA7 moderately long seta, situated anteromesally to PA5, at midlength between PA6 and PA8 but laterally to line connecting PA6 and PA8. Two rather long setae (PA12–13) and one long seta (PA14) on median part of dorsal surface of parietale, forming triangular group; PA12 and PA13 closely aggregated, located posteriorly to PA14; PA12 mesally to PA13; FR 14 posteriorly to stemmata. Seta PA8 long, behind antennal socket close to frontal line; seta PA9 long, situated posterolaterally to antennal socket. Pore-like sensillum PA10 at midlength between PA8 and 14, between mesal two stemmata. PA11 long seta, situated laterally to PA9–10, close and posteriorly to anterior row of stemmata. PA15–18 forming irregular transverse row on anterior one-third of lateral surface of head capsule; PA15 and PA17 pore-like, PA16 rather long seta and PA18 long seta; PA18 located dorsally to remaining ones, PA17 ventrally to remaining ones; PA16 between PA18 and PA17; PA15 posteroventrally to PA18. Anterior corner of epicranium with one pore-like sensillum (PA19), two rather long setae (PA20–21), and one long seta (PA22); PA19 located dorsally to remaining ones; PA20 between PA19 and PA21; PA22 ventrally to remaining sensilla. Three pore-like sensilla (PA23–25) on anterior margin close to ventral mandibular acetabulum; PA23 on outer face; PA24 and PA25 closely aggregated, on inner face; PA24 behind PA25. Two long setae (PA26 and PA28) and one pore-like sensillum (PA27) forming triangular group, situated ventrally on median part of parietale; PA26 and PA28 undistinguishable (the position of PA26 and PA28 shows intraspecific variation). Two pore-like sensilla (PA29–30) on posterior part of ventral parietale; PA29 situated mesally to PA30; PA30 on posterolateral corner.

Antenna (Fig. 2A–B). Antennomere 1 with five pore-like sensilla (AN1–5) and several very small sensilla-like structures; AN1 situated dorsally on basal 0.29, AN2 dorsally on basal 0.34, AN3 laterally on distal margin of intersegmental membrane between antennomeres 1 and 2; AN4 subapically on inner face of sclerite; AN5 ventrally on distal margin. Antennomere 2 with one pore-like sensillum (AN6) situated dorsally on basal 0.43 of sclerite; three setae (AN7–9) and sensorium SE1 on small membranous area completely surrounded by sclerite, located subapically on outer face of anten-

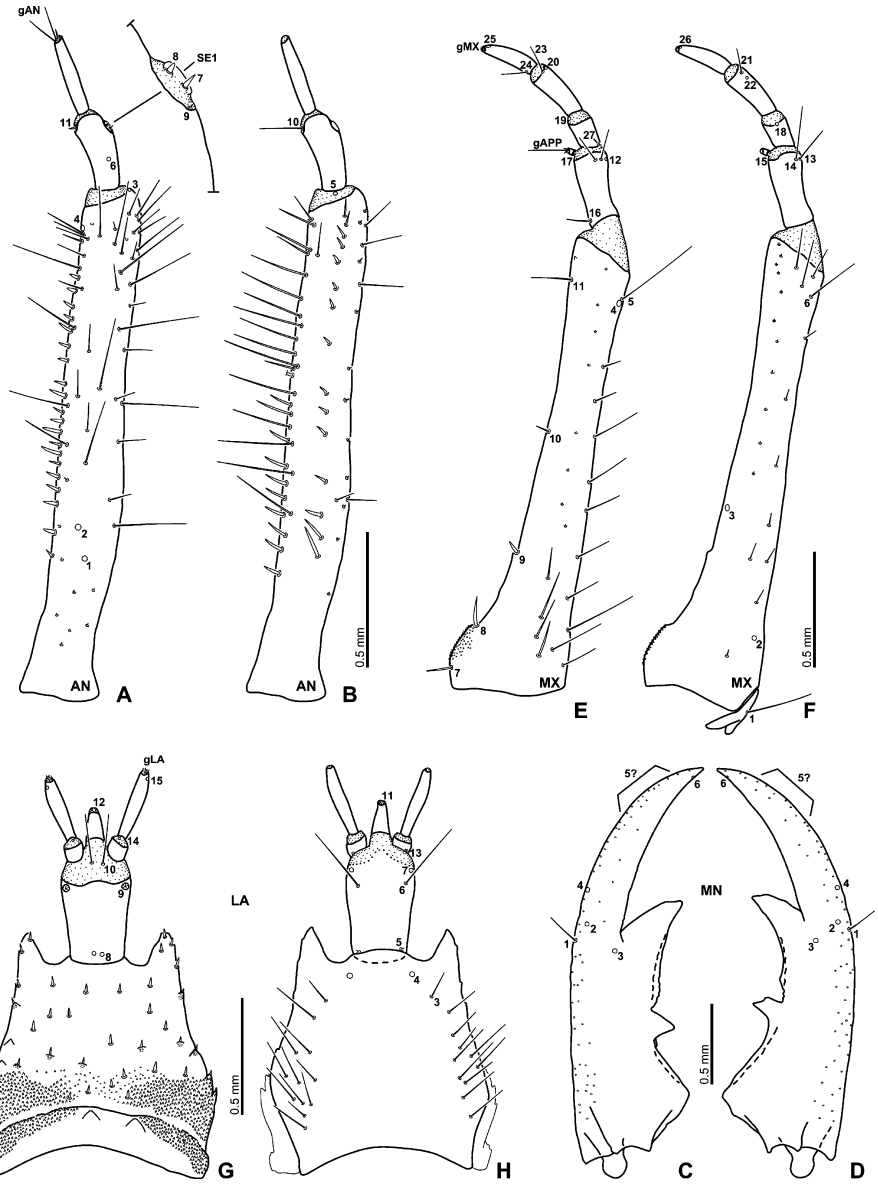

Fig. 6. Head appendages of *Hydrochara libera* (SHARP, 1884), third instar. —— A–B, Antenna, dorsal (A) and ventral (B) view; C–D, mandible, dorsal view; E–F, maxilla, dorsal (E) and ventral (F) view; G–H, labium, dorsal (G) and ventral (H) view.

nomere 2; AN7 short, stout; AN8 short, rounded apically, between AN7 and AN9; AN9 small; position of AN7–9 variable intraspecifically; SE1 small, rounded, below AN7–9 in dorsal view; AN10–11 on inner face of intersegmentary membrane between antennomeres 2 and 3, AN10 rather long, AN11 short, stout, both setae close to each other. Antennomere 3 with apical sensilla (gAN) in apical membranous area; gAN with two rather long setae and a few short setae.

Mandible (Fig. 2C–D) with two setae (MN1 and MN5) and four pore-like sensilla (MN2–4 and

MN6), and with several very small sensilla-like structures. MN1 rather short, on median part of outer face of mandible. Sensilla MN2–4 pore-like, on about midlength of dorsal surface; MN2 located laterally of line connecting MN3 and MN4, close to MN1; MN3 situated posteromesally to MN2; MN4 on lateral face, anteriorly to MN1–3. MN5 minute, situated subapically on outer face of mandible; MN6 subapically on inner face of mandible.

Maxilla (Fig. 2E–F). Cardo with one rather short ventral seta (MX1). Stipes with a row of five short to rather long setae (MX7–11) situated dorsally along inner face; MX7–9 and MX9–11 almost equidistant from each other; MX7–8 short, rather stout, MX8 longer and stouter than MN7; MX9 stout, shorter than MX8; MX10 trichoid, longer than MX9; MX11 the longest, trichoid. Pore-like sensillum MX2 situated ventrally on basal 0.18 of lateral portion of sclerite; pore-like sensillum MX3 on basal 0.43 of inner face; one pore-like sensillum (MX4) and two rather long setae (MX5–6) situated lateroventrally on outer face of apical part of sclerite; MX4 on lateral face; MX5 between MX4 and MX6, longer than MX6; MX6 ventrally to MX4–5. Dorsal surface of palpomere 1 with one moderately long, trichoid seta (MX16) situated basally on dorsal inner face; lateral to ventral surface of sclerite of palpomere 1 with three sensilla (MX12–14) close to distal margin of sclerite; MX12 pore-like on lateral face, MX13 rather short seta between MX12 and MX14, MX14 long seta on midwidth of ventral surface of sclerite. Two rather small pore-like sensilla (MX15 and MX17) on membrane behind inner appendage; MX17 on dorsal surface, MX15 on ventral surface. Inner appendage with one rather long seta and a few short setae (gAPP). Palpomere 2 with two pore-like sensilla (MX18 and MX19) and one minute seta (MX27); MX18 situated ventrally on median part of distal margin of sclerite; MX19 on inner face of intersegmental membrane between palpomeres 2 and 3; MX27 at base of outer face of sclerite. Palpomere 3 with two rather long setae (MX21 and MX23) and two pore-like sensilla (MX20 and MX22); MX20 on outer face close to distal margin of sclerite; MX21 located ventrally on lateral part of distal margin of sclerite; MX22 behind MX21; MX23 dorsally on outer part close to distal margin, mesally to MX20. Palpomere 4 with one rather long seta (MX24) situated basally on inner face, and with digitiform sensillum (MX25) and pore-like sensillum (MX26) apically on outer face of sclerite; MX25 dorsally, MX26 ventrally. Apical membranous area of palpomere 4 with several minute setae (gMX).

Labium (Figs. 1B, 2G–H). Submentum with two pairs of setae (LA1–2); LA1 long, in each lateral corner, LA2 short, situated anteromesally to LA1 (Fig. 1B). Ventral surface of mentum with one pair of rather short setae (LA3) and pore-like sensilla (LA4) distally; LA3 behind LA4, LA4 close to distal margin. Dorsal surface of prementum with two pairs of pore-like sensilla (LA8–9); LA8 at midwidth of prementum close to basal margin; LA9 slightly digitiform, on small membranous area completely surrounded by sclerite of prementum; the area situated subapically and laterally on sclerite of prementum. Ventral surface of prementum with three pairs of sensilla (LA5–7) on lateral portion; seta LA5 very short on basal margin of sclerite; very long seta LA6 and pore-like sensillum LA7 on distal margin of sclerite; LA6 situated posteromesally to PA7. Membrane between prementum and palpi with one pair of rather long setae (LA10) close to base of ligula. Ligula with two pairs of pore-like sensilla on apical membranous area; LA12 situated dorsally, LA11 ventrally. Palpomere 1 with one minute seta (LA13) situated ventrally on basal margin of sclerite; LA14 on dorsal face of intersegmental membrane between palpomeres 1 and 2. Palpomere 2 with one subapical pore-like sensillum (LA15) on outer face. Apical membranous area of palpomere 2 with several short setae (gLA).

Chaetotaxy of head. Second instar larva. Parietale bearing about 17 rather short secondary setae on anterior part of lateral surface.

Antenna (Fig. 4A–C). Antennomere 1 bearing numerous secondary setae. Dorsal surface with several small secondary setae on basal one-third, variable length of secondary setae on distal one-

third, remaining part with several, variable length of secondary setae, setation of the part spacer than that of distal part; inner face with densely arranged, variable length of secondary setae; short to rather short setae stout, rather long setae trichoid. Ventral face with a longitudinal row of short, stout secondary setae. Lateral face with variable length of secondary setae, setation of lateral face sparser than that of inner face.

Mandible (Fig. 4D–E). Outer face of mandible bearing numerous small secondary setae.

Maxilla (Fig. 4F–G). Outer face of stipes with a row of rather short secondary setae; one long secondary seta close to MX5–6; dorsal surface of stipes with a longitudinal row of secondary setae; basal four rather short, stout (number and shape of these setae show intraspecific variation), remaining setae very short; ventral surface of sclerite of stipes with numerous short secondary setae. Palpomere 1 with two rather long secondary setae dorsally on apical part of sclerite, close to MX12.

Labium (Fig. 2H–I). Dorsal and lateral surface of mentum with about 30–40 short secondary setae altogether, a part of these setae below tooth-like cuticular projections; ventral surface of mentum with about 10 rather short, secondary setae on each lateromedian part of ventral surface.

Chaetotaxy of head. Third instar larva. Closely similar to second instar larva (Figs. 5C, 6).

Description of pupa (Figs. 7, 8C–D, 9D). Colour. Slightly greenish milky white in living individuals (Fig. 9D), white in the specimens fixed with boiling water, entirely greyish white or greyish white with partially black in the specimens fixed with ethanol; eyes of pharate adult reddish brown.

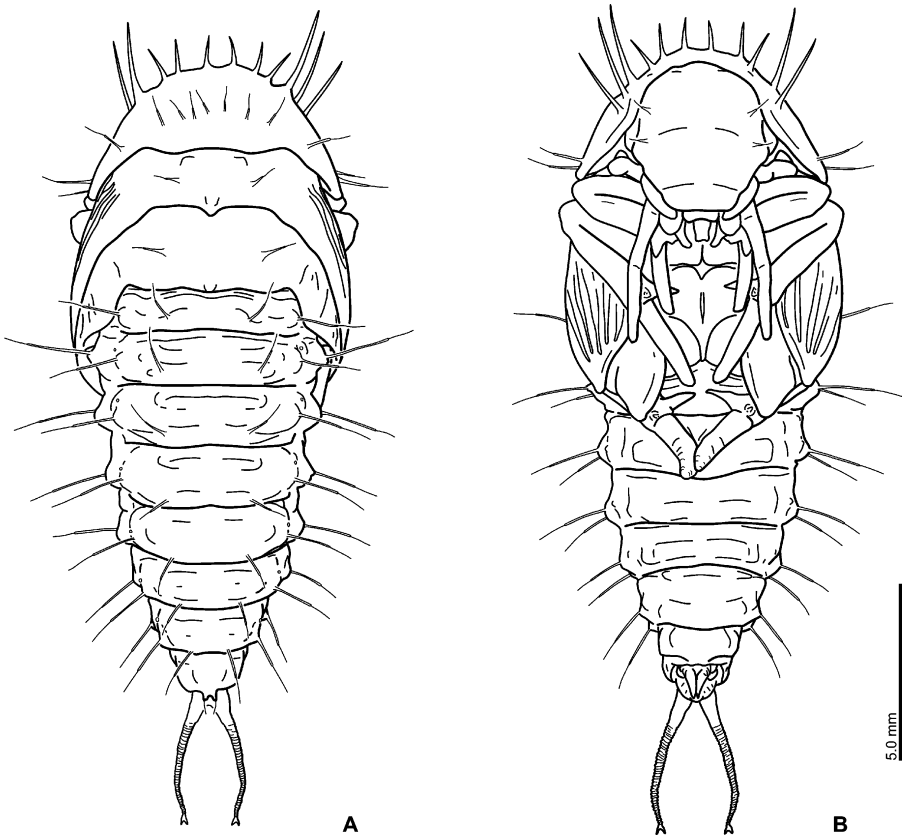

Fig. 7. Pupa of *Hydrochara libera* (SHARP, 1884), dorsal (A) and ventral (B) view.

Body somewhat broad, abdomen more slender. Head, thorax and abdomen with styli, without simple setae; only styli on medioanterior face of pronotum without apical seta (Fig. 8C), remaining ones with apical seta (Fig. 8D).

Head. Head deflexed towards ventral face, completely covered by pronotum in dorsal view; head and eyes partially covered by pronotum in ventral view. Antennae completely covered by head and pronotum in dorsal view, partially visible in ventral view; mouthparts visible in ventral view, maxillary palpi long, projecting close to median part of mesotarsi. Styli on head: two pairs of short supraorbital styli close to inner margin of each eye.

Thorax. Pro-, meso- and metathoracic legs visible in ventral view but metathoracic pair covered by wingpads, only distal portion of tibiae to tarsi visible; metathoracic leg strongly projecting ventrally. Styli on thorax: pronotum with 24 styli on the figured specimen (number of styli on pronotum slightly variable intraspecifically, range of number of styli see below); styli on medioanterior surface stout, without apical seta, remaining ones narrow. Six to seven short styli on medioanterior surface of pronotum; two to three moderately long styli located each lateral side of the median short ones, curved dorsally (Fig. 8C); these styli on anterior surface with finely serrate surface. Seven to eight short styli forming transverse row on pronotal disc, one pair of short styli located laterally to these ones; each posterolateral corner of pronotum with two somewhat long styli. Meso- and metanotum with one pair of styli on each median portion.

Abdomen. Abdomen with nine segments, attenuate towards apex. Segment 9 with well-developed, non-articulated urogomphi; urogomphi bifurcate at apex. Styli on abdomen: segment 1 with two

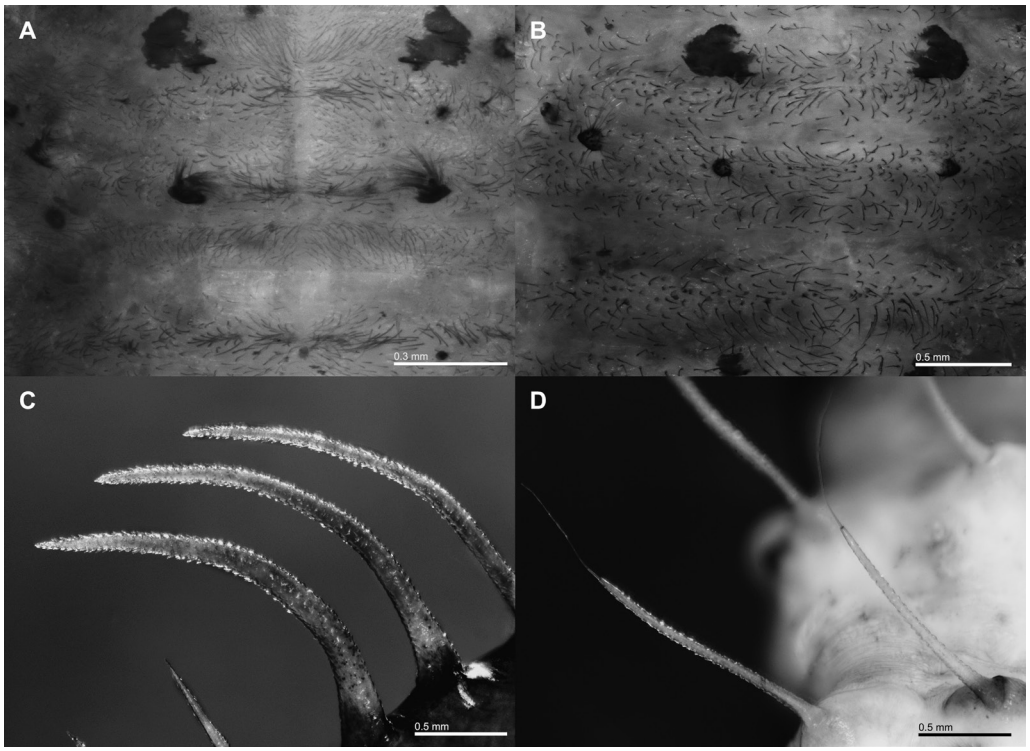

Fig. 8. *Hydrochara libera* (Sharp, 1884). —— A–B, Larva, detail of the dorsal surface of the first abdominal segment, first (A) and second (B) instar; C–D, pupa, styli on anterior face of pronotum (A) and abdomen (B).

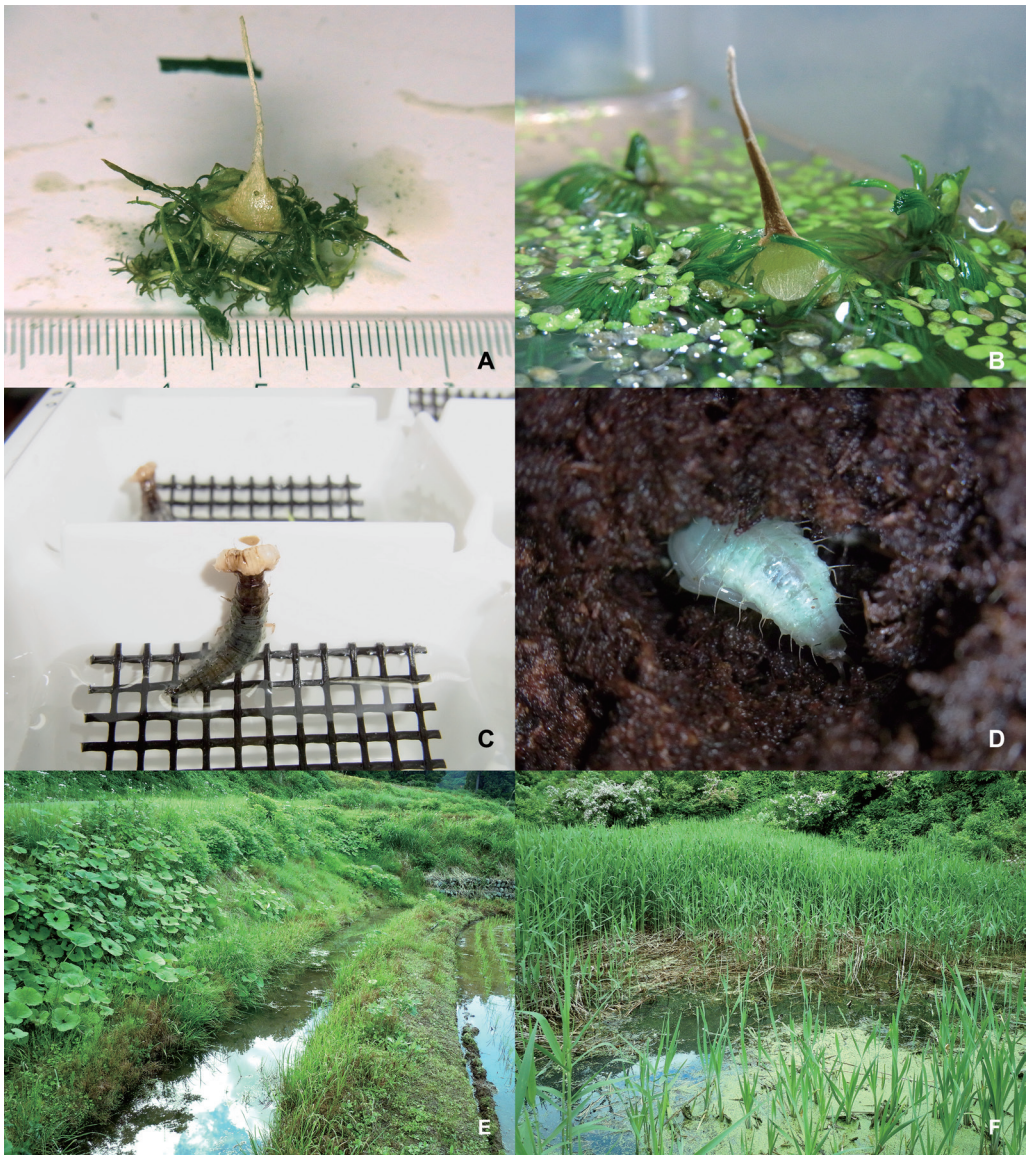

Fig. 9. Immature stages and habitats of *Hydrochara libera* (SHARP, 1884). —— A–B, Egg-case, collected in the field (A) and under rearing (B); C, third instar larva, feeding off maggot under rearing; D, pupa and pupal chamber, under rearing (the pupa moved actively while the author was taking photos, therefore the pupa in pupal chamber may be artificial situation). E–F, Habitats of *H. libera*, Hanzôgane, Nagaoka-shi, Niigata Prefecture, Japan, 14 June, 2011. Photos A–D by Y. MINOSHIMA, E–F by Y. IWATA.

pairs of rather long styli, one on median part, one laterally to the remaining stylus, close to spiracle; segments 2 to 7 with a transverse row of six styli, median two pairs similar in shape and position to these on segment 1, remaining one pair on lateral face. Segment 8 with one pair of stylus distally on median part.

Biology. The habitat of *H. libera* has been reported by previous studies (*e.g.*, NOMURA & HAYASHI, 1998; HAYASHI & YAGI, 2000; HAYASHI, 2011). The environment of our collecting sites of adults (Niigata Prefecture) and egg-case and larvae (Hokkaidô) seem to be fit to previous reports. They were collected in the pond (Fig. 9F) or in the very slowly flowing water (Fig. 9E); these collecting sites are rich in aquatic or semi-aquatic plants and situated in sunny location. Larvae live in the same habitats as adults.

Immature development takes about one month under the room temperature (its length may depend on the temperature and the amount of food), the period of each developmental stages as follows: 5–7 days in the egg, 3–4 days in the first instar larva, 3–4 days in the second instar larva, 5–6 days in the third instar larva. After that, larvae leave water for pupation. Pupation takes place in a pupal chamber in the soil. Prepupa and pupa take about one week in total.

Remarks. Five species of the genus *Hydrochara* have been described on the morphology of their immature stages in the previous studies: *Hydrochara affinis* (HOSOI, 1947; MORIOKA, 1955; MINOSHIMA & HAYASHI, 2011), *H. caraboides* (LINNAEUS, 1758) (*e.g.*, BØVING & HENRIKSEN, 1938; ARCHANGELSKY, 1997), *H. obtusata* (SAY, 1823) (*e.g.*, RICHMOND, 1920; WILSON, 1923), *H. occulta* (ORCHYMONT, 1933) (MATTA, 1982), and *H. soror* SMETANA, 1980 (MATTA, 1982). The abdominal lateral projections within all *Hydrochara* species, larval morphology of which have been known, are distinctly longer than those of other representatives of the family Hydrophilidae (*Berosus* LEACH, 1817 has long lateral 'tracheal gills'; see *e.g.*, ARCHANGELSKY (1997, 2008)). However, the larva of *H. libera* is characterised by the short lateral projections on abdominal segments within the known larvae of the *Hydrochara*. The lateral projections of *H. libera* are more or less proportionally longer than those of other representatives of the Hydrophilini (RICHMOND, 1920; WILSON, 1923; BØVING & HENRIKSEN, 1938; ARCHANGELSKY, 1997; WATTS, 2002; TORRES *et al.*, 2008; MINOSHIMA & HAYASHI, 2011) but the projections are often indistinct especially in the third instar larvae; this is due to the condition of specimen. To identify the undetermined Hydrophilini larvae with the naked eye, it is essential to observe other characters and to pay attention to short lateral projections like *H. libera*.

Key to the Species of the Larva of Genus *Hydrochara* from Japan (All Instars)*

1. Inner face of antennomere 1 with distinct tooth-like cuticular projections. Head and its appendages without secondary sensilla. (First instar) ····· 2
— Inner face of antennomere 1 smooth. Head and its appendages bearing secondary sensilla. (Second and third instars) ····· 3
2. Inner face of antennomere 1 with large, very strong tooth-like cuticular projections. ····· *Hydrochara affinis* (SHARP)
— Inner face of antennomere 1 with a few, small but distinct tooth-like cuticular projections. ····· *Hydrochara libera* (SHARP)
3. Clypeolabrum almost symmetrical, lateral projections on abdominal segments long. ····· *Hydrochara affinis* (SHARP)
— Clypeolabrum somewhat asymmetrical, lateral projections on abdominal segments short. ····· *Hydrochara libera* (SHARP)

* Second instar larva of *Hydrochara affinis* was not examined.

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We thank Masahiro ÔHARA (Hokkaido University Museum) for his kind advice and critical reading of the draft of the manuscript and Yuuki KAMITE (Nagoya City Public Health Research Institute) for providing us with his larval materials of *Hydrochara libera*. We also thank Martin FIKÁČEK (National Museum, Praha) for his constructive criticism on the manuscript. This study was partly supported by the Grant-in-Aid for JSPS Fellows (10J03421) to Y. MINOSHIMA.

要　　約

蓑島悠介・岩田泰幸・林　成多：エゾコガムシ（鞘翅目ガムシ科）の卵嚢・幼虫・蛹の形態．―― 日本産コガムシ属はコガムシ *Hydrochara affinis* SHARP とエゾコガムシ *H. libera* SHARP の 2 種が知られている．前者は，これまでに卵嚢・幼虫・蛹の形態が記載されているが，後者は未知であった．筆者らは，飼育と野外採卵・採集によって得た日本産の材料をもとに，エゾコガムシの卵嚢・幼虫・蛹の形態を詳細に記載し，コガムシとの比較を行った．

本種の未成熟期は，以下の形態的特徴でコガムシとの区別が可能である．卵嚢：マストは細く長い（コガムシは太く短い）（図 9A–B）．幼虫：1) 1 齢幼虫の触角第 1 節内面に，わずかな数の小さな，しかし明瞭な突起を備える（コガムシはより多くの，大きな突起を備える）（図 2A–B）；2) Clypeolabrum はやや左右非対称（コガムシはほぼ左右対称）（図 1C–D, 3A, 5C）；3) 腹部側面の突起は短い（コガムシは非常に長い）（図 3B–C, 5A）．蛹については，細井 (1947) によるコガムシの先行研究と比較して，1) 前胸背板前縁部の長い突起が 3 もしくは 2 対（コガムシでは 4 対）；2) 前胸背板前縁部の短い突起が 6–7 本（コガムシでは 5 本）であることから区別が可能であるように思われる（図 7）．しかし，本研究で前胸背板の突起の数に個体変異があることが判明した．細井 (1947) では個体変異に触れられていないため，確実な同定が可能かどうかは現時点では不明である．

これまで，コガムシ属幼虫の属レベルでの同定は，腹部側面の突起が顕著に長いことで容易とされてきた（ゴマフガムシ属 *Berosus* は，腹部側面に長い糸状の突起を備えるが，この突起は気管鰓であり，コガムシ属のもつ突起とは異なる形質である）．しかし，本種の腹部側面突起は短い．これはコガムシ属の既知幼虫 5 種と比較しても特異な形質状態である．この突起は固定時に体が膨張すれば明瞭であるが，特に三齢幼虫においてしばしば目立たない．そのため，本属幼虫の肉眼による同定においては，本種のような短い突起を持つ種が他に存在する可能性にも留意し行わなければならないだろう．

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