

## Functional morphology and evolution of specialized mouthparts of Cephenniini (Insecta, Coleoptera, Staphylinidae, Scydmaeninae)

Paweł Jałoszyński<sup>a,\*</sup>, Rolf G. Beutel<sup>b</sup>

<sup>a</sup> Museum of Natural History, Wrocław University, Sienkiewicza 21, 50-335 Wrocław, Poland

<sup>b</sup> Institut für Spezielle Zoologie und Evolutionsbiologie, FSU Jena, Jena, Germany

### ARTICLE INFO

#### Article history:

Received 23 December 2011

Accepted 16 July 2012

#### Keywords:

Scydmaeninae

Cephenniini

Anatomy

Labium

Evolution

### ABSTRACT

We present the results of a morphological study of the labium and labial (premental) discs in Cephenniini, ant-like stone beetles feeding on oribatid mites. The discs are composed of a cuticular plate connected by a circumferential ring with the hypopharyngeal suspensorium. The discs have likely developed from the premental cuticle and from internal sclerotizations of the labium. The shape of the external plate can be changed from flat to concave and vice versa by contractions and relaxations of the labial muscles. Contractions result in a flat or only slightly concave shape whereas during relaxation the discs become strongly concave and adhere tightly to the captured mite. Once this is achieved, detaining of the prey is energy-free. Based on known hypotheses concerning the evolution of Oribatida and Staphylinidae, we exclude the possibility of a co-evolution of “proto-Cephenniini” with yet unarmored “proto-Oribatida”, and suggest three alternative scenarios: i) the predators co-evolved with a particular early lineage of Oribatida that has acquired the hard armor relatively recently; ii) ancestors of Cephenniini gradually shifted from feeding on other types of prey towards fully armored Oribatida; or iii) the labial discs have originally developed for functions not related to feeding.

© 2012 Elsevier Ltd. All rights reserved.

### 1. Introduction

The Cephenniini is one of the largest tribes of Scydmaeninae, currently comprising nearly 400 species grouped in 18 genera and distributed worldwide, except for subpolar zones. Nearly all of them inhabit leaf litter or rotting wood in forests, with the single remarkable exception of *Cephennodes araiorum* (Jałoszyński, 2003), the only known intertidal ant-like stone beetle. In two Oriental genera, *Trurlia* Jałoszyński, 2009 and *Trichokrater* Jałoszyński, 2011, structures resembling trichomes were found on the pronotum, suggesting a possible association with ants or termites, but nothing is known about the biology of these beetles. Cephenniini have remarkably stout bodies (Fig. 1A). They are almost always oval and strongly convex, and the head is not constricted behind the vertex and is retracted into the pronotum up to the posterior margin of the compound eyes. This compact form differs strikingly from the typical slender body of other Scydmaeninae, which are usually strongly constricted between the head and pronotum and between the pronotum and elytra, which gives them the conspicuous ant-like appearance. Cephenniini are very small, typically about 1 mm

in length, with the largest known species measuring about 2.3 mm, and the smallest barely reaching 0.5 mm. The adult morphology of most genera is relatively well known (e.g. Jałoszyński, 2011a, b), while the larvae (Fig. 1B) remain exceptionally poorly studied. Previously published results of phylogenetic analyses (Jałoszyński, 2011a, b) provide a sound basis for reconstructing the evolution of this tribe and supported three major branches: the *Cephennium*, *Cephennodes* and *Cephennomicrus* groups of genera. Recently, strong support for the monophyly of Cephenniini and its placement as the sister group of Eutheiniini was found (Jałoszyński, 2012a).

Despite the commonly accepted view that Scydmaeninae are predators specialized on armored oribatid mites, and possibly also on other small organisms (e.g., Newton, 1991; Newton and Franz, 1998; O'Keefe, 2005), published observations or results of prey preference experiments are exceptionally scarce. Members of some genera (e.g., *Scydmaenus* Latreille, 1802) were observed to feed on live armored mites (Schuster, 1966; Schmid, 1988; Molleman and Walter, 2001) and on live springtails or soft-bodied acarid mites (Jałoszyński, 2012b), or scavenging on various dead arthropods (Molleman and Walter, 2001; Jałoszyński, 2012c). Members of Cephenniini, however, were observed feeding only on live Oribatida and Uropodina, with strong preferences towards the former mites (Fig. 2) (Schuster, 1966; Schmid, 1988; Jałoszyński, unpublished obs.).

\* Corresponding author.

E-mail address: [scydmaenus@yahoo.com](mailto:scydmaenus@yahoo.com) (P. Jałoszyński).



Fig. 1. *Cephennium thoracicum*. (A) Adult; (B) larva.



**Fig. 2.** Examples of typical feeding posture of adult *Cephennium majus*. The prey, armored Oribatida, are captured and immobilized using labial discs, the cuticle is penetrated by scraping movements of mandibles, digestive juices are injected through a drilled hole, and liquefied tissues are ingested by the predator.

Schuster (1966) was the first author to describe how the blunt and rather short mandibles of *Cephennium* Müller et Kunze, 1822 are used to penetrate the mite cuticle by sustained scraping, and Schmid (1988) described unusual paired labial discs in the same genus, located on the large prementum, between broadly separated bases of the labial palps. These discs are used to capture and detain the relatively large, subglobose, often smooth and sparsely setose prey. Although the discs were often referred to as suckers, their internal structure was never studied, and an alternative mechanism of adhesion by a sticky secretion could not be excluded. It was observed that a mite is captured by the flexible frontal part of the prementum, and the discs are involved in this process. Subsequently, the mandibles drill a small hole in the cuticle (an example of a typical damage is shown in Fig. 3), digestive juices are injected and the liquefied tissues are slowly ingested by the adult or larva of *Cephennium*. The entire process may take several hours and the predators produce only liquid faeces (Schuster, 1966).

Other Scydmaeninae feeding on armored mites (e.g., Cyrtoscydmini or some Scydmaenini) utilize a different technique. They capture the prey and often lift it up with the mandibles and fore legs, insert one mandible into a natural body opening (the mouth, anal or genital opening), break apart any movable parts around the insertion site (mouthparts, anal or genital plates), and then inject digestive juices (Schmid, 1988; Molleman and Walter, 2001;

Jałoszyński and Olszanowski, in preparation). With a single known exception, ant-like stone beetles using this technique do not possess labial discs. Their prementum is small and unmodified, typically with a median pair of long, bristle-like sensilla, and approximate bases of the labial palps (e.g., Jałoszyński, 2005). The only exception is *Stenichnus* Thomson, 1859 (Cyrtoscydmini), which has a single pair of small and weakly sclerotized adhesive pads on the prementum. However, the mandibles in this genus are always long and falciform, and are not used to drill a hole in the mite cuticle. Instead, *Stenichnus* was observed to use the labial pads to attack and sometimes lift up the prey. But after that, the mandibles and fore legs immediately take over the holding function and one mandible is inserted into a body opening (Schmid, 1988; Molleman and Walter, 2001). Moreover, various species of *Stenichnus*, despite having very uniform mouthparts, show different prey preferences, some feeding mostly on lens-shaped, densely setose and strongly microsculptured Uropodina, and not on nearly spherical, sparsely setose and smooth Oribatida, or even feeding on soft-bodied mites and other small organisms (Jałoszyński, unpublished observations). In such cases the beetles use the prementum only in the very initial stage of attack.

Although published observations or feeding experiments concerning Cephenniini were only carried out with some European species of *Cephennium*, the labial discs located on the prementum between broadly separated palp bases can be found in all genera of

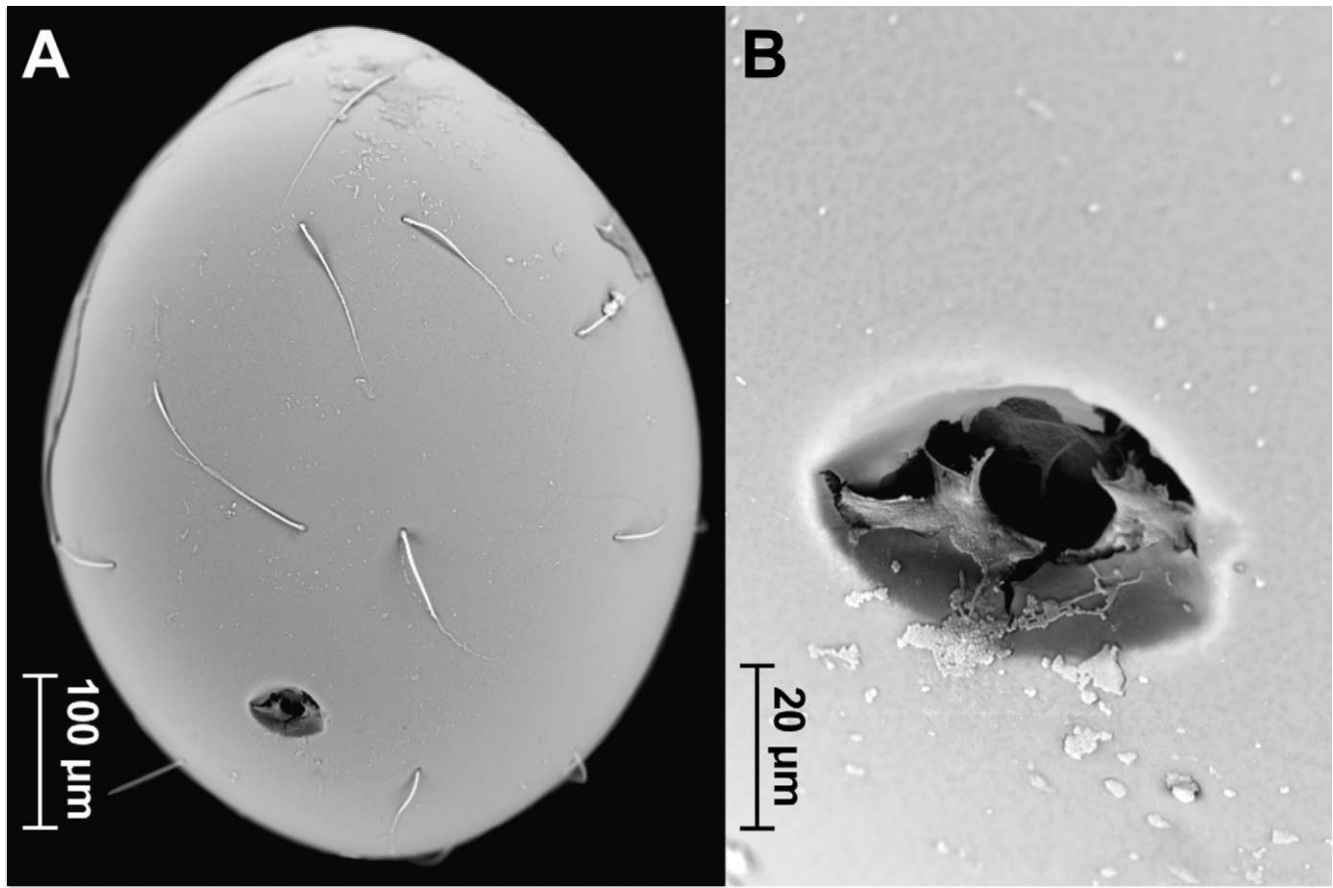
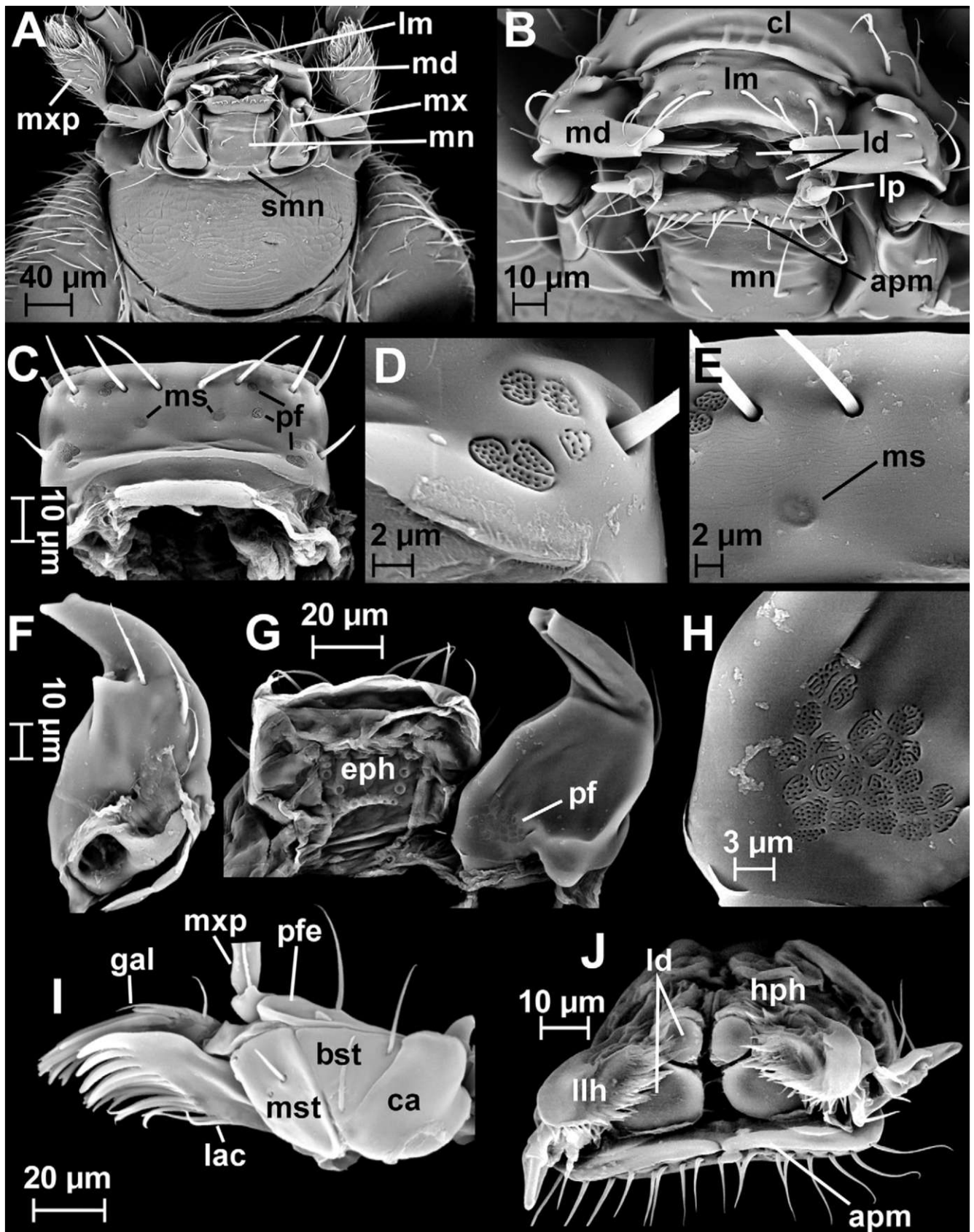


Fig. 3. Typical damage caused in armored mite cuticle by feeding of adult *Cephennium*. (A) Phthiracarid mite remains after feeding of *Cephennium majus*; (B) close-up of the drilled hole.

the tribe. This suggests that feeding habits and techniques among all Cephenniini may be similar, and the preferred or exclusive prey for all genera are oribatid or uropodine mites. Feeding on armored mites, the presence of labial discs and the characteristic scraping action of the mandibles to penetrate hard cuticle were not only observed in adults, but also in larvae of *Cephennium* (Schuster, 1966; Schmid, 1988). This makes the Cephenniini a rare example of similar and highly unusual morphological adaptations both in adults and larvae, used for the same specialized mode of feeding. The common occurrence and abundance of armored mites in virtually all types of forest soils make them a suitable source of food for predators that managed to develop morphological and behavioral mechanisms to compromise the defensive systems of these arthropods. The Cephenniini have achieved this goal and gained an access to a very reach source of food. It seems plausible that they exert a significant impact on soil biology in subtropical and tropical forests, where both Oribatida and Scydmaeninae are common and abundant. Moreover, Cephenniini, which can be as small as 0.46 mm (undescribed species of *Cephennococcus* Jałoszyński, 2011b from Borneo), are the smallest known predacious beetles, at least in terms of body length. Only some extremely slender Staphylinidae belonging to Leptotyphlinae or Euaesthetinae may have a similar body volume, but are longer (A. Newton, pers. comm.) and do not have modified mouthparts. Therefore, a detailed functional characterization of these unusually specialized mouthparts may help to understand structural limitations of miniaturization in predators, and their possible co-evolution with a well-protected prey.

The present study was carried out to address the following morphological, functional and evolutionary issues: i) are the labial discs suckers or sticky pads?; ii) is detaining a prey with the discs for several hours an energy-consuming process? iii) from which structure(s) evolved the discs? In order to answer these questions, we examined the external and internal structure of the specialized mouthparts of Cephenniini. The genus *Cephennium* was selected for study, as it is apparently strictly specialized to feed on armored mites (Schuster, 1966; Schmid, 1988). In long-term rearing and prey preference experiments, the first author confirmed that three European species of *Cephennium* (*Cephennium majus* Reitter, 1882, *Cephennium thoracicum* (Müller et Kunze, 1822) and *Cephennium slovenicum* Machulka, 1931) indeed choose exclusively oribatid mites from a broad spectrum of live and dead prey (these results will be described in detail elsewhere). Based on these experiments, the strategy of prey capture and functions of various elements of the mouthparts reported by previous authors were confirmed. The morphology of the mouthparts of adult *Cephennium* was then studied by scanning electron microscopy and the internal structure of the labium was examined using histological sections. Additionally, the morphology of the prementum of species of six other genera of Cephenniini was studied (the taxa were selected to represent all three informal groups of genera currently recognized), and the internal structure of the labium of *Cephennodes* sp. was also taken into consideration. The data obtained for adult mouthparts were compared with the morphology and histology of the larval labium of *Cephennium gallicum* Ganglbauer, 1899. Based on the results, we discuss possible evolutionary



**Fig. 4.** Mouthparts of male (A–I) and female (J) of adult *Cephennium thoracicum*. (A) Head in frontoventral view; (B) anterior part of head in frontal view; (C) labrum in dorsal view; (D) lateral view of labrum; (E) median sensillum of labrum; (F) right mandible in dorsal view; (G) epipharynx and left mandible in ventral view; (H) ventral basal porous fields of mandible; (I) left maxilla in ventral view (maxillary palpomeres II–IV omitted); (J) labium in dorsofrontal view. Abbreviations: apm, anterior premental margin; bst, basistipes; ca, cardo; cl, clypeus; eph, epipharynx; gal, galea; hph, hypopharynx; lac, lacinia; ld, labial disc; llh, lateral lobe of hypopharynx; lm, labium; lp, labial palp; md, mandible; mst, mesostipes; mn, mentum; ms, median sensilla of labrum; mx, maxilla; mxp, maxillary palpus; pf, porous fields; pfe, palpifer; smn, submentum.

scenarios leading to the unusual modifications of the labium of Cephenniini, as an important aspect of the possible prey-predator co-evolution.

## 2. Materials and methods

### 2.1. Taxa examined

**Adults.** Species of seven genera of Cephenniini were included in the study. They represent all three evolutionary lineages within the tribe, which were confirmed as monophyletic by Jałoszyński (2011a) and have a worldwide distribution. The general morphology of the mouthparts was examined with scanning electron microscopy (SEM) using males and females of *C. thoracicum* (Müller et Kunze, 1822) (representing the *Cephennium* group; coll. in Germany). Additionally, the prementum was studied also using SEM in two genera of the *Cephennodes* group: *Cephennodes* sp. (Japan) and *Hlavaciellus* sp. (Borneo); three taxa of the *Cephennomicrus* group: *Cephennomicrus delicatissimus* Jałoszyński, 2009 (Japan), *Trurlia* sp. (Malay Peninsula) and *Cephennula* sp. (Borneo); and a Neotropical species *Cephazteca matagalpana* Jałoszyński, 2011a, b (Nicaragua) of yet unclear position within the tribe. For histological study, 75% ethanol-preserved specimens of *C. thoracicum* (Germany) and *Cephennodes* sp. (Japan) were used. All specimens are deposited in the collection of the first author (Wrocław, Poland).

**Larvae.** Larvae of Cephenniini are very rare in collections and specimens are often unsuitable for histological study. Therefore, the general morphology of the mouthparts of a mature larva of *C. thoracicum* was studied using SEM, while a single mature larva of *C. gallicum* Ganglbauer, 1899 was serially sectioned to examine internal structures. Both larvae were collected in Switzerland and preserved in an unknown liquid medium. The morphology of the mouthparts of larvae of these two species is nearly identical, as confirmed by examination of several additional specimens from the same sample in transparent slide preparations under light stereoscopic and compound microscopes.

The specimens are deposited in the collection of the first author (Wrocław, Poland).

### 2.2. Scanning electron microscopy (SEM)

Dry-mounted adult beetles were relaxed in warm water, dissected, macerated for 10–20 min in a warm 10% solution of NaOH, thoroughly washed, dehydrated in absolute ethanol and air-dried. The studied larva was transferred from the original, unknown liquid medium to FAE (3.4 vol. 35% formalin, 1 vol. acetic acid, 6.7 vol. ethanol) for 2 h, briefly washed in a series of 70/40/20% ethanol and then in distilled water, cleaned for 1 min in 10% NaOH at ambient temp., thoroughly washed in water, transferred to absolute ethanol through a series of 20/40/75% ethanol, kept for 1 h in acetone, and subsequently for 1 h in hexamethyldisilazane. This procedure proved to be useful in the first author's laboratory to prepare very small and fragile larvae for SEM, simultaneously cleaning specimens from various impurities contained in previous preservation fluids, which often tend to crystallize on the specimen surface. Dehydrated specimens were mounted on SEM stubs with carbon tabs and sputter-coated with gold (Pirani 501, Edwards). Microscopy was performed on a LEO 435 VP or an Evo 40 scanning electron microscope.

### 2.3. Histology

Adult and larval specimens stored in liquid media were transferred to 96% ethanol, embedded in Araldite, cut at 1 µm with

a Microm microtome (HM 360), and stained with methylene blue and acid fuchsine (adults) or only with methylene blue (larva).

### 2.4. Photography and image processing

The sagittal sections were photographed with a PixeLINK PL-A622C digital camera on a Zeiss Axioskop. A specimen of *C. thoracicum* was photographed with a Nikon Coolpix 4500 digital camera mounted on a Nikon SMZ1500 microscope. Final image adjustments and annotations were made in Corel PhotoPaint.

## 3. Results

### 3.1. Morphology of mouthparts of adult *C. thoracicum* (Fig. 4A–J)

The labrum (lm) (Fig. 4B–E) is more than twice as broad as long, subrectangular, with a nearly straight anterior margin. Its dorsal surface (Fig. 4C) bears a row of eight symmetrically distributed, anteriorly directed setae close to the anterior margin, an ante-basal transverse groove and numerous sensory or glandular structures. Lateral parts of the labrum (Fig. 4C, D) are occupied by several circular, oval or irregularly-shaped porous fields (pf), some of which are isolated and some adjacent to each other. The median part of the labrum (Fig. 4C, E) bears a pair of median campaniform sensilla or pseudopores (ms).

The epipharynx (eph) (Fig. 4G) is devoid of macro- and microtrichia and its membranous surface bears two mediolateral longitudinal rows of large glandular openings; similar openings form an arcuate transverse row near the base of the epipharynx.

The mandibles (md) (Fig. 4B, F, G, H) are symmetrical, subtriangular in shape and curved ventrad. The broad mandibular base is dorsally convex and ventrally concave, and bears a dense group of more than twenty porous fields (pf) on its ventral surface. A setose prostheca is entirely missing. The robust apical tooth is curved mesad; its apex is a relatively blunt. The apical part of the mandible in ventral view is irregularly impressed, and the apical tooth bears a ventral longitudinal groove. A short and broadly subtriangular subapical tooth is present dorso-medially. Long and thick mandibular setae are present; two are inserted dorsally near the external mandibular margin and one dorso-medially at the base of the subapical tooth.

The maxillae (mx) (Fig. 4A, I) are moderately long. Each is composed of a broad aetose cardo (ca), a subtriangular basistipes (bst) with two basal setae, an elongated mesostipes (mst) with two submedian and one apical seta, an elongate palpifer (pfe) with a single median seta, and an elongate lacinia (lac) and galea (gal). The lacinia (Fig. 4I; lac) bears a robust and curved mesoapical trichial brush. The galea (Fig. 4I; gal) is strongly elongated; it bears a single sub-basal seta on the external margin and a robust and curved mesoapical trichial brush. The maxillary palp (Fig. 4A; mxp) is long, with a minute palpomere I, a strongly elongate and curved palpomere II, a very large palpomere III which is broadened in its distal half; the short but broad, subconical palpomere IV has a truncate apex and is obliquely inserted on the broad apex of the palpomere III.

The labium (Fig. 4A, B, J) is large and elongate, with a strongly sclerotized, subrectangular mentum (mn) bearing several paired setae and a very short, broad submentum (smn) with a single pair of setae. The prementum (Fig. 4J) is connected with the mentum by a narrow flexible membrane; its very broad anterior margin (apm) is divided medially by a short notch and bears a fringe of long and thick antero-ventrally directed setae and two pairs of labial discs (ld). In males the discs (Fig. 4B; ld) are subequal in diameter and composed of a broadly separated anterolateral pair and an approximate posterior pair; in females (Fig. 4J; ld) the anterior discs are distinctly larger than the posterior ones and both pairs are

narrowly separated medially. The labial palps (Fig. 4B,J; lp) are very small; the small palpomeres I and II are about as long as broad; the apex of the strongly elongate palpomere III is blunt.

Moderately large, flexible lateral lobes (llh) densely set with microtrichia are present on the distal region of the hypopharynx (hph) (Fig. 4J); its basal part is medially divided by a distinct longitudinal groove and laterally densely covered with microtrichia.

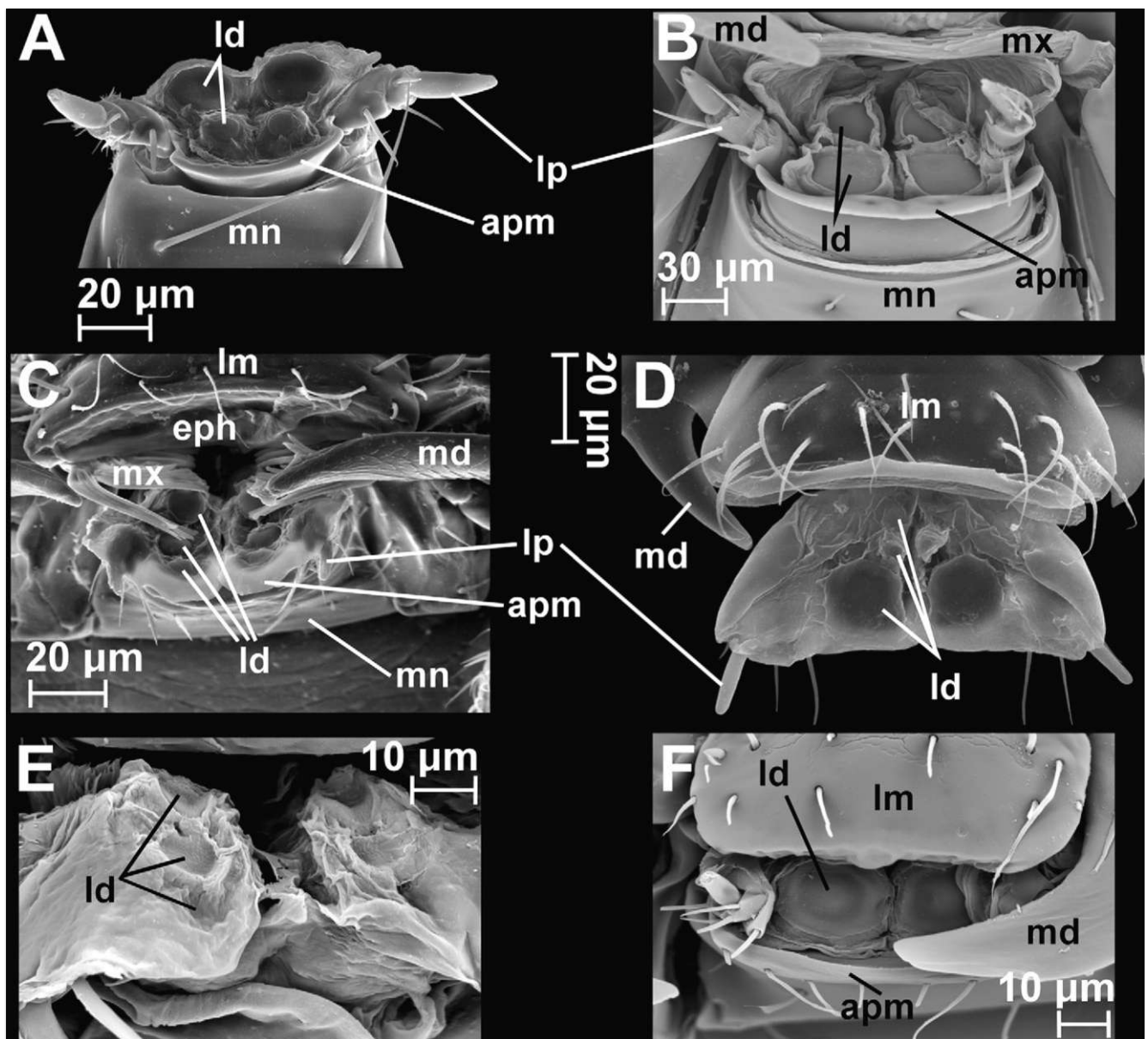
### 3.2. The prementum of other adult Cephenniini (Fig. 5A–F)

The general structure of the prementum in all studied Cephenniini is similar, with broadly separated insertions of small labial palps (lp), a median longitudinal groove, and paired labial discs (ld) occupying most of the anterior surface. Differences between genera can be seen in the number and diameter of the discs. In *Cephennodes* (Fig. 5A), *Hlavaciellus* (Fig. 5B) and *Cephazteca*

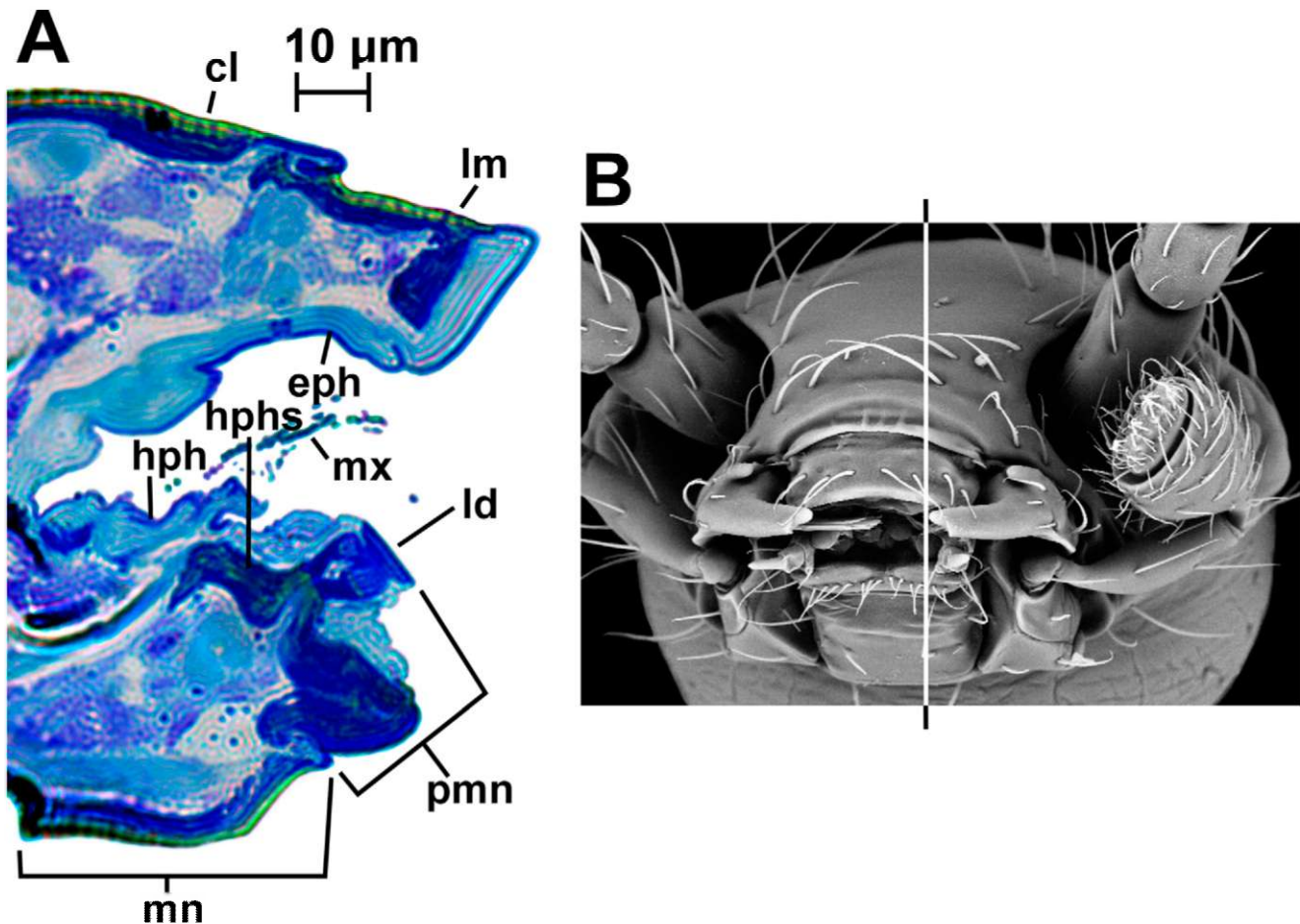
(Fig. 5F) there are two pairs of large, sharply demarcated discs narrowly separated medially (only the anterior pair is visible in Fig. 5F). In all three genera of the *Cephennomicrus* group, i.e. *Cephennomicrus* (Fig. 5C), *Cephennula* (Fig. 5D) and *Trurlia* (Fig. 5E), there are three pairs of discs with moderately distinct margins. Only in *Cephennula* (Fig. 5D) one pair of discs (the anterior pair) is as large as those of *Cephennium*, *Cephennodes*, *Hlavaciellus* and *Cephazteca*; the median and posterior pair of discs are very small. All three pairs of discs are small in *Cephennomicrus* (Fig. 5C) and *Trurlia* (Fig. 5E), and the anterior pair is very weakly developed, as circular impressions without marginal rims.

### 3.3. Anatomy of the labio-hypopharyngeal complex of adult *C. thoracicum* and *Cephennodes* sp. (Figs. 6A, 7A–C, 8, 9A–I)

The prementum (pmn) of *Cephennium* and *Cephennodes* is ventrally separated from the mentum (mn) by a transverse mento-



**Fig. 5.** Prementum of Cephenniini. (A) *Cephennodes* sp. (frontoventral view); (B) *Hlavaciellus* sp. (frontoventral view); (C) *Cephennomicrus delicatissimus* (frontoventral view); (D) *Cephennula* sp. (frontodorsal view); (E) *Trurlia* sp. (frontodorsal view); (F) *Cephazteca matagalpana* (frontodorsal view). Abbreviations: apm, anterior premental margin; eph, epipharynx; ld, labial disc; lm, labrum; lp, labial palpus; md, mandible; mn, mentum; mx, maxilla.



**Fig. 6.** Male of *Cephennium thoracicum*. (A) Sagittal section of anterior part of head; (B) chart of section. Abbreviations: cl, clypeus; eph, epipharynx; hph, hypopharynx; hphs, hypopharyngeal suspensorium; ld, labial disc; lm, labrum; mn, mentum; mx, maxilla; pmn, prementum.

premental fold (mpmf), and dorsally from the hypopharynx (hph) by a transverse hypopharyngeal-premental fold (hpmf) (Figs. 6A, 7A–C, 9A). The hypopharyngeal suspensorium (hphs) (Fig. 7A–C, 8) is very thick and strongly developed; it occupies a significant part of the prementum (pmn) and the anterior part of the mentum (mn). The endo- and exoskeletal labio-hypopharyngeal structures, i.e. the internal sclerites of the palpigers, the hypopharyngeal suspensorium (hphs), the anterior premental margin (apm), the ventral premental surface, and the sclerotized mento-premental fold (mpmf) are fused together thus forming a massive internal scaffold, on which the labial discs (ld) are inserted (Figs. 6A, 7A–C, 8, 9A–I). The discs have a complicated internal structure, with distinctly delimited internal and external plates. The external plate (ep) (Figs. 8 and 9H, I) is a solid circular structure continuous with the cuticle of the anterior surface of the prementum. The internal plate (ip) (Fig. 9H, I) is a similar circular structure, but with a central opening; the base (i.e. the posterior part) of the plate is continuous with the anterior part of the hypopharyngeal suspensorium (hphs). The internal and external plates are connected by a circumferential ring (r) of cuticle composed of longitudinal chitinous fibers (Fig. 9I). The space enclosed anteriorly by the external plate and laterally by the ring forms a lumen (lu) that communicates with the internal labial part through the median opening of the internal disc (Fig. 9H).

The integrated internal premental scaffold and the anterior part of the mentum provide insertion sites for four labial muscles: *M. submentopraementalis* (*M.* 28) (Fig. 8) inserting medially on the

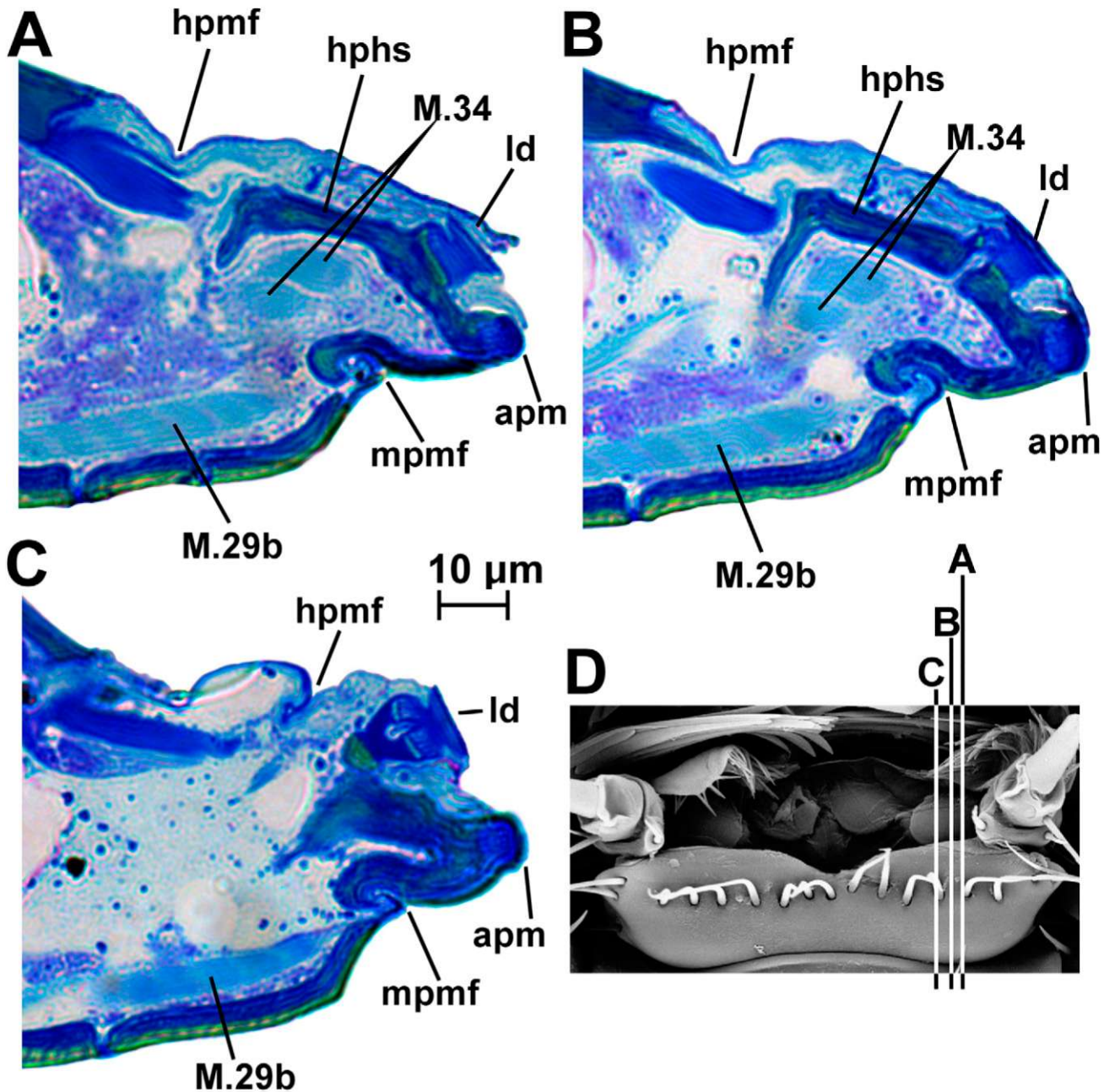
mento-premental fold; *M. tentoriopraementalis inferior* (*M.* 29a, b) with the subcomponent *a* (Fig. 9C) inserting laterally on the hypopharyngeal suspensorium near the internal sclerite of the palpiger, and the subcomponent *b* (Figs. 7A–C, 9A–C) inserting ventrolaterally on the mento-premental fold; *M. tentoriopraementalis superior* (*M.* 30) (Fig. 9E–G) inserting dorso-laterally on the hypopharyngeal-premental fold; and *M. praementopalpalis externus* (*M.* 34) (Fig. 7A–B) inserting on the ventral base of palpomere I. Due to the strongly developed hypopharyngeal suspensorium with its posterior fusion to the converging internal sclerites of the palpigers, *M.* 34 is recurved and strongly oblique in relation to the longitudinal axis of the labium. Additionally, posterolateral parts of the hypopharyngeal suspensorium are insertion sites for *M. frontohypopharyngalis* (*M.* 41) (not shown), which has its origin on the frons and functions as the elevator of the suspensorium.

#### 3.4. The mouthparts of larval *C. gallicum* (Fig. 10A–D)

The labrum (lm) (Fig. 10 A, B) is membranous, semioval, broader than long, and devoid of setae.

The mandibles (md) (Fig. 10B, C) are subtriangular, with a broad base and a relatively slender distal part strongly curved inwards; subapical teeth are lacking. The mesal margin is smooth, while the external margin bears a row of transverse indentations. Two long sub-basal setae are inserted dorsally on the external margin.

The maxillae (mx) (Fig. 10B, D) are large but shorter than the labium, with cardines mesally fused to the submentum. The mala is



**Fig. 7.** Male of *Cephennium thoracicum*. (A–C) Sagittal sections of labium; (D) chart of sections. Abbreviations: apm, anterior premental margin; hphs, hypopharyngeal suspensorium; hpmf, hypopharyngeal-premental fold; ld, labial disc; M. 29b, subcomponent b of *M. tentoriopraementalis inferior*; M. 34, *M. praementopalpalis externus*; mpmf, mento-premental fold.

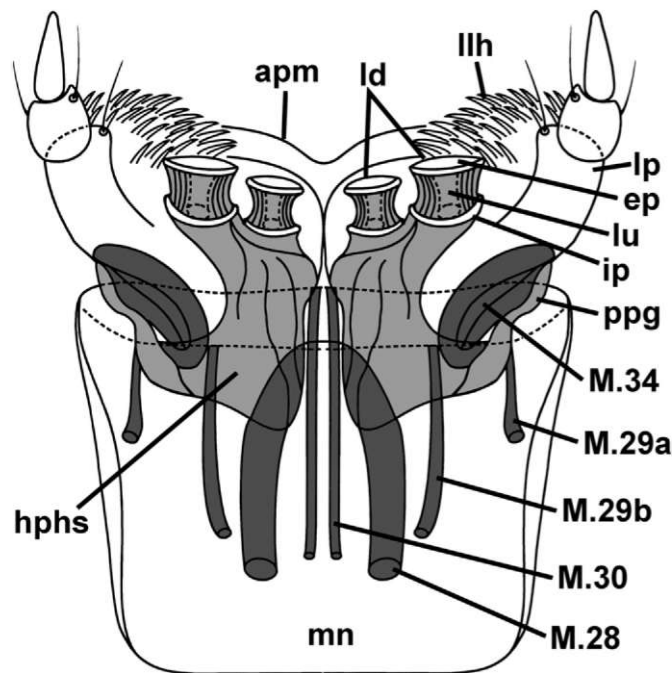
distinctly bilobed, with the apex of each lobe bearing only 2–3 large and thick trichia. The maxillary palps (mxp) are 3-segmented, long and strongly narrowed distally, with a particularly elongate palpomere III.

The labium (Fig. 10B, D) is large in relation to the remaining mouthparts, with a subrectangular mentum and a particularly well developed prementum. The prementum bears three labial discs (ld): a very large median disc or pad, which is approximately hexagonal in shape with well marked margins, and a lateral pair of much smaller, sharply defined circular discs. Two-segmented labial palps (lp) are inserted below each lateral disc, below the anterior premental margin (apm); palpomere I is short and broad; palpomere II is very long and slightly narrowing distally.

### 3.5. Anatomy of anterior alimentary tract and labio-hypopharyngeal complex of larval *C. gallicum* (Fig. 11A–H)

The anterior part of the alimentary tract (Fig. 11A) is distinctly divided into the anterior (precerebral) pharynx (aph), posterior (postcerebral) pharynx (pph) and the oesophagus (oes). These components are distinctly delimited by an array of ventral and dorsal folds forming valves, the anteroventral pharyngeal valve (aphv), the posteroventral pharyngeal valve (pphv), and the dorsal pharyngo-oesophageal valve (poesv) (Fig. 11A).

The very thick labium (lb) is fused with the large hypopharynx (hph) (Fig. 11A–H) and indistinctly divided into a long mentum and a very short prementum. Most of the anterior surface of the



**Fig. 8.** Schematic placement of hypopharyngeal suspensorium in labium of adult male of *Cephennium thoracicum* (dorsal view). Abbreviations: apm, anterior premental margin; ep, external plate of labial disc; hphs, hypopharyngeal suspensorium; ip, internal plate of labial disc; ld, labial disc; llh, lateral lobe of hypopharynx; lp, labial palp; mn, mentum; M. 28, *M. submentopraementalis*; M. 29a, b, subcomponents a and b of *M. tentoriopraementalis inferior*; M. 30, *M. tentoriopraementalis superior*; M. 34, *M. praementopalpalis externus*; ppg, process of palpiger.

prementum is occupied by three labial discs (ld). The central labial disc shows a complex internal structure in sagittal sections (Fig. 11B–H). It is composed of a thin and solid external plate (ep) continuous with the anterior premental cuticle, and a very thick internal plate (ip) ventrally continuous with the cuticle and internal sclerotization of the anterior premental margin (Fig. 11G–H). The internal plate is hollow and the lumen of the labial disc (lu) is connected with the internal space of the labium. Other internal sclerotized structures of the hypopharynx and prementum are poorly developed.

The internal plate (ip) of the labial disc provides insertion sites on the ventral side for the subcomponent b of *M. tentoriopraementalis inferior* (M. 29a, b) (Fig. 11C–E) and dorsally for the *M. praementopalpalis externus* (M. 34) (Fig. 11B–F). The indistinctly marked ventral mento-premental fold is an insertion site of the *M. submentopraementalis* (M. 28) (Fig. 11E–F).

**4. Discussion**

The labial discs of adults and larvae are complex structures. They are not restricted to the surface, but their various parts are continuous with external and internal skeletal elements of the labio-hypopharyngeal complex. The arrangement of cuticular plates and labial muscles provides clues for the mechanism used to capture and immobilize the prey.

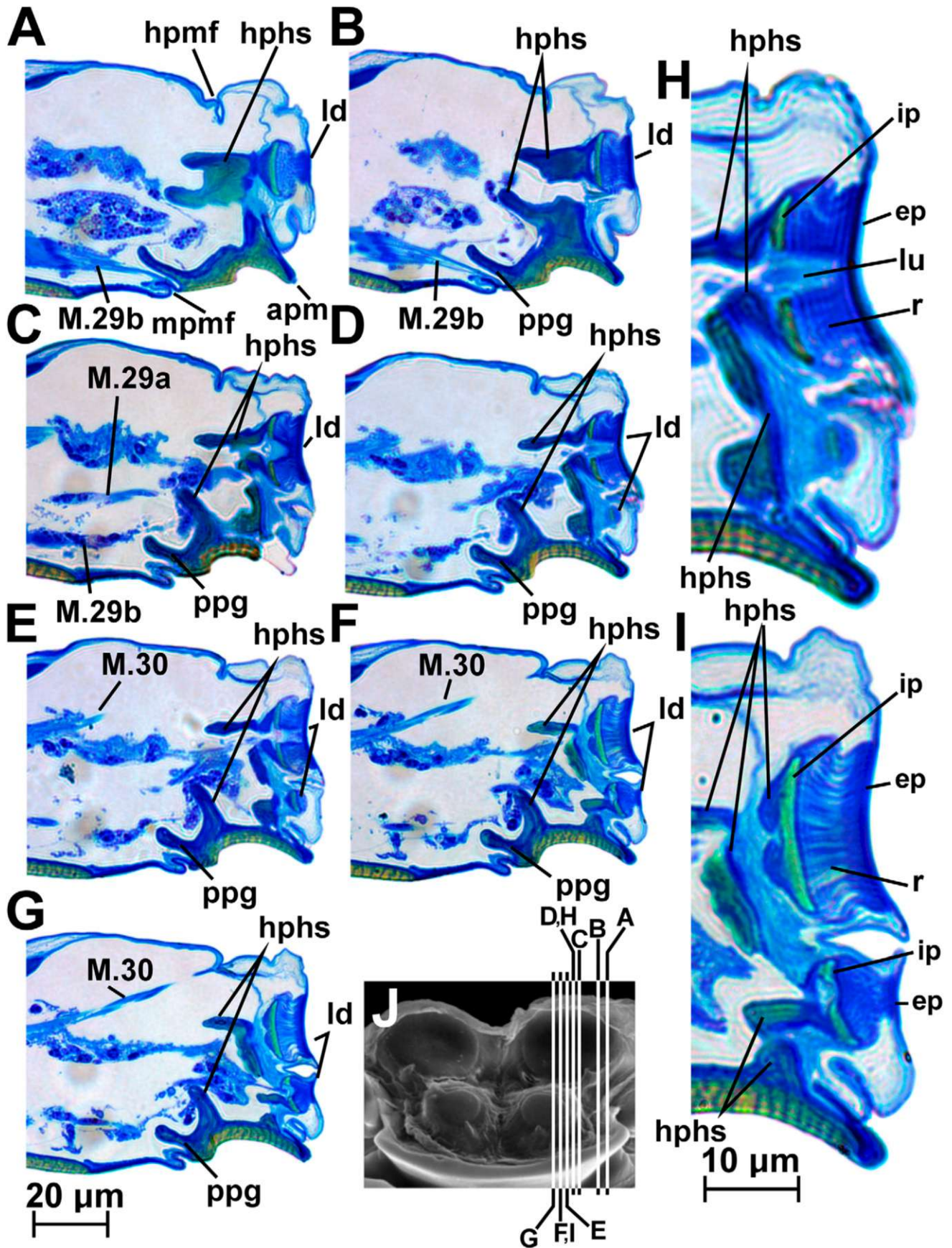
**4.1. Labial discs of Cephenniini as compound suckers of premento-hypopharyngeal origin**

The labial discs (ld) of Cephenniini are composed of external (ep) and internal (ip) plates connected by a circumferential

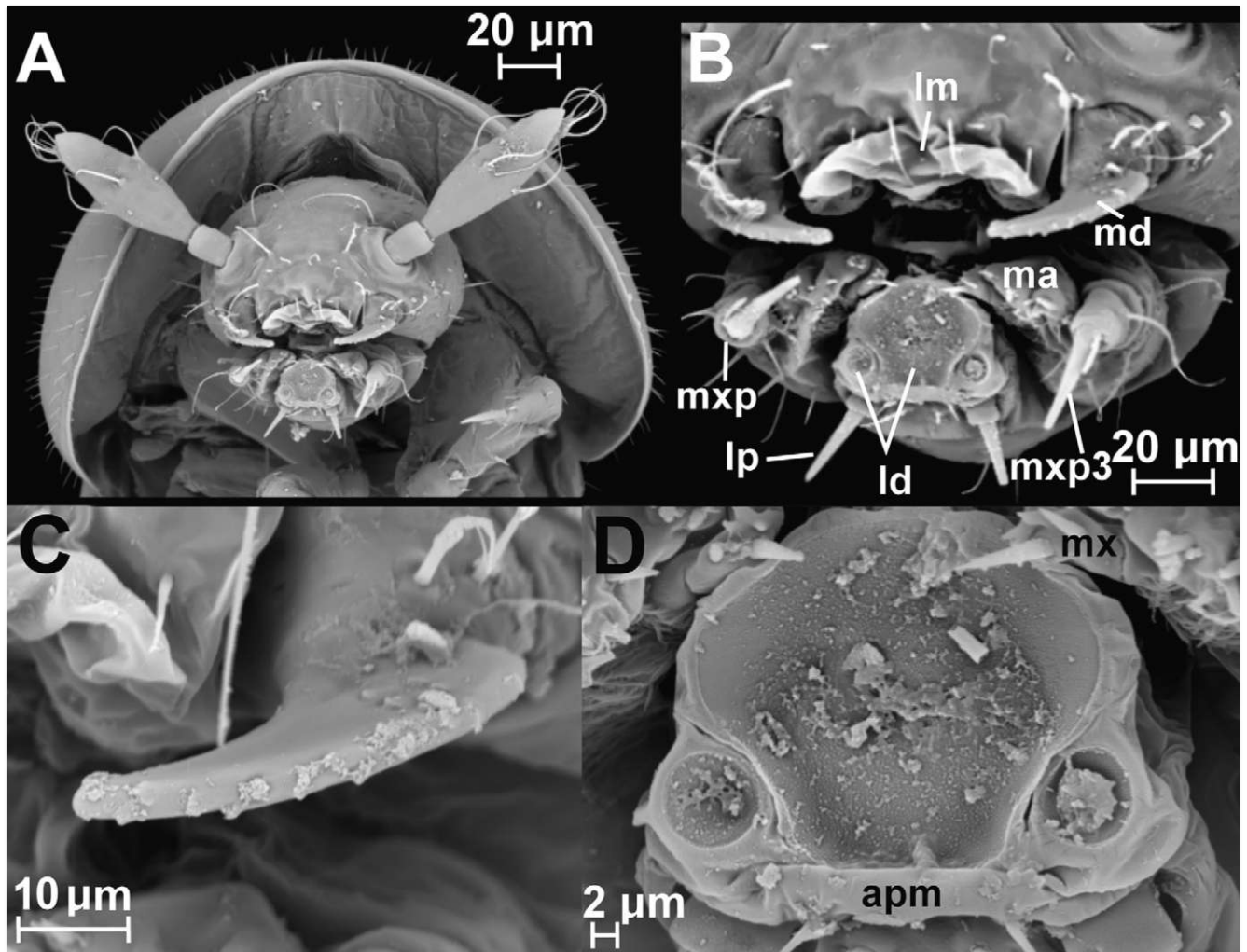
cuticular ring (r) (Figs. 8, 9H–I, 11G–H). The internal architecture of the hollow discs in larvae and adults is similar, suggesting similar evolutionary origins. However, the arrangement of muscles and their insertion sites is not identical, e.g. in larvae *M. 29b* inserts on the internal plate, a hypopharyngeal structure, while in adults on the mento-premental fold (mpmf). Nevertheless, it seems clear that the external plate of the labial disc is apparently an external cuticular structure, while the internal plate is an integral part of the hypopharyngeal suspensorium (hphs).

The internal structure of the labial discs is characteristic of suckers, i.e. organs capable of adhering to smooth surfaces by creating a pressure lower than ambient. The central area of the external plate is flexible and its shape can be changed between flat and concave by contractions and relaxations of longitudinal labial muscles (*M. 28* and *M. 30*), which usually function as retractors of the prementum. This action may be possibly modulated by *M. 29a, b*, also premental retractors, and *M. 34*, primarily responsible for movements of labial palps. A diagram of the suction cycle inferred from the structure of the labium is shown on Fig. 12. In the resting position (Fig. 12A) the labial muscles are relaxed, and the anterior surface of the prementum including the labial discs is strongly concave. Before a mite can be detained, the shape of the labial discs must be changed to less concave or flat, and this can be achieved either by retracting the dorsal or ventral part of the prementum by contraction of *M. 30* (Fig. 12B) or *M. 28*, and possibly also *M. 29b* (Fig. 12C). Because the observed attacks of *Cephennium* on oribatid mites always took place from above (Fig. 2), the retraction of the ventral part of the prementum seems more plausible. The shape of labial discs may be modulated not only by muscles responsible for movements of the labial palps, i.e. *M. 34*, but also by *M. 41*, the elevator of the suspensorium. The beetle has to contract the labial muscles (and ‘load’ the labial discs) only shortly before attacking a mite. When the labium is pressed against the mite’s cuticle, the muscles are relaxed, the labial discs become more concave, thus decreasing pressure between the external plate and the prey, and the mite is detained (Fig. 12D). This mechanism enables Cephenniini to feed on an immobilized oribatid mite for a prolonged period of time without expending energy. An alternative or supplementary mechanism can be hypothesized, in which the shape of labial discs is modulated by changes of the hydrostatic pressure inside the labium, also driven by contractions and relaxations of the labial muscles (changing the internal volume of the labium). The complex structure of the labial discs, with the central opening of the internal plate and the lumen communicating with the internal space of the labium suggest that such a hydrostatic mechanism is not entirely impossible. However, this would require that an increased pressure inside the labium is not dissipated towards the lumen of the head capsule, and morphological structures that could be interpreted as adaptations for closing the internal labial cavity were not found.

Since the typical prey is subglobose in shape, the median longitudinal groove dividing the prementum of adult beetles (Fig. 4J, Fig. 5A–F) and the lateral position of the suckers likely guarantee that the premental surface fits tightly to the mite cuticle. It can be hypothesized that the unusually recurved *M. 34* (Fig. 7A–B), which is inserted on the ventral base of the palpo-mere I and originates on the posterior part of the strongly curved internal sclerite of the palpiger, can modulate this fit by antero-lateral movements of the premental halves. In the studied larva, *M. 34* is particularly robust and unusually shaped (Fig. 11E–F), and may have a similar function. However, in this case the prementum is not divided into lateral halves. It is conceivable that the adjustment is achieved by bending the lateral premental parts bearing the small lateral suckers (Fig. 10B–D).



**Fig. 9.** Male of *Cephennodes* sp. (A–I) Sagittal sections of labium; (J) chart of sections. Abbreviations: apm, anterior premental margin; ep, external plate of labial disc; hphs, hypopharyngeal suspensorium; hpmf, hypopharyngeal-premental fold; ip, internal plate of labial disc; ld, labial disc; lu, lumen of labial sucker; *M. 29a*, subcomponent *a* of *M. tentoriopraementalis inferior*; *M. 29b*, subcomponent *b* of *M. tentoriopraementalis inferior*; *M. 30*, *M. tentoriopraementalis superior*; mpmf, mento-premental fold; ppg, process of palpiger; r, circumferential ring of labial sucker.



**Fig. 10.** Mouthparts of a mature larva of *Cephennium gallicum*. (A) Head in frontal view; (B) mouthparts in frontal view; (C) mandible in dorsofrontal view; (D) prementum in frontal view. Abbreviations: apm, anterior premental margin; ld, labial disc; lm, labrum; lp, labial palpus; ma, maxillary mala; md, mandible; mx, maxilla; mxp, maxillary palpus; mxp3, maxillary palpomere III.

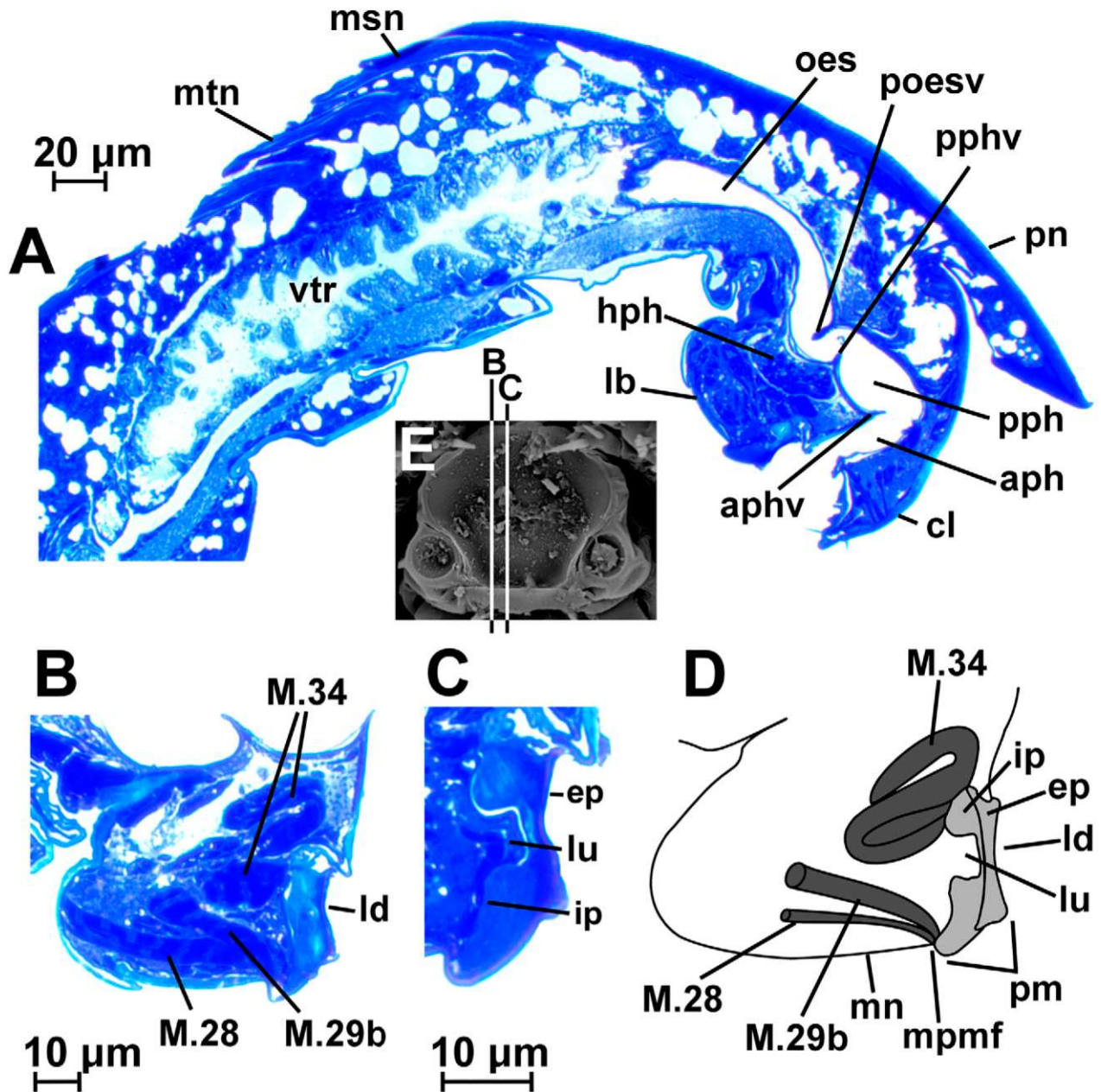
#### 4.2. The feeding mechanism of *Cephenniini*

The capture and immobilization of prey by means of labial suckers and drilling a small hole in the cuticle require a specialized mechanism of food ingestion. In adult *Cephenniini* the main components of the mouthparts are heavily sclerotized and rigid, but the membranous epipharynx (eph), and especially its anterior and lateral margins, and the flexible prementum (pnm) may form a sealing device tightly surrounding the hole drilled in the mite integument. Membranous margins of the epipharynx projecting laterally and anteriorly and visible in dorsal view as a transparent “velum” were previously found in various genera of *Cephenniini* and suggested to be a part of such a sealing mechanism (e.g., Jajoszyński, 2009). The mandibles (md) are not only relatively short but also curved ventrally and in a fully extended position do not interfere with the tight connection between the mouth and the damage site. In the studied larva (Fig. 10B) the sealing structures are more evident. When the short mandibles are fully extended, the membranous labrum (lm), a pair of broad and short malae with only a few trichia and the large, flexible prementum form a ring surrounding the hole drilled in the mite cuticle. Digestive fluids can be injected directly into the prey and the liquefied tissues are pumped back into the pharynx.

Specific morphological adaptations of the foregut for pumping liquefied food were not found in adult *Cephenniini*, whereas the studied larva of *Cephennium* shows an unusual array of pharyngeal valves that is likely part of a pumping mechanism (Fig. 11A). Adult *Cephennium* and *Cephennodes* lack the valves (not shown). The number and placement of these structures suggest a specific functional mechanism. When the pharyngo-oesophageal (poesv) and posteroventral pharyngeal (pphv) valves are in contact thus closing the pharynx posteriorly, and the anteroventral pharyngeal valve (aphv) is open at the same time, the voluminous posterior pharynx (pph) can be filled with liquid food. By closing the anteroventral pharyngeal valve and opening the posterior set of valves, the larva can release the content of the posterior pharynx into the oesophagus (oes). The very large ventriculus (vtr) with the lumen greatly enlarged by multiple folds ensures an efficient absorption of nutrients from the large volume of liquid food ingested by the predator during several hours of continuous feeding.

#### 4.3. Evolution of feeding specialization in *Cephenniini* and co-evolution with prey mites

The phylogeny of Scydmaeninae is presently only partly reconstructed, but at the current state of knowledge it appears

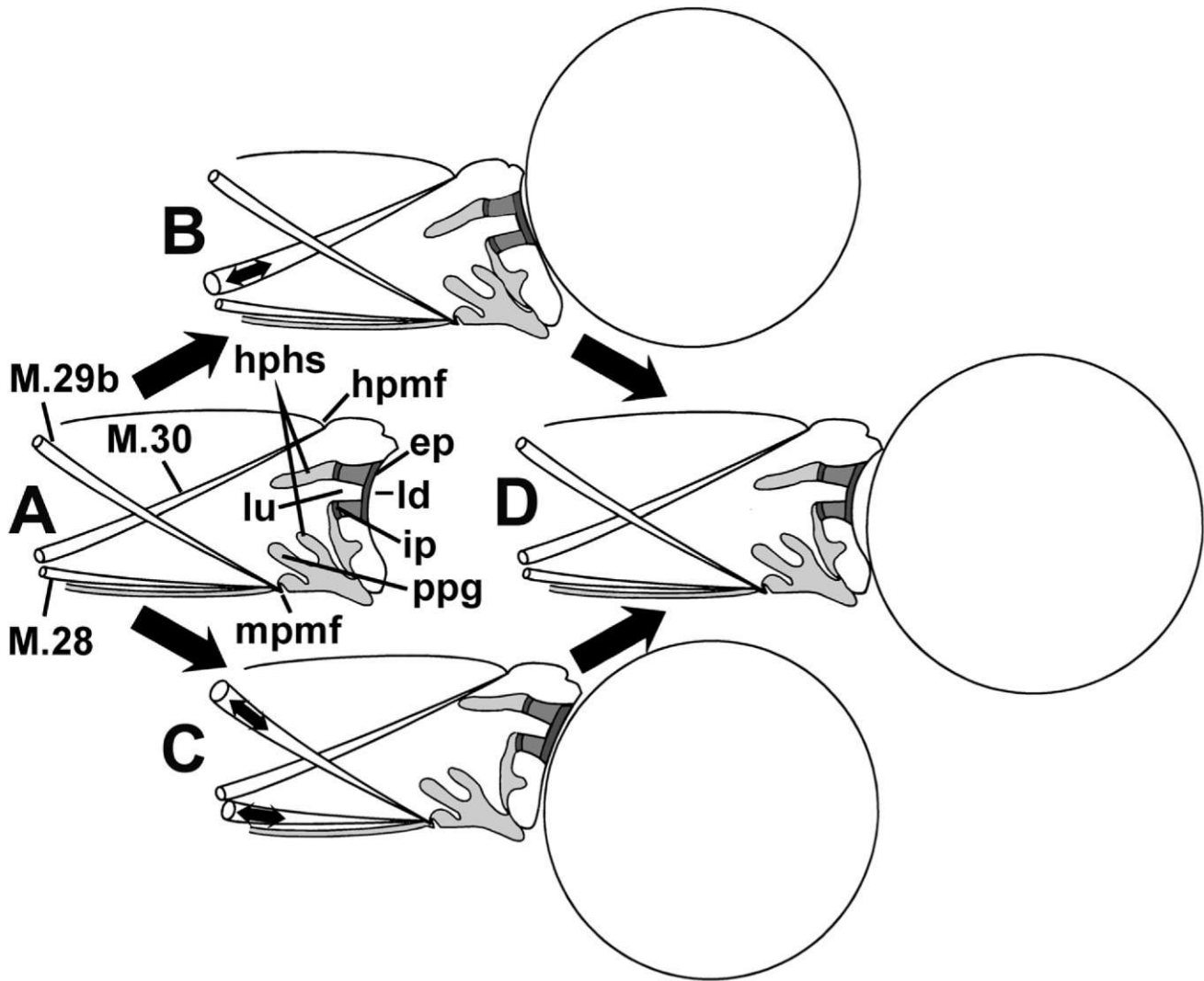


**Fig. 11.** Mature larva of *Cephennium gallicum*. (A) submedian sagittal section of head, thorax and anterior part of abdomen; (B, C) sagittal sections of labium (B) and central labial disc (C); (D) schematic structure of central labial disc in submedian sagittal section; (E) chart of sections. Abbreviations: aph, anterior pharynx; aphv, anteroventral pharyngeal valve; blp, base of labial palp; cl, clypeus; ep, external plate of labial disc; hph, hypopharynx; ip, internal plate of labial disc; lb, labium; ld, labial disc; lu, lumen of labial sucker; M. 28, *M. submentopraementalis*; M. 29b, subcomponent b of *M. tentoriopraementalis inferior*; M. 34, *M. praementopalpalis externus*; mn, mentum; msn, mesonotum; mtn, metanotum; oes, oesophagus; poesv, pharyngeo-oesophageal valve; pm, prementum; pn, pronotum; pph, posterior pharynx; pphv, posteroventral pharyngeal valve; vtr, ventriculus.

that Cephenniini belongs to the most specialized tribes (Grebennikov and Newton, 2009; Jajoszyński, 2012a). Therefore, the modified prementum with labial suckers and broadly separated bases of very small labial palps is apomorphic, whereas the non-modified prementum without suckers and with approximate bases of large labial palps is a plesiomorphy. Similarly, the strategy of other Scydmaeninae grasping mites with the mandibles and front legs and inserting one mandible into the mouth, anal or genital opening, seems to be an adaptation which has evolved early in the subfamily. The published data (Schuster, 1966; Schmid, 1988; Molleman and Walter, 2001), combined with yet unpublished experiments and observations of the first author, suggest that at least some species of these genera using this technique are either opportunistic predators or scavengers, while Cephenniini

are strictly specialized on armored mites. It is likely that this specialization developed from unspecialized feeding on various soil organisms, including soft-bodied or only partly armored mites, possibly belonging to the ancestors or stemlineage of Oribatida. The unique morphological adaptations of Cephenniini restrict this tribe to one specific type of prey and exclude other organisms from their diet.

The fossil record of Cephenniini is exceptionally scarce. The tribe is only known from the relatively young Eocene amber (Jajoszyński, unpublished). The genera *Ektatotrichia* Chatzimanolis, Engel & Newton and *Electroatopos* Chatzimanolis, Engel & Newton described from mid-Cretaceous amber resemble some Cephenniini, but their true affinities remain unknown and details of their labia were not studied. Therefore, the morphology of the known extinct



**Fig. 12.** Schematic sagittal section of the labium of adult *Cephennodes* showing the suction cycle of the labial discs (mite represented by large circle). (A) Resting position – longitudinal labial muscles (*M. 28*, *M. 29b* and *M. 30*) relaxed and labial discs strongly concave; (B, C) labium prepared for suction – longitudinal muscles inserting dorsally (*M. 30*; B) or ventrally (*M. 28* and *M. 29b*); (C) contracted and labial discs slightly concave or flat; (D) mite detained – longitudinal muscles relaxed, labial discs strongly concave. Abbreviations: ep, external plate of labial disc; hphs, hypopharyngeal suspensorium; hpmf, hypopharyngeal–premental fold; ip, internal plate of labial disc; ld, labial disc; lu, lumen of labial sucker; *M. 28*, *M. submentopraementalis inferior*; *M. 29b*, subcomponent *b* of *M. tentoriopraementalis inferior*; *M. 30*, *M. tentoriopraementalis superior*; mpmf, mento–premental fold; ppg, process of palpiiger.

Scydmaeninae cannot be used to reconstruct the evolution of the labial discs. However, the continuity of the internal plates of the discs with the hypopharyngeal suspensorium may suggest a possible evolutionary mechanism. The non-modified prementum in Scydmaeninae is usually relatively weakly sclerotized compared to the mentum. If precursors of extant Cephenniini fed on soft-bodied mites, certain areas of the premental cuticle were likely pressed against the internal sclerotization during feeding. The position of such contact sites might have been constant and determined by the shape of the suspensorium. Therefore, the prey integument was pressed onto the same (paired) areas of the prementum. A hardening of the premental cuticle at the contact sites might have been favored by selection, especially if combined with excreting a sticky secretion from labial glands to stabilize the captured prey. The next step was a gradual acquisition of a sclerotized connection between the sticky cuticular pads or plates with the hypopharyngeal suspensorium. By this process, constant, pre-specialized and paired contact areas between the prementum and the prey were developed; the prementum was gradually enlarged,

and the labial palps and mandibles shortened or curved downwards to expose the capturing device. The labial muscles, originally only involved in movements of the prementum and its parts, already generated changes in the shape of premental surface. Therefore, a transition from sticky plates to true suckers might have been simple, requiring only the development of a thinner central area on the disc to transform the passive sticky plate into an active sucker.

The structural data seem support this hypothesis. However, it appears unlikely that the labial suckers have developed during a process of co-evolution between “proto-Cephenniini” and “proto-Oribatida”. The earliest Oribatida fossils are dated to the Middle Devonian, but molecular clock estimates even place this group of Acari among the earliest arthropods that have colonized terrestrial habitats, with the earliest radiation of major lineages in aquatic environments of the Cambrian (Schaefer et al., 2010). In very clear contrast to this, the oldest Scydmaeninae are known from mid-Cretaceous amber (Poinar and Brown, 2004; Chatzimanolis et al., 2010), and the Staphylinine group of Staphylinidae (to which

Scydmaeninae belong) is believed to have originated in the Jurassic (Clarke and Chatzimanolis, 2009). Therefore, it is highly unlikely that “proto-Cephenniini” have co-evolved with “proto-Oribatida”, which are very likely much older. Cephenniini might have i) co-evolved with a particular, relatively young group of “proto-Oribatida” that has acquired the hard armor relatively recently (in relation to the postulated very old history of Oribatida), or ii) ancestors of Cephenniini gradually shifted from feeding on other types of prey towards fully armored Oribatida, or iii) the labial discs have originally developed for functions not related to feeding. The hypotheses ii) and iii) can be combined in one, if the labial discs have evolved for other purposes than feeding and only later were adopted to capture fully armored oribatid mites. With the extremely poor fossil record of Scydmaeninae it is not possible to support any of these hypotheses. However, the mechanism of developing the compound and complicated labial suckers in response to the contact of the prementum with prey, proposed in the previous paragraph, seems plausible. Detailed morphological study of possible fossil Cephenniini in the oldest ambers from Lebanon and Myanmar may help to clarify this problem. Furthermore, the small and indistinctly defined labial discs found in the genera of the *Cephennomicrus* group (*Cephennomicrus*, *Cephennula*, *Trurlia*) may represent an intermediary state between an unmodified prementum and one equipped with large, strongly sclerotized and sharply demarcated suckers as they are present in *Cephennium* or *Cephennodes*. Observations of live beetles of the *Cephennomicrus* group during feeding and studying their prey preferences may help to reconstruct the evolution of such an unusually modified labium.

### Acknowledgements

We thank Alfred Newton and Margaret Thayer (Field Museum of Natural History, Chicago, USA) and Giulio Cuccodoro (Muséum d'histoire naturelle de la Ville de Genève, Switzerland) for their help in locating well-preserved larval material used in our study. The SEM images were taken by Krzysztof Kalmski (Wrocław University of Environmental and Life Sciences, Poland) and Zbigniew Adamski (Electron and Confocal Microscope Laboratory, Faculty of Biology, Adam Mickiewicz University, Poznań).

### References

- Chatzimanolis, S., Engel, M.S., Newton, A.F., Grimaldi, D.A., 2010. New ant-like stone beetles in mid-Cretaceous amber from Myanmar (Coleoptera: Staphylinidae: Scydmaeninae). *Cretaceous Research* 31, 77–84.
- Clarke, D.J., Chatzimanolis, S., 2009. Antiquity and long-term morphological stasis in a group of rove beetles (Coleoptera: Staphylinidae): description of the oldest *Octavius* species from Cretaceous Burmese amber and a review of the “Euaesthetine subgroup” fossil record. *Cretaceous Research* 30, 1426–1434.
- Grebennikov, V.V., Newton, A.F., 2009. Good-bye Scydmaenidae, or why the ant-like stone beetles should become megadiverse Staphylinidae sensu latissimo (Coleoptera). *European Journal of Entomology* 106, 275–301.
- Jatoszyński, P., 2003. *Chelonoidum araiorum* sp. nov., an unusual species of the Cephenniini (Coleoptera, Scydmaenidae) from the intertidal zone of the Pacific coast of Central Honshu, Japan. *Bulletin of the National Science Museum, Tokyo* 29, 225–228.
- Jatoszyński, P., 2005. Revision of the oriental genus *Loebrites* Franz (Coleoptera, Scydmaenidae). *Revue suisse de Zoologie* 112, 351–369.
- Jatoszyński, P., 2009. *Cephennomicrus* Reitter (Coleoptera, Staphylinidae, Scydmaeninae) of Japan and Taiwan: taxonomic notes, ten new species and comparative morphology of *nomurai* and *taiwanensis* species groups. *Zootaxa* 2145, 1–35.
- Jatoszyński, P., 2011a. *Hlavaciellus* Jatoszyński, 2006: eleven new species, detailed morphology and systematic position within Cephenniini (Coleoptera, Staphylinidae, Scydmaeninae). *Zootaxa* 2763, 1–33.
- Jatoszyński, P., 2011b. Cephenniini with prothoracic glands and internal ‘cavities’: new taxa, enigmatic characters and phylogeny of the *Cephennomicrus* group of genera (Coleoptera, Staphylinidae, Scydmaeninae). *Systematic Entomology* 36, 470–496.
- Jatoszyński, P., 2012a. Beetles with ‘trochantelli’: phylogeny of Cephenniini (Coleoptera: Staphylinidae: Scydmaeninae) focusing on Neotropical genera. *Systematic Entomology* 37, 448–477.
- Jatoszyński, P., 2012b. Adults of European ant-like stone beetles (Coleoptera: Staphylinidae: Scydmaeninae) *Scydmaenus tarsatus* Müller & Kunze and *S. hellwigii* (Herbst) prey on soft-bodied arthropods. *Entomological Science* 15, 35–41.
- Jatoszyński, P., 2012c. Observations on cannibalism and feeding on dead arthropods in *Scydmaenus tarsatus* Müller & Kunze. *Genus* 23, 25–31.
- Molleman, F., Walter, D.E., 2001. Niche segregation and can-openers: scydmaenid beetles as predators of armoured mites in Australia. In: Halliday, R.B., Walter, D.E., Proctor, H.C., Norton, R.A., Colloff, M.J. (Eds.), *Acarology: Proceedings of the 10th International Congress*. CSIRO Publishing, Melbourne, pp. 283–288.
- Newton, A.F., 1991. Scydmaenidae (Staphylinodea). In: Stehr, F.W. (Ed.), *An Introduction to Immature Insects of North America*, vol. 2. Kendall/Hunt Publishing Co, Dubuque, Iowa.
- Newton, A.F., Franz, H., 1998. World catalog of the genera of Scydmaenidae (Coleoptera). *Koleopterologische Rundschau* 68, 137–165.
- O’Keefe, S.T., 2005. Scydmaenidae Leach, 1815. In: Beutel, R.G., Leschen, R.A.B. (Eds.), *Coleoptera, Beetles. Morphology and Systematics (Archostemata, Adephaga, Myxophaga, Polyphaga Partim)*. Handbook of Zoology, vol. 1. De Gruyter, Berlin, New York.
- Poinar Jr., G., Brown, A.E., 2004. A new subfamily of Cretaceous antlike stone beetles (Coleoptera: Scydmaenidae: Hapsomelinae) with an extra leg segment. *Proceedings of the Entomological Society of Washington* 106, 789–796.
- Schaefer, I., Norton, R.A., Scheu, S., Maraun, M., 2010. Arthropod colonization of land – linking molecular and fossil oribatid mites (Acari, Oribatida). *Molecular Phylogenetics and Evolution* 57, 113–121.
- Schmid, R., 1988. Morphologische Anpassungen in einem Räuber-Beute-System: Ameisenkäfer (Scydmaenidae, Staphylinodea) und gepanzerte Milben (Acari). *Zoologische Jahrbücher, Abteilung für Systematik, Ökologie und Geographie der Tiere* 115, 207–228.
- Schuster, R., 1966. Scydmaeniden-Larven als Milbenräuber. *Naturwissenschaften* 53, 439–440.