Mitt. Mus. Nat.kd. Berl., Dtsch. entomol. Z. **50** (2003) 1, 143–150 30. 06. 2003

Description of larval instars of *Neodryinus typhlocybae* (Ashmead, 1893) (Hymenoptera Dryinidae), with remarks on its biology

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With 10 figures

Abstract

Larval instars III (immature) and IV (mature) of *Neodryinus typhlocybae* (Ashmead) are described for the first time. Besides, some data on other aspects of the postembryonic development are provided. The affinities among the immature and mature larvae of *N. typhlocybae* and corresponding instars of other species of the same subfamily are discussed.

Key words: Chrysidoidea, Gonatopodinae, Homoptera, Fulgoromorpha, development, morphology, parasitism.

Introduction

Dryinidae is a family of Hymenoptera Aculeata including ten subfamilies. As parasitoids of Homoptera Auchenorrhyncha, they are effective for the biological control of plant pests.

Unfortunately the knowledge on the biology and particularly on the postembryonic development of this interesting group of insects is scarce (Olmi 1994). Hypermetamorphosis occurs with two types of larvae differing in morphology, physiology and ethology: larvae of the first 3–4 instars are called 'immature', larva of the last instar 'mature'.

The few, and often uncomplete, available descriptions of both types of larvae are based on one species of the subfamily Aphelopinae (*Aphelopus indivisus* Kieffer), two species of Dryininae (*Dryinus poecilopterae* (Richards), *D. stantoni* Ashmead) and seven species of Gonatopodinae (*G. peculiaris* Brues, *G. americae* Olmi, *G. caraibicus* (Olmi), *G. lunatus* Klug, *G. chilensis* (Olmi), *Haplogonatopus hernandezae* Olmi, *H. oratorius* (Westwood)).

Among the Dryinidae, Gonatopodinae is one of the subfamilies most rich in species (470), distributed with 12 genera in all biogeographic regions. In the palaearctic region the genera *Echthrodelphax*, *Gonatopus*, *Neodryinus* and *Haplogonatopus* are recorded.

The genus *Neodryinus* includes 37 species worldwide. Two of them, *Neodryinus somniatus* Brues, 1933, a fossil species found in baltic amber, and *N. typhlocybae* (Ashmead, 1893), imported in order to biological control, are recorded in the palaearctic region.

N. typhlocybae, a nearctic species, is a parasitoid of Fulgoromorpha species belonging to the family Flatidae: *Ormenis septentrionalis* (Spinola), *Anormenis chloris* (Melichar), *Metcalfa pruinosa* (Say), *M. regularis* (Fowler) (Guglielmino & Olmi 1997). This species has been recently introduced to Italy (Girolami & Camporese 1994), France (Malausa 1999), Slovenia (Sala & Foschi 2000) and Switzerland (Jermini, Brunetti, Bonavia 2000) in the course of programmes adressed to the biological control of *Metcalfa pruinosa* (Say), a nearctic Flatid recorded in Europe since the late 1970ies.

Some aspects of the biology of *N. typhlocybae* have been described, but the larval instars are not known for any species of this genus. In this contribution we describe the larval instars III (immature) and IV (mature) of this interesting parasitoid. Besides, some data on other aspects of the postembryonic development are provided.

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Received January 2002, accepted November 2002

 1435-1951/03/0106-0143 $ 17.50+.50/0

Material and Methods

N. typhlocybae larvae of both types were obtained from parasitized nymphs of *Metcalfa pruinosa*.

Measurements and drawings were made on specimens preserved in 70% alcohol. To test the permeability of the cuticle of the cephalic vesicles, live immature larvae were embedded in methylene blue; to deduce the number of immature instars from the number of exuviae, some specimens were treated for 15 minutes with 10% KOH.

For the investigation of the morphology of head, thorax and abdomen, mature larvae were cleared for about 30 minutes in 10% KOH, slide-mounted in Faure liquid and observed with a Leitz Laborlux 12.

Results

Females of *N. typhlocybae* insert the eggs into the intersegmental membrane below the mesothorax wingpads of *Metcalfa pruinosa* nymphs. *N. typhlocybae* has four larval instars. As the first three ('immature') instars differ mainly in the number of their exuviae and in their size, the following description of immature larvae concentrates to the instar III.

Larval instar III (Figs 1–3)

Length: 1.44 –2.20 mm; Width: 0.92–1.24 mm.

White-yellowish in colour, protruding from the host body below the mesothorax wingpad (Fig. 1). As the other Gonatopodinae immature larvae, with two kidney-shaped vesicles in the cephalic region; posterior region of the body bent ventrally. Segmentation of the body indistinct, respiratory system peripneustic with nine pairs of lateral spiracles (one thoracic + eight abdominal) (Fig. 2).

Body partially covered by the discarded exuviae of the two previous larval instars (Fig. 2), immersed into the host's haemocoel with its anterior part, the posterior part protruding outside.

The position of the exuviae shows that for each moult, the cuticle breaks along a preformed mediodorsal line.

Cephalic region covered by a sclerotized case and therefore with the mouth closed (Fig. 3), with the cephalic vesicles as the only structures with permeable cuticle as evidenced by methylene blue coloration.

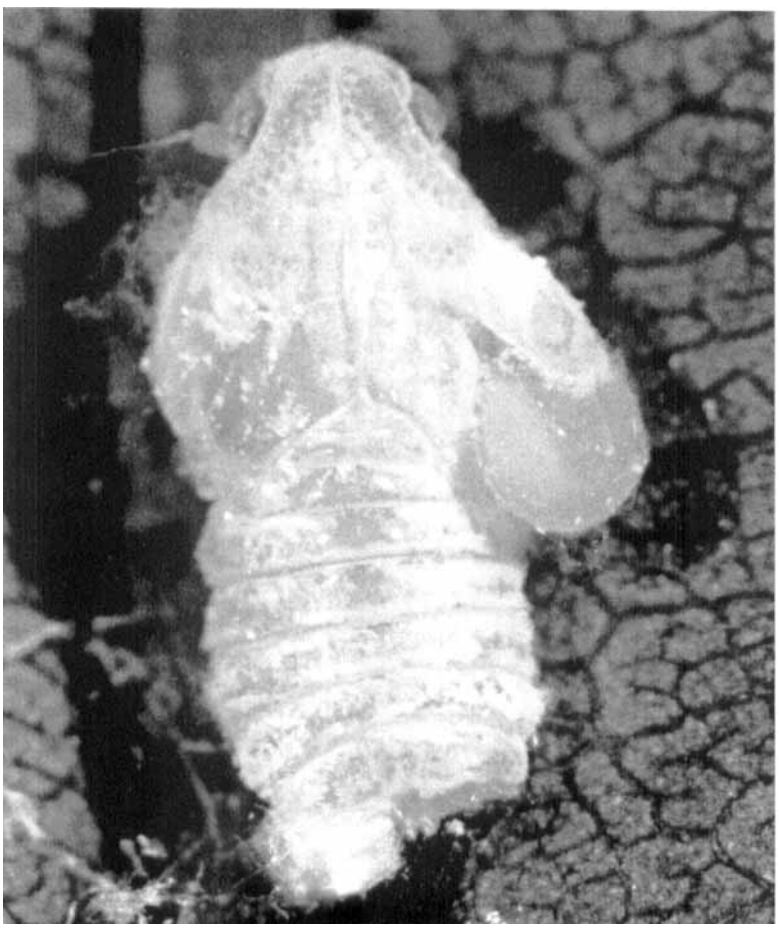

Fig. 1: Nymph of *Metcalfa pruinosa* (Say) parasitized by an immature larva of *Neodryinus typhlocybae* (Ashmead).

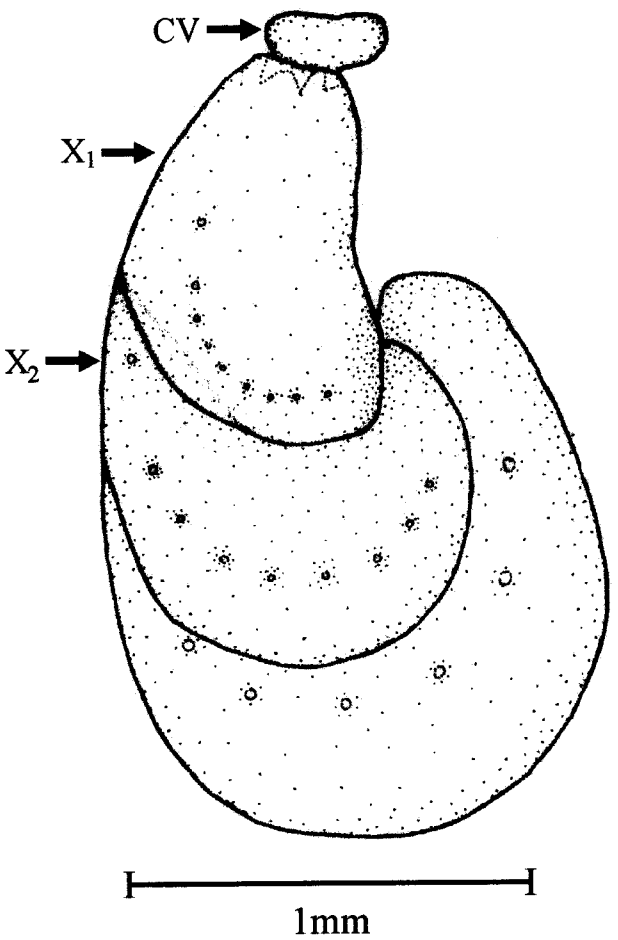

Fig. 2: *Neodryinus typhlocybae* (Ashmead), immature larva of larval instar III, lateral view. – CV: Cephalic vescicle; X_1: Exuvia of larval instar I; X_2: Exuvia of larval instar II

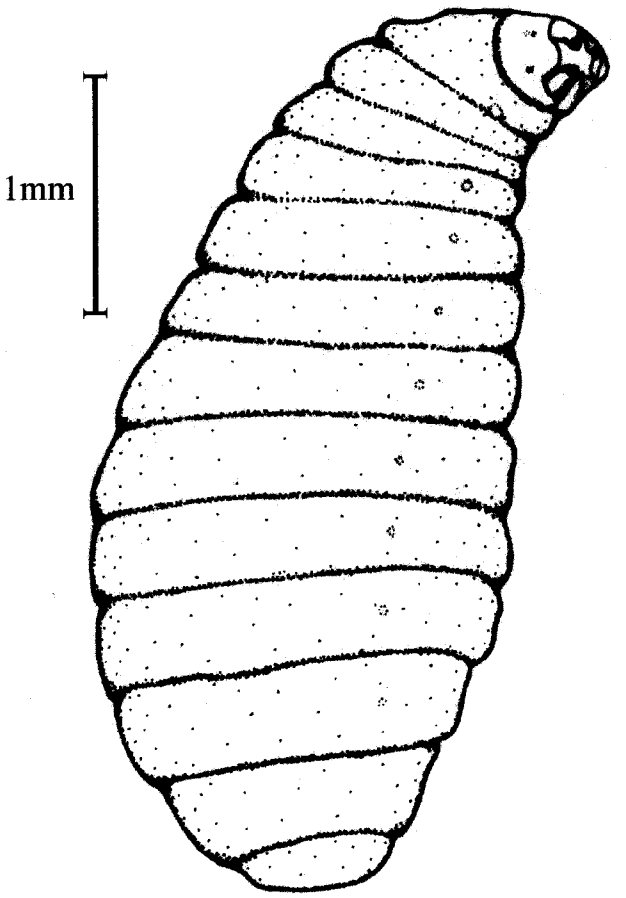

Fig. 4: *Neodryinus typhlocybae* (Ashmead), mature larva, dorsolateral view.

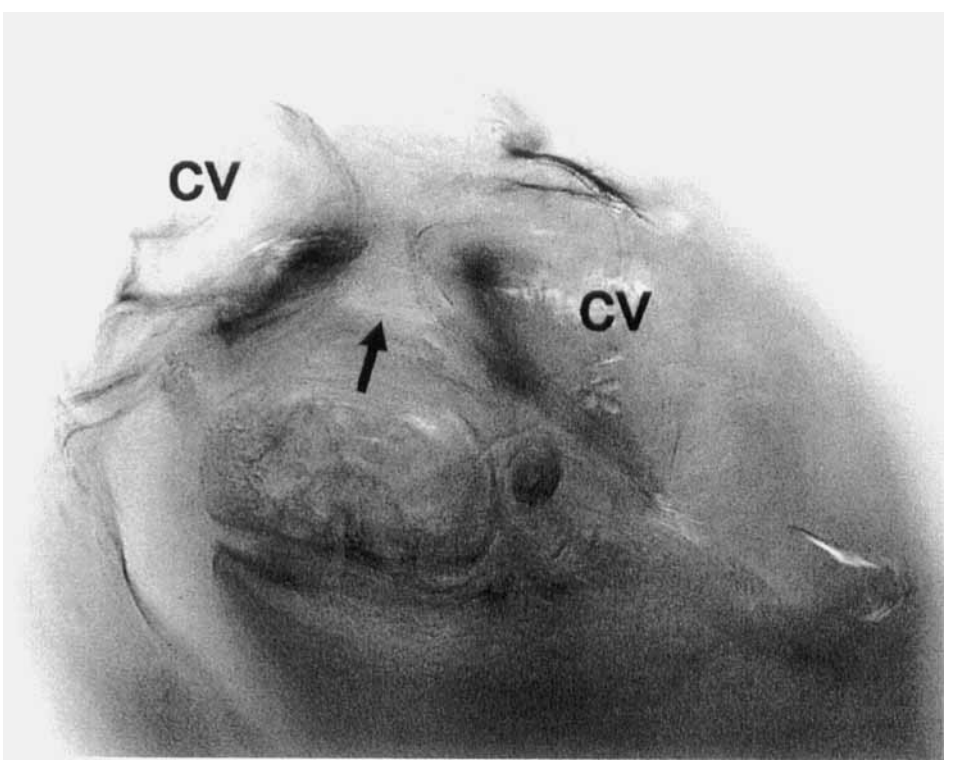

Fig. 3: *Neodryinus typhlocybae* (Ashmead), cephalic cuticular case of immature larva – CV: Cephalic vesicle; arrow: oral region closed by cuticle, without recognizable mouth opening.

As supposed by Carcupino et al. (1998) and Guglielmino (2002) for other species of Gonatopodinae and Dryininae, probably also in the immature larval instars of *N. typhlocybae* the cephalic vesicles absorb the nutrients from the haemolymph of the host and pass them to the haemolymph of the larva.

Ventral process, a structure probably with anchorage function, observed in *Gonatopus contortulus* Patton (Fenton 1918), *G. peculiaris* Brues (Barrett et al. 1965) and *G. caraibicus* (Olmi) (Virla 1992) could not be found.

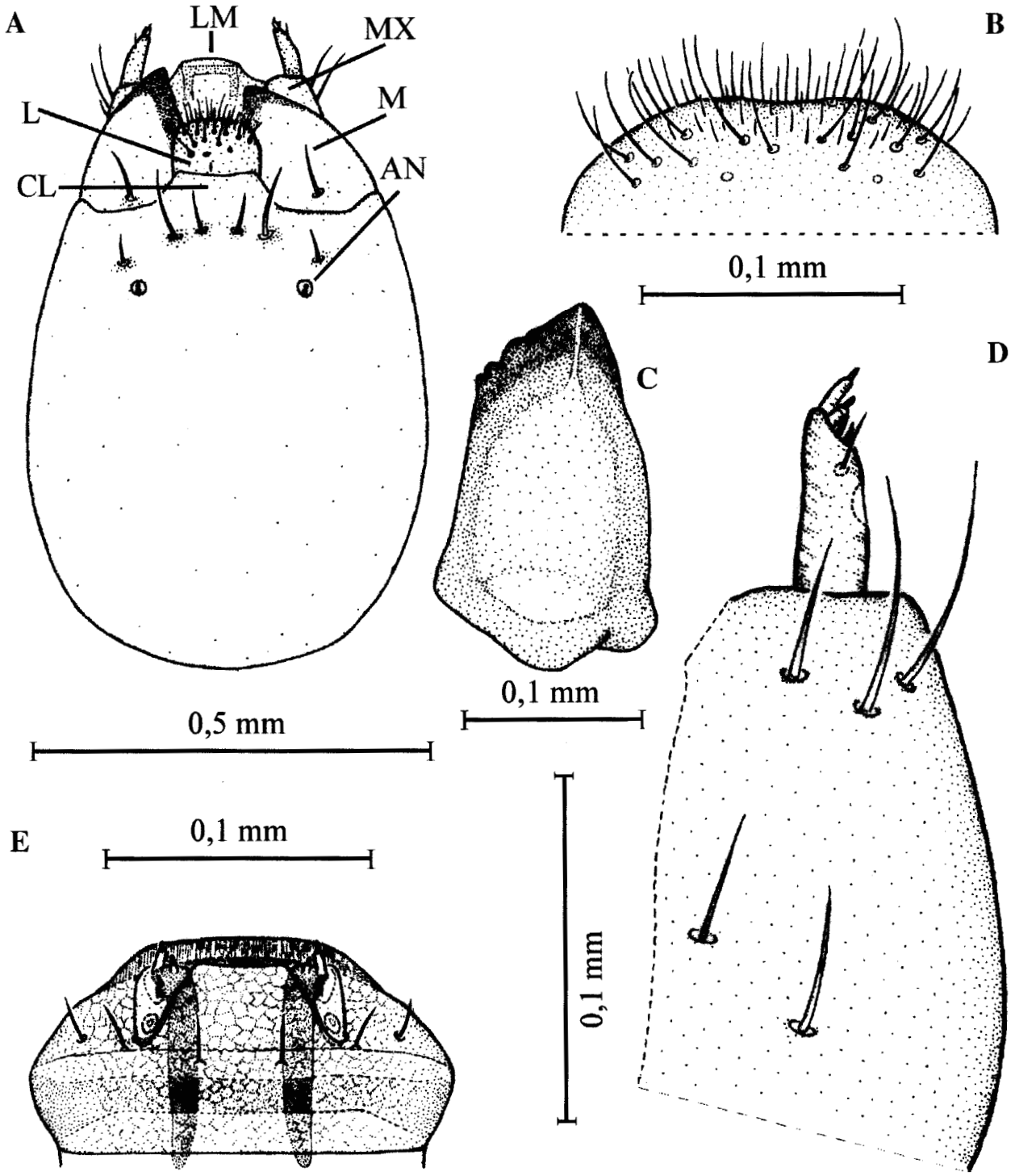

Fig. 5: *Neodryinus typhlocybae* (Ashmead), mature larva. – **A**: head, dorsal view. – **B**: labrum, dorsal view. – **C**: right mandible, dorsal view. – **D**: distal part of left maxilla, ventral view. – **E**: median lobe of labium, ventral view – *AN*: Antenna; *CL*: Clypeus; *L*: Labrum; *M*: Mandible; *MX*: Maxilla; *ML*: Median lobe.

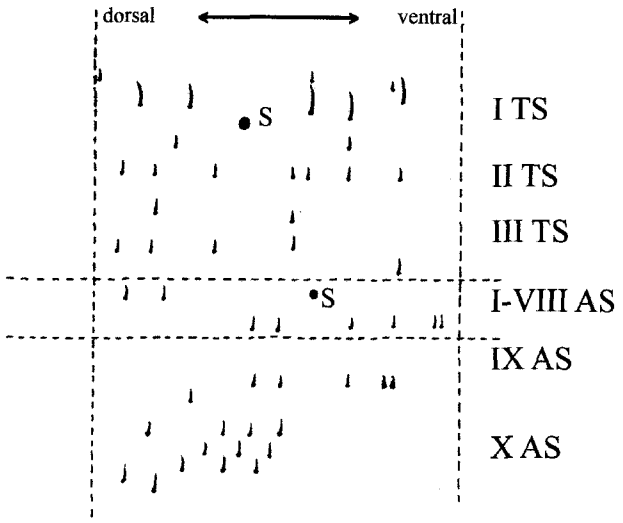

Fig. 6: *Neodryinus typhlocybae* (Ashmead), mature larva, thoracal and abdominal chaetotaxy in schematic view – TS: thoracal segments; AS: abdominal segments; S: spiracles.

Larval instar IV (Figs 4–6)

Length: 2.60–5.60 mm; Width: 1.20–1.64 mm.

Creamy white or pink in colour, with the typical facies of mature Gonatopodinae larvae: head well-developed, three thoracal and ten abdominal segments (Fig. 4).

Head prognathous, longer than wide (Fig. 5A). Antennae very simple in structure, reduced to a pair of circular areas surrounded by a thickened cuticular ring each, and with two small conical sensilla in the middle. Clypeus subtrapezoidal, with concave anterior margin, not well distinct from the frontal region, lacking sense organs. Labrum (Fig. 5B) with convex lateral margins and rounded anterior corners, bearing numerous slender sensory bristles and few sensilla.

Mandibles (Fig. 5C) strongly sclerotized, subrectangular, about 1.5 times longer than their maximum width, bearing a glandular canal opening on their tip, distally slightly narrowed and with three-five small teeth.

Maxillae (Fig. 5D) made up by two weakly sclerified pieces bearing each two slender proximal bristles near the lateral border of the labium, three distal bristles and an apical palp; this palp subcylindric, with one short bristle and a large sensorial pit in latero-ventral position, bearing four papillae on the apex, two of which enlarged and ending with a minute sensillum.

Labium consisting of the postmentum and a median lobe (Fig. 5E) formed by praementum and hypopharynx (Buyckx 1948) and bearing the spinning apparatus opening. Postmentum weakly sclerified and subtrapezoidal in shape. Palpi inserted on the median lobe (Fig. 5E), short and broad, each with three slender bristles near the base, two large sensorial pits, one near the base, the other on the apex and bearing four papillae, two of which enlarged and with a minute apical sensillum.

Thorax and abdomen with indistinct segmentation, covered by minute bristles; Fig. 6 gives a schematic view of chaetotaxy. Respiratory system consisting of one pair of big thoracic spiracles and eight pairs located on the first abdominal segments.

Fig. 7: *Metcalfa pruinosa* (Say), residues of nymph with the hatching opening of the mature larva of *Neodryinus typhlocybae*.

Fig. 8. *Neodryinus typhlocybae* (Ashmead), cocoon.

Behaviour of the mature larva

Just formed, the mature larva begins to feed, breaking the anterior portion of the previous exuviae and entering the host body to devour its organs. Due to the continuous peristaltic movements of the larva, its activity can be observed both directly through the fine exuviae, by the contractions of the posterior part of the body, and indirectly from the deformation of the host's body walls while it is gradually deprived of its internal organs.

Killed the host (Fig. 7), the mature larva begins to build a cocoon on which it attaches also the residues of the dead host (Fig. 8). The cocoon is whitish, oval, with a double wall, in some cases the larva incorporates also tufts of wax torn from the body of the dead host.

The behaviour of the mature larva of *N. typhlocybae* differs from that of all *Gonatopus* species known from this point of view, which leave the body of the host and search for a place suitable to build their cocoon and pupate.

Prepupa and pupa

The mature larva, protected inside the cocoon, after a quiescent phase transforms first into the prepupal, and then into the pupal stage. Like all the other species of Dryinidae, also *N. typhlo-*

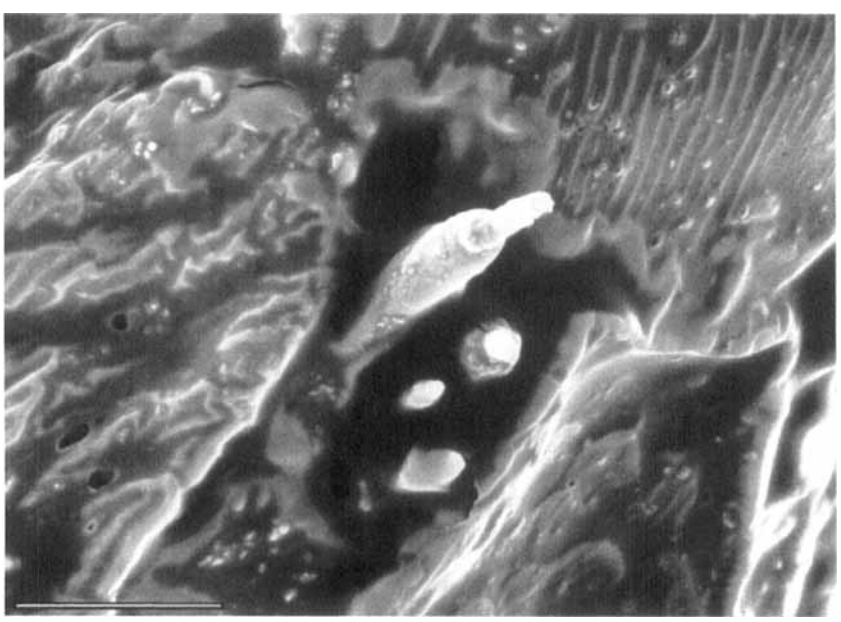

Fig. 9. *Gonatopus lunatus* Klug, mature larva, detail of right labial palp showing number and structure of sensilla. Scale = 10 µm.

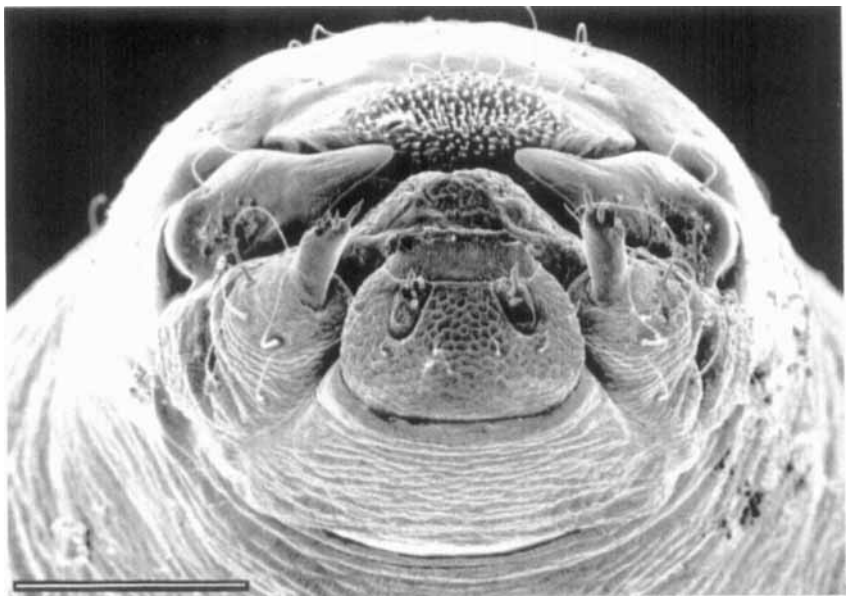

Fig. 10. *Gonatopus lunatus* Klug, mature larva, cephalic region. Scale = 100 µm.

cybae has a pupa exarata. Metamorphosis completed, the adult cuts the anterior wall of the cocoon with its mandibles. At first come out the antennae, which frenetically are moved, and then the imago frees itself, opening a large passage by the mandibles.

Discussion

At present, the few, and often uncomplete, data on the postembryonic development of the Gonatopodinae do not allow to value the taxonomic significance of the larval characters.

From comparison of immature and mature larvae of *N. typhlocybae* with the corresponding instars of other species of the same subfamily, it is possible to deduce some differences.

The immature larva of *N. typhlocybae* differs from:

1) *Gonatopus bicolor* (Haliday) (Heikinheimo 1957, as *Dicondylus lindbergi* Heikinheimo), *G. helleni* (Raatikainen) (Raatikainen 1961, as *Dicondylus helleni* Raatikainen), *G. peculiaris* Brues (Barrett et al. 1965, as *Pachygonatopus minimus* Fenton), *G. americae* Olmi (Giri & Freytag 1986, as *Dicondylus americanus* (Pekins)), *G. lunatus* Klug (Guglielmino & Virla 1998) and *Haplogonatopus hernandezae* Olmi (Hernandez & Belloti 1984) in the colour of the body (white-yellowish in *N. typhlocybae*, greyish-brown in *G. bicolor* e *G. helleni*, brown or black in *G. peculiaris*, *G. americae*, *G. lunatus*, *H. hernandezae*);
2) *G. caraibicus* (Olmi) (Virla 1992, as *Tetrodontochelys peculiaris* (Brues)), *G. contortulus* Patton (Fenton 1918) and *G. peculiaris* Brues in the absence of the ventral process;
3) *G. chilensis* (Olmi) (Virla & Mangione 2000) in the mouth closed by cuticle (open in *G. chilensis*), as well as in the way of breaking of the cuticle during each moult (mediodorsal fracture line in *G. chilensis* extending to the anterior and ventral region, Figs 4–5 in Virla & Mangione 2000);
4) *H. oratorius* (Westwood) (Kitamura 1985, as *Haplogonatopus atratus* Esaki & Hashimoto), *G. lunatus* Klug, *G. caraibicus* (Olmi), *G. chilensis* (Olmi) and *G. peculiaris* Brues in the number of immature larval instars (3 in *N. typhlocybae*, 4 in the other species).

The mature larva of *N. typhlocybae* differs from:

1) *G. chilensis* (Olmi), *G. lunatus* Klug, *H. hernandezae* Olmi in the subrectangular mandibles (triangular or subtriangular in the other species).
2) *G. chilensis* (Olmi) in the presence of four sensilla on the labial palpi (only two in *G. chilensis*).

With this regard, *N. typhlocybae* agrees with *G. lunatus*. In the description of that species, Guglielmino e Virla (1998) erroneously indicated the presence of two papillae instead of four on each labial palp (Fig. 9).

However, beyond these differences, it is possible to observe the extreme similarity of some structures in species belonging to different genera. For example, the shape and structure of the maxillae, labium and corresponding palpi, as well as the number and position of the bristles located on these appendages in the mature larva

of *N. typhlocybae* and *G. lunatus* (Figs 5D–E, 9, 10). Therefore, it is possible to presume that these characters at lower taxonomic level are of little significance for systematics.

Acknowledgements

We thank Dr. S. Foschi (Bioplanet, Cesena, Italy) for providing parasitized nymphs of *Metcalfa pruinosa*, Mr. M. Vollaro (University of Tuscia, Viterbo) for the production of macrophotographs, and Mr. H. Schoppmann (University of Tübingen) for technical assistance on the SEM. We thank also Prof. M. Olmi (University of Tuscia, Viterbo), Prof. G. Pilato (Univesity of Catania) and Dr. R. Gerecke (Tübingen) for their valuable suggestions.

This study was supported by a grant of the University of Tuscia, Viterbo (ex 60%).

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