

## Morphology and Ontogeny of Immature Stages of Helotidae Based on Descriptions of *Helota thoracica* Ritsema and *H. gemmata* Gorham (Insecta: Coleoptera: Cucujoidea)

Chi-Feng Lee<sup>1,\*</sup>, Sadatomo Hisamatsu<sup>2</sup>, and Ping-Shih Yang<sup>3</sup>

<sup>1</sup>Applied Zoology Division, Taiwan Agricultural Research Institute, 189 Chung-Cheng Road, Taichung 413, Taiwan

<sup>2</sup>Entomological Laboratory, Faculty of Agriculture, Ehime University 5-7, Tarumi 3-chhōme, Matsuyama, 790-8566 Japan

E-mail:sthisamatsu@yahoo.co.jp

<sup>3</sup>Department of Entomology, National Taiwan University, Taipei 106, Taiwan. E-mail:psyang@ntu.edu.tw

(Accepted June 22, 2007)

**Chi-Feng Lee, Sadatomo Hisamatsu, and Ping-Shih Yang (2007)** Morphology and ontogeny of immature stages of Helotidae based on descriptions of *Helota thoracica* Ritsema and *H. gemmata* Gorham (Insecta: Coleoptera: Cucujoidea). *Zoological Studies* 46(6): 760-769. Three larval instars and the pupae of *Helota thoracica* Ritsema, 1895 are described and figured for the first time. The larval chaetotaxy of the head, pro- and mesonota, abdominal terga III, legs, abdominal sterna III, and urogomphi is described. Almost all setae present in larval instar I are primary. Subprimary setae appear on the abdominal sternites, head, and legs. Pupae are very conservative since no differences were observed between *H. thoracica* and *H. gemmata*. A novel method for rearing adult and larval helotids is also provided. <http://zoolstud.sinica.edu.tw/Journals/46.6/760.pdf>

**Key words:** Taxonomy, Larval chaetotaxy.

The Helotidae is a small cucujoid family containing 106 species. Their biology and immature stages are poorly known. Adults and larvae of *Helota gemmata* Gorham feed on sap exuding from trees (Olliff 1883, Fukuda 1943). Larvae and pupae of this species were field-collected and described (Fukuda 1943 1944). However, these general descriptions did not address either the chaetotaxy or 1st-instar larvae. In the present paper, a new approach for rearing adult and larval helotids is provided to facilitate studies of larval chaetotaxy. All larval instars and pupae of *H. thoracica* Ritsema were studied. In addition, they are compared with those of *H. gemmata* to determine interspecific differences and phylogenetically informative characters of the family.

Immatures provide a source of characters to supplement those of adults. Larval chaetotaxy may provide a number of important character sets

for phylogenetic analyses (e.g., Alarie and Balke 1999). The larval chaetotaxy for a number of families of Coleoptera has been studied, including most families of the Archostemata (Grebennikov 2004), Carabidae (Bousquet and Goulet 1984), and Dytiscidae (Alarie and Balke 1999) in the Adephaga, and the Staphylinidae (Ashe and Watrous 1984), Histeridae (Kovarik and Passoa 1993), Ptiliidae (Grebennikov and Beutel 2002), Hydraenidae (Delgado and Soler 1997), and Leiodidae (Wheeler 1990) in the Staphyliniformia of the Polyphaga. This is the 1st paper to discuss the larval chaetotaxy of a family within the Cucujoidea.

### METHODS AND MATERIALS

Adults of *Helota thoracica* (Taiwan) and *H.*

\*To whom correspondence and reprint requests should be addressed. Tel: 886-4-23302301 ext. 631. Fax: 886-4-23302804. E-mail:cflee@gate.sinica.edu.tw

*gemmata* (Japan) were collected at rotten fruit-baited traps. Specimens were placed into plastic boxes containing some pineapple and short tree branches. Females were observed to lay eggs between crevices on the tree branches, and additional substrates were tested to determine which substrate females preferred for oviposition. Further observations suggested that coconut fiber was favored by females since most eggs were laid there. Hatched larvae also fed on pineapple and exhibited low mortality. Mature larvae tended to bore into the tree branches and construct a chamber for pupation. However, because larvae normally pupate inside tree branches, it is impossible to observe when pupation occurs. Also, when the pupal chamber is disrupted prior to pupation, the larva will search for another site. Such larvae often die, having depleted too much of their energy reserves. Polystyrene was found to provide a very good substitute substrate for larval pupation. Because polystyrene is soft and translucent, larvae were able to easily bore into it, and pupation could easily be observed. Using this method, more than 100 larvae and 10 pupae of *H. thoracica*, as well as 20 larvae and 3 pupae of *H. gemmata* were available for examination.

Two principal systems of setal nomenclature have been proposed for larval Coleoptera: the Ashe-Watrous system (Ashe and Watrous 1984) and the Bousquet-Goulet system (Bousquet and Goulet 1984). The former is better suited to larval Helotidae since setal orientation is longitudinal. We followed a modification of the Ashe-Watrous system proposed by Wheeler (1990) since it is more easily represented. Labels proceed in general from anterior to posterior in rows, and from mesad to laterad in the order of rows. Simple designations of position are used to name rows, such as D for dorsal, L for lateral, and so forth. Setae are numbered within rows from anterior to posterior beginning with 1, and rows lettered from midline to laterad beginning with the letter "a". Morphological area codes are uppercase letters; rows lowercase letters; and setal numbers are given as Arabic numerals. The head bears a number of posterior setae (e.g., Fig. 1, P1-P4), but such setae cannot be applied to the thoracic and abdominal terga since the number of setae of posterior rows on these terga is consistent with those of anterior ones. Regarding the posterior rows as regular rows is more convenient. In the Ashe-Watrous system, each leg segment is divided into 4 fields: anterior, posterior, dorsal, and ventral. Setae in the center of each field are labeled as

anterolateral, posterolateral, dorsal, and ventral, respectively. Setae that are offset from the center of the anterior and posterior fields are labeled as dorsal or ventral according to their relative position. The dorsal and ventral fields are not subdivided. Setae in the same subfield or row are numbered from the base to the apex beginning with 1. While most of the leg chaetotaxy is interpretable from this diagram, that of the coxa is too variable to warrant further comment.

## RESULTS

### *Helota thoracica* Ritsema, 1895 (Lee et al. 2006)

*Egg*: Length 2.9-3.0 mm. Pale yellow, elongate, cylindrical, rounded at both ends.

*Larva*: *Head*: Instar I (Fig. 1). Maximum width 1.0-1.1 mm. Stemmata 6. Dorsal chaetotaxy: with 5 rows, row Da with 3 setae (Da1-3); row Db with 3 setae (Db1-3); row Dc with 2 setae (Dc1, 2); row Dd with 4 setae (Dd1-4); row De with 3 setae (De1-3); 2 lateral setae (L1, 2); posterior row with 4 setae (P1-4); campaniform sensilla: 1 (a) between Db1 and Dc1, 1 (b) between Db2 and Db3, 1 (c) between Da2 and Da3, 1 (d) close to frontal suture and posterior to Dc2, 1 (e) outer to posterior setae, and 1 (f) between posterior to De1. Antenna: antennomere I with 2 dorsal campaniform sensilla; antennomere II with 1 subapical seta dorsally and 1 subapical seta and with 2 dorsal campaniform sensilla; apex with 2 sensoria and 1 seta. Labrum with 10 setae along each side, mostly along lateral margins. Venter (Fig. 2): 4 setae (V1-4) and 2 campaniform sensilla posterior to V4. Maxillae: cardo externally divided, inner sclerite with 1 seta; stipes with 2 setae. Labium: submentum and mentum each with 1 seta; prementum with 2 setae.

Instar II: Maximum width 1.5-1.6 mm. Chaetotaxy same as that of instar III.

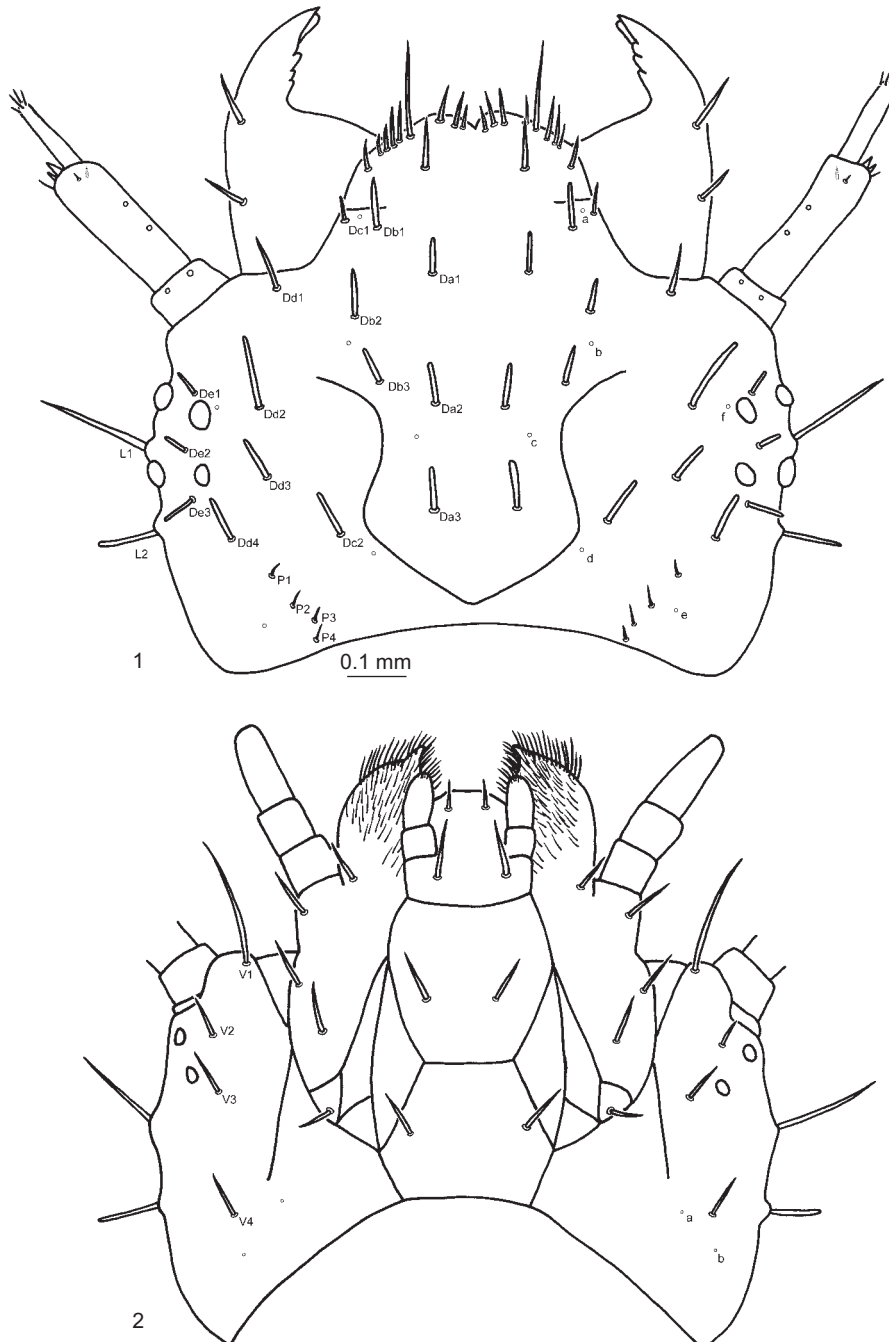
Instar III: Maximum width 2.1-2.3 mm. Dorsal chaetotaxy (Fig. 3) same as in instar I but smaller, except for 3 secondary setae between Db2 and Db3, 1 between Da3 and Db3, 4 among Db3, Dc2, Dd2, and Dd3; 2 more campaniform sensilla (g, h) between Dd2 and Dd3. Venter (Fig. 4) with 2 additional setae between L1 and L2.

*Pronotum*: Instar I (Fig. 5). Chaetotaxy: 4 rows (Da-Dd) with 3 setae each; 2 lateral setae (L1, 2); with 1 additional lateral seta above L1; all but L1 and L2 paddle-shaped; L1 and L2 longest and L1 longer than L2; Dc1 and Dd1 similar and a

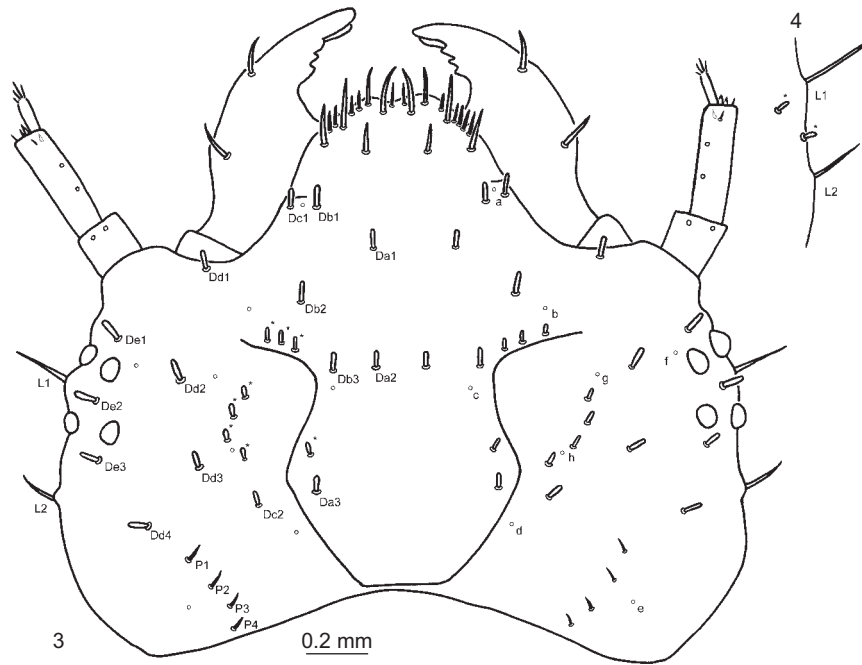
little shorter than L1 and L2, much longer than Da1 and Db1; Da2, Db2, Dc2, and Dd2 similar and shortest, Db3 and Dd3 similar and longer than Da3 and Dc3. Campaniform sensilla: 1 (a) between middle line and Da1, 1 (b) between Da1 and Db1, 1 (c) or 2 (d) between Db1 and Dc1, 1 (e) between Dc1 and Dd1, 1 (f) between Dd1 and L1, 1 (g) between L1 and L2, 1 (h) between L2 and Dd3, 1

(i) between Dc3 and Dd3, 1 (j) between Db3 and Dc3, 1 (k) between Da3 and Db3, 1 (l) between Da2 and Db2, 1 (m) outer to Dd2.

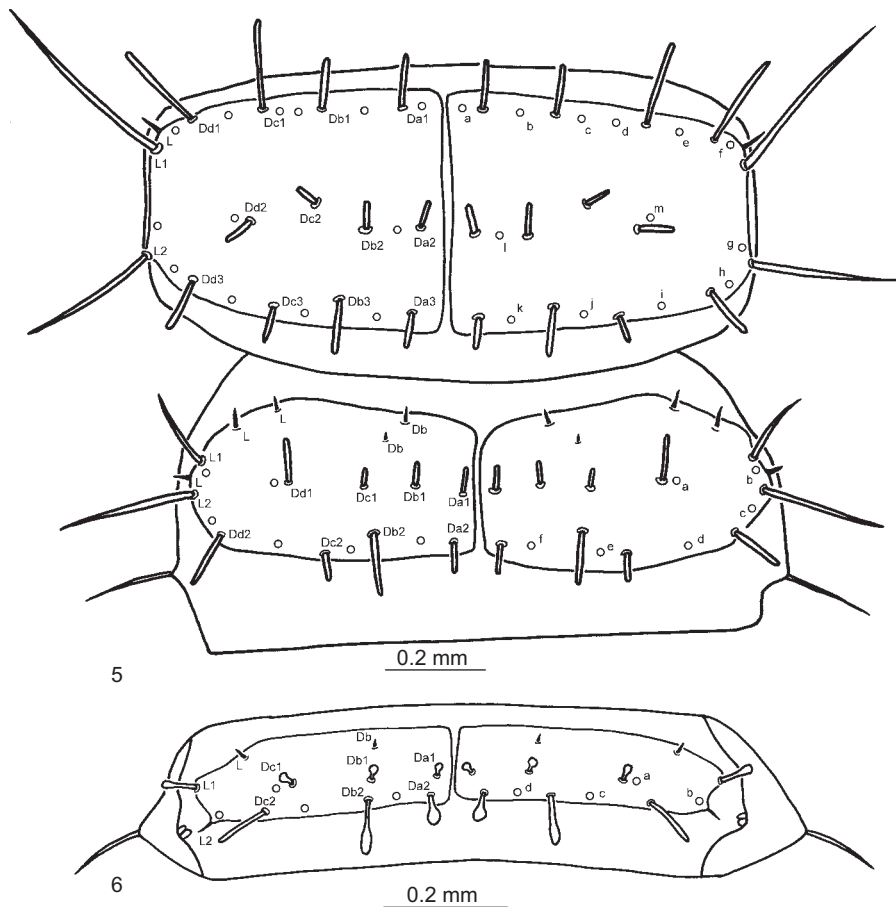
Instar II (Fig. 7): Chaetotaxy similar to that of instar I, but surface with dense, short, secondary, paddled-shaped setae; Dc1, Dd1, L1, L2, and Dd3 much longer than others. Campaniform sensilla: similar to those of instar I, but sometimes with 1



**Figs. 1, 2.** *Helota thoracica*, larval instar I, head. 1. Dorsal view; 2. ventral view.



**Figs. 3, 4.** *Helota thoracica*, larval instar III, head. 3. Dorsal view; 4. ventral view.



**Figs. 5, 6.** *Helota gemmata*, larval instar I. 5. Pro- and mesonota; 6. abdominal tergum III.

more sensillum (n) present between Dc3 and Dd3, sensillum l absent.

Instar III (Fig. 9): Chaetotaxy similar to that of instar II, but secondary setae at lateral margins well developed; secondary seta (L) between Dd1 and L1 very small. Campaniform sensilla: similar to those of instar II, but sensillum (f) between Dd1 and L1, and 1 (h) between L2 and Dd3 absent.

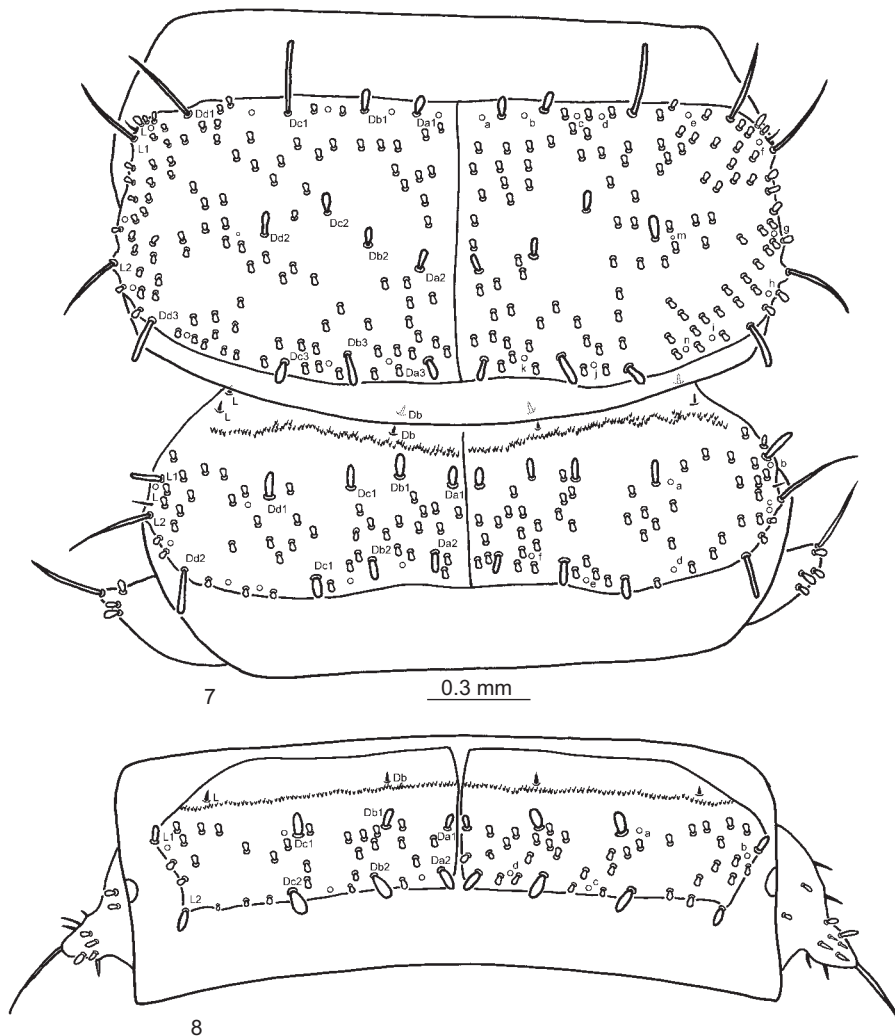
*Mesonotum*: Instar I (Fig. 5). Chaetotaxy: 4 rows (Da-Dd) with 2 setae each; 2 lateral setae (L1, 2); with 2 additional setae above row Db; with 3 additional lateral setae, 2 above L1, 1 between L1 and L2; all but L1 and L2 paddle-shaped; L2 longest and much longer than L1; Dd1 shorter than L1 and much longer than Da1, Db1, and Dc1; Db2 and Dd2 similar to L1 and much longer than Da2 and Dc2. Campaniform sensilla: 1 sensillum between Dd1 and L1 (a), L1 and L2 (b), L2 and

Dd2 (c), Dc2 and Dd2 (d), Db2 and Dc2 (e), and Da2 and Db2 (f). Metanotum similar to mesonotum.

Instar II (Fig. 7): Chaetotaxy: similar to that of instar I, but surface with dense, short, secondary paddled-shaped setae; only L1, L2, and Dd2 much longer than others.

Instar III (Fig. 9): Chaetotaxy: similar to that of instar II, but secondary seta (L) between L1 and L2 very small. Campaniform sensilla: similar to those of instars I and II, but sensillum (c) between L2 and Dd2 absent.

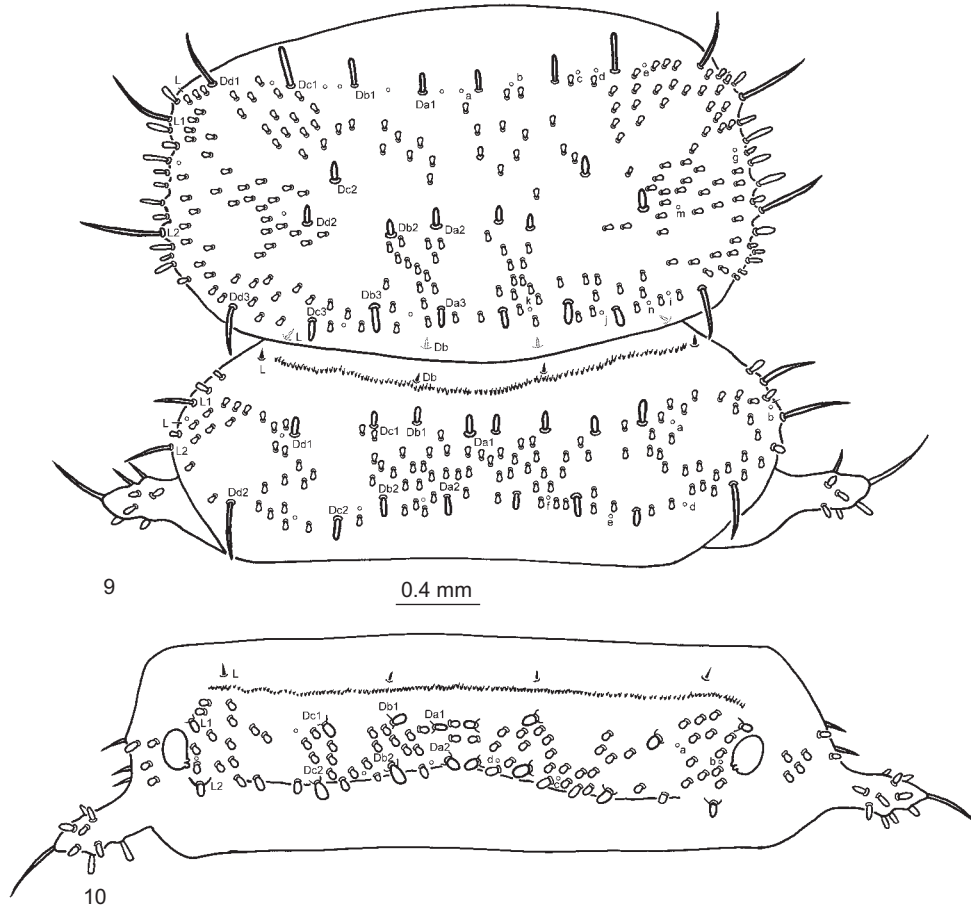
*Legs*: Instar III (Fig. 11): Trochanter with 1 anterodorsal seta (Ad1), 1 anteroventral seta (Av1), 1 ventral seta (V1), 1 posterolateral seta (Pl1), and 1 posteroventral seta (Pv1). Femur with 2 anterodorsal setae (Ad1, 2), 1 anterolateral seta (Al1), 2 ventral setae (V1, 2), 5 posterodorsal



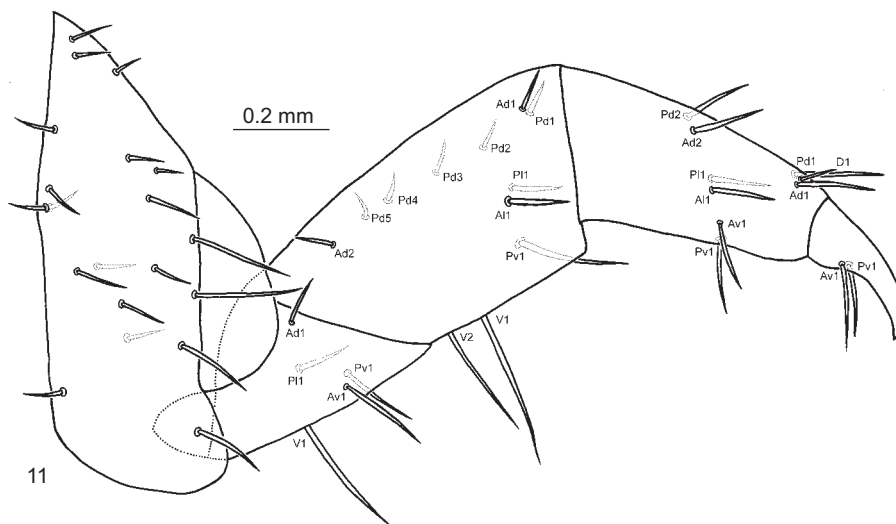
Figs. 7, 8. *Helota thoracica*, larval instar II. 7. Pro- and mesonota; 8. abdominal tergum III, same scale as in figure 7.

setae (Pd1-5), 1 posterolateral seta (PI1), and 1 posteroventral seta (Pv1). Tibia with 1 dorsal seta (D1), 2 anterodorsal setae (Ad1, 2), 1 anterolateral

seta (Al1), 1 anteroventral seta (Av1), 2 posterodorsal setae (Pd1, 2), 1 posterolateral seta (PI1), and posteroventral setae (Pv1). Tarsungulus with a pair



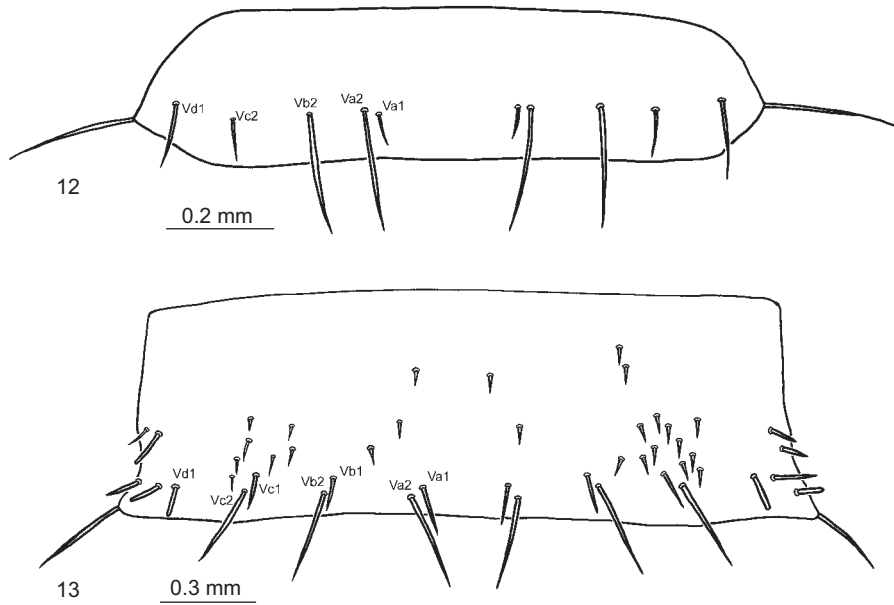
**Figs. 9, 10.** *Helota thoracica*, larval instar III. **9.** Pro- and mesonota; **10.** abdominal tergum III, same scale as in figure 9.



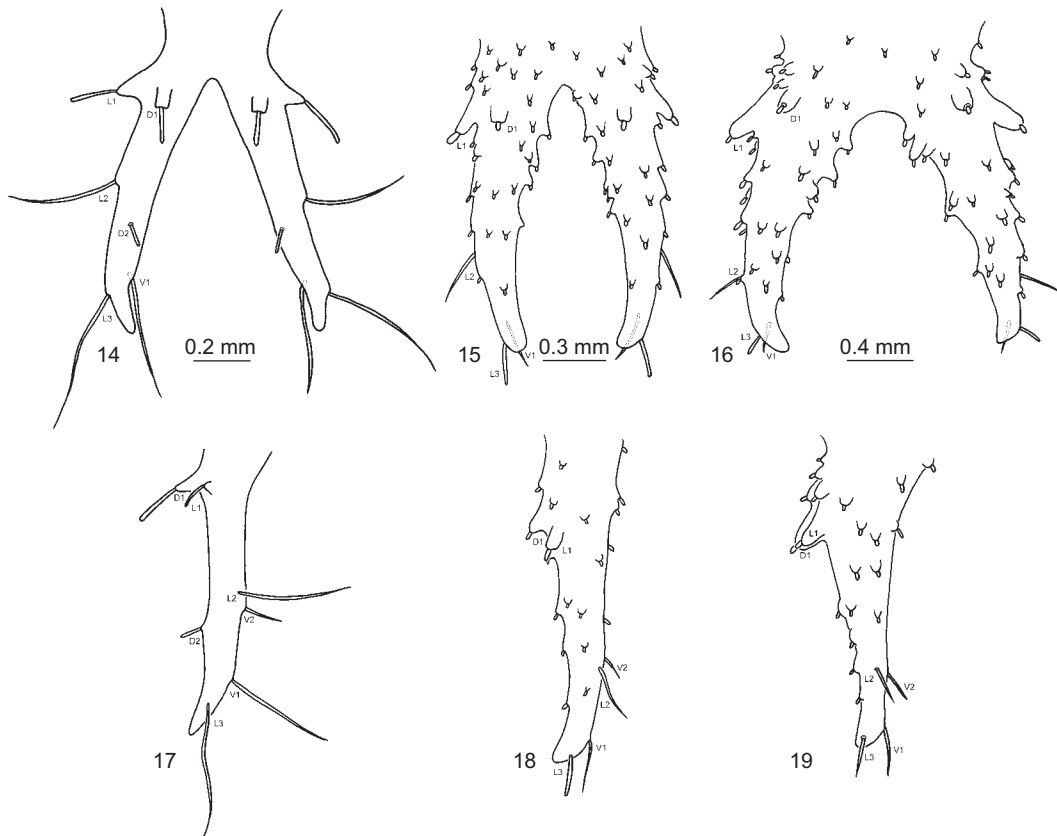
**Fig. 11.** *Helota thoracica*, larval instar III, leg, anterior view.

of setae (Av1, Pv1). Instar I similar to instar III but lacking 4 posterodorsal femoral setae (Pd2-5). Instar II similar to instar III.

*Abdominal tergum III*: Instar I (Fig. 6). Chaetotaxy: 3 rows (Da-Dc) with 2 setae each; 2 lateral setae (L1, 2); with 1 additional seta on later-



**Figs. 12, 13.** *Helota thoracica*, abdominal sternum III. **12.** Larval instar I; **13.** larval instar II.



**Figs. 14-19.** *Helota thoracica*, urogomphi. **14.** Larval instar I, dorsal view; **15.** larval instar II, dorsal view; **16.** larval instar III, dorsal view; **17.** larval instar I, lateral view; **18.** larval instar II, lateral view; **19.** larval instar III, lateral view.

al row (D) and 1 seta on row Db; all but L2 paddle-shaped; Db2 and Dc2 longest and much longer than Da2; Da1-Dc1 similar and shorter than Da1; L1 similar to Da1 and much longer than L2. Campaniform sensilla: 1 sensillum between Dc1 and L1 (a), L1 and L2 (b), Db2 and Dc2 (c), and Da2 and Db2 (d). Other abdominal terga similar to tergum III.

Instar II (Fig. 8): Chaetotaxy: similar to that of instar I, but surface with dense, short, secondary paddle-shaped setae; all primary setae similar and a little larger than secondary setae.

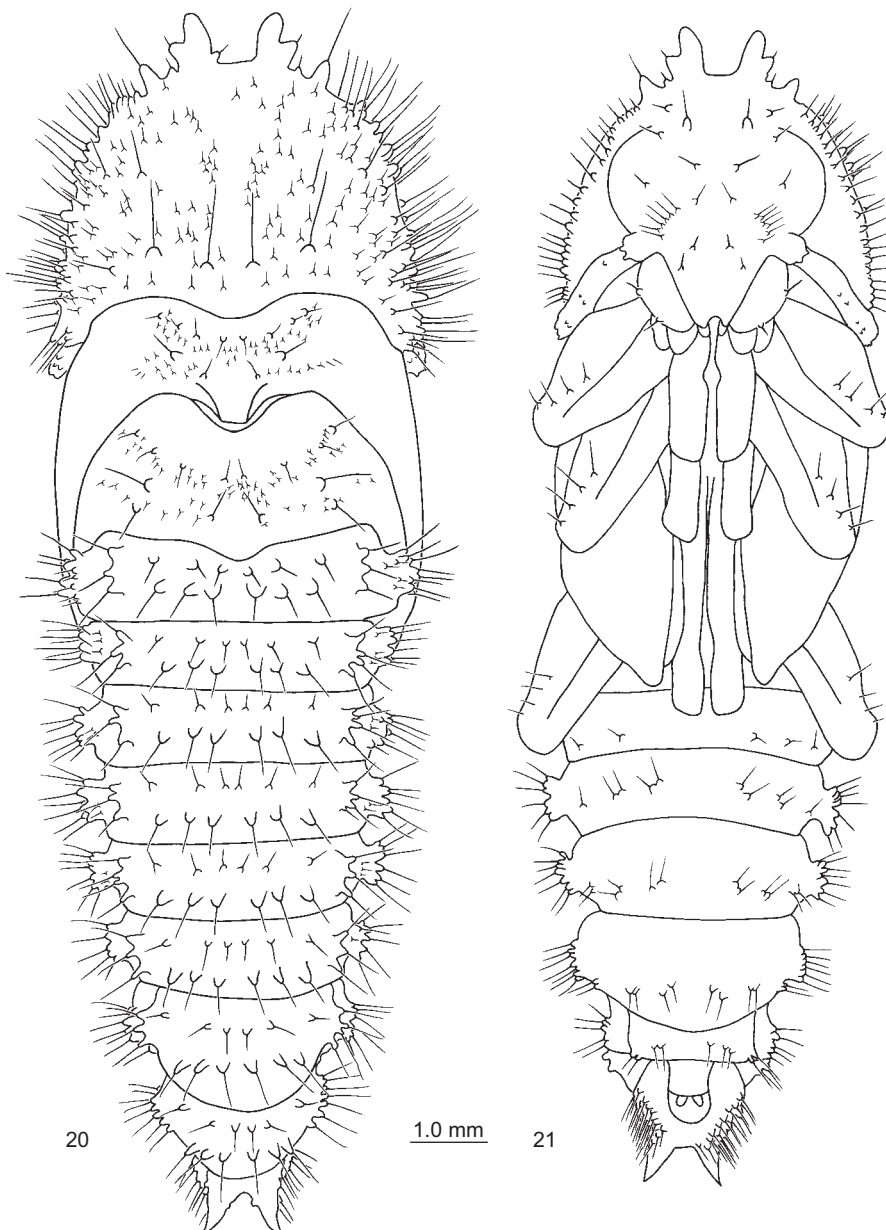
Instar III (Fig. 10): Similar to instar II.

*Abdominal sternum III*: Instar I (Fig. 12). Chaetotaxy: 4 rows (Va-Vd), 1st row with 2 setae (Va1, 2), others with 1 each (Vb2, Vc2, and Vd2). Other abdominal sternites similar to sternite III.

Instar II (Fig. 13): Chaetotaxy: similar to that of instar I, but 1 more setae present on rows Vb and Vc (Vb1, Vc1), with small scattered secondary setae.

Instar III: similar to instar II.

*Urogomphi*: Instar I. Surface without tubercles except for D1 and L1. Chaetotaxy: dorsum with 2



Figs. 20, 21. *Helota thoracica*, pupa. 20. Dorsal view; 21. ventral view.

setae (D1, 2), lateral margins with 3 setae (L1-3), venter with 2 setae (V1, 2); L3 and V1 near apices, D2 anterior to L3 and V1, L2 and V2 anterior to D2 and at middle, D1 and L1 anterior to L2 and V2 and near base; L1, D1, and D2 elongate, paddle-shaped; others long hair-like.

Instar II: Surface with scarce tubercles, 1 small paddle-shaped seta on apex of each tubercle. Chaetotaxy: D2 absent, others much shorter than instar I.

Instar III: Similar to instar II, but wider and directed outwardly.

*Pupa*: Dorsum (Fig. 20): Two short processes at anterior margin near midline. Pronotum with sparse tubercles, 1 seta located at tip of each tubercle, those on medial portion relatively smaller. Meso- and metanota with minute setae and several prominent setae. Abdominal terga I-VII with 2 rows mesially, each row with 3 setae, anterior setae smaller than posterior setae; 1 cluster of 3-6 setae at sides; another cluster of 7-12 setae at lateral margins. Urogomphi present.

Venter (Fig. 21): Lateral margins of pronotum with dense setae; several setae between eyes; a cluster of 6 setae at bases of eyes; 2 setae near base of each mandible; 4 setae near apex of each femur. Abdominal sternites IV-VIII with 3 pair of setae at each side, those on sternites IV and VIII somewhat reduced.

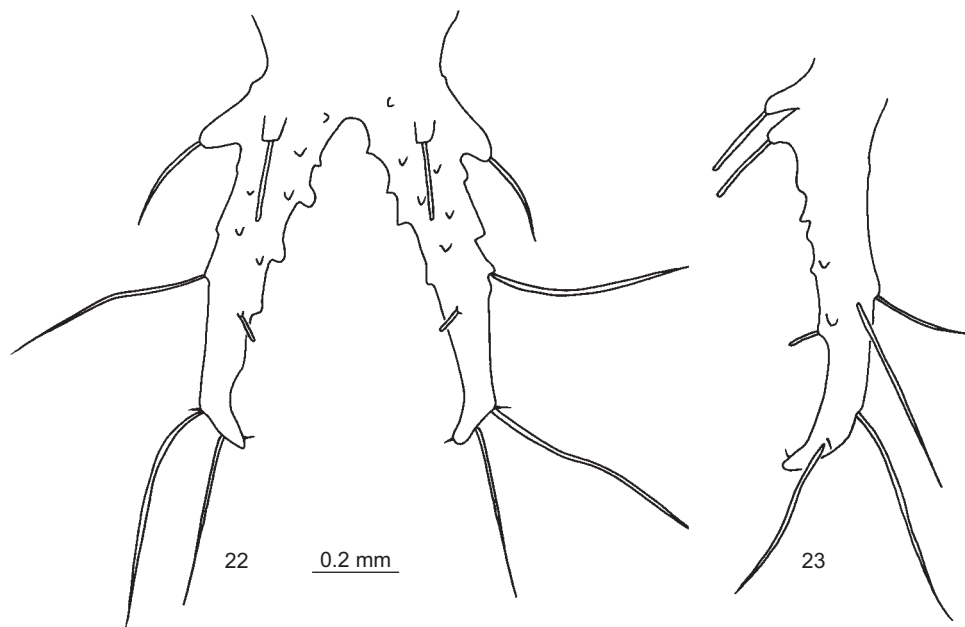
*Diagnosis*: Immature stages of *H. gemmata* are very similar to those of *H. thoracica*, except

that the urogomphi of the larval instar I have scattered tubercles and 1 small seta on the apices and bases of V3, respectively (Figs. 22, 23). Both setae also appear in subsequent instars.

*Biology*: All immatures ( $n = 32$ ) were reared at the laboratory ( $25 \pm 2^\circ\text{C}$ ). Eggs lasted  $5.69 \pm 0.78$  d, 1st stadium lasted  $4.50 \pm 0.57$  d, the 2nd stadium lasted  $5.00 \pm 0.67$  d, the 3rd stadium lasted  $16.75 \pm 2.37$  d, and the pupal stadium lasted  $10.13 \pm 1.01$  d.

## DISCUSSION

Larval setae were added at the molt between instars I and II and retained in similar numbers and positions in instar III. The ontogeny of larval setae is similar to Wheeler's observation (1990) of the Leiodidae. He defined 3 types of setae as follows: primary setae are those of constant number and placement present in neonatal larvae; secondary setae are of variable size, placement, and presence, generally added in later instars, but of comparatively little taxonomic value; and subprimary setae are constant in number and placement, but added after the 1st instar. On larval helotids, almost all setae present in larval instar I, including minute setae on the thoracic and abdominal tergites (such as L and Db) can be regarded as primary setae. Shortening primary setae in later instars is a trend throughout all morphological



Figs. 22, 23. *Helota gemmata*, urogomphi, larval instar I. 22. Dorsal view; 23. lateral view.

areas, in particular for those setae on the thoracic and abdominal tergites and urogomphi. Primary setae on the thoracic and abdominal tergites in larval instars II and III (Figs. 8, 10) are so short that they are difficult to distinguish from secondary setae, except for the lateral setae of thoracic tergites (such as L1, L2, and Db2). Primary setae L1, D1, and D2 on the urogomphi of later instars are too small and similar to the secondary setae to be recognizable (Figs. 14-19). However, L1 and D1 can be distinguished because they are located at the tops of prominent tubercles.

Subprimary setae appear on abdominal sternites (Fig. 13: Vb1 and Vc1), head, and legs. The presence of subprimary setae Vb1 and Vc1 on the abdominal sterna is special and interesting.

Adults of *H. thoracica* can easily be distinguished from those of *H. gemmata* by the patterns of punctures on the elytra and genital structures. Tiny differences between larvae of *Helota thoracica* and *H. gemmata* (see diagnosis) imply that the larval chaetotaxy is more conservative than adult characters. Pupae of *Helota* may be much more conservative since no significant differences between the species were observed.

**Acknowledgments:** We thank Ms. Yi-Ching Yu for assistance with rearing helotids, especially for new ideas relating to female oviposition and larval pupation. This project was supported financially by a grant (NSC96-2313-B-055-005-MY2) from the National Science Council, Taiwan. Finally, we thank Mike C. Thomas and Daniel K. Young for reading the manuscript.

## REFERENCES

- Alarie Y, M Balke. 1999. A study of larva of *Carabdytes upin* Balke, Hendrich and Wewalka (Coleoptera: Adephaga: Dytiscidae), with comments on the phylogeny of the Colymbetinae. *Coleopt. Bull.* **53**: 146-154.
- Ashe JS, LE Watrous. 1984. Larval chaetotaxy of Aleocharinae (Staphyliinoidea) based on a description of *Atheta coriarica* Kraatz. *Coleopt. Bull.* **38**: 165-179.
- Bousquet Y, H Goulet. 1984. Notation of primary setae and pores on larvae of Carabidae (Coleoptera: Adephaga). *Can. J. Zool.* **62**: 573-588.
- Delgado JA, AG Soler. 1997. Morphology and chaetotaxy of larval Hydraenidae (Coleoptera) I: genus *Limnebius* Leach, 1815 based on a description of *Limnebius cordobanus* d'Orchymont. *Aquat. Insect.* **19**: 37-49.
- Fukuda A. 1943. Description of the larva of *Helota gemmata* Gorham. *Trans. Nat. Hist. Soc. Formosa* **33**: 15-18. (in Japanese)
- Fukuda A. 1944. Description of the pupa of *Helota gemmata* Gorham. *Trans. Nat. Hist. Soc. Formosa* **34**: 119-122. (in Japanese)
- Grebennikov VV. 2004. Review of larval morphology of beetles of the suborder Archostemata (Insecta: Coleoptera), including first-instar chaetotaxy. *Eur. J. Entomol.* **101**: 273-292.
- Grebennikov VV, RG Beutel. 2002. Morphology of the minute larva of *Ptinella tenella*, with special reference to effects of miniaturization and the systematic position of Ptiliidae (Coleoptera: Staphyliinoidea). *Arthropod. Struct. Dev.* **31**: 157-172.
- Kovarik PW, S Passoa. 1993. Chaetotaxy of larval Histeridae (Coleoptera: Hydrophiloidea) based on a description of *Onthophilus nodataus* Leconte. *Ann. Entomol. Soc. Am.* **86**: 560-576.
- Lee CF, M Satô. 2006. The Helotidae of Taiwan (Coleoptera: Cucujoidea). *Zool. Stud.* **45**: 529-552.
- Oloff AS. 1883. Descriptions of two larvae and new genera and species of Clavicorn Coleoptera, and a synopsis of the genus *Helota*, MacLeay. *Cist. Entomol.* **3**: 49-61.
- Ritsema C. 1895. A new species of the genus *Helota* from Thibet. *Notes Leyden Mus.* **17**: 49-50.
- Wheeler QD. 1990. Morphology and ontogeny of postembryonic larval *Agathidium* and *Anisotoma* (Coleoptera: Leiodidae). *Am. Mus. Novit.* **2986**: 1-46.