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Article

Immature stages of *Pseudregolis wedah* (Kollar, 1844) (Lepidoptera, Nymphalidae)[1)2)]

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Abstract

The external morphology of the immature stages of *Pseudergolis wedah* (Kollar, 1844) are described and illustrated in detail together with some notes on larval behavior. The most conspicuous feature of the immature stages of this species appears in the pupal morphology. That is, the 8th and the subsequent abdominal segments are located along the posterior margin of the 7th segment, and arranged dorso-ventrally to the body axis of anterior segments just under the posteriorly elongated dorsal part of the 7th segment. Consequently, the cremaster is just behind the 7th segment on the abdominal plane. The egg and larvae are similar to those of *Dichorragia* and *Stibochiona*, but the head horns of 4th and 5th instars are provided with many distinctive processes, and the 5th instar has a dorsal protuberance on the 3rd abdominal segment.

The larvae of earlier stages, at least 1st and 2nd instars, construct a resting perch with their own frass pellets and silk at the leaf edge of the host plant.

Key words: immature stages, *Pseudergolis wedah*, Lepidoptera, Nymphalidae, morphology, behavior.

Introduction

The genus *Pseudergolis* C. et R. Felder, [1867], is a nymphalid genus comprising only two species, *P. avesta* (C. et R. Felder, [1867]) from Sulawesi and *P. wedah* (Kollar, 1844) from India to Central China. This genus is currently classified in the subfamily Pseudergolinae of the family Nymphalidae (the tribe Pseudergolini of the subfamily Nymphalinae (s. lat.)) together with the genera *Dichorragia, Stibochiona* and *Amnosia*.

Mackinnon and Nicéville (1898) reported on the larva and pupa of *P. wedah* for the first time and recorded *Debregeasia bicolor* (Urticaceae) as a larval host plant of this species. Later, Moore (1901-1903), Bingham (1905), and Stichel (1909) described the larva and/or pupa based mainly on the original description and drawings of Mackinnon and Nicéville. However, the descriptions and figures given by them are so brief and rough that it is difficult to compare the early stages of *Pseudergolis* with those of its relatives based solely on their works. Recently, Watanabe (1996) published a short report on the immature stages of this species, in which he recorded *Debregeasia edulis* Wedd. as a host plant, and provided photographs of an early and a final instars in the field and of the pupa and the adult. Further detailed observations on the morphology and behavior of the immatures of this genus are essential.

We had the opportunities to rear the immature stages of *Pseudergolis wedah* and to observe the morphology and behavior of all the immature stages of this butterfly at several localities in Sichuan (1993), Yunnan (1994) and Guizhou (1995) in south-western China. Although we had only poor specimens for the first and the last instars, we obtained considerable

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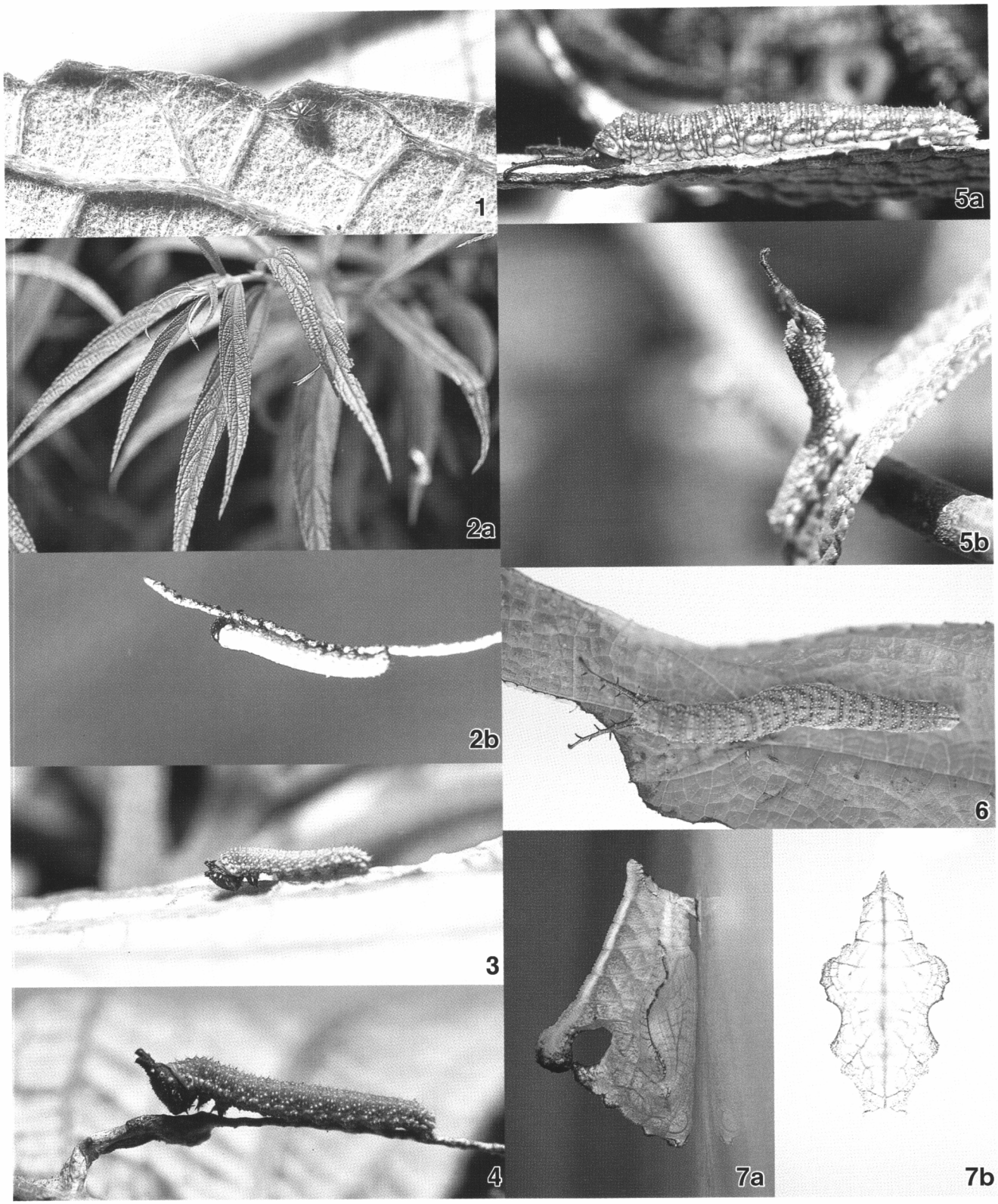


Figs. 1-7. Immatures of *Pseudergolis wedah* (Koll.). 1: Egg on the under surface of leaf of the host plant. From this egg one hymenopterous parasitoid emerged. 2a, 2b: First instar larva on the resting perch. 3: Second instar larva. 4: Third instar larva. 5a: Fourth instar larva in rest. 5b: Ditto, taking threatening posture against some stimulus. 6: Full grown fifth (last) instar larva on the upper surface of the host plant leaf. 7a: Greenish pupa. 7b: Brownish gray pupa.

new information about these larval stages together with other stages.

In the present paper we describe the external morphology of the immature stages of this species together with brief notes on larval behavior.

Materials and method

The eggs were obtained from the host plant, *Debregeasia bicolor*, in the field in Gairi, Guizhou Province, of which only one was observed being deposited by a female of this species in the field. In addition, eggs dissected from one dried female specimen were used for the study[1)]

All the examined larvae were collected from the host plant in Guizhou Province (Gairi and Injian in late July, 1995), in Sichuan Province (Tian-quan-xian, Tian-quan-he, 30° 5′ N, 102° 35′ E, about 1,300 m above sea level, on August 29, 1993), and in Yunnan Province (Kung-shan, 27°45′N, 98°40′E on June 19, 1994). We raised three pupae from the larvae on this host plant, and two adult males from the pupae. We also collected a live pupa from the host plant in Gairi. The larvae of all instars and one pupa were fixed and preserved in 70 % ethanol. The exuviae of pupae were also used for the description together with the ethanol preserved pupa. Descriptions of coloration were made from observation of live and preserved speci-ens and from color slides of live material. The external characters of larvae and pupae were observed under a binocular microscope. The chaetotaxy of the 1st and 2nd instar larvae was examined under a compound microscope (up to x 600) based on the larvae macerated with 10 % KOH solution.

The terminology of the first instar larva follows Hinton (1946) with some modifications (cf. Nakanishi, 1988), and the terminology of the pupa follows Mosher (1916).

Results

Morphology of Immature Stages

Egg (Fig.1). White when oviposited, becoming yellowish thereafter; barrel-shaped, 0.56 mm in diameter and 0.62 mm high, with 13-15 longitudinal ribs traversed by faint cross-braces.

First instar larva (Figs. 2, 8, 13). Length 3.5 mm; width of head 0.36mm; height of head 0.3 mm measured from vertex to apical margin of clypeus. Head piceous black, body greenish, scattered with numerous minute white spots. Setae on both head and body black, very short and almost straight; D1 seta of second abdominal segment about 0.14 times as long as the height of the segment. Head with 17 primary setae on each side as is usual in nymphalid species. The setae of D, SD and L groups of all the segments,

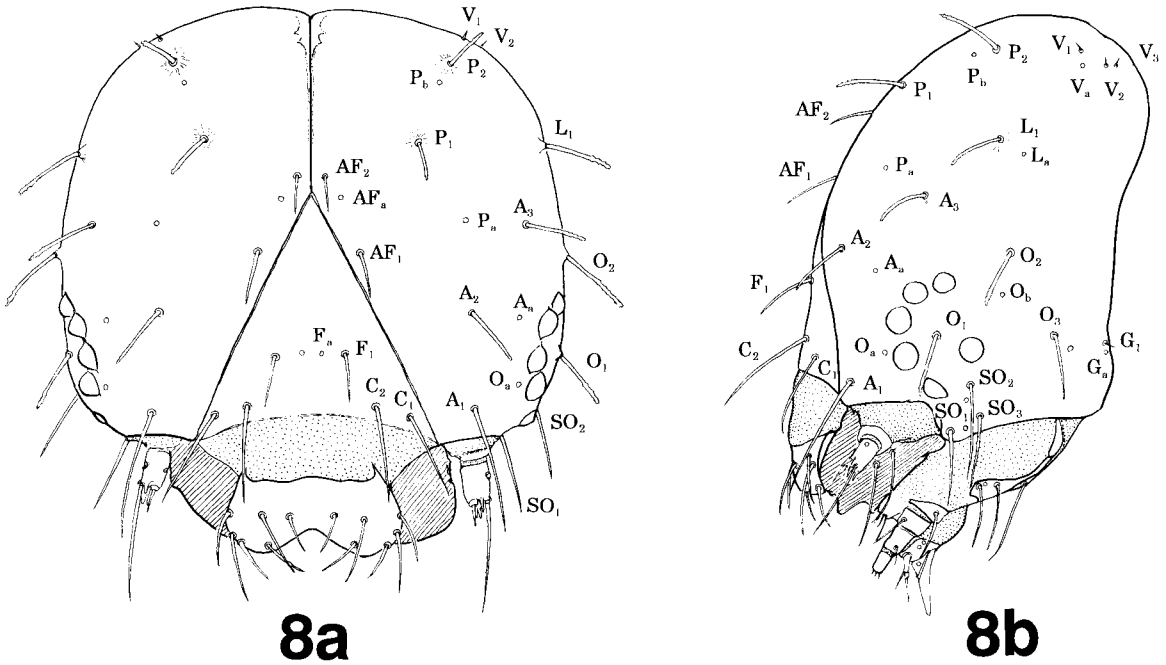
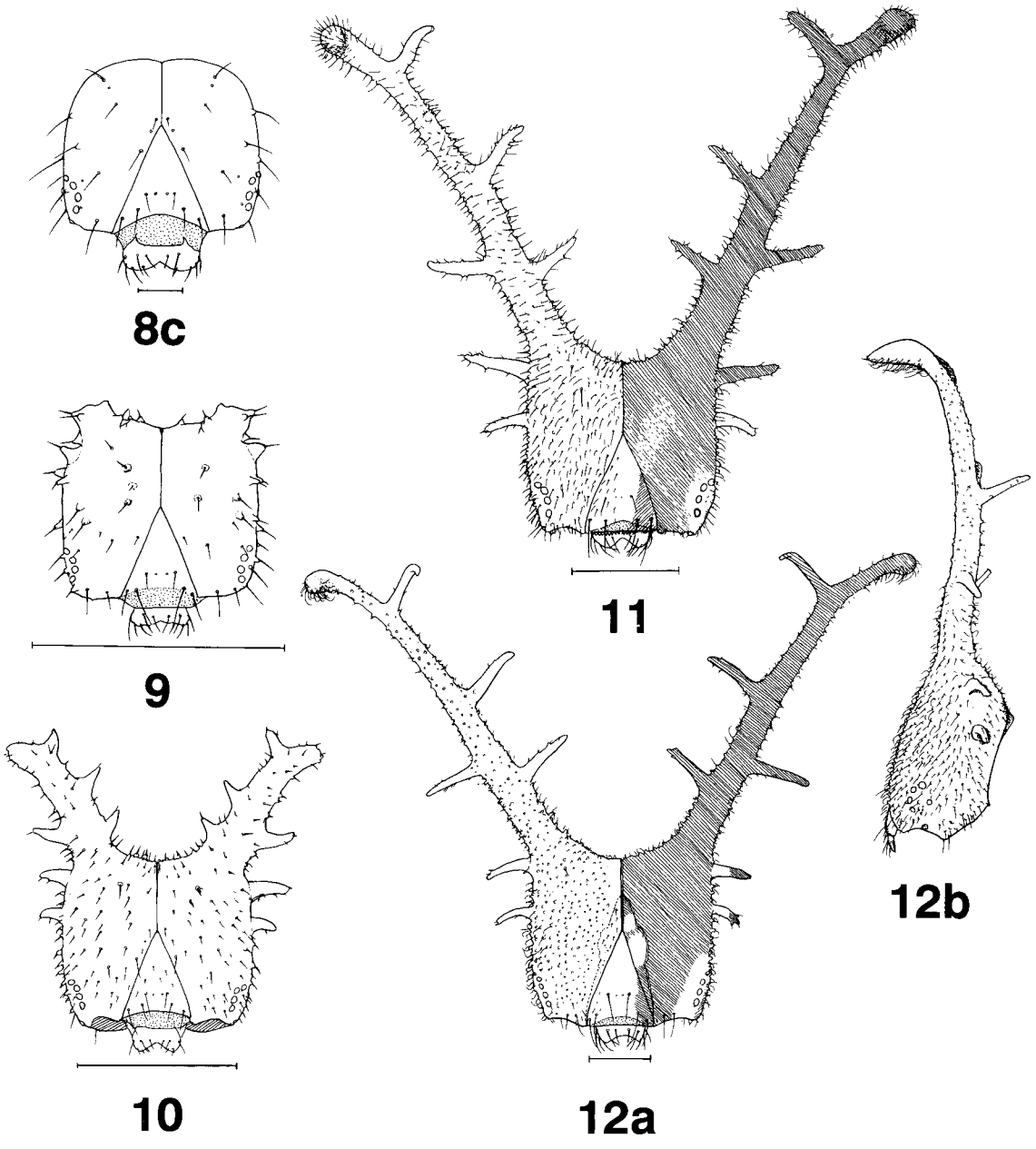


Fig. 8. Head of 1st instar. 8a: Frontal aspect. 8b: Lateral aspect.

1) The entire abdomen of the female was boiled in 7% solution of potassium hydroxide for 5 minutes, and then washed in freshwater. The matured eggs were carefully isolated from other substances in the abdomen under a binocular microscope. The eggs were then transferred into 70% ethanol and observed.



Figs. 8c–12. Cranium of larvae; frontal aspect except for 12b. 8c: First instar. 9: Second instar. 10: Third instar. 11: Fourth instar. 12a: Last instar. 12b: Ditto, lateral aspect. Scales 0.1mm for fig. 8 and 1mm for figs. 9–12.

of SV groups of thorax and the 1st abdominal segment and an external seta of the anal proleg shield more or less thick, often bluntly ended, minutely plumose apically. The setae of SV and V groups, and D2 and each one of SD and L setae of the prothorax, one of L setae of 8th, L seta of 9th abdominal segment, and an apical seta on the anal plate simple. Most of the macrosetae of the body arising from a raised socket on a small pinaculum. The microsetae as in Fig. 13; those on SV and V areas of the prothorax are omitted because of damage to the only available specimen. Prothorax (T1) : Thoracic shield bilaterally divided into two small sclerites, each of which carries two XD and two D, with D2 just posterior to XD1; two punctures arranged transversely between XD1 and D2; two SD on a common pinaculum anterodorsad to spiracle, SD2 simple and long; two L on a common pinaculum anterior to spiracle, L2 short and simple; two SV on a common pinaculum dorsal to leg; one V minute and slightly posterior to leg. Meso- and metathorax (T2 and T3) : D1 near dorsomedian line and approximately halfway along the segment; D2 ventral to D1; two SD on a small prominence ventral to D2, SD2 shorter and simple; L1 ventral to the prominence carrying SD; SV1 dorsal to leg; V1 as on T1. First abdominal segment (A1) : D1 slightly anterior to halfway of the segment; D2 posteroventral to D1 and posterior to half way of the segment; SD1, L1, SV1 and V1 arranged almost on a vertical line situated nearly half way along the segment ; L2 posterodorsal to L1, at the same level of the spiracle; V1 near the ventromedian line. Second abdominal segment (A2) similar to A1 except for the following points: L1 ventrad to spiracle, L2 a little posteroventral to spiracle, and the presence of SV2 between SV1 and V1. Third to sixth abdominal segments (A3-A6) similar to A2 except for two SV being on proleg shield. Both 7th and 8th abdominal segments (A7 and A8) very similar to A1 in chaetotaxy; A8 with spiracles larger than those of the preceding segments, and with simple L2. Ninth abdominal segment (A9) similar to A1 except for the absence of L2 and V1; SV long and fine. Tenth abdominal segment (A10) bearing 4 pairs of setae on anal plate, 5 setae on proleg shield and other 4 setae, P1, SP1, SV1 and V1 (cf. Kitching, 1984), and with many minute spines around anus. Prolegs with uniordinal crochets (6) arranged in a semicircle; anal proleg with uniordinal crochets (7) arranged in a semicircle.

Second instar larva (Figs. 3, 9, 14) . Body length 5-6 mm; width of head 0.7-0.8 mm; height of head 0.54 mm. Similar to first instar in coloration. Head less shiny than in first instar, with short thick horns on apices of epicrania; head horn about 0.13 times as long as height of head, with several short setae on dorsal and ventral regions; those on the dorsal region and on horns each arising from a short but distinct white conical tubercle. Chaetotaxy of second instar as follows: T1 similar to that in 1st instar, two pairs of darkened sclerites present posterior to D1, not covering the bases of XD and D setae; 2 minute secondary setae between D group and SD group; an additional SD seta posterodorsal to spiracle; L group consisting of 5 setae; SV with an additional seta. T2 to A7 each transversely divided into 5 ridges by furrows above the spiracular area; each ridge bearing 3-5 pairs of setae. T2 and T3 with the anteriormost (1st) ridge bearing 4 pairs of setae; 2nd ridge with 3 pairs of setae above the spiracular level; 2nd ridge also with 2 setae, each at the spiracular and subspiracular levels, and the former stronger than the latter; 3rd ridge with 3 setae dorsally and a strong seta at the spiracular level and presumably homologous with L seta of the 1st instar; 4th ridge with 3-4 pairs of setae in addition to a short and extremely fine seta below the lowermost seta, which seems to be homologous with one of SD setae, and 2 strong dorsal setae being homologous with D1 and D2 of 1st instar; the 5th ridge with 6 pairs of setae, of which the lowest three are arranged in an inverted triangle and related to L group; a seta posteroventral of the lowest seta of the 5th ridge; 2-3 setae possibly belonging to L group below the triangle; 3-4 SV setae lateral to coxa; V seta minute and posterior to coxa. A1 with a similar chaetotaxy as T2 and T3, and with 1st ridge without the most dorsal seta; submesal seta of 1st ridge is D1; SD1 represented by the lowest seta of 3rd ridge; 4th ridge with 4 pairs of setae, of which the submesal one is D2, and the lower two are not homologous with SD2; 5th ridge with 4 pairs of setae, of which the lowest one is at the spiracular level and just in front of the lower seta of the 1st ridge of the next segment; setae of L group arranged in three subgroups, one just in front of the spiracle and of 1 seta, the second group below the spiracle and consisting of 3 setae (the anteriormost strong, below spiracle, the second, L2, similar to the former and posteroventral to the spiracle; the last being posteroventral to the second) , the third group consisting of 3 setae, two on a swelling below the 2nd group and one posterior to the swelling; SV group consisting of a long seta and a small one posteroventral tothe former; V1 short and close to the ventromedian line. A2 to A7 fundamentally similar to A1, but 1st ridge with one seta at supraspiracular level and one L seta; the 4th ridge with 5 pairs of setae from A2 to A6, 3 pairs in A7 segment; the second group of L setae consisting of 6 setae (the anteriormost small, the next the lowermost and strongest, the 3 following arranged in an inverted

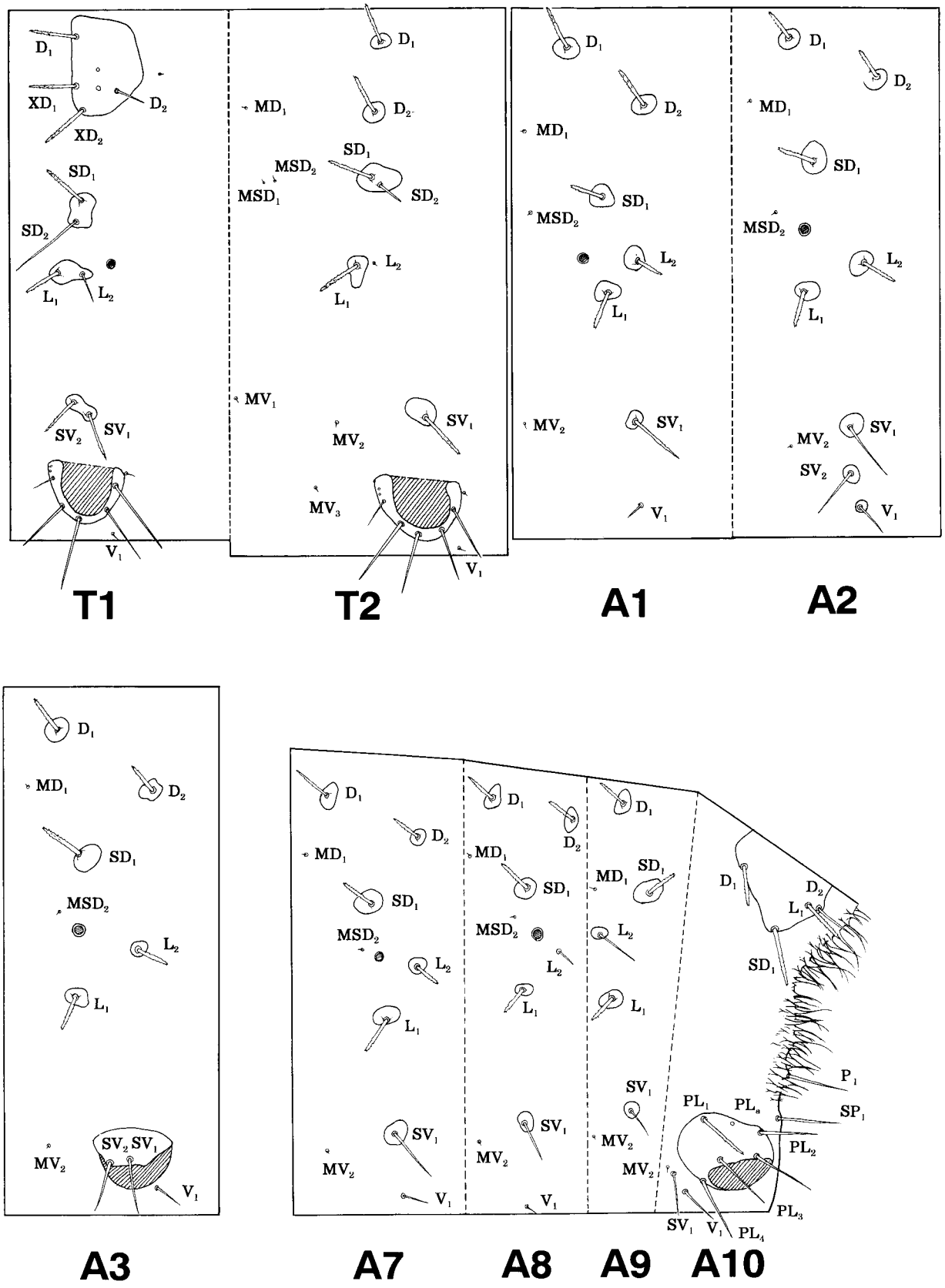


Fig. 13. Body chaetotaxy of first instar. T1: Prothorax. T2: Mesothorax. A1–A10: First to 10th abdominal segments.

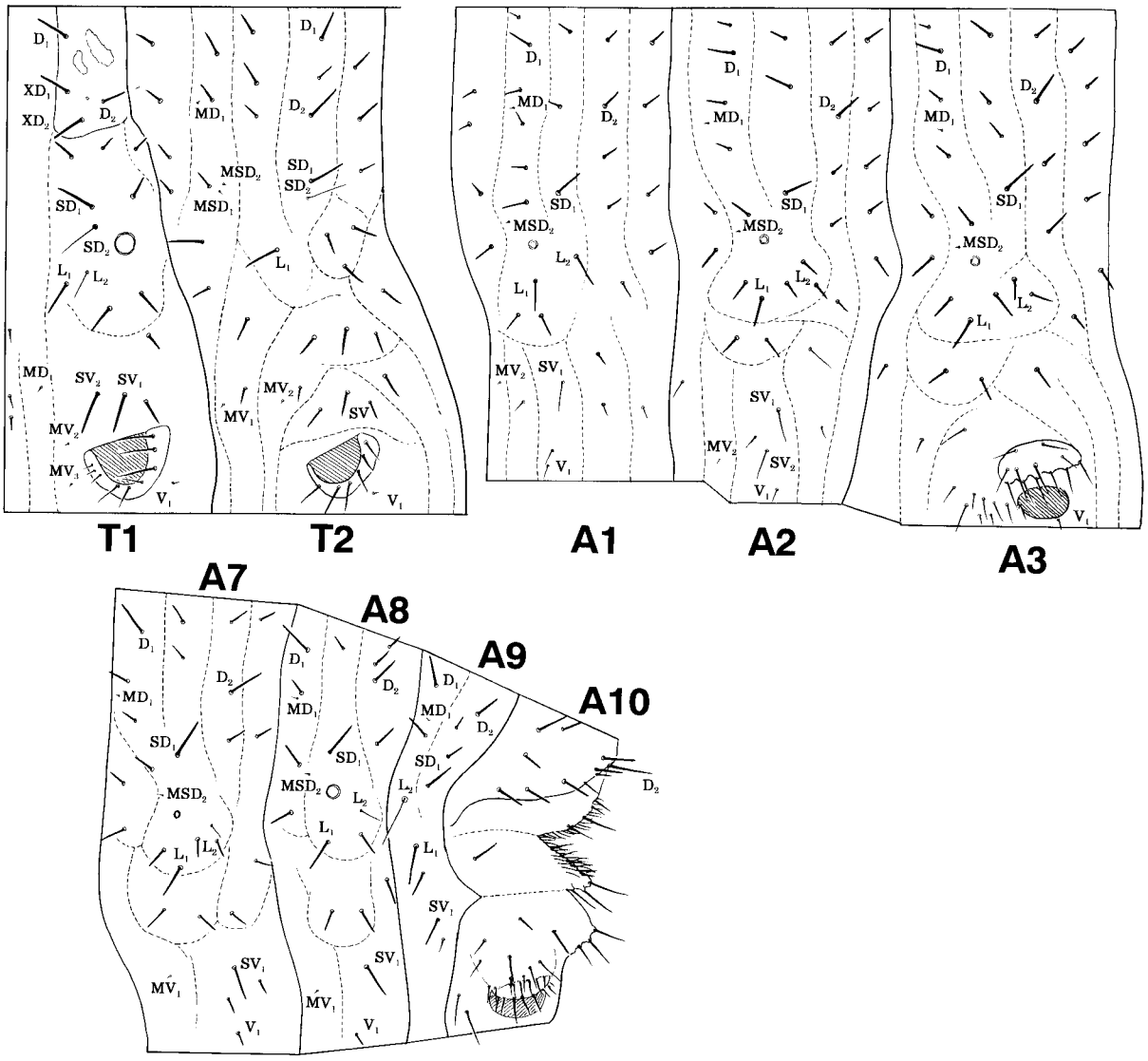
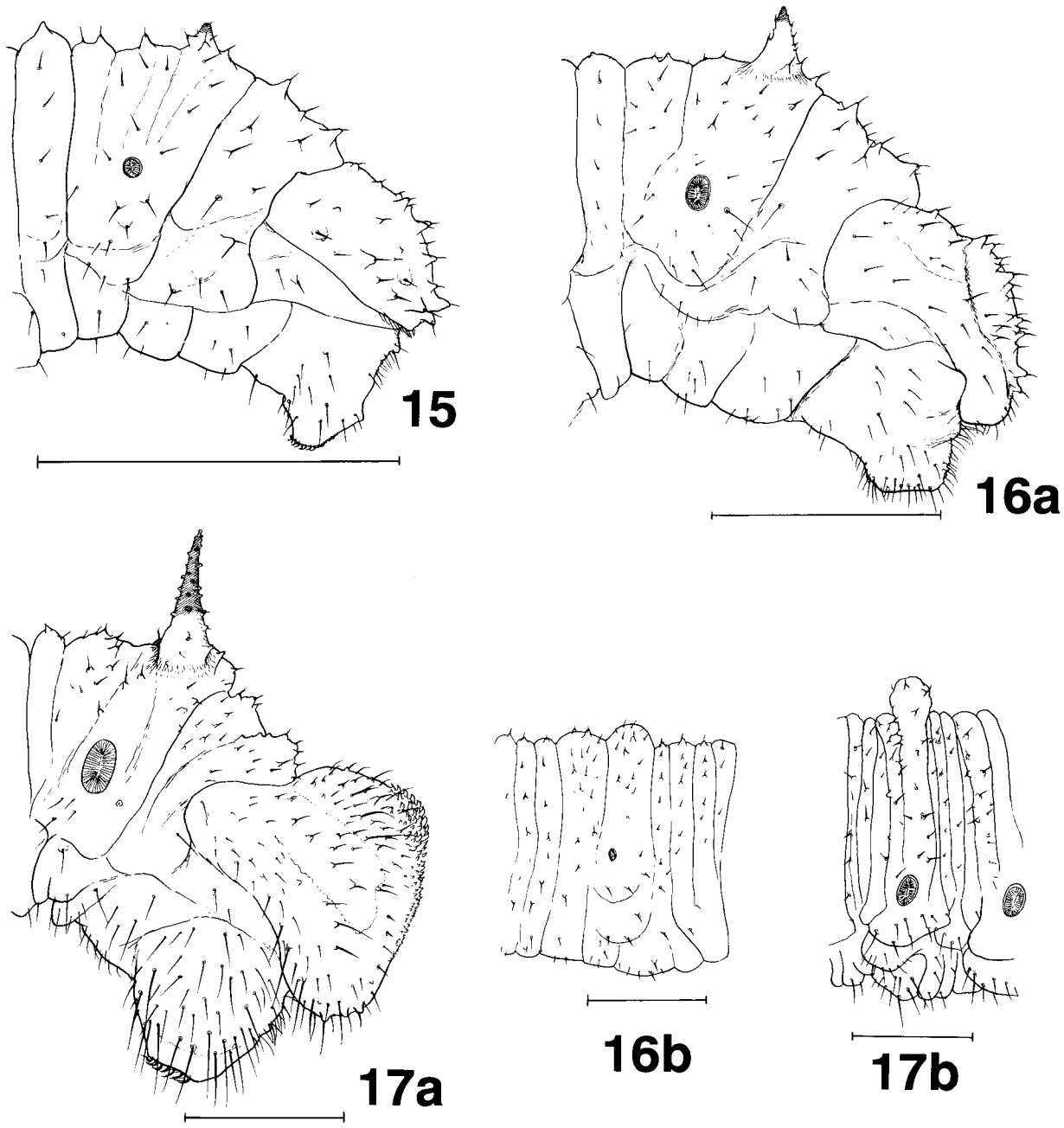


Fig. 14. Body chaetotaxy of second instar. T1: Prothorax. T2: Mesothorax. A1–A10: First to 10th abdominal segments.

triangle and the last posteroventral to the triangle); sometimes with a small seta ventral the level of the lowest L group seta; in A2 and A7, 1–2 setae between the longer SV and V setae; in A3 to A6 a long seta anterodorsal to proleg shield, which has many setae, and several setae of varying lengths in front of the proleg. A8 with furrows between ridges rather vague and spiracle large, nearly twice as large as that in A7 in diameter; 1st ridge with only 1 seta in front of the spiracle; 2nd ridge with 2 pairs of setae of which the mesal one is D1; 3rd ridge with 2 pairs of setae, the lowest one SD; 4th ridge with 3–4 pairs of setae, intermediate pair being D2, which is not so specialized as in the succeeding instars; 5th ridge without setae; second group of L of 3 setae; lower group of L bisetose; each one SV and V seta. A9 with setae similar to those of 1st instar; 3 short setae transversely arranged between D2 and SD, the latter long and much finer; each one seta below L and SV setae. A10 with dorsal area bearing 3 pairs of discal setae and 7 pairs of marginal setae; proleg shield bearing many short setae; each one SV and V as in 1st instar.

Third instar larva (Figs. 4, 10, 15). Body length 7.5–8.5 mm; width of head 1.2 mm. Colored as in second instar, except head tinged with brown. Very similar to second instar. Head with horns elongated, about 0.6 times as long as height of head; conical tubercles appearing in second instar longer and stouter, colored white; the tubercle near apex of horn bearing several short setae, other tubercles having only one seta at apex as in second instar. Secondary setae on both head and body increasing in number, with their basal tubercles somewhat elongated. T1 shield absent. Chaetotaxy of body similar



Figs. 15, 16a, 17a. Posterior abdominal segments, lateral aspect. 15: Third instar. 16a: Fourth instar. 17a: Fifth (last) instar. **Figs. 16b, 17b.** Second abdominal segment showing a dorsal protuberance, lateral aspect. 16b: Fourth insta. 17b: Fifth (last) instar. Scales 1mm.

to that of 2nd instar, but setae somewhat increased in number; A8 with D2 setae, each on a conical protuberance 0.7 × as high as its base.

Fourth instar larva (Figs. 5, 11, 16). Body length about 18 mm; width of head 1.8 mm. Head with long six-branched horns, one of the branches arising from posterior surface of horn; horn about 2.2 times as long as height of head, with numerous short setae on the shaft and branches. Head blackish brown, with frontoclypeus whitish; cranium marked with three pairs of vertical whitish bands in frontal view; two projections on each side of posterodorsal portion of cranium, of which the ventral one is transparent white, the dorsal one sometimes transparent white on its apical 2/3. Body dark green in ground color, but somewhat whitish ventral to the spiracular line; spiracles black, ringed by yellow; A1–A8 each with vague dark stripe extending from in front of spiracle, running obliquely dorsally to near the dark transverse dorsal band, the segments somewhat tinged blackish posteriorly to the spiracle. Chaetotaxy of body similar to that of 3rd instar, but number of setae somewhat increased; A2 with a weak dorsal swelling on the 3rd ridge; A8 with D2 group of setae each on a process 1.5 × as long as basally wide, and darkened apically; anal plate normal with several prominent

tubercles for the primary setae and several weaker sockets.

Fifth instar larva (Figs. 6, 12, 17). Body length about 45 mm; width of head 4.0 mm. The head horns become much longer, but its proportion to head height almost as in 4th instar. Head darker than in 4th instar, and with fine, white adfrontal sutures. Chaetotaxy of body similar to that of 4th instar, but setae increased in number; A2 with the dorsal protuberance on the 3rd ridge, which is slightly longer than basally thick, bluntly ended, and bearing a few short setae; A8 with D2 group each on an elongated process 3 × as long as basally thick, white on basal 1/3, black on apical 2/3, bearing many spinules; anal plate with a dorsomedian narrow band of dense black spinules, each of which consists of dorsally (anteriorly) curved, blunt-ended base and a short colorless apical spine.

Pupa (Figs. 7, 18). Length about 19.0 mm, width 9.7 mm, height 9.5 mm. Chartreuse green with outer ridge of bulge on forewing and dorsomedian portions of T1-A2 brown, and scattered with dark brown maculae on T2 and A2. A field collected pupa and one pupated in the breeding cylinder under artificial conditions, both from Guizhou Province, are pale brownish gray, and finely striped with brown as illustrated in Fig.7b. Head somehow projected forward on gena; labium recognized as a small elongated pentagonal sclerite between mandibles; labrum not clearly defined; proboscis (maxillae) and antennae extending to caudal margin of wings, and nearly reaching anterior margin of A5; bases of antennae closely approximating each other on dorsum of head; T1 and T2, particularly the latter, strongly projecting dorsally to make the strong keel, which distinctly extends backward on T2 along dorsomedian line. Pupa strongly expanded laterally in dorsal or ventral view due to strong bulges of forewings, but the bulge broadly excavated medially. A4 with weak lateral expansions under spiracles. Abdomen with a well-developed dorsal keel from A2 to A7; the keel produced into a enormously developed dorsal projection on A2, which extends forward and touches the dorsal keel of T2. A7 prolonged posteromedially on dorsum, very narrow on venter; A7 is the posteriormost segment which can be seen in the dorsal view. As the abdomen strongly bent ventrally beyond the 7th abdominal segment, A8 and the subsequent abdominal segments are located along the posterior margin of the 7th segment arranged dorso-ventrally just under the elongated dorsal part of the 7th segment. Consequently, the cremaster is just

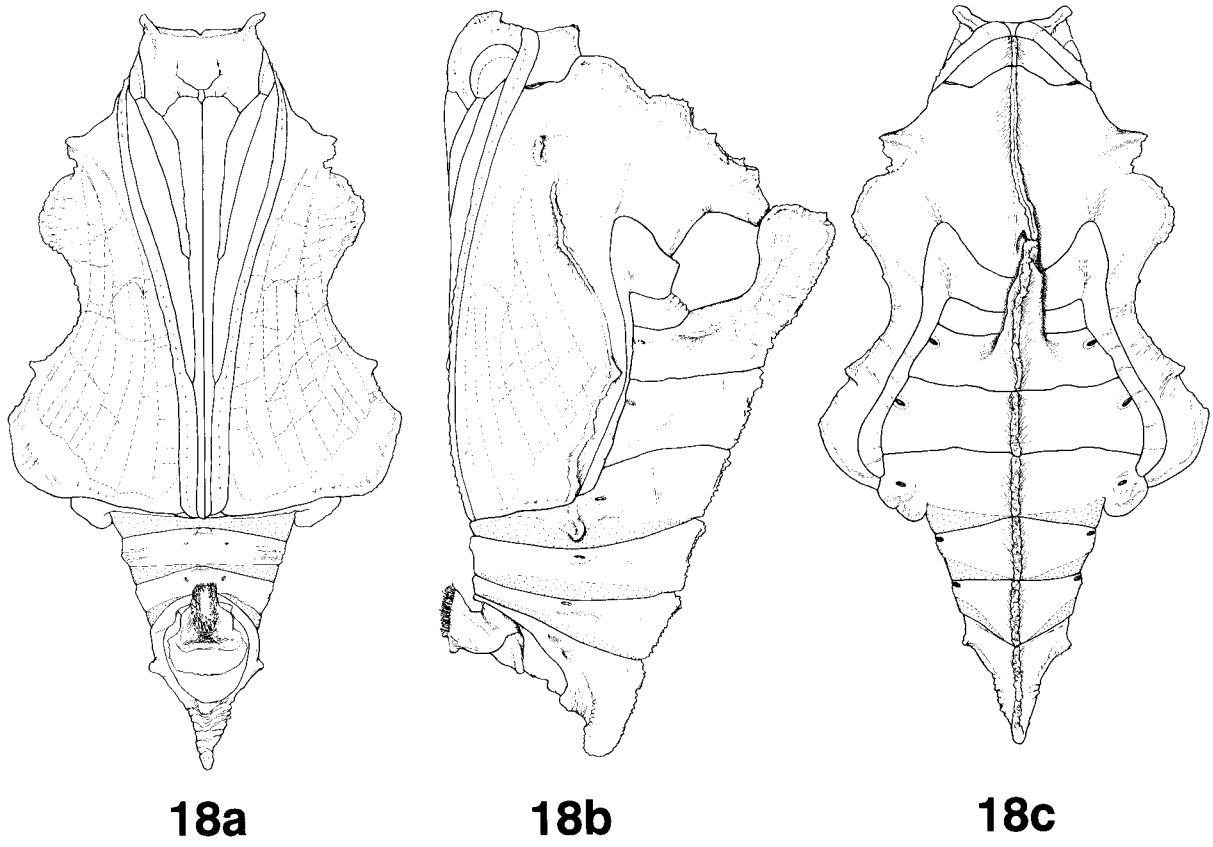


Fig. 18. Pupa. 18a: Ventral aspect. 18b: Lateral aspect. 18c: Dorsal aspect.

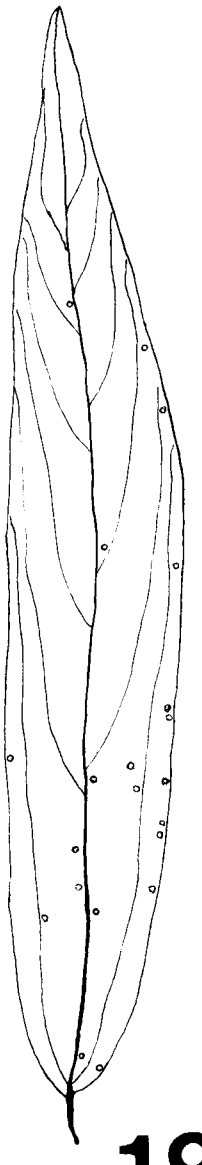
behind the 7th segment which is located parallel to the body axis.

Biological Notes

The eggs and larvae were found on leaves of *Debregeasia bicolor* Wedd. (Urticaceae), which seems to be the only host plant of this species in the surveyed areas.

In Gairi we observed one fast flying female alighted on the undersurface of a leaf of the host plant for oviposition. She deposited one egg on the same surface and then flew away. A similar ovipositing behavior was observed in another female at Lishon. She deposited several eggs singly on the undersurface of leaves of a host plant growing along the forest edge. Twenty-one eggs were discovered by checking several thousands of leaves of field-collected host plant in Gairi. They were all deposited on the undersurfaces of the leaves. Their distribution on the leaves is schematically shown in Fig. 19. Judging from this, the female may deposit eggs irregularly on a leaf, with a tendency of depositing along its edge.

Although we did not see larvae of this species sufficiently in the field, we are able to present some notes concerning larval behavior. The young larvae, such as the first, second and perhaps the third instars, begin to eat leaves from the edge judging from the trace of the eating left on the leaves of the host plant, on which the larvae stayed (Fig.20). The larvae of earlier stages, at least 1st and 2nd instars, con-



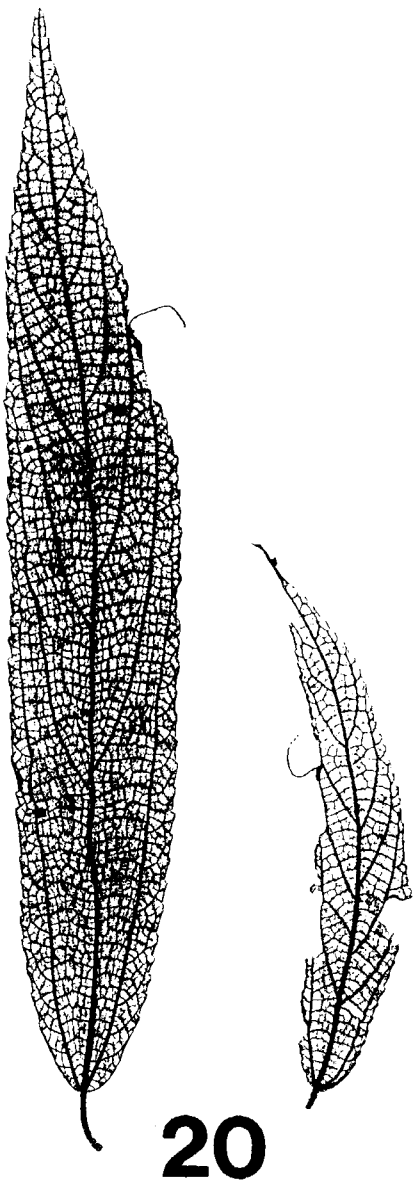


Fig. 19. Distribution of deposited eggs of *Pseudergolis wedah* (Koll.) on leaf of host plant. **Fig. 20.** Resting perches and traces of eating of *Pseudergolis wedah* (Koll.) on leaves of host plant.

struct a resting perch by spinning their own frass pellets at the edge of the leaf of the host plant. The perch is a slender and straight extension, at most 15 mm in length, and projects almost horizontal to the surface of the leaf. They usually stay on the perch, from which they move to the edge of the leaf to feed. In one case, a first instar larva stayed on the underside of the perch with its head towards the tip of the perch as shown in Fig. 2 clinging to the perch using all legs and prolegs. The middle to the last instar larvae remained on the midrib of the upper surface of a leaf directing their heads towards the petiole of the leaf.

In Gairi, we observed that adult males became very active towards evening, they appeared in a clearing by a brook, perched on a leaf of a bush with their wings open, and pursued other butterflies flying near their perching spots.

Discussion

The early stages of *Pseudergolis wedah* were reported for the first time in 1898 by Mackinnon and Nicéville, and later, Moore (1901-1903), Bingham (1905), and Stichel (1909) gave descriptions and drawings of larva and pupa as mentioned in the introduction. Although they did not state the exact instar of the larva they treated, we may safely infer that it was the last instar based on the descriptions and drawings in their papers. Watanabe (1996) provided a photograph of an early instar in the field, but the photograph is too small to judge its instar or to determine the detailed structure. Therefore, the first to 4th instars are described and illustrated in detail for the first time in the present paper.

In the first instar larva of *P. wedah*, the chaetotaxy is nearly the same as the basic pattern of the subfamily Nymphalinae (cf. Nakanishi, 1988), which is also widely seen in other subfamilies of the family Nymphalidae of a narow sense. It is difficult to show the conspicuous features of the first instar larva of this species. Accordingly we list the distinctive characteristics as follows.

1. The setae are very short, at most 0.14 times the height of the body segment.
2. The prothoracic shield is bilaterally divided into two sclerites.
3. The V1 seta is absent from the 9th abdominal segment.

For the last instar larva, the above-mentioned authors listed the following features as distinctive: the presence of a short fleshy dorsal pointed tubercle on the 5th segment (the 2nd abdominal segment) and two fleshy, sharp-pointed, erect, black spines on the 11th segment (the 8th abdominal segment). In the specimen we investigated, one low wart-like swelling was present instead of a fleshy pointed tubercle on the 2nd abdominal segment.

For the pupa, Moore (1901-1903) described "anal end with an upward obtuse protuberance" and Bingham (1905) described "anal end curved upwards". However, the protuberance stated by Moore is the posterior prolongation of the dorsal part of the 7th abdominal segment. Moore (1901-1903) stated "Pupa suspended by the tail, but in a horizontal position". This is not the usual manner in Nymphalidae. This corresponds with the peculiar apical abdominal structure of the pupa described above. In the three cases we observed in the breeding glass cylinder of 12 cm diameter and 18 cm high, the full-grown last instar larvae spun silk on the sides and not on the top and they hung on the silk by cremaster. In this case the pupa is in a vertical position just as Fig.9c of Pl.U of Mackinnon and Nicéville (1898). Therefore it may be a usual behavior that the pupa of *Pseudergolis wedah* suspends itself in a vertical position.

As described above there are two types of coloration in pupa, one is pale green and the other pale brownish gray. Only the green type had been known, but recently Watanabe (1996) illustrated a gray pupa. The brownish grey type is finely striped with dark brown as illustrated in Fig. 7b. We have no information about the causes of these different pupal colorations.

The habit to make a resting tower by young larvae is observed in a variety of nymphalid groups. *Cyrestis* and *Stibochiona* construct the extension by spinning frass pellets (Igarashi & Fukuda, 1996); *Dichorragia* constructs the extension with cut pieces of the host plant leaf and silk. Many limenitines, *e. g. Neptis* and *Limenitis*, construct the perch leaving the midrib of the host plant leaf when they cut it. In *Pseudergolis wedah*, the tower is constructed as in *Cyrestis* and *Stibochiona*. The tower is constructed at the tip of the leaf in most nymphalids. On the contrary, the tower-making behavior of this species is peculiar in that it is made on the straight lateral edge of the leaf.

The systematic position of the genus *Pseudergolis* within the Nymphalidae has not been decided. Eliot (1978) stressed the importance of the early stages in the systematics of family Nymphalidae and placed *Pseudergolis* in the subfamily Pseudergolinae together with the genera *Dichorragia, Amnosia* and *Stibochiona*, or in the tribe Pseudergolini of the subfamily Nymphalinae together with the same three genera (1992). According to him the combined taxon has the following combination of characters; larvae smooth or nearly so, with a pair of long horns on the head and a pair of upright spines or tubercles on the eighth

abdominal segment. In contrast, Harvey (1991) treated Eliot's Pseudergolinae as "a group of uncertrain status and subfamilial placement" and at the same time he transferred *Dichorragia* to Cyrestidini and *Amnosia* to Kallimini in the subfamily Nymphalinae based on the larval morphology.

Regarding the pupa, *Pseudergolis* and *Dichorragia* are very similar in having a long curved horn arising from the thorax and with one or a pair of distinct keels on the abdomen. In contrast, in the genera *Amnosia* and *Stibochiona*, the pupae are similar to that of usual nymphaline type (i.e. *Vanessa*) (Igarashi & Fukuda, 1997). In addition, the pupa of *Rhinopalpa polynice* is somewhat similar to that of *Dichorragia* (Igarashi & Fukuda, 1997). We only detected some resemblance of the pupa between *Pseudergolis* and *Dichorragia*, which may be a simple convergence, and could not summarize the pupal characters of Pseudergolinae.

The presence of long head horns and a pair of dorsal processes close to each other on the 8th abdominal segment in the later instars are common features shared by the genera assembled in the Pseudergolinae; but these characters are not unique to this subfamily. We could not compare detailed morphological characters of the early stages of *Pseudergolis* with those of its relatives, and consequently could not use these characters to discuss precisely the phylogenetic position of the subfamily.

However, we may safely say that *Pseudergolis* resembles *Dichorragia* based on the dorsal keel of the thorax in the pupa, and that *Pseudergolis* resembles *Stibociona* in the construction of its perching tower, and the branched head horns. The genera *Pseudergolis*, *Stibochiona* and *Dichorragia* show a transformation from branched head horns to simple head horns lacking the branches, and they share the apically clavate head horns.

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