CHAPTER - IV. NYMPHAL MORPHOLOGY

The study of the immature stages is not only interesting but also important for a complete investigation of the Zoocoenoses and it necessitates the determination of species of all the ascertained specimens. It is also important for the purpose of explaining the relationship and phylogeny of species and for determining the higher systematic categories.

In most species of the family Tingidae, the nymphal instars have striking morphological characters, on the basis of which it is possible to determine the instars of various species to a certain extent. It is quite often found to be difficult to determine the characters which are phylogenetically and adaptively significant. With the morphology of the nymphs of more number of species being brought to light, the problems of phylogeny and interrelationships will become better defined. With this aim in view, the various categories of cuticular and body outgrowths of the nymphal instars of about 25 species and the phenomenon of sweating in a few selected species of Tingidae have been morphologically and histologically investigated.

IV - 1. Materials and methods:

For morphological studies, the nymphal instars were treated in cellosolve for 2-3 hours and mounted in
polyvinyl-lactophenol. For histological studies, the nymphal instars were fixed in Bouin's fluid for 24 hrs. After several changes in 70% alcohol, the materials were processed following Peterfi's celloidin paraffin embedding method. Longitudinal and transverse sections of the entire nymphal instars were cut at 6 μ. The sections were stained in Heidenhain's Hematoxylin-eosin, dehydrated and then mounted in DPX.

IV-2. Body outgrowths:

All the five nymphal instars of all the species exhibit remarkable variety of cuticular and body outgrowths. The nomenclature employed here is after Livingstone (1962c). They are classified as cuticular outgrowths and outgrowths of the body wall. The former develop as process of the cuticle whereas the latter develop as the evagination of the body wall and contain the hypodermis which is continuous with the rest of the hypodermis of the body wall. They also carry a fine branch of the tracheole.

Distribution of the various body outgrowths of the different species has been represented in the Table 4.

IV-2-a. Cuticular outgrowths

a. Tiny excrescences (Fig. 18a):

They are very minute outgrowths of the cuticle and they form a uniform covering on the dorsal surface of
<table>
<thead>
<tr>
<th>S.No.</th>
<th>Type of outgrowths</th>
<th>Species</th>
<th>Distribution</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Cuticular outgrowths</td>
<td></td>
<td></td>
</tr>
<tr>
<td>1.</td>
<td>Tiny excrescences</td>
<td>Afrotogis phanuelii sp.nov., Ammianus ravanus, Cochlochila bullita,</td>
<td>Dorsum of the body except antenna and leg</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Dasytingis rudis, Dasytingis semota, Dictyla karnatica sp.nov.</td>
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<tr>
<td></td>
<td></td>
<td>Phaenotropis cleopatra, Urentius hystricellus</td>
<td></td>
</tr>
<tr>
<td>2.</td>
<td>Spherules</td>
<td>Abdastartus atrus, Phatnoma costalis</td>
<td>&quot;</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Agramma hupehanum, Agramma graminii sp.nov</td>
<td>&quot;</td>
</tr>
<tr>
<td>3.</td>
<td>Cuticular spines</td>
<td>Corythauma ayyari, Corythauma gibbossa sp.nov.</td>
<td>&quot;</td>
</tr>
<tr>
<td>4.</td>
<td>Biradiate scoli</td>
<td>Agramma graminii sp.nov., Agramma hupehanum</td>
<td>&quot;</td>
</tr>
<tr>
<td>5.</td>
<td>Triradiate scoli</td>
<td>Phatnoma costalis</td>
<td>&quot;</td>
</tr>
<tr>
<td>6.</td>
<td>Tetraradiate scoli</td>
<td>Afrotogis phanuelii sp.nov., Phatnoma castalis</td>
<td>&quot;</td>
</tr>
<tr>
<td>7.</td>
<td>Stellate type</td>
<td>Abdastartus atrus</td>
<td>&quot;</td>
</tr>
<tr>
<td>8.</td>
<td>Spatulate hairs</td>
<td>Abdastartus, Agramma, Corythauma gibbossa sp.nov. and Dulinius conchatus</td>
<td>Distipalmus</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>lateral, dorsal, abdominal tubercles, leg, antenna</td>
</tr>
<tr>
<td>9.</td>
<td>Nonglobulated spines</td>
<td>Agramma, Haedus, and all other species</td>
<td></td>
</tr>
<tr>
<td>E.No.</td>
<td>Types of Outgrowths</td>
<td>Species</td>
<td>Distribution</td>
</tr>
<tr>
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<td>-------------------------------------------------------------------------</td>
<td>-----------------------------------</td>
</tr>
<tr>
<td></td>
<td>Outgrowths of the body wall</td>
<td>Dulinius conchatus, cysteochila javensis, Cysteochila incolans, Dasytingis rudis, Pontanus puerilis, Lasiancantha ruellii sp. nov. Urentius hystricellus, U. euonymus, Teleonemia scrupulosa.</td>
<td>Dorsum of the body</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Dulinius conchatus, Corythauma ayyari, Lasiancantha spp. Tingis tomentosii, Cysteochila javensis, Pontanus puerilis, Dasytingis rudis, Urentius euonymus, U. hystricellus, Cochlochila bullita, Habrochila laeta</td>
<td>Dorsal and lateral abdominal tubercles</td>
</tr>
</tbody>
</table>
Fig. 18 - NYMPHAL OUTGROWTHS

(Magnification: a to r - 10 x 45 X, s & t -15 x10X)

Cuticular outgrowths

a - Tiny excrescences
b - Cuticular spherules
c - Biradiate scoli
d - Cuticular spines
e - Triradiate scoli
f - Tetraradiate scoli
g - Stellate hairs.

Body outgrowths

h - Dulinius conchatus
i - Cysteochila javensis
j - Cysteochila incolana
k - Dasytingis rudis
l - Pontanus puerilis
m - Lasiacantha ruellii sp.nov.
n - Urentius hystricellus
o - Urentius euonymus
p - Physatocheila asiatica sp.nov.
q - Teleonemia scrupulosa
r - Tingis tomentosii sp.nov.
s - Haedus yacobii sp.nov.
t - Haedus manii sp. nov.
FIG 10 - NYMPHAL OUTGROWTHS

CUTICULAR OUTGROWTHS

a
b
c
d

f

BODY OUTGROWTHS

h
i
j
k

l
m
n
o
p
q
r

s
t
PLATE - XV - CUTICULAR AND BODY OUTGROWTHS

(Magnification - 15 x 10 X)

Fig. 1 Phatnoma costalis

2 Abdastartus atrus

3 Agramma graminii sp.nov.

4 Phaenotropis cleopatra

5 Aconchus urbanus

6 Haedus yicobii sp.nov.

7 Haedus manii sp.nov.

8 Haedus grewii sp.nov.

9 Perissonemia ecmeles

10 Stephanitis typica

11 Stephanitia charies

12 Stephanitis macranthaii sp.nov.
the body and these types of cuticular outgrowths are absent in the antennal and leg segments.

b. **Spherules or circular structures** (Fig. 18,b):

These structures are present on the dorsal surface of the body of the *Abdastartus atrus* and *Agramma graminii* sp.nov.

c. **Scoli** (Fig. 18, c,e,f & g)

These are very minute outgrowths of the cuticle. They form an uniform covering on the dorsal surface of the body and they are absent on the antennal and leg segments. The scoli are the open tubes and they do not carry any secretory globules terminally. Structural variations are seen among scoli. They may be biradiate, triradiate, tetraradiate or stellate. The distribution of these scoli in different species has been indicated in Table-4.

d. **Cuticular spines** (Fig. 18,d)

These are the spinous outgrowth of the cuticle which are present on the dorsal surface of the body surface of the few species.

e. **Spatulate hairs** (Plate XVII, Fig.9 & 10, SPH):

They are restricted only to the ventral surface of the distitarsus and are present only in very few species viz., *Agramma graminii* sp.nov, *Agramma hupehanum*, *Corythauma gibbossa* sp.nov. and *Dulinius conchatus*. The functional significance is not known.
PLATE - XVI - CUTICULAR AND BODY OUTGROWTHS

(Magnification - 15 x 10 X)

Fig. 1  *Afrotingis phanuelii* sp.nov.
2  *Eteoneus cinchonii* sp.nov.
3  *Physatocheila asiatica* sp.nov.
4  *Dulinius conchatus*
5  *Corythauma ayyari*
6  *Corythauma gibbossa* sp.nov.
7  *Lasiacantha ruellii* sp.nov.
8  *Tingis tomentosii* sp.nov.
9  *Cysteochila javensis*
10  *Pontanus puerilis*
11  *Teleonemia scrupulosa*
12  *Dasytingis semota*
f. Non-globulated spines:

There is a wide range of structural variations in the non-globulated spines found in the different species. They may be short and long spines; long and blunt spines or setaceous spines. These types of spines are present in the tip of the antennal segment and the leg.

IV-2-b. Outgrowths of the body wall:

1. Open tubes (Fig. 18, h-q).

These develop as the evagination of the body wall and contain hypodermis of the body wall. As they are found in larger number they are mainly responsible for the extrusion of secretion and they are distributed on the dorsal surface of the body, all over. Structural variations are seen among these tubes.

2. Simple globulated spines (Fig. 18, r):

They are simple spines, each borne on a base or pedicel and terminally expanded in the form of an open cup that often contains the secretory globules and hence the name globulated spines. They are distributed on the antennal segments and on the dorsal surface of the body. They invariably arise in succession and increase in length in the successive stages.
PLATE - XVII - CUTICULAR AND BODY OUTGROWTHS

(Magnification - Figs. 1 to 8 - 15 x 10 X; Figs. 9 & 10 - 10x 40X)

Fig. 1 Urentius euonymus

2 Urentius hystricellus

3 Dictyla karnatica sp.nov.

4 Cochlochila bullita

5 Naochila nigra sp.nov.

6 Naochila minuta sp.nov.

7 Habrochila laeta

8 Haedus ruthii sp.nov.

9 Abdastartus atrus

10 Corythauma gibbossa sp.nov.
Fig. 19 - NYMPHAL OUTGROWTHS
(Magnification - 15 x 10\'X)

a - Pontanus puerilis
b - Tingis tomentosii sp.nov.
c - Dasytingis rudis
d - Physatocheila asiatica sp.nov.
e - Eteoneus cinchonii sp.nov. lateral abdominal tubercle
f - Eteoneus cinchonii sp.nov. dorsal abdominal tubercle.
g - Naohila minuta sp.nov.
h - Naohila nigra sp.nov.
i - Stephanitis charies
j - Stephanitis macranthii sp.nov.
k - Stephanitis cinnamomii sp.nov.
l - Dulinius conchatus
m - Stephanitis typica
n - Habrochila laeta
o - Cochlochila bullita
p - Cochlochila bullita
q - Perissonemia ecmeles lateral abdominal tubercle.
r - Perissonemia ecmeles dorsal abdominal tubercle.
s - Urentius hystricellus
t - Urentius euonymus
u - Teleonemia scrupulosa
v - Teleonemia scrupulosa
w - Phaenotropis cleopatra
x - Agramma gramini sp.nov.
y - Dictyla karnatica sp.nov.
z - Lasiacantha ruellii sp.nov.
FIG. 19 - NYMPHAL OUTGROWTHS

BODY OUTGROWTHS - TUBERCLES
3. Tubercles (Fig. 19, a - y):

Tubercles are the typical evaginations of the body wall. All the prominent outgrowths of the body surface are the tubercles. The base of each tubercles is invariably padded. On the basis of the complexity of their development in the successive stages of the nymphal instars, the tubercles are classified as simple tubercles and compound tubercles, the former differ from the latter in having a smooth and simple base which may be slightly padded. In Phatnoma costalis and Abdastartus atrus the body is smooth without any tuberculate outgrowth (Plate XV, Figs. 1 & 2). All the tubercles of the first instars are simple tubercles. In Agramma graminii sp. nov. Phaenotropis cleopatra and in Aconchus urbanus, the fifth instar nymph possess the simple tubercle (Plate XV, Figs. 3, 4 & 5).

A compound tubercle bears one to several prominent globulated spines, each carrying in its cup fine granules or spherules of secretory material that becomes viscous as the instar advances in age. In Dictyla sp. the compound tubercles are very much reduced (Plate XVII, Fig. 3). The increase in the complexity of the tubercles in the successive stages is brought about by the addition of more and more globulated spines usually in the form of whorls. (Plate XV, Fig. 9, Plate XVI, Figs. 1, 2, 3, 4, 5, 6, 8, 9 & 11; Plate XVII, Figs. 1 & 2). In Dasytingis rudis,
in the members of the genus Lasiacantha, and on the basis of the order of their appearance, the tubercles of the thoracic and abdominal margins are designated as primaries, secondaries, tertiaries, and quartnaries and so on. The primaries always mark the posterolateral margin of the each segment. All the other subsequent tubercles arise anterior to the primaries. In the members of the genus Stephanitis and Dulinius conchatus the increase in the complexity of the tubercle is brought about by the addition of spines on the tubercle without any whorls (Plate XV, Figs. 10, 11 & 12 & Plate XVI, Fig. 4) In the case of Habrochila laeta (plate XVII, Fig. 7) there is a special category of tubercle designated here as capitulate type in which the compound tubercle has a swollen head that bears circlets of short spines through which the secretion exudes.

The extreme development of compound tubercle has been found in the genus Haedus. Here the nymphs donot manifest any form of secretory globules the tubercles are enormously elongated and most arborescent (Plate XV, Figs. 6, 7, 8 & Fig. 18, s & t). The compound tubercle of Nacchila & Cochlochila resemble each other in having a minaret shape. Terminally the tubercle expands into an ornamented tower that carries the secretory globule. It is probable that the capitulate tubercles of Habrochila laeta is a better specialized
form of minaret type of Cochlochila bullita, Naochila minuta and Naochila nigra (Plate XVII, Fig. 4, 5, 6, & Figs 19-g, h & p)

Considerable translocation of tubercles of the pterothoracic segments has taken place in the first instar nymph due to the extension of the wing pads up to the posterior margin of the fourth abdominal segment. Translocation of the two simple tubercles of the metathorax and the primaries of the second abdominal segment takes place posterior to the primaries of the mesothorax and this kind of translocation appears to be an universal phenomenon in Tingidae.

In Dasytingis rudis, D. semota and in Pontanus puerilis, the primaries of the metathorax never increase in size and complexity in the successive stages, unlike those of the other segments. The primaries of the third abdominal segments, that remained in their entire complexity in the fourth instar nymph are totally lost in the fifth instar nymph. Until the fourth instar stage all the eight abdominal segments possess the lateral marginal tubercles, but in the last instar nymph, only six segments possess them, the tubercles of the second segment being deleted as reported by (Livingstone, 1976) & Livingstone et al (1981a). Similar translocation of marginal tubercles has been reported in several other species of Tingidae (Stusak & Stys, 1959, Livingstone, 1968c) but deletion of this type has escaped the observations of earlier workers.
PLATE - XVIII

(V - instar nymphs showing the secretory globules 10 x 15 X )

Fig. 1 Cochlochila bullita

2 Habrochila laeta

3 Corythauma ayyari

4 Dulinius conchatus

5 Pontanus puerilis

6 Stephanitis typical

7 Urentius hystricellus

8 Teleonemia scrupulosa

9 Haedus yacobii sp.nov.
   Notice the nonsecretory nature of the tubercles

GL - Globule
IV-3. Histology of globule secretion

The globulated spines and the open tubes are characteristically developed in the nymphal instars of Tingidae. The globulated spines are so named because they bear 'waxy' rather sticky exudation at their tip. This exudation appears as shining globule and is present in different sizes, depending on the nature of the outgrowth that bears it. The exudation progressively increases in size and gradually becomes viscous as the nymph advances in age (Plate XVIII, Figs. 1 - 8).

Sections of the various nymphal instars of Tingidae reveal prominent display of oenocytes just beneath the hypodermis. In Agramma spp. (Plate XIX, Fig. 1) which do not possess the globulated spines, the sweating mechanism is totally absent and the oenocytes in this insect occupy the regions just beneath the anterior and ventrolateral margins of the abdominal tergites. In the sections of the nymphal instars of summer species such as Urentius hystricellus, Cochlochila bullita, Habrochila laeta, Corythauma ayyari, Dictylakarnatica & in Lasiacantha ruellii sp.nov. in addition to these oenocytes there are, certain isolated spherical as well as sub spherical cells scattered amidst oenocytes beneath the hypodermis (Plate XIX, Fig. 6, Plate XX, Figs. 7 & 9). These cells are here described as globule secreting cells (GC). The distribution and number of these cells beneath the hypodermis vary according to the type and size of the
PLATE - XIX - HISTOLOGY OF SECRETORY GLOBULES

(Magnification - 10 x 40 X)

Fig. 1 *Agramma graminii* sp.nov.- Notice the nonsecretary nature of spines and absence of globule secreting cells.

2 *Cochlochila bullita* - Sagittal section (SS) through pronotal evagination of the V-Instar nymph showing secreting cell at the base of globule body tubercle.

3 *Cochlochila bullita* - S.S. through the abdominal region of the V-Instar nymph showing globule secreting cells densely packed with secretion.

4 *Cochlochila bullita* - Sagittal section through the cephalic tubercle of the V-Instar nymph showing globule secreting cell at the base.

5 *Cochlochila bullita* - Sagittal section through the dorsal abdominal tubercle of the V-Instar nymph (newly emerged) showing three prominent globule secreting cells (non secreting phase).

6 *Cochlochila bullita* - C.S. through the dorsal abdominal region of the V-Instar nymph, showing the single actively secreting cell surrounded by the group of oenocytes.

7 *Cochlochila bullita* - C.S. through the third abdominal segment of the V-Instar nymph showing the globule secreting cells and the oenocytes in the fore wing pad.

8 *Habrochila laeta* - C.S. of the V-Instar nymph showing the globule secreting cell in the fore wing pad and the secretary cells of the tubercle head.

9 *Habrochila laeta* - Enlarged view of the tubercle head showing the arrangement of globule secreting cells beneath each globulated spine (15 x 40 X).

FW - Fore wing pad; GC - Globule secreting cell;

HW - Hind wing pad; OE - Oenocyte; TH - Thorax.
body outgrowths. Unlike the oenocytes, these globule secreting cells are restricted only to the nympha! instars of the species that manifest the phenomenon of 'sweating'. The globule secreting cells differ markedly from the oenocytes in their histology and distribution and often found to be associated with the latter.

The size and the shape of the globule secreting cells vary, depending on the secretory activity (0.032 mm to 0.05 mm). Immediately after emergence, these cells are spherical and clear, the cytoplasm without any trace of secretory granules. The body outgrowths of this stage donot present any evidence of "sweating". As the nymph starts feeding, the globule secreting cells present a rather dense and opaque picture. The nucleus is hard to differentiate from the cytoplasm, the latter being fully packed with strongly eosinophilic granules (Plate XIX, Fig.5). The discharged secretion in the form of fine eosinophilic granules accumulates in the subcuticular space, at the base of the body outgrowths. As they accumulate, they are gradually ooed out through the tubercular outgrowths and accumulate at their tips in the form of shining globules, steadily increasing in size and viscosity, giving a real picture of a "sweating" phenomenon. Such "sweating" phenomenon is never seen in any freshly emerged nymphs of summer forms or any of the nymphs of the winter forms, or adult of any form.

Globule secreting cells are distributed rather uniformly on the dorsum, sub hypodermally. Unlike the
PLATE - XX - HISTOLOGY OF SECRETORY GLOBULES

(Magnification - 10 x 40 X)

Fig. 1 Corythauma ayyari - Globule secreting cell at the base of the spine on the fore wing pad.

2 Dictyla karnatica sp.nov. - C.S. through the pronotal anterior evagination hood of the V Instar nymph showing a single large globule secreting cell.

3 Dictyla karnatica sp.nov. - C.S. through the abdominal segment of the V-Instar nymph showing the globule secreting cells in the fore wing pad.

4 Lasiacantha ruellii sp.nov - L.S. through the cephalic tubercle of the V-Instar nymph showing the globule secreting cells at the base of the globulated spines.

5 Teleonemia scrupulosa - Sagittal section through abdominal segment of the V-Instar nymph showing the distribution of oenocytes.

6 Teleonemia scrupulosa - C.S. through the 3rd abdominal segment of the V-Instar nymph showing the single large globule secreting cell.

7 Teleonemia scrupulosa - C.S. through the abdominal region showing the large globule secreting cell surrounded by group of oenocytes.

8 Urentius hystricellus - Sagittal section through the median dorsal tubercles of V-Instar nymph showing globule secreting cell at the base of the globulated spines.

9 Urentius hystricellus - Sagittal section through the abdominal region of the V-Instar nymph showing the globule secreting cells and the oenocytes.

AS - Abdominal segment; FW - Fore wing pad
GC - Globule secreting cell; HW - Hind wing pad;
OE - Oenocytes.
oenocytes, the globule secreting cells occupy areas of the head, thorax, prothoracic anterior, lateral and posterior evaginations and are embedded in the upper layer of the hypodermis of the forewing pad and other integumentary evaginations (Plate XIX, Figs. 2, 7 & 8, Plate XX. Figs. 2, 3 & 6). However, they are totally absent beneath the hypodermis of the sternum and the hindwing pad. The number and disposition of these cells depend on the nature and type of the body outgrowths to which they are associated. Larger and more complex tubercles will always have more than one actively secreting cells (Plate XIX, Fig. 3, 4 & 5) whereas the smaller porous spines invariably will have one cell (Plate XX, Figs. 1, 4 & 8).

Sections of Habrochila laeta, exhibit a different in being pattern of distribution of globule secreting cells/arranged one beneath each spine of the capitulum of the tubercles. So the secretion of the globule at the tip of the tubercle head is the accumulation of the secretion of the individual globule secreting cell exuded through the respective spine (Plate XIX, Figs. 8 & 9).

The arrangement of the oenocytes vary from the arrangement of the globule secreting cell. They form sheets of closely associated cells, with or without dense cytoplasm, occupying regions just beneath the anterior and venterolateral margins of the abdominal tergites. They are never found at
any region anterior to the abdomen as reported by Livingstone (1978) in Tingidae and by Kramer and Wigglesworth (1959) in *Periplaneta americana*. A similar arrangement of oenocytes has been seen in *Teleonemia scrupulosa* (Plate XX, Fig. 5).

Nymphs of summer species of Tingidae, by their elaborate development of body outgrowths and their associated secretory cells, have thus evolved a special mechanism for the elimination of water and other non-essential products.

It is interesting that on the basis of the "sweating" phenomenon, Livingstone (1976) included *Stephanitis typica* under winter forms that do not "sweat". Examination of the nymphal instars of all the species of *Stephanitis* recorded in Southern India show fine, relatively smaller secretory globules suggesting that the development of this phenomenon is related to the prevailing climatic conditions. In Southern India where winter and summer are not well defined in a scrub jungle ecosystem, more number of species are recorded here to be "sweating" and the non "sweating" *Haedos* species (Plate XVIII, Fig. 9) are definitely from the tropical rainforests. Further examination of the life stages of more number of species will provide confirmatory evidences for this phenomenon.