exterior in larval Tenthredinoidea, he does not give details of structure which are required to develop this idea.

Serial sections of larvae about 3 mm long (presumably in their second instar) show that the hypo-pleural organ is already present at this early stage. Parkin (1942) was unable to find these organs either in sections or dissections until the larvae were \frac{1}{4} to \frac{1}{3} grown. He realised that the size of the organ and the number of pits forming it increased as the larva grew. The small larvae of S. noctilio showed only from 8-10 cavities (Fig. 6a). Moreover, the openings to the cavities are wider in proportion to their depth in the small larvae than they are in the older larvae. (Fig. 6b). No cultures were made from the hypopleural organs of these small larvae, but the contents took up the fungal stain Methyl Green.

#### Layers in the Cuticle.

The layers in the insect cuticle are clearly differentiated by Lower's Trichrome stain. The extremely thin epicuticle, when visible, is stained red, the exocuticle becomes pale yellow, the meso-cuticle turns orange and the endo-cuticle is coloured green. The cellular hypodermis, with orange nuclei is stained pale purple.

In sections of the hypo-pleural organ in the inter-moult condition prepared for examination under the light microscope and stained with Lower's Trichrome, the epicuticle could not be detected. The exocuticle is absent from the cuticle of the hypo-pleural organ except for the tips of the spines. According to Lower (1964) the exocuticle is absent from the cuticle of most immature insects.

The meso-cuticle forming the bases of the spines and the outer visible layer of the cuticle stained strongly with Orange G.

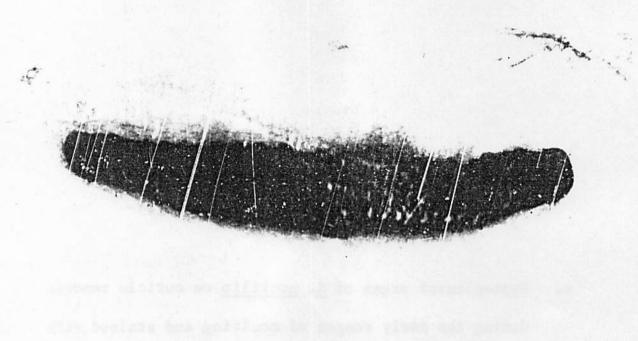
The green endo-cuticle is the innermost layer of the cuticle. In the region of the hypo-pleural organ the purplish eals of the hypodermis are elongated, spindle-shaped. On histological grounds, it is likely that they are secretory cells.

#### Moulting.

From the description of the cuticle which reveals that there is no tanned or sclerotised layer (exocuticle) in the integument of the larva, it is to be expected that the pits of the hypoplaural organ will collapse as the supporting endocuticle is digested away by the moulting fluid. Serial sections of a moulting pre-pupa show that the endocuticle, both within the septa of the hypo-pleural organ and in the cuticle generally, has been digested. Only the orange meso-cuticle remains. It is also evident from these sections that, as the sides of the septa have shrunk together, the external openings of the pits of the hypo-pleural organ have become wider (Fig. 6c).

# Figure 7.

- a. Hypo-pleural organ of S. noctilio on cuticle removed during the early stages of moulting and stained with Aniline Blue. Mag. 115x.
- b. Seven coiled bundles found in the region of the hypopleural organ on a cast larval skin. Mag. 140x.



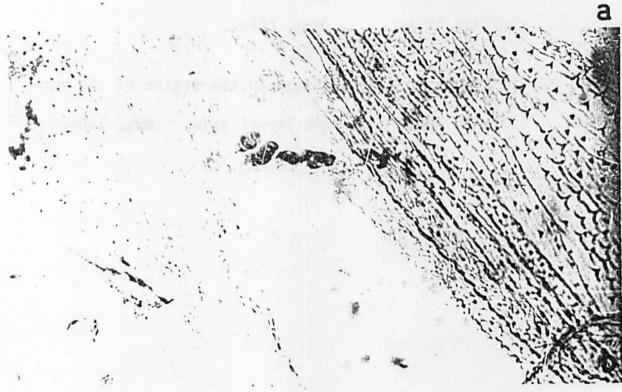


FIG. 7

#### Figure 8.

- e. Glistening ridge of wax packets on the moist prepupal exuviae of S. noctilio. Mag. 72x.
- b. The wrinkled remains of the hypo-pleural organ after
  the wax packets have sloughed off the pre-pupal exuviae.
  Two terminal pits are clearly defined. Mag. 56x.
- c. Three terminal cavities is all that remains of the hypo-pleural organ on a cast pre-pupal exuviae. Mag. 400x.
- d. A group of seven wax packets sloughed off the prepupal exuviae of <u>S</u>. noctilio and viewed end-on. Mag. 200x.

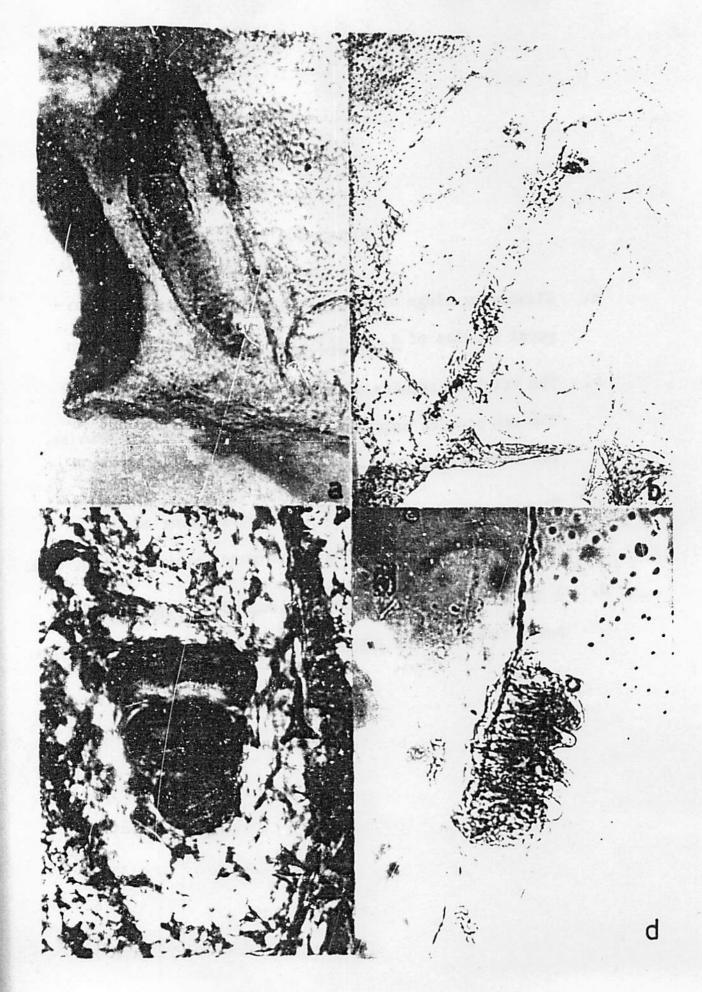


FIG. 8

Parkin (1942) first speculated about the changes which would take place in the hypo-pleural organ during moulting. He assumed that the cuticular pits which contain the fungus would be shed during a larval/larval moult, in this way breaking the association between the larve and the fungus at each instar. Observations were made during this study on a moulting larva preserved in formalin. These showed that his assumption was correct. In this larva the old cuticle had become detached in the region of the head, but had not yet ruptured, and was distended like a balloon. After excising one of the hypo-pleural organs it became apparent that the old cuticle could simply be lifted off. Fig. 7a shows this piece of cuticle with the pits of the hypo-pleural organ stil clearly defined but without any surrounding glandular issue (Fig. 3b). The darkened coils of fungus are visible within the pits. It is suggested that this hypo-pleural organ had not collapsed because the moulting process was incomplete and therefore the septa of the pits were still supported by endocuticle.

On the soft cuticle of the new exceskeleton, the shallow cavities of the new organ could be seen. Measurements taken under high magnification showed that the new organ was slightly longer than the old one. The cavities appeared free of fungus and attempts to stain any contents with the fungal stain Aniline Blue were unsuccessful.

# Figure 9.

Three wax packets of S. noctilio viewed from the side to show the scalloped base with a thick coat of wax, and the flat top edge formed at the surface of the pit. Mag. 650x.

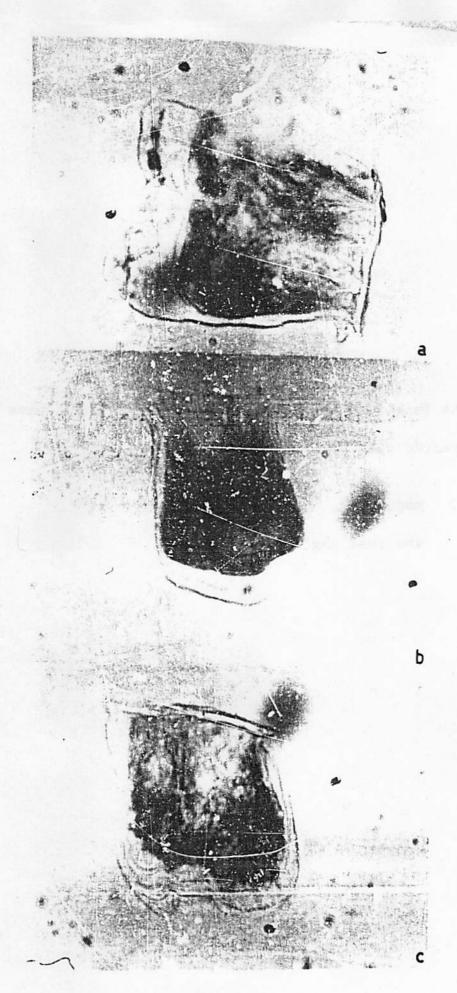


FIG. 9

Figure 10.

The fungal contents of the wax packets which have been treated with wax solvents.

the rest mag. 1,000x.

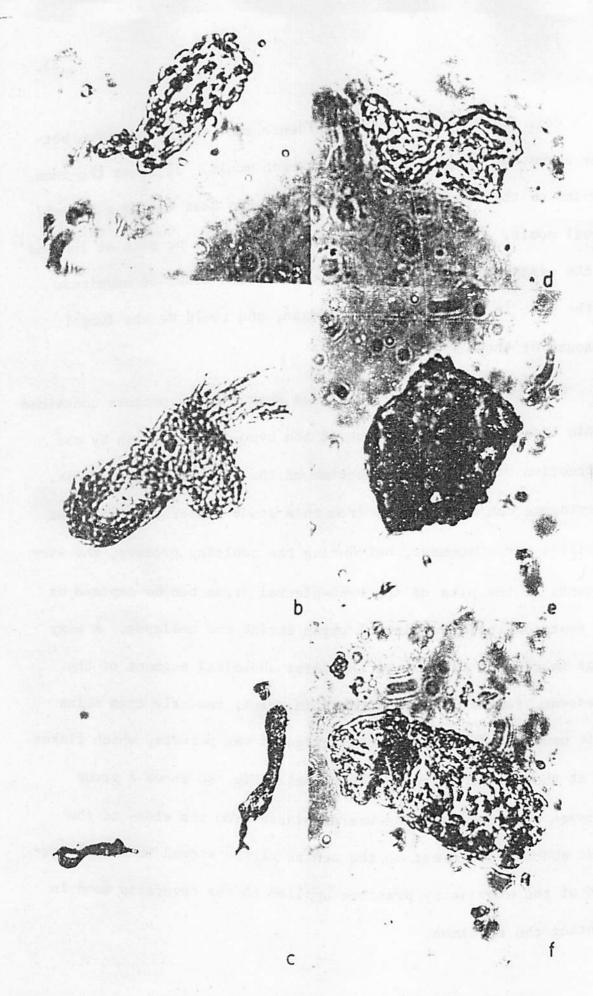


FIG. 10

rig. 7b presents further evidence that the association between larva and fungus is broken at each moult. It shows the torn remains of the hypo-pleural organ on a skin cast during a larval/larval moult. Seven tightly coiled bundles can be seen at one end of the organ. These bundles are of the same order of magnitude as the pits in the hypo-pleural organ, and could be the fungal contents of the organ.

Francke-Grosmann (1957) states that the wax packets contained within the pits are forced out of the hypo-pleural organ by the contraction of muscles at the time of the pre-pupal/pupal moult. No evidence can be produced from this study either to confirm or criticise her statement, but during the moulting process, the waxy contents of the pits of the hypo-pleural organ become exposed as the septa of the hypo-pleural organ shrink and collapse. A waxy ridge is clearly visible on the first abdominal segment of the quiescent, female pre-pupa. Only in moist, recently cast skins is it usual to find intact this ridge of wax packets, which flakes off at the slightest touch. (Fig. 8a). Fig. 8d shows a group of seven wax packets which was displaced from the sides of the first abdominal segment to the centre of the second abdominal segment of the exuvise by pressure applied to the coverslip ased in mounting the specimen.

#### (c) Wax Packets.

- 1. Examinations of cast skins show that an elongate, elliptical area of short parallel wrinkles is often all that remains of the septa of the hypo-pleural organ. Sometimes a few pits near the tip of the organ are still clearly defined on the cast skin. (Fig. 8b & 8c).
- 2. Under high magnification the outer layer of the wax packets appears to be stratified (Fig. 8d and Fig. 9c). There are two possible interpretations of this observation. Either the stratifications represent numerous scales which will flake off the packets or they represent the progressive but discontinuous deposition of wax around a bundle of fungus.
- 3. Examinations of numerous wax packets, stained, unstained and after treatment with wax solvents, provide evidence to substantiate the second interpretation. (See Appendix I and Figure 10).

### (ii) Female Reproductive System.

Francke-Grosmann (1939) described the comparative morphology of the ovipositors of X. spectrum, U. gigas and S. juvencus and illustrated the reproductive system and the inter-segmental sac of S. juvencus. The same general description applies to the morphology and anatomy of these structures in S. noctilio. However, a more precise knowledge of the structure of the ovipositor and accessory glands is required for an appreciation of later discussions regard-

#### Figure 11.

#### Ventral view of ovipositor of S. noctilio adult.

is inter-segmental sacs

cg club gland

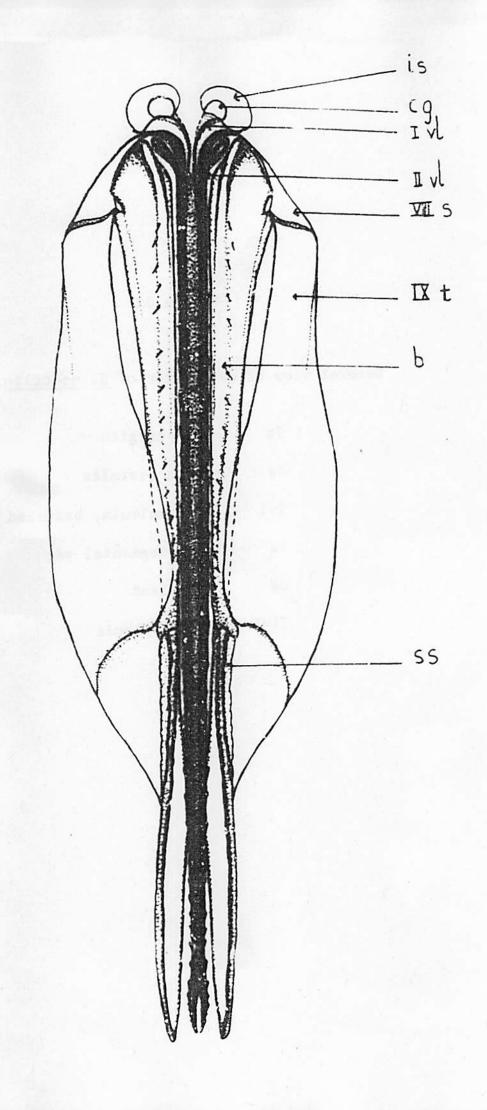
Ivl first valvula

VIIIs eighth stermite (triangular plate)

IXt ninth tergite (quadrate plate)

b base of sheath (oblong plate)

38 saw-sheath



# Figure 12.

# Lateral view of ovipositor of S. noctilio.

9t ninth tergite

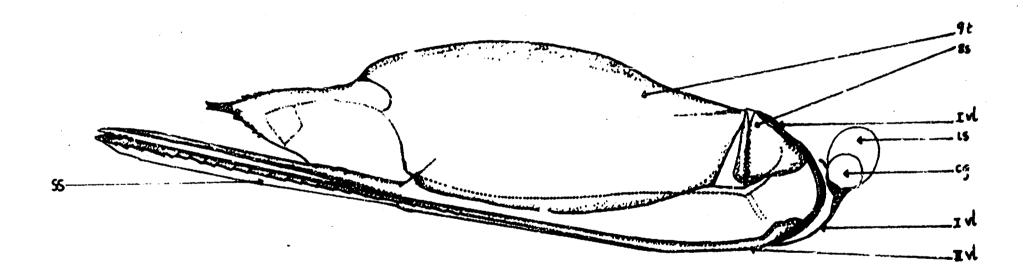
8s oighth stermite

Ivl first valvula, branched

is inter-segmental sac

cg club gland

Ilvl second valvula



4- 1

# Figure 13.

The first valvula of the ovipositor of S. noctilio.

VIII st eight stermite

is inter-segmental sac

cg club gland

#### Figure 14.

Section of the club gland and inter-segmental sac of a pupa of <u>S. noctilio</u>, showing the unicellular glands with stout ducts opening into the sac.



FIG. 13

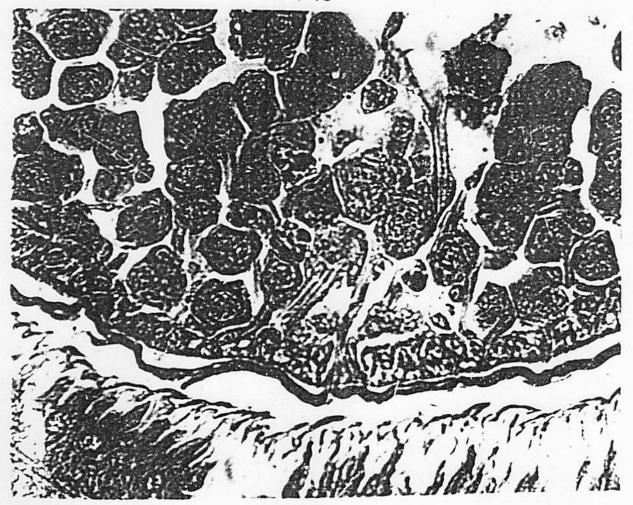


FIG. 14

# Figure 15.

# Ventral view of bases of first and second valvulas.

is inter-segmental sac

cg club gland

Ivl first valvula

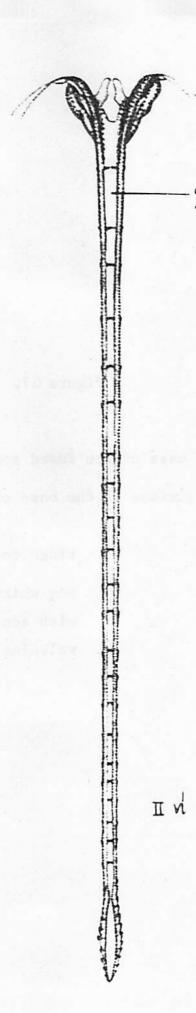
Ilvl second valvula

VIIIst eighth stermite

Figure 16.

Ventral view of fused second pair of valvulae of S. noctilio.

g ventral groove



#### Figure 17.

Dorsal view of base of the fused second pair of valvulae and the inner surface of the base of the sheath.

- b ridge on sheath base
- p peg which articulates with second pair of valvulae.

