Intracellular development of the cotton-rat filaria *Litomosoides carinii* in the vector mite *Ornithonyssus bacoti*

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Ornithonyssus bacoti, the vector of experimental cotton-rat filariasis, is the only mite yet described as an intermediate host of a filariid. However, little is known about the site of development of *Litomosoides carinii* (see Hughes, 1950; Freer, 1953), particularly whether or not the developing stages are intracellular parasites.

Methods

Mites were naturally infected by feeding on patent cotton-rats and were kept without further blood meals at 27°C and 70 to 80% relative humidity. At daily intervals, samples of 10 to 20 mites were either examined alive in saline and then subsequently crushed for measurement of the developing filarial stages, or they were fixed in hot Carnoy or Bouin's fluid for paraffin serial sections and stained with haematoxylin and eosin.

Results

The development from the ingested microfilaria to the infective third-stage larva was accomplished within 13 days, the two moults taking place at days 6 to 7 and 10 to 12. The microfilariae measured 85 μm in length, i.e. about one tenth of the body of the mite, whereas the infective larvae were as long as the mite (850 μm) (Fig. 1). One day after the bloodmeal, microfilariae could be seen through the dorsal plate of the living mite in the space between the two antero-dorsal caeca of the blood-filled gut, where they were actively curling between and within the large granulated cells of the two salivary glands. In sections, the intracellular localization of those first-stage larvae could be confirmed as well as the subsequent disintegration and destruction of the infected cells. In heavily infected mites, the large cells of the salivary glands were almost completely destroyed within the first two or three days after the blood meal.

Other larvae were seen in the haemocoele, in particular in the podosoma, dorsal to the coxae and above the sternal plate lying between, or partially in, the cells of a parenchymatous tissue, which forms a double cell-layer under the epidermis of the podosoma. Functionally, these cells correspond to the fat-body of insects. Most larvae had reached the early sausage-stage within four days, regardless of the sites where they were found.

At the first moult, most larvae were found in syncytia formed by the infected cells of the "fat-body", the nuclei and nucleoli of which were enlarged and the cytoplasm of which had become eosinophilic. No similar induction of hypertrophy and no formation of syncytia occurred in the infected salivary glands. Often, several worms developed close together in the same syncytium, usually in the anterior podosoma, dorso-lateral to the coxae and near to the salivary glands (Fig. 2). In a few cases, developing larvae were found in the secretory glands of the uterus, in the vaginal glands or in the coxal glands, always embedded in a syncytium of hypertrophic cells. The second moult took place within those syncytia. The larvae were completely immobile from the late sausage-stage (day 5) to the early III-stage (day 11) (Fig. 3).

A degenerative development was observed rarely in those larvae which had successfully left the gut after the blood-meal. Most of the larvae that were unable to continue development died at the early sausage-stage, i.e. before they reached the sites of development in the syncytia. Disintegration of developing larvae in the syncytia was accompanied by the formation of crystals in the cytoplasm of the parasitized cells and the cuticula of these larvae was darkly coloured with melanin.

Discussion

Litomosoides carinii develops intracellularly in several organs of the same vector which can be parasitized simultaneously. However, no larvae were found in the Malpighian tubules or in muscle cells, common sites of development in other vectors (Schacher, 1973), and they were not seen in other organs of a suitable size, e.g. the nervous or digestive systems, the ovary or the lyrate organ. *Dipetalonema viteae* develops in two distinct organs, namely, in the muscles and in parts of the salivary glands of *Ornithodorus tartakowskyi* (see Bain, 1967) and this may suggest that the site of development of filariae in the acarine host is less tissue-specific than it is in dipteran vectors.

It is obviously difficult to decide if all larvae develop completely—or at least partially—in the cytoplasm of the syncytia, especially in a heavily infected mite. Therefore, the intracellular localization between the two moults remains uncertain as a "conditio sine qua non". However, it would help to explain the high variability in the rate of development in the intermediate host (Scott *et al.*, 1951; Freer, 1953) and the fact that, hitherto, cultivation of filariae *in vitro* has only been successful up to the early sausage-stages, even when these survived for several weeks (Devaney & Howells, 1979).

References

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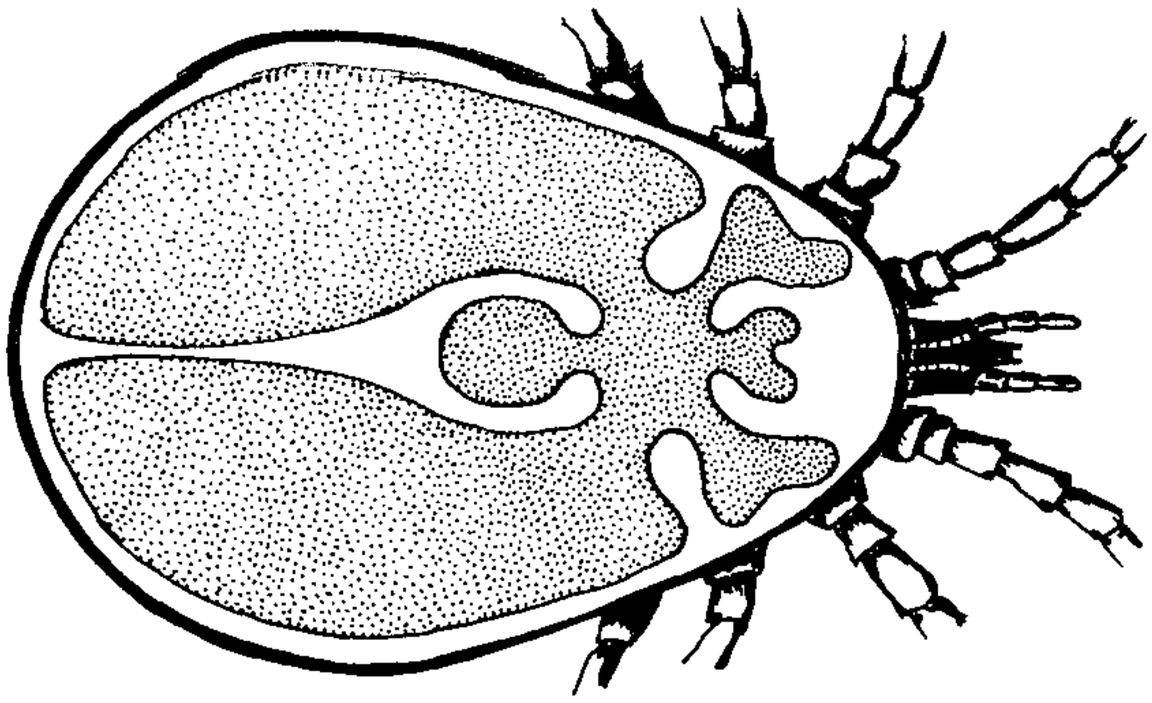

Fig. 1. Size of a freshly engorged mite as compared to the length of developing filarial stages.

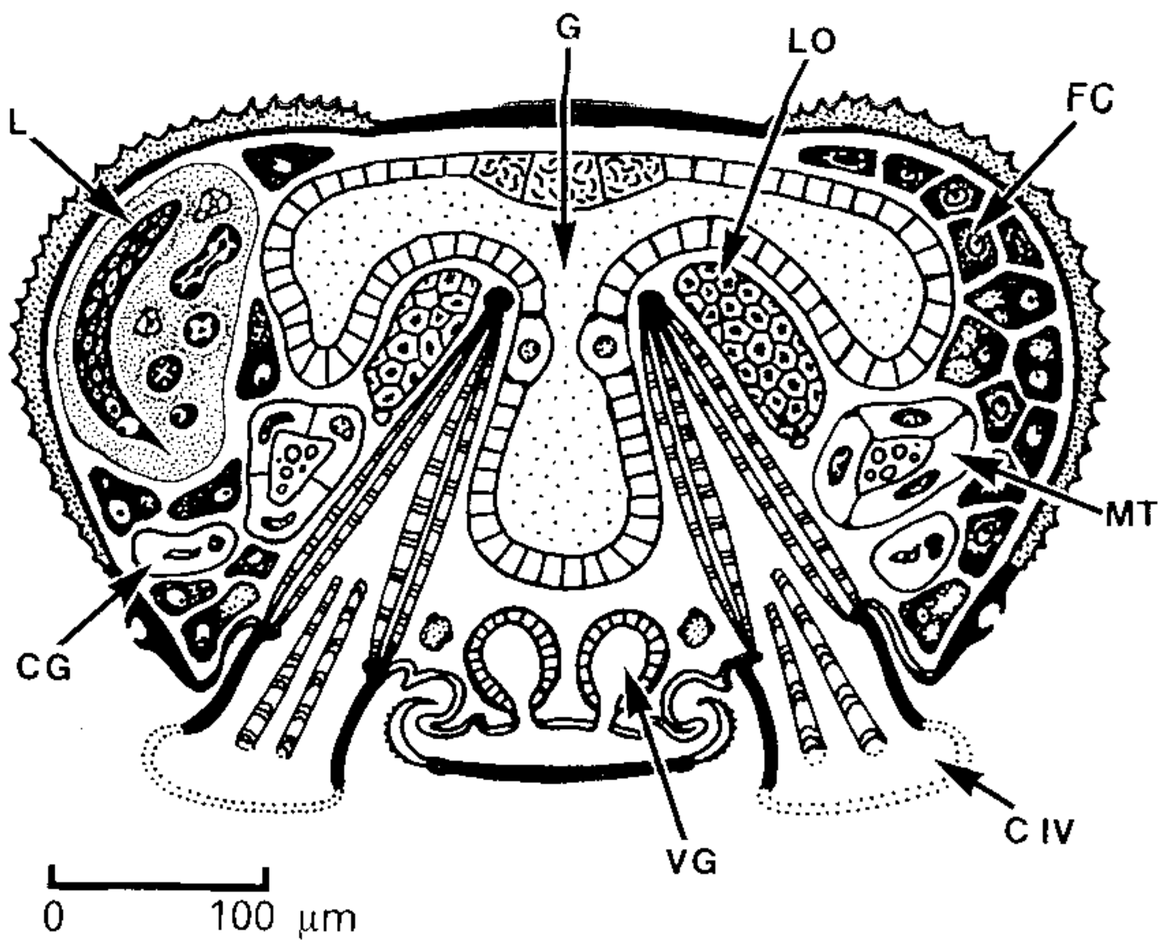

Fig. 2. Transverse section, 4 days after infection.

CG: coxal gland
C IV: coxa IV
FC: fat-cell
G: gut
L: sausage-stage larvae in a syncytium of the fat-cells
LO: lyrate organ
MT: Malpighian tubules
VG: vaginal gland

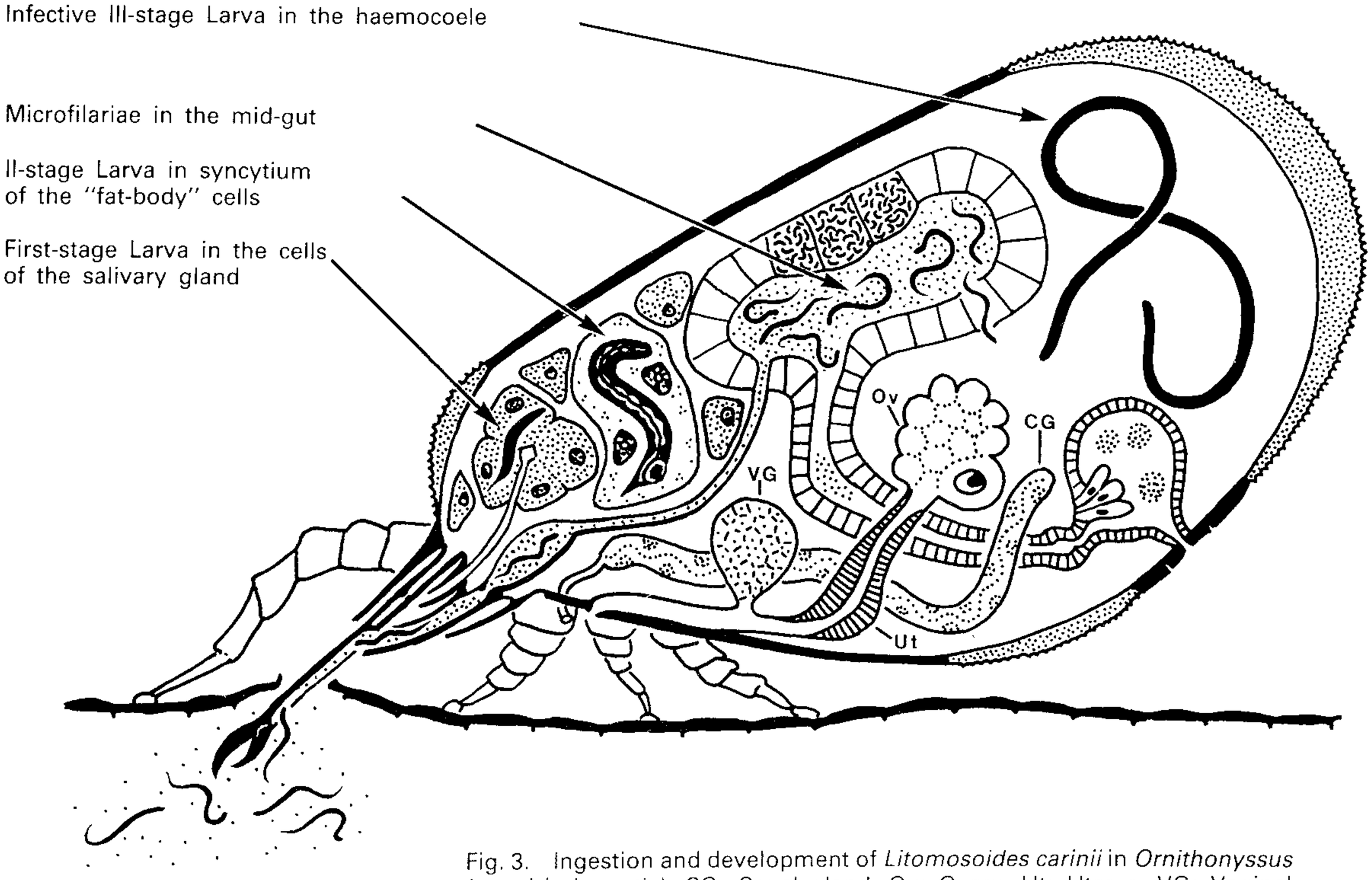

Fig. 3. Ingestion and development of *Litomosoides carinii* in *Ornithonyssus bacoti* (schematic). CG: Coxal gland. Ov: Ovary. Ut: Uterus. VG: Vaginal gland.

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