THE INFECTIVE LARVA OF *LITOMOSOIDES YUTAJENSIS* GUERRERO *ET AL.*, 2003 (NEMATODA: ONCHOCERCIDAE), A *WOLBACHIA*-FREE FILARIA FROM BAT

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Summary:

The infective larva of *Litomosoides yutajensis* Guerrero *et al.*, 2003, a parasite of the bat *Pteronotus parnellii*, is described; it is distinct from congeneric infective larvae by the absence of caudal lappets. The life cycles of five other species of *Litomosoides* are known; three are parasites of rodents, one of a marsupial and one of a bat. As with these species, the experimental vector of *L. yutajensis* used was the macronyssid mite *Ornithonyssus bacoti*. In nature, the main vectors are probably other macronyssids but transmission by *O. bacoti*, with its large host-range, could account for the characteristic host-switchings in the evolution of *Litomosoides*. Unlike the murine model *L. sigmodontis* Chandler, 1931, *L. yutajensis* is devoid of the endosymbiontic bacteria *Wolbachia* and may be of great interest.

KEY WORDS : Nematoda, Filarioidea, infective larva, *Litomosoides*, bat, mite, *Wolbachia*.

Résumé : LARVE INFECTANTE DE *LITOMOSOIDES YUTAJENSIS* GUERRERO *ET AL.*, 2003 (NEMATODA: ONCHOCERCIDAE), FILAIRE DÉPOURVUE DE *WOLBACHIA*, PARASITE DE CHAUVES-SOURIS

Description de la larve infectante de Litomosoides yutajensis *Guerrero* et al., *2003, parasite de* Pteronotus parnellii *; elle est distincte des autres espèces du genre par l'absence de languettes caudales. Comme dans les cycles de* Litomosoides *élucidés, trois parasites de rongeurs, un de marsupial, et un de chauve-souris, le vecteur expérimental est* Ornithonyssus bacoti. *D'autres Macronyssidae sont probabement les principaux vecteurs naturels, mais* O. bacoti, *avec son large spectre d'hôtes, pourrait expliquer que les captures sont caractéristiques de l'évolution du genre* Litomosoides. L. yutajensis *est dépourvu de la bactérie endosymbionte* Wolbachia, *contrairement au modèle murin* L. sigmodontis, *et pourrait avoir un grand intérêt.*

MOTS CLÉS : *Nematoda, Filarioidea, larve infectante,* Litomosoides, *chiroptère, acarien,* Wolbachia.

INTRODUCTION

The species of *Litomosoides* Chandler,1931 are parasites of neotropical bats and terrestrial mammals, including didelphid marsupials and murid rodents (Bain *et al.*,1980; Brant & Gardner, 1997; Guerrero *et al.,* 2002; Bain *et al.,* 2003; Notarnicola, 2005). Life-cycles have been elucidated for only five of the 31 described species, three from sigmodontines (Scott *et al.*, 1951; Bain *et al.*, 1980; Diagne *et al.,* 1989), one from marsupials (Bain *et al.*, 1980), and one from a bat (Bain *et al.,* 2002). For all of them the experimental vector used was the same, the mite *Ornithonyssus bacoti* (Hirst, 1913) (Macronyssidae).

One of these species, *L. sigmodontis* Chandler, 1931 (incorrectly named *L. carinii* in the past; see Bain *et al.,* 1989), has become a much used murine model for filarial immunological analyses (Bain & Philipp, 1991; Martin *et al.*, 2001; Bain, 2002; Allen *et al.,* 2000; Hoerauf *et al.*, 2005). Like the majority of human filariae and other onchocercids, most species of the genus *Litomosoides* are associated with the endosymbiotic bacterium *Wolbachia pipientis* Hertig, 1936. However *L. yutajensis* Guerrero *et al.,* 2003 is free of this endosymbiont (Casiraghi *et al.*, 2004). As in other filarial nematodes, there is evidence that this bacterium is required for the development and reproduction of its filarial host (Hoerauf *et al.*, 1999; Casiraghi *et al.,* 2002), knowledge of the life cycle of *L. yutajensis*, which parasites a bat (Guerrero *et al.*, 2003), would be of particular interest.

MATERIALS AND METHODS

Bats were collected in Yutaje, Amazonas, Venezuela (5° 36' 30'' N, 66° 06' 51'' W) using three mist nets between 19:00 and 21:00 in October 2004 at the end of the rainy season. Nets were placed 1-3 metres above ground level, close to a small stream in an undisturbed area of tropical humid forest. Bats were collected alive and placed individually in cotton bags. Next morning, 2 µl of blood were taken by clipping a hind claw and immediately examined microscopically for living microfilariae.

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Ornithonyssus bacoti was used as experimental vector. The specimens, originating from a laboratory colony, were kept in 6 × 1.5 cm glass tubes, each containing 25 mites (Diagne *et al.,* 1990). Mites were released to feed on a microfilarial bat which had been immobilized in soft wire mesh and then placed overnight above a container filled with water in case any mites escaped. The next morning the fed mites were recovered, placed in tubes, and kept in an ice-box with wet cotton wool to maintain 80 % humidity and a temperature close to 25-27° C.

RESULTS

Filarial infection

Many bats were collected and immediately released but seventeen *Pteronotus parnellii* (Gray) (Chiroptera: Mormoopidae), the type host of *L. yutajensis*, were collected and examined. Of these, three had only microfilariae and one had both adults and microfilariae, about 300/10 µl; this bat (number 39 YU) was used to feed the mites. A total of 420 fully engorged male and female *O. bacoti* were recovered and, after 15 days, two motile larvae were recovered from 191 surviving mites. One of these was a fully infective larva and the other was a late second stage.

Morphology of the infective larva

Body 750 µm long, 15 µm wide; tubular buccal capsule 19 µm long; nerve ring and excretory pore 65 µm and 85 µm from anterior extremity, respectively; oesophagus 290 µm long, with anterior muscular part of 160 µm; tail 52 µm long; tail extremity slightly distorted and attenuated; no lateral lappets (Fig. 1A-C).

DISCUSSION

The infective larva of *L. yutajensis* had the same morphology as those previously described in *Litomosoides* species: *L. sigmodontis* Chandler, 1931; *L. legerae* Bain *et al.,* 1980 and *L. galizai* Bain *et al.,* 1989, parasitic in sigmodontines; *L. petteri* Bain *et al.,* 1980; parasitic in didelphid marsupials; and *L. chandleri* Esslinger, 1973, parasitic in bats. The infective larvae of all these species have a long tubular buccal capsule nearly 20 µm long, a distinctive character of the genus (Bain & Chabaud, 1986). *L. yutajensis*, the only *Litomosoides* species without *Wolbachia*, is distinct from the other species in the absence of the two lateral small lappets at the caudal extremity.

Only two *L. yutajensis* larvae were found. The low density of blood microfilariae of the bat used to feed the

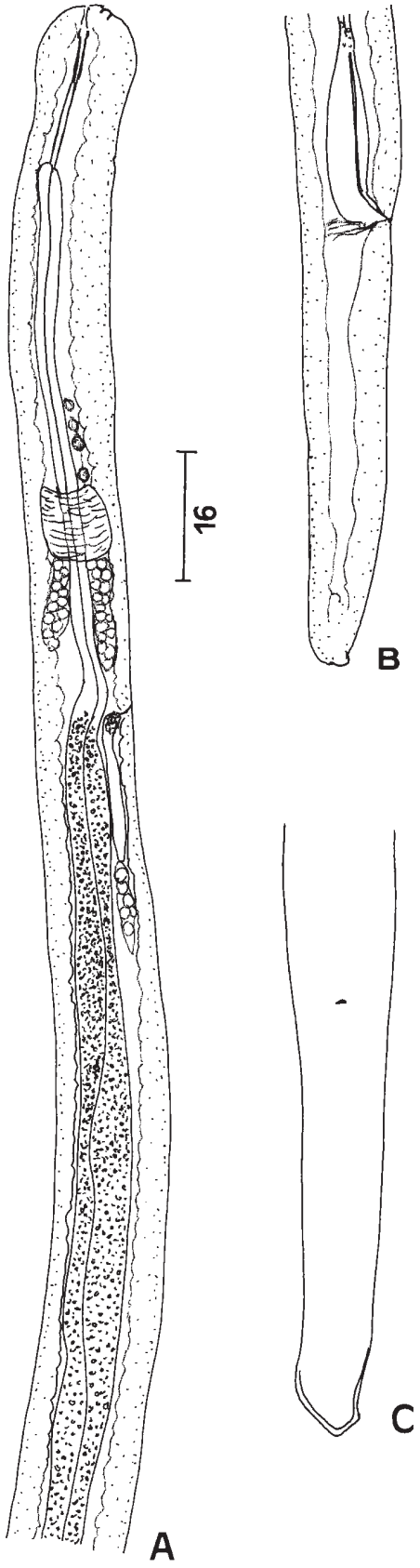

Fig. 1. – Infective larva of *Litomosoides yutajensis*. A: anterior part, right lateral view. B and C: caudal part, right lateral and ventral views, respectively. Scale bars: 16 µm.

mites was a restricting factor, since a high proporion of ingested microfilariae are destroyed by the teeth of the buccal armature (Diagne *et al.,* 1990). Moreover *O. bacoti* is probably not the natural vector, at least not the predominant one. *O. bacoti* is a widespread mite on birds and mammals (Micherdzinski, 1980; Guerrero, 1985) but it is found less commonly on bats than on terrestrial rodents and marsupials. Other macronyssids, such as *Radfordiella* spp. (Radovsky *et al.,* 1971), have been recorded on several species of bats including *Carollia perspicillata* (Linnaeus), *Anoura caudifer* (E. Geoffroy), *Glossophaga soricina* (Pallas) which also hosts *Litomosoides* species (Esslinger, 1973; Guerrero *et al*., 2002; Bain *et al.,* 2003). An undescribed species of *Radfordiella* has been reported from *Pteronotus parnellii*, the type host of *L. yutajensis,* by Radovsky (1967), Saunders (1975) and Guerrero *et al.* (2003). Interestingly, Yunker & Chitwood (1972) reported a macronyssid, *Macronyssoides kochi* (Fonseca), containing two filarial larvae on the bat *Artibeus jamaicensis* Leach, in Mexico. The worms' size (742 µm long, 14 µm wide), the host and its geographical origin leave little doubt that they represent a species of *Litomosoides*, although the buccal capsule was not observed by these authors.

Several species of macronyssids naturally infecting bats are probably the natural vectors of *Litomosoides* species of these hosts. *O. bacoti*, although a less suitable vector, might play a role in favouring dispersion among the taxonomically unrelated mammals constituting the wide host-range of *Litomosoides*, and may be responsible for the host-switchings which seem to be a mark of the evolution of this filarial genus.

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