# ULTRASTRUCTURE OF THE TEGUMENT OF *Metamicrocotyla macracantha* (ALEXANDER, 1954) KORATHA, 1955 (MONOGENEA, MICROCOTYLIDAE)

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## ABSTRACT

The ultrastructure of the body tegument of *Metamicrocotyla macracantha* (Alexander, 1954) Koratha, 1955, parasite of *Mugil liza* from Brazil, was studied by transmission electron microscopy. The body tegument is composed of an external syncytial layer, musculature, and an inner layer containing tegumental cells. The syncytium consists of a matrix containing three types of body inclusions and mitochondria. The musculature is constituted of several layers of longitudinal and circular muscle fibers. The tegumental cells present a well-developed nucleus, cytoplasm filled with ribosomes, rough endoplasmatic reticulum and mitochondria, and characteristic organelles of tegumental cells.

*Key words*: ultrastructure, tegument, *Metamicrocotyla macracantha*, Monogenea.

## RESUMO

### Ultra-estrutura do tegumento de *Metamicrocotyla macracantha* (Alexandre, 1954) Koratha, 1955 (Monogenea, Microcotylidae)

Foi realizado o estudo do tegumento do corpo de *Metamicrocotyla macracantha* (Alexander, 1954) Koratha, 1955, parasito de *Mugil liza* (tainha) do Canal de Marapendi, Rio de Janeiro, Brasil, pela microscopia eletrônica de transmissão. O tegumento é formado por uma camada externa sincicial, uma camada muscular e uma camada interna contendo células tegumentares. O sincício consiste em matriz com três tipos de corpos de inclusão e mitocôndrias. A musculatura é formada por diversas camadas de fibras musculares longitudinais e circulares. As células tegumentares apresentam núcleo bem desenvolvido, citoplasma preenchido por ribossomas, retículo endoplasmático rugoso e mitocôndrias, organelas características das células tegumentares.

*Palavras-chave*: ultra-estrutura, tegumento, *Metamicrocotyla macracantha*, Mongenea.

## INTRODUCTION

The body tegument has been studied by transmission electron microscopy in only a few species of the Microcotylidae: *Atriaster* sp. by Justine (1992), *Microcotyle* sp. by Justine & Bonami (1993), *Atriaster heterodus* by Santos & Lanfredi (2000), and in *Paranaella luquei* by Cohen *et al.* (2001).

*Metamicrocotyla macracantha* (Alexander, 1954) has been reported from marine fishes belonging to the genus *Mugil* at several places worldwide (Koratha, 1955; Hargis, 1956; Bravo-Hollis, 1966, 1982; Young, 1970; Tantaleán, 1974; Skinner, 1975, 1978; Rawson, 1976; Minchew, 1977; Collins, 1985; Conroy *et al*., 1985, 1986; Garcia & Williams, 1985; Oliva & Munoz, 1985; Bargiela, 1987; Juarez-Arroyo

& Salgado-Maldonado, 1989; Kohn *et al*., 1994; Luque, 1994; Knoff *et al.,* 1997).

The surface topography of *M. macracantha* was described through scanning electron microscopy by Kohn *et al.* (1994), with their transmission electron microscopic observations related to those for spermiogenesis and spermatozoa as reported by Baptista-Farias *et al.* (1995) and vitelline cells by Baptista-Farias & Kohn (1998).

The present study describes the tegument ultrastructure of *M. macracantha*, a parasite from *Mugil liza* from Rio de Janeiro.

## MATERIALS AND METHODS

Specimens of *Metamicrocotyla macracantha* were collected from the gills of *Mugil liza* (Val.) (Mugilidae) obtained from fishermen of Canal de Marapendi, Barra da Tijuca, Rio de Janeiro, Brasil. The helminths were fixed in 0.1 M phosphate-buffer 2.5% glutaraldehyde, post-fixed for 1 h in 1% osmium tetroxide in the same buffer, dehydrated in an increasing graded ethanol series, and embedded in Epon (Luft, 1961). Ultrathin sections obtained using a Reichert Ultracut E-microtome were collected in 200 mesh grids, contrasted with uranyl acetate for 30 min and Reynold's solution of lead citrate for 5 min, and observed under a Zeiss EM electron microscope.

## RESULTS

The body tegument of *Metamicrocotyla macracantha* is composed of an external syncytial layer and musculature, and an inner layer containing tegumental cells (Fig. 1). The external syncytial layer is delimited by the apical and basal plasma membranes (Figs. 2 and 4, respectively). The apical plasma membrane is coated by the glycocalyx which presents ridges at regular intervals. A fibrous terminal web is observed immediately beneath the apical plasma membrane (Fig. 2).

The basal plasma membrane presents thin projections which extend into the syncytial layer continuous with the basal lamina, which is just beneath and presents uniform, thickness, without folds (Fig. 4).

The syncytium consists of a matrix containing three different types of body inclusions: round bodies (Fig. 2, n. 1); electrondense bodies (Fig. 2, n. 2); and electronlucent vesicles (Fig. 2, n. 3), which in some regions of the syncytium seem to be fused, forming a multivesicular body. The density of these body inclusions varies, according to the region of the tegument. In some regions, the electronlucent vesicles are more abundant (Fig. 2) while, in others, the electrondense bodies predominate (Fig. 3). Round to elongate mitochondria presenting about six cristae and sparse distribution (Figs. 2 and 4) were also observed.

The musculature is constituted by several layers of longitudinal and circular muscle fibers, presenting large and round to elongate associated mitochondria, which are surrounded by a limiting membrane, and present one to six well-defined cristae (Fig. 5). The space between the muscle fibers and mitochondria is filled with interstitial material, and neuromuscular plates are observed in the muscle fiber extremities (Fig. 5).

The irregularly shaped tegumental cells present nuclei without defined regions of heterochromatin, cytoplasm filled with ribosomes, rough endoplasmatic reticulum, and mitochondria with many cristae (Fig. 1).

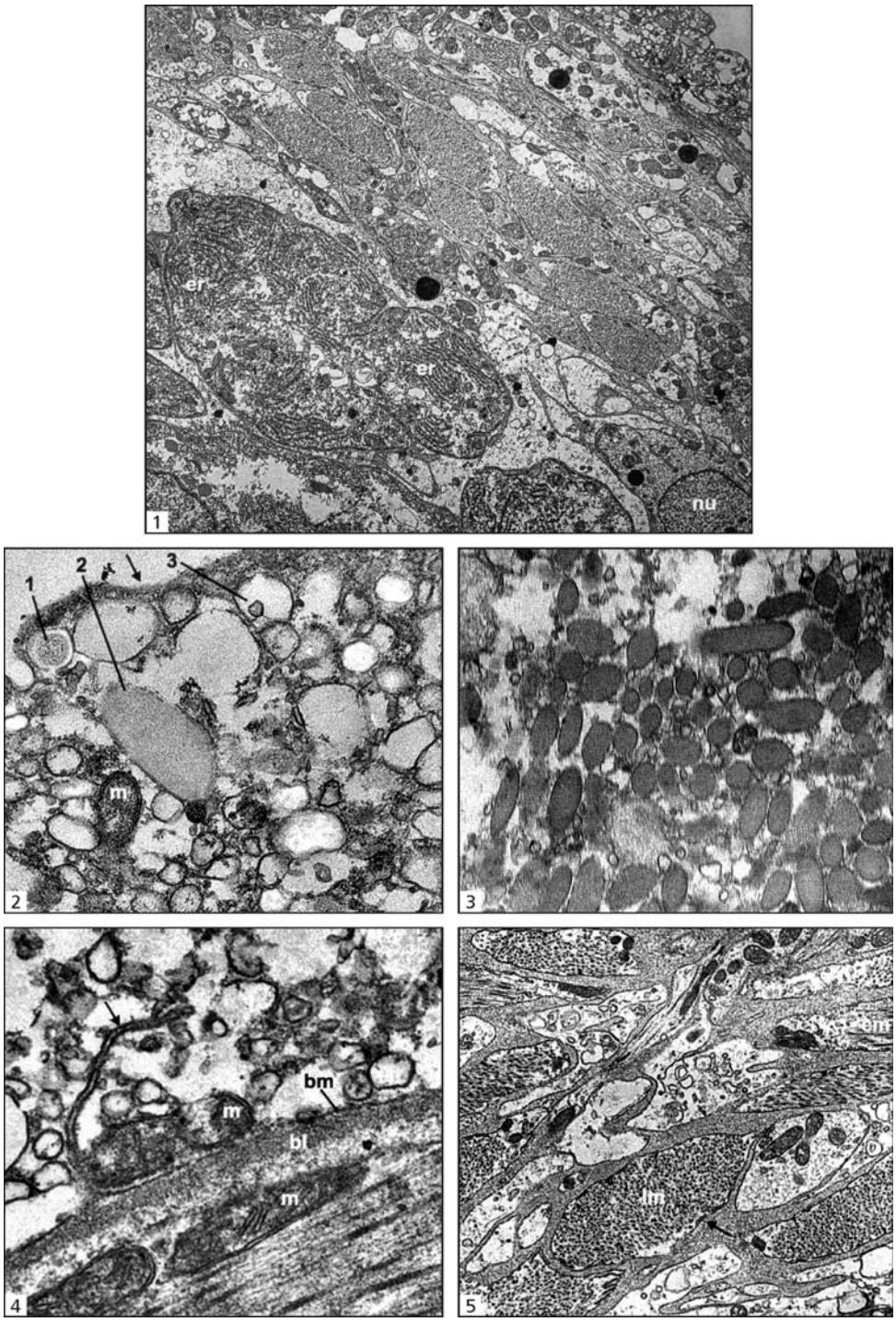
## DISCUSSION

The body tegument of *Metamicrocotyla macracantha* follows the general pattern described in monogenean species already studied, although presenting a few differences.

Microvillous-like tegumental projections have been observed in the tegument of some microcotylid species, as in *Atriaster* sp. by Justine (1992), *Atriaster heterodus* by Santos & Lanfredi (2000), and *Paranaella luquei* by Cohen *et al.* (2001). They had not been observed in the present species, as in *Microcotyle* sp. by Justine & Bonami (1993).

*Metamicrocotyla macracantha* was found coiled in the branchial filaments, as referred to by Minchew (1977) and Conroy *et al.* (1985, 1986). This characteristic position could explain the absence of microvillous-like tegumental projections in the body tegument, since these structures could be prejudicial to the host tissue. According to Halton (1979), the absence of microvillous proportions better contact between host and parasite.

The differences in the morphology and in the electrondensity of body inclusions found in the tegument suggest that they probably have different biochemical properties and functions (Ramasamy & Bhuvaneswari, 1993). Similar inclusions presenting variations in pattern and distribution were reported on the tegument of many Monogenea (Halton, 1982; Smyth & Halton, 1983; Threadgold, 1984).




**Figs. 1-5** — Transmission electron micrographs of the tegument of *Metamicrocotyla macracantha*. **Fig. 1** — Transversal section of body showing the general view of the tegument, composed by an external syncytial layer, musculature and tegumental cells with nucleus (nu) and endoplasmatic reticulum (er). The 5,000 x. **Fig. 2** — Apical region of the tegument showing the external syncytial layer presenting a terminal web (arrow), three types of body inclusions: round bodies (n. 1), electrondense bodies (n. 2), and electronlucent vesicles (n. 3) and mitochondria (m). The 40,000 x. **Fig. 3** — Detail of the external syncytial layer showing a region where the electrondense bodies are more abundant. The 19,000 x. **Fig. 4** — High magnification of the basal region of the external syncytial layer showing the thin projections (arrow) of the basal plasma membrane (bm), mitochondria (m), and basal lamina (bl). The 50,000 x. **Fig. 5** — Musculature of body constituted by circular and longitudinal fibers (lm), associated mitochondria, and detail of neuromuscular plates (arrow), 10,000 x.

The basal invaginations observed in the external syncytial layer suggest that the tegument has a hole for obtaining nutrients, for excretion, and for osmorregulation (Brennan & Ramasamy, 1996). According to Lyons (1972), thin projections of this membrane, as observed in *M. macracantha*, are filled by a reticulin-like substance forming the cytoskeleton and are related to the sustenance of the tegumental external layer.

The body musculature of *M. macracantha* is well developed. The folds observed are due to contraction of longitudinal and circular muscle fibers. Neuromuscular plates were observed in the extremity of the fibers, as in the musculature of *Pricea multae* (Ramasamy *et al.,* 1986). Mitochondria of the muscular layer are smaller than those found in the external syncytium, as in other Polyopisthocotylea such as *Vallisia indica* (Ramasamy *et al*., 1987), although in *P. luquei* (Cohen *et al*., 2001) the mitochondria of the external layer are smaller.

The fibrous terminal web, observed just beneath the apical plasma membrane, was also observed in other Microcotylidae, as *Microcotyle* sp. by Justine & Bonami (1993) and in *Atriaster* sp. by Justine (1992) and *P. luquei* (Cohen *et al.*, 2001). According to Lyons (1973), this web is a feature displayed by Polyopisthocotylea and had not been observed in Monopisthocotylea.

The synthesis of tegumental inclusions involves the rough endoplasmatic reticulum and Golgi complex, characteristic organelles of tegumental cells. In the tegument of *M. macracantha,* only one type of tegumental cell, which produces the different kinds of body inclusions, was observed.

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