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SURFACE TOPOGRAPHICAL AND ULTRASTRUCTURAL STUDIES ON A CARYOPHYLLIDEAN CESTODE, *LYTOCESTUS INDICUS* (LYTOCESTIDAE)

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ABSTRACT: The body of *Lytocestus indicus* is divided into scolex, neck and non-segmented strobila. The scolex region lacks well differentiated structures for anchorage and has a smooth surface, while the post scolex region has narrowly spaced, fine transverse foldings. The worm's body surface is covered with microtriches; also, pit like structures and papillae are found in the tegument of the mid body. Ultrastructurally, the tegument has a cytoplasmic zone lined externally and internally by plasma membrane, with invagination in the outer one. Rod shaped structures are abundantly found in the cytoplasmic zone. The cytoplasmic zone is followed by muscular and parenchymal layers.
Key words: *Lytocestus indicus*, Caryophyllidea, Cestode, Scanning electron microscopy, Transmission electron microscopy, Tegument

INTRODUCTION

In the absence of a conventional digestive system, the tegument of cestodes serves as the only interface in the host-parasite system and plays an important role in the physiological functions of the parasite. The surface topography and structure of the tegument with regard to cyclophyllidean cestodes, larval (Voge *et al*, 1979; Novak and Dowsett, 1983; Conn, 1988; Mizinska- Boevska *et al*, 1989) and adult stages (Morseth, 1966; Thompson *et al*, 1982; Velichko and Skvortsova, 1984, 1985; Holy and Oaks, 1986; Davydov *et al*, 1990; Irshadullah *et al*, 1990; Rasheed and Lewis, 1992) have been studied. Information with regard to pseudophyllidean cestodes is also available (Anderson, 1975a; Boyce, 1976; Tedesco and Coggins, 1980; Yamane *et al*, 1982).

Regarding the monozoic cestodes, studies on this aspect are rather scanty. The available accounts mainly pertain to some caryophyllids like *Hunterella nodulosa* by Hayunga and Mackiewicz (1975), *Caryophyllaeus laticeps* by Richards and Arme (1981, 1982) and *Lytocestus indicus* by Bhattacharjee and Tandon (1990). The present communication aims to investigate into the surface topography and ultrastructural organization of the tegument in *Lytocestus indicus*, a common caryophyllid parasite of siluroid fishes in India.

METHODOLOGY

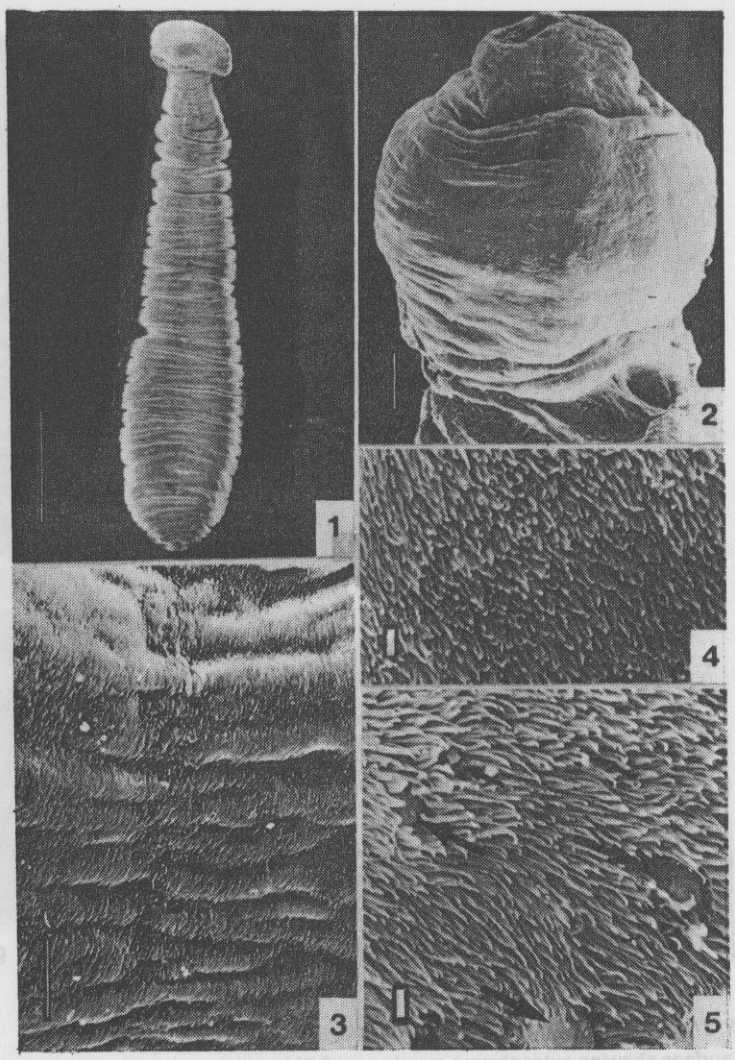
Live specimens of *Lytocestus indicus* (Moghe, 1925) Woodland, 1926 were recovered from the duodenum of freshly killed catfish, *Clarias batrachus* (L) in 0.7% saline and processed for scanning and transmission electron microscopic study. For scanning electron microscopy (SEM), the specimens were fixed in 10% neutral buffered formalin, dehydrated through an ascending alcohol series and critical-point dried. In lieu of critical-point drying, tetramethylsilane was also used following Dey *et al* (1989). Gold plated specimens were observed under scanning electron microscope JEOL-JSM- 35 CF. For transmission electron microscopy small-sized pieces of the worm were first fixed in 3% glutaraldehyde in 0.2 M cacodylate buffer, followed by post fixation in 1% Osmium tetroxide in 0.2 M cacodylate buffer. Alcohol and toluene were used for dehydration and clearing, respectively. CY 12 was used as an embedding medium. Ultrathin sections were cut using an LKB 2088 Bromma Ultrotome, double stained with alcoholic uranyl acetate and aqueous citrate and viewed in JEOL JEM- 100 CX- II.

RESULTS AND DISCUSSION

The body of *L. indicus* is divided into three parts: scolex, an indistinct neck and non-segmented strobila. The scolex region which lacks well differentiated anchoring structures has indistinct furrows or bothria. The indistinguishable neck is merging between the scolex and strobila. The scolex region has a relatively smooth surface while most of the post-scolex length of the body shows narrowly spaced, fine transverse foldings which give the body surface a highly wrinkled texture (Figs. 1 & 2). The whole body of the worm is covered with densely distributed microtriches. In all the three regions, microtriches are simple without showing any branching (Figs. 3-5).

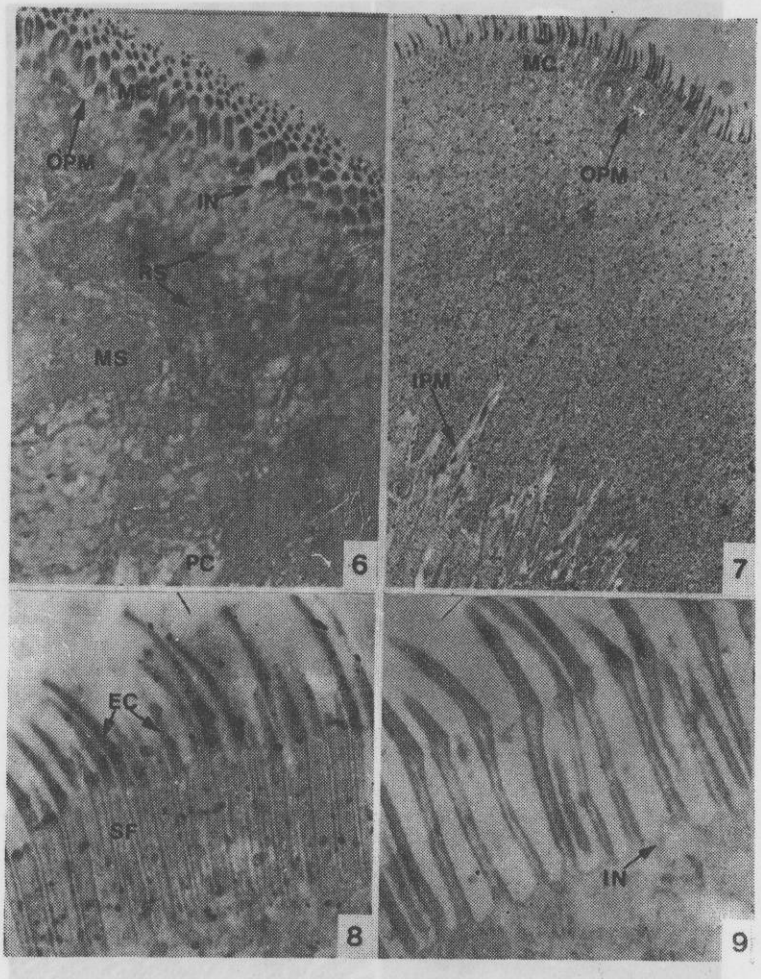
In the tegument of the mid-body region of the worm pit-like structures are revealed scattered here and there and surrounded by microtriches. Prominent dome-shaped rounded elevations or papillae are also observed bulging out at places from the microtrichial tuft in the mid-body. These papillae have smooth surface and are devoid of the microtriches (Fig. 5).

Ultrastructurally, the body surface is elaborated by the presence of a cytoplasmic zone which is bordered externally by an outer plasma membrane and internally by an inner plasma membrane (Figs. 6 & 7). The outer plasma membrane has got inpushings in the form of small invaginations; it is also in continuity with the outer covering of the microtriches (Figs. 6-9). The inner plasma membrane has an irregular border or outline (Fig. 7). Each microthrix is



Figs. 1-5: Scanning electron micrographs of *Lytocestus indicus*.

1. Entire worm. Scale bar, 1mm; 2. Scolex region. Scale bar, 100 µm; 3. A region in the scolex showing microtriches. Scale bar, 10 µm; 4. Microtriches in the neck region. Scale bar, 1 µm; 5. A closer view of the strobilar tegument; papillae (large arrows) and pit-like structures (small arrows) in between the microtriches are seen at certain places. Scale bar, 1 µm.



Figs. 6-9: Transmission electron micrographs of *Lytocestus indicus*.

6. Body surface showing outer plasma membrane (OPM), with invagination (IN), microtriches (MC), rod-shaped structures (RS); muscle (MS) and parenchymal cells (PC). Scolex region. x13,400.
7. Body surface showing microtriches (MC), outer and irregularly arranged inner plasma membrane (OPM, IPM). Neck region. x10,000.
8. Microtriches as seen at higher resolution; the electron dense cap (EC) and the shaft (SF) are clearly seen. Neck region. x40,000.
9. Microtriches from yet another field; invagination (IN) of outer plasma membrane is also shown. Scolex region. x5,400.

elongated and distinguishable into two parts, an electron dense cap and a shaft. These two parts are almost equal in length. The electron dense cap has a pointed tip, while the shaft has an electron lucent core (Figs. 8 & 9). In the cytoplasmic zone of the scolex region, rod shaped structures are abundantly scattered. The cytoplasmic zone is followed by the muscular zone where muscle cells are observed. Below the muscular zone is the parenchymal zone(Fig. 6) .

As also observed by Bhattacharjee and Tandon (1990) the scolex of *L. indicus* lacks any defined anchoring structures and is provided with indistinct furrows or bothria. However, in other caryophyllidean cestodes like, *Djombangia penetrans* a terminal introvert of glandular nature is present (Fuhrman, 1931; Mackiewicz, 1972). In pseudophyllidean cestodes, *Diphyllobothrium spp.*, the scolex possesses bothria on the ventral and dorsal side (Anderson, 1975b).

L. indicus has a microtrich- carpetted scolex surface, an observation which is similar to that of a cyclophyllidean cestode, *Echinococcus granulosus* (Irshadullah *et al*, 1990). The occurrence of simple and unbranched microtriches confers with the observation made by Bhattacharjee and Tandon (1990) in the same species. Three types of microtriches have been observed in *E. granulosus*: the long filamentous type, thin blade like and thick blade like (Irshadullah *et al*, 1990). Boyce (1976) opined that the microtriches are involved in the uptake of nutrients. Microtriches have thus been analogized with the microvilli which constitute the brush border of many other invertebrate and vertebrate transport epithelial systems (Read, 1955; Rosario, 1962: Lumsden, 1966; Beguin, 1966; Rifkin *et al*, 1970). The long filamentous microtriches are seemingly involved in absorptive function (Lumsden *et al*, 1982).

The pit-like structures observed in *L. indicus* in the present study show resemblance to the openings of pore canals in diphyllobothriid cestodes (Yamane *et al*, 1975). The dome-shaped structures or papillae interrupted by scattered pits which are also free of microtriches appear to be similar to the tumuli in the pseudophyllidean cestode, *Eubothrium salvelini* as reported by Boyce (1976). These structures were not detected in the ultrathin sections, and this could be due to the reason that they were missed during sectioning.

The cytoplasmic zone bound internally by the inner plasma membrane in *L. indicus* is comparable with the distal cytoplasm and fibrous zone in the cyclophyllidean species; in the latter also, the presence of muscles, parenchymatous cells and microtriches covered by a membrane that is continuous over the outer surface of the distal cytoplasm have been reported (Morseth, 1966).

In *L. indicus* each microthrix is differentiated into two almost equal parts: an electron dense cap with a pointed tip and a lightly stained shaft, similar to the

conventionally known structure of microtriches (Rothman, 1960, 1963; Yamane *et al*, 1982). However, Richards and Arme (1981) regarded the microthrix of *C. laticeps* as comprising three regions; the proximal shaft, the electron dense spine and a distal extension.

The rod-shaped bodies found in the cytoplasmic zone of *L. indicus* are comparable with variously described tegumentary bodies (Rothman, 1963; Beguin, 1966; Braten, 1968; Morris and Finnegan, 1969; Lumsden *et al*, 1974; Threadgold and Hopkins, 1981) and their presence is regarded as typical of the caryophyllidean tegument (Davydov and Poddybnaya, 1988).

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