Ultrastructural studies of the Mehlis´ gland in *Metamicrocotyla macracantha* (Monogenea, Microcotylidae) parasite of *Mugil liza* (Teleostei)

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Abstract

The ultrastructure of Mehlis' gland of *Metamicrocotyla macracantha*, a gill parasite collected from *Mugil liza* from Rio de Janeiro, Brazil, was studied by transmission electron microscopy. The Mehlis' gland consists of two types of secretory cells, S1 and S2, each producing a different secretory body. The S1 bodies are spherical, lamellae-like and were observed in different stages of development in the cytoplasm of these cells. The S2 bodies are spherical to ovoid with dense content, showing a crystalline structure. The cytoplasm of Mehlis' gland cells contains also free ribosomes, granular endoplasmatic reticulum and Golgi complex, characteristic organelles of secretory cells.

Key-words:
Mehlis´gland.
Ultrastructure.
Monogenea.
Metamicrocotyla macracantha.
Transmission electron microscopy.

Introduction

Mehlis' gland is a feature of the female reproductive system of parasitic platyhelminthes, being associated with the ootype in which the egg is assembled. Much variations in numbers, types and arrangement of these glands has been documented[1]. Most studies of Mehlis' gland have been conducted in Digenea[2,3,4,5,6,7,8,9,10,11,12], but ultrastructural studies of the following monogeneans have also been published: *Diplozoon paradoxum* and *Calicotyle kroyeri*[13], *Cichlidogyrus halii typicus*[14], *Entobdella hipoglossi*[15] and *Entobdella soleae*[15,16].

Kearn[1] emphasised the need for ultrastructural studies of Mehlis' gland in a greater range of monogeneans and, with this in mind our attention focussed on *Metamicrocotyla macracantha*, (Alexander, 1954) Koratha, 1955, a polyopisthocotylean gill parasite of marine fish *Mugil liza*. Previous ultrastructural studies of the reproductive system of this parasite have been concerned with spermatogenesis, spermiogenesis and spermatozoa[17], vitelline cells[18], tegument[19] and, in the presente study, the Mehlis´ gland of this species was examined by transmission electron microscopy.

Materials and Methods

The parasites were collected from the gills of *Mugil liza*, caught off Rio de Janeiro, Brazil. Worms were fixed in 0.1M phosphate-buffered 2.5% glutaraldehyde, postfixed for one hr in 1% osmium tetroxide in the same buffer, dehydrated in a graded ethanol series, and embedded in Epon. Ultrathin sections were collected on copper grids, double-stained with 2% alcoholic uranyl acetate and lead citrate and observed in a Zeiss EM 900 electron microscope.

Results and Discussion

The Mehlis' gland of M. *macracantha* was found to consist of two types of secretory cells, referred to here as S1 and S2. Each cell type produces a different secretory body.

The Mehlis´gland secretory cells are lobed, slender, with ducts opening on each side into the lumen of the ootype.

The secretory cells S1 contain a large

nucleus with nuclear pores, a prominent nucleolus and homogeneous heterochromatin (Figure 1). The plasma membrane presents infoldings into the cytoplasm, toward the cell nucleus. The cytoplasm contains many free ribosomes and profiles of granular endoplasmic reticulum. Mitochondria are usually found, mainly near the plasma membrane.The spherical S1 bodies were observed in different developmental stages in the cytoplasm according to its maturation (Figure 3), involving a progressive condensation of its contents, which become lamellae-like (Figure 2).

The secretory cells S2 are lobed, presenting deep folds in the membrane, reaching the proximities of the nucleus and separated from each other by interstitial material. These cells contain a large central and irregular nucleus (Figure 4). The peripheral cytoplasm contains numerous free ribosomes and granular endoplasmic reticulum in abundance, containing cisternae with finely granular material together with Golgi complexes. A few mitochondria were observed distributed throughout the cytoplasm. The space without organelles within the cytoplasm is filled with spherical to ovoid S2 secretory bodies. The dense contents of these bodies show a crystalline structure, with the units arranged in parallel, alternating electron-dense and electron-lucent lines. The electrondensity of these S2 bodies was observed to vary, according to the stage of development (Figure 5)

The cytoplasmic extensions (ducts) of the secretory cells carry secretory products to the ootype lumen. These ducts, besides secretory bodies, also contain mitochondria and ribosomes (Figure 6)

The secretory products of the two types of cells present few changes in the structure during the migration through the duct. The S1 bodies become dilated and irregular in the terminal portion of the duct, resulting in a space between the limiting membrane of the secretory body and the enclosed group of microvesicles. S2 secretory bodies have not been observed within the duct (Figure 6)

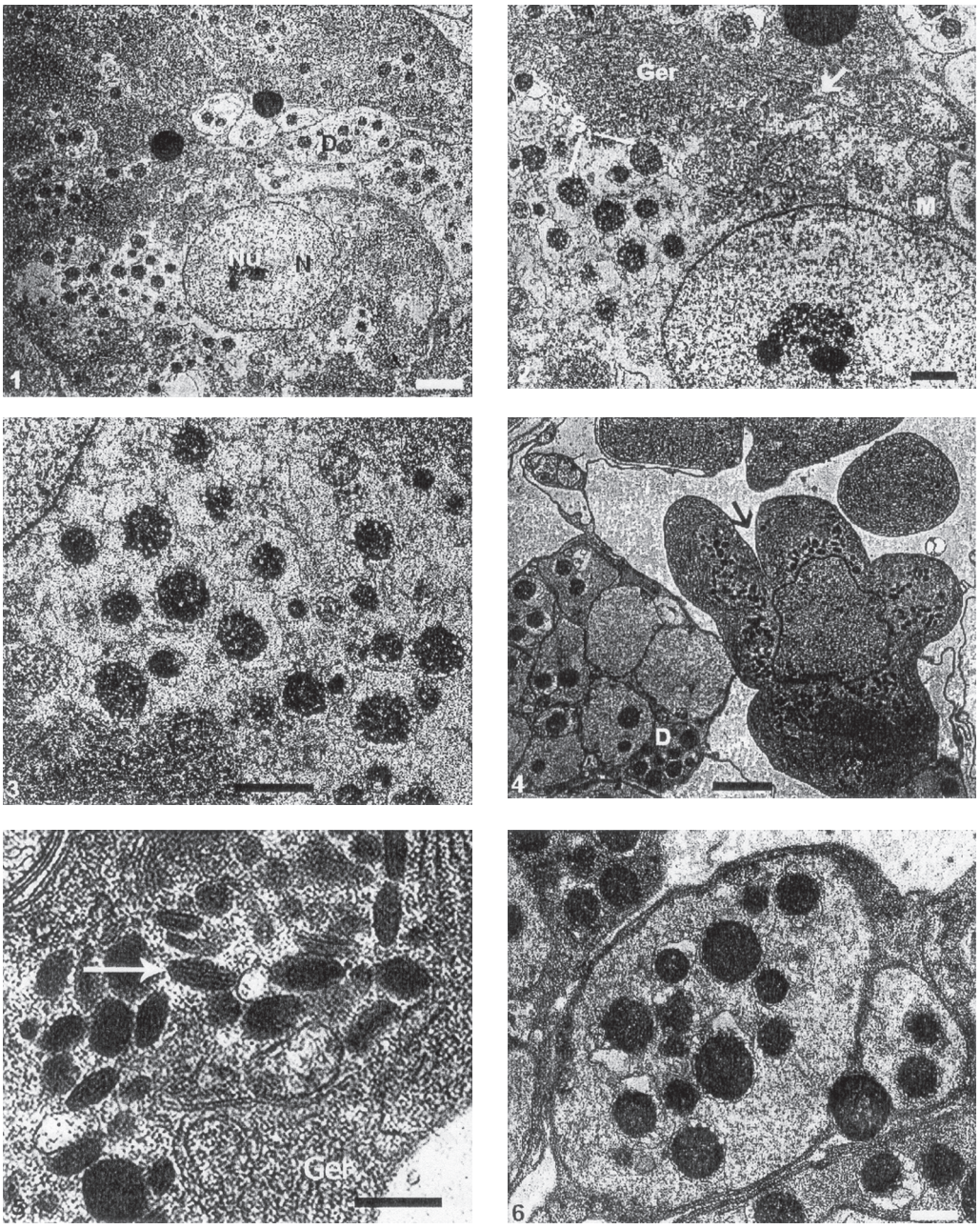
The ultrastructure of the Mehlis' gland of *Metamicrocotyla macracantha* is similar to that of other monogeneans and most digeneans already studied, presenting two types of secretory cells, producing distinctly different types of secretory body. These two kinds of cells are referred to here as S1 and S2, according to the nomenclature proposed by Threadgold and Irwin[6] and Stranock and Halton[13]. These cells have been also denominated as alpha and beta[9,15,16] and G1 and G2[14].

The exceptions are *Schistosoma mansoni*[7,20] and *S. margrebowiei*[10], which possess only one type of secretory cell.

The two types of Mehlis' gland cells observed in *M. macracantha* are similar to those observed in *Diplozoon paradoxum* and *Calicotyle kroyeri* studied by Stranock and Halton[13], *Cichlidogyrus halii typicus* by El Naggar, Khidr and Kearn[14], *Entobdella soleae* and *E. hippoglossi* by Tappenden and Kearn[15,16]. The S1 cells are predominant and characterized by granular endoplasmic reticulum and Golgi stacks and the secretory bodies are spherical, irregular in shape. The S2 cells contain GER in abundance, few mitochondria and the secretory bodies are dense, showing a crystalline structure, as observed in other monogeneans already studied[13,14,15,16].

The S1 bodies are derived from the fusion of small vesicles probably from the adjacent Golgi apparatus. When first formed, the S1 bodies appear to be empty but, during the maturation process, each S1 vacuole increases in size and its boundary membrane repeatedly invaginates budding off small vesicles which are characteristic of the contents of mature S1 bodies. These bodies are distributed throughout the cytoplasm with the mature form located in the perinuclear region[13].

Based on work on digenean *Halipegus eccentricus*, Holy and Wittrock[9]



Figures 1 – 6 - TEM sections through the Mehlis' gland of *Metamicrocotyla macracantha*. Figure 1. Secretory cell S1, with a large central nucleus (N), with prominent nucleolus (Nu) and homogeneous heterochromatin. Mehlis' gland ducts are also present (D). Bar- 2.5m. Figure 2. S1 cell showing an invagination of the plasma membrane (arrow), mitochondrion (M), granular endoplasmic reticulum (Ger) and secretory bodies (S1). Bar – 1m. Figure 3. Part of an S1 cell, showing the secretory bodies at different stages of development. Bar – 1.1m. Figure 4. Lobed S2 cell. Arrow indicates deep infoldings of all surface. Ducts of S1 cells also present (D). Bar- 2.5 m. Figure 5. S2 cell showing the secretory bodies with crystalline structure (arrow) and the granular endoplasmic reticulum (Ger) at the cell periphery. Bar – 0.5m. Figure 6. Gland duct showing S1 secretory bodies in different stages of maturation and free ribosomes. Bar – 1 m.

proposed a morphological categorization of Mehlis' gland cells taking into account four basic characteristics: (i) shape of GER cisternae; (ii) shape of Golgi; (iii) morphology of the secretory bodies and (iv) presence of secretory body material in the ootype lumen. The alpha cells observed in *H. eccentricus* are similar to the S1 cells observed herein in *M. macracantha* and the beta cells correspond to the S2 cells of the present species. In both species the secretory bodies are morphologically similar and the S2 secretory bodies were not observed within the lumen of the ootype.

The S2 secretory bodies were not observed in the ducts. According to Tappenden and Kearn[16], this is a evidence that these bodies dissociate before the entry the female reproductive tract.

The arrangement of the Mehlis' gland cells in *M. macracantha* disposed in two groups, one on each side of the ootype, is in agreement with the pattern observed in *Calicotyle* kroyeri, while in *Diplozoon paradoxum* they are arranged radially around the ootype[13].

The presence of endoplasmic reticulum and Golgi complexes in the S1 and S2 cytoplasm suggests that assembly and release of secretory bodies by these cells may be continuous.

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Estudos ultraestruturais da glândula de Mehlis de Metamicrocotyla macracantha (Monogenea, Microcotylidae) parasito de Mugil liza (Teleostei)

Resumo

A ultraestrutura da glândula de Mehlis de *Metamicrocotyla macracantha*, parasita de brânquia coletado de *Mugil liza* do Rio de Janeiro, Brasil, foi estudado através da microscopia eletrônica de transmissão. A glândula de Mehlis consiste de dois tipos de células secretoras, S1 e S2, cada uma produzindo um corpo secretor diferente. Os corpos S1 são esféricos, em forma de lamelas e observados em diferentes estágios de desenvolvimentos no citoplasma dessas células. Os corpos S2 são esféricos a ovais com conteúdos densos, apresentando uma estrutura cristalina. O citoplasma das células da glândula de Mehlis apresenta também ribossmas livres, retículo endoplasmático granular e complexo de Golgi, organelas características de células secretoras.

Palavras-chave:
Glândula de Mehlis.
Ultraestrutura.
Monogenea.
Metamicrocotyla macracantha.
Microscopia eletrônica de transmissão.

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