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Light and Electron Microscope Studies on the Cells of the Labyrinth in the "Green Gland" of Cambarus Sp.

Everett Anderson
State University of Iowa

H. W. Beams
State University of Iowa

Light and Electron Microscope Studies on the Cells of the Labyrinth in the "Green Gland" of Cambarus Sp.[1]

By Everett Anderson and H. W. Beams

Introduction

In the course of studying the distal portion of the crayfish nephron tubule (Beams, Anderson and Press, 1956) cytoplasmic inclusion bodies of unknown origin were encountered in the cells of the labyrinth with a peculiar fibrous or laminated internal structure. This paper deals primarily with an investigation of these.

Materials and Methods

The material used in this study was obtained from *Cambarus sp.* The "green gland" was dissected out and cut in small pieces. Some pieces were fixed in Champy's and Reguad's solutions and stained with Delafield's hematoxylin for light microscopic observations. For electron microscopy a few pieces were fixed in a one percent buffered (pH 7.25) solution of osmium tetroxide for approximately 30 minutes. They were washed, dehydrated, infiltrated and embedded in a methacrylate monomer. Thin sections were obtained by use of a glass knife, mounted on an International Minot rotary microtome and subsequently examined with an RCA model EMU-2B electron microscope without the removal of the embedding medium.

Description

Light microscope preparations stained with Delafield's hematoxylin show the epithelium of the labyrinth highly involuted and composed of a single layer of large cells which are in contact with a thin basement membrane. Each cell consists of a relatively large nucleus, basal striations and a number of secretion granules. The apical portions of the cells show a prominent "brush border" and on the surface of this border, in some preparations, may be seen clear droplets.

From electron micrographs it may be observed that the "brush border" is composed of a number of vertically arranged protoplasmic processes (Microvilli) which are extensions of the apical cytoplasm (BB, figure 1). Another feature of the "brush border" is the characteristic bleb formations (B, figure 1).

The basal striations, when observed with the electron micro-

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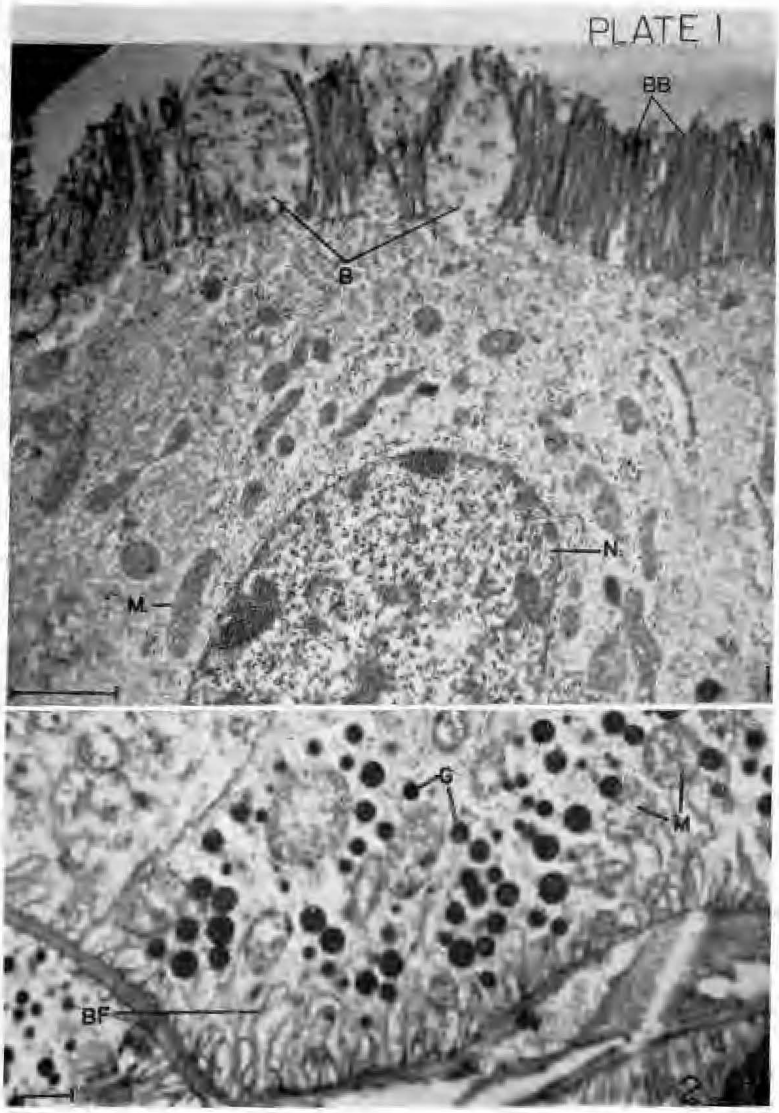

Description of Plates

The line drawn on each figure equals one micron.

Plate I

Figure 1. Section of the apical portion of a labyrinth cell showing the "brush border" (BB), bleb formations (B), nucleus (N) and mitochondrion (M).

Figure 2. Section showing the infolding of the plasma membrane (BF), secretion granules (G), and mitochondrion (M).

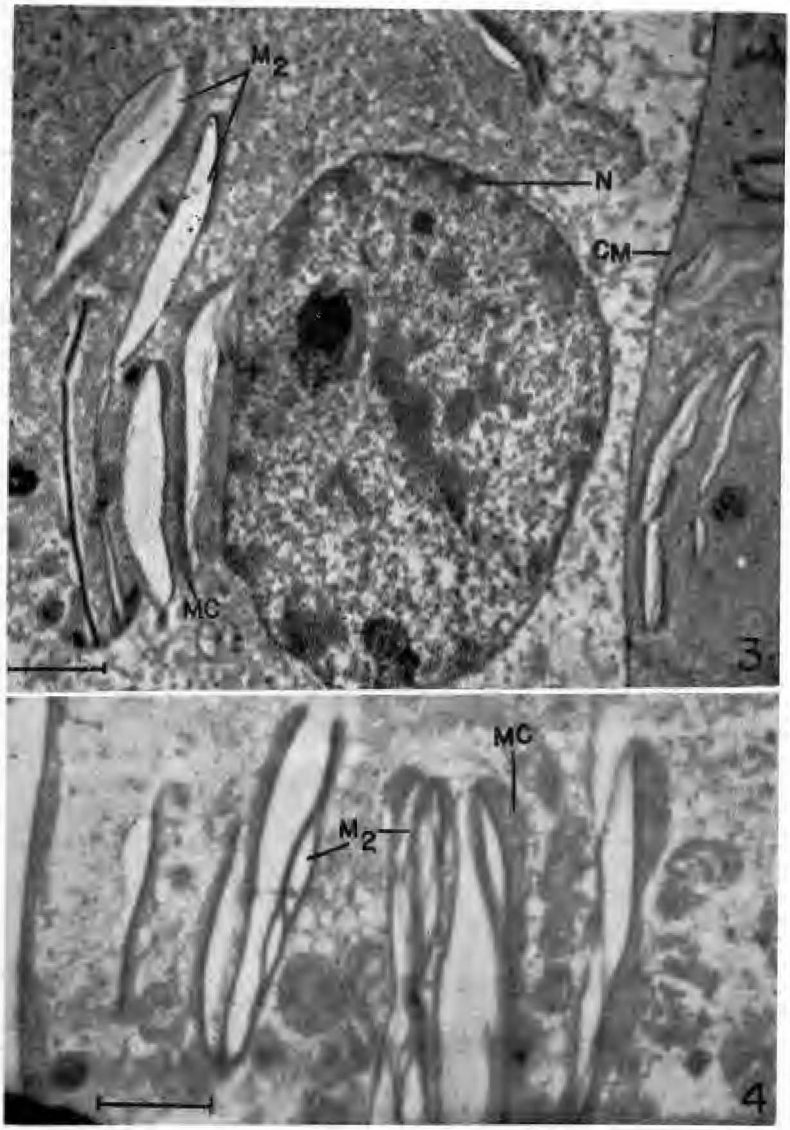

Plate II

Figures 3 and 4. Section through an area between the basal and apical portion of the cell showing cytoplasmic inclusion bodies (M_2) and the cortical area (MC). The nucleus (N) and cell membrane (CM) is shown in figure 3.

scope, are seen as a number of folds (BF, figure 2). These folds are here interpreted as an infolding of the plasma membrane dividing the basal part of the cell into small compartments. This membrane is not related to the endoplasmic reticulum, for this organelle was observed in other electron micrographs, showing no apparent relationship with the basal folds.

Between the apical and basal portion of the cell is observed the relatively large nucleus (N, figure 1 and 3). Likewise, the cytoplasm contains a number of filamentous and spherical mitochondria. These were also observed in preparations prepared in Champy's and Regaud's solutions. The internal structure of a few of these organelles show cristae (M, figure 2), typical of mitochondrial morphology as reported by a number of investigators.

Perhaps one of the most striking characteristics of a number of cells composing the labyrinth are the elongated cytoplasmic inclusion bodies found distributed between the apical and basal portion of the cell. Their number in a cell varies and in some cases they appear to be in close contact with the nucleus (figure 3), but showing no visible effects on its morphology. These bodies appear to have an internal structure composed of fibers or lamellae running longitudinal to the long axis and surrounded by a homogeneous cortical area (MC, figures 3 and 4). It may be pointed out that in light microscope preparations no abnormality was observed among the formed granules or bodies in the cells.

Discussion

It is difficult to correlate the cytoplasmic inclusion bodies observed with the function of the labyrinth of the crayfish kidney. However, some investigators have observed cytoplasmic inclusion bodies in cornea cells of both normal and infected rabbits (Lucus and Herrmann, 1935), and infected ciliated epithelium of the bronchi of mice (Harford and Hamlin, 1952). The origin of these bodies observed in the crayfish is not known. Whether they are pathological is not certain; but when they appear in such great numbers, distorting the ground substance of the cytoplasm, it is difficult to regard them as normal cell organelles. However, these inclusions may represent results of some viral infection.

Existing knowledge of excretion in the crayfish suggests that the labyrinth secrets material outwardly (Maluf, 1939, 1941). This excretion is accomplished by the formation of globules or blebs in the protoplasmic processes of the "brush border" and later pinched off into the lumen. Apparently, the function of the "brush border" of these cells differs from that of other excretory organs of other organisms. However, it should be pointed out that it has been demonstrated by Beams, Tahmisian and Devine, (1955) that mitochondria of the grasshopper malphigian tubules are

excreted by migrating into the protoplasmic processes of the "brush border" which are later pinched off into the lumen. Perhaps some of the secretory granules, cytoplasmic inclusion bodies, and mitochondria may be excreted in this same manner from the cells composing the labyrinth of the crayfish kidney.

Summary

1. The cells composing the labyrinth of the crayfish kidney were investigated with the aid of the light and electron microscope. They consist of a "brush border" composed of a number of protoplasmic processes and a basal part displaying simple infolding of the plasma membrane.

2. Cytoplasmic inclusions of unknown derivation were observed displaying an internal structure composed of fibers or lamellae running longitudinal to the long axis and surrounded by a homogeneous cortical area.

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Department of Zoology
State University of Iowa
Iowa City Iowa