# Acta Medica Okayama

Volume 14, Issue 4

1960

Article 4

DECEMBER 1960

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## **Abstract**

Electronmicroscope pictures of the thin sections of the shell of an ovum of Capillaria hepatica have been demonstrated. The shell is of two layers, an outer one with holes and an inner thick one. The inner layer is composed of several opaque sublapes 2 to 3 microns in width. The plug is 4 to 6 microns in diameter and has a granular zone near the outer surface.

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Acta Med. Okavama 14, 261-264 (1960)

# SUBMICROSCOPIC STRUCTURE OF THE EGG SHELL OF HELMINTH

## III. A STUDY ON CAPILLARIA HEPATICA

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Received for pabulication, May 20, 1960

The morphologic structures of the egg of *Capillaria hepatica* have been studied in detail by many authors. Particularly, ASADA<sup>3</sup> (1925), KAMIYAMA<sup>4</sup> (1928), and BAYLIS<sup>5</sup> (1931) have published several exellent works. All of these researches done by using light microscope show us that the eggs are of barrel-shaped, are provided with polar plugs at both poles, and have a thick shell of double layers, but observations of the submicroscopic structures of the shell and plug have not so far been published. The purpose of the present investigation was to reveal the fine structures of the egg shell of *Capillaria hepatica* with an electron microscope.

## MATERIAL AND METHOD

Small pieces of fresh liver of *Rattus norvegicus* containing the egg of *Capillaria hepatica* (*Bancroft*, 1893) *Travassos*, 1915, collected in Okayama City, were fixed in 1% buffered osmium tetroxide solution at pH 7.2 for 2 hours, dehydrated through a graded series of ethanol, embedded in methacrylate, thin sectioned and observed under electron microscope of Akashi TRS-50B. The procedures are essentially the same as in the previous work<sup>1</sup>. Observations by light microscope have been carried out on the wet samples by the routine method.

# OBSERVATIONS AND DISCUSSION

Under light microscope the eggs of *Capillaria hepatica* appear transparent and colorless and are 40 to 52 by 52 to 27 microns in size. They are of barrelshaped and have two characteristic mucoid plugs at both poles which do not protrude out of the surface of the egg shell. These eggs have a transparent shell of two layers, outer and inner ones. Many radial fine lines are found in the outer layer.

Under electron microscope two layers can be distinguished in the shell, less opaque outer one and dark thick inner one. The outer layer is approximately

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0. 25 micron in thickness and has numerous holes arranged radially. These holes contain homogeneous substances some moderately opaque and separated from each other by thin opaque walls, which are connected to the inner membrane and the limiting membrane of the egg shell surface. The inner layer appears as electron dense compact thick line (Figs. 2 and 5), 2 to 3 microns in thickness. This layer is made up of eight opaque zones, arranged in parallel sandwiching less opaque zones, just like the egg shell of *Trichuris vulpis*. Each of these deeply dark zones are 0.06 to 0.08 micron in width and less opaque zones are 0.8 to 0.16 micron in thickness, excepting the outer one which is approximately 0.8 micron in thickness. The deeply dark zones should be much more compact than the less opaque zones that have many ripple figures. The borders of the outer and inner surfaces of this layer are not distinct. The outer surface of this layer is rough with many short processes, which connect limiting membranes of the holes in the outer layer.

The plug is 4 to 5 microns in thickness and 4 to 6 microns in diameter and appears less opaque while the border line of the outer and inner surfaces of the plug is not so clear. However, there are granular zones about 0.4 micron in thickness near the outer surface of plug, which is composed of numerous granules about 0.06 micron in diameter (Figs. 3 and 4) differing from the plugs of *Trichuris*. These observations show that the egg shell of *Capillaria hepatica* are composed of two layers, an outer layer with numerous holes and an inner thick layer. But it is not certain whether or not these two layers correspond to the outer and inner layers respectively seen under light microscope. The structure of the inner layer is the same as the egg shell of *Trichuris vulpis*.

#### **SUMMARY**

Electronmicroscope pictures of the thin sections of the shell of an ovum of Capillaria hepatica have been demonstrated. The shell is of two layers, an outer one with holes and an inner thick one. The inner layer is composed of several opaque sublapes 2 to 3 microns in width. The plug is 4 to 6 microns in diameter and has a granular zone near the outer surface.

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#### EXPLANATION OF PLATES

One micrograph (Fig. 1.) is taken under light microscope and other five micrographs under electron microscope.

- Fig. 1. Living fertilized egg. The egg shell appears transparent colorless and barrel-shaped. The egg shell have two layers, outer and inner ones. It has two characteristic mucoid plugs at both poles which do not protrude out of the surface of the egg shell. ×1200.
- Fig. 2. Section cut longitudinally through both poles of an egg shell. Two layers can be distinguished in the egg shell. The outer layer has numerous holes arranged radially. These holes contain homogeneous substance and separated from each other by thin opaque walls which are connected to the inner membrane and the limiting membrane of the egg shell surface. The inner layer appears as electron dense thick zone. The polar plugs are situated on both poles of the egg shell. ×1000.
- Fig. 3. Section cut longitudinally through the plug of an egg shell. The plugs appear transparent and are less opaque. The border line of the outer and inner surfaces of the plug is not so clear. There are granular zones near the outer surface of plug, containing numerous granules. ×8000.
- Fis. 4. Section cut longitudinally through the plug of an egg shell. The same as in Fig. 3.  $\times 34000$
- Fig. 5. Cross section through a fertilized egg shell. The outer layer has numerous holes, which contain homogenous substance and is separated by thin walls. These walls are connected to the inner and limiting membrane of the egg shell surface. The inner layer is electron dense. ×27000.
- Fig. 6. Section cut longitudinally through a fertilized egg shell. The holes of outer layer contain homogeneous substance. The inner layer is made up of eight opaque zones, arranged in parallel sandwiching less opaque zones which have many ripple figures. The borders of the outer and inner surfaces of this layer are not distinct. ×16000.

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