Optical and scanning electron microscope description of the tapeworm Cotugnia sp. (Fam: Davaineidae) in Streptopelia senegalensis.

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Abstract
Davaineidae is a very large family and contains the most genus of Cestodes, as it contains 54 species of Cotugnia , which was discovered in birds in different places of the world . Isolating this type of tapeworm from small intestine of ***Streptopelia senegalensis*** from Najaf city in Iraq. The optical microscope and the scanning electron microscope revealed. the worm has a square head which contains four suckers in the corners, rostellum containing hooks, neck, mature segments and gravid segments which contain eggs.
Keywords: Cotugnia , tapeworm , Streptopelia senegalensis , Davaineidae , scanning electron microscope.

1. Introduction

Most of the tapeworms of this species Cotugnia [1] are 62 mm long, white in color and have a head, a neck, a mature segment and an immature segment, and the head is square in shape. The head contains four suckers, rounded oval, muscular, found at all angles. The rostellum is large, with an oval shape in the front, and with the rostellum rings, there are rostellum hooks, the rostellum hooks number 100-120, and they are arranged in one circle, after the scolex there is a medium neck, a little wide in front and a narrow back. The mature segment is greater in width than the length, and about five times larger than the long and medium, with a slightly concave lateral edge, as all the body segments contain a double set of genitals. 115-120 [2].
life cycle: Gravid pieces come out to the periphery with feces, and eggs can live for a long period of years. In the intermediate family, such as ants (Pheidole and Tetramorium), beetles Calathus (Amara), grasshoppers, flies, and earthworms, they are infected by eating eggs individually, after which the embryo hatches from a larva from the egg in the intestine of the host intermediate, then the larva turns into a cysticercoid and remains in the body of the intermediate host until it is swallowed by before the definitive host and is activated by the bile of the definitive host, then the cysticercoid attaches to the mucous membrane of the small intestine and then the process of developing body segments begins directly, where the period of infection to the onset of symptoms ranges from two to three weeks [3].
Symptoms and diseases: Internal parasites infect the intestine, causing nodules and acute inflammation in the intestine, and thus the ability of the intestine to absorb nutrients and vitamins is weak by the host [4,5] and studies have shown that infection may cause weight loss for the body, delay in growth, and decrease product Eggs weaken the body's immunity and may cause death.
And the results showed [3] that sp. Cotugnia in the infected intestines of chickens affected the inner layers of the tissues of the digestive system and the effect was in varying proportions in the four layers of the digestive system. The important effects of the parasite were: the blood vessels become congested in the mucous membrane and the outer muscle layer, and a fibrosis bundle appears in the connective tissues (lamina propria). Some of the intestinal glands swell and their lumen expands. The blood vessels expand and the tissues become necrotic in the damaged areas of the villi. A fibrous bundle forms in the damaged part of the villi in the longitudinal section of it, and the epithelial tissue of the villi suffers dissolution and necrosis in parts of it. Other parts showed bundle fibrosis in the villi, neovascularization and detachment of epithelial tissue lining.
Epidemiological: Infection of chickens with parasitic infection leads to the loss of their economic value. Domestic poultry works to complete the life cycle of the parasite in addition to the intermediate host such as earthworms, ants, flies, locusts and beetles [6]. And showed [7] that tapeworms can lead to the majority of lesions that cause the formation of nodules in the intestinal mucosa, inflammation and bleeding. Parasitic diseases are among the important and essential factors in determining the low productivity of chickens and are often neglected because of the rarity of causing death [8,9].
Helminthiasis is a major problem affecting local chickens, and helminths are the cause of deterioration in the productivity and health of birds, especially domestic ones, in different regions of Ethiopia [10]. Cotugnia is one of the most prevalent species in domestic birds, ducks and pigeons, which are C. digonopora and C. digonopora. protogerys and C. fastigata. These are tapeworms that have a significant impact in causing many diseases, and severe infection affects the health of birds [11].

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2. Material and Methods

Sample collection: This study was conducted During February to June 2022, a total of 60 specimens captured and from the store designated for the sale of birds. from Al-Najaf city in Iraq. Birds were examined for the cestode immediately or within a few hours of slathering.

Bird examination: After slaughtering the birds, they were brought to the laboratory of the Department of Life Sciences / College of Education for Girls. The examination process was carried out by dissecting the birds according to the method of [12] by opening the bird's body longitudinally using a very sharp scalpel from the compound passing through the abdomen and chest after removing its feathers. Conducting a macroscopic examination of the alimentary canal to note any damages that may be present on the outer surface of the alimentary canal of the bird, then the alimentary canal was separated from the body and placed in a petri dish containing saline solution (9%) at 37 °C to preserve it in its natural state, and examining the body cavity with a manual magnifying lens in search of parasites Or its larval stages, the alimentary canal was divided into four parts, which are the trachea, liver, esophageal bile sac, crop, gizzard, small intestine, large intestine, and the complex area, so that each part is separate, then each part was opened longitudinally by means of sharp scissors in a petri dish placed on a black background.

After completing the process of opening these four parts, the process of examination and search for intestinal worms begins using a magnifying glass and a dissecting microscope to isolate large and medium-sized worms. As for small-sized worms, they are isolated using a fine needle.

Visual examination: After isolating the different worms, they were classified according to shape and with the help of a dissecting microscope, where their numbers were counted and kept in small plastic bottles containing 70% ethyl alcohol and glycerin after washing them with tap water to remove impurities and mucous materials attached to them.

Tapeworm isolation: The tapeworms extracted from the intestine of the palm treptopia seengalensis were removed and the tapeworms were washed with tap water, then placed in a solution of Vezlji, then I kept a section of worms in a bottle containing an ethical alcohol 70%, in addition to a few drops of kliserin for the purpose of fixing and later dyeing them for the purpose of diagnosing optical microscope. I preserved a section of worms in a bottle containing 10% until the electronic microscope was diagnosed. I preserved a section of worms in bottles containing absolute ethnic alcohol and placed in the strawberries until the molecular diagnosis [13].

Microscopic examination of tapeworms

After measuring the lengths of the tapeworms by means of a ruler, they were cut into suitable pieces and then dyed using the ready-made acetocarmine, where several drops of this dye were placed on the samples by means of a dropper in a watch bottle and with continuous examination of the dyed samples until they acquired the appropriate redness, and in the event that the sample acquired a dye It is shortened by adding several drops of hydrochloric acid (HCL) 10%. It is placed on the dye away from the model, after which the head is isolated from it. As for the rest of the dyed pieces, they were placed between two strips and tied using rubber bands and placed in 70% ethyl alcohol for 24 hours at room temperature.

After that, the compressed forms were opened and placed in ascending concentrations of alcohol: 70% (10-15 minutes), 80% (5-10 minutes), 90% (5 minutes), and then absolute alcohol (100%) for one minute, then into absolute alcohol and xylol at a ratio of (1:1) for one minute, then transferred to xylol for one minute to clarify the internal structures of the parasite. Finally, permanent segments of the parasite were prepared by loading it onto a slide using Canada balsam [14]. Pictures were taken with a digital camera that diagnosed the worms based on the diagnosis of the Natural History Museum and Research Center / University of Baghdad.

Scanning electron microscope examination of worms

Samples were examined at the University of Kufa / College of Science, supervised by Dr. Ahmed Hussein Muhammad Al-antaki. Whereas, after the drying process, the sample is loaded on the sample holders through a double -faced adhesive tape and then placed in the gold cover room that works to cover it by measuring the nanometer to keep the sample from damage due to the enormous energy from the electrons involved from the device. The samples are placed in their places to examine the use of Scanning electron microscope (SEM)/Inspect S50 /FEI company .made in Netherland [15].

3. Results and Discussion

Optical microscope description of the tapeworm Cotugnia sp. isolated during the current study:

This type of tapeworm was isolated from the small intestine of Streptopelia senegalensis. The worm's body length was 18 cm and its width was 12.3 mm. The head (scolex) were square in shape, containing four muscular capsules, the diameter of the size was 16 micrometers, and the head contained a retractable rostellum with a diameter of 11 micrometers, and it contained hooks with a length of 6 µm under 100 x, 40x power (Fig.1), the mature segment is 29 µm long and 90 µm wide. It contains two pairs of reproductive organs and contains two genital openings on each side under 100 x power (Fig. 2). The gravid segments contained eggs, Under

a force of 40x, Figure (3).

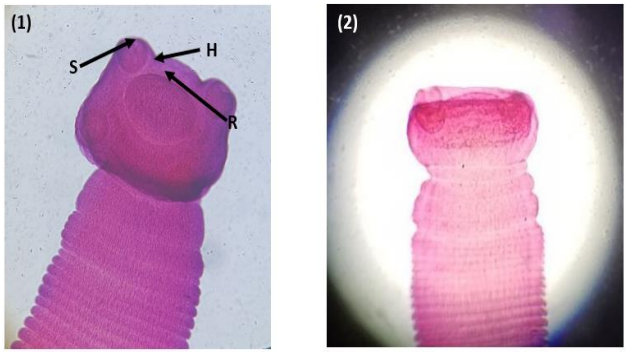

Figure. 1: The scolex of Cotugnia sp. (S) muscular suckers (R) Rostellum (H) hooks, ***Streptopelia senegalensis host*** *(1) magnification 100x (2) magnification 40x*

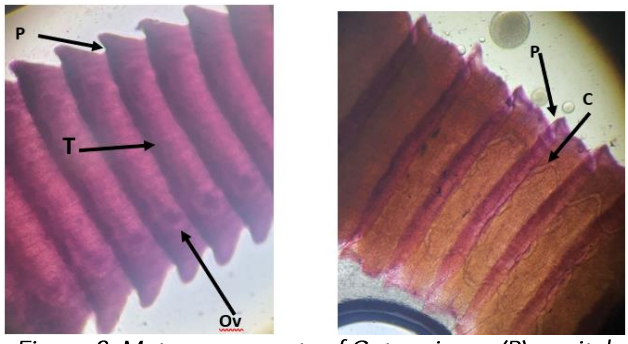

Figure 2: Mature segments of Cotugnia sp. (P) genital pore (T) testes (ov) ovary (C) cirrus, ***Streptopelia senegalensis*** *host. 100x magnification power*

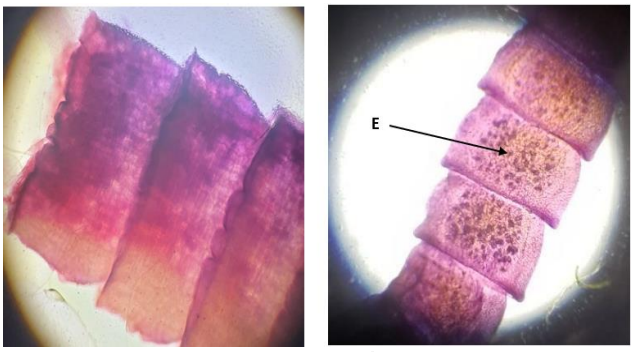

Figure 3: The gravid segments of Cotugnia sp. (E) eggs. ***Streptopelia senegalensis*** *host. magnification 40x.*

Description of the scanning electron microscope Cotugnia sp. isolated during the current study:

Phase pigeons as host Scanning electron microscopy showed the head of Cotugnia sp. Square in shape, containing four muscular receptacles, the length of the retractor is 55 micrometers, and the rostellum is 97 micrometers in diameter, retractablem, Bar=100µm, Bar=300µm, Figure (4). Behind the location of the hooks is a wide circular band measuring 5 micrometers. Numerous very flat, scale-like spines were found, showing bilateral symmetry and a slightly convex appearance, Bar=50µm Figure (5). the mature segment is contains two pairs of reproductive organs and contains two genital openings on each side The segments are rectangular in shape and overlapped with the next piece, The length of the mature segment is 0.8 mm, and its width is 4 mm, Bar=500 mm, Figure (6) and Bar=100, Bar=300 Figure (7) . The gravid segments contained eggs, Bar=4mm, Bar=100µm Figure (8)

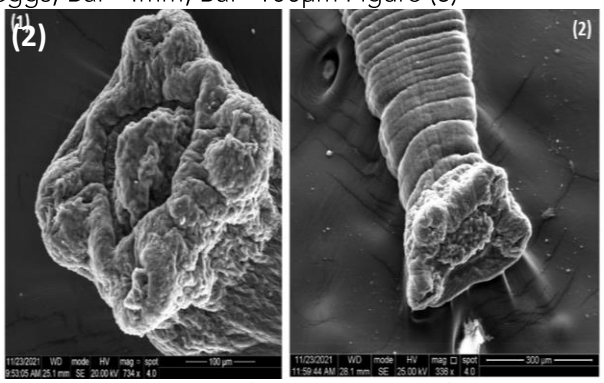

Figure 4: The scolex of Cotugnia sp. (S) four suckers (R) Rostellum, ***Streptopelia senegalensis host*** *(1) Bar=100µm, (2) Bar=300µm.*

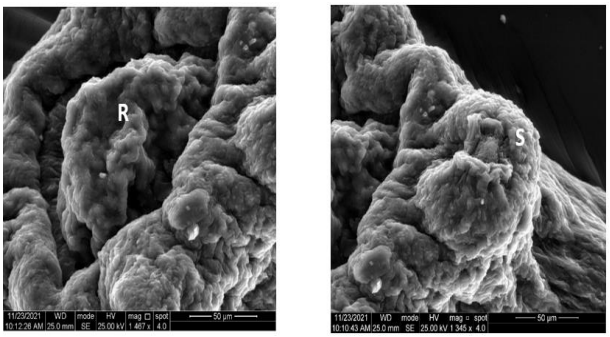

Figure 5: The scolex of Cotugnia sp. (S) four suckers (R) Rostellum, ***Streptopelia senegalensis host****, Bar=50µm,*

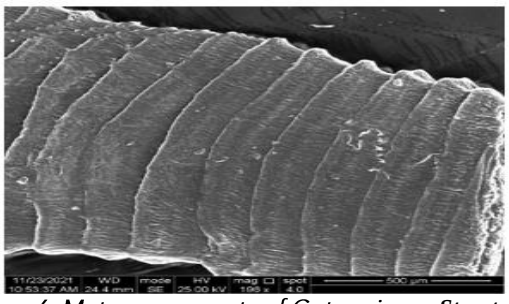

Figure 6: Mature segments of Cotugnia sp, ***Streptopelia senegalensis*** *host, Bar=500µm*

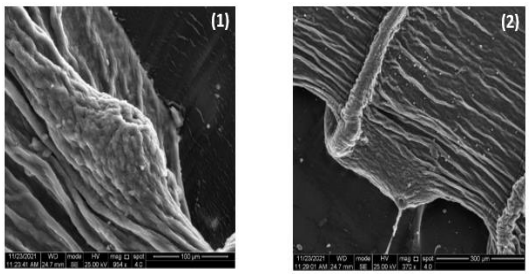

Figure 7: Mature segments of Cotugnia sp. (P) genital pore, ***Streptopelia senegalensis*** *host, (1) Bar=100µm, (2) Bar=300µm*

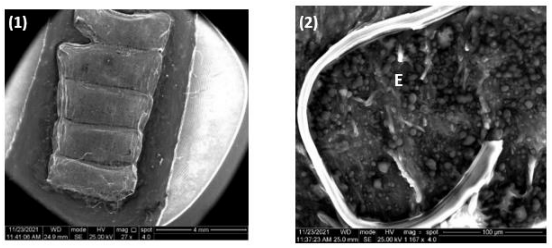

Figure 8: The gravid segments of Cotugnia sp. (E) eggs. ***Streptopelia senegalensis*** *host, (1) Bar=4mm, (2) Bar=100µm*

Examination of the light microscope and electron microscope of the tapeworm Cotugnia sp. In this study, some morphological characteristics were relied on by light microscopy and scanning electron microscopy were used to determine the surface structures of parasites, as they are of great importance in identifying parasites on hand as mentioned [16].

Cotugnia sp., recorded in the current study, is close to the characteristics of the previously mentioned species in terms of the general phenotypic characteristics of the members, but differs from C. digonopora [1,17]. in the presence of head with a diameter of 1.7 mm, a rostellum diameter of 0.16 mm, the presence of many hooks, and a cirrhosis of 0.400 mm in length, which differs from C. polyacontha [18] whose head are 0.47 mm in diameter. , a rostellum diameter of 0.23 mm and a cauliflower 0.182 mm long. The present specimen differs from C.cuneatea [19] in which the head is rounded, with a diameter of 0.28 mm and a rounded rostellum with a diameter of 0.11, and differs from C. joyeuxi [20] in that the diameter of the head is 0.66 mm, the hooks are sloping, and the cirrus sac is 0.078 long. mm. The present form differs from C. parva [20] because of the head 0.48-0.66 x 0.68-0.87 mm in length and width, rostellum drop 0.13 mm, and the icicle 0.198-0.108 mm in length. The worm in this study differs from C. fleari [21] by the presence of a head diameter of 0.47-0.59, and a cirrus sac 0.28-0.33 mm long. The shape of the worm under discussion differs from that of C. bhali [22] in that the head are 0.52 mm in diameter, the rostellum is 0.36 mm in diameter, the hooks are, and the cirrus sac is 0.217-0.225 mm long. It differs from C. intermedia (Johri, 1934) by the presence of a head diameter of 0.46-0.527 mm, a cloaca sac of 0.217-0.227 mm in length. The present form differs from C. noctua [22] in that the head is 0.53 mm in diameter, the rostellum is 0.227 mm in diameter, and the cirrus sac is 0.177-0.300 mm in length. It differs from C. taiwanensis [23], where the head is 0.56-0.77 mm in diameter, and the rostellum is 0.46 mm in diameter. This study differs from C. magna [24] in the presence of a head 0.59-0.65 mm in diameter, and a rostellum with a diameter of 0.287-0.287 mm. 0.316 mm, cirrus cyst 0.239-0.272 mm in length. This page is done

The current study showed that Cotugnia sp. Different from C. polyacantha with a square-shaped head with a diameter of 0.3-0.4 x 0.2-0.3 mm, the presence of four unarmed circular muscular suckers with a diameter of 0.08-0.2 mm, and a large oval rostellum capable of retracting, with a diameter of 0.2-0.3 mm and armed with hooks arranged in two rows, and behind the hooks there are many spines Very flat, scale-like at the base of the hooks, the mature segments have two sets of reproductive organs and measure 0.7-0.9 x 2.7-2.9 mm with two genital openings. Where the genital opening is located randomly in the center of the lateral part on each side, the size of the gestational pieces is 1.5-3 x 5-6 mm, and it is filled with egg capsules and each capsule contains only one egg [25].

It is clear from the above discussion that the species under current study are new to and different from other known species of the genus Cotugnia according to the taxonomic characteristics and the existence of differences. Where, by scanning electron microscope, [26] described for the first time scale-like spines called "cuticle pointed plate" on the rostellum of R. (R) C. micracantha.

The results of the current study showed that Cotugnia sp. It is characterized by a square-shaped area and contains four unarmed circular suckers and a large oval rostellum that is able to contract and is armed with hooks arranged in two rows. Behind the hooks there are many flat spines, as the shape of these spines resembles scales, as they form an indentation at the base of the rostellum, and this is consistent with the description he mentioned [27]. In the current study, the scale-like spines covering the base of the rostellum in the tapeworm Cotugnia sp. By scanning electron microscopy, we believe that the scale-like spines are a distinctive feature found in all family Davaineidae. Further scanning electron microscopy studies of a large number of species and genera belonging to Davaineidae are recommended and necessary to support this study.

4. Conclusions

Tapeworms Cotugnia sp, It infects the small intestine of Streptopelia senegalensis, and by visual examination, we find that this infection led to severe inflammation and bleeding. It was detected by light microscopy and scanning electron microscopy.The morphological characteristics of this worm showed that it has a square-shaped head that contains four cupholes in its four corners and a large rostellum that contains spines. on the eggs.

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