



Intestinal worms parasitic in some types of birds in Tikrit city and their effect on the intestine histological picture

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ABSTRACT

The study was conducted by collecting 756 birds, including 152 birds from *Columba livia*, 138 from *C. oenas*, 113 birds from *C. alumbus*, 144 from *Streptopelia turtur*, and 209 from *S. senegalensis*, for the period from November 2021 to April 2022. The cestodes *Raillietina microcanthata*, *Cotugnia intermedia*, *C. infundibulum*, *Hymenolepis* sp. were diagnosed, and the nematode *Ascaridia columbae*. With varying proportions and severity of infection. As *R. microcanthata* were recorded parasitic in the intestines of *C. livia* by 1.39%, and in *C. oenas* 8.61%, and in *C. oenas* by 3.87%. . . And *C. intermedia* was recorded as an intruder in the small and large intestines of birds (*C. livia*, *C. oenas*, *C. alumbus*, *S. turtur*, and *S. senegalensis*) with varying rates of infection. The infection rate was 1.32% in males and 2.64% in females. It was isolated from pigeons (*C. livia*, *C. oenas*, *C. alumbus*, *S. senegalensis*). The highest infection rate was recorded in females of *C. livia* with a rate of 5.08%, and the lowest infection rate in males with a rate of 0.93%.

Hymenolepis sp. was isolated from each of males and females of the species (*C. livia*, *C. oenas*, *C. alumbus*, *S. turtur*) with an infection rate of 6.93%, 2.89%, 3.87%, 2.77%, respectively. The highest infection rate was in the females of the *C. livia*, at a rate of 5.08%, and in the females of *C. oenas*, at a rate of 2.94%.

A. columbae were recorded in the small and large intestines of pigeons, with an infection rate of 5.23%, 5.79%, 2.84%, and 4.1%, respectively. *C. alumbus* increased by 0.98%.

Where it appeared that there was a wide rupture of the intestinal mucosa due to loss of villi and their shedding with their epithelial cells in intestinal cavity and many white blood cells, macrophages and parasite eggs were found in the main page around the degenerated intestinal glands, its glandular cells. White blood cells around it in the main page, and the intestinal cavity contained masses of epithelial cells degenerated from the villi, with the presence of shrinkage of some mucous intestinal glands with thin-walled blood vessels devoid of blood.

Keywords: *Intestinal parasitic worms, birds, Tikrit city, Iraq, histopathology, including 152 from C. livia, 138 from C. oenas, 113 from C. alumbus, 144 from S. turtur and 209 from S. senegalensis, from Tikrit / Iraq from*

INTRODUCTION

A parasite is an organism that lives on or in another organism (Cabodevilla et al., 2020). Domestic pigeons are raised for hobbies and as a source of food in some countries (Alkharigy, 2018). Both domestic and wild pigeons are potential hidden reservoirs for many human diseases, and they can also transmit parasitic diseases to animals and poultry (Ali et al., 2020).

Domestic pigeons are an important source of essential protein for people as pigeons gain weight more quickly than other birds, are easy to breed and pigeons can be infected with a wide range of pathogens and serve as a reservoir for parasitic diseases. The proximity of pigeons to other domestic birds increases the risk of parasitic infection in poultry (Alkharigy et al., 2018).

Columba livia pigeons belong to the order Columbiformes and the family Columbidae. (Santos et al., 2020).

Cestodes tapeworms require at least one intermediate host before they can complete their life cycle in birds. These worms can cause nutritional deficiencies, weight loss, and diarrhea. Infection can be diagnosed by finding eggs or sometimes proglottids in the stool (Harrison and Lightfoot, 2006).

Severely infected birds with nematodes may show nutrient malabsorption, anorexia, weight loss and diarrhea. In severe cases, the worms can cause intestinal obstruction and death (Harrison and Lightfoot, 2006).

The effect of parasitism is often the tissue damage of the host, either through the penetration of the parasite itself or its larvae between the organs and tissues and multiplying in the host, or fixing itself in the host's tissues by means of fixation, as well as causing blockage of blood vessels and tissue necrosis by infection with tapeworms. Histological changes were recorded in 50 birds represented by degenerative signs and necrosis in the apparent tissue of the villi, and the villi appeared disjointed, torn, and lost their original features, as was the case of chronic inflammation with the accumulation of lymphocytes, plasma cells, and macrophages, as well as the presence of hyperinflation, as recorded (Mohammad and Awad, 2020) infected with two types of nematodes, *Tetrameres* sp and

Microtetrameres spiralis, isolated from some waterfowl, where the pathological examination of the infected tissues showed expansion in the cavity of the endometrial gland, atrophy, necrosis, and degeneration of the glandular cells, and severe cases may lead to the death of birds.

(Rizwana et al., 2019) described the damage caused by the nematode worm *Dasyurotaenia robusta* to the intestines of the house crow *Corvus splendens*, including necrosis and atrophy of villi.

The study aims to diagnose intestinal parasitic worms that infect types of pigeons, the proportions and severity of their infection, and to study the histopathological changes of the intestine resulting from intestinal worms in each type of (pigeon) under study.

MATERIALS AND METHODS

1- Sample collection: Samples were collected during the period from November 2021 until April 2022, which 756 samples of pigeons were collected, which included five species, namely (*C. Livia*, *C. Oenas*, *C. alumbus*, *S. turtur*, and *S. Sengalensis*) that were collected. Collected from Tikrit city / Salah al-Din Governorate, after the process of collecting pigeons, samples were taken to the parasites laboratory in Department of Biology / College of Education for women / Tikrit University, and the process of isolating parasites was carried out.

2- Isolation of endoparasites: The pigeons were dissected after anesthesia with a chloroform solution, then the bird's body was opened longitudinally, starting from the exit area, passing through the abdomen and chest, after removing the feathers from it. Then, the gastrointestinal tract was visually examined, to note if there were damages on its outer surface of the duct. The alimentary canal of the bird was then separated from the body and placed in a Petri dish containing the physiological solution, then opened by scissors and forceps longitudinally in a Petri dish placed on a black background. Worms (Tylor & muller, 1971).

1-2: Isolation of intestinal worms and washing them: The worms were kept in medium-sized plastic bottles containing 70% ethyl alcohol solution after washing them with distilled water to remove the impurities and mucous materials.

2-2: Fixation and staining of tapeworms: The washing process was done with distilled water, then they were placed in the physiological solution, and the process was done after taking their lengths with a ruler, after which they were kept in plastic bottles containing ethyl alcohol at a concentration of 70% for the purpose of protecting them from damage.

Tapeworms were stained with Aceto Carmine Stain (Ash, 1979).

2-3: Nematodes

The nematodes were kept in warm 70% ethyl alcohol and left there until they were liquefied using lactophenol solution, and then loaded onto glass slides using glycerine gel (Ergens et al., 1992).

3- Preparation of histological sections: Histological sections were prepared after taking certain organs from healthy and infected birds, where the organs were fixed after dissection directly with 10% formalin fixative, then the histological sections were prepared according to (Banchroft and Stevens, 1982).

4- Statistical analysis: The results were analyzed statistically by applying the Minitab statistical program using the ANOVA test and the Chi-square test, and the arithmetic means were compared with the (Duncan) multinomial test with a probability level of $0.05 \leq p$. Materials and methods:

RESULTS AND DISCUSSION

The study was conducted by collecting 756 birds, including 152 from *C. livia*, 138 from *C. oenas*, 113 from *C. alubus*, 144 from *S. turtur*, and 209 from *S. sengalensis*, as resident birds in the city of Tikrit / Iraq for the period from November 2021 to April 2022.

R. microcanthata were diagnosed according to (Yamaguti, 1959; Khalil et al, 1994), with a length ranging from 130-170 mm and a width of 800 μ m. The body segments are close in size and long in relation to their width, while the head is smaller than the segments and has a diameter of 190- 210 micrometers, and the head contains receptacles equipped with two rows of hooks, and the snout carries 160-200 canines with a length of about 8 micrometers. The reproductive system consists of 13-16 testicles containing seminal vesicles. The number of eggs per capsule is 5-7 eggs. These worms were recorded

parasitizing in The intestines of *S. turtur* increased by 1.39%, the intestines of *C. oenas* 8.61%, and the intestines of *S. sengalensis* by 3.87%. The highest rate of infection was recorded in the males of *S. turtur* at a rate of 5.63%, and the lowest rate of infection was 0.11% in the males of *C. livia*, as in Table (1).

(Albiaty, 2011) stated that the overall prevalence rate was 73.01%. Three genera of nematodes diagnosed and identified: *Aporina delafondi*, *Cotugnia intermedia*, and *Raillietina*. A study (Shubber et al., 2006) showed that pigeons were infected with *Raillietina* sp. with a rate of 38.94%.

C. intermedia was diagnosed according to (Khalil et al., 1994), and the body length was about 40-50 mm and its width was 3.5-4 mm. The head was equipped with a snout armed with two rows of hooks whose length ranged between 12-14 micrometers. The edge, and the genital openings are bilateral, located in the front half of the edge of the body segment on both sides, the number of testes ranges between 50-90 testicles combined in two groups, and the ciliary sac is 17-25 micrometers long, and the diameter of the egg ranges between 60-80 micrometers.

Parasites were recorded in the small and large intestines of birds (*C. livia*, *C. oenas*, *C. alubus*, *S. turtur* , *S. sengalensis*) with varying infection rates (Table 2). The highest infection rate was recorded in females, with a rate of 5.33%, and males, with a rate of 4.34%, compared to the rest of the species.

In a study (Mizher et al., 2020), *C. cotugnia* parasite was diagnosed with an infection rate of 10%.

C. infundibulum was diagnosed according to (Yamaguti, 1959; Khalil et al., 1994), this type of tapeworm has a length of between 200-250 mm, the head is equipped with a large snout armed with one row of hooks that number 15-20 Clubs 25-30 micrometers long for one dog, and the receptacles are large in size without hooks, and the body pieces at the end of the worm are wider than at the front of the worm, and the genital openings are alternately arranged and open laterally near the beginning of the body piece, and the number of testicles is about 20-30 testicles combined in The posterior section of the mature segment, and the lobed ovary is located in the anterior third of the mature segment.

This species was recorded in the small and large intestines, with an infection rate of 1.32% in males and 2.64% in females. It was isolated from the pigeons (*C. livia*, *C. alumbus*, *S. turtur*, *S. senegalensis*) under study (Table 3). Male hummingbirds by 0.93%.

The intestines of 107 birds of doves in Sulaymaniyah province were examined for intestinal parasites, and the tapeworms *Choanotaenia masculosa* and *Acanthocephalan* spp. were diagnosed. with a prevalence of 8.56% and 13.91%, respectively (Abdulrahman et al., 2019).

Hymenolepis sp. was diagnosed according to (Yamaguit,1961;Calnek et al.,1994), and it is a tapeworm about 20 mm long and 2 mm wide. The body piece, and the testes are few in number, three testicles arranged in the middle of the body piece, and the eggs are spherical in shape, small in size, and contain a six-spined embryo.

These tapeworms were isolated from both males and females of the species under study, which included (*C. livia*, *C. oenas*, *C. alumbus*, *S. turtur*), with an infection rate of 6.93%, 2.89%, 3.87%, and 2.77%, respectively (Table 4). The highest infection rate was in the females of *C. livia*, at a rate of 5.08%, and in the females of *C. alumbus*, at a rate of 2.94%.

In a study (ibrahem et al., 2012), an infection rate of *Raillietina* parasite was recorded at 1.07% in Khorramabad, western Iran, and *Hymenolepis* parasite, at an infection rate of 4.2%, and *Eimeria* parasite, at an infection rate of 7.1%.

A. columbae were diagnosed according to (Yamaguit, 1961; Calnek et al., 1994), where the length of the females is about 40-100 mm and they are white or transparent yellow, and they are characterized by the fact that the mouth opening is surrounded by three lips and the genital opening is located in the center of the body is approximately and the posterior end of the worm is tapering, and the eggs are spherical in shape and have a thick shell that contains many bumps.

This type of nematode was recorded in the small and large intestine regions of the pigeons of the studied species, with an infection rate of 5.23%, 5.79%, 2.84%, and 4.1%, respectively (Table 5).

The infection rate was in female hummingbirds at a rate of 0.98%.

Ascaridia parasite was diagnosed in Saudi Arabia in domestic pigeons with an infection rate of 83%, and in a study (batel, 2020) . *Ascaridia columbae* parasite was diagnosed with an infection rate of 69%, compared to what was diagnosed in Bangladesh with an infection rate of 86.0%, and in India, with an infection rate of 92%.

The results of the study showed the presence of macroscopic and microscopic pathological changes in the affected birds' organs, which showed clinical signs characterized by general weakness, poor appetite, and weight loss in the affected birds. In *C. livia*, a wide rupture of the intestinal mucosa appeared, due to the loss of villi and their shedding with their epithelial cells in the intestinal cavity, and many white blood cells, macrophages, and parasite eggs were found on the main page around the degenerated intestinal glands, their glandular cells. (fig. 1).

The intestinal glands of the doves were surrounded by loose connective tissue, white blood cells, and the smooth muscle layer of intestine that forms the intestinal wall, composed of bundles of internal smooth muscle fibers in a circular direction (Fig. 2). The features of villi did not appear in intestinal tissue with a total destruction of the intestinal glands and the proliferation of some white blood cells around them in the primary page. The intestinal cavity also contained masses of degenerated epithelial cells from the villi, parasitic eggs and white blood cells. (fig. 3).

In the intestines of the infected *C. oenas*, the intestinal villi were destroyed and infarcted from the surface of the intestine. Many epithelial cells have degeneration and sloughing, and many of the intestinal glands have shrinkage and atrophy of their glandular cells, with the presence of inflammatory cells of white blood cells, as shown in fig. (4), and rupture also appeared. Acute intestinal villi and the shedding of many of those cells affiliated with them in the lumen of the intestine with inflammatory white blood cells as those cells were found in the core of villi and the underlying page with parasite eggs as in and the presence of parasite eggs as in fig. (5).

In *S. turtur*, a massive infiltration of white blood cells and macrophages was found on the main page around the intestinal glands, with atrophy of the cells of some of those glands and their shrinkage from the basement membrane on which the gland cells rest. A total shedding of the intestinal villi cells from the surface was observed in the intestines of the infected lunar pigeons (fig. 6). . As for the intestinal mucosa, vestigial villi were found on its surface or the loss of these villi, and the shedding of numbers of epithelial cells in the cavity of the intestine. (fig.7).

In *S. senegalensis* it was found that there was damage to some villi and intestinal glands connected to the base of the villi and lined with glandular cells that secrete yeasts, and between those glands there was infiltration of some white blood cells and parasite eggs, fig. (8).

The results agreed with the researcher Bahrami et al., (2012) who found necrosis in the small intestine of infected pigeons, especially in the villi and the mucosa and musculature of the intestine, as Soulsby and Adang (2008)

mentioned that in many cases, the intestinal mucosa shows inflammatory effects and bleeding resulting On the movement of parasites, based on previous studies, *R. echinobothrida* parasite is a tapeworm that has great economic importance, as it harms the host by destroying intestinal tissues such as villi atrophy, intestinal inflammation, and the formation of granulomas, (Al-Marsomy and Al-Hamadaani, 2016). . Abed et al., noted in (2014) that there are two types of intestinal tapeworms, *Cotugnia* spp. and *Raillietina* spp, which were diagnosed in birds that suffer from an enlarged heart and liver, and some severely infected birds had semi-necrotic intestines with abundant mucous secretions as well as an increase in thickness. Mucous membranes and the presence of hemorrhagic spots in abundance in the intestine with hypertrophy of inflammatory cells. This is attributed to the fact that adult worms stick to the small intestine by scolex and change their location in the intestinal wall continuously due to their migration in the intestinal cavity to obtain food (jasim et al., 2019; jasim et al., 2017).

TABLE 1: Percentage of birds under study infected with *R. microcantha* and by sex

| birdes | female | | | male | | |
|------------------------|--|----------|------|---|----------|------|
| | examined | infected | % | examined | infected | % |
| <i>Columba llivia</i> | 78 | 1 | 1.28 | 84 | 1 | 0.11 |
| <i>C. oenas</i> | 67 | 2 | 2.98 | 71 | 4 | 5.63 |
| <i>S. senegalensis</i> | 102 | 3 | 2.94 | 107 | 1 | 0.93 |
| total | 247 | 6 | 2.42 | 262 | 6 | 2.29 |
| Statistical analysis | ns Chi-Square = 0.633 P-Value = 0.729 | | | * Chi-Square = 4.880 P-Value = 0.047 | | |
| | ns Chi-Square = 1.667 P-Value = 0.435 | | | | | |

TABLE 2: Percentage of birds under study infected with *C. intermedia* and by sex

| birdes | female | | | male | | |
|------------------------|--|----------|------|--|----------|------|
| | examined | infected | % | examined | infected | % |
| <i>S. turtur</i> | 75 | 4 | 5.33 | 69 | 3 | 4.34 |
| <i>C. oenas</i> | 67 | 1 | 1.49 | 71 | 3 | 4.22 |
| <i>S. senegalensis</i> | 102 | 2 | 1.96 | 107 | 1 | 0.93 |
| <i>C. alumbus</i> | 59 | -- | -- | 54 | 2 | 3.70 |
| total | 303 | 7 | 2.31 | 301 | 8 | 2.65 |
| Statistical analysis | ns Chi-Square = 4.337 P-Value = 0.227 | | | ns Chi-Square = 2.465 P-Value = 0.482 | | |
| | ns Chi-Square = 3.277 P-Value = 0.611 | | | | | |

TABLE 3: Percentage of birds under study infected with *C. infundibulum* and by sex

| birdes | female | | | male | | |
|----------------------|--|----------|------|--|----------|------|
| | examined | infected | % | examined | infected | % |
| Columba llivia | 59 | 3 | 5.08 | 54 | 1 | 1.85 |
| C. oenas | 67 | 1 | 1.49 | 71 | 1 | 1.40 |
| C. alumbus | 102 | 3 | 2.94 | 107 | 1 | 0.93 |
| S. turtur | 75 | 1 | 1.33 | 69 | 1 | 1.44 |
| total | 303 | 8 | 2.64 | 301 | 4 | 1.32 |
| Statistical analysis | ns Chi-Square = 2.249 P-Value = 0.522 | | | ns Chi-Square = 0.251 P-Value = 0.920 | | |
| | ns Chi-Square = 0.750 P-Value = 0.943 | | | | | |

TABLE 4: Percentage of birds under study infected with *Hymenolepis* sp. and by sex

| birdes | female | | | male | | |
|----------------------|--|----------|------|--|----------|------|
| | examined | infected | % | examined | infected | % |
| Columba llivia | 59 | 1 | 1.69 | 54 | 1 | 1.85 |
| C. oenas | 67 | 1 | 1.49 | 71 | 1 | 1.40 |
| C. alumbus | 102 | 1 | 0.98 | 107 | 1 | 0.93 |
| S.turtur | 75 | 3 | 5.26 | 69 | 1 | 1.44 |
| total | 303 | 6 | 1.98 | 301 | 4 | 1.32 |
| Statistical analysis | ns Chi-Square = 2.208 P-Value = 0.530 | | | ns Chi-Square = 0.251 P-Value = 0.920 | | |
| | ns Chi-Square = 0.625 P-Value = 0.916 | | | | | |

TABLE 5: Percentage of birds under study infected with *A. columbae* and by sex

| birdes | female | | | male | | |
|----------------------|--|----------|------|--|----------|------|
| | examined | infected | % | examined | infected | % |
| Columba llivia | 59 | 2 | 3.38 | 54 | 1 | 1.85 |
| C. oenas | 67 | 2 | 2.98 | 71 | 2 | 2.81 |
| C. alumbus | 102 | 1 | 0.98 | 107 | 2 | 1.86 |
| S. turtur | 75 | 2 | 2.66 | 69 | 1 | 1.44 |
| total | 303 | 7 | 2.31 | 301 | 6 | 1.99 |
| Statistical analysis | ns Chi-Square = 1.281 P-Value = 0.734 | | | ns Chi-Square = 0.365 P-Value = 0.947 | | |
| | ns Chi-Square = 0.929 P-Value = 0.819 | | | | | |

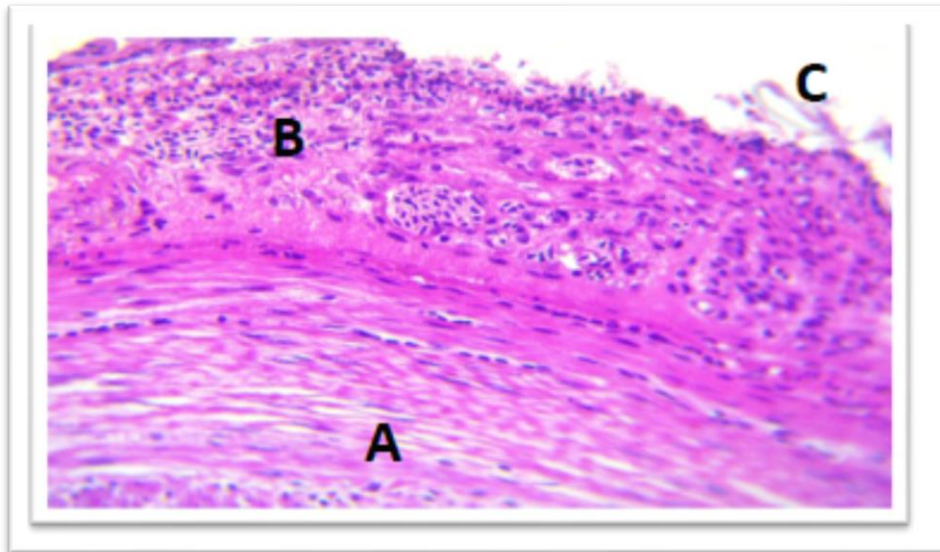


FIGURE 1: intestine of *C. livia*

A: disintegration of the smooth muscle fibers of intestinal wall, B: infiltration of leukocytes from the muscular layer, C: rupture of the serous membrane covering the intestine (H&E, 400X).

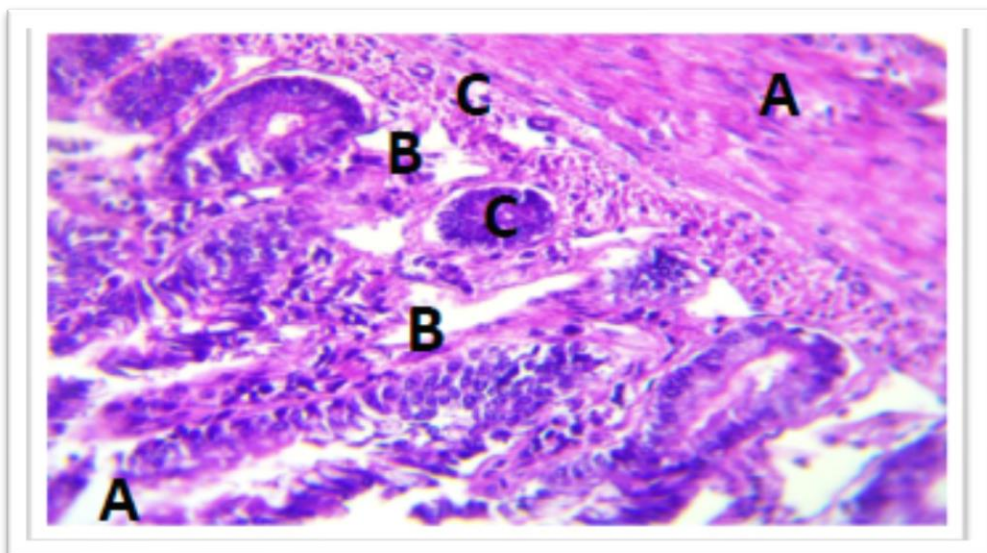


FIG. 2 The intestines of infected *C. oenas*

A: shedding of the epithelial cells lining the villi, B: infiltration of white blood cells between the villi, C: white blood cells containing the menisci between the intestinal glands (H&E, 400X).

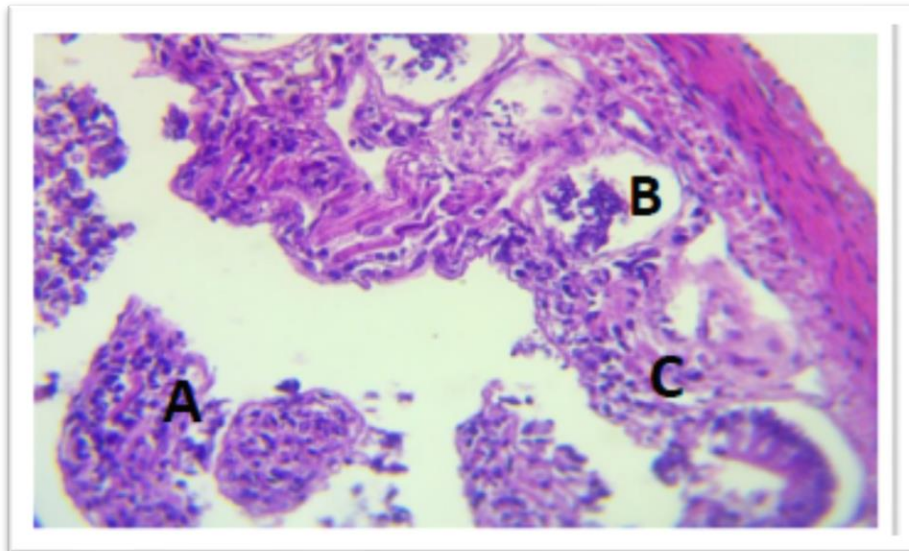


FIG. 3: the intestines of infected *C. oenas*
A: damage to the intestinal mucosa and shedding of the villi in the lumen of the intestine, B: complete degeneration of the cells of the intestinal glands, C: infiltration of white blood cells and parasite eggs (H&E)

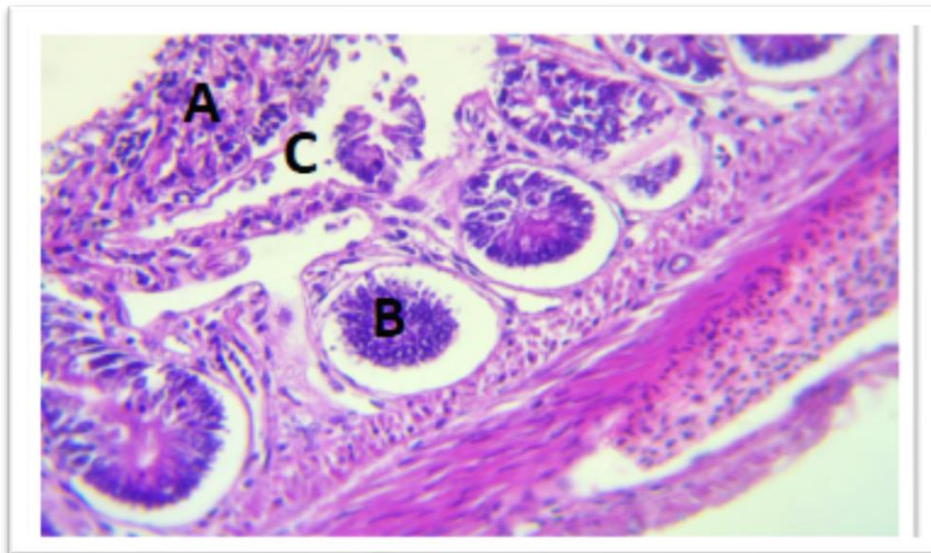


FIG. 4: the intestines of infected *C. alumbus*
A: Breakdown of villi and proliferation of white blood cells and parasite eggs in the pulp of broken villi. B: Shrinkage of the intestinal glands and degeneration of their cells. C: Presence of parasite eggs in the intestinal glands (H&E). (400X)

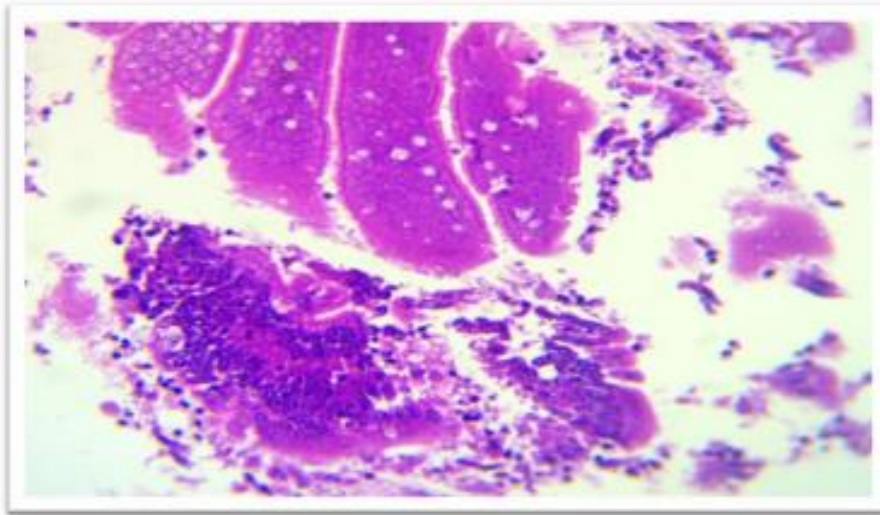


FIG. 5: the intestines of the infected *C. alumbus*
The intestinal cavity contains masses of crushed epithelial cells with white blood cells, mucous masses, and parasite eggs (H&E,400X)

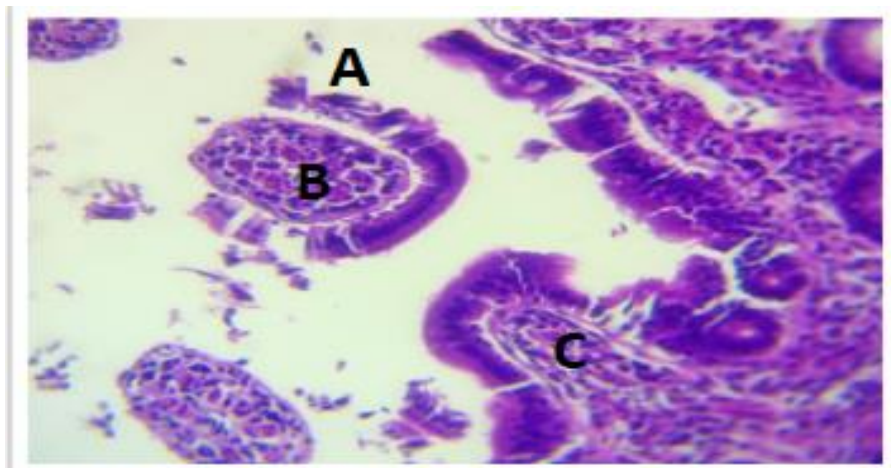


FIG. 6 the intestines of infected *S. turtur*
A: rupture and sloughing of epithelial cells and parts of villi in the lumen of the intestine, B: parasitic eggs, C: infiltration of white blood cells into the villi core. (H&E,400X)

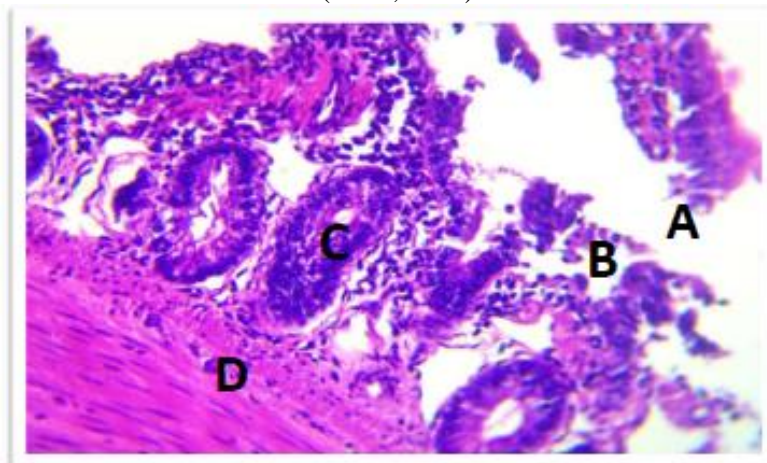


FIG. 7: the intestines of infected *S. turtur*
A: shedding and degeneration of villus cells, B: dispersal of white blood cells, C: degeneration of enteric gland cells, D: parasite eggs (H&E,400X).

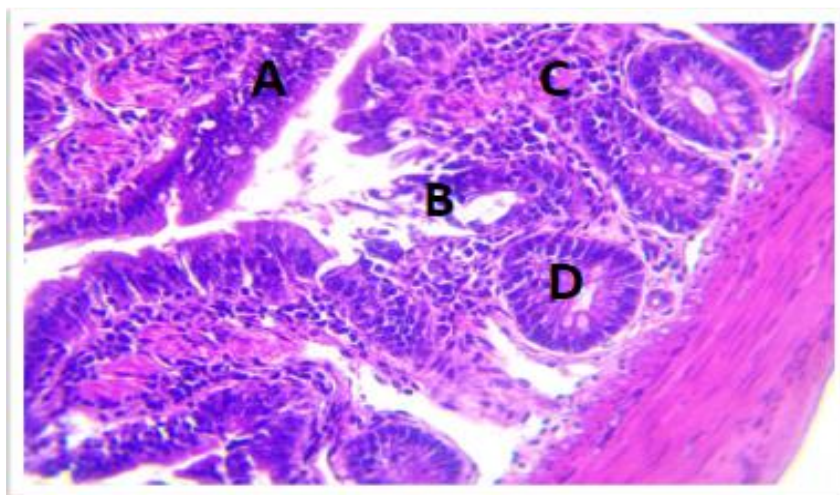


FIG. 8 the intestines of infected *S. senegalensis*

A: villus moulting, B: villus loss, C: infiltration of leukocytes and parasite eggs into the basal part, D: degeneration of the intestinal glands (H&E, 400X).

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