



Histopathological Changes in the Intestine of Infected Pigeons (*Columba livia domestica*) by Helminth Parasites from Al-Qassim, Saudi Arabia

Mashaal A. Aldamigh^{1*} , Amaal H. Hassan^{1,2}, Ahlam A. Alahmadi¹

¹Biological Sciences Department, Science College, King Abdulaziz University, Jeddah, Saudi Arabia

²Zoology Department, Science Faculty, Sohag University, Sohag, Egypt

Email: *ma.aldamigh@gmail.com

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Abstract

It has been reported that parasite infection can significantly harm domestic pigeons. In this study, we wanted to assess the histopathological changes caused by helminth infection. A total of thirty-five domestic pigeons (*C. l. domestica*) purchased from Al-Qassim region in Saudi Arabia were included in this study. Among those, nine pigeons (25.71%) were infected with helminth parasites of a cestode (*Raillietina* species) and a nematode (*Ascaridia columbae*). It was observed that the infected pigeons demonstrated growth retardation, emaciation, weakness, droopiness, and diarrhea. Several histopathological changes were observed in the intestine of infected pigeons including atrophy and distortion of villi, infiltration of inflammatory lymphocytic cells, erosion, and loss of the intestine integrity, necrosis in villi, and blood vessels congestion. We concluded for the first time from Al-Qassim region in Saudi Arabia that the infection with helminth parasites can cause significant histopathological changes in the intestine of the infected pigeons. Consequently, this may lead to a high mortality rate of the infected pigeons. Future studies are required to determine the prevalence and biological factors that have a significant impact on the helminth parasites infection from different regions in Saudi Arabia.

Subject Areas

Animal Behavior, Parasitology, Veterinary Medicine, Zoology

Keywords

C. l. domestica, Histopathological Changes, Helminth Parasites, Intestine,

Saudi Arabia

1. Introduction

Generally, pigeons are considered universal birds (Sari *et al.*, 2008) [1]. Among the domestic pigeons, *Columba livia domestica* is considered the main source of meat and eggs. The domestic pigeons are a host of several ecto- and endoparasites (Marques *et al.*, 2007 [2]; Sivajothi and Reddy, 2014 [3]), including helminth parasites (Ghazi *et al.*, 2002) [4]. Infection with helminth parasites may lead to severe health problems in domestic pigeons.

Previous studies from Nigeria, Egypt, and Iraq have reported cases of pigeons infected with helminth parasites. The prevalence of reported pigeons with helminth infections was 48.3%, 58.3% and 66.3%, respectively (Adang *et al.*, 2008 [5]; Abed *et al.*, 2014 [6]; Ibrahim *et al.*, 2018 [7]). Infection with helminth parasites causes economic losses and serious damage to infected pigeons (Cheng, 1973 [8]; Adang *et al.*, 2008 [5]).

Pigeons normally consume worms and insects that may carry helminth parasite (Soulsby, 1982) [9]. The most significant group of helminth parasites infecting pigeons is known to be nematode; the main genera involve Ascaridia, Syngamus, Capillaria, and Heterakis (Matur and Dawam, 2010). *Ascaridia columbae* is the most frequent parasite-infected pigeons (Tadelle and Ogle, 2001). The main genera of cestode infecting pigeons involve Davainea, Hymenolep, *Raillietina*, and Choanotaenia (Jadhav and Gore, 2004). *Raillietina* sp. is the common cestodes infecting pigeons (Tadelle and Ogle, 2001). Infection of pigeons with helminths can be very harmful causing growth retardation, emaciation, and weakness (Cheng, 1973) [8].

The intestine of the pigeons is a suitable environment for the helminth parasites infection. It supplies the parasites with sufficient food and safe shelter (Bernard and Matthews, 2001) [10]. The infected pigeons can be presented with several symptoms. Some of these symptoms include diarrhea, weakness, and decreased growth. Additionally, the presence of the helminth parasites can lead to obstruction of the digestive tract and hemorrhage in the intestine (Kaufmann, 1996) [11]. As a result, severe damage to the intestinal tissue of the infected pigeons can be presented (Hoste, 2001) [12], leading to increased death (Tanveer *et al.*, 2011) [13].

Therefore, one way to investigate the infection is through the application of histopathology. Histopathological characterization can play a crucial role in diagnosing the infection. Unfortunately, only a few studies have reported the histopathological changes in the intestine of the infected pigeons. Some of the reported histopathological changes include epithelial cell degeneration, destruction of epithelium secretory gland, ulceration, enteritis, and lymphocyte macrophage infiltration (Abed *et al.*, 2014) [6].

In Saudi Arabia, birds including pigeons are considered a source of meat and eggs. Therefore, the aim of this study is to present and characterize the histopathological changes in the intestine of infected pigeons with helminths. To the best of our knowledge, this is the first study to characterize the histopathological changes of infected pigeons and evaluate the prevalence of helminths infection in Al-Qassim, Saudi Arabia.

2. Materials and Methods

2.1. Study Area

The total number of domestic pigeons (*C. l. domestica*) included in this study was thirty-five pigeons. The pigeons were purchased from a local market to sell birds in AL-Qassim region. Al-Qassim region is one of the largest regions in Saudi Arabia. Al-Qassim region is located in the near center of Saudi Arabia. Among the cities in Al-Qassim region, the capital city Buraydah, Ar-Rass, Al-Bukairiah, and Unaizah, the weather in Al-Qassim is a typical dessert climate with rainy winters and hot summers.

2.2. Parasite Identification

The experiment in this study was conducted at the department of Biological Sciences at King Abdulaziz University, Jeddah, Saudi Arabia. We followed the guidelines of the Institutional Animal Ethics Committee at the department of biological sciences in the collage of sciences at the university. All pigeons were directly dissected after purchasing from the bird's market. The pigeons were not maintained nor fed in the facility. Accordingly, the pigeons used in this study did not require any ethical clearance.

For dissecting the pigeons, we followed the procedure described by Al-Hussaini and Demian (1982) [14]. The body cavity of each bird was opened by a longitudinal incision, and the contents of each internal organ were taken out separately in a petri dish with normal saline (0.9%) (37°C). The intestines of pigeons were carefully removed and examined for the presence of helminth parasites. Collected helminth parasites from infected pigeons were isolated and washed several times in saline solution to remove mucus and debris and then prepared for staining. The helminth parasites were identified and confirmed using light and SEM microscopies according to Soulsby (1982) [9].

2.3. Histopathological Examination

The main aim of this study is to study the histopathological changes in the intestine of infected pigeons. Histopathological examinations were conducted on the tissue samples that were obtained from the intestine of the infected and non-infected pigeons. The collected tissue samples were fixed in 10% formalin. After 24 hours, the tissues were washed with 70% alcohol for dehydration. For dehydration, the tissue samples were then serially dehydrated in 70% to 100% alcohol. Next, the tissue samples were cleared in xylene and embedded in paraf-

fin wax. To prepare for examination, thick sections of six-microns were placed on slides and kept on top of 40°C plates. After heating the sections, they were then stained with hematoxylin and eosin. The light microscope was used to examine the fixed tissue sections on the slides according to the helminthological keys of Soulsby (1982) [9] and Yamaguti (1961) [15].

The helminth parasites were fixed in 2.5% glutaraldehyde solution for 24 hours at 25°C. For dehydration, the samples were washed in 0.1 M sodium cacodylate buffer (pH 7.2) and postfixed in 1% osmium tetroxide in the same buffer. The samples were dehydrated in a graded ethanol series. The samples were mounted on aluminum stub and were dried then coated by gold sputtering for 60 s using Auto Fine Coater (JFC-1600) and viewed with a FEI Quanta FEG 450 Scanning Electron Microscope (SEM).

3. Results

A total of 35 pigeons were purchased from the bird's market and examined (Figure 1A). Out of the 35 pigeons examined, 9 pigeons were infected with helminth parasites (Figure 1B). Two helminth parasites species are recorded from the gut of the infected pigeons. They are one cestode species (*Raillietina* species) and one nematode species (*Ascaridia columbae*). Images of both *Raillietina* species and *Ascaridia columbae* species were confirmed using the light microscope (Figure 1E & Figure 1F). No trematodes species were recorded. The overall prevalence of infection was 25.71%. It was observed that the infected pigeons had growth retardation, emaciation, weakness, droopiness, and diarrhea.

As a general observation post dissection, the intestine of infected pigeons appeared to be thickened with mucus excretion compared to the healthy intestine. This thickness is due to the presence of large load of helminth parasites in the infected intestine.

Moreover, several locations in the intestine of the infected pigeons developed hemorrhagic hematoma (Figure 1C & Figure 1D).

Normally, the intestine has a multi-layer structure with mucosal and submucosal layers, as well as circular and longitudinal muscles. Villi, a folded structure, outlines the anterior surface of the intestine. The villi help in increasing the surface range and absorbent area of the intestine (Figure 2A).

In this study, we assessed a cross-section from the intestine of healthy and infected pigeons. The cross-sections demonstrated remarkable histopathological changes associated with the helminths infection. Among the infected pigeons, the infected intestine demonstrated atrophy and distortion of villi and glands, infiltration of lymphocytes, and desquamation of the lining epithelium inside the lumen with erosion (Figure 2B). As a result, the integrity of the typical intestinal structure was lost. Moreover, the wall of the blood vessels was slightly thickened and congested (Figure 2C). At different points, the intestine demonstrated necrosis of the intestinal mucosa (Figure 2D).

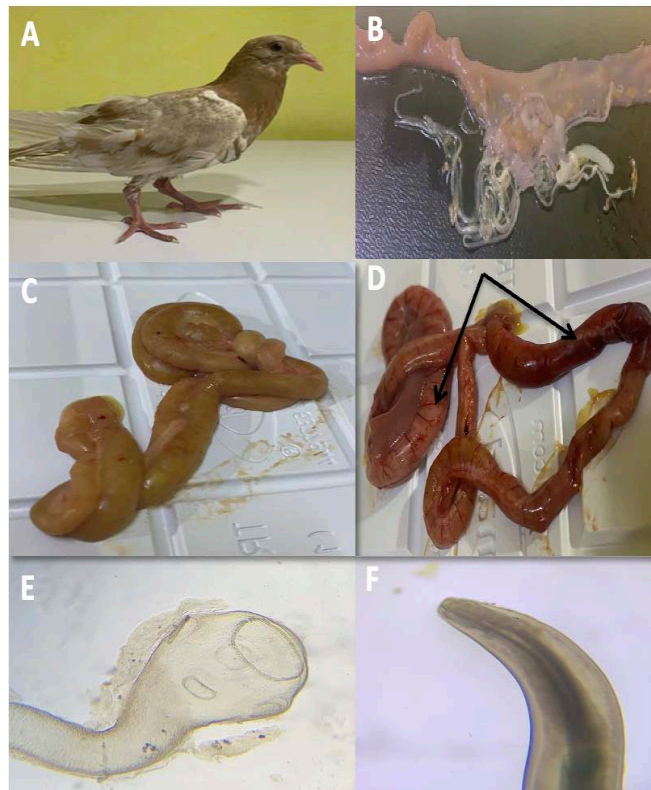


Figure 1. (A) Representative image of an infected domestic pigeons purchased from the bird's market in Al-Qassim region; (B) An image of an intestine obtained from an infected pigeon containing the helminth parasites; (C) An image showing a normal intestine obtained from a healthy pigeon; (D) A representative image of an congested intestine obtained from an infected pigeon showing haemorrhagic haematoma; (E) An image of cestode of *Raillietina* species; (F) An image of a nematode of *Ascaridia columbae* species; the cestode and nematode images were taken under the light microscope (10 \times).

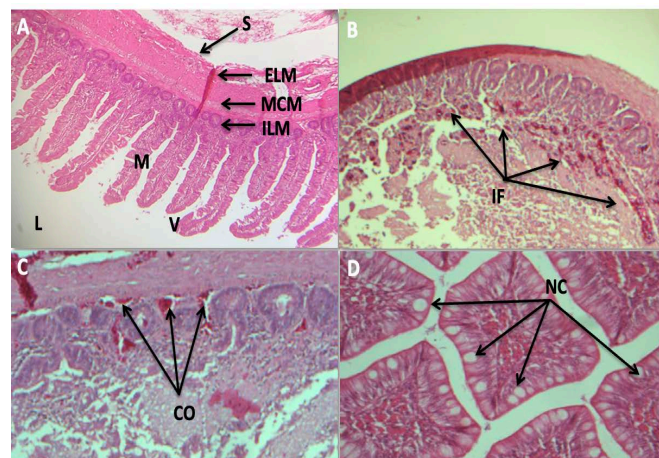


Figure 2. Representative cross sections from the intestine of (A) uninfected pigeon demonstrating several layers, Serosa (S), longitudinal muscle layer (LML), circular muscle layer (CML) and longitudinal muscle layer (LML), mucosa (M), villi (V) and lumen (L); (B) infected pigeon demonstrating distortion of villi and infiltration of lymphocytes (X10); (C) infected intestine showing congested blood vessels (20 \times); (D) infected pigeon with necrotic tissue (NC) (40 \times); H & E stain.

4. Discussion

Domestic pigeons play an essential role in social economy all over the world. In Saudi Arabia, domestic pigeons are considered as a supplementary source of meat and eggs. However, the production of domestic pigeons is hampered by the presence of parasite infection such as helminth parasites. Parasitic infections are responsible for severe health problems in domestic pigeons. The intestine serves as an important host for the helminth parasites. Therefore, assessing the histopathological changes in the infected intestine can play a crucial role in diagnosing helminths infection in domestic pigeons. The main of the study is to assess the histopathological changes in the intestine of the infected pigeons, for the first time, in Al-Qassim region in Saudi Arabia. As previously mentioned, Al-Qassim region is one of the largest regions in Saudi Arabia.

In the present study, the overall prevalence of infection in this study was 25.71%. This is slightly lower than the previously reported prevalence of infection in Nigeria (48.3%), Egypt (58.3%), and Iraq (66.3%) (Adang *et al.*, 2008 [5]; Abed *et al.*, 2014 [6]; Ibrahim *et al.*, 2018 [7]). Other studies also reported a higher prevalence of infection with helminth parasite with over than 96% in Iran (Eslami *et al.*, 2009) [16], and 89% in Morocco (89.9%) (Hassouni *et al.*, 2006) [17], and over 91% in Ethiopia (Eshetu *et al.*, 2001) [18]. The low prevalence rate observed in our study could be due to the small sample size and the limited geographical region where these pigeons purchased. Future studies with larger sample size are required.

The result of this study clearly demonstrates that the infection with helminth parasite can be very harmful. In accordance with previous reports, the domestic pigeons infected with helminth parasites represented some symptoms including growth retardation, emaciation, weakness, and diarrhea (Kaufmann, 1996) [11]. In accordance with previously reported findings (Kamal *et al.*, 2020) [19], the intestines of the infected pigeons were congested by the helminth parasite. This congestion resulted in the development of hemorrhagic hematoma throughout the intestine. Moreover, the blood vessels were congested with mild thickening of their walls. In the light of previously reported studies, these symptoms appear to be common in all infected pigeons with helminth parasites.

Although few studies identified and examined the presence of the parasites using molecular methods, several studies have considered analyzing the histopathological changes in the intestinal tissue of the infected pigeons as a crucial diagnostic tool to confirm helminths infection. Among the several histopathological changes detected in the intestine of the infected pigeons were atrophy and distortion of villi and glands with erosion, losing the typical structure of the intestine, infiltration of inflammatory lymphocytic cells, and increased necrosis of the intestinal mucosa with desquamation of the lining epithelium inside the lumen.

The observed histopathological changes in this study were in line with previously reported histopathological changes caused by cestode species (*R. tetragona*) (Molin 1858) and nematode species (*A. columbae*) (Gmelin, 1780) (Shaikh

et al., 2016) [20]. In this study, other histopathological changes were observed including architectural disintegration of the muscular layer, destruction of Brunner's and crypt glands, necrosis of serosal layer, migratory tunnels formed along with fibrosis, atrophy of villi, and infiltration of mononuclear (macrophages and lymphocytes) in lamina propria. Additionally, the findings from this study of the histopathological changes are consistent with the previously reported findings in the intestines of domestic pigeons (*C. l. domestica*) caused by helminths infection in Egypt (Ibrahim *et al.*, 2018) [7], and the intestines of domestic chickens that infected with the cestode *Cotugnia* sp. from Iraq (Mahdi *et al.*, 2018) [21].

Altogether, the general appearance of the infected pigeons, observation of helminth parasites in the gut of the infected pigeons after dissection, identification of helminth parasites under the light and SEM microscopes as well as the histopathological changes in the intestine of the infected pigeons can serve as confirmatory testing for helminths infection.

It is important to note that the present study has several limitations. The main limitation of this study is the small sample size collected from one site. Additionally, these pigeons were purchased during one month from the same season. It was previously reported that the helminth parasite infection increases during wet seasons. This could have an impact on the prevalence of infection and type of helminth parasite species detected. Another limitation of this study is the inadequate description of the infected pigeons including gender. Moreover, the isolated helminth parasite species were not adequately identified and characterized using molecular techniques. Future studies are required to study the surveillance of helminths infection from different regions in Saudi Arabia as there are huge geographical variations between the different regions in Saudi Arabia. This can have a huge impact on the prevalence as well as the type of helminth parasite species identified.

5. Conclusion

In conclusion, our study clearly demonstrated the histopathological changes in the intestine of the infected domestic pigeons. Based on our knowledge, this is the first study to study the intestine of the infected pigeons with helminth parasites. The infection of domestic pigeons with helminth parasites can lead to an increase in mortality and has severe economic consequences. Future studies are required to determine the prevalence and the key factors that can lead to the increase of helminths infection in Saudi Arabia.

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Conflicts of Interest

The authors declare no conflicts of interest.

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