Research Article

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Comparative Study of the Histopathological Changes of Liver and Lung of Two Lines of laying Chickens Experimentally Infected by *Ascaridia galli*

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Abstract

This work aimed to determine the effect of *A. galli* on the histological changes in the liver and lung of two distinct lines of laying chickens and study the resistance of each line. In this study, fifty laying hens 8 weeks of age were used, (25 of which were Lohmann brown-classic and 25 of which were Lohmann lsl-lite). Each line was divided into two groups: the infected group (N=15) these chickens were infected with a single dose of 500 embryonated eggs through oral inoculation, and the second group control group (N=10) was given normal saline. After 8 weeks the chickens were killed to examination the histological changes. The results showed a histopathological lesion of the liver of the Lohmann brown-classic infected group showed fatty degeneration, blood vessels congestion and the fibrin and inflammatory cells in the lumina of the dilated veins these inflammatory cell infiltrations include eosinophil, while Lohmann lsl-lite showed congestion of blood vessels, as well as lymphocyte infiltration of the inflammatory cells. The lung of Lohmann brown-classic showed congestion in inflammatory cell blood vessels and infiltration with alveolar congestion and eosinophils inflammation and necrotic patches, whereas histopathological changes of the lung of Lohmann lsl-lite showed eosinophils proliferation within an alveolar wall, deterioration of their lumina as a result of haemorrhage. While the control group of two-line showed normal liver and lung. Concluded that the *A. galli* had an effect on the histological structure of the liver and lung, additionally, Lohmann lsl-lite was more affected and less resistant compare to Lohmann brown-classic.

Keywards: *Ascaridia galli,* laying hens, liver, lung, histopathology.

دراسة مقارنة للتغيرات النسيجية المرضية للكبد والرئة لنوعين من الدجاج البياض المصابة تجريبياً بالإسكارس

تهدف الدراسة الى تقدير تأثير الاسكارس *Ascaridia Galli* على التغيرات النسيجية في الكبد والرئة لنوعين مختلفين من الدجاج البياض ودراسة مقاومة كل نوع. في هذه الدراسة ، تم استخدام خمسين دجاجة بياضة عمرها 8 أسابيع ، (25 منها كانت Lohmann Brown-Classic و 25 منها Lohmann lsl-lite). تم تقسيم كل نوع إلى مجموعتين: المجموعة المصابة (N = 15) تم اصابتها بجرعة واحدة من 500 البيوض الحاوية على الكتلة الجنينية عن طريق الفم ، والمجموعة الثانية مجموعة السيطرة (N = 10) أعطيت محلول ملحي. بعد 8 أسابيع تم قتل الدجاج لفحص التغيرات النسيجية. أظهرت النتائج وجود تغيرات مرضية نسيجية في الكبد في مجموعة Lohmann Brown-Classic المصابة والتي أظهرت تنكسًا دهنيًا واحتقانًا في الأوعية الدموية وخلايا الفبرين والتهابات في الصفحة اللبادية والأوردة كانت متوسعة ،و كان هناك ارتشاح خلايا حمضات ، بينما أظهر Lohmann lsl-lite احتقان الأوعية الدموية وكذلك ارتشاح الخلايا الليمفاوية للخلايا الالتهابية. أظهرت رئة مجموعة Lohmann Brown-Classic احتقانًا في الأوعية الدموية للخلايا الالتهابية مع احتقان الاسناخ وارتشاح خلايا الحمضات وضهور بقع نخرية ، في حين أظهرت التغيرات النسيجية المرضية لرئة Lohmann lsl-lite زيادة في ارتشاح خلايا الحمضات داخل الجدار سنخي ، وتنكس في الصفحة البادية نتيجة النزيف. بينما أظهرت مجموعة السيطرة لكلا النوعين انسجة طبيعية لعضوي الكبد والرئة. نستنتج إلى أن الاسكارس *Ascaridia galli* كان له تأثير على التركيب النسيجي للكبد والرئة ، بالإضافة إلى أن Lohmann lsl-lite كان أكثر تأثراً وأقل مقاومة مقارنةً بـ Lohmann brown-classic.

Introduction

Ascaridia galli is the most frequent parasitic nematode in birds all over the world. They infect a variety of birds, including chickens, geese, guinea fowl, turkeys, and other wild birds (1,2,3). However, the chicken is most likely the most commercially and epidemiologically significant host (2). *A. galli* is a serious threat, the adults are found in the small intestine, and their eggs have a high amount of resilience, making it difficult to totally remove them once they have entered a particular environment (3). According to reports, *A. galli* (22 to 84 %) is the most common helminth parasite in free-range chickens (5,6) . The frequency of helminthiasis infection in chickens was found to be quite high, with *A. galli* infection accounting for 31% of the total (7). An abundance of parasitic infections continue to affect chicken production, according to statistics from the Food and Agriculture Organization (FAO). Typically, *A. galli* worm infection results in significant damage to the gut during the worm migration phase. This migration happens in the intestinal mucosa and results in hemorrhagic enteritis, as well as decreased digestion and nutritional assimilation, which affects the epithelium's mucin synthesis (8). The migration of the larvae during the tissue phase of the life cycle may be linked to the histopathological effects, particularly hemorrhagic lesions in the liver and lungs (9). Fatty degeneration and areas of coagulation necrosis of the hepatic cells were found in the livers of infected birds, most notably in the portal sections. In the necrotized sections, there were mononuclear and polymorphonuclear cellular infiltrations. The sick birds' livers had congested blood vessels and sinusoids, while their lungs had hemorrhagic regions, congested blood vessels, and haemosiderosis, the peribronchiolar and interalveolar septae had mononuclear and polymorphonuclear cellular infiltration, which extended and filled certain alveoli (10). The liver was microscopically examined and revealed an inflammatory cell infiltrate surrounding the portal veins, mainly consisting of mononuclear cells, as well as subcapsular haemorrhage, a fatty deterioration was discovered in the livers of some afflicted birds. Interstitial edoema and degenerative changes in the tubular epithelium were the most prominent histological changes in the kidneys, which were primarily associated with an intensive lymphocyte and macrophage infiltration of the interstitium (multifocal interstitial nephritis) (11) The histological changes in the liver of bird, which negatively affects its vital functions, and effect on the productivity of bird (12).

Chicken genotypes fluctuate in their sensitivity to *A. galli*, and various isolates of *A. galli* have variable infectious abilities, according to the assumptions. Due to the genetic development of the immune system, resistance to diseases is a common occurrence in all animal species, and individual variances in chickens (13). On the other side, few research have investigated at genetic resistance to illness in indigenous chicken breeds, and little is known about genetic vulnerability to parasite infections in hens, especially helminth infections. The main objective of this study was to assess the resistance of two commercial lines (Lohmann white & red) against *A. galli* infection.

Materials and Methods

Preparation and maturation of *Ascaridia galli* eggs:

Adult female of *A. galli* were collected from infected chickens. The eggs were extracted from crushed female worms in distilled water using a mortar and pestle, and filtered into a beaker with a mesh sieve of 0.01

mm diameters. The sediments were rinsed in a beaker with a 0.5 N sodium hydroxide solution. Then the eggs were incubated for two weeks at 30 C° in a 0.1 % (w/v) potassium dichromate ($K_2Cr_2O_7$) (14).

Experimental animals

Fifty laying hens 8 weeks of age were used, (25 Lohmann brown-classic and 25 Lohmann lsl-lite), each line was divided into two groups, infected group (N=15) were infected with a single dose of 500 embryonated eggs through orally inoculation by using a plastic pasteur pipette according to (15). and control (N=10) group giving normal saline.

Tissue Sample collection processing:

Tissue sample of liver and lung were collected after chicken killed and dissected according to the ethical protocol of the College of Veterinary Medicine, University of Baghdad, using cervical dislocation. Sample kept in container with 10% formalin for histopathological examination. The fixed tissue samples were cut and processed in an automated tissue processor in separate chambers containing varied alcohol concentrations, (70, 80, 95 and 100%, 100%, 100%, 100%), then cleaned in xylene and embedded in paraffin for fine block production. haematoxyline and eosin (H & E) was used to stain the blocks, which were cut at 5 m, dewaxed, rehydrated, then dewaxed again. Then mounted on canada balsam and left to dry, the slides were examined using a light microscope (16).

Results and discussion

liver examination

Histopathological examination of liver of Lohmann brown-classic infected group showed fatty degeneration, blood vessels congestion and the fibrin and inflammatory cells in the lumina of the dilated veins these inflammatory cell infiltration include eosinophil (fig. 1), While the Histopathological changes of liver of infected Lohmann lsl-lite showed congestion of blood vessels, as well as lymphocyte infiltration of the inflammatory cells. blood vessels filled with inflammatory cell. There was significant necrosis of the hepatocytes, as well as widespread regions of haemorrhage and necrosis- pyknotic nuclei (fig. 3). Furthermore liver of the control group of each group showed a healthy tissue (fig. 2 and 4).

lung examination

Histopathological changes of lung of Lohmann brown-classic showed blood vessels congestion inflammatory cell infiltration with alveolar congestion and eosinophils inflammation and necrotic patches with consolidation contents were generally observed (fig. 5) Whereas Histopathological changes of lung of Lohmann lsl-lite showed eosinophils proliferation within alveolar wall, deterioration of their Lumina as a result of haemorrhage, In addition, there was severe congestion of the pulmonary blood vessels and extensive smooth muscle Hypertrophy (fig. 7). However, the lung of the control group of each group showed a healthy tissue (fig. 6 and 8).

Histopathological examination of liver of Lohmann brown-classic infected group are agree with (17), In the studied liver and lungs, histology indicated bleeding, clogged blood vessels, fatty degeneration, necrosis, and leukocytic infiltration. While the Histopathological changes of liver of infected Lohmann lsl-lite were agree with (18) who investigated the laying hens intentionally infected with *A. galli* indicated significant pathological abnormalities in the liver and lungs of the animals. These lesion due to *A. galli* infection the other study that conducted on the Broiler artificially infected by *A. galli* postmortem examinations revealed severe pathological lesions infected liver. The infected liver had a reddish color. Exudate from a lung lobe showed signs of severe congestion and persistent pneumonia (19). Furthermore liver of

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the control group of each group showed a healthy tissue (fig. 3 and 4).

The finding of current study of Histopathological changes of lung were in agreement with (17), who investigated the laying hens experimentally infected with *A. galli* have pathological abnormalities in the lungs. These lesions are related to larval migration throughout the tissue phase of the life cycle. Larvae stage III migrated up and down the intestinal lumen including liver and lung (20). This results were agreement with (10). Moreover, when compared to Lohmann lsl-lite, Lohmann brown-classic showed better resistance and fewer lesions. These return to the genetic structure, the genetic background determined the immune responses and infection level (21). The diversity in genetic structure that is acquired by prolonged environmental adaptation promotes disease resistance (22).

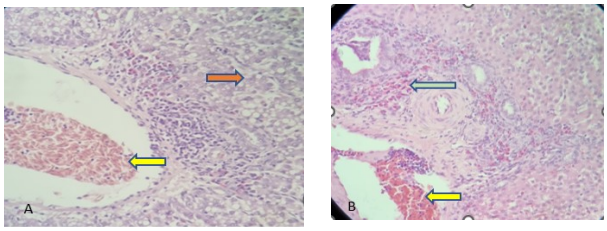

Figure 1. Histopathological image of liver Lohmann brown-classic by infected *A.galli* (infected group) (Central area A and portal area B) from inf-1 group showed fatty degeneration (Red arrow), blood vessels congestion(yellow arrow) and inflammatory cell infiltration mainly eosinophil (Green arrow). H&E X40

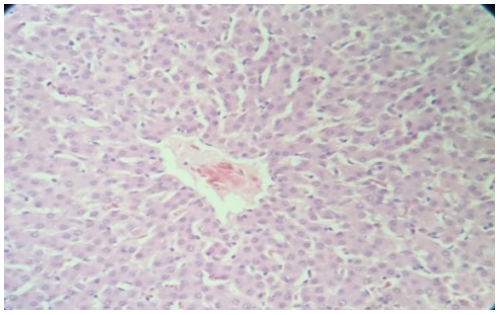

Figure 2. Histopathological image of liver health Lohmann brown-classic control group showed normal histology architecture. H&E X40

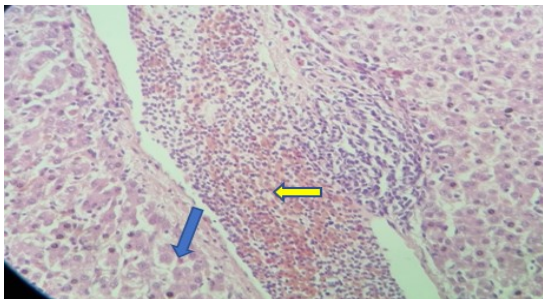

Figure 3. Histopathological image of liver infected Lohmann lsl-lite by *A. galli* (infected group) showed blood vessels filled with inflammatory cell (yellow arrow), hepatocyte with necrosis - pyknotic nuclei (blue arrow) H&E X40

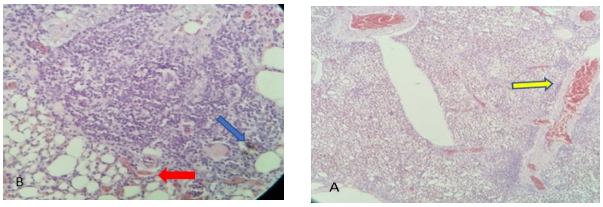

Figure 5. Histopathological image of lung of infected Lohmann brown-classic by infected *A. galli* (infected group) showed blood vessels congestion(yellow arrow), inflammatory cell infiltration with alveolar congestion (red arrow) and eosinophils (blue arrow) H&E A X10 and B X40

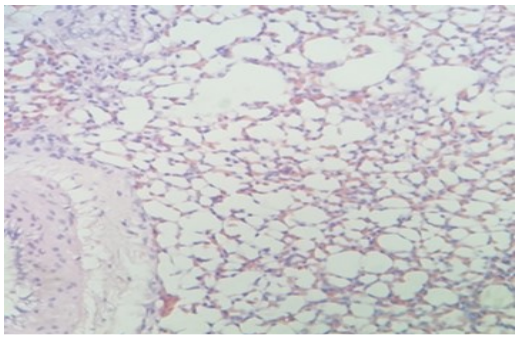

Figure 6. Histopathological image of lung of healthy Lohmann brown-classic control group showed normal histology architecture. H&E X40

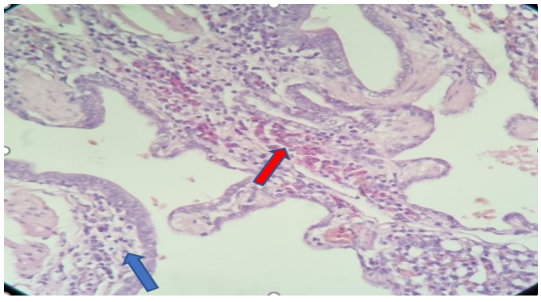

Figure 7. Histopathological image of lung infected Lohmann lsl-lite by *A. galli* (infected group) showed eosinophils proliferation within alveolar wall (red arrow) and smooth muscle Hypertrophy (blue arrow). H&E X40.

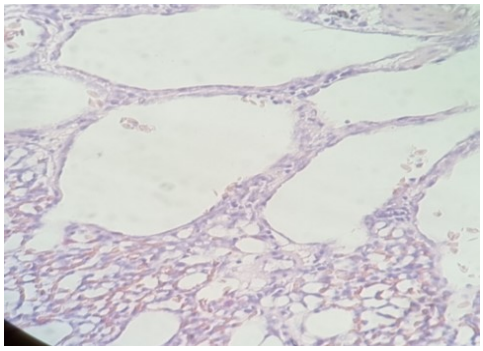

Figure 8. Histopathological image of lung of healthy Lohmann lsl-lite control group showed normal histology architecture. H&E X40

Conclusion

Concluded that the *Ascaridia galli* had effect on histological structure of liver and lung, additionally, Lohmann brown-classic recorded more resistance and less lesions compared to the Lohmann lsl-lite.

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Conflict interest

There are no conflicts of interest with this manuscript's publishing.

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