

## Clinical and histopathological study of inclusion body hepatitis in broilers in Basrah governorate

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**Abstract** Inclusion body hepatitis outbreaks and hydropericardium hepatitis syndrome, particularly in commercial broiler flocks, have been detected in various geographic regions of Iraq over the past five years, indicating the widespread distribution of FAdVs throughout our nation from the north to the south. The Basrah governorate in Iraq has seen significant economic losses as a result of elevated mortality rates and poor performance in chicken farms. 180 cases of sick and fresh dead birds, ages 17 to 28 days, were removed from broiler chicken farms located throughout the Basrah province between 2023 and 2024. The goal of this study was to examine the postmortem lesions and clinical findings of suspected IBH, as well as to obtain histopathological sections of the affected tissues (liver, kidney, and heart). Acute symptoms included ruffled feathers, anorexia, depression, low body weight, inconsistent flock size, and a high mortality rate (10–25%). pale and straw-colored fluids in the pericardial sac, enlarged and hemorrhagic kidneys, and an expanded, congested spleen are other symptoms of an enlarged, friable liver with yellow muroid dropping as a result of excess bile acids and lesions identified during the postmortem investigation. Furthermore, according to a histopathological examination, the infected chicken's damaged kidneys showed evidence of renal tubular epithelium vacuolation, interstitial tissue hemorrhage, infiltration of inflammatory cells, and severe hepatitis with intranuclear inclusion bodies and degenerative changes. The macrophage was particularly well-represented among these cells. The study concluded that loss of biosecurity and vaccination programs against IBH infection in poultry populations particularly in Basra governorate make them more prone to infection.

**Keywords:** Avian adenovirus, Basrah Province, broilers, Inclusion body hepatitis

**Introduction** The avian adenovirus, which belongs to the adenoviridae family and infects a variety of poultry including falcons, raptors, parrots, and ostriches, is known as the fowl adenovirus (FAdVs), and it reflects several syndromes in the production of chickens, including inclusion body hepatitis (IBH) and hepatitis hydro pericardium syndrome (HHS). However, the primary hosts of the virus are chickens initially (1). FAdVs are categorised into 12 serotypes (FAdV-1 to 8a and 8b to 11) and five species (FAdV-A to FAdV-E) based on the cross-neutralization assay. FAdVs are members of the Adenoviridae family of viruses. (2, 3). Other bird species with documented cases of IBH include geese (4), turkeys (5), pigeons (6, 7), psittacine birds (8, 9). IBH and HHS were found in the liver and pancreas of three chicken samples, and

were reported in numerous countries, including Egypt (10, 11), Iraq and Japan (12), and India (13). The liver's grossly visible signs include pallor, enlargement, and tiny foci. Additionally, glomerulonephritis is linked to kidney swelling (14). Histological examination of the livers revealed severe hepatic damage with presence of large basophilic intranuclear inclusion bodies (8). At necropsy, turkeys appeared anemic and had pale yellow livers. Histopathologic examination of affected livers revealed diffuse hepatic degeneration and multifocal necrosis, with approximately 70% of the hepatocytes containing large, basophilic, intranuclear inclusion bodies (5). Broilers often get IBH between three and seven weeks but reports of cases date back up to 20 weeks (12). In hens, the illness is usually detected when the death rate ranges from 2 to 40%. It is typical to observe high death rates in hens under

three weeks old. Owing to the virus's virulence, fatality rates in cases of secondary co-infection have been recorded to reach 80%. The maximum death rate often occurs in three to four days and declines in nine to fourteen days. The purpose of the current study was to examine the histological effects of IBH in broiler chickens. In the late periods after increase poultry industries in Iraq, there are Cases of digestive systems problems associated with depression and mortality especially in young chicks. The primary diagnosis based on clinical signs and case history addition to postmortem lesions were adeno virus infections, therefore the present research aimed to study clinical and postmortem finding along with histopathological changes caused by such viruses in broilers in Basrah province

#### Materials and methods

##### Ethical approve

This study was approved for the animal care at the College of Veterinary Medicine, University of Al-Qadisiyah, Under the No. 4955 dated 17/11/2024

##### Samples collection

The samples used in the present study (60 samples) were collected from different location of seven broiler flocks infected with FAdV in Basrah Governorate, Information regarding the broiler flocks was recorded and included the following: location of the flocks, date of infection with the disease, age, number of birds, motility rate, and use of IBH vaccine. Formalin-fixed tissue block samples previously tested positive with Fowl Adenovirus serotype 8b in an infected broiler flock in Basrah Governorate, detected, and submitted to GenBank under accession numbers PP860569.1 and PP860570.1 in a previous study by (15), and were used for histopathological analysis in the present study.

##### Histopathological preparation

Liver, heart, and kidney fixed by using 10% formalin (BDH / limited Poole /England /40%), formalin block samples dehydrating by using ethyl alcohol (B.D.H (UK) / 95%) passed through ascending concentrations (70-80-90-100), the clearing by xylene (BDH / limited Poole /England) was done by passing the specimens in two steps of pure xylene (5-10) minutes for each step, the tissue embedded in paraffin wax (BDH / limited Poole /England) at (45) degree, the tissue was stained with hematoxylin and Eosin based on (9).

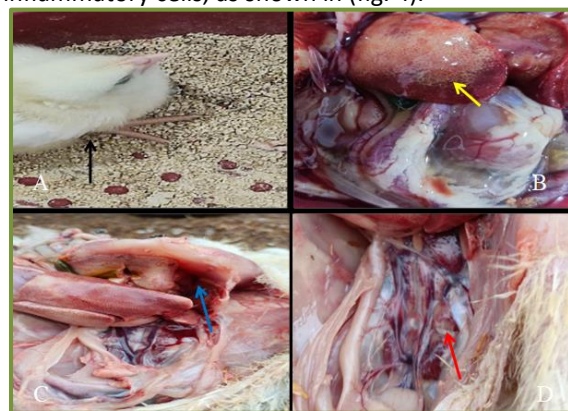
##### Clinical finding and gross lesions

Broiler chicks under three weeks old from the affected farms under study experienced sudden death with the course of the disease extended to 5-7 days. The

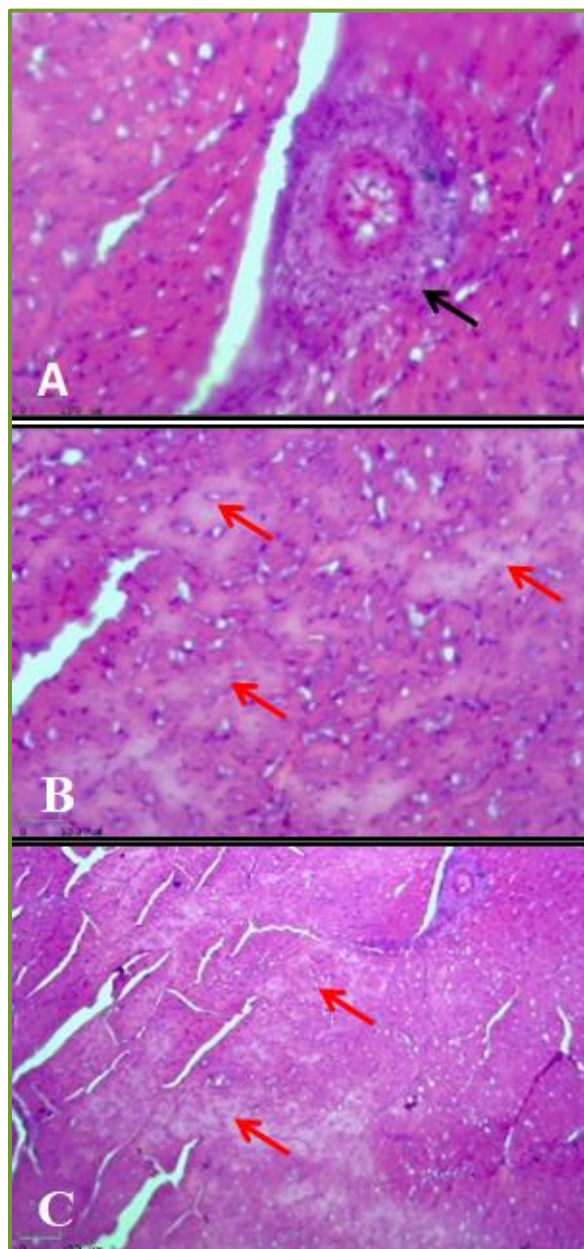
infections with IBH had a mortality rate ranging from 2% to 10%. The unexpected death of many flocks infected by IBH was seen, and certain birds had non-specific clinical indications such as huddled-up lethargy, feather-ruffling, and yellow mucoid droppings that resembled sulfur due to an excess of bile acids—four days passed during the IBH mortality period; the liver is frequently enlarged, swollen, and pale, yellowish in color, with multiple focal red hemorrhagic spots. And enlarged kidneys and dilatation of renal tubules were also observed (Fig .1).

##### Histopathological characterization

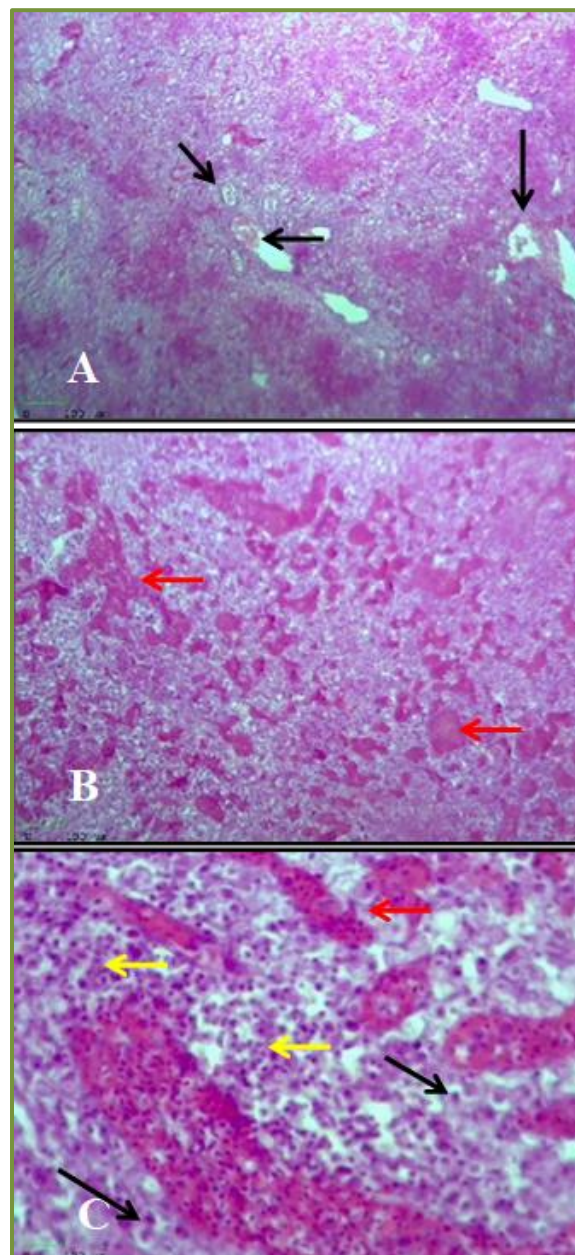
The histopathological changes caused by IBH viral infection in the current study include the presence of the necrosis of the cardiac muscle and thickening of the wall of the blood vessel was noted in (fig. 2). While the obvious and characteristic lesions of this infection especially in the main target organ (liver); presence intra nuclear Inclusion bodies, hyperemia of the arteries and hepatic sinusoids, sinusoidal dilatation, and hemorrhagic area. Severely infiltration of inflammatory cells especially the macrophage (fig .3). presences of multi areas of necrosis (fig. 3). Whereas the damaged kidneys of infected chicken showing the presence of vacuolation of the renal tubular epithelium, interstitial tissue hemorrhage with congested blood vessel and infiltration of inflammatory cells, as shown in (fig. 4).



**Figure 1:** Gross images of bird infected with IBH: A- depression (arrow), B and C- enlargement, hemorrhagic (blue arrow), necrotic and friable liver (yellow arrow), D- enlargement, congestion and pale kidney (red arrow).

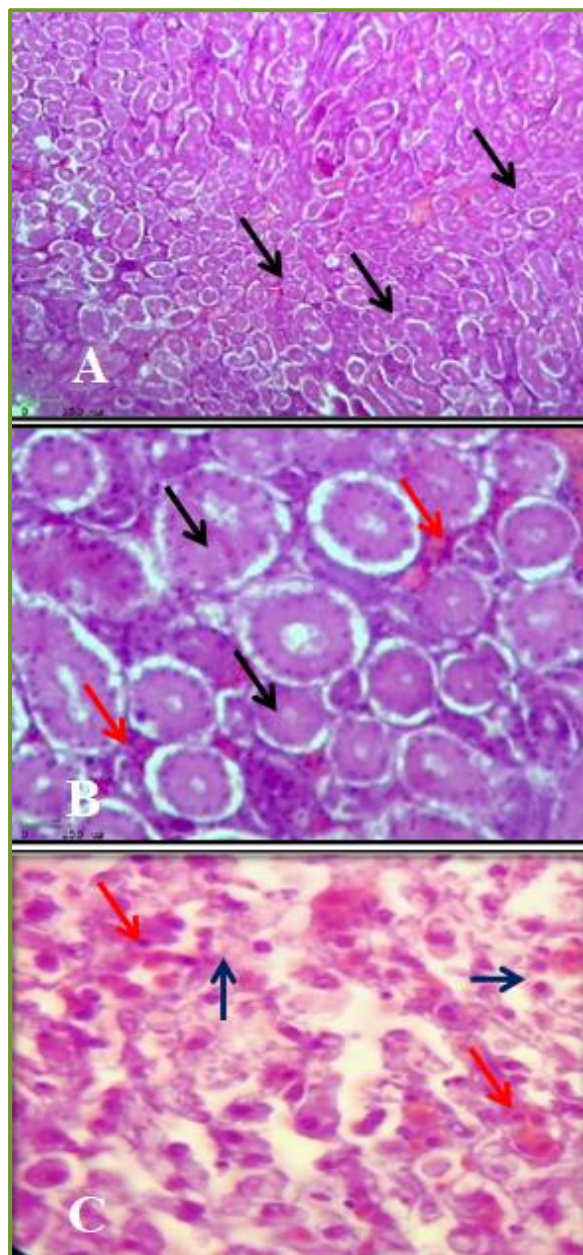


**Figure 2:** Histopathological section of infected chicken heart: A- the thickening of the wall of the blood vessel (black arrow), B, C-presence of the necrosis of the cardiac muscle (red arrows) (H&E 40X).



**Figure 3:** Histopathological section of infected chicken liver; A, B-presence of hemorrhagic area (red arrows), congestion of C.V. (black arrows). (H&E 10X), C-presence Inclusion body (black arrows), hemorrhagic area (red arrows) and sever infiltration of inflammatory cells (yellow arrows) specially the macrophage (H&E 40X).





**Figure 4:** Histopathological section of infected chicken kidney: A. presence of vacuolation of the renal tubular epithelium (black arrows), (H&E 4X). B- the presence of vacuolation of the renal tubular epithelium (black arrows), interstitial tissue hemorrhage (red arrows), (H&E 10X), C- presence of vacuolation of the renal tubular epithelium, interstitial tissue hemorrhage (red arrows), and infiltration of inflammatory cells (blue arrows), (H&E 40X).

## Discussion

Fowl avian adenoviruses have been detected in numerous avian species, including domestic poultry and wild birds, across the globe in recent times (16, 17). Numerous authors in Iraq have claimed that the disease has severe effects on the economic losses of broilers, and that further research is required to emphasize the disease's function (15). Due to an overabundance of bile acids, the affected birds displayed dullness, depression, mortality, and yellow mucoid dropping. Infected farms saw a drop in body weight gain and feed conversion ratio. These findings were connected to (18, 19) and showed a sudden and high mortality rate that persisted for four to five days. In the postmortem analysis, the majority of the liver it had areas of localized or widespread necrosis and was pale, friable, and swollen. In certain instances, the hepatic parenchyma may experience pinpoint or Ecchymotic hemorrhage. In some of the deceased broiler chickens, there were either isolated liver or hemorrhages. A drop in osmotic blood pressure and the accumulation of straw-colored fluid in the pericardial sac are the results of degenerative and necrotic changes in the infected liver, which are the cause of liver dysfunction (20).

Broilers are more vulnerable to bigger mortality than other avian species because of the severe metabolic imbalance and the substantial damage of the pancreas and liver (21). Pathognomonic for HHS is hydropericardium, which is characterized by accumulation of clear, straw-colored fluid in the pericardial sac (22). Pulmonary edema, a larger, discolored liver, and enlarged kidneys with dilated tubules are typical lesions. When the osmotic pressure of colloidal plasma is decreased, fluid leaks into the pericardial sac. While serum levels of uric acid, potassium, calcium, and triglycerides were significantly greater, blood glucose and plasma protein levels were decreased. This could be because of fluid buildup in the pericardial sac and the abdomen (23).

Histopathologically significant lesions were seen in the liver, kidney, and heart of the infected broiler chicks. necrosis of the cardiac muscle and thickening of the wall of the blood vessel, while the obvious and characteristic lesions of this infection especially in the main target organ (liver); presence intra nuclear Inclusion bodies, (hemorrhagic area and sever infiltration of inflammatory cells specially the macrophage, these results were related to that (24)

who found Large irregular basophilic intranuclear inclusion bodies were seen in hepatocytes in broiler chicken farm in India. where the damaged kidneys of infected chicken of the current study showing the presence of vacuolation of the renal tubular epithelium; interstitial tissue hemorrhage and infiltration of inflammatory cells such finding in the line with that of (25); who reported the kidneys of the infected chickens in China were edematous with renal tubule degeneration and necrosis .

### Conclusions

We recorded a abrupt high mortality rate in infected broiler flocks in Basra Governorate, southern Iraq, where some clinical signs were observed as well as post-mortem lesions characteristic of this disease, which were confirmed by histopathological changes in the infected organs with this virus, knowing that most of the infections were diagnosed in Chicks that hatched from eggs imported from outside Iraq. Finally, Loss of biosecurity and vaccination programs against IBH infection in poultry populations particularly in Basra governorate make them more prone to infection.

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### Conflict of interest

The authors declare that there is no conflict of interest.

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