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Diagnosis of Fowl Cholera in Broiler Chicken Collected from Banda Aceh and Aceh Besar

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ABSTRACT

Fowl cholera is a highly contagious bacterial disease affecting poultry worldwide including broiler chickens. This study aimed at diagnosing fowl cholera in broiler chicken collected from Banda Aceh and Aceh Besar based on anatomical pathology and histopathological lesions. Approximately 1,400 broilers chicken's cadaver were collected from poultry market around Banda Aceh and Aceh Besar within 2022-2024, then necropsied at Pathology Laboratory, Veterinary Medicine Faculty, USK. All chicken were subjected to an anatomical pathology examination, followed by histopathological observation using hematoxylin-eosin staining method. The macroscopic results revealed multifocal necrosis in the liver, hemorrhages in several internal organs, oedema in lungs, ascites in thorax cavity, and arthritis. Histopathological examination highlighted characteristic lesions including vasculitis, submucosal edema in trachea, multifocal necrosis in liver, and desquamation of intestinal villi. The prevalence rate of fowl cholera disease during 2022, 2023, and 2024 were 8,2%, 11,2%, and 14.0%, respectively. In conclusion, broilers with fowl cholera shows inflammation lesions in several organs, focal necrosis in liver, ascites in thorax and abdominal cavity in acute form, and arthritis in chronic form.

Keywords: *Fowl cholera, Pasteurella multocida, Broiler, Histopathology*

INTRODUCTION

Fowl cholera is caused by *Pasteurella multocida*, a Gram-negative, non-motile coccobacillus bacterium. The primary strain responsible for fowl cholera is *P. multocida* subspecies *multocida*, although occasional outbreaks are caused by the *septica* or *gallicida* subspecies. This disease is highly contagious and can affect a wide variety of avian species, both wild and domestic. While commonly associated with chickens and turkeys, it is also a significant concern for ducks and geese (Reuben et al. 2021). Fowl cholera is also referred to as avian cholera, avian pasteurellosis, or avian haemorrhagic septicaemia.

Several studies have reported the prevalence of fowl cholera globally. For instance, Kuczkwoski et al. (2006) documented 123 cases of fowl cholera over a three-year period on farms raising geese and turkeys in South-West Poland. In Indonesia, Purnomo (1980) reported that deaths from pasteurellosis in ducks reached 62% of a population of 1,400 ducks. In West Java and Jakarta, the mortality rate due to cholera in ducks ranged from 30% to 50% (Tati and Supar, 2008). Additionally, a study by Krisdiyantoro et al. (2023) found that a sample from a broiler farm in Rhee District, Sumbawa Regency, tested positive for poultry cholera. Moreover, Tangkondan et al. (2023) identified three isolates of *Pasteurella multocida* from 23 samples tested in Kupang Regency.

Pasteurella multocida holds substantial economic significance worldwide. Avian pasteurellosis, caused by this pathogen, can result in high morbidity and mortality rates, manifesting in peracute, acute, chronic, or localized forms (Singh et al. 2014). The peracute form is the most virulent and contagious, often causes sudden death in seemingly healthy birds without prior symptoms. In the acute form, affected birds may exhibit symptoms such as anorexia, high fever, mucous discharge from

the beak, green foul-smelling diarrhoea, and cyanosis of the comb a few hours before death (Raji et al. 2010).

In the chronic form of avian pasteurellosis, birds displayed various symptoms, with respiratory problems being the most common. Torticollis (wry-neck) was occasionally noted, along with swollen eyes, wattles, or ear lobes. Some birds exhibited lameness caused by swollen joints. Birds that succumbed suddenly often showed minimal lesions during necropsy, though petechiae were commonly observed on the heart and serous membranes, especially over the gizzard fat. In milder cases, small necrotic foci were present in the liver. Lesions associated with chronic disease were characterized by the accumulation of yellowish, caseous exudate in areas such as the air sacs, ears, ear lobes, wattles, or joints (Dorsey and Harshfield, 1959).

Stressors such as environmental changes, nutritional shifts, and injuries can influence the progression of the disease. Birds that are chronically infected or in the convalescent phase serve as major sources of infection, as carrier birds can shed *P. multocida* for life. Chronic infections can lead to localized forms of the disease. *P. multocida* can survive in the environment for up to two weeks and in dried blood for up to eight days. However, when separated from organic material, the bacterium is easily destroyed by common disinfectants, sunlight, drying, and temperatures above 56°C (Blakey et al. 2019).

Certain strains of *P. multocida*, an enigmatic pathogen, are capable of colonizing the nasopharynx, respiratory tract, and gastrointestinal tract of animals, including poultry, leading to severe diseases (Reuben et al. 2021). Evidence gathered on fowl cholera highlights the respiratory tract as the primary entry point, along with the conjunctiva and cutaneous wounds. Additionally, *P. multocida* is considered part of the normal microflora in the upper respiratory tract of many healthy animals, including poultry, where it can persist in the oropharynx for extended periods without causing illness. However, virulent strains of *P. multocida* can invade the mucosa of the upper respiratory tract, subsequently infecting the air sacs and lungs of birds. By an unclear mechanism, potentially involving migration within macrophages of the upper respiratory tract, the bacteria may enter the bloodstream from the mucosa and proliferate in various tissues, particularly in the liver and spleen (Reuben et al. 2021).

MATERIALS AND METHODS

The data for this study was obtained from the Pathology Laboratory of the Veterinary Medicine Faculty, Universitas Syiah Kuala, Banda Aceh between the year 2022 and 2024. The result is presented yearly reflecting number of samples sent to the laboratory monthly from poultry market around Banda Aceh and Aceh Besar.

Necropsy Procedure

The bird was positioned dorsal recumbency with wings and legs extended. A midline incision was made from the cloaca to the neck, and the sternum bone was removed to expose the thoracic and abdominal cavities. External and internal examinations were conducted systematically to assess the condition of the skin, musculature, and internal organs, including the respiratory, gastrointestinal, hepatic, and cardiovascular systems. Observations included the presence of lesions, discoloration, or abnormal fluid accumulation. Samples collected from affected tissues and organs were preserved in 10% formalin prior to histopathology procedure.

Histopathology Technique

Tissues were fixed in 10% neutral buffered formalin, dehydrated in graded alcohol and cleared in xylol, then embedded in paraffin wax to form blocks for sectioning with the thickness of 5 µm. The tissues sections were then placed on glass slides. The staining method were apply using hematoxylin and eosin (Kiernan, 2015). The observation was performed using a microscope (Olympus CX23, Japan) then documented using photomicrograph (Olympus DP20, Japan). The data was analyzed descriptively.

RESULTS AND DISCUSSION

Macroscopic examination of broilers infected with fowl cholera revealed several anatomical pathology lesions for instance local infections in the joints (arthritis), liver necrosis, and haemorrhages in the intestines, heart, and trachea, which contained exudates. The lungs and kidneys showed swelling and haemorrhagic discoloration. In addition, ascites also found in thorax cavity. The macroscopic lesions are demonstrated in Figure 1.

Similar findings were reported by Krisdiyantoro et al. (2023) in broiler chickens, which exhibited clinical signs of joint infections and bleeding in the heart and intestines. The acute form of fowl cholera lesions was also associated with the presence of acute septicaemia which was characterized by bleeding as in the peracute form. Petechial haemorrhages and ecchymoses were usually observed in various tissues such as in subepicardial and subserosa areas, lungs, abdominal fat tissue and intestinal mucosa. Hydropericardium and ascites were often characterized as the acute form of fowl cholera (Tabbu, 2004). The ascites was primarily a result of inflammatory vascular damage, liver dysfunction, and systemic changes that disrupt fluid homeostasis. Its presence often indicates severe systemic involvement and poor prognosis in affected birds (Wilkie et al. 2012).

Pasteurella multocida has three primarily tissues and cell target, the first is endothelial cells. *P multocida* endotoxins and virulence factors can damage endothelial cells lining the blood vessels, leading to vasculitis, increased vascular permeability, and haemorrhage. This is a key feature of its pathogenicity, resulting in systemic inflammation and organ damage. Secondly, the upper respiratory tract, including the trachea and lungs, is often the primary site of infection. The bacteria cause inflammation, oedema, and necrosis in these tissues. And third, the liver and spleen are common sites of colonization and damage to the systemic spread of the bacteria. Multifocal necrosis in the liver and inflammation in the spleen are frequently observed in infections.

The bacterium's endotoxins and other virulence factors (such as capsule, lipopolysaccharides, and adhesins) allow it to adhere to and damage endothelial cells which lead to vasculitis, oedema, and haemorrhages (Boyce and Adler, 2000).

Histopathological observation results showed that hyperaemia and vasculitis were observed, along with submucosal oedema and rupture of villi (desquamation of epithelium) in trachea. In the liver, multifocal necrosis was observed (Figure 2). The lungs exhibited characteristic changes, including intravascular oedema, similar to findings reported by Ghaly et al. (2017), where a case study of *P. multocida* in poultry showed pathological changes such as perivascular oedema. Moreover, the intestines and proventriculus showed desquamation and thickening of the villi. Multifocal necrosis in the liver highlights systemic involvement of the disease. The presence of necrotic lesions suggests a

severe disruption of liver function, likely caused by bacterial dissemination and localized inflammatory responses (Goncalves and Tonin, 2018). Characteristic intravascular oedema in the lungs emphasizes the impact of fowl cholera on the respiratory system. This finding, indicative of fluid accumulation, compromises gas exchange and exacerbates respiratory symptoms (Wille et al. 2016).

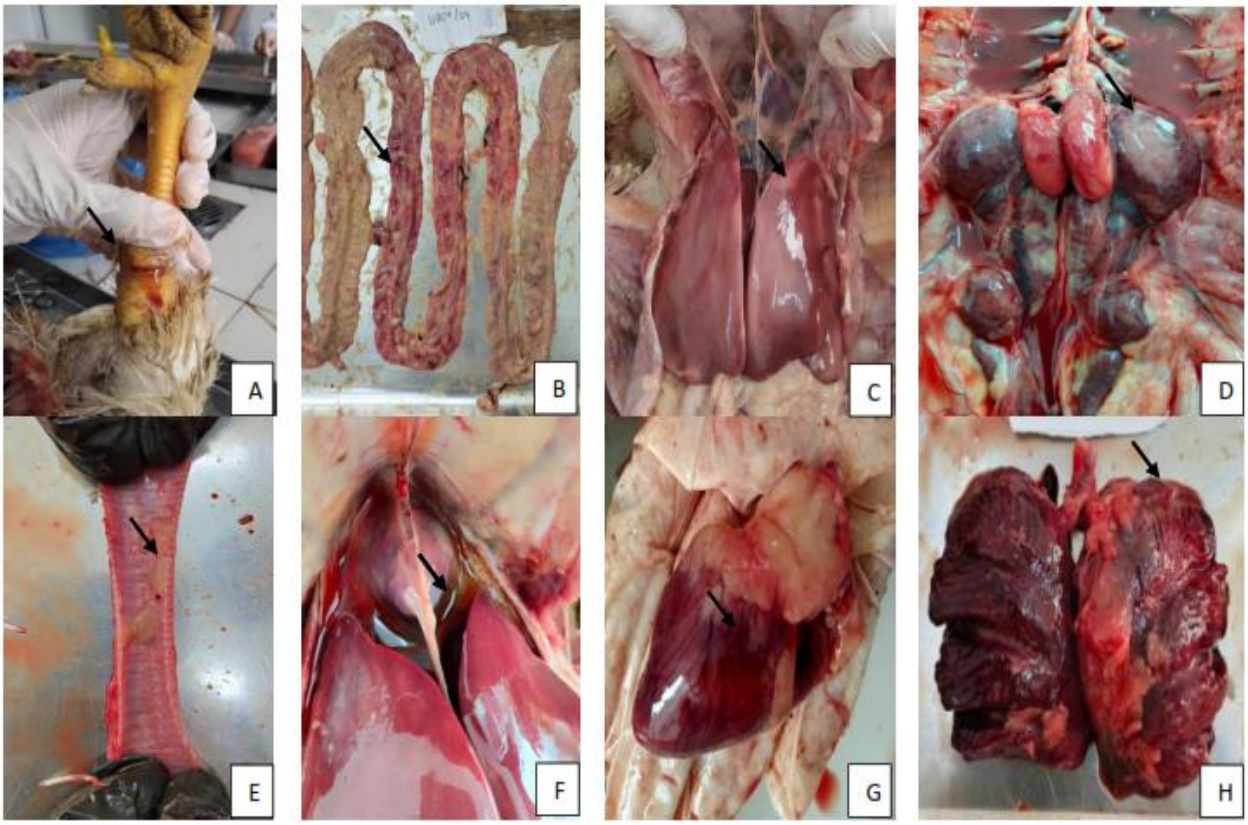

Figure 1. Macroscopic lesions of broiler chicken with fowl cholera, local arthritis (A), ptechi of intestine (B), focal necrosis on liver (C), swollen kidney (D), hemorrhage and exudate in lumen surface of trachea (E), ascites in thorax cavity (F), hemorrhage on heart (G), swollen and hemorrhage of lungs (H).

The lesions that arise from peracute form are generally associated with damage to the blood vessel endothelium, which causes haemorrhage in various visceral organs. The liver will experience congestion and is sometimes accompanied by multifocal coagulation necrosis. In the acute form, the most important microscopic changes include damage to the vascular endothelium which causes bleeding in various organs (Blackey et al.,2019). Acute forms of avian cholera can cause multifocal coagulation necrosis, accompanied by heterophil infiltration. Heterophil infiltration can also be found in the lungs. In the chronic form, the most important microscopic changes include suppurative inflammation characterized by areas of necrosis, heterophil infiltration, fibrin formation, multinucleated giant cells, and fibroblast proliferation. Local meningitis is often found which is characterized by infiltration of heterophils and lymphocytes in the meninges area (Wille et al. 2016).

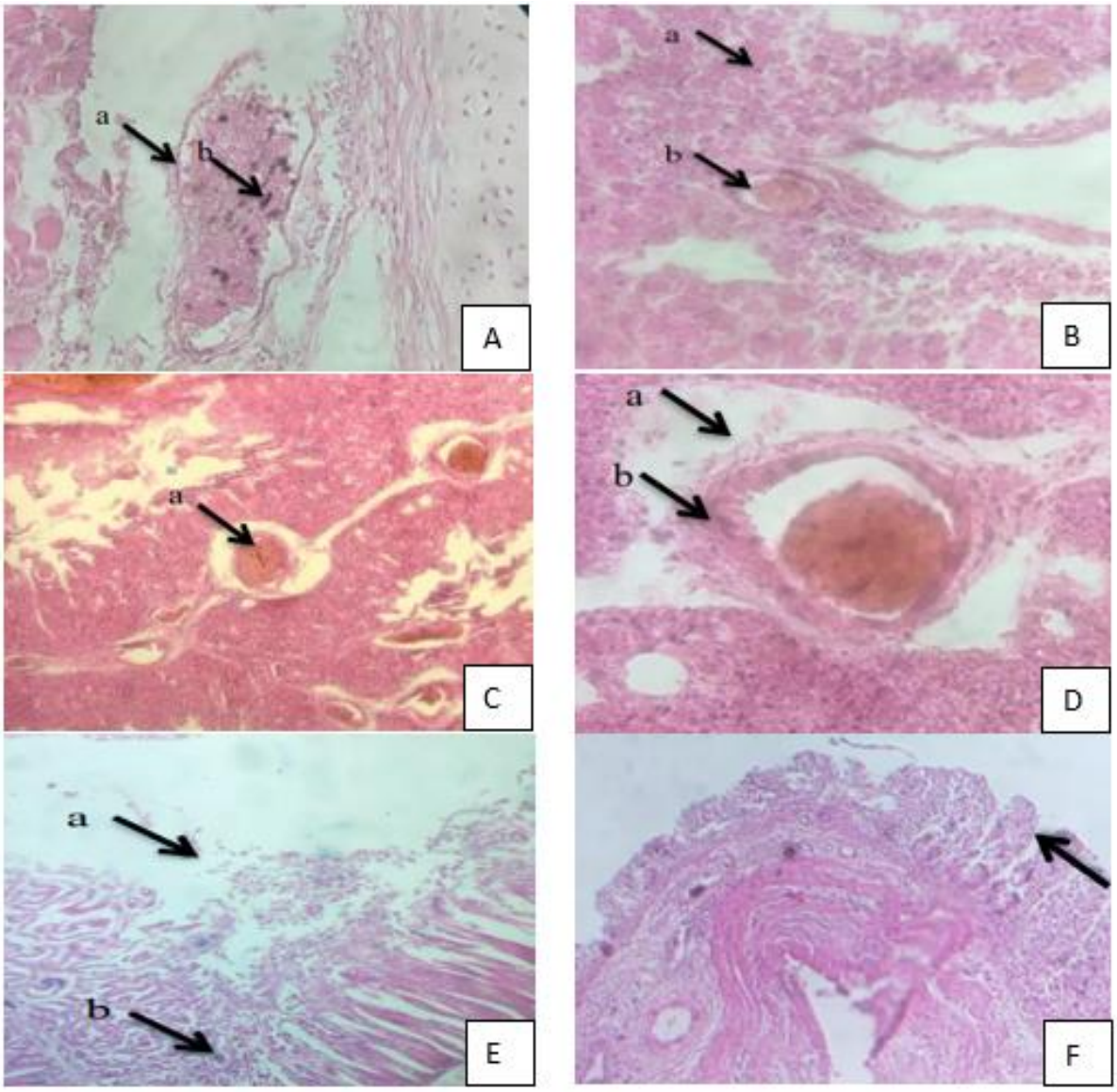

Figure 2. Histopathological figure of broiler chicken infected with fowl cholera, dilation and rupture of blood vessel in trachea (A), multifocal necrosis in liver (a), hyperemia (b) (B), hemorrhage in lung (C), edema intravascular in lung (a), arterial wall thickening (vasculitis) (b) (D), desquamation of proventriculus epithelial gland (a) and pyknosis (b) (F), thickening of intestine villi (F) (H&E, 100X, 400X)

The necropsy results of avian pasteurellosis conducted at the Pathology Laboratory, Veterinary Medicine Faculty over the study years revealed prevalence rates of 8.2%, 11.2%, and 14.0% for 2022, 2023, and 2024, respectively (Table 1). The overall prevalence rate for the study period from 2022 to 2024 was 11%. These findings indicated a rising trend in the prevalence of avian pasteurellosis over the observed years.

Table 1. The yearly distribution of fowl cholera cases in chicken cadavers at Pathology Laboratory

No	Year	Number of broilers cadavers necropsied at Pathology Laboratory	Number of broilers positive cholera	Prevalence (%)
1	2022	499	41	8.2
2	2023	488	55	11.2
3	2024	413	58	14.0

This prevalence is higher from the report of Masdooq et al. (2008) who found overall prevalence of 6.4% in Jos-Plateu State, and Raji et al. (2010) in Zaria-Kaduna State Nigeria with the prevalence rate of 4.7%. The prevalence rate of avian pasteurellosis might be affected by several factors including seasonal variation during the year. For example, heavy rainfall made it possible for the organism to survive for longer time in the environment.

Warm temperatures and high humidity create suitable conditions for the survival and spread of *Pasteurella multocida*. Overcrowded poultry farms accelerate disease transmission, and certain poultry breeds may be more susceptible to infection (Wille et al. 2016). Additionally, inadequate hygiene and poor biosecurity practices further heighten the risk of outbreaks. Addressing these issues requires comprehensive strategies, including enhancing biosecurity measures, implementing effective vaccination programs, and educating farmers on disease prevention and management (Boyce et al. 2010).

CONCLUSION

To conclude, broilers with fowl cholera showed inflammation lesions in several organs, focal necrosis in liver, ascites in thorax and abdominal cavity in acute form, and arthritis in chronic form. The prevalence rate of the disease is higher than other places that might be caused by seasonal variation during the year.

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