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Current Symptoms and Pathological Changes of Bursa of Fabricius from Commercial Farming Broiler Ldto Infectious Bursal Disease

Gejala dan Perubahan Patologis Terkini Organ Bursa Fabricius dari Peternakan Komersial Broiler pada Infectious Bursal Disease Bentuk Akut dan Kronis

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Abstrak

Infectious Bursal Disease (IBD) atau Gumboro disebabkan oleh virus IBD dari *family* Birnaviridae. Penyakit ini bersifat akut dan sangat menular pada unggas muda. Infeksi virus ini menyebabkan kerusakan parah pada organ limfoid yaitu bursa Fabricius yang menyebabkan imunosupresi. Angka kesakitan penyakit ini dapat mencapai 100%, sedangkan angka kematian berkisar antara 20 hingga 100% sehingga menimbulkan kerugian ekonomi yang besar. Penyakit IBD masih menjadi ancaman yang signifikan meskipun program vaksinasi telah diterapkan. Penelitian ini bertujuan untuk mengetahui perubahan patologi khas bursa Fabricius pada ayam broiler komersil yang menunjukkan gejala IBD. Sampel diperoleh dari peternakan ayam broiler komersial di Sragen, Wonogiri, Batang, dan Sleman. Pemeriksaan lesi patologi makroskopis menunjukkan pembesaran bursa Fabricius disertai masa gelatin pada permukaan serosal, edema dengan akumulasi cairan di lumen, hemoraghi pada permukaan serosal, atrofi, dan eksudat kaseosa di lumen. Perubahan histopatologi IBD akut meliputi hemoraghi, kongesti, nekrosis limfosit, akumulasi fibrin, edema dan infiltrasi heterofil pada jaringan interfolikuler. Perubahan mikroskopis pada IBD kronis (5-7 hari pasca infeksi IBDV) antara lain atrofi folikuler, nekrosis limfosit, vakuolisasi folikel, dan proliferasi fibroblas dan jaringan ikat pada ruang interfolikuler bursa Fabricius. Kesimpulannya, gambaran perubahan patologis bursa Fabricius yang dominan pada IBD akut adalah lesi patologi makroskopis (pembengkakan dan edema, plica bursa Fabricius menebal dan membesar, hemoraghi), lesi mikroskopis (kongesti, hemoraghi, infiltrasi heterofil) atau infeksi IBD kronis pada ayam broiler. lesi patologi makroskopis (atrofi bursa Fabricius, atrofi disertai eksudat perkejuan pada lumen bursa Fabricius), lesi mikroskopis (nekrosis limfosit, vakuolisasi folikel bursa Fabricius, proliferasi fibroblas dan jaringan ikat interfolikuler).

Kata kunci: ayam broiler komersil; bursa Fabricius; Gumboro; perubahan patologis

Abstract

Infectious Bursal Disease (IBD) or Gumboro is caused by the IBD virus of the Birnaviridae family. The disease is acute and highly contagious in young birds. The virus infection causes severe damage to the lymphoid organs i.e. bursa of Fabricius leading to immunosuppression. The disease morbidity may reach 100%, while

the mortality varies from 20 to 100%, causing high economic losses. Infectious Bursal Disease has remained significant threat although vaccination has been applied. This study aimed to determine the current typical pathological changes in the bursa of Fabricius of commercialbroilers showing IBD symptoms. The samples were obtained from commercial broiler farms in Sragen, Wonogiri, Batang, and Sleman. Gross lesion examination showed enlargement of the bursa of Fabricius with gelatinous material on the serosal surface, oedema with fluid accumulation in the lumen, hemorrhages of the serosal surface, atrophy, and caseous exudate in the lumen. Histopathology changes of acute IBD include hemorrhages, congestion, lymphocyte necrosis, accumulation of fibrin, oedematous and heterophils infiltration in the interfollicular tissues. Microscopic changes in chronic IBD (5-7 days post infection IBDV) including follicular atrophy, lymphocyte necrosis, vacuolization of the follicle, and proliferation of fibroblast and connective tissue in the interfollicular space. Pathological lesion both of macroscopic and microscopic of acute and chronic suspected IBD from commercial broiler farming were similar to previous studies. Sample with clinical signs of birds in the study were depression, trembling, weak, white and watery diarrhea, ruffled feathers and feathers surrounded vent covered with urates have clinical symptoms of classical types of IBD.

Keywords: broiler commercial farming; bursa of Fabricius; Gumboro; pathological changes

Introduction

Infectious Bursal Disease (IBD) or Gumboro is an acute and highly contagious disease. The virus is a single-shelled, non-enveloped virion with icosahedral symmetry and a diameter varying from 55–65 nm(Eterradosi & Saif, 2013). The IBD virus is Avibirnavirus with two segmented double-stranded RNA segments (Muller *et al*., 1979). Segment A encodes 2 structural proteins (VP2 and VP3), a viral protease (VP4), and a nonstructural protein (VP5) (Raja *et al*., 2016). VP2 is the major structural protein, which is involved in antigenicity, cell tropism, virulence, and apoptosis (Coulibaly *et al*., 2005). The smaller segment B encodes for the RNA-dependent RNA polymerase VP1 protein (Jackwood *et al*., 1985).

The primary target organ for IBDV is the bursa of Fabricius (Eterradosi & Saif, 2013). The characteristic lesions of IBD are necrosis of the bursa of Fabricius with subsequent inflammation and depleted the causing athrophy to the bursa of Fabricius (Schat & Skinner, 2008). According to Kegne & Chanie (2014), IBDV causes lymphoid depletion of the bursa of Fabricius and if this occurs in the first 2 weeks of life, significant depression of the humoral antibody response may result.

Infectious Bursal Disease is an immunosuppressive disease in poultry causing huge economic losses for the industry with mortality reaching 20% or more in young chickens and prolonged immunosuppressive effects (Gao *et al*., 2014; Michel & Jackwood, 2017; Orakpoghenor *et al*., 2020). Mortality is determined by IBDV virulence, infection dose, age, breed and passive immunity (Ingrao *et al*., 2013). According to (Zeleke A *et al*., 2005), mortality due to IBD infection in the field in broiler chicken reached 59.09% and commercial layer chicken 25.08%. In very malignant cases IBDV infection (vvIBDV) can even reach 100% (Parede *et al*., 2003). The farm owners have reported frequent occurrences of the diseases even in vaccinated flocks (Nwagbo *et al*., 2023). The IBD is economically important for the poultry industry since it can cause immune depression (Orakpoghenor *et al*., 2020; Pikuła *et al*., 2020). Strict biosecurity together with the use of conventional inactivated and live vaccines had been successfully controlling IBD until the emergence of antigenic variants in early 1980s (Rathnayake & Kalupahana, 2019). In addition, vaccination against the vvIBD strains encounters many challenges as they can breakthrough antibody protection, and therefore highly efficient vaccination is required (Khan *et al*., 2017).

Infectious Bursal Disease cases are still considered as threat due to high economic loss (Eterradosi & Saif, 2013; Khan *et al*., 2009; Orakpoghenor *et al*., 2020). This study aims to characterize pathological changes in bursa of Fabricius of commercial farming broilers showing IBD symptoms from the latest outbreak

between acute and chronic form IBD. Clinical signs and pathological characteristic changes of bursa of Fabricius as main target organ could assist early diagnosis in the field.

Materials and Methods

The samples were obtained from commercial broiler farming in Sragen, Wonogiri, Batang, and Sleman regencies. The bursa of Fabricius specimens were collected from birds showing symptoms of depression, trembling, white and watery diarrhea, ruffled feathers and feathers surround vent covered with urates. The observed gross changes of the bursa of Fabricius were enlarged and edematous, gelatinous, hemorrhage, atrophy, and caseous exudate in the lumen. Collected bursa of Fabricius tissue were fixed in 10% neutral buffered formalin and routinely processed for histology preparations stained with hematoxylin and eosin (Slaoui & Fiette, 2011) at the Disease Investigation Centre, Wates, Yogyakarta. The histopathology examination was carried out at the Department of Pathology at the Faculty of Veterinary Medicine UGM. The bursa of Fabricius was categorized into chronic and acute based on farm recording. A case is chategorise as chronic when the infection has passed mortality peak (5 – 7 days).

Results and Discussion

The observed clinical signs of birds in the study were depression, trembling, weak, white and watery diarrhea, ruffled feathers and feathers surrounded vent covered with urates. The majority of the study on IBDV reported clinical symptoms as general weakness, whitish diarrhea, dirty cloaca and tremors. (Liang *et al*., 2015; Oluwayelu *et al*., 2002; Omer & Khalafalla, 2022; Zannah *et al*., 2020). The disease is characterized with very short course, sudden onset of depression, followed by prostration and reluctance to move, with ruffled feathers and frequently watery or white diarrhea (Ignjatovic, 2014). This study has recorded similar symptoms of IBD (Figure. 1).

However, initially such symptoms as sudden onset, morbidity, and ruffled feathers were suggested as an acute coccidiosis outbreak (Eterradosi & Saif, 2013). Also ruffled feathers, anorexia, dirty vent, and diarrhea was similar to Salmonellosis (Nazir *et al*., 2012). Clinical symptoms induced by vvIBDV are similar to those induced by classical virulent strains of IBDV, except that the disease is more pronounced, acute and accompanied with high mortality. Onset of the acute phase is faster and the acute phase is more generalized (Ignjatovic, 2014).

Figure 1. Symptoms showed by suspect IBD birds in the field. (A) depression; (B) trembling; (C, D) weakness; (E) ruffled feathers; (F) vent feathers covered with white and watery diarrhea.

The bursa of Fabricius is the main organ affected by IBDV. Lesions in the bursa of Fabricius generally induce immunosuppression which results in secondary infections and suppress production performance(Sassia et al., 2017). The main lesions of the bursa of Fabricius were reported as inflame, edematous, hyperemic, hemorrhagic, and atrophy (Khan et al., 2009). Figure 2 showed pathological changes of acute IBD. Figure 2. Acute form of IBD in bursa Fabricius; (A) Swelling and edema; (B) Plica bursa is thickened and enlarged; (C, D) hemorrhage; (E) Microscopic lesion of bursa Fabricius; (e) edema, (h) hemorrhage, (k) congestion, (i) heterophil infiltration; Lesions are indicated by black arrow.

An acute clinical outbreak of classical IBDV are characterised by sudden onset, high morbidity, spiking mortality curves, with birds dying within the first 4 days following clinical symptom and a rapid recovery time of about 5–7 days post clinical signs (Eterradosi & Saif, 2013; Ignjatovic, 2014; van den Berg *et al*., 2000). Macroscopic lesion in acute phase showed bursa of Fabricius swelling and edema (Figure. 2A), thickened and enlarged in plica of bursa of Fabricius (Figure. 2B), hemorrhage in the bursa of Fabricius (Figure. 2C, 2D). This research was similar with other research that enlarged and hemorrhage in the serosa of bursa Fabricius, slight hemorrhage on the plica bursa of Fabricius were showed (Akter *et al*., 2018b; Bharathi *et al*., 2021; Chowdhury *et al*., 1996; Kulsum *et al*., 2018; Orakpoghenor *et al*., 2021). The acute inflammatory swelling or enlargement of the bursa of Fabricius would occur in few days into the outbreak (Igwe *et al*., 2017). Bursa of Fabricius would be swollen (enlarged) with increased weight, because of congestion, exudation of fluid into the interfollicular tissues, accumulation of fluid in the necrotized lymphoid follicles forming cysts and presence of mucous, caseous or gelatinous exudate on mucosal folds lining the inside of the bursal lumen (Igwe *et al*., 2017; Wang *et al*., 2011).

Microscopic lesion in acute phase showed hemorrhage in the follicle bursa of Fabricius (Figure. 2E-h), congestion (Figure. 2E-k), heterophil infiltration (Figure. 2E-i). Ignjatovic et al., (2004) reported the presence widespread acute lymphoid necrosis, follicular haemorrhage and stromal oedema in acute IBD (Figure. 2E). Reactive cells infiltration by heterophils and macrophages in the interfollicular space were detected (Kulsum et al., 2018). Zeryehun et al., (2012) reported that histology bursa of Fabricius 4 days pos infection with VVIBD revealed congested blood vessels and hemorragic bursa of Fabricius with some activity of RBCs escaping through damaged blood vessel wall. These strongly suggested damage to the blood vessel and a possible disseminated intravascular coagulation (DIC) (Zeryehun et al., 2012).Figure 3 showed chronic type of IBD. Two forms of

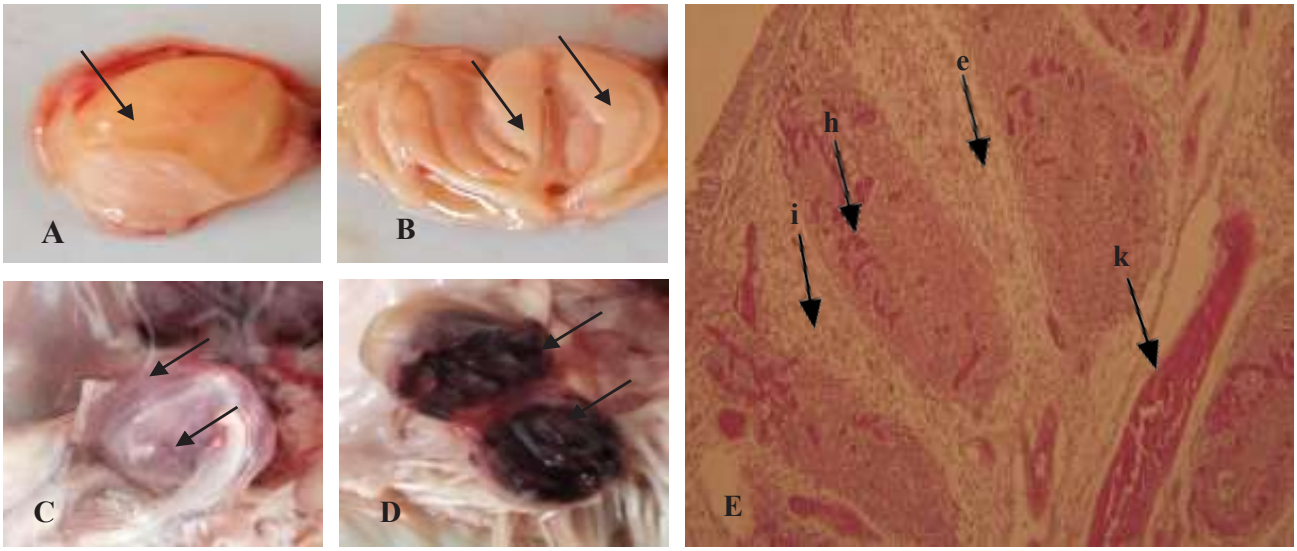

Figure 2. Acute form of IBD in bursa Fabricius; (A) Gross lesion swelling and edema; (B) Plica bursa thickened and enlarged; (C, D) hemorrhage; (E) Microscopic lesion of bursa Fabricus; edema (e), hemorrhage (h), congestion (k), heterophil infiltration (i); The lesion is indicated by black arrow.

chronic lesion of bursa of Fabricius that were atrophy and atrophy IBD with exudate caseous. In this study, atrophic lesion was mostly observed in all samples compared to enlarged, gelatinous materials covering the serosal surface of bursa of Fabricius or ecchymosis hemorrhages on the mucosal surface.

Macroscopic lesion in chronic phase showed atrophy of bursa of Fabricius (Figure. 3A) and atrophy with exudate caseous in the lumen bursa of Fabricius (Figure. 3B). The results in this study was as previously reported by Bharathi *et al* (2021). Caseous exudate lesions in the lumen and atrophy of the bursa of Fabricius were found in India (Bharathi *et al*., 2021). The main lesions were reported as inflame, edematous, hyperemic and hemorrhagic, and atrophy (Khan, 2009). In this study, atrophic lesion was mostly observed (50%) in all samples which collected post infection peak. Juranova *et al*., (2001) has reported atrophy in bursa of Fabricius after 3 – 5 days post infection in chronic or sub-acute IBD (Figure. 3A & 3B) and another researcher has reported significant size decrease in size (Latief *et al*., 2017). Atrophy bursa of Fabricius occurred when the inflammation had receded and atrophy was due to lymphocytic depletion from necrosis and apoptosis. Size reduction of bursa of Fabricius was considered a consequence of resolution of acute inflammation in the transition to sub-acute. Chronic inflammation involved the clearance of inflammatory exudates after diminution of the actions of virus-induced pro-inflammatory cytokines and onset of fibroplasia in the interstices (Badau *et al*., 2023). According to Orakpoghenor *et al*., (2021), the subsequent decreases in bursa of Fabricius weight were possibly due to atrophy following recovery from extensive damage by the IBDV infection.

The histopathology of the bursa of Fabricius samples showed lymphocytic destruction and vacuolization, also severe lymphocyte depletion (Figure. 3C). Complete depletion of lymphocytes resulting in atrophy of follicles, fibroplasia around the follicles of bursa of Fabricius (Chowdhury *et al*., 1996). These lesions are similar to those described by Akter *et al*. (2018). The chronic type showed mild to severe lymphoid depletion in follicles, follicular atrophy, and vacuolization of follicles. The bursal follicles are then replaced by cysts lined by columnar epithelium with fibroblastic interfollicular stroma (Bharathi *et al*., 2021; Okoye & Uzoukwu, 1990). The cystic cavities contain globules of mucin, which indicate regression of the inflammatory reaction (Eterradosi & Saif, 2013). There is appearance of scattered foci of lymphocytes in the bursal

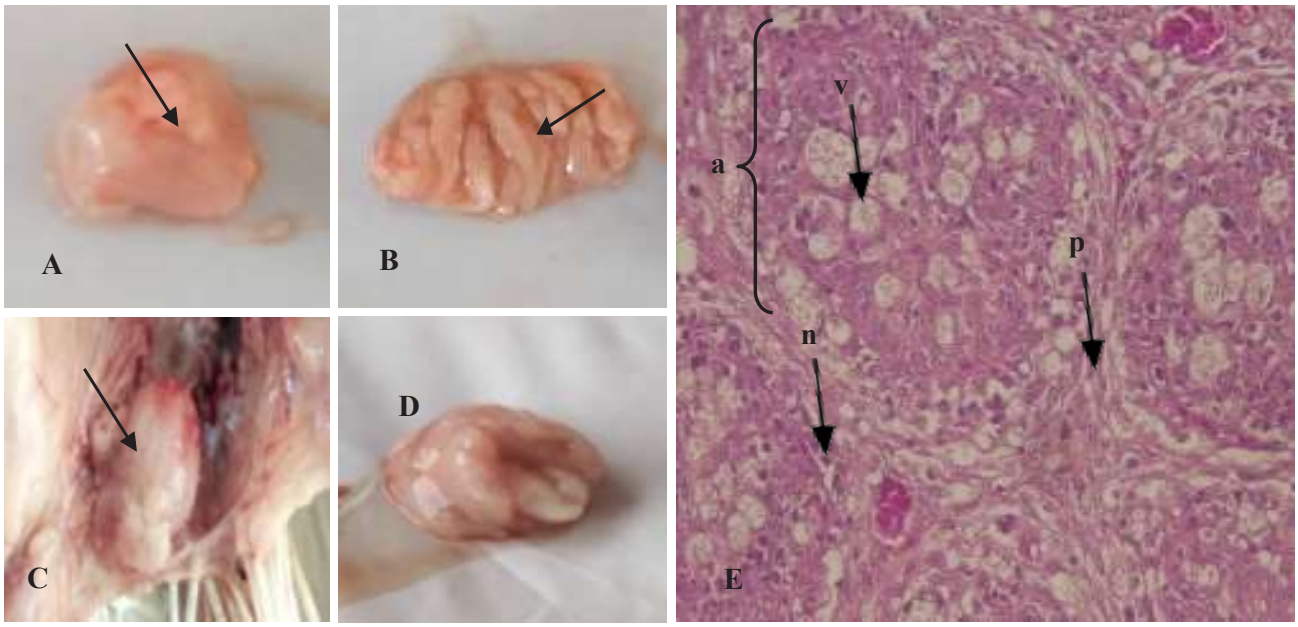

Figure 3. Chronic form of IBD in bursa Fabricius; (A) Atrophy; (B) Gross lesion atrophy bursa Fabricus; (C) Atrophy bursa of Fabricius with ecxudate caseous in the lumen; (D) Gross lesion atrophy bursa of Fabricius with ecxudate caseous in the lumen; (E) Microscopic lesion atrophy bursa Fabricius; (a) atrophy follicle, (n) lymphocyte necrosis, (v) vacuolization of bursa follicles, (p) proliferation of fibroblasts and interfollicular connective tissue. The lesion is indicated by black arrow.

follicles during the recovery period (Eterradosi & Saif, 2013). Atrophy bursa of Fabricius found in majority of the cases which attributed to the samples collected at the post-peak period of outbreak when majority of the bursa of Fabricius had undergone atrophy (Hoque *et al*., 2001; Islam *et al*., 2008; Rudd *et al*., 2002). Ignjatovic (2004) reported the presence widespread acute lymphoid necrosis, follicular hemorrhage and stromal oedema in acute IBD (Figure. 2A).

Conclusion

The bursa of Fabricius pathological lesion of suspected acute and chronic IBD from commercial broiler farming were similar to previous studies. This finding applies to both macroscopic and microscopic. Also, the clinical symptoms were consistent to classical type.

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