



MUCORMYCOSIS IN A 35-DAY-OLD ISA BROWN PULLET: A CASE REPORT

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Abstract

Mucormycosis is an opportunistic mycotic infection caused by fungi of the order Mucorales that primarily affects the respiratory system and can spread systemically in immunocompromised hosts, posing a significant threat to poultry production systems. This study aimed to investigate the cause of mortality in a 35-day-old ISA Brown layer pullet through a comprehensive clinicopathological and microbiological approach. Data were obtained through epidemiological investigation, necropsy, gross pathological and histopathological examinations, fungal and bacterial culture and identification, and parasitological examination, followed by descriptive qualitative analysis. Clinically, the affected chicken exhibited dyspnea, difficulty breathing, inability to stand, dull feathers, lethargy, reduced feed intake, and white diarrhea, with morbidity, mortality, and case fatality rates of 1.13%, 0.5%, and 44.44%, respectively. Poor housing sanitation, inadequate biosecurity, and open feed storage near a forest area were identified as important predisposing factors. Gross lesions consisted of white nodules in the lungs, air sacs, trachea, liver, and kidneys, while histopathology revealed non-septate fungal hyphae associated with pneumonia haemorrhagica et necroticans, giant cell formation, and inflammatory cell infiltration, indicating active fungal invasion with immune response. Lymphoid depletion of the bursa of Fabricius suggested concurrent Infectious Bursal Disease-associated immunosuppression. Fungal culture confirmed *Mucor circinelloides* and *Rhizopus* spp. as the primary etiological agents isolated from the lung, liver, and kidney, while *Escherichia coli* was isolated exclusively from the intestine, and *Eimeria* spp. oocysts (4,300 OPG) indicated a mild coccidial infection in the recovery phase. These findings confirm disseminated mucormycosis complicated by IBD-associated immunosuppression as the cause of mortality, driven largely by poor environmental management and biosecurity lapses. Therefore, proper housing management, hygienic feed storage, structured vaccination programs including IBD booster, and strict biosecurity measures are strongly recommended to prevent mucormycosis in poultry production systems.

Keywords: *Mucor circinelloides*, biosecurity, immunosuppression, layer pullet, coccidiosis.

INTRODUCTION

Fungal infections in poultry are an important concern in the modern poultry industry because they can cause health disorders, decreased production performance, and increased morbidity and mortality, particularly in young chickens. Common mycoses in poultry include aspergillosis, candidiasis, and mucormycosis, most of which are opportunistic infections closely associated with host immunosuppression. Pathogenic fungi are widely distributed in farm environments, including litter, dust, feed, and decaying organic matter; however, birds with a competent immune system are generally able to inhibit the development of infection. Conversely, under conditions of impaired immunity, fungal spores can more readily colonize, germinate, and invade tissues, resulting in severe systemic lesions (Veterinaria Digital, 2024; Rodrigues et al., 2018). One important group of opportunistic fungi in poultry is the order Mucorales, which is known to cause mucormycosis involving the respiratory and gastrointestinal tracts, eyes, and other visceral organs (Chauhan & Roy, 2013).

Mucormycosis is a mycotic disease caused by members of the order Mucorales, including the genera *Mucor*, *Rhizopus*, and *Lichtheimia*. Among these species, *Mucor circinelloides* is one of the species most frequently isolated from clinical cases of mucormycosis and has high pathogenic potential. Infection occurs through inhalation or ingestion of spores, which subsequently germinate into invasive filamentous hyphae within host tissues. These fungi are capable of causing angioinvasion, vascular thrombosis, infarction, and tissue necrosis, resulting in progressive and potentially fatal lesions. Histopathologically, mucormycosis is characterized by broad, ribbon-like, non-septate or sparsely septate (pauci-septate) hyphae with irregular, wide-angle branching, approximately 90°, accompanied by inflammatory responses such as heterophilic or neutrophilic infiltration, granuloma formation, and chronic pyogranulomatous inflammation (Rodrigues et al., 2018). In poultry and other avian species, mucormycosis lesions are most frequently found in the lungs and gastrointestinal tract, although dissemination to the liver, brain, sinuses, skin, and other organs may occur in advanced cases (Galosi et al., 2022).

The involvement of the fungus *Mucor circinelloides* as a causative agent of mucormycosis in birds has been confirmed through histopathological and molecular approaches. Galosi et al. reported a case of mucormycosis in a psittacine bird (*Agapornis roseicollis*) caused by *M. circinelloides*, which was confirmed using molecular analysis of formalin-fixed paraffin-embedded (FFPE) tissue. Histopathological examination revealed broad, non-septate hyphae in the lungs and liver accompanied by multifocal granulomatous lesions. Granuloma formation represents part of the host defense mechanism against Mucorales infection. An experimental study by Inglesfield et al. demonstrated that phagocytes are rapidly recruited to the site of infection and form innate granulomas as an attempt to limit fungal dissemination. However, under immunosuppressive conditions, these protective mechanisms become ineffective, allowing the infection to progress to a more severe and systemic form.

One of the major predisposing factors involved in opportunistic fungal infections in poultry is immunosuppressive disease, particularly Infectious Bursal Disease (IBD). This disease, caused by IBD virus, affects primary lymphoid organs, particularly the bursa of Fabricius, and exhibits tropism for immature B lymphocytes, resulting in impaired humoral immunity in chickens. Damage to lymphoid tissue caused by IBD renders chickens more susceptible to secondary infections caused by bacteria, viruses, and opportunistic fungi (Sharma et al., 2000). Under these conditions, the ability of phagocytes and the adaptive immune response to eliminate fungal spores is reduced, allowing Mucorales to develop more readily within tissues. Therefore, the occurrence of mucormycosis in poultry not only indicates high environmental exposure to

fungal spores but may also be associated with underlying immunological disorders, including Infectious Bursal Disease.

MATERIALS AND METHODS

Ethical Approval for Animal Subjects

The case animal, protocol number 247/N/26, was a 5-week-old female layer chicken originating from a private layer farm in Sebudi Village, Selat District, Karangasem Regency, Bali Province, Indonesia, which used a semi-closed-house housing system. The chicken was reported to have developed health problems before eventually dying and was subsequently submitted as a case for diagnostic investigation.

Anamnesis and Epidemiology

Epidemiological data were collected through direct observation at the farm and interviews with the farmer or farm manager. The information collected included the farmer's identity, general description of the poultry enterprise, housing system and conditions, health history of the chickens, management measures that had been implemented, and environmental conditions of the poultry house. All collected data were subsequently analyzed to determine morbidity, mortality, and case fatality rates (CFR), and were also used as supporting information for establishing the diagnosis (Khan et al., 2024).

Anatomical and Histopathological Pathology

The deceased case chicken was necropsied at the Laboratory of Veterinary Pathology, Faculty of Veterinary Medicine, Udayana University, for examination, documentation, and collection of samples based on gross pathological changes. The examination included evaluation of the shape, size, color, consistency of the organs, and the presence of characteristic lesions in the tissues. Organs showing gross pathological changes were collected and cut into approximately $1 \times 1 \times 1$ cm pieces and then fixed in 10% Neutral Buffered Formalin (NBF). The fixed tissues were subsequently processed for histopathological examination through graded dehydration, clearing with toluene, paraffin infiltration and embedding, sectioning at 5 μ m thickness, routine Harris hematoxylin-eosin (H&E) staining, and examination under a light microscope (Sewoyo et al., 2026).

Fungal Examination

Fungal isolation was performed by culturing samples on Sabouraud Dextrose Agar (SDA). Lung and tracheal tissues were aseptically inoculated onto the medium and incubated at room temperature for 3–7 days. The resulting colonies were subsequently examined macroscopically, followed by microscopic examination using 10% KOH preparations and methylene blue staining to identify fungal morphological characteristics (Suarjana, Rompis, Tono, Besung, & Mahatmi, 2024).

Bacterial Examination

Bacteriological examination was performed to determine the possibility of secondary bacterial infection in addition to parasitic agents. All procedures were conducted at the Laboratory of Veterinary Bacteriology and Mycology, Faculty of Veterinary Medicine, Udayana University. Organ samples consisting of the lungs, liver, kidneys, and intestines were inoculated onto Nutrient Agar (NA) and MacConkey Agar (MCA) using the streak-plate method and incubated at 37°C for 18–24 hours. Following incubation, bacterial colonies were examined based on their macroscopic characteristics (Apriyanthi and Widayanti, 2022).

Further identification included Gram staining and a series of biochemical tests, namely catalase, Triple Sugar Iron Agar (TSIA), and IMViC tests, including Sulfide Indole Motility (SIM), Methyl Red–Voges-Proskauer (MR-VP), Simmons Citrate Agar (SCA), and sugar fermentation tests. All tests were incubated at 37°C for 18–24 hours, and the results were evaluated based on changes in medium color, gas production, H₂S production, and bacterial growth patterns (Deptan, 1999; Apriani et al., 2023).

Parasitological Examination

Parasitological examination was performed using routine fecal analysis, including native smears, sedimentation, and flotation. Egg per Gram (EPG) counts were determined using the McMaster method, and preserved worm specimens were prepared.

Data Analysis

All epidemiological data, gross pathological and histopathological findings, and fungal, bacterial, and parasitological identification results were analyzed descriptively and qualitatively. The findings were used as the basis for determining the cause of death in the case chicken.

RESULTS AND DISCUSSION

Results

The case animal was a 35-day-old female ISA Brown pullet from a flock of approximately 800 birds. The farm used a semi-closed-house system and was located near a forested area. The housing was poorly maintained, with infrequent cleaning and litter replacement. Feed was stored in an open area, potentially increasing exposure to environmental fungal spores. Biosecurity measures were also inadequate, as access to the farm was not restricted and no disinfection facilities were available at the entrance. Clinically, the case chicken showed dull feathers, yellowish-white fecal contamination around the cloacal region, and dry legs. The bird also exhibited dyspnea, difficulty breathing, weakness, inability to stand, reduced activity, and decreased feed intake. These clinical and external findings are presented in Figure 1A. Within the flock, 9 chickens were reported to have developed clinical signs and 4 subsequently died. Based on a flock population of 800 birds, the morbidity, mortality, and case fatality rates were 1.13%, 0.50%, and 44.44%, respectively.

Necropsy revealed multifocal white nodules in the thoracic region, particularly involving the respiratory organs. White nodules were observed around the thoracic area, as shown in Figure 1B. The air sacs appeared cloudy and contained multifocal white nodules (Figure 1C). The lungs showed multifocal white nodules accompanied by hemorrhagic areas (Figure 1D). Similar white nodules were also observed on the liver and kidneys, with mild hemorrhage noted in the kidneys (Figure 1E–F). The gastrointestinal tract showed uneven reddish-brown discoloration involving both the small and large intestines (Figure 1G). The bursa of Fabricius showed mild reddish discoloration (Figure 1H). These gross pathological findings indicated that the lesions were predominantly associated with the respiratory system, with additional lesions involving visceral organs, particularly the liver and kidneys.

Histopathological Findings. Histopathological examination of the lungs revealed areas of necrosis and congestion associated with pneumonia haemorrhagica et necroticans (Figure 2A–B). At higher magnification, giant cells and inflammatory cell infiltration consisting of lymphocytes, heterophils, and macrophages were observed (Figure 2C–D). Non-septate fungal hyphae were also identified within the pulmonary tissue (Figure 2E), supporting the presence of a fungal infection in the lungs. Histopathological examination of the liver demonstrated

hepatic congestion (Figure 3A). The kidneys showed infiltration of macrophages and lymphocytes in the intertubular areas, accompanied by glomerulitis and necrosis (Figure 3B). The bursa of Fabricius exhibited lymphoid depletion (Figure 3C). In the small intestine, villous erosion and lymphocytic infiltration in the lamina propria were observed (Figure 3D). In the large intestine, particularly the cecal region, heterophilic and lymphocytic infiltration in the lamina propria, necrosis of the crypts of Lieberkühn, and hemorrhage were observed (Figure 3E).

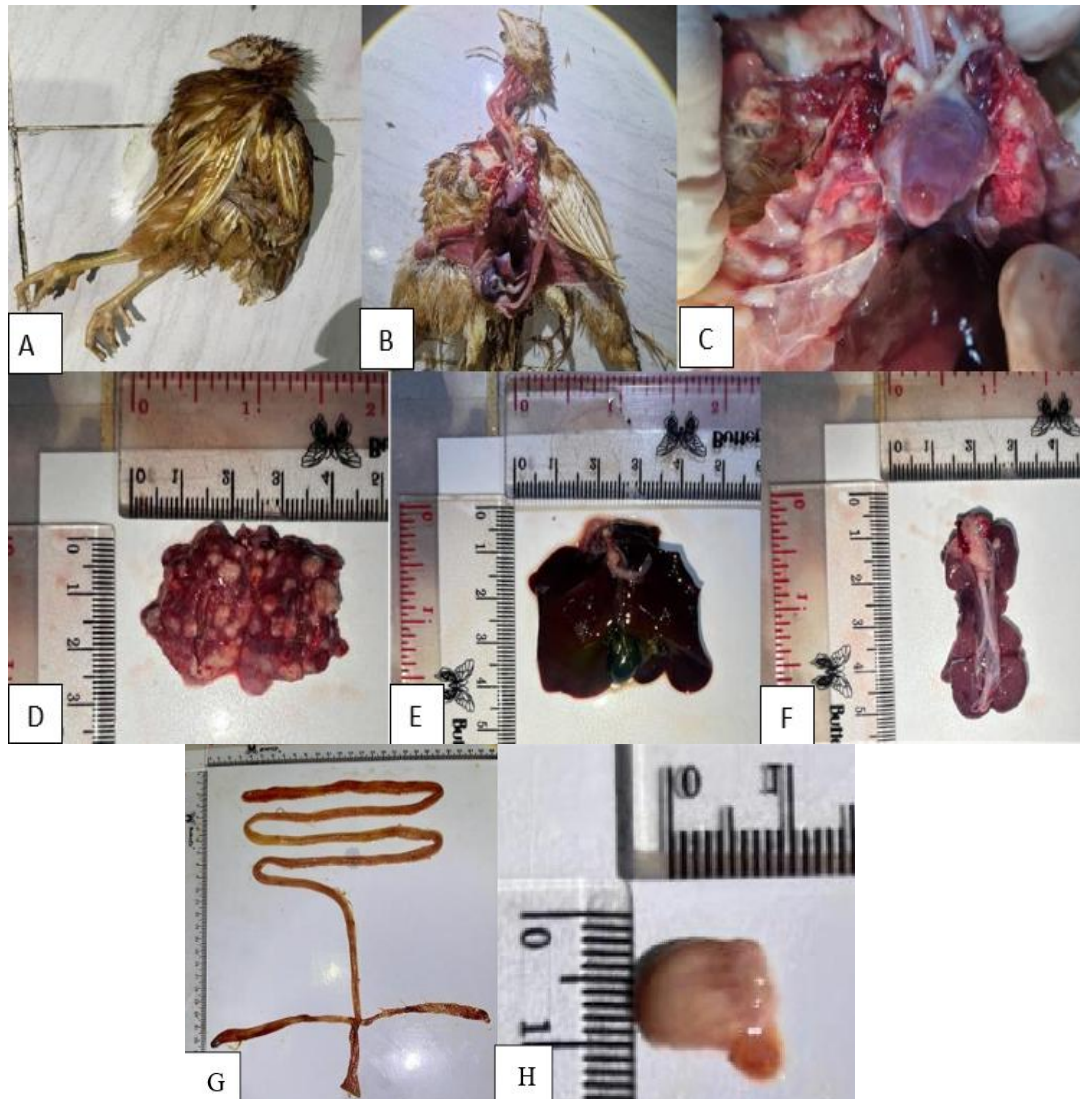


Figure 1. Gross pathological findings of the case chicken; (A) Condition of the case chicken before necropsy; the feathers appeared dull, yellowish-white fecal material was present, and the legs appeared dry. (B) Condition of the chicken after necropsy; white nodules were observed in the thoracic region. (C) White nodules and cloudy appearance of the air sacs. (D) Multifocal white nodules and hemorrhage in the lungs. (E, F) The liver and kidneys showed white nodules on the surface, with mild hemorrhage observed in the kidneys. (G) Uneven reddish-brown discoloration of the small and large intestines. (H) Slight reddish discoloration of the bursa of Fabricius.

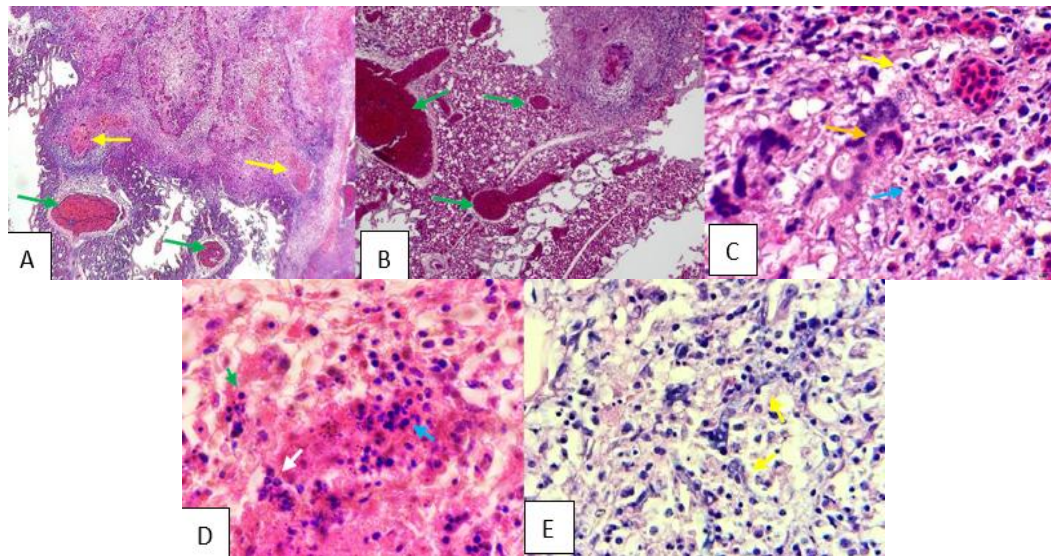


Figure 2. Gross pathological and histopathological findings of the lungs; (A, B) Pneumonia haemorrhagica et necroticans, with areas of necrosis (yellow arrows) and congestion (green arrows) (H&E, 100×). (C) Giant cells (orange arrow) and infiltration of lymphocytes (yellow arrow), heterophils (white arrow), and macrophages (blue arrow) (H&E, 1000×). (D) Infiltration of heterophils (white arrow), lymphocytes (green arrow), and macrophages (blue arrow) (H&E, 1000×). (E) Non-septate fungal hyphae in the lung tissue (yellow arrow) (H&E, 1000×).

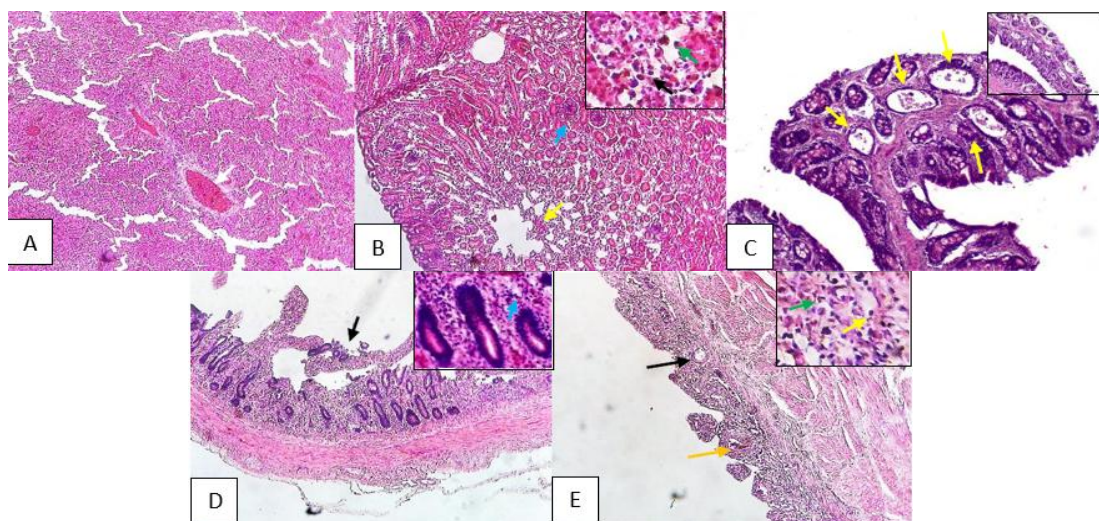


Figure 3. Gross pathological and histopathological findings of the organs; (A) Liver (H&E, 400×), showing hepatic congestion (white arrow). (B) Kidney (H&E, 400× and 1000×), showing infiltration of macrophages (green arrow) and lymphocytes in the intertubular area (black arrow), glomerulitis (blue arrow), and necrosis (yellow arrow). (C) Bursa of Fabricius (H&E, 100× and 400×), showing lymphoid depletion (yellow arrow). (D) Small intestine (H&E, 400× and 1000×), showing villous erosion (black arrow) and lymphocytic inflammatory cell infiltration in the lamina propria (blue arrow). (E) Large intestine (H&E, 400× and 1000×), showing heterophilic inflammatory cell infiltration (green arrow) and lymphocytes in the lamina propria (yellow arrow), crypts of Lieberkühn necrosis (black arrow), and hemorrhage (orange arrow).

Mycological Examination. Fungal culture of the lung tissue on Sabouraud Dextrose Agar (SDA) produced fungal colonies that were subsequently examined using KOH and methylene blue preparations. Microscopic examination identified fungal elements consistent with *Mucor* sp. and *Rhizopus* sp. (Figure 4-1A–B). Fungal cultures obtained from the liver and kidney tissues also demonstrated the presence of *Mucor* sp. and *Rhizopus* sp., as identified by microscopic examination using methylene blue and KOH preparations (Figure 4-2A–B). The isolation of these fungi from the lungs, liver, and kidneys was consistent with fungal involvement extending beyond the respiratory system.

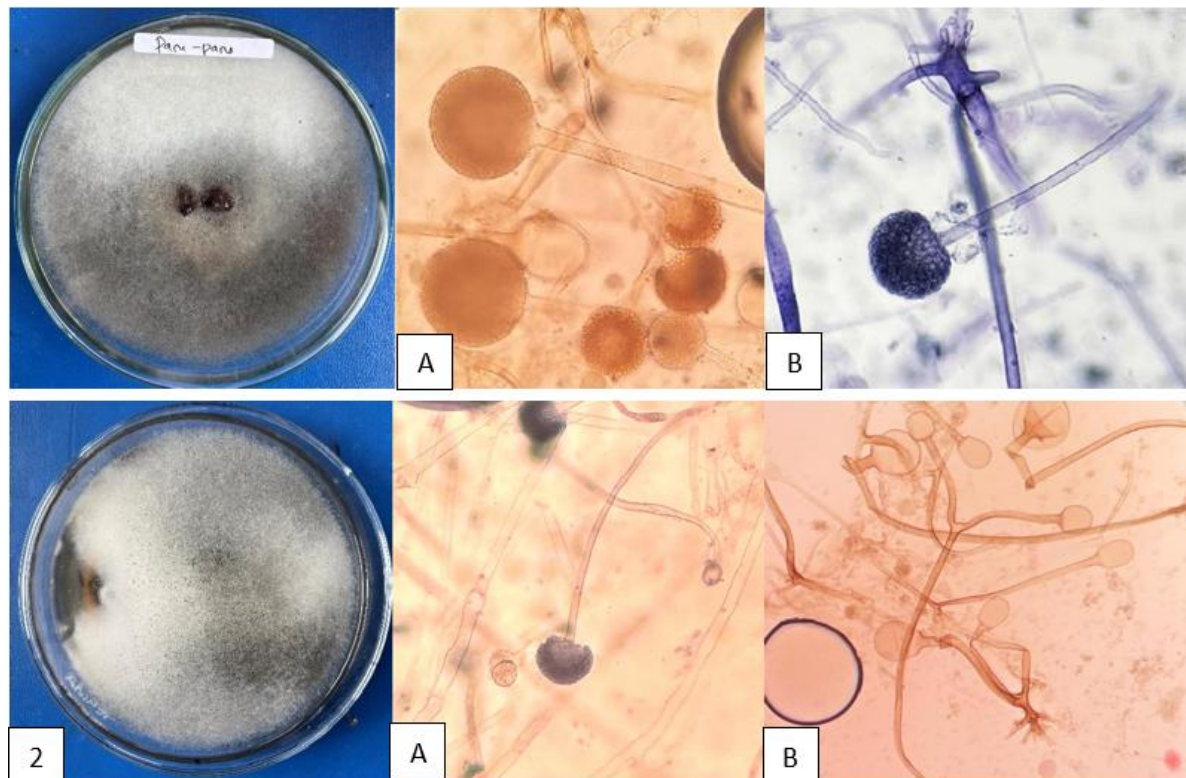


Figure 4. Fungal culture and identification from organs of the case chicken; (1) Fungal culture on Sabouraud Dextrose Agar (SDA) using lung tissue; (1A) KOH preparation (400×) and (1B) methylene blue staining (400×), identifying *Mucor* sp. and *Rhizopus* sp. (2) Fungal culture on Sabouraud Dextrose Agar (SDA) using liver and kidney tissues; (2A) methylene blue staining (400×) and KOH preparation (100×), identifying *Mucor* sp. and *Rhizopus* sp.

Bacteriological and Parasitological Findings. Bacterial culture on Nutrient Agar (NA) and MacConkey Agar (MCA) demonstrated bacterial growth (Figure 5A–B). Gram staining revealed pink, rod-shaped, Gram-negative bacteria occurring singly (Figure 5C). Based on the biochemical identification tests, the isolate was identified as *Escherichia coli*. Parasitological examination revealed an unsporulated *Eimeria* sp. oocyst, characterized by an oval shape and a completely formed oocyst wall without visible sporocysts or sporozoites (Figure 5D). Quantitative fecal examination using the McMaster method revealed an oocyst count of 4,300 oocysts per gram (OPG).

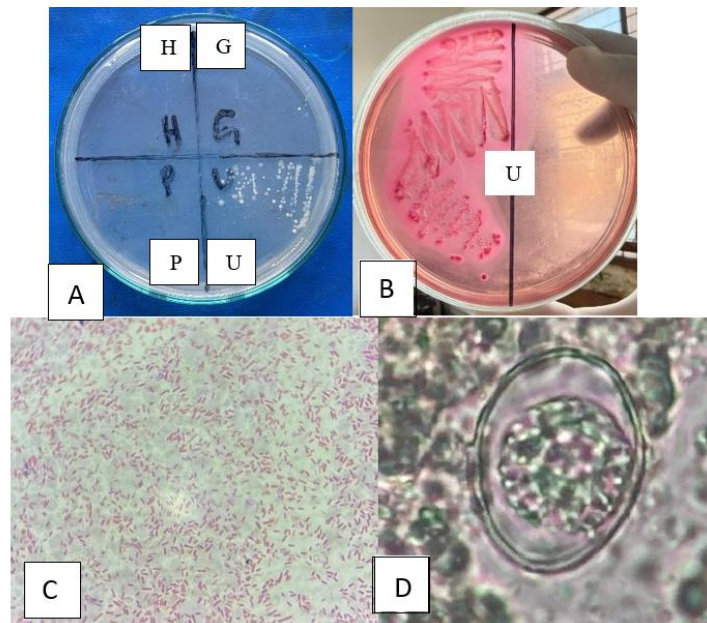


Figure 5. Bacterial and parasitological examination; (A) Bacterial culture on general-purpose medium (NA). (B) Bacterial culture on MacConkey Agar (MCA). (C) Gram staining showing Gram-negative, single rod-shaped bacteria appearing pink. (D) Unsporulated *Eimeria* sp. oocyst (oval-shaped, without sporocysts or sporozoites, with a fully formed oocyst wall).

Discussion

Mucormycosis is an opportunistic fungal infection caused by fungi belonging to the order Mucorales, including the genera *Mucor*, *Rhizopus*, and *Absidia*, which are generally saprophytic but may become pathogenic in immunocompromised hosts or following exposure to high concentrations of spores (Chayakulkeeree et al., 2006). Unlike aspergillosis, which is more frequently reported in poultry, mucormycosis is relatively rarely documented, although its causative agents are ubiquitous and can be found in litter, contaminated feed, and moist poultry-house dust (Refai et al., 2016). In the present case, the semi-closed-house system, minimal cleaning frequency, infrequent litter replacement, open feed storage, and inadequate biosecurity measures at the farm located near a forested area in Sebudi, Karangasem, Bali, were suspected to be the major predisposing factors increasing the concentration of fungal spores in the air and consequently increasing the likelihood of inhalation by the chickens (Tablante, 2013; Sousa et al., 2018). The tropical climate of Indonesia, characterized by high temperature and humidity, also supports optimal growth of fungi belonging to the order Mucorales in poultry-house environments (Ribes et al., 2000).

The clinical signs observed in the case chicken, including dyspnea, difficulty breathing, inability to stand, dull feathers, lethargy, decreased appetite, and white diarrhea, were consistent with the clinical manifestations of respiratory mucormycosis in poultry, which occurs due to fungal hyphal invasion of the pulmonary and air sac tissues (Raper & Fennell, 1965; Chayakulkeeree et al., 2006). The severe respiratory distress observed in this case was most likely caused by the growth of angioinvasive *Mucor circinelloides* hyphae, which can invade blood vessels and cause thrombosis, tissue necrosis, and severe impairment of oxygen exchange (Dannaoui, 2017).

This interpretation was supported by the gross pathological findings of white nodules in the lungs, air sacs, trachea, liver, and kidneys, which represent characteristic lesions of avian mucormycosis with systemic dissemination (Refai et al., 2016). The histopathological findings

of non-septate fungal hyphae accompanied by pneumonia haemorrhagica et necroticans further supported the diagnosis of mucormycosis in this case. The broad, non-septate (pauci-septate) hyphae are characteristic of fungi belonging to the order Mucorales and distinguish them histopathologically from *Aspergillus* spp., which possess septate hyphae with acute-angle branching (Walls et al., 2014; Spellberg et al., 2005). In addition, giant cells, lymphocytic, heterophilic, and macrophage infiltration were observed in the necrotic areas, reflecting a granulomatous immune response aimed at limiting fungal dissemination. Giant cells, or multinucleated giant cells, are formed through the fusion of macrophages that are unable to phagocytose the fungal hyphae individually because of their large size and are generally observed at the margins of necrotic areas, forming a boundary between necrotic and healthy tissue (Lim et al., 2023).

Heterophilic infiltration reflects an acute innate immune response, with heterophils acting as early effector cells recruited to the site of infection for phagocytosis and lysosomal enzyme release. Meanwhile, lymphocytic and macrophage infiltration indicates the involvement of the adaptive immune response in eliminating the infectious agent (Kogut et al., 2017; Ibrahim et al., 2012). Despite the formation of these cellular immune responses, their effectiveness was insufficient to control fungal growth completely because of the immunosuppressive condition experienced by the case chicken, as discussed further below (Spellberg et al., 2005; Dannaoui, 2017). The systemic dissemination observed in this case was closely associated with the finding of lymphoid depletion in the bursa of Fabricius on histopathological examination. Lymphoid depletion of the bursa of Fabricius is a pathognomonic lesion of Infectious Bursal Disease (IBD), an immunosuppressive disease caused by an avibirnavirus belonging to the family Birnaviridae that targets proliferating B lymphocytes in the bursa of Fabricius (van den Berg, 2000).

Chickens infected with IBD experience destruction of bursal immune cells, resulting in severe immunosuppression and an inadequate immune response against opportunistic pathogens, including Mucorales fungi (Dormitorio et al., 2013). In this case, although IBD vaccination was reportedly administered only at the DOC stage at the hatchery without subsequent booster vaccination, failure of vaccine protection or reinfection with a more virulent field strain at 35 days of age was considered possible. This is a period when chickens are particularly susceptible to IBD (Mahardika et al., 2016). The immunosuppressive condition associated with IBD was therefore suspected to be a major factor enabling *Mucor circinelloides* to progress from a localized pulmonary infection to disseminated mucormycosis involving the liver and kidneys (Refai et al., 2016). The pathogenesis of this case began with *Mucor circinelloides* spores being inhaled from the contaminated poultry-house environment and deposited in the lower respiratory tract and lungs. In chickens with normal immunity, these spores would be phagocytosed by alveolar macrophages and neutralized. However, in chickens experiencing immunosuppression due to IBD infection, impaired phagocytic function allows the spores to germinate into invasive hyphae (Spellberg et al., 2005).

The extensive invasion and tissue damage are mediated not only by the mechanical angioinvasive properties of the hyphae but also by toxins produced directly by Mucorales fungi. The major toxin identified is mucoricin, a 17-kDa protein with structural and functional features resembling plant-derived ricin, including the ability to inhibit host-cell protein synthesis through N-glycosidase activity, the presence of a motif that mediates vascular leakage, and a lectin sequence. Antibodies against this toxin have been shown to prevent hypovolemic shock, organ necrosis, and death in mucormycosis infection models (Soliman et al., 2021). At the cellular level, mucoricin inactivates host-cell ribosomes, disrupts vascular permeability through the LDV domain, and induces both necrosis and apoptosis of host cells

simultaneously. Consequently, the resulting tissue damage occurs through two mechanisms: inhibition of protein synthesis and direct cell death (Soliman et al., 2021).

Angioinvasive *Mucor* hyphae subsequently penetrate blood vessel walls, facilitated by a specific interaction between CotH surface proteins of Mucorales and the GRP78 receptor on endothelial cells. This interaction is specific to Mucorales and has not been observed in *Candida* spp. or *Aspergillus* spp. The resulting vascular thrombosis, infarction, and pulmonary tissue necrosis manifest histopathologically as pneumonia haemorrhagica et necroticans (Baldin & Ibrahim, 2017; Dannaoui, 2017). In *Rhizopus microsporus*, which may be present as a co-infecting agent, additional toxin mechanisms include rhizoxin, an antimitotic agent produced by the endosymbiotic bacterium *Burkholderia rhizoxinica* residing within the fungal cytosol, and rhizonin, a cyclopeptide with fatal hepatotoxic effects that may synergistically aggravate visceral organ damage, including that of the liver and kidneys (Partida-Martinez & Hertweck, 2005; Partida-Martinez et al., 2007).

Continued fungal growth within the resulting necrotic tissues is also supported by iron acquisition through the siderophore rhizoferrin secreted by Mucorales. Deletion of the gene encoding rhizoferrin biosynthesis in *Mucor lusitanicus* has been shown to significantly reduce virulence, confirming that iron acquisition is an important component of infection persistence (Pérez-Arques et al., 2022). Through the bloodstream, fungal hyphae or fungal elements disseminate to other visceral organs, including the liver and kidneys, producing multifocal infectious foci, as reflected by fungal isolation from both organs. In chickens experiencing IBD-associated immunosuppression, this entire pathological cascade—from mucorin production that disrupts host-cell protein synthesis, to CotH–GRP78-mediated angioinvasion, thrombosis and ischemic necrosis, and iron acquisition that sustains fungal growth within tissues—proceeds with inadequate immune defense, ultimately resulting in multiorgan failure and death of the case chicken (Chayakulkeeree et al., 2006; Soliman et al., 2021).

Bacteriological examination revealed the presence of *Escherichia coli* isolated from the intestinal organ. *Escherichia coli* is a Gram-negative bacterium that naturally constitutes part of the normal gastrointestinal flora of poultry in limited numbers, but it can act as an opportunistic pathogen when the microbial balance is disrupted or intestinal mucosal integrity is compromised (Oladokun & Sharif, 2024). The presence of *E. coli* exclusively in the intestine and its absence from the lungs, liver, and kidneys suggests that the bacterium had not undergone systemic translocation and therefore more likely represented normal flora or a localized infection associated with intestinal mucosal alterations (Barnes et al., 2008). Intestinal lesions characterized by lymphocytic and heterophilic inflammatory cell infiltration observed on histopathology were more likely to reflect inflammatory responses associated with *Eimeria* spp. infection and IBD-associated immunosuppression than primary *E. coli* infection, although the presence of this bacterium should still be monitored because it may aggravate clinical conditions under immunosuppressive circumstances (Argarini et al., 2023).

The detection of *Eimeria* spp. oocysts at 4,300 OPG indicated mild to subclinical coccidiosis, considering that severe infection is generally characterized by oocyst counts exceeding 10,000 OPG, whereas values below 5,000 OPG are classified as subclinical infections (Ogedengbe et al., 2023). The absence of schizont stages on intestinal histopathology indicated that the asexual stage responsible for the greatest tissue damage had already occurred and that the case was in the recovery phase of coccidiosis (Damer et al., 2022). Based on the overall epidemiological, clinical, gross pathological, histopathological, and microorganism isolation findings, the death of the 35-day-old female ISA Brown layer pullet in this case resulted from a complex interaction between pulmonary and systemic mucormycosis caused by *Mucor circinelloides* as the primary disease, aggravated by IBD-associated immunosuppression, accompanied by mild

coccidiosis in the recovery phase and the presence of *E. coli* as normal flora with potential opportunistic pathogenicity.

This case report emphasizes the importance of proper poultry-house management, hygienic feed storage, structured vaccination programs including IBD booster vaccination, and strict biosecurity measures to prevent mucormycosis and immunosuppressive diseases in poultry production systems.

CONCLUSIONS AND SUGGESTIONS

Conclusions

The death of the 35-day-old female ISA Brown layer pullet in this case was caused by pulmonary mucormycosis associated with *Mucor circinelloides* infection, supported by clinical, pathological, histopathological, and microbiological findings. Immunosuppression associated with IBD infection was suspected to have contributed as an aggravating factor, allowing systemic dissemination of the fungus to the liver and kidneys. Mild coccidiosis in the recovery phase and the presence of *Escherichia coli* in the intestine were also identified as concurrent conditions. Poor housing management, unhygienic feed storage, and the absence of a subsequent vaccination program were suspected to be the major predisposing factors in this case.

Suggestions

Prevention and control of mucormycosis in poultry can be achieved through improved routine poultry-house sanitation and stricter implementation of biosecurity throughout the farm. Improvements in housing management should include optimization of ventilation, routine litter replacement, storage of feed in closed containers protected from moisture, and appropriate stocking density according to the capacity of the poultry house. These measures should be implemented to minimize fungal spore contamination in the environment. In addition, a structured vaccination program, including administration of an IBD booster vaccine at the appropriate age, is strongly recommended to prevent immunosuppressive conditions that may aggravate opportunistic infections in chickens.

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