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Mannheimia haemolytica infections in broiler breeder farms of poultry

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Abstract

Avian Pastuerellosis caused by *Pasteurella spp* has found to cause economic significance in Broiler Breeder farms in intensive poultry producing areas and persists as an infection in specific integrations or farms. Acute outbreak associated with the environment or managemental stress, may result in depression in egg production. In this study, Broiler breeder birds were bought to the laboratory with a history of mortality characterized by respiratory distress. On post mortem, air sacculitis, pericarditis along with fibrinous perihepatitis and necrotic foci like lesions were observed in the liver. The liver samples were collected for the culture of *Pasteurella* organisms and Biochemical tests were conducted to identify the species of *Pasteurella*. Bacteriological evaluation was done by culturing the samples directly into Nutrient agar, Mac Conkey agar and Blood agar. The pure colonies were isolated and stained using Gram's staining technique and based on morphology and biochemical tests, the causal organism were found to be *Mannheimia Haemolytica*.

Keywords: broiler breeder, isolation and identification, *Mannheimia Haemolytica*

Introduction

Mannheimia haemolytica (Formerly known as *Pasteurella haemolytica*), a gram negative, non motile coco-bacillus, is usually present as commensal in the upper respiratory tract of various animal species and can act as an opportunistic pathogen, causing mild to severe respiratory infections under stress conditions (DeRosa *et al.*, 2000) [5]. *M. haemolytica* is also a usual flora of the respiratory tract of chicken and animals and plays a drastic role of opportunist under stress factors by causing the respiratory disease (Taylor *et al.*, 2010) [10]. Antiabong *et al.*, 2006 [3] identifies *M. haemolytica* as secondary or co-pathogen in chicken infected with respiratory viral pathogens like Infectious Bronchitis virus (IBV) and Infectious Laryngotracheitis virus (ILT) and as a primary respiratory pathogen after its isolation from clinically ill and dead chicken. Ali *et al.*, 2015 [1] also identifies *M. haemolytica* as a primary source of disease causing severe respiratory distress in the adult poultry birds besides resulting in significant mortality and loss of production in mature chicken. A raised incidence of *Pasteurella haemolytica* with increase in age was noted both with regard to flocks and the number of chickens examined (M Bisgaard, 1977) [4]. Isolations of *P. haemolytica* from sporadic cases with different pathological manifestations involving the respiratory and digestive tracts, liver, spleen and oviduct have been made in poultry without determining whether *P. haemolytica* was a primary or secondary pathogen (Greenham and Hill, 1962) [6]. The necropsy findings of *M. haemolytica* infections revealed air sacculitis, pericarditis, perihepatitis, congested and flaccid ova with egg peritonitis (Setta, A. *et al* 2017) [8].

Materials and Methods

Dead birds around 10 weeks of age from a broiler breeder farm with a history of respiratory distress and mortality were received in the laboratory. The birds were necropsied and tissue samples from liver were collected for further laboratory investigation. Increased mortality were reported the following day and based on the post mortem lesions, samples were collected for culture of Fowl cholera organism. Bacteriological evaluation was done by culturing the samples directly into Nutrient agar, Mac Conkey agar and 5% sheep blood agar. The pure colonies were isolated and stained using Gram's staining technique and the causal organism were determined based on Morphology and biochemical tests.

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Fig 1: Air Sacculitis with Pericarditis and Fibrinous Hepatitis



Fig 2: Necrotic white foci on pale liver

Results

On post mortem examination, the major lesions observed was air sacculitis, pericarditis, fibrinous perihepatitis with necrotic white spot on liver resembling like fowl cholera lesions. The bacteriological evaluation showed white, grey, smooth colonies on nutrient agar and shiny white grayish colonies with hemolysis on 5% sheep blood agar after 24-48 hours incubation. There was no growth on Mac conkey agar. The isolated bacteria were gram negative, cocco bacilli showing silimar morphology like *Pasteurella spp.* The biochemical tests showed Maltose negative, Dulcitol Negative, Sucrose positive, Fructose positive, Mannose positive, Urease negative, Indole negative and fermented Glucose and sucrose with no production of H₂S. These biochemical evaluations revealed the presence of *Mannheimia haemolytica* (*Pasteurella haemolytica*).

Culture report



Fig 3: Nutrient Agar: white, grey, smooth colonies are seen



Fig 4: Mac Conkey agar: No growth seen



Fig 5: Blood agar: Shiny white grayish colonies seen on day 1.



Fig 6: Blood agar: Hemolysis seen on day 2.

Biochemical Tests results



Fig 7: Maltose and Dulcitol Negative.



Fig 8: Mannose Positive

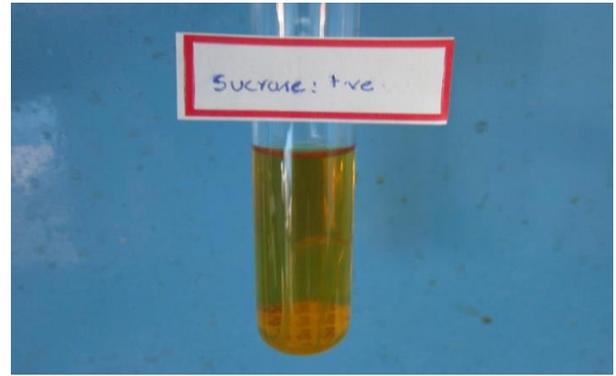


Fig 12: Sucrose Positive



Fig 9: TSI: Yellow slant/Yellow butt, No H₂S production



Fig 13: Indole Negative

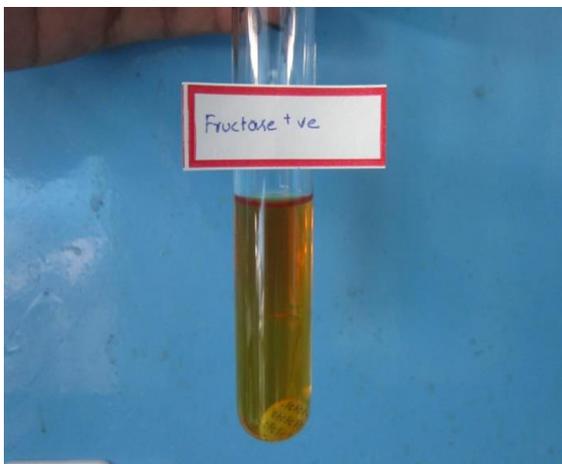


Fig 10: Fructose Positive



Fig 11: Urease Negative

Discussion

Mannheimia consists of Gram-negative, evenly stained short rods (Smith and Phillips, 1990) [9]. Ali *et al.*, 2015 [1] isolated *Mannheimia haemolytica* from adult commercial poultry flocks, initially reported with severe respiratory distress and necropsy findings were quite similar to those found in Fowl cholera infection. However *Mannheimia haemolytica* was identified as a primary source of disease which has been reported to cause a severe respiratory distress in the adult poultry birds besides resulting in significant mortality and loss of production in mature chicken.

Biscaard M (1997) found that none of the *P. Haemolytica* strains isolated from different pathological conditions were able to grow on MacConkey agar. Hawari *et al.*, 2008 [7] also found that all isolates presumed to belong to *M. haemolytica* did not produce indole and grew in MacConkey agar. All strains of *Mannheimia* ferment mannitol, glucose, maltose, sorbitol, and sucrose without gas production. Indole, urease, methyl blue (MB) and Voges-Proskauer (VP) reactions are negative. Catalase (almost always) and oxidase are positive (Smith and Phillips, 1990) [9]. *Mannheimia* can be separated from genus *Pasteurella* by not producing acid from D mannose (Angen *et al.*, 1999) [2]. Based on the cultural characteristics and biochemical tests, it can be indicated that the causative agent is *Mannheimia hemolytica*.

Conclusions

Intensive breeder farming practices have led to a variety of new emerging diseases under different kinds of stress and management practices. Prevention of Avian Pasteurelosis is based on good management practices, sanitation measures and exclusion of wild birds, rodents and other animals. Secondary infection with other pathogenic organisms have evolves the need to understand the causative organism for containment of

the disease. In this study *Mannheimia Haemolytica* was found to be pathogenic organism which was resembling *Pasteurella Multocida* in clinical signs and on post mortem examination, however the culture and biochemical tests revealed *Mannheimia Haemolytica* as the pathogen. Further molecular detection through PCR for more specific identification and confirmation of the organism is to be focused in the future.

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