Clinicopathological studies of Brooder pneumonia in Broiler Chicken

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Abstract
The present investigation was carried out to investigate the clinicopathology of aspergillosis in broiler chicken at Kamrup district of Assam. The disease was diagnosed on the basis of clinical signs, gross & microscopic lesions and isolation of the fungus. Gross lesions were characterized by presence of white-yellowish caseous nodules in the lung, airsacs, heart, thoracic wall and abdominal serosa. Microscopically focal granulomatous reaction was observed in the lungs which were characterized by central necrotic area with infiltration of heterophils, macrophages, epithelioid cells and formation of giant cell. Invasion of fungal hyphae at the peribronchiolar and interstitial tissue and haemorrhages were noticed. On the 4th day post incubation, appearance of velvety, bluish green colonies were observed on the SDA plates. Therapeutic treatment with copper sulphate orally and replacement of the litter responded rapidly with decreased mortality.

Keywords: Aspergillosis, brooder pneumonia, broiler, clinicopathology

1. Introduction
Aspergillosis is the most common fungal disease of the avian respiratory system. It is an infectious, non-contagious fungal disease caused by species in the ubiquitous opportunistic saprophytic genus *Aspergillus*, in particular *Aspergillus fumigatus* [3]. Aspergillosis also called as brooder pneumonia and have economic importance in the poultry industry, and is a frequent cause mortality in companion, aviary and free-ranging birds [8, 10]. Although aspergillosis is predominantly a disease of the respiratory tract, all organs can be involved. The disease may be chronic and insidious, or it may cause peracute death. Established aspergillosis infections are clinically challenging to resolve. The warm, humid environment of the farm sheds, feed stores, floor etc., favor its growth. It is a contaminant of every environment because of its adaptability to growth substrates and the production of spores that remain viable under extremely harsh conditions. Inhalation of air borne conidia is the principal mode of exposure. Aspergillosis in young chicks and pullets is commonly associated with overwhelming exposure to large numbers of conidia from heavily contaminated feed, litter, or the hatchery environment [4]. The present communication deals with the clinicopathological and mycological studies of Aspergillosis in a broiler flock.

2. Materials & Methods
2.1 Sample collection: The study was conducted to diagnose aspergillosis in broiler during physical visit of the farms and when submitted for post-mortem examination to the Department of Pathology, CVSc, AAU, Khanapara, Guwahati-22 (Assam). The clinical signs exhibited by the affected birds and the history provided by the farmers were properly recorded.

2.2 Necropsy and Histopathological examination: The dead birds submitted for post-mortem were examined systematically following standard protocol. The physical conditions of the birds and the gross alterations indifferent organs were carefully recorded. For histopathological examination, represented tissue samples were collected in 10% formal saline solution. After proper fixation, paraffin embedded tissue sections of 4-6µ were prepared and stained by routine Haematoxylin & Eosin technique for microscopic examination [7]. Duplicate sections were stained with Gridleys stain for demonstration of fungi in the tissue. Direct microscopic examination was performed by using Lactophenol cotton blue in the impression smear for detection of the fungi.
2.3 Isolation of the Fungus: The isolation of the *Aspergillus* spp. was carried out from the affected organs i.e. lung, liver, air sac. The samples were inoculated to Sabouraud Dextrose Agar (SDA) plates and incubated at 37 °C for 5 days. Identification was done on the basis of the colony characteristics and microscopic appearance. Suspected colonies were examined by the Wet mount method using Lactophenol cotton blue stain.

3. Results and Discussion
The affected birds showed moderate to severe dyspnoea, gasping (Fig. 1) and death. Whitish watery diarrhea, anorexia, ruffled feathers, progressive emaciation, dehydration, increased thirst were also recorded. The results are in agreement with the earlier findings \[11, 13\], who reported dyspnoea, gasping and nasal discharge in acute form of aspergillosis.

On necropsy, there was presence of white-yellowish caseous nodules in the lung (Fig. 2&3), air sac, heart, thoracic wall and abdominal serosa (Fig. 4&5). The size of the nodules varies from 2- 10 mm in diameter. The lung parenchyma was consolidated. Similar findings of nodular lesions were reported \[1, 6\]. Microscopically, focal granulomatous reaction was observed in the lungs. The granulomas were characterized by central necrotic (Fig. 6) area with infiltration of heterophils, macrophages, epithelioid cells and formation of giant cell. Invasion of fungal hyphae at the peribronchiolar and interstitial tissue and haemorrhages were noticed (Fig. 7). The alveoli and the bronchiolar lumina were filled with heterophils and necrotic debrices. Similar alterations like presence of central coagulative necrotic area in the aspergillosis lung was described \[5\]. Femenia et al., 2007 reported more severe, inflammatory lesions on the pleura and the underlying pulmonary lobules of lungs in experimentally infected *Aspergillus fumigatus* infection.

The air sac membranes were thickened due to infiltration of heterophils, lymphocytes & macrophages. Giant cells were also seen in the membrane. Duplicate section stained with Gridley’s demonstrated the fungi morphologically similar with *Aspergillus* spp.

On the 4th day post incubation, appearance of velvety, bluish green colonies were observed on the SDA plates. Microscopic examination revealed presence of unbranched conidiophores with a characteristic dome shaped vesicle at the tip, which bore long chains of conidia (Fig. 8).

The disease condition might be occurred due to improper hygiene and sanitation with improper ventilation and high humidity which led to mouldy litter at the onset of rainy season \[10\].

*Aspergillus* commonly grows in damp soils, decaying materials, organic debris and free grains. High numbers of conidia are released into the atmosphere and are inhaled by human, bird and other animals. These spores travel through upper respiratory tract to the lungs. If spores are localized in the lungs, the fungi may be disseminated to other parts of the body and the diseases often leading to death \[12\]. The diagnosis can be confirmed by demonstration of characteristic organism with their septed hyphae in tissue section \[8\]. In the present study, the disease was diagnosed on the basis of history, clinical signs, gross & microscopic examination and isolation of the fungus. The birds of the affected flock was treated with Copper sulphate solution. Fresh stock solution was prepared by dissolving 50 gm of copper sulphate in a mixture of 250 ml vinegar and 750 ml clean water. Then 2 ml of the stock solution/ litre of drinking water was given for 5 days. At the same time, Griseofulvin tab was given @ 2mg/ litre drinking water for 5 days. Apart from this, litter materials were replaced. The treatment of the flock & the litter with copper sulphate was found to be effective as the severity of the clinical sign in the affected birds reduced from 3rd day and recovered completely within 5-7 days from commencement of the treatment \[13\].
4. **Conclusion:** In conclusion, the present investigation reported the clinico-pathological study of Brooder pneumonia in broiler chicken. The disease was diagnosed on the basis of clinical signs, gross & microscopic lesions and isolation of the fungus. Therapeutic treatment with copper sulphate, Griseofulvin orally and replacement of the litter responded rapidly with decreased mortality.

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6. **Reference**