Pulmonary aspergillosis in a seventeen-day old ostrich chick (*Struthio camelus*)

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Abstract
Aspergillosis is an acute fatal to chronic disease in birds. Tissues of lung and liver from 17 days old ostrich chicks submitted to the Department of Veterinary Pathology, Madras Veterinary College, Chennai – 7 for histopathological examination from Arignar Anna Zoological Park, Vandalur, Chennai. History of sudden death and subsequent conduction of post-mortem examination was reported. Gross examination of the lung revealed multifocal grey white coloured nodules measuring about 0.5 cm to 3 cm diameter. Nodule extended deep into the parenchyma upon incision. Liver was pale in appearance. Histopathological examination revealed multiple granulomatous nodule with central caseo-necrotic areas surrounded by inflammatory cells predominantly lymphocytes, macrophages and heterophils in the lung. Multinucleated giant cells were seen. Fungal mycelia was seen with routine Haematoxylin and Eosin and Gomori’s Grocott silver stain (GG) which showed black colored fungal septate branching hyphae and was suggestive of *Aspergillus* spp. Liver showed congestion, micro vesicular to macro vesicular fatty degenerative changes. Based on the gross and histopathological examination, it was confirmed as Aspergillosis.

Keywords: Ostrich chick, pulmonary aspergillosis, histopathology, fungal hyphae, gomori grocott stain

1. Introduction
Ostrich (*Struthio camelus*) is a large flightless bird used in semi-arid and desert areas, some African countries [1]. Aspergillosis is one of the common respiratory tract infections in poultry due to managemental problems [2]. Aspergillosis is caused by *Aspergillus fumigatus* which is the primary species responsible for infection in different avian species. Aspergillosis is an acute fatal to chronic disease in birds [3]. Aspergillosis is also caused by high population density, poor ventilation, excessive administration of antibiotics, nutritional deficiency and immunosuppressive agents are important factors for the development [4, 5]. Aspergillosis is mainly caused by aspergillus fumigatus [4]. Acute aspergillosis in young chicks produces higher morbidity and mortality and chronic form particularly in older birds causes sporadic morbidity and mortality [4]. In addition to that, reduction in body weight, difficulty in respiration [6], encephalitis, ophthalmitis, osteomyelitis, dermatitis, systemic forms [7] were observed in birds.

2. Materials and Methods
Tissues of lung and liver from a 17 days old ostrich chicks submitted to the Department of Veterinary Pathology, Madras Veterinary College, Chennai – 7 for histopathological examination from Arignar Anna Zoological Park, Vandalur, Chennai. History revealed that the bird suddenly died and post-mortem examination conducted. Samples were received in 10 per cent neutral buffered formalin (NBF). The formalinised tissue samples were processed and paraffin embedded tissue sections were cut into 4-6µm thickness and stained with haematoxylin and eosin stain (H&E) [8]. The stained slides were examined under light microscope (Olympus CX20).

3. Results
Gross examination of the lung revealed multifocal grey white coloured nodules measuring about 0.5 cm to 3 cm diameter. Upon incision, the nodule was found to extend deep into the parenchyma. Liver was pale in appearance. Microscopical examination of the lung revealed multiple granulomatous nodule with central caseo-necrotic areas (Fig.1) surrounded by...
inflammatory cells predominantly lymphocytes, macrophages and heterophils. Multinucleated giant cells were also seen (Fig. 2). Fungal mycelia was seen with routine Haematoxylin and Eosin (Fig. 3) and special stain of Gomori’s Grocott methanamine silver nitrate stain (GG) showed black colored fungal septate branching hyphae suggestive of *Aspergillus* spp. (Fig. 4). Liver showed congestion, micro vesicular to macro vesicular fatty degenerative changes (Fig. 5). Based on the gross and histopathological examination (H&E staining and GG stain), it was confirmed as Aspergillosis.

**Fig 1:** Lung – Histopathology - Multiple granulomatous nodule with central caseo-necrotic areas H&E 100µm

**Fig 2:** Lung – Histopathology - Multinucleated giant cells H&E 20µm

**Fig 3:** Lung – Histopathology - Fungal mycelia with routine Haematoxylin and Eosin stain - 10µm

**Fig 4:** Lung – Special stain - Gomori’s Grocott methanamine silver nitrate stain (GG) showed black colored fungal septate branching hyphae suggestive of *Aspergillus* spp. H&E 20µm

**Fig 5:** Liver – Histopathology - Congestion, micro vesicular to macro vesicular fatty degenerative changes H&E 50µm

### 4. Discussion

Aspergillus can be seen in soil, plant debris, wood and air and the most common pathogenic species is *Aspergillus fumigatus*. In addition to that, other aspergillus species including *Aspergillus flavus*, *Aspergillus terreus*, *Aspergillus niger* and *Aspergillus versicolor* might cause aspergillosis [9, 10]. Aspergillosis generally develops in birds due to secondary causes such as stress, immunosuppression, infected hatchery, contaminated feed and water [11, 7]. In the present case, the infection could have occurred due to immunosuppression. Gross examination revealed grey white nodules of about 0.5 to 2 cm diameter. Similar gross findings were recorded by previous workers [12-14]. Histopathological examination revealed multifocal granulomatous nodule with central caseo-necrotic areas and infiltration of lymphocyte, macrophage and heterophils with multinucleated giant cells, fungal hyphae were also seen. Histopathological findings in the present study is in agreement with earlier reports of Khosravi *et al.* (2008); Araghi *et al.* (2014); Fitzgerald and Moisan (1995); Perelman and Kuttin (1992) [13, 8, 7, 10]. Gomori grocott methanamine silver nitrate stain (GG) also revealed black coloured fungal hyphae in the present study which was also reported by Khosravi *et al.* (2008); Fitzgerald and Moisan (1995) [13, 7].

In the present study, acute aspergillosis was recorded in a 17 day old ostrich chick. However, aspergillosis sporadically occurs in adult with outbreak of high morbidity and mortality.
in young birds \cite{13, 14}. Yokota et al. \cite{15} reported two year old ostrich affected with aspergillosis in lung and air sacs. Sancak and Paracikoulu \cite{16} recorded aspergillosis in air sac of 3 months old ostrich. Perelman and Kuttin \cite{14} recorded aspergillosis due to brooder borne infection with pulmonary involvement.

5. Conclusion
In the present study, to conclude, the acute aspergillosis observed in the seventeen day old ostrich could be caused by the toxins released by the fungus causing immunosuppression effects followed by death.

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7. References