PATHOLOGICAL CHANGES IN COCKATOOS (Cacatua alba) NATURALLY INFECTED WITH PSITTACINE BEAK AND FEATHER DISEASE (PBFD)

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ABSTRACT

This paper describes the signs and post-mortem findings in sixteen cockatoos that were naturally infected with Psittacine Beak and Feather Disease (PBFD) virus (psittacine circovirus). The examination of the general outer body condition showed beak surface irregularity and feather loss with skin discoloration, while the internal organs showed various changes such as hydropericardium, liver paling, spleen mottling and various degrees of enteritis. Microscopic examination of tissues and organs obtained at necropsy revealed purple globular inclusion bodies within the nucleus of the feather pulp epidermis and cytoplasm of the feather pulp macrophages, crypt's epithelium of the intestine and within some lymphoid cells of the spleen and Bursa Fabricious among the young birds. Some additional findings observed, included blood microfilarisis, inclusion bodies of Pacheco's disease, intestinal cestode and blood parasites.

KeyWords: Circovirus; Cocckatoos Disease; Psittacine Disease; PBFD in Indonesia.

INTRODUCTION

The Psittacine Beak Feather Disease (PBFD) is caused by the *circovirus* which belongs to the circoviridae family. Psittacine circovirus is the smallest virus known to be capable of causing disease at only 16 nm (nanometers) in diameter. The virus has a broad host range and could infect birds of the Cacatuidae, Psittacidae and Loridae families. The circovirus infections also have been identified in other than avian species such as pigs (Todd 2004). The PBFD infection has been reported world wide in many countries such Australia (Pass and Perry 1984; Mc Orist *et al.* 1984), Thailand (Kiatipattanasakul-Banlunara *et al.* 2002), Brazil (Werther 1998), United States of America (Jergens *et al.* 1988), Germany (Rahaus and Wolff 2003) and serologically in Indonesia. This paper is to describe the first pathological examination of those PCR positive birds in Indonesia.

MATERIALS AND METHODS

Sixteen PCR positive for circovirus cockatoos of various ages, which were culled and terminated in a commercial breeding center were donated to our laboratories for a pathological examination. Detailed necropsy was performed and tissues were fixed in 10%

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phosphate buffered formalin. Numerous areas of skin and other systemic organs were embedded in paraffin, sectioned at 4 μ m, and stained with hematoxylin and eosin (HE).

RESULTS AND DISCUSSION

Although the PBFD is already distributed worldwide, this case report represents the first documented case of PBFD in Indonesia. These cockatoos had been hatched and reared in a commercial breeding center and had been kept relatively isolated. The source of the infection was undetermined, but probably originated from birds that had been captured from the wild.

On necropsy, there were beak surface irregularity and feather loss with skin discoloration, while the internal organs showed various abnormalities such hydropericardium, liver paling, spleen mottling and various degrees of enteritis (Figure 1).

Microscopically, the skin of the examined birds showed hyperkeratinization, lymphocytic folliculitis and feather pulp necrosis (Figure 2). Some inclusions were found in the follicular epithelium and in the feather pulp's macrophages. The follicular epithelium contained glassy, basophilic nuclear inclusions. While the macrophages of the feather pulp contained multiple, globular, basophilic, and cytoplasmic inclusions (Figure 3).

The Bursa Fabricious of the young birds revealed severe depletion of the lymphoid tissue and also multifocal necrosis with stromal oedema, accompanied by moderate infiltration of lymphocytes and plasma cells. Circovirus basophilic inclusions were also apparent in these organs (Figure 4a).

There was lymphoid depletion in the spleen. There was necrosis and degeneration of the medullary macrophages and reticular epithelial cells accompanied by numerous large intracytoplasmic and few intranuclear basophilic inclusion bodies, consistent with circovirus (Figure 4b)

The lamina propria of the intestines showed infiltration of lymphocytes, plasma cells and macrophage. Occasionally basophilic cytoplasmic inclusion bodies were apparent within the crypt's epithelium.

The hepatocytes of the liver showed a diffused hydropic degeneration. Occasionally, intranuclear basophilic inclusion were apparent within the Kupfer's cells (Figure 4c). Three of the birds examined showed intranuclear eosinophilic inclusion bodies similar with the Pacheko's disease inclusions (Figure 4d).

Edematous lesions of the brain were found constantly within the brain perivascular, heart, lung and other organs of all the birds examined. This circulatory disturbance caused by numerous findings of microfilarias within the blood vessels of all the organs. In addition, blood smear examinations revealed microfilariasis and the *Haemoproteus* sp. blood parasites

Instead of the positive PCR result, the PBFD diagnosis of these birds was based on the appearance of the basophilic inclusion bodies, which were consistent with circoviral inclusion bodies as was previously reported by Ritchie et al. 1990 and Werther et al. 1998. The inclusion bodies were large, globular, multiple, basophilic, intracytoplasmically or occasionally

intranuclearly located. The inclusions were found in the skin, intestinal crypts, spleen and the Bursa of Fabricious. The extracutaneous inclusion's organ distribution was associated with the alimentary tract and lymphoreticular organ as were reported in the previous cases (Latimer et al. 1990).

The pathological changes of PBFD are highly variable. They depends on the bird age, the infection time, the disease severity and the presence or absence of secondary infections. Although the name "PBFD" is due to the external lesion of the disease, not all sick birds showed the feather and beak abnormality, as reported in the circovirus acute infection of the African grey parrots (Schoemaker 2000; Doneley 2003)

The circovirus have learned to choose dividing cells for replication, so it will attack the tissues of young birds, and those that are growing rapidly or are replaced frequently. The organs which showed active dividing cells are skin, feathers, beak, oesophagus, crop and organs of the immune system such as the thymus, cloacal bursa, and bone marrow (Raidal 1995). Viral replication of the feathers and beak will produce typical deformities in their growth, infection of the digestive organs will reduce the food digestion process and damage the immune organs resulting in depression of the immune system and thereby making it vulnerable to secondary infections by other viruses, bacteria, or fungi.

In this case, all of the examined birds showed to have heavy filarial infections, *Haemoproteus* sp blood parasites and intestinal tapeworms. While three birds have Pacheko's inclusion bodies within their liver. Circovirus Co-infection by other infectious agents were reported frequently, such as the co-infection by the pancreatic trematodiasis (Werther *et al.* 1998), cryptosporidiosis and other secondary diseases including septicemia, peritonitis, chlamydiosis, mycotic ventriculitis (Latimer 1992) and *Pseudomonas aeruginosa* (Doneley 2003).

The inclusion bodies organ distribution reflected on how the infection spread within the breeding center. PBFD is spread by inhalation of feather dust containing virus particles or by viral ingestion through crop secretions and fecal material (Ritchie et al. 1991; Pesek 1996).

There are no known treatment that has been established against circovirus infection although experimental vaccines are under development. Well developed management plans based on current knowledge of the disease can assist in reducing the disease within cockatoo populations. To control the spread of PBFD it is important to disinfect any contaminated areas and prevent contact of the infected birds with susceptible birds.

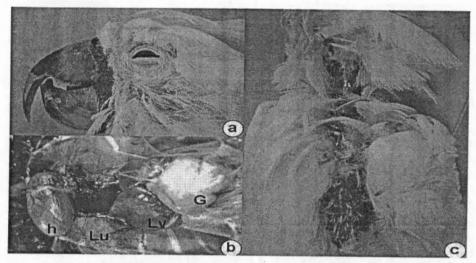


Figure 1. Beak surface irregularity (a) and feather loss with skin discoloration (c), while the internal organs (b) showed various changes such as a rounded heart (h) and pale liver (Lv). There were no significant lesions in the lung (Lu) nor the gizzard (G), but the intestines showed various degrees of inflammation

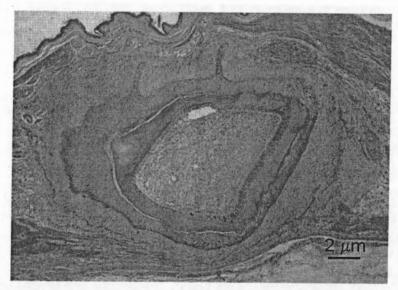


Figure 2. The skin showed hyperkeratinization, lymphocytic folliculitis and necrotic feather pulp; HE staining

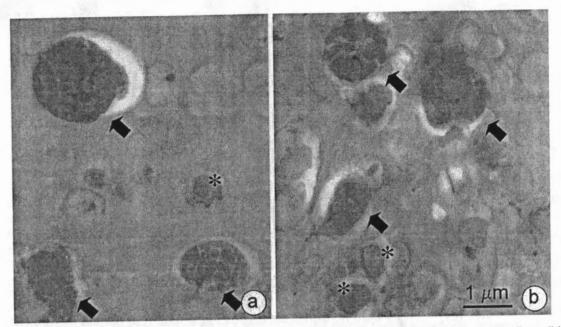


Figure 3. Circovirus inclusion bodies within the feather pulp (a) and feather epithelium (b). The cytoplasmic inclusions were multiple, globular and basophilic (arrow), while the nuclear inclusion were glassy and basophilic (a); HE staining

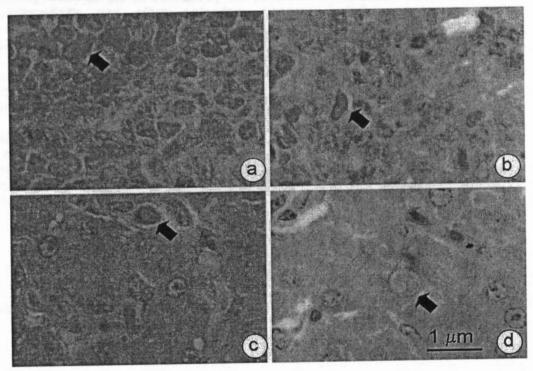


Figure 4. Extracutaneus inclusion bodies (arrows). Circovirus inclusion body of the Bursa Fabricius (a), and the spleen's necrotic follicle lymphoid (b), Kupffer's cell of the liver (c); HE staining

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