



Case Report

Disseminated avian tuberculosis in captive *Ara macao*

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Abstract

Avian mycobacteriosis was diagnosed in a captive scarlet macaw (*Ara macao*) that presented multifocal granulomas on subcutaneous tissue, sciatic nerves, infraorbital sinus, trachea, air sacs, muscles, spleen and liver. Microscopically, central areas of caseous necrosis surrounded by epithelioid macrophage, multinucleated giant cells, and lymphocytes were observed. Acid-fast bacilli were demonstrated by Ziehl-Neelsen stain. Inoculation into Löwenstein-Jensen, Stonebrink and Petagnani media, yielded *Mycobacterium* spp, which was identified as *Mycobacterium avium* by polymerase chain reaction technique (PCR).

Keywords: *Ara macao*, *Mycobacterium avium*

Mycobacterial infections are an important cause of morbidity and mortality in pet and zoological birds (20). Birds are commonly infected by *Mycobacterium avium* and *M. genavense* (20, 9) which are potentially zoonotic and most commonly infect very young, geriatric, or immunosuppressed individuals (13). Infections by *M. bovis* and *M. tuberculosis* have also been reported in birds (3, 5).

Avian tuberculosis caused by *Mycobacterium avium* have been reported in several species, including hooded merganser (*Lophodytes cucullatus*) (17), ostrich (*Struthio camelus*) (6), screech owl (*Otus asio*) (4), lesser flamingos (*Phoeniconaias minor*) (10), emus (*Dromaius novaehollandiae*) (12,18), rheas (*Rhea americana*) (16) and pheasants (*Phasianus colchicus*) (19). In canaries (*Serinus canarius*) (14) was diagnosed *Mycobacterium genavense*. In geese (*Anser anser*) (11) and maroon-faced parakeet (*Pyrrhura leucotis*) (23) and other psittacine species (15).

A 31 years old male scarlet macaw (*Ara macao*), from a zoological garden, weighting 985 grams, was attended in the Veterinary Hospital, UNESP-Jaboticabal, Brazil, with a history of four months right facial nodular swollen, one near the nostril and other under the

infraorbital sinus. The bird was bright, alert and responsive, without any behavior disturbance, with good feathering, mild beak desquamation, and normal appetite. The animal presented moderate dyspnea, evidenced by an open beak breath and an up and down tail movement with every breath.

Cardiomegaly, hepatomegaly and hyperventilation were observed in the radiographic exam. The facial swollen showed at radiography a density similar to soft tissues. A fine needle aspiration cytology of the facial mass revealed severe inflammatory infiltrate (heterophilic and granulomatous inflammation with erythrophagocytosis of unknown etiology). The bird was treated with tilosin (20 mg/ml) added in drinking water during 10 days and a single dose of vitamin A (30.000 IU/Kg), intramuscular without improvement. The animal died under anesthesia during the procedure for excisional biopsy of the mass. Complete necropsy was undertaken. The time between the treatments and the death of the animal was two months.

The necropsy was performed at the Department of Veterinary Pathology, FCAV – UNESP. The animal presented poor body condition. Grossly, several nodular

yellowish firm masses with 0.5 - 1.0 cm in diameter, were found intermingled in the right facial muscles and intermandibularis subcutaneous space (Figure 1). Similar masses were in the tracheal lumen, left abdominal air sac, right hind limb musculature (adjacent to femur-tibiotarsic joint and the sciatic nerve), and left forelimb musculature (adjacent the radio-humeral joint). Also the head presented an encapsulated and yellowish mass near the nostril (Figure 2). At cut surface of the masses there were numerous small yellowish granules with 1 - 2 mm in diameter. Similar lesions varying in size from 0.5 – 1.0cm were within myocardium and endocardium (Figure 3 and 4). There were 20 ml of hydropericardium and focal edema of the epicardium. The liver and the spleen were enlarged with several whitish superficial and parenchymal masses with 1-2 mm in diameter. Petechiae were observed on the intestinal mucosa.

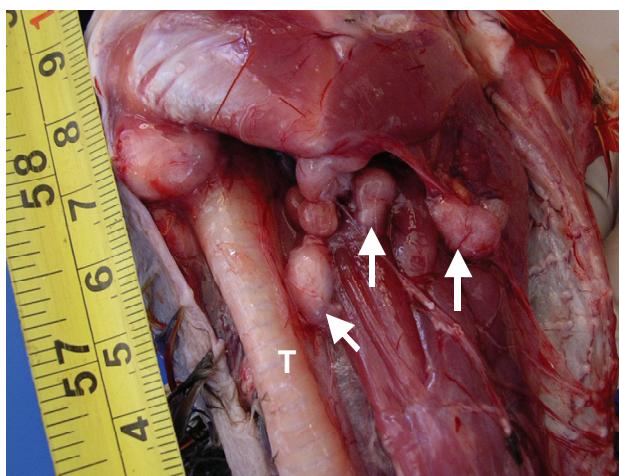


Figure 1 - Head; *Ara macao*. Multiple subcutaneous nodules (arrows) within submandibular region. T: Trachea

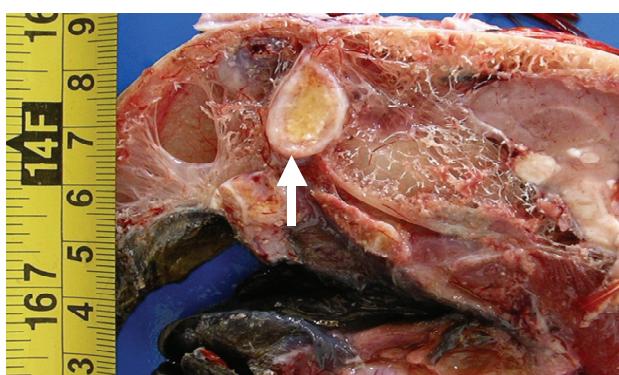


Figure 2 - Head; *Ara macao*. Notes the encapsulated and yellowish mass near the nostril (arrow).

Samples from diverse tissues were fixed in 10% neutral phosphate buffered formalin solution and processed as routinely. Paraffin sections were stained by hematoxylin and eosin (H&E) and Ziehl-Neelsen methods. The polymerase chain reaction (PCR) technique was applied in frozen samples of the granulomatous lesions.

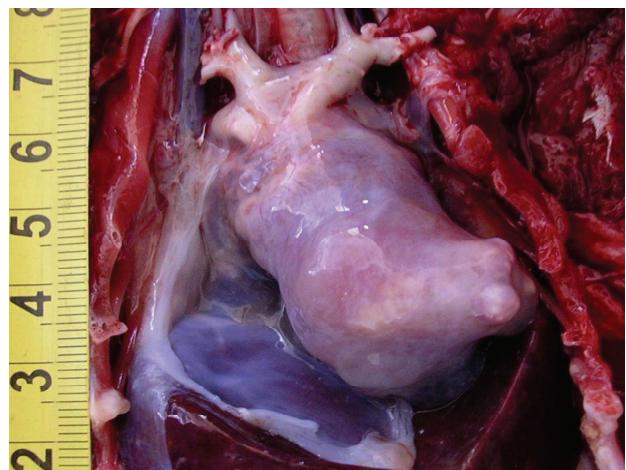


Figure 3 - Heart; *Ara macao*. Multiple whitish and firm nodules at the epicardium, under the pericardial sac.

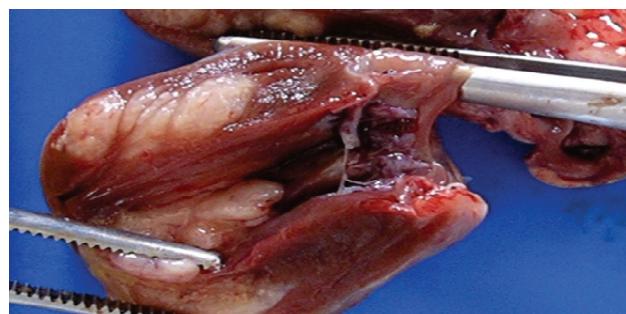


Figure 4 - Heart; *Ara macao*. Multiple whitish and firm nodules (arrow) within ventricular myocardium and endocardium

The histopathology findings were those typical of avian tuberculosis (Figure 5). Granulomas containing a central caseous necrosis, surrounded by macrophages, heterophils, lymphocytes and moderate amounts of multinucleated giant cells (Langhan's type), were present within liver, spleen, heart and nodular masses. Acid fast positive bacilli were present within necrotic tissues and cytoplasm of macrophages (Figure 6).

Frozen samples from the nodular masses and the affected organs (liver, spleen and heart) were sent to the Microbiology Laboratory from the University of São Paulo (Laboratorio de Doenças Infectiosas-Bacteriologia e Micologia – FMVZ – USP) for culture and identification. They were decontaminated according to Petroff technique, spread into Löwenstein - Jensen, Stonebrink and Petragnani culture media and maintained on 37°C for 60 days (2). The cultures were positive for *Mycobacterium* sp. 32 days after inoculation on Löwenstein - Jensen and Stonebrink media, and 35 days after inoculation on Petragnani media. The polymerase chain reaction (PCR) technique was applied in frozen samples of the nodular masses, revealing *Mycobacterium avium* as the involved organism.

Mycobacterium avium infection is relatively frequent in captive and wild birds, but no description in *Ara macao* was detected in the literature. The appearance

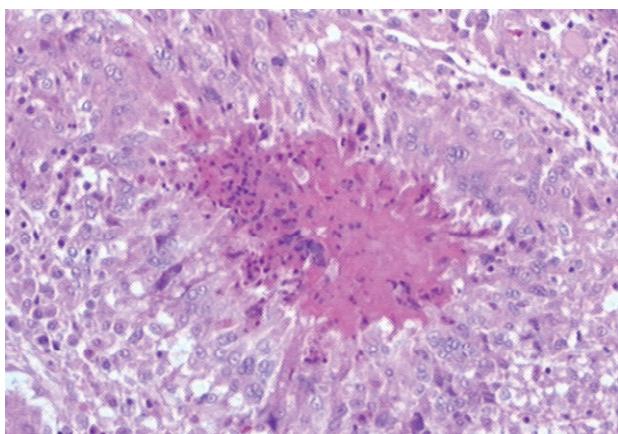


Figure 5 - Heart, *Ara macao*. Photomicrograph of the nodular mass exhibiting a typical granuloma with central necrotic core surrounded by large number of epithelioid macrophages, lymphocytes, plasma cells and multinucleated giant cells. Heart, *Ara macao* (Hematoxilin & Eosin, obj. 20 x).

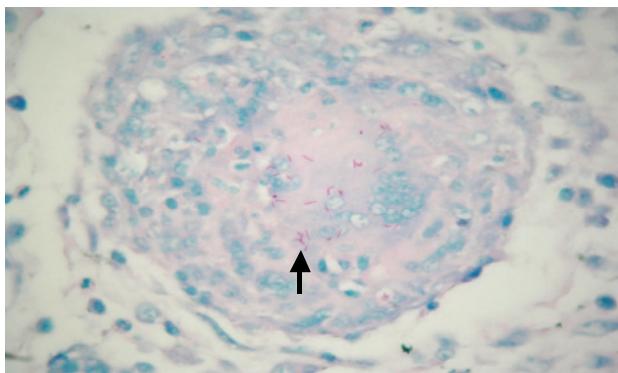


Figure 6 - Heart, *Ara macao*. Photomicrograph of the nodular mass exhibiting acid fast bacilli (arrow) (Ziehl Neelsen acid fast stain, obj. 100 X).

of clinical signs such as emaciation, muscular atrophy, and serous atrophy of subcutaneous and intracavitory fat deposits are frequent findings in birds infected by *M. avium* (7). Due to an oral route transmission, *M. avium* infection often results on gastrointestinal tract, hepatic and spleen lesions, without involvement of lungs, skeletal or other tissues (3,21). On this case report, the lesions affected not single organs, but were distributed in several organs and tissues – nostrils, trachea, air sacs, liver, spleen, heart, skeletal muscles and subcutaneous.

Mycobacterium usually causes chronic necrosis and ulceration on the tongue and granulomatous lesions in the skin, near nostrils and behind the orbital tissues, especially on Amazon parrots. Granulomas in the infraorbital sinus have already been described in poultry and an inhalator route entrance was supposed (7). Histopathologic findings of the present case were consistent to those reported by other authors (6,12). Despite absent in this case, medullar bone granulomas are occasionally present (3).

Avian mycobacteriosis in captive birds represents a serious problem due to public health concerns and the potential threat to endangered or rare species, when these ill animal was maintained in collections of zoological garden. Mycobacterial isolation and identification is important because of the emergence of *M. genavense* in the avian tuberculosis disease complex. The PCR test for this case was important and definitive for the diagnostic of the etiologic agent as *M. avium*.

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