



CASE REPORT

Kidney Myelolipoma and Amyloidosis Associated with Lung Osseous Metaplasia in Broiler Chicken

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ABSTRACT

A case of renal myelolipoma and amyloidosis associated with osseous metaplasia in a 14 day old broiler chicken in an experimental group is described. Beginning with their 6th day of life, forty broiler chickens (Ross 308) were randomly separated into two groups of 20 subjects each. The experimental group was treated with ochratoxin A (OTA) for 21 days (50 µg/kg/day) and the other one constituted the control group. Histological examination of the kidney revealed a proliferation of mature adipocytes, accumulation of numerous myelocytes, erythroblasts, eosinophilic myelocytes and heterophils, delimited by an osseous tissue surrounded by renal parenchyma. A similar ectopic metaplastic tissue was observed in the lungs. Additionally, in kidneys, skin and liver an amorphous deposit of amyloid was observed.

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INTRODUCTION

Myelolipoma is an uncommon benign extramedullary tumor of the adrenal gland, spleen and liver, composed of well-differentiated adipose tissue and a variable amount of hematopoietic cells of both lymphoid and myeloid lines. Recent experimental evidence suggests that both the myeloid and lipomatous elements have a monoclonal origin, which strongly supports the hypothesis that myelolipoma are neoplastic lesions. In veterinary literature, myelolipoma is rarely described in spleen, liver, spinal cord and adrenal gland in dogs, cats and also in ferret (Porter *et al.*, 2004; Ueno *et al.*, 2007; Dan *et al.*, 2012; Storms and Janssens, 2013).

Myelolipoma is extremely rare in birds; only few cases being reported in domestic and exotic birds, with cutaneous, subcutaneous, thoracoabdominal, splenic and liver localization.

In birds, myelolipoma may be associated with amyloidosis. Neoplastic lesions, consisting in osseous metaplasia within deposits of amyloid were previously reported, but a single case in bird Swan Goose (*Ansercygnoides*) documented the pathological features of myelolipoma and amyloidosis with osseous metaplasia in the liver (Hatai *et al.*, 2009) while it has never been reported in broiler chickens.

MATERIALS AND METHODS

The lesions were identified in a previous study (Solcan *et al.*, 2013) concerning the consequences of ochratoxin A intoxication performed on chickens. Forty 6 day old Ross 308 chickens were randomly divided in two groups: experimental (E) and control (C). The experimental group received daily, by gavage, pure ochratoxin A (99.50%) (Sigma Aldrich, Poole, UK) dissolved in sterilized sunflower oil at a dose of 50 µg/kg BW, during 21 days. The control group received diluent (sterilized sunflower oil) only. Five chickens were randomly selected from each group and were euthanized on the 7th, 14th and 21st days of the experiment. The organs including kidney, liver, spleen, skin, brain, lung, heart, intestine and ovaries were sampled and fixed in 10% neutral buffered formalin and embedded in paraffin. The sections, 5µm thick, were stained with hematoxylin and eosin for histological examination. Other staining techniques used were Giemsa in order to reveal myelolipoma cellular details, von Kossa for osseous tissue and Congo red for amyloid identification.

Findings: On necropsy examination the internal organs showed no appreciable macroscopic lesions. Microscopic examination of the lung, liver and the kidney from a 14 day old chicken revealed some unusual lesions.

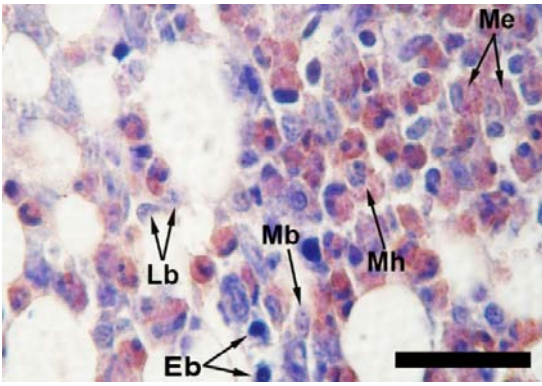


Fig. 1: Kidney myelolipoma in a 14 day old chicken. Lb - lipoblast, Eb - erythroblast; Mb - myeloblast; Me - eosinophilic myelocyte; Mh - heterophilic myelocyte. Giemsa; Bar=50 μ m.

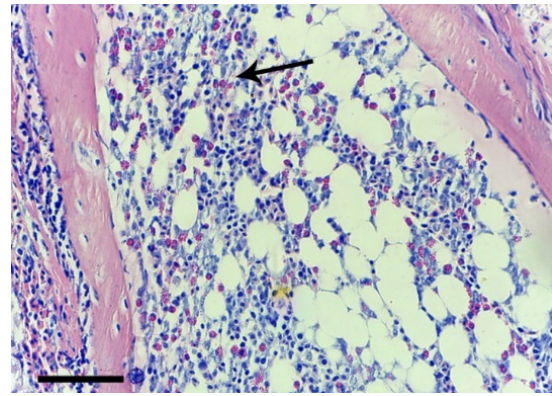


Fig. 4: Kidney myelolipoma specific cells in a 14 day old chicken. Giemsa; Bar=50 μ m.

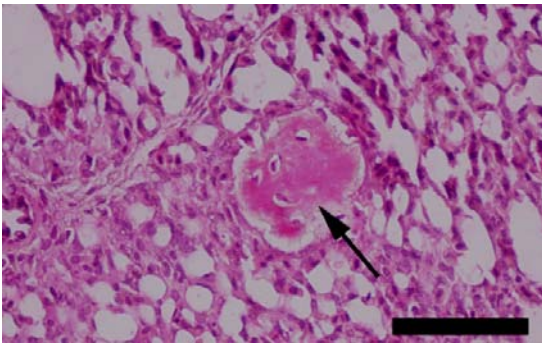


Fig. 2: Lung from 14 day old chicken. Ectopic osseous tissue (arrow). H & E; Bar=100 μ m.

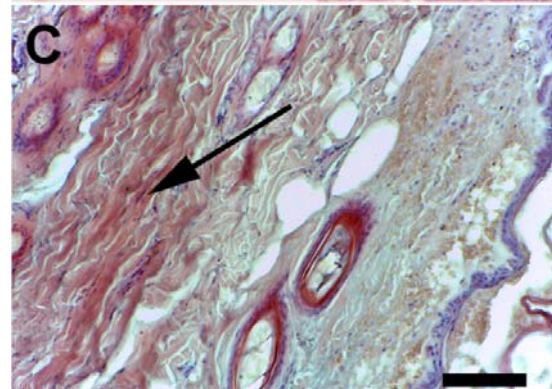
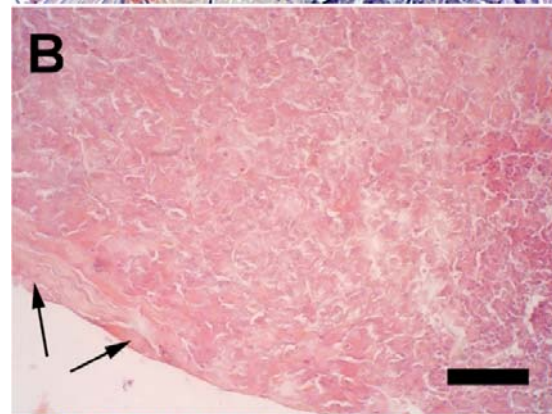
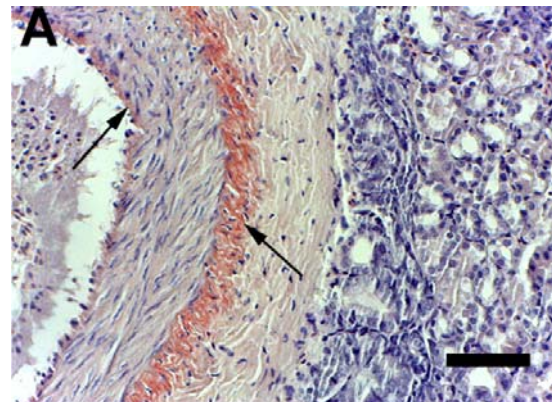


Fig. 5: A) Kidney from a 14 day old chicken. Amyloid deposits placed in arteriolar wall (arrows); B) Liver from a 14 day old chicken. Deposits of amyloid under the hepatic capsule (arrows) and C) Amyloid deposits in breast skin from a 14 day old chicken (arrow). Congo red; Bar=50 μ m A and C; Bar=100 μ m B.

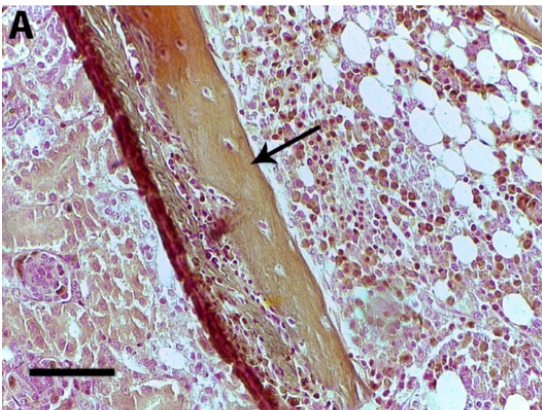


Fig. 3: A) Osseous tissue surrounding kidney myelolipoma in a 14 day old chicken (arrow). B) Ectopic osseous tissue in lung from a 14 day old chicken (arrow). Von Kossa; Bar=50 (A) and 100 μ m (B).

In kidney, proliferation of mature adipocytes, accumulation of numerous myeloblasts, erythroblasts, eosinophilic and heterophilic myelocytes was observed.

The lesions were well delimited by an acidophilic structure, surrounded by renal parenchyma (Fig. 1). Surprisingly, such a structure was also seen in the lung resembling with an osseous tissue (Fig. 2). Von Kossa staining of the kidney and lung sections reveals the osseous tissue at the border of the kidney myelolipoma (Fig. 3a) and the abnormal presence of ectopic osseous tissue in the lung (Fig. 3b). Eosinophilic myelocytes and heterophils with intracytoplasmic granules and round nuclei were identified in kidney by Giemsa stain (Fig. 4). Additionally, deposits of amyloid were identified in kidneys, liver and skin of investigated chickens. In kidney, the amyloid deposits were present in the arteriolar wall (Fig. 5a), interstitial space and in the basal membrane of the renal tubules. In liver, a mild to moderate deposits of amyloid were noted mainly in arterial walls, under hepatic capsule (Fig. 5b) and in Disse spaces. Also, amyloid deposits were present in skin from chest region (Fig. 5c). Corroborating all these aforementioned lesions, this case was diagnosed as renal myelolipoma associated with a mild amyloidosis and osseous metaplasia in the lungs.

DISCUSSION

Myelolipoma is rarely encountered in humans, the majority occurs within the adrenal glands, but several extra-adrenal ones have been reported (Talwalkar and Shaheen, 2006; Zieker *et al.*, 2008). These lesions are occasionally incidental findings and are highlighted during of diagnosis procedure (computed tomography) for other affections (Porter *et al.*, 2004). In veterinary literature, myelolipoma was reported in dogs, cats, and nonhuman primates. Like in human counterpart, myelolipoma in animal is diagnosed incidentally at necropsy, with no previous symptoms related to the lesions, except the cases when the tumor size might affect a normal function of an organ, such in a dog with spinal myelolipoma responsible for acute paraplegia.

Until now, 8 cases of myelolipoma were reported in exotic and domestic birds, with different locations: 3 in liver, 1 in cutaneous tissue, 3 subcutaneous and 1 located in thoracoabdominal region. To the author's knowledge, this is the first report of kidney myelolipoma in a broiler chicken. Some authors suggested that the proliferation of the hematopoietic cells might be a consequence of the association of myelolipoma tissue with bone marrow and therefore an infiltration with immature stem cells is induced.

Since the origin of myelolipoma is uncertain, it has been hypothesized that the starting point of the neoplasm might be represented by direct extension of bone marrow from the adjacent interosseous bone, activated by hematopoietic stimuli (Hatai *et al.*, 2008), such as prolonged hypoxia and anemia. Another speculation about the tumor origin is represented by a choristoma, which is defined as normal tissue with an abnormal localization, with the possibility to arise from choristomatous hematopoietic stem cells. Some authors suggested that since the histogenesis is not completely understood, the

myelolipoma could represent hamartoma or choristoma rather than true neoplasms. Some authors stated that few species of birds are prone to systemic amyloidosis; for *Anseriformes* this pathological disorder is recognized as an important cause of death and since the symptoms of amyloidosis are generally non-specific, for a right diagnosis is necessary histopathology examination following biopsy or necropsy. In chickens, in this respect, it has been described in 2 young layer flocks. Amyloidosis was associated with chronic inflammatory diseases, such as lameness in commercial brown layer hens (Ueno *et al.*, 2007) and inflamed joints induced by *Mycoplasma synoviae* in commercial breed of turkey and bumblefoot in swan goose (Suzuki *et al.*, 2010) have been reported. Although a concise relationship between amyloidosis and a chronic inflammation is not established, the present case might be considered as an amyloidosis consecutively to secondary infections associated with the immunosuppression caused by a prolonged ingestion of high doses of ochratoxin A.

Also, foci of ossification have been found in myelolipoma of animals. The ectopic ossification has been described in other subtypes: heterotopic ossification and osseous choristoma. Osseous choristoma is a lesion that is microscopically characterized by normal bone in abnormal sites, while the heterotopic ossification is an alteration that usually occurs as secondary to metastatic mineralization from chronic disease or dystrophic mineralization. However, in human and animal species, the formation of metaplastic bone was remarked to be associated with adrenal myelolipoma and with pulmonary amyloid deposits. It is believed, since the mechanism of ossification observed both in amyloidosis and myelolipoma is unknown, the underlying mechanism responsible is represented by a reactive non-specific lesion. As was suggested by other authors, association of the cutaneous myelolipoma with bone tissue formation is secondary to the stromal tissue formation of the myelolipoma.

Briefly, the presence of the hematopoietic cells, adipose cells with amyloid deposits and bone ossification is consistent with a diagnosis of myelolipoma associated with amyloidosis and osseous metaplasia. The findings suggest that the present lesions may be associated with ochratoxin intoxication and thus, the possible implications, remain to be investigated.

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