Unusual metastases of renal cell carcinoma in the thyroid in a dog

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SUMMARY

This report describes the case of a bilateral RCC (renal cell carcinoma) with unusual thyroid metastases in a 9-year-old male Anatolian Karabash dog with a history of lethargy, anorexia, depression, dyspnoea and snoring. At necropsy, external and internal solitary masses were found in the left and right kidneys respectively and tumoral nodules were detected in the left adrenal gland, in the regional lymphatic nodes, in lungs and also in the totally necrotic left and right thyroid glands. Histologically, the neoplastic tissues showed solid and tubular cellular structures typical of carcinoma. The RCC diagnosis was confirmed by the double vimentin (V9) and cytokeratin (AE1/AE3) immuno-labelling in the primary renal tumours as well as in the secondary tumours. This is the first report showing thyroid metastases from RCC in the dog.

Keywords: Dog, renal cell carcinoma (RCC), thyroid metastases, immunohistochemistry, vimentin, cytokeratins.

Introduction

In dogs, renal cell carcinomas (RCCs) are unilateral and uncommon and primarily metastasize to the lung and to the regional lymph nodes [8, 18]. On the other hand, they are bilateral and more common in human beings [13, 22]. Moreover, metastases into the thyroid gland besides the other organs [5, 13] can be observed in humans whereas there is no report of thyroid RCC metastases in animals.

Some tumour markers such as neuron specific enolase (NSE), S-100 protein and epithelial membrane antigen (EMA) [9] allow the diagnosis of RCC and combination of two tumour markers is usually human [20]. To the author knowledge, there are only few studies using double tumour labelling in animals such as horse and cow [15, 19, 21] and this information is lacking in the dog.

In the present case report, pathological findings and immunohistochemical characterization by both vimentin and cytokeratin labelling of a renal cell carcinoma and its thyroid metastases were described in an Anatolian Karabash dog.

Case history

A 9-year-old male Anatolian Karabash dog from the Dog Rehabilitation Centre of Kayseri Metropolitan Municipality was presented to the Department of Pathology, Faculty of Veterinary Medicine, Erciyes University for necropsy. Lethargy, anorexia, depression, dyspnoea and constantly snoring were previously reported by the referring veterinarian.

The main necropsy findings were the following:

- Firstly, a mass (12x12x5 cm³) starting from the arcus between the left and right regions of the laryngea cricoidea cartilage and extending through the thoracius cranialis aperture with the 15th trachea segment was noticed. The mass was constituted by the left and right thyroid glands and the retropharyngeal lymph nodes, oesophagus, the both sides of the arteria carotis communis, the vague nerve, the truncus sympathetic and neck muscles crossed the mass area. The mass sections were haemorrhagic and necrotic.
- Secondly, a dorsomedial mass of the left kidney originating...
TING from the front edge to the hilus renalis was observed. The cross cortical sections of this mass were partly haemorrhagic, greyish, white and yellowish.

- Thirdly, in contrast to the left kidney, the cross sections of the right kidney without an outside mass revealed the presence of a mass in the renal pelvis extending to the cortex.

- Fourthly, greyish white foci were observed within the left adrenal gland and the left regional lymphatic nodes.

- Finally, lungs were haemorrhagic, greyish-white and yellowish with multiple self limited masses that were 0.5-3.0 cm in size (Figure 1). These lung masses were haemorrhagic, necrotic, greyish-white and yellowish.

The other organs were roughly normal.

**Pathological and immunohistochemical analyses**

Abnormal tissue samples were fixed in 10% neutral-buffered formalin, embedded in paraffin wax, sectioned at a thickness 5 μm, mounted on glass slides, stained with haematoxylin and eosin, and examined with a light microscope (Olympus, BX50).

The avidin-biotin-peroxidase complex method (ABC-Shandon) was applied to detect the origin of the neoplasm. Endogenous peroxidase activity was inactivated with 3% hydrogen peroxide in methanol for 20 min. After PBS washing, 10% normal goat serum (Shandon, Pittsburgh, USA) was

<table>
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<th>Primary antibodies</th>
<th>Clones</th>
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<tbody>
<tr>
<td>Mouse anti-vimentin</td>
<td>V9</td>
<td>1:50</td>
<td>Neomarkers</td>
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<tr>
<td>Mouse anti-cytokeratin</td>
<td>AE1/AE3</td>
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<td>Neomarkers</td>
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<td>Mouse anti-thyroglobulin</td>
<td>2H11+6E1</td>
<td>1:50</td>
<td>Neomarkers</td>
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<tr>
<td>Rabbit anti-calcitonin</td>
<td>SP17</td>
<td>1:50</td>
<td>Neomarkers</td>
</tr>
<tr>
<td>Rabbit anti-synaptophysin</td>
<td>Polyclonal</td>
<td>1:50</td>
<td>Neomarkers</td>
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*Table 1: Immunohistochemical analyses: Primary antibodies used.*
Results

In kidneys, pleomorphic, cubical to columnar epithelial cells forming irregular tubules were evidenced by microscopic examination. Neoplastic cells have round to oval nuclei and mitotic figures as well as necrotic cells were frequently observed. Solid areas consisting of cells with clear cytoplasm and polygonal cells with basophilic nucleus and eosinophil cytoplasm were evidenced. These solid structures were surrounded by fibrous tissue. Dilated tubules containing hyaline cylinders were observed in the medulla.

Similar histopathological changes with solid and tubular structures (Figure 2) were also detected in the injured areas of the thyroids, the left adrenal gland, regional lymphatic nodes and lungs. Necrosis and haemorrhage were also found in the centre of large tumour nodules. Furthermore, necrotic foci were particularly abundant in the thyroid tumoral nodules. The tumoral masses were delimited by a thin parenchyma zone.

A marked vimentin immunostaining (the number of vimentin positive cells exceeded 100 in the microscopic fields (X200 and X400) presented in Figures 3a-d) was observed in the clear cells and in solid tubular structures from the primary renal tumours and from metastases in the other organs (left adrenal gland, lymphatic nodes, lungs and thyroids). In parallel, the tubular structures observed in the damaged tissues were also strongly immuno-labelled by the anti-cytokeratin antibody (the number of cytokeratin positive cells exceeded 20 and 100 in the microscopic fields presented in Figures 3e and 3f respectively). On the other hand, no significant staining of these neoplasm structures was obtained with the anti-calcitonin, anti-synaptophysin and anti-thyroglobulin antibodies.

Discussion

In the present clinical case, a bilateral renal cell carcinoma was diagnosed in a dog by conventional histology and by the double vimentin and cytokeratin immunolabelling. Moreover, metastases were evidenced in the left adrenal gland and in regional lymphatic nodes, in lungs but also in thyroids. This is the first report of RCC metastasis into the thyroid in a dog whereas the thyroid localization of the RCC metastases is relatively frequent in human beings [5, 13]. Tumoral nodules were detected in both left and right thyroid glands and this is consistent with the left and right metastasis localization observed in the human thyroid [5]. The rare occurrence of the RCC metastasis into the dog thyroid may be due to the unpredictable course of the RCC leading to non-specific clinical manifestations and radiographic findings and to a probable under-estimation of thyroid metastases. Furthermore, frequent necrotic areas were evidenced in the thyroid tumoral nodules in the present case. The similarity with thyroid carcinomas which are necrotic and haemorrhagic in the dog [6] could be related to the decreased vascularization of the thyroid glands induced by neoplasm associated to the increased secretion of iodated thyroid hormones.

Renal cell carcinomas are mainly encountered in adult (middle aged and old) and in males in dogs and in humans [1, 3, 11]. In agreement with these epidemiological data, the affected dog was adult (9 year old) and a male.

Histological and cytological patterns of the present tumours were solid/tubular and cubical/columnar respectively, in accordance with the classical histological criteria described by NIELSEN et al. [14]. Oncocytoma, a granular cell tumour in the kidney, must be distinguished from RCC. The differential expression pattern of intermediate filament proteins in subtypes of RCC and in renal oncocytomas may allow the distinction between these 2 classes of renal cell tumours [4]; whereas vimentin and cytokeratin are co-expressed in RCC, cytokeratin alone is produced by chromophobe cell carcinomas and oncocytomas. Consequently, a double positive vimentin/cytokeratin labelling is only obtained in the case of RCC. Moreover, non renal granular cell tumours are positive for vimentin and negative for epithelial cell markers [4]. The RCC diagnosis is usually confirmed by immunohistochemistry using vimentin and cytokeratin as tumour markers in humans [9, 20] and more rarely in horses [15, 21] and in cows [19]. In RCC, the expression of intermediate filament proteins is correlated with the specific morphologic appearance [16]. The prognostic value of immuno-markers depends on the histological grade of malignancy.

Immunohistochemical studies revealed that thyroglobulin, calcitonin gene related peptide, neuron-specific enolase, somatostatin and neurotensin are specific for canine thyroid gland tumours [2]. Furthermore, thyroglobulin and calcitonin are useful sensitive and specific genetic markers for follicular and parafollicular cells derived from thyroid carcinomas [7]. RAMOS-VARA et al. [17] reported that the sensitivity of thyroglobulin to detect primary thyroid carcinomas is as high as 100 % in agreement with the results of MOORE et al. [12]. In the present study, the local origin of the carcinoma nodules found in thyroids of the dog was ruled out because of the
negative thyroglobulin and calcitonin labelling. Synaptophysin, which shows immunoreactivity with 100% of carcinoma tumours, has been known as a sensitive and specific marker for tumours with neuroendocrine differentiation [10]. Again, as synaptophysin labelling was negative in this dog, such a type of tumour was discarded. RCC is usually unilateral in dog while few studies reported the scarce occurrence of bilateral RCC [3, 8, 11]. In the present case, RCC was simultaneously and unusually detected in the 2 kidneys.

As a conclusion, this adult male dog has exhibited an unusual bilateral RCC which has produced regional metastases (adrenal gland and lymphatic nodes) and distal secondary tumours in the lungs but also in the thyroids, the thyroid localisation of the metastases being quite rare in dogs contrary to humans. Consequently, when secondary carcinomas were detected in the dog thyroids, it would be relevant to look for the occurrence of a RCC.

Figure 2: a. Kidney of the 9 year old male dog. Solid (S) and tubular areas (T) (with hyperchromatic nucleus (arrowheads) in the tubular area) divided by a fibrous stroma (white arrows). Polygonal cells with basophilic nucleus and granular cytoplasm (black arrows) in solid area. Haematoxylin and eosin, X400. b. Kidney of the 9 year old male dog. Clear cells with vacuolated cytoplasm that appears honeycomb (black arrows). Haematoxylin and eosin, X400. c. Thyroid of the 9 year old male dog. Solid areas of RCC metastases (black arrows). Haematoxylin and eosin, X200. d. Lung of the 9 year old male dog. Solid areas of RCC metastases (black arrows). Haematoxylin and eosin, X200.
FIGURE 3: a. Kidney of the 9 year old male dog. Solid areas stained with vimentin. ABC method, Harris’ haematoxylin counterstain, X400. b. Kidney of the 9 year old male dog. Clear cells areas stained with vimentin. ABC method, Harris’ haematoxylin counterstain, X400. c. Thyroid of the 9 year old male dog. Metastatic tumour cells stained with vimentin. ABC method, Harris’ haematoxylin counterstain, X200. d. Lung of the 9 year old male dog. Metastatic renal cell carcinoma with interspersing solid (S) and tubular areas (T) stained with vimentin. ABC method, Harris’ haematoxylin counterstain, X200. e. Kidney of the 9 year old male dog. Tubular structures (arrow) stained with cytokeratin ABC method, Harris’ haematoxylin counterstain, X400. f. Lung of the 9 year old male dog. Metastatic tumour cells stained with cytokeratin. ABC method, Harris’ haematoxylin counterstain, X200.
References


