Mast cell leukemia in a dog

Mastcelleukemie bij een hond

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ABSTRACT

Cutaneous mast cell tumors are common in dogs, but systemic involvement is rare. In these rare cases, however, neoplastic mast cells can appear in large numbers in the peripheral blood, giving rise to mast cell leukemia or systemic mastocytosis. In both cases, the liver, spleen and bone marrow are often infiltrated with mast cells. Mast cell leukemia originates in the bone marrow and is characterized by a diffuse bone marrow infiltration with mast cells. The present case report describes the results of the blood cell count (CBC), serum biochemical panel, autopsy, histopathology and immunohistology of a 14-year-old male Jack Russell terrier with mast cell leukemia without cutaneous involvement.

SAMENVATTING

Cutane mastceltumoren komen vaak voor bij de hond. Bij mastcelleukemie en systemische mastocytose bevindt zich een groot aantal tumorale mastcellen in het perifeer bloed. Bij deze gevallen zijn de lever, de milt en het beenmerg vaak geïnfiltreerd met mastcellen. Mastcelleukemie ontstaat in het beenmerg en wordt gekenmerkt door een diffuse infiltratie van het beenmerg met mastcellen. In deze casereport worden de resultaten beschreven van de hematologie, biochemie, autopsie, histopathologie en immunohistochemie van een 14 jaar oude Jack Russell terriër reu met mastcelleukemie zonder cutane mastocytoma's.

INTRODUCTION

One biological characteristic of canine cutaneous mast cell tumors is that they may spread systemically, behaving like a hematopoietic malignancy (i.e. lymphoma or leukemia). In these cases, there is usually a history of a previously excised cutaneous mastocytoma (Couto, 1992). In some instances, neoplastic mast cells appear in the peripheral blood in considerable numbers, giving rise to mast cell leukemia without a skin tumor or other peripheral tissue involvement (Jain, 1986).

Mast cell tumors (MCTs) are common tumors of the canine skin, estimated to represent up to 20% of all skin tumors in this species (Dorn and others, 1968). Primary gastrointestinal MCTs have been reported and should be distinguished from visceral MCTs. Visceral or systemic MCTs in dogs are mostly associated with a high grade primary cutaneous MCT (Ozaki *et al.*, 2002). Very rarely, a form of mast cell leukemia has been reported. (Dobson and Scase, 2007)

Mast cells originate from myeloid stem cells; therefore mast cell leukemia should be regarded as a myeloproliferative disorder (Metcalfe *et al.*, 1997). Mast cell leukemia originates in the bone marrow and is characterized by uncontrolled and progressive proliferation and infiltration of mast cells into various organs (Jacobs *et al.*, 2002). Mast cells and basophils originate in the bone marrow from the same pluripotent stem cell (CD34+). After release from the bone marrow, mast cells are distributed to various peripheral tissues via the blood (hematogenous) as nongranulated mononuclear cells. In the peripheral tissues, mast cells further differentiate (i.e. develop granules) and have a lifespan ranging from weeks to months (Dahm *et al.*, 2001). Normally, mast cells do not circulate in the peripheral blood, but they are present in most tissues of the body (Dahm, 2001). Although mast cells and basophils share certain similarities in mediator content, histochemical characteristics and function, they are two different cell lineages (Jacobs *et al.*, 2002). The development and the secretory function of mast cells are stem cell factor dependent and the ligand for the receptor is encoded by c-kit, which is not the case for basophils (Jacobs *et al.*, 2002). The pathogenesis of mast cell leukemia is largely unknown, but in humans and dogs a derangement of the c-kit receptor and/or its ligand likely plays a primary role (London *et al.*, 1999). Also, the differentiation from systemic mastocytosis is not obvious, as this disorder also often involves the liver, spleen, lymph nodes and bone marrow, and the presence of neoplastic mast cells in

the blood (Dahm *et al.*, 2001). Once the tumor is identified in all tissues, including the bone marrow, the terms mast cell leukemia and systemic mastocytosis are really indistinguishable (R. Alleman, personal communication).

The present case gives a description of a dog diagnosed with mast cell leukemia without cutaneous involvement based on the results of CBC, the abnormalities found on autopsy, and the results of histopathologic and immunohistochemical examination of the bone marrow, liver and spleen.

CASE REPORT

A 14-year-old intact male Jack Russell terrier was presented to the local veterinarian with diarrhea and a painful abdomen for 1 week. According to the veterinarian, the dog did not have a history of other diseases during the last 8 years when it had been under his supervision. No significant abnormalities were noticed on general examination, except for a moderately swollen abdomen. An abdominal radiograph (left lateral) did not reveal any abnormalities. Blood was sampled for a CBC and serum biochemistry profile. The results of CBC and biochemistry can be found in Table 1. There was a marked thrombocytopenia (74.000/µl (ref. value 200.000 – 400.000/µl)). The leukocyte count was within normal levels, but showed a marked basophilia in the leukocyte differential count. In the present case, these cells were identified as mast cells, based on the cytological findings of the peripheral blood smear. Mast cells differ from basophils in that the mast cell nucleus is round and often covered by densely stained, purplish granules that also fill the cytoplasm (Jain, 1993), while basophils have a polymorphic nucleus. The results of the biochemistry profile showed a moderate increase in serum alkaline phosphatase concentration (268 U/l – 10-50 U/l), serum bile acid concentration (39 µmol/l - <20) and serum gamma-globuline concentration (13 g/l – 3-8).

A peripheral blood smear (Modified Wright stain, Sigma Aldrich) showed a marked increase of mast cells (i.e. 28%) (Figure 1), characterized as round cells, with a round to oval nucleus and a variable amount of cytoplasm filled with metachromatic granules. The granules partially obscured the nuclear morphology. There was also moderate anisocytosis and an overall increase of cell size.

One week later, the dog was euthanized because its condition had deteriorated rapidly and the presumptive diagnosis of mast cell leukemia has a very bad prognosis.

On autopsy, a severely enlarged, pale and thickened spleen, a normal sized and colored liver with multiple pale nodules and normal appearing (reddish) bone marrow were noted. No abnormalities were observed in the gastrointestinal tract, lymph nodes or skin. The other organ systems also appeared normal. Samples of the liver, spleen, and bone marrow were collected for histopathologic examination.

The result of bone marrow cytology/histology (Mo-

Table 1. Results of hematologic and biochemistry examination.

HEMATOLOGY				
ERYTHROCYTES	5.66	10^{12}/L	5.5 - 7.5	
LEUCOCYTES	11.6	10^{9}/L	6.0 - 12.0	
LEUKOC. FORMULA				
Segments	65.0	%	50.0 - 63.0	
Eosinophils	0.0	%	0.0 - 3.0	
Basophils	28.0	%	0.0 - 1.0	+
Lymphocytes	14.5	%	13 - 30	
Monocytes	2.0	%	3.0 - 7.0	
Segments abs.	7.6	10^{9}/L	3.2 - 11.8	-
Eosinophils abs.	0	10^{9}/L	0.05 - 1.18	+
Basophils abs.	3.27	10^{9}/L	0 - 0.1	
Lymphocytes abs.	1.7	10^{9}/L	1.0 - 4.8	
Monocytes abs.	0.23	10^{9}/L	0.05 - 1.24	
HEMOGLOBIN	8.2	mmol/L	8.5 - 12.0	
HEMATOCRIT	443.0	ml/L	420 - 540	
MCV	78	fl	67 - 80	
MCH	14	fmol	13.0 - 16.0	
MCHC	18	mmol/L	20.0 - 23.0	-
BIOCHEMISTRY				
FRUCTOSAMIN	176	umol/L	< 372	
TOTAL PROTEIN	67.0	g/L	58.0 - 75.0	
PR.ELECTROPH. alb	44.5	%	43 - 54	
alfa 1	7.4	%	4 - 8	
alfa 2	16.3	%	12 - 22	
beta	12.4	%	15 - 23	-
gamma	19.4	%	6 - 13	+
albumine abs.	29.8	g/L	28 - 38	
alfa 1 abs.	5.0	g/L	3 - 6	
alfa 2 abs.	10.9	g/L	6 - 14	
beta abs.	8.3	g/L	10 - 14	-
gamma abs.	13.0	g/L	3 - 8	+
UREA	4.3	mmol/L	2.00 - 6.70	
CREATININ	65.41	umol/L	50+1/kg bodyw.	
UREA/CREATININ	65.98			
GPT (ALT)	40	U/L	25 - 55	
Y GT	5	U/L	2 - 10	
ALK. FOSF.	268	U/L	10 - 50	+
BILE ACIDS	39	umol/L	< 20	+

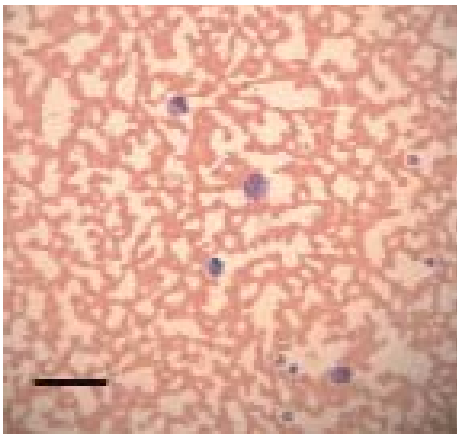

Figure 1. Microphotograph of the blood smear. Three mast cells are present in the peripheral blood (arrow). Modified Wright stain – Bar = 50 µm.

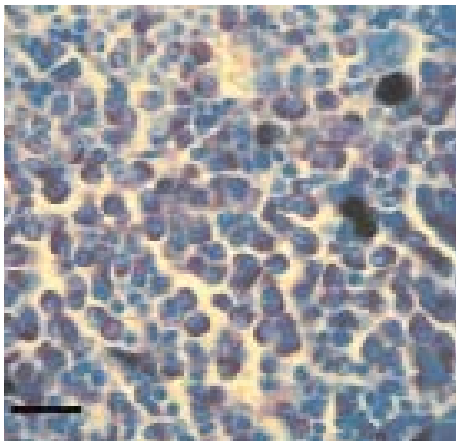

Figure 2. Microphotograph of the bone marrow. Note the metachromatic staining granules in the cytoplasm of the mast cells, diffusely infiltrated between the normal hematopoietic cells (without metachromatic staining cytoplasm). Giemsa stain – Bar = 25 µm.

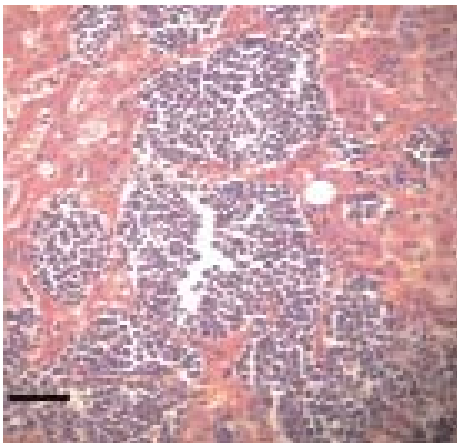

Figure 3. Microphotograph of the liver showing a severe, diffusely infiltrative growth of mast cells. HE staining – Bar = 50 µm.

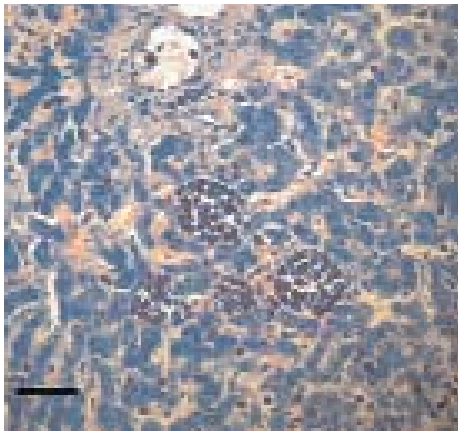

Figure 4. Microphotograph of the liver. Note the metachromatic staining of the cytoplasm of the infiltrating mast cells. Giemsa stain – Bar = 50 µm.

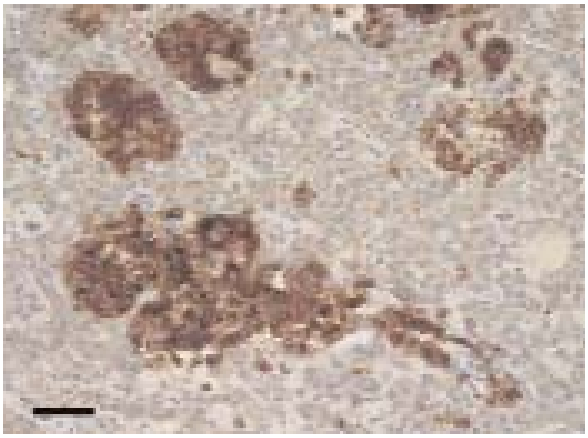

Figure 5. Microphotograph of the liver. Immunohistochemical staining for c-kit. Note the brown cytoplasmic staining of the infiltrating mast cells. Bar = 50 µm.

dified Wright stain, Hematoxylin-Eosin, Giemsa) showed hypercellularity and a diffuse infiltration with abundant variably sized mast cells characterized by abundant cytoplasm with metachromatic staining granules, which obscure the round nucleus, anisocytosis and anisokaryosis, and occasionally a mitotic figure. Some areas consisted completely of these mast cells, while other areas still showed small remnants of normal hematopoiesis with a diffuse infiltration of mast cells. This was clearly demonstrated with a Giemsa stain (Figure 2). Megakaryocytes were almost absent, which explained the thrombocytopenia.

Histopathologic examination (Hematoxylin-Eosin & Giemsa) of the liver also revealed a marked ad random infiltrative proliferation of round cells with an eccentric nucleus and a large amount of dense eosinophilic cytoplasm (Figure 3). The spleen showed a complete loss of normal architecture and a massive diffuse proliferation of the same cells described in the liver. The cytoplasm of these cells stained metachromatically or showed metachromatic granules with Giemsa staining (Figure 4).

A strong positive c-kit (CD117) immunoreactivity was observed in the mast cells of the epithelial and hematopoietic tissues. The infiltrating mast cells in the liver (Figure 5), spleen and bone marrow showed a strong brownish cytoplasmic coloration.

The findings in the present case are compatible with a diagnosis of mast cell leukemia.

DISCUSSION

Mast cells are not normally present in the peripheral blood of healthy dogs and cats. The causes of mastocytemia include: primary mast cell tumors involving the skin, the spleen or the intestinal tract, disseminated mast cell neoplasia (systemic mastocytosis involving the spleen, liver, lymph nodes and bone marrow), mast cell leukemia and severe inflammatory/allergic disorders (Latimer and Twedten, 1999). Mastocytemia in dogs has also been described in enteritis (parvovirus), fibrinous pericarditis and pleuritis, bacterial peritonitis, aspiration pneumonia, acute pancreatic necrosis, immune mediated hemolytic ane-

mias, renal failure associated with acute inflammation, inflammatory skin disease, and hemorrhage secondary to hemophilia and gastric torsion (Scott and Stockham, 2000).

Mast cell leukemia originates in the bone marrow and the diagnosis is based upon a moderate to marked leukocytosis involving a significant proportion of mast cells (3-74%) within the complete blood cell count, as well as a diffuse bone marrow infiltration with these cells. In humans, if more than 10% of the cells in the blood are mast cells, then the diagnosis of mast cell leukemia is indicated (Torrey *et al.*, 1990). The present case did not demonstrate leucocytosis, but it did reveal a marked presence of mast cells (28%) in the peripheral blood. The bone marrow was also severely and diffusely infiltrated with variably sized mast cells, which suggests the origin of the tumor in the present case. The term leukemia really indicates that the tumor begins in the bone marrow.

Most dogs with systemic mast cell disease present with lethargy, anorexia, vomiting and weight loss, in association with splenomegaly, hepatomegaly, pallor and, occasionally, detectable cutaneous masses. The complete blood counts in affected dogs commonly reveal cytopenias, with or without circulating mast cells (Couto, 1992). Here, splenomegaly, hepatomegaly and thrombocytopenia and circulating mast cells in the peripheral blood were noted. The moderate increase of serum alkaline phosphatase concentrations and serum bile acids can be explained due to the infiltrative growth of the neoplastic mast cells in the liver, causing obstruction of the bile ducts and reduced excretion of bile. Hypergammaglobulinemia is often observed with neoplasia. Cats with hemolymphatic involvement are classified as having systemic mast cell disease (or mast cell leukemia), since bone marrow, spleen, liver and blood are commonly involved. Most cats present with nonspecific signs, such as anorexia and vomiting; however, abdominal distension due to splenomegaly is a consistent feature. As in dogs, hematologic abnormalities in cats with systemic mast cell disease include cytopenias and the presence of circulating mast cells or basophilia. However, a high percentage of cats may have normal complete blood counts (Couto, 1992).

CD117 or c-kit is a receptor tyrosine kinase thought to play a key role in human and canine mast cell neoplasms. Normal (membrane-associated) and aberrant (cytoplasmic, focal or diffuse) CD117 immuno-expression patterns have been identified in canine mast cell tumors. Cytoplasmic CD117 expression has been found to correlate with higher histological grade and with a worse post-surgical prognosis (Gil da Costa *et al.*, 2007).

Once a patient develops metastatic or disseminated mast cell tumors, a cure is rarely obtained. Treatment in these cases is aimed at palliating the neoplasm and its complications by using chemotherapy and supportive therapy (Couto, 1992). However, there are still many questions about how to best manage mast cell tumors (Dobson and Scase, 2007).

ACKNOWLEDGEMENTS

We would like to thank the staff of Medisch Labo Bruyland and Labo Pathologie Yperman for their assistance. Dr. Y. Temmerman (Marke) is acknowledged for his cooperation.

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