

# What is your diagnosis? Abnormal cells on a blood smear from a dog

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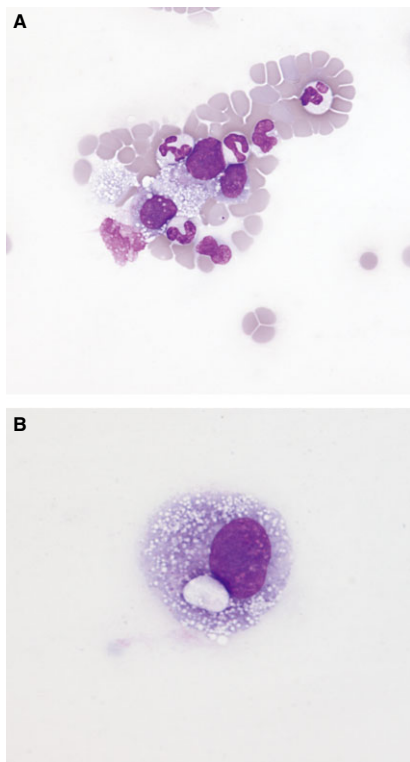
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## Case Presentation

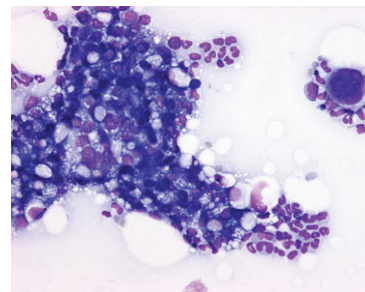
A 7-year-old intact female Australian Shepherd dog was referred to the Veterinary School of Toulouse for acute weakness, dyspnea, and marked thrombocytopenia. On presentation, the dog was laterally recumbent, depressed, and

hypothermic. Physical examination showed numerous petechiae, scleral hemorrhage, and a small mammary mass (7 mm diameter) at the fifth left mammary gland.

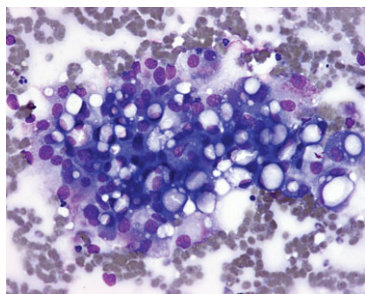
A CBC revealed marked thrombocytopenia ( $37 \times 10^9/L$ ; reference interval [RI]  $108\text{--}562 \times 10^9/L$ ), mild leukocytosis ( $26.7 \times 10^9/L$ ; RI  $5.6\text{--}20.4 \times 10^9/L$ ), moderate neutrophilia ( $22.9 \times 10^9/L$ ; RI  $2.9\text{--}13.6 \times 10^9/L$ ), and a low normal hematocrit (39%; RI 35–52%) accompanied by mild reticulocytosis ( $180.6 \times 10^9/L$ ; RI  $19.4\text{--}150 \times 10^9/L$ ). A biochemistry profile showed mild hypercalcemia (ionized calcium 1.54 mmol/L; RI 1.2–1.5 mmol/L). A coagulation profile revealed prolonged PT ( $> 120$  seconds; RI 7.1–9 seconds) and APTT ( $> 180$  seconds; RI 12.8–17.2 seconds), non-detectable fibrinogen concentration, and increased fibrin(ogen) degradation products ( $> 20$  mg/L; RI  $< 5$  mg/L). The petechiae, thrombocytopenia and the changes in the coagulation profile were highly suggestive of disseminated intravascular coagulation (DIC). Thoracic X-rays were unremarkable. An abdominal ultrasound revealed changes in the structure and echogenicity of the left iliac lymph node suggestive of a metastatic neoplastic process; there was no evidence of other masses in the abdominal and thoracic cavities. A blood (Figure 1) and a bone marrow smear (Figure 2) were evaluated.



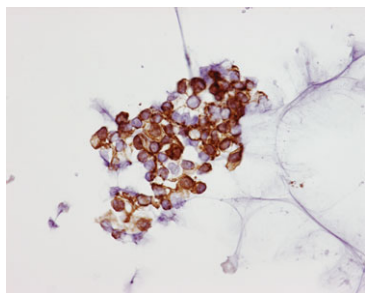
**Figure 1.** Blood smear from an Australian Shepherd with a small mammary mass and disseminated intravascular coagulation. May–Grunwald–Giemsa, (A)  $\times 40$  objective oil immersion, (B)  $\times 100$  objective oil immersion.



**Figure 2.** Bone marrow smear from an Australian Shepherd with a small mammary mass and disseminated intravascular coagulation. May–Grunwald–Giemsa,  $\times 100$  objective oil immersion.



**Figure 3.** Cytology of a fine-needle aspirate of a small mammary mass in an Australian Shepherd with disseminated intravascular coagulation. Note the papillary cluster of cells with vacuolated cytoplasm suggestive of epithelial cells. May–Grünwald–Giemsa,  $\times 40$  objective oil immersion.



**Figure 4.** Immunohistochemistry for Cytokeratin on bone marrow smear from an Australian Shepherd with disseminated intravascular coagulation and a small mammary mass using mouse monoclonal anti-human cytokeratin clone MNF116 (detection of cytokeratins 5, 6, 8, 17, and probably 19) from Dako, France. Diaminobenzidin chromogen.  $\times 100$  objective oil immersion.

**Diagnosis:** Circulating and metastatic carcinomatous cells in peripheral blood and bone marrow, respectively.

### Interpretation

Cytologic evaluation of a FNA of the mammary mass (Figure 3) revealed a highly cellular sample with large amount of necrotic debris in a hemorrhagic inflammatory background. A population of cohesive cells organized in papillary clusters, with moderate-to-high N:C ratio, basophilic granular cytoplasm containing achromatic, round and sharply contrasting globules, and with eccentric nuclei containing variably sized nucleoli, were highly suggestive of epithelial cells, characterized by lipid accumulation. Marked anisocytosis and anisokaryosis were observed. A similar population was observed in bone marrow smears. An immunohistochemical stain of a bone marrow smear was positive for cytokeratin (Figure 4). After euthanasia, necropsy revealed multiple subcutaneous, vesical, renal, and subarachnoid hemorrhagic lesions, and a neoplasm of the left inguinal mammary gland. Metastases were found in the left iliac lymph node, mediastinum, lungs, spleen and kidneys. Histopathology confirmed a poorly differentiated lipid-rich mammary carcinoma (MC) with nodal, hepatic, splenic, pulmonary, medullary and renal metastases.

### Discussion

MC with DIC, bone marrow involvement, and circulating tumor cells (CTCs) has been described previously.<sup>1</sup> The characteristics were similar to the present one, except that hypercalcemia was not reported.

In canine MC, as in human breast cancer, CTCs are rare. The presence of CTCs has prognostic relevance and their detection has been facilitated with the use of reverse transcription polymerase chain reaction. In 2 studies, a set of potential markers for canine mammary gland CTCs was identified.<sup>2,3</sup> Crystallin alpha B (CRYAB) was the most sensitive (35%) and specific (100%) single marker to detect CTCs in metastatic MC.<sup>4</sup> In the case presented here, CRYAB was not performed because atypical cells were observed on the blood smear and their epithelial origin was confirmed by immunocytochemistry on bone marrow smears.

Disseminated intravascular coagulation has been associated with solid tumors, including MC.<sup>1</sup> The magnitude of the coagulation changes is related to the cancer stage and the presence of distant metastasis.<sup>5</sup> Although the exact mechanism is still unknown, the presence of inflammatory cytokines and the production of pro-coagulant and heparin-like molecules by the tumor are probably contributing factors. In the case presented here, neoplasia was the most likely cause for the DIC.

Hypercalcemia is frequently associated with lymphomas and carcinomas in dogs. In the case presented here, paraneoplastic hypercalcemia seemed most likely; however, other causes of hypercalcemia were not investigated.

**Key Words:** Bone marrow, circulating tumor cells, dog, hypercalcemia, mammary tumor.

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