Hemangiosarcoma (HSA) is an aggressive, malignant tumor that arises from either the vascular endothelium or endothelial precursor cells [1]. The origin of tumour is frequently in the spleen (28–50%), right atrium and auricle (3–50%) and subcutaneous tissues (13%) [3-6]. HAS can also primarily occur in liver, kidney, bladder, prostate, peritoneum, lung, pulmonary artery, aorta, muscle, bone, oral cavity, tongue, vertebral body, and central nervous system [6,7].

Metastasis occurs early in the disease, via hematogenous routes, because of the access to the systemic circulation, but also through transabdominal implantation following rupture. Around 80% of the dogs can have metastasis by the time of diagnosis. Up to 63% of dogs with right atrial HSA have concurrent metastatic disease [8-10]. The overall prevalence of HSA is reported to be 0.3–2.0% of all tumors in dogs, and represent about 5-7% of all malignancies in dogs [5,7,11].

Ware and Hopper emphasize that HSA is the most common cardiac tumor identified in dogs, with an overall incidence of 0.19%. HSA represent 69% of all cardiac tumors frequently in the spleen (28–50%) and pericardial cavity (hemopericardium) and also the rupture of the right atrium. The spleen was sampled for histopathological examination. Hemangiosarcoma does not cause pain and there are no clinical signs of the disease 

A post-mortem examination of an 8 years old German Shepherd dog was performed in order to find out the death cause. The macroscopic examination revealed a large amount of blood in the abdominal (hemoperitoneum) and pericardial cavity (hemopericardium) and also the rupture of the right atrium. The spleen was sampled for histopathological examination. Hemangiosarcoma does not cause pain and there are no clinical signs of the disease that allow the blood to escape into the abdomen, heart, and splenic veins, the blood to leak into the pericardial cavity, and the blood to enter the systemic circulation, but also through transabdominal implantation following rupture. Around 80% of the dogs have metastasis by the time of diagnosis. Up to 63% of dogs with right atrial HSA have concurrent metastatic disease [8-10]. The overall prevalence of HSA is reported to be 0.3–2.0% of all tumors in dogs, and represent about 5-7% of all malignancies in dogs [5,7,11].

Results and discussions

The gross examination of the German Shepherd dog cadaver revealed no pathological signs. Following necropsy, a large amount of blood was observed in the abdominal cavity (hemoperitoneum) (fig. 1). Figure 1. Blood collection in the abdominal cavity (hemoperitoneum).

The pericardial cavity opening emphasizes local blood accumulation (hemopericardium) (fig. 2). On heart gross examination it was observed a rupture in the right atrium and the heart sectioning revealed a dark-red homogenous tissue.

**Experimental part**

**Materials and methods**

It was performed an 8 years old German Shepherd dog post-mortem examination, following a sudden death. There were taken spleen samples for histopathological examination.

The samples preparation was carried out as follows: 24 h alcohol fixation at room temperature (prevent the tissue alteration due to the enzymes activity; preserve the tissue texture; improves the optical differentiation), alcohol dehydration (five steps: 70, 80, 90, 100% and 100% alcohol, each step for two hours), clearing with benzene, paraffin wax at 56°C, embedding tissues into paraffin blocks, trimming of paraffin blocks (6 μm), sections mounting on the glass slides (using Meyer albumin), hematoxylin-eosin-methylene blue staining [16,17].

Staining was performed as follows: deparaffination of tissue sections in benzene, rehydration using decreasing concentrations of alcohol, rinsing in distilled water, hematoxylin staining, alcohol eosin staining and methylene blue staining, water removal using increasing concentrations of alcohol, cover slide mounting [17]. Hematoxylin will stain the nuclei in blue and the mucus in light blue. Eosin will stain the cytoplasm in pink, collagen in pale pink, red blood cells in bright red, and colloid in red. Methylene blue improves the blue colour of the nuclei, making them more observable [17].

The microscopical examination is useful as differentiating diagnosis method only if chemical preparation of samples is applied [18].

**Key words:** hemangiosarcoma, German Shepherd dog, hematoxylin-eosin-methylene blue staining.
It was assumed that the death occurred following a super acute hemorrhage.

The spleen gross examination revealed the presence of two round formations, nonencapsulated, of different sizes, localized on the spleen head; respectively tail (fig. 3). Following the spleen sectioning, a homogeneous dark red tissue was observed (fig. 4).

The complete examination of the cadaver does not show other pathological lesions.

Based on the gross examination findings the presumptive diagnosis was HSA. The cadaver age (8 years) fits in the normal range specific for HSA. The scientific data reveal the adult age (8-13 years) as being specific for HSA occurrence [4,7,9,11,13].

The diagnosis is also supported by the unicity of the case during the last 5 years of necropsies. The specific literature emphasize that the frequency of HSA is generally rare 0.3–2.0% [5] or 5-7% [11] of all tumors seen in dogs.

Applying the “double two thirds rule” according to which approximately 2/3 of dogs with splenic masses will have malignant tumors and approximately 2/3 of those malignant tumors will be diagnosed as HAS (43%) [19,20], and taking into account that 63 to 70% of dogs presenting splenic masses with nontraumatic hemobdenomen were HAS [21] we assumed that the found out formation was HSA.

The concomitant occurrence of the lesions in the spleen and right heart in 25% of HAS cases [5,7,10] supports our presumption, even if some authors suggest that this percentage at initial presentation is significantly lower (5% or less) [22].

Also, on notice that the cardiac or pericardial tumors are responsible for approximately 60% of cases of pericardial effusion in dogs [23].

Specific literature data emphasize that several splenic mass lesions (e.g., HSA, hematoma, hemangioma) can have a similar gross and ultrasonographic appearance, and large masses do not necessarily denote malignancy [24]. According to the mentioned affirmation, we consider to perform the histopathological examination of spleen samples for a definitive diagnosis.

Following the hematoxylin-eosin-methylene blue staining, the microscopically examination of the spleen samples emphasizes immature, endothelial cells forming vascular spaces containing variable amounts of blood and/or thrombi (fig. 5), according to the specific literature data [25].

We know that if histopathological features are minimal but HSA is suspected, immunohistochemistry for von Willebrand’s factor (factor VIII–related antigen) or CD31/platelet endothelial cell-adhesion molecule (PECAM) can be used to demonstrate endothelial derivation and support the diagnosis of HAS [25,26,27], but our findings were sufficient to confirm the diagnosis.

We could not specify which was the primary site of tumor occurrence, the right atrium or the spleen, but according scientific papers, the origin of the tumors is equal noticed (50 %) [3,5,6].

![Fig. 1. Blood collection in the abdominal cavity (hemoperitoneum)](image1)

![Fig. 2. Heart - rupture of the right atrium](image2)

![Fig. 3. Spleen - round reddish-black formations.](image3)

![Fig. 4. Spleen - hemorrhagic aspect.](image4)

![Fig. 5. Spleen - abnormal blood vessels and blood-filled channels and spaces. Hematoxylin-eosin-methylene blue staining, 20x](image5)

**Conclusions**

The German Shepherd dog post mortem gross examination revealed hemoperitoneum, hemopericardium and right atrium rupture that, based on the specific literature data, lead to the presumption of hemangiosarcoma.

The confirmation of hemangiosarcoma diagnosis was made taking into account the histopathological examination of the spleen samples, following hematoxylin-eosin-methylene blue staining. The presence of the specific histopathological aspects (blood-filled channels or spaces) confirmed the HSA diagnosis, without demanding immunohistochemistry tests.

The rare founded pathological aspects (once in 5 years), show that HSA has quite low frequency in dogs.

**References**

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