FINDINGS IN CEPHALIC CLINICAL MANIFESTATIONS CAUSED BY SECONDARY ORGANOPATHIES RESULTING FROM CANINE BABESIOSIS

A. SALLAY*, B. BŢCESCU*, N. BERCARU*, R. CONDRUŢ**

Abstract

This paper aimed at describing the cephalic clinical manifestations brought about by the secondary organopathies resulting from babesiosis in 42 dogs that were examined and studied in the Medical Pathology Clinic of the Faculty of Veterinary Medicine, Spiru Haret University and the Tiovet Centre for Diagnosis and Treatment in Bucharest. The dogs that were diagnosed with babesiosis had an indifferent facies, photophobia with xerophthalmia and slower palpebral reflexes, the conjunctival and gingival mucous membrane being quite pale in 27 dogs (64.28 %), sub-icteric in 10 dogs (23.8 %) and icteric in 15 dogs (11.9 %). We noticed the thickened tongue with the molar tooth print in 23 dogs (54.76 %), with white-greyish accumulations of matter in 8 cases (19.04 %) and xerostomia in 19 subjects (45.23 %). The kidney and liver functional changes were biochemically confirmed by determining uraemia (between 40 and 50 mg/dl in 15 cases, between 51 and 100 mg/dl in 24 cases, over 100 mg/dl in 3 cases) and the alanine aminotransferase (between 60 and 85 UI/l in 2 cases, between 90 and 100 UI/l in 28 cases, over 110 mg/dl in 12 cases). The main urine changes referred to haemoglobinuria (between 0.2 and 1 mg/dl in 24 cases, over 1 mg/dl in 18 cases). The biochemical establishment of babesiosis was based on the determination of the haemoglobinaemia (under 5 mg/dl in 3 cases, between 5 and 6 mg/dl in 24 cases, between 6 and 7 mg/dl in 5 cases; reference values: 8-12 g/dl). The disease was confirmed by revealing the pear-shaped sporozoites in the red cells. Peripheral blood smears were done by using the May-Grünwald-Giemsa staining method. The disease was confirmed by the presence of Babesia canis.

Key words: *babesiosis, ammonia halitosis, anaemia, jaundice, xerostomia*

Introduction

Babesiosis was found all over the world and in 1984 Prof. Nicolae Dulceanu diagnosed it in Romania too. The spread of the disease depends on the presence of biotopes with ticks. The number of ailments reaches a peak in the interval April-October, but, as an exception, cases were also registered in the cold season. The disease appears in common dogs and improved breed dogs as well, with the secondary organopathies caused by paraclinical changes subsequent to the infestation with *Babesia canis* being amplified by immunosuppressive factors or intercurrent diseases of the infested animals [4].

After contaminating the red blood cell, *Babesia canis* sporozoites trigger the process of erythrocytholisis responsible for the appearance of haemolytic anaemia; haemoglobinuria, proteinuria, cylindruria and jaundice are the first signs of the animal falling ill [6]. The body reacts by rising temperature (39.8º-40.5ºC) due to the lipopolysaccharides released from the membranes of the sporozoites that affect the hypothalamic nervous centres. Higher body temperature is accompanied by cardio-circulatory, digestive and excretory changes, as well as by metabolic imbalances [5, 10]. The main degenerative histological changes were found in the kidneys, with degenerescence of the proximal contorted tubes and necrotic processes accompanied by the detachment of renal epithelial cells from the basement membrane. Acute tubular necrosis is probably the result of the hypoxic renal lesion caused by haemolytic anaemia and systemic hypotension. These ultrastructural renal features could explain the increase in urea and serum creatinine [8, 11].

* Faculty of Veterinary Medicine, *Spiru Haret* University.
** Tiovet.

Kidney malfunction, frequently encountered in infections with *Babesia canis*, must be identified in due time and diagnosed correctly for the adequate prophylactic and therapeutic measures to be taken.

Materials and methods

Personal research consisted of the clinical and paraclinical study on case records of the Medical Pathology Clinic of the *Spiru Haret* Faculty of Veterinary Medicine and of the Tiovet Diagnosis and Treatment Center.

Between May 2007 and February 2008, we examined and studied 42 dogs of various breeds, age between 2 and 14: 32 subjects were studied in the clinic (18 male dogs and 14 female dogs), whereas the rest of 10 (6 males and 4 females) were examined at the Tiovet Diagnosis and Treatment Center.

Using the examination methods described hereinafter, the cephalic and systemic clinical manifestations were observed, analyzed and monitored. Both the main examination methods (inspection, palpation, percussion, auscultation and thermometry) and complementary methods (blood smear staining, biochemical testing of blood and urine) were applied.

Blood samples were taken from each dog in several stages, which were used to prepare the smear and for determining and interpreting the values of the following biochemical blood parameters: ureic nitrogen, creatinine, haemoglobine, alanin aminotransferase, aspartate aminotransferase and total bilirubin. The Revlovet Plus device was used for the biochemical blood testing, and the smears were prepared by using the May-Grünwald-Giemsa method.

Urine analysis consisted of the physical (quantity, colour, smell, specific weight, and viscosity), chemical (albuminuria, haemoglobinuria, glycosuria, ketonuria, bilirubinuria, nitrite content, urobilinogen and pH) and microscopic examination (examination of the urine sediment). The physical examination was both visual and olfactory, whereas the chemical test was performed with the Pocket Chem Arkay UA-PU 4210 automatic analyzer for the determination of urinary concentrations.

Results and discussion

The dogs were diagnosed between May 2007 and February 2008, in 29 cases (69.04%), the infection with *Babesia canis* being caused by failure to perform external prophylactic disinfesting of the dogs and by massive tick infestation following strolls on the Bucharest outskirts in 13 cases (30.96%).

The onset of the symptomatology was sudden, and the rapid aggravation of the dog's condition required in most cases a medical consultation in 48 hours since the owner's noticing the onset of the status of prostration.

Of the 42 subjects, 38 (90.47%) presented at the first clinical examination hyperthermia (39.9º-40.2ºC), the rest of 4 subjects (9.52%) having subfebrile temperature (39.6º-39.8ºC). Body temperature was monitored all through the period of investigation, with the day-time oscillations of body temperature presenting the specific see-saw pattern.

The anamnetic investigation and clinical examination revealed that all the subjects, irrespective of their corporal thermal condition, were in a state of prostration, with lack of appetite, indifferent expression, exaggerated calm, photophobia with xerophthalmia and slower palpebral reflexes; pallor of conjunctival and gum

mucous membranes was found in 27 dogs (64.28%), sub-icteric colour was found in 10 dogs (23.80%) and icteric colour in 15 dogs (11.90%).

The olfactory examination of halitosis in 34 subjects (80.95%) revealed the ammonia smell specific to uraemic intoxication. Accompanying cephalic manifestations were noticed in 29 cases (69.04%), the ammonia halitosis being perceived both in icteric dogs and in those with pale mucous membranes.

Thickened tongue with molar tooth print was noticeable in 23 dogs (54.76%), with white-greyish coating found in 8 cases (19.04%); xerostomia was found in 19 subjects (45.23%). The dry, chapped and discoloured nose was noticed in 12 subjects (28.57%).

Previous to performing the biochemical blood test, urine samples were collected, as the results of the chemical analysis of urine give clues to the configuration of the blood profile. Urine was collected by urethral catheterism, and only in few cases by spontaneous miction; the second is a disadvantageous variant because of urethral, vaginal or prepuce secretions potentially contaminating the urine. The values obtained following the biochemical urinary analysis were monitored in table 1.

Urine with macroscopic turbidity was centrifuged (500 rot./min.) and the sediment thus obtained was examined under the microscope. The microscopic examination tracked and established the structure of the organized sediment represented by epithelial cells, leukocytes, red cells and urinary cylinders in 29 subjects (69.04%). Unorganized sediment was found only in 4 dogs, consisting of calcium oxalates in 3 cases (7.14%) and ammonia-magnesian phosphate in one case (2.38%).

Table 1

Values of the parameters investigated by urine biochemistry

Investigated parameters	**Reference values**	**Dogs with icteric mucous membranes**	**Dogs with pale mucous membranes**
Glycosuria	negative	negative in 15 dogs	negative in 27 dogs
Albuminuria	≤ 30 mg/dl	negative in 8 dogs above 100 mg/dl in 7 dogs	negative la 5 dogs below 100 mg/dl in 22 dogs
Urobilinogen	negative	between 2 and 4 mg/dl in 5 dogs; above 4 mg/dl in 10 dogs	negative in 27 dogs
Bilirubinuria	negative	between 1 and 3 mg/dl in 5 dogs; above mg/dl la 10 dogs	negative in 20 dogs; below 1 mg/dl in 7 dogs
Haemoglobinuria	negative	below 1 mg/dl in 3 dogs; above 1 mg/dl in 12 dogs	below 1 mg/dl in 4 dogs; above 1 mg/dl in 23 dogs
Ketonuria	negative	negative in 15 dogs	negative in dogs
Density	≈ 1.030	normal limits in 15 dogs	normal limits in 27 dogs
pH	5,5 – 6,5	normal limits in 15 dogs	normal limits in 27 dogs
Nitrites	negative	negative in 15 dogs	negative in 27 dogs
Leukocyturia	≤ 75 leu/µl	75 leu/µl in 9 dogs	75 leu/µl in 22 dogs

The results obtained led to the conclusion that the main urinary biochemical changes involve haemoglobinuria (42 cases – 100%), albuminuria (29 cases – 69.04%), bilirubinuria (22 cases – 52.38%) and urobilinogen (15 cases – 35.71%).

The biochemical analysis of the blood was used to confirm the diagnosis. In the first stage, the haemoglobine, ureic nitrogen and alanin aminotransferase (ALT) blood content was determined. The parameters were selected according to the

clinical manifestations, colour changes perceived in cephalic membranes, the presence of ammonia halitosis and urinary biochemical results; the values obtained are presented in tables 2, 3 and 4.

The values obtained confirmed for all the subjects the condition of anaemia, retention of ureic nitrogen, and enzymatic hepatic changes. Creatinine, aspartate aminotransferase (AST GOT) and total bilirubin were determined only in some of the dogs, the values obtained confirming the presence of kidney and hepatic failure.

Table 2

Synopsis of ureic nitrogen values in examined dogs

Parameter	**Reference value**	**Between 30 and 50 mg/dl**	**Between 51 and 100 mg/dl**	**› 100 mg/dl**
Ureic nitrogen	8-26 mg/dl	15 cases (35.71%)	24 cases (57.14%)	3 cases (7.14%)

Table 3

Synopsis of values of alanin aminotransferase (ALT) in examined dogs

Parameter	**Reference value**	**Between 50 and 60 mg/dl**	**Between 60 and 70 mg/dl**	**› 70 mg/dl**
ALT GPT	8.9-48.5 mg/dl	12 cases (28.57%)	28 cases (66.66%)	2 cases (4.76%)

Table 4

Synopsis of haemoglobine values in examined dogs

Parameter	**Reference value**	**Between 6 and 7 g/dl**	**Between 5 and 6 g/dl**	**Below 5 g/dl**
Haemoglobine	8-12 g/dl	5 cases (11.90%)	34 cases (80.95%)	3 cases (7.14%)

The correlation between renal necrosis, the cause of deficient blood filtration and systemic hypotension caused by anaemia, could be assessed and inferred to a certain extent from the study of the values obtained in the determination of ureic nitrogen and haemoglobinemia [1, 2, 3, 7, 12]. In other words, the amount of seric urea is directly proportional to the degree of anaemia. In the present work, three cases confirm this hypothesis (see tables 2 and 4). The hepatic enzyme diagnosis was changed mainly in subjects with haemoglobinemia below 6 g/dl (see tables 3, 4).

The anamnetic data point to a fulminating and galloping evolution of the disease, in absence of a medical record regarding the hepatic-renal pathology in the examined subjects. The fast feverish reaction maintained on the background of a hepatic-renal failure, the sad expression, the state of prostration, anaemia and haemoglobinuria were decisive in the reinterpretation of the diagnosis. To this purpose, blood smears were prepared with a view to confirming the infestation with *Babesia canis*. The examination of the smears highlighted the presence of pear-shaped sporozoites in the red cells (fig. 1).

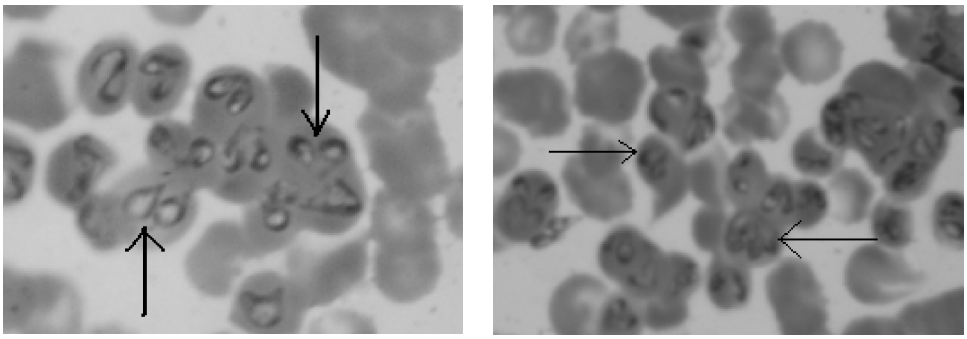

Fig. 1. Pear-shaped sporozoites in dog red cells. MGG staining method, lens immersion.
(Photo: Răzvan Condruț)

Their size was larger than the radius of the red cell and they were placed in twinned arrangements. Based on this data, the cause of the dogs falling ill was identified as infestation with *Babesia canis*, and hepatic-renal failure was considered as complication from babesiosis [9, 12].

Conclusions

1. The feverish reaction maintained against the background of organ functioning failure, the sad expression, the state of prostration, anaemia and haemoglobinuria are all symptoms that lead to the suspicion of an infection with *Babesia canis*.

2. Confirmation of babesiosis is rendered by tracking the pear-shaped sporozoites inside the red cells, on the peripheral blood smears (May-Grünwald-Giemsa staining method, examination by lens immersion).

3. The cephalic manifestations were mainly determined by erythrocytholisis (pallor of cephalic mucous membranes, icterus, changes of the nose) and retention of ureic nitrogen (ammonia halitosis, coated tongue, photophobia, xerophthalmia, xerostomia, slower palpebral reflexes). These are not manifestations specific to babesiosis, but their presence can lead to the suspicion of the disease.

4. The main blood and urinary biochemical determinations whenever a *Babesia canis* infection is suspected refer to haemoglobinaemia and haemoglobinuria.

5. The correlation between the kidney necrosis and systemic hypotension could be assessed and inferred from the study of the values of ureic nitrogen and haemoglobinaemia determinations, the quantity of serum urea standing in direct proportion to the degree of anaemia.

6. The hepatic enzyme diagnosis was changed especially for the subjects with haemoglobinaemia below 6 g/dl.

Reference

1. Alan, R. K., Laurie, E., Dunbar, A., C. Girard, 1991, *Concurrent Ehrlichiosis and Babesiosis in a Dog*, Can. Vet. J. May, 32 (5), pp. 305-307.

2. Breitschwerdt, E. B., Malone, J. B., MacWilliams, P., 1983, *Babesiosis in the Greyhound,* J., Am., Vet., Med., Assoc., 182 (9), pp. 978-982.

3. Camacho, A. T., Pallas, E., Gestal, J. J., 2002, *Babesia Canis Infection in a Splenectomized Dog,* Bull., Soc., Pathol., Exot., 95 (1), pp. 17-19.

4. Coman, S., Bogdan, B., Bercaru, N., Petruţ, T., 2008, *Boli parazitare şi micoze la animalele de companie,* Fundaţia *România de Mâine* Publishing House, pp. 96-97.

5. Curcă, C. D., 1995, *Fiziopatologie*, Fundaţia *România de Mâine* Publishing House, vol. I, pp. 350-369.

6. Dantas, Torres F., Figueredo, L., 2006, *Canine Babesiosis. A Brazilian Perspective*, Vet., Parasitol., 24 (7), pp. 134-150.

7. Irwin, P. J., Hutchinson, G. W., 1991, *Clinical and Pathological Findings of Babesia Infection in Dogs*, Aust., Vet., J., 68 (6), pp. 204-209.

8. Lobetti, R. G., Reyers, F., Nesbit, J. W., 1996, *The Comparative Role of Haemoglobinaemia and Hypoxia in the Development of Canine Babesial Nephropathy*, J. S. Afr. Vet. Assoc., 67, pp. 188-198.

9. Lobetti, R. G., 2005, *Cardiac Involvement in Canine Babesiosis,* J., S., Afr., Vet., Assoc., 76 (1), pp. 4-8.

10. Máthé, A., Vörös, K., Papp, L., Reiczigel, J., 2006, *Clinical Manifestations of Canine Babesiosis in Hungary (63 cases)*, Acta., Vet., Hung., 54 (3), pp. 367-385.

11. Máthé, A., Dobos, Kovács M., Vörös, K., 2007, *Histological and Ultrastructural Studies of Renal Lesions in Babesia Canis Infected Dogs Treated with Imidocarb,* Acta., Vet., Hung., 55 (4), pp. 511-523.

12. M. L. F. van Velthuysen, S. Florquin, 2000, *Glomerulopathy Associated with Parasitic Infections*, Clin., Microbiol., Rev., 13 (1), pp. 55-66.

13. Nel, M., Lobetti, R. G., Keller, N., 2004, *Prognostic Value of Blood Lactate, Blood Glucose, and Hematocrit in Canine Babesiosis,* J., Vet., Intern., Med., 18 (4), pp. 471-476.

14. Ruiz de Gopegui, R., Peñalba, B., Goicoa, A., 2006, *Clinico-Pathological Findings and Coagulation Disorders in 45 Cases of Canine Babesiosis in Spain,* J., Vet., 42 (8), pp. 234-257.

15. Scally, M. P., Leisewitz, A. L., Lobetti, R. G., 2006, *The Elevated Serum Urea: Creatinine Ratio in Canine Babesiosis in South Africa Is Not of Renal Origin*, J., S., Afr., Vet., Assoc., 77 (4), pp. 175-178.

16. Schetters, T., 2005, *Diagnosis of Babesiosis in the Dog*, Tijdschr., Diergeneeskd., 130 (13), p. 421.

17. Zygner, W., Gójska, O., Rapacka, G., 2006, *Hematological Changes During the Course of Canine Babesiosis Caused by Large Babesia in Domestic Dogs in Warsaw (Poland),* Vet., Parasitol., 34 (7), pp. 234-251.