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Abstracts expanded - oral presentation

CANINE EHRLICHIOSIS: CLINICAL, HEMATOLOGICAL AND SEROLOGICAL INVESTIGATION OF 100 DOGS - 640
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Introduction
The canine ehrlichiosis has emerge as one of the most important infectious diseases that affect dogs (Moreira et al. 2003, Sousa et al. 2004). It is a disease transmitted by the tick Rhipicephalus sanguineus, with worldwide occurrence and prevalence throughout the year and is more common during or after the hot months (Breitschwerdt 2004, Couto 2003). In Brazil, the most common Ehrlichia species are E. canis and E. phlyctis, being the first the one that causes more severe clinical disease (Nelson & Couto 2006). Clinical signs vary according to the duration of infection, immunocompetence of the host and factors related to the microorganism (Couto 2003, Nelson & Couto, 2006). The events occur after an incubation period of 7 to 21 days in three different stages: acute, subclinical and chronic (Couto 2003). The signals observed in the acute phase are in general are nonspecific, including fever, anorexia, depression and weight loss (Breitschwerdt 2004, Mac Candlish 2001). Dogs remain asymptomatic during subclinical phase, although the microorganism persists intracellularly, progressing to chronic phase, featuring among the main signs of mucosal pallor, petechiae, spontaneous bleeding, splenic and hepatomegaly (Breitschwerdt 2004, Mac Candlish 2001, Nelson & Couto 2006). The immune response is maintained pr-
Materials and Methods
We used 100 dogs in different races, sex and ages, suspected to have (or not) canine ehrlichiosis, which were seen at Unime Veterinary Hospital, all living in Lauro de Freitas, Salvador and surrounding towns, in the period of September to December, 2007. Blood sample of the animals were subjected to hemogram, blood smears and serological test for *E. canis* together with the analysis of the history and clinical signs of each animal. Blood samples were obtained by venous collection with the volume at 1.5 to 3 mL of blood carried out by the Veterinary Medical of the HOSVET. The samples were packed into a tube with anticoagulant and sent to the Laboratory of Clinical Pathology to perform the hemogram and blood smear. The technique of indirect ELISA for serological analysis of the dogs was done with the plasma by the commercial kit Hélica ®. Some parameters were analyzed for clinical research, such as: race, sex, age, geographical distribution, history of tick, temperature, body score, appetite, color of mucous, depression, spleno and hepatomegaly, vomiting, nasal or ocular discharge, epistaxis, petechiae, ocular alterations, presence of anemia and other laboratory changes consistent with the disease under study.

Results
The analysis of the results demonstrated the prevalence of 75% of pure-bred dogs in comparison with the mixed breed (25%), being Poodle and Rottweiler the main races of occurrence. In an overall assessment, it was observed that the total sample 55% were female and 45% male, and the age ranged from 1 month to 12 years. According to the clinical parameters, 68% of the dogs had consistent signs of canine ehrlichiosis, and 85% had a history of contact with ticks. The main signs were pale mucosa (31%), depression (29%), hyporexia (22%), anorexia (19%), nasal or ocular discharge (19%) and fever (8%), considered to T° > 39.5 °C. We also observed less frequently vomiting (8%), epistaxis (5%), petechiae (3%), uveitis (3%) and acute blindness (1%). The predominant hematological changes were anemia (47%) and thrombocytopenia (47%), followed by lymphopenia (30%) and neutrophilia (30%). Pancytopenia (13%) was an uncommon finding in this study. In the blood smears were viewed morulas of *E. platys* in 60% of the samples, while in the indirect ELISA 49% of the samples were not reacting to the test and 51% reagents. Among the animals with no reactive samples, 79% demonstrated clinical and / or hematological changes that is frequently identified in the ehrlichia infection. While among the animals that had reagents to test samples, 43% were asymptomatic. Comparing the results obtained using the two techniques, we found that among 51% of the population diagnosed with infection by *E. canis* through the technique of indirect ELISA and 60% with the diagnosis of infection by *E. platys* through the technique of blood smear, 24% of animals studied had a diagnosis of infection by both species. In addition to these results the clinical and hematological findings, it was observed that 12% had simultaneous diagnosis of infection by *E. canis* and *E. platys* associated with clinical and hematological signs.

Discussion
The data from this study demonstrated the variety and nonspecific clinical and hematological signs in the canine ehrlichiosis and their association with the serodiagnostic and blood smear. The paller of the mucous was the predominant clinical sign, similar to the study of Munhoz & Babo (1998) that diagnosed 227 dogs that was attended in the Veterinary Hospital of the University of Mato Grosso do Sul with ehrlichiosis and report this finding as the second most frequent. More pathognomonic signs of this disease were observed in lower frequency, such as epistaxis and splenomegaly, described by Breitschwerdt (2004), according to Nelson & Couto (2006) as characteristic of chronic disease. Epistaxis, despite having been observed in five animals in this study, only two of them had the diagnosis confirmed by the indirect ELISA. The presence of petechiae is a classic sign, however, seems to be a rarely found. The uveitis, a finding also characteristic of chronic disease and observed in three animals in this study, was also present only in one animal with serodiagnostic. Oriá et al. (2004) describe the erlichiosis as a major cause of uveitis, which can occur without a definitive standard. The meeting of anemia and thrombocytopenia as predominant hematological findings corroborated with the study of Moreira et al. (2005), that found 70.3% of anemia in animals, followed by thrombocytopenia in 50%. Mendonça et al. (2005) analyzed the changes in the blood of 109 dogs with a presence of the morula by *Ehrlichia* spp. in leukocytes related thrombocytopenia occurred more frequently (87.15%), followed by anemia (77.98%). The serologic test for indirect ELISA showed that several animals were asymptomatic reagents, reinforcing the idea suggested by Belanger et al. (2002), that does not distinguish serodiagnostic exposure or infection and also the possibility of cross-reactions with other species, suggested by Waner et al. (2001). Moreover, others too, with compatible symptoms were not reacting to the test, possibly because they have not yet suffered seroconversion or because they express the chronic phase of the disease and may not present itself reagent at the serological tests due to the profile of predominant cellular immune response. The diagnosis of simultaneous infection by *E. canis*, through the technique of indirect ELISA, and *E. platys* through the technique of blood smears observed in 24% of animals studied, evidence the suggests by Harrus et al. (1997), that concurrent infections by *E. canis* and *E. platys* are frequent and the infection by *E. canis* may contribute to the pathogenesis of *E. platys*. The meeting of 12 animals with a diagnosis of infection by both species associated with clinical and hematological findings demonstrates once more the nonspecific and insidious character of the disease, reinforcing Munhoz & Babo (1998) idea that is necessary the association of the hemogram, clinical findings, serology and blood smears for the diagnosis of the canine ehrlichiosis, especially in endemic regions to detect asymptomatic animals, since the exclusive serodiagnostic can be sometimes confusing, because of its many variables, such as cross-reactions with other pathogens, time of seroconversion, immunocompetence of the animal and not distinguish between exposure and infection.

Conclusion
The technique of indirect ELISA is an important tool, however is not...
reliable when used as single parameter, because of its many variables. The blood smear technique is fast, easy and reliable in the detection of the *E. platys* morulas. When associated with hematological and/or clinical findings compatible with ehrlichiosis the diagnosis can be conclusive. The results of this study point to the need of using more than one means of diagnosis associated with clinical and hematological parameters for an early detection of this disease. Currently, canine ehrlichiosis manifests itself without a classic pattern, often silent or with mild symptoms, common in many other diseases, passing unnoticed by the owners, confusing the clinicians and leading to late diagnosis, with consequent prognosis reseverd. The growing number of cases, the various clinical manifestations in different stages of the disease and their chronic and insidious nature, reinforce the idea that is essential use the methods of diagnosis to improving the prognosis of our patients

**References**


**Keywords:** Ehrlichiosis, *Ehrlichia canis*, *Ehrlichia platys*, dog, thrombocytopenia