ORIGINAL ARTICLE

https://doi.org/10.21708/avb.2024.18.2.12047

Epidemiology and hematological analysis of dogs naturally infected by *Hepatozoon* spp. from central Brazil - retrospective study

Análise epidemiológica e hematológica de cães naturalmente infectados por Hepatozoon spp. da região central do Brasil – estudo retrospectivo

Ana Julia de Almeida Martins[1], Vinícius Cruz Silva Sousa[2], Raiany Borges Duarte[1], Karla Irigaray Nogueira Borges[3], Dirceu Guilherme de Souza Ramos[1], Ísis Assis Braga[1,3]*

ABSTRACT: Hepatozoonosis is a hemoparasitosis caused by intracellular protozoa of the genus *Hepatozoon*, transmitted to dogs and other carnivores by parasitized ticks, such as *Rhipicephalus sanguineus* and *Amblyomma* spp. Its prevalence varies by geographic region, favoring tropical and subtropical areas. *Hepatozoon* spp. infection cause nonspecific clinical manifestation. This longitudinal retrospective study aims to highlight the epidemiological and hematological aspects of dogs naturally infected with *Hepatozoon* spp. in the municipality of Mineiros, state of Goiás, Brazil, between 2018–2022. This study considered the following information: age, sex, breed, distribution of cases over the years, seasonality, and hematological profile. A total of 272 domestic dogs were found to be parasitized by *Hepatozoon* spp. using the blood smear technique. Adult dogs were more likely to be affected by hepatozoonosis, with no breed or sexual predisposition. This study showed an exponential increase in cases diagnosed over the years, particularly in autumn and winter (the dry season). Within the study's evaluation criteria, most dogs had anemia, thrombocytopenia, and hyperproteinemia; however, there were no changes in the total leukocyte count.

KEYWORDS: coinfection; hemoparasites; hepatozoonosis; leukocytes; tick-borne disease.

RESUMO: A hepatozoonose é uma hemoparasitose causada por protozoários intracelulares do gênero *Hepatozoon*, transmitida a cães e outros carnívoros por carrapatos parasitados, como *Rhipicephalus sanguineus* e *Amblyomma* spp. Sua prevalência varia de acordo com a região geográfica, favorecendo áreas tropicais e subtropicais. A infecção por *Hepatozoon* spp. causa manifestações clínicas inespecíficas. Este estudo retrospectivo longitudinal tem como objetivo destacar os aspectos epidemiológicos e hematológicos de cães naturalmente infectados por *Hepatozoon* spp. no município de Mineiros, estado de Goiás, Brasil, entre 2018-2022. Este estudo considerou as seguintes informações: idade, sexo, raça, distribuição dos casos ao longo dos anos, sazonalidade e perfil hematológico. Um total de 272 cães domésticos foi encontrado parasitados por *Hepatozoon* spp. usando a técnica de esfregaço de sangue. Os cães adultos apresentaram maior probabilidade de serem afetados pela hepatozoonose, sem predisposição sexual ou raça. Esse estudo mostrou um aumento exponencial nos casos diagnosticados ao longo dos anos, especialmente no outono e no inverno (estação seca). Dentro dos critérios de avaliação do estudo, a maioria dos cães apresentou anemia, trombocitopenia e hiperproteinemia; no entanto, não houve alterações na contagem total de leucócitos.

PALAVRAS-CHAVE: coinfecção; hemoparasitas; hepatozoonoses; leucócitos; doença transmitida por carrapato.

INTRODUCTION

Hepatozoonosis is a hemoparasitic disease caused by intracellular protozoa of the genus *Hepatozoon*, with *Hepatozoon americanum* and *Hepatozoon canis* being the predominant species infecting domestic animals (O' Dwyer, 2011). *H. canis* infects neutrophils and monocytes and is transmitted to dogs and other carnivores by parasitized ticks, such as *Rhipicephalus sanguineus* and *Amblyomma* spp., which are considered the main vectors of infection in dogs in South America (O' Dwyer; Massard; Souza, 2001; Forlano *et al.*, 2007; Garrido *et al.*, 2022).

The prevalence of hepatozoonosis varies across geographic regions, with higher rates in tropical and subtropical areas.

[1]Programa de Pós-Graduação em Biociência Animal da Universidade Federal de Jataí, Jataí/GO, Brasil
[2]Médico Veterinário pelo Centro Universitário de Mineiros, Mineiros/GO, Brasil
[3]Centro Universitário de Mineiros, Mineiros/GO, Brasil
*Corresponding author: isis@unifimes.edu.br
Received: 07/25/2023. Accepted: 05/09/2024

Transmission by ticks has been reported from different regions of Asia, Africa, Europe, South America, and the United States (Baneth; Samish; Shkap, 2007; Baneth, 2011; Miranda, 2013). In Brazil, infections by *Hepatozoon* spp. have been described in states in the southeast (Rubini *et al.*, 2005; Spolidorio *et al.*, 2009; Pereira *et al.*, 2011), northeast (Ramos *et al.*, 2010), south (Lasta *et al.*, 2009) and midwest regions (Paludo *et al.*, 2003; Mundim *et al.*, 2008a; Spolidorio *et al.*, 2011).

The pathogenesis of the infection is closely associated with the level of parasitemia, host immune response, and presence of coinfection, culminating in variable and non-specific clinical presentations, along with intermittent clinical signs, including phases of recovery and recurrence (Mundim *et al.*, 2008b; Cicuttina; De Salvo, 2017). Diagnosis is based on the microscopic detection of intracytoplasmic gamonts in blood smear cells (Baneth *et al.*, 2003). Often, the infection is incidentally detected during routine clinical practice. When underdiagnosed and untreated, *Hepatozoon* spp. infection can lead to complications and even fatality (Pereira *et al.*, 2011; Garrido *et al.*, 2022).

Previous studies have primarily focused on the clinical and epidemiological findings related to *Hepatozoon* spp. infection in dogs (Antunes *et al.*, 2015; Oliveira *et al.*, 2021; Peres *et al.*, 2022). Therefore, the objective of this study was to highlight the epidemiological and hematological aspects of a significant sample of dogs naturally infected with *Hepatozoon* spp. from the central region of Brazil across a five-year period from 2018–2022. Given the generic nature of clinical manifestations associated with the plasticity of vectors in regions with diverse climatic characteristics, the results of this study will further our understanding of the epidemiology and hematological aspects of hepatozoonosis.

MATERIAL AND METHODS

This retrospective longitudinal study aimed analyzed the epidemiological and hematological aspects of dogs naturally infected with *Hepatozoon* spp. from the Mineiros-GO municipality. All data related to hemoparasitosis in domestic dogs issued by the Veterinary Clinical Pathology Laboratory of the municipality from 2018 to 2022 were analyzed.

Samples selected for analysis underwent automatic cell counting using the Celltac α hematological analyzer (Nihon Kohden MEK 6500). Subsequently, a differential count and search for hemoparasites were carried out using blood smears stained in panoptic dye and visualized under immersion microscopy.

The results of the hemoparasite research were tabulated based on the positive diagnosis of *Hepatozoon* spp., determined by observing the protozoan's gametes in leukocytes from blood smears (Figure 1). Data compilation involved obtaining, analyzing, and grouping the following information: age, sex, breed, distribution of cases over the years, seasonality, and hematological profile, using the reference values from Jain (1993) and Meyer and Harvey (2004). Prior to conducting the study, a free informed consent form (TCLE) was sent to the veterinary laboratory to ensure confidentiality of patient and guardian data. The frequency of the variables was calculated with a 95% confidence interval using the Epi info™ version 7.2.

RESULTS AND DISCUSSION

Profiles data of 272 dogs naturally infected with *Hepatozoon* spp. are shown in Table 1. Regarding breed distribution, 135 (49.63%) animals were mixed-breed, while 137 (50.37%) were specific breeds. Among the identified breeds were: Akita (n=1), American (n=7), Blue Heeler (n=6), Border Collie (n=7), Boxer (n=1), Bull Terrier (n=2), Chow Chow (n=8), Cocker Spaniel (n=1), Dachshund (n=5), Dogue Bordeaux (n=1), Fila (n=1), Brazilian Terrier (n=1), Siberian Husky (n=2), Labrador (n=1), Maltese (n=1), German Shepherd (n=3), Belgian Shepherd (n=1), Pinscher (n=9), Pit Bull (n=14), Pointer (n=1), Poodle (n=7), Pug (n=2), Rottweiler (n=7), Shar-Pei (n=3), Shih Tzu (n=13), and German Spitz (n=1).

Canine hepatozoonosis has been described in several countries and is typically more prevalent in younger animals, as reported by Chhabra *et al.* (2013) and Mundim *et al.* (1994, 2008b). Younger animals are predisposed to the development of diseases due to their immature immune systems and because they present the acute phase of infection, at which the number of gametocytes in the peripheral blood is higher (Gomes *et al.*, 2010; Antunes *et al.*, 2015; Garrido *et al.*, 2022). Contrary to these findings, this study observed a higher incidence of hepatozoonosis in adult animals, consistent with the observations made by Miranda (2013), who attributed this higher incidence in adults to their prolonged exposure to the *H. canis* vector and

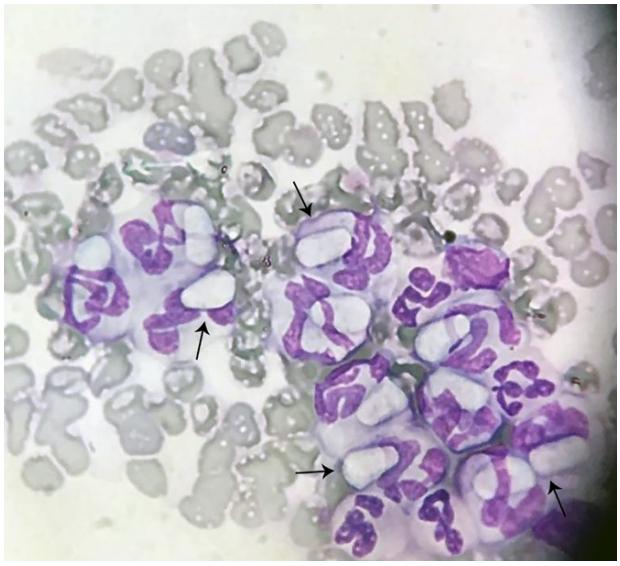

Figure 1. Photomicrograph of intracytoplasmic *Hepatozoon* spp. gamonts (arrows) in neutrophils from dogs in the municipality of Mineiros. Blood smear stained with rapid panoptic dye, viewed under immersion microscopy - 100x objective.

the prophylactic measures typically focused on puppies, leading to reduced tick infestation in younger animals.

The results of this study demonstrated equivalence between the sexes, confirming the absence of a sexual predisposition, consistent with findings reported by Antunes *et al.* (2015) and Garrido *et al.* (2022). Feliciano *et al.* (2012) suggested immunological instability in females due to hormonal variations during pregnancy and the estrus period, potentially predisposing them to infections, given that *Hepatozoon* spp. are opportunistic pathogens. In contrast, other studies have reported a higher prevalence of infected males, attributing this difference to factors such as predominance, population, and behavioral aspects (Gavazza; Bizzet; Papini, 2003; Mundim *et al.*, 2008b; Miranda *et al.*, 2013).

In the evaluation of the presence of *Hepatozoon* spp. regarding breed, previous studies have noted a higher prevalence of no defined breed has been found in dogs (Gomes *et al.*, 2010; Garrido *et al.*, 2022). Despite this, breed is not recognized as a predisposing factor for hepatozoonosis (Antunes *et al.*, 2015). The epidemiological relevance of this infection lies in the presence of tick infestations as its transmission depends on the ingestion of infected ticks (Oliveira *et al.*, 2021).

Analysis of the distribution of cases over a five-year period showed an annual increase in the number of dogs diagnosed with parasitism by *Hepatozoon* spp. Notably, seasonality was found to influence their occurrence, with a higher number of cases diagnosed in autumn and winter (dry season) compared to spring and summer (rainy season), as shown in Figure 2. The lower rates of diagnosed cases in the rainy season can be attributed to the reduced presence of the vector at this time. It is well-documented that rainfall has a deleterious effect on the reproduction of ticks, leading to a decline in their population and, consequently, a lower risk of transmission of pathogens to animals (Adejinmi, 2011).

Analysis of the hematological profile of dogs infected with *Hepatozoon* spp. using an erythrogram verified that a large proportion of the animals had red blood cell counts and hematocrit values below the reference values. The presence or absence of erythrocyte regenerativity can be determined by evaluating the percentage of reticulocytes in red blood cells, which classifies the type of anemia as regenerative or non-regenerative (Rahmeier; Holsbach, 2022). However, the absence of these data in the present study is attributed to clinicians' lack of adherence to request examination.

To determine the presence of anemia, the values of red blood cells, hemoglobin, and hematocrit were evaluated and correlated. Within the study's evaluation criteria, 76.84% of the dogs had anemia, of which 61.40% had red blood cells with anisocytosis and polychromasia, in addition to 18.48% displaying metabolic cells. Assessment of bone marrow regeneration considered the presence of anisocytosis (RDW), polychromasia and metarubricytes (Table 2). However, some studies have described anemia in dogs infected with *H. canis* to be normocytic, normochromic, and nonregenerative (Mundim *et al.*, 2008b; Chhabra *et al.*, 2013; Paiz *et al.*, 2016).

In the leukogram profile, it was observed that 61.03% of the dogs showed no changes in the total leukocyte count, remaining within the reference values for the species (Table 2). Leukocytosis, which is sometimes neutrophilic, is associated with high parasitic loads in animals (Mundim *et al.*, 2008b; Chiareli, 2009; Chhabra *et al.*, 2013; Peres *et al.*, 2022). However, it can be inferred that most dogs in this study had low parasitemia.

Checking the leukocyte cytology, 26.10% of activated monocytes, 2.94% of toxic neutrophils, 2.21% showing both changes, and 13.43% of reactive lymphocytes were identified. Notably, abnormalities in the neutrophils, monocytes, and lymphocytes counts in the parasitized dogs were not significant in this study. However, Peres *et al.* (2022) reported that monocytosis is a common finding in hepatozoonosis.

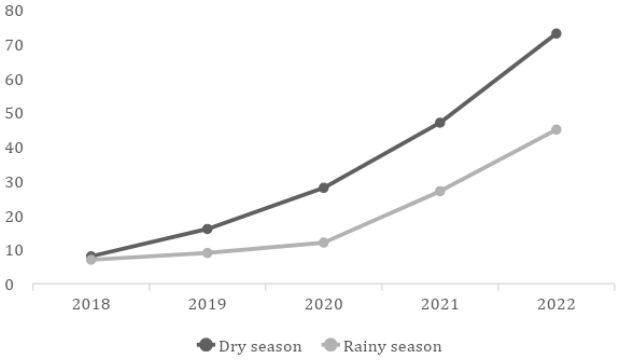

Figure 2. Cases of dogs parasitized by *Hepatotoon* spp. related to seasonality, from 2018 to 2022, in the municipality of Mineiros, Goiás state.

Table 1. Profile of dogs parasitized by *Hepatozoon* spp., from 2018 to 2022, in the municipality of Mineiros, Goiás state.

		N (272)	Frequency (%)	CI%
Age range	Young	46	16.91	12.66 – 21.91
	Adult	226	83.09	78.09 – 87.34
Sex	Female	153	56.25	50.13 – 62.23
	Male	119	43.75	37.77 – 49.87
Breed	Specific breed	137	50.37	44.27 – 56.46
	Mixed-breed	135	49.63	43.54 – 55.73

Young (≤ 12 months); adult (> 12 months); CI = confidence interval

Table 2. Hematological profile of dogs diagnosed with *Hepatozoon* spp., through visualization of gamontes in blood smears, from 2018 to 2022, in the municipality of Mineiros, state of Goiás.

		N (272)	Frequency (%)	CI%
Anemia	Not anemic	63	23.16	18.28 – 28.64
	Normocytic and Normochromic	42	15.44	11.36 – 20.29
	Anisocytosis and Polychromasia	167	61.40	55.33 – 67.21
Total leukocytes	Leukocytosis	52	19.12	14.62 – 24.30
	N/C	166	61.03	54.96 – 66.86
	Leukopenia	54	19.85	15.28 – 25.10
Platelets	Thrombocytosis	26	9.56	6.34 – 13.69
	N/C	68	25.00	19.97 – 30.59
	Thrombocytopenia	178	65.44	59.46 – 71.08
Total proteins	Hyperproteinemia	120	44.12	38.13 – 50.24
	N/C	111	40.81	34.91 – 46.91
	Hypoproteinemia	41	15.07	11.04 – 19.89

CI = Confidence interval; N/C = no change

Furthermore, eosinopenia occurred in 47.06% of cases, corroborating the results of Chhabra *et al.* (2013) and Mundim *et al.* (2008b), who attributed eosinopenia to the destruction of eosinophils by corticosteroids and catecholamines released into the body in response to the stress caused by the infection.

The platelet counts revealed thrombocytopenia in 65.44% of the parasitized dogs, similar to what was reported by Chiareli (2009). The precise mechanism of thrombocytopenia in cases of hepatozoonosis remains unknown; however, some studies have reported the presence of coinfections with other hemoparasites as a contributing factor (Baneth, 2006; Costa, 2011; Peres *et al.*, 2022).

Plasma proteins were elevated in 44.12 % of the cases, a finding identified as common by Paiz *et al.* (2016), generated by the stimulation of the humoral response induced by the hemopathogen (Table 2).

Among the infected animals, 7.58% presented coinfections on cytological examination. However, it's important to note that other infections should not be ruled out because of the low sensitivity of the technique applied. Coinfections are commonly observed in canine hepatozoonosis, as *Hepatozoon* spp. are opportunistic parasites. Yet, in this study, coinfections were identified in only 8.82% of the parasitized dogs, of which four were infected with *Anaplasma platys*, three with *Babesia* spp., and fourteen with *Ehrlichia* spp. (Figure 3). Notably, Lentz corpuscles were identified in three animals.

Hepatozoon spp. are typically found inside monocytes and neutrophils (Cruz *et al.*, 2023). Instances of invasion and coinfection of the same neutrophil are rare, as is the detection of ehrlichial morulae in canine neutrophils is uncommon. However, Aguiar *et al.* (2019) previously reported the coinfection of neutrophils with *Hepatozoon* spp. and *Ehrlichia spp.* morulae, reinforcing the results found in this study.

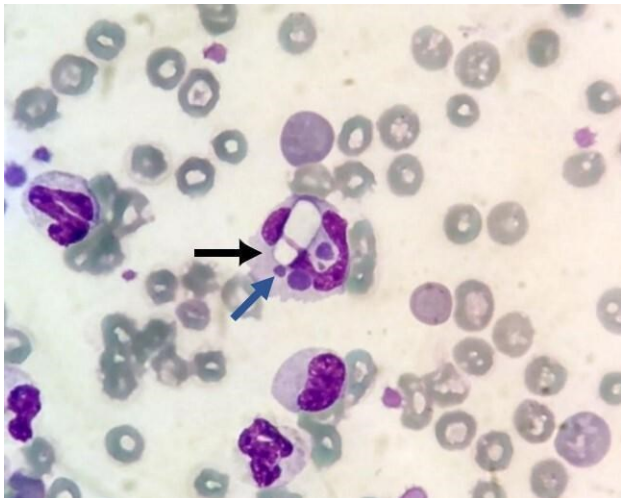

Figure 3. Canine blood smear sample, made available by the Veterinary Clinical Pathology Laboratory in the city of Mineiros-GO, stained by rapid panoptic visualized under immersion microscopy, under a 100x objective, showing neutrophils co-infected by gamontes of *Hepatozoon* spp. (black arrow) and *Ehrlichia* spp. morula (blue arrow).

CONCLUSION

Hepatozoonosis occurs predominantly in regions with a hot climate, which explains the exponential increase in cases over the five-year study period, particularly during the dry season. This trend was notable in the central-western region, where as the semi-humid tropical climate prevails. However, no specific predilection for age, sex, or breed was observed in this study. The observed erythrocyte and platelet changes suggest hematological compromise due to *Hepatozoon* spp., and exacerbation of these parameters may be associated with coinfection with other pathogens.

REFERENCES

ADEJINMI, J. O. Effect of water flooding on the oviposition capacity of engorged adult females and hatchability of eggs of dog ticks: *Rhipicephalus sanguineus* and *Haemaphysalis leachi leachi*. **Journal of Parasitology Research**, v. 2011, p. 1-6, 2011.

AGUIAR, D. M. *et al*. Uncommon *Ehrlichia canis* infection associated with morulae in neutrophils from naturally infected dogs in Brazil. **Transboundary and emerging diseases**, v. 67, p. 135-141, 2019.

ANTUNES, T. R. *et al*. Aspectos hematológicos e epidemiológicos de cães naturalmente infectados por *Hepatozoon* spp. no município de Campo Grande, Mato Grosso do Sul, Brasil. **Acta Veterinaria Brasilica**, v. 9, n. 3, p. 324-328, 2015.

BANETH, G. *et al*. Canine hepatozoonosis: two disease syndromes caused by separate *Hepatozoon* spp. **Trends in Parasitology**, v. 19, n. 1, p. 27-31, 2003.

BANETH, G. *Hepatozoon canis* infection. *In:* GREENE, C. E. **Infectious diseases of the dog and cat, Hepatozoonosis.** Canada: Saunders, 2006. p. 698-711.

BANETH, G.; SAMISH, M.; SHKAP, V. Life cycle of *Hepatozoon canis* (Apicomplexa: Adeleorina: Hepatozoidae) in the tick *Rhipicephalus sanguineus* and domestic dog (*Canis familiaris*). **Journal of Parasitology**, v. 93, n. 2, p. 283-299, 2007.

BANETH, G. Perspectives on canine and feline hepatozoonosis. **Veterinary Parasitology**, v. 181, n. 1, p. 3-11, 2011.

CHHABRA, S. *et al*. Retrospective study of clinical and hematological aspects associated with dogs naturally infected by *Hepatozoon canis* in Ludhiana, Punjab, India. **Asian Pacific Journal of Tropical Biomedicine**, v. 3, n. 6, p. 483-486, 2013.

CHIARELI, R. A**. Investigação clínico-laboratorial e molecular da infecção por *Hepatozoon canis* em cães da região periurbana de Brasília.** 2009. 84 f. Dissertação (Mestrado em Saúde Animal) – Universidade de Brasília, Brasília, 2009.

CICUTTINA, G. L.; DE SALVO, M. N. Detección molecular y análisis filogenético de *Hepatozoon canis* (Eucoccidiorida: Haemogregarinidae) en perros clínicamente sanos de Bahía Blanca (Buenos Aires). **FAVE Ciencias Veterinarias**, v. 16, n. 1, p. 46–49, 2017.

COSTA, H. X. **Interação de hemoparasitos e hemoparasitoses em casos clínicos de trombocitopenia em cães no município de Goiânia.** 2011. 72 f. Dissertação (Mestrado em Ciência Animal) – Universidade Federal de Goiás, Goiânia, 2011.

CRUZ, C. O. *et al*. *Hepatozoon* spp. em cães na Zona da Mata Mineira–Relato de Casos. **Revista Brasileira de Higiene e Sanidade Animal**, v. 17, n. 2, p. 1-9, 2023.

FELICIANO, M. A. R. *et al*. Imunologia na gestação de cadelas: revisão de literatura. **Revista Brasileira de Reprodução Animal**, v. 36, n. 3, p. 158-162, 2012.

FORLANO, M. D. *et al*. Molecular characterization of *Hepatozoon* sp. from Brazilian dogs and its phylogenetic relationship with other *Hepatozoon* spp. **Veterinary Parasitology**, v. 145, n. 1-2, p. 21-30, 2007.

GARRIDO, A. C. N. *et al*. Aspectos hematológicos e sazonais da infecção natural por *Hepatozoon* spp. em cães no sertão da Paraíba. **Ciência Animal**, v. 32, n. 3, p. 9-17, 2022.

GAVAZZA, A.; BIZZET, M.; PAPINI, R. Observations on dogs found naturally infected with *Hepatozoon canis* in Italy. **Revue de Médecine Vétérinaire**, v. 154, n. 8-9, p. 565-571, 2003.

GOMES, P. V. *et al*. Occurrence of *Hepatozoon* sp. in dogs in the urban area originating from a municipality in southeastern Brazil. **Veterinary parasitology**, v. 174, n. 1-2, p. 155-161, 2010.

LASTA, C. S. *et al*. Infecção por *Hepatozoon canis* em canino doméstico na região Sul do Brasil confirmada por técnicas moleculares. **Ciência Rural**, v. 39, p. 2135-2140, 2009.

MIRANDA, R. M. **Prevalência e caracterização molecular da espécie *Hepatozoon* e parâmetros hematológicos e bioquímicos de cães (*Canis familiaris*) naturalmente infectados procedentes da microrregião de Uberlândia – MG.** 2013. 74 f. Tese (Doutorado em Imunologia e Parasitologia Aplicadas) – Universidade Federal de Uberlândia, Uberlândia, 2013.

MUNDIM, A. V. *et al*. *Hepatozoon canis*: estudo retrospectivo de 22 casos de infecção natural em cães de Uberlândia, MG. **Revista do Centro de Ciências Biomédicas da Universidade Federal de Uberlândia**, v. 10, n. 1, p. 89-95, 1994.

MUNDIM, E. C. S. *et al*. Incidência de hemoparasitoses em cães (*Canis familiares*) de rua capturados pelo Centro de Controle de Zoonoses (CCZ) da cidade de Anápolis-GO. **Ensaios e Ciência: Ciências Biológicas, Agrárias e da Saúde**, v. 12, n. 2, p. 107-115, 2008a.

MUNDIM, A. V. *et al*. Clinical and hematological signs associated with dogs naturally infected by *Hepatozoon* sp. and with other hematozoa: a retrospective study in Uberlândia, Minas Gerais, Brazil. **Veterinary Parasitology**, v. 153, n. 1-2, p. 3-8, 2008b.

O'DWYER, L. H.; MASSARD, C. L.; DE SOUZA, J. C. P. *Hepatozoon canis* infection associated with dog ticks of rural areas of Rio de Janeiro State, Brazil. **Veterinary Parasitology**, v. 94, n. 3, p. 143-150, 2001.

O'DWYER, L. H. Brazilian canine hepatozoonosis. **Revista Brasileira de Parasitologia Veterinária**, v. 20, n. 3, p. 181-193, 2011.

OLIVEIRA, L. V. S. *et al*. Hematological, clinical and epidemiological aspects of *Hepatozoon canis* infection by parasitological detection in dogs from the rural area of Sousa, Paraíba, Brazil**. Ciência Rural**, v. 51, n. 2, p. 1-8, 2021.

PAIZ, L. M. *et al*. Hematological disorders detected in dogs infected by *Hepatozoon canis* in a municipality in Mato Grosso do Sul State, Brazil. **Arquivo Brasileiro de Medicina Veterinária e Zootecnia**, v. 68, p. 1187-1194, 2016.

PALUDO, G. R. *et al*. *Hepatozoon* spp.: report of some cases in dogs in Brasília, Brazil. **Veterinary Parasitology**, v. 118, n. 3-4, p. 243-248, 2003.

PEREIRA, A. M. *et al*. Ocorrência de *Hepatozoon* sp. em caninos naturalmente infectados no município de Piraí, Rio de Janeiro, Brasil. **Revista Brasileira de Ciência Veterinária**, v. 18, n. 2-3, 2011.

PERES, P. C. O. *et al*. Aspectos Hematológicos de Cães Infectados por *Hepatozoon* em Goiânia, Goiás. **Concilium**, v. 22, n. 1, p. 1-9, 2022.

RAHMEIER, G.; HOLSBACH, V. Estudo eritrocitário comparativo de cães anêmicos**. Arquivos Brasileiros de Medicina Veterinária FAG,** v. 5, n. 2, 2022.

RAMOS, R. *et al*. Molecular survey and genetic characterization of tick-borne pathogens in dogs in metropolitan Recife (north-eastern Brazil). **Parasitology research**, v. 107, p. 1115-1120, 2010.

RUBINI, A. S. *et al*. Molecular identification and characterization of canine *Hepatozoon* species from Brazil. **Parasitology Research**, v. 97, p. 91-93, 2005.

SPOLIDORIO, M. G. *et al*. *Hepatozoon canis* infecting dogs in the State of Espírito Santo, southeastern Brazil. **Veterinary Parasitology**, v. 163, n. 4, p. 357-361, 2009.

SPOLIDORIO, M. G. *et al*. Molecular detection of *Hepatozoon canis* and *Babesia canis vogeli* in domestic dogs from Cuiabá, Brazil. **Revista Brasileira de Parasitologia Veterinária**, v. 20, p. 253-255, 2011.