



## *Hepatozoon canis* - An Insidious Pathogen and its Morphological Alterations in Chippiparai Dogs

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### Abstract

This study presents about the pathogenicity of *Hepatozoon canis* in native dogs of southern districts of Tamil Nadu, India. During the study period from January 2020 to December 2020, a total of 275 Chippiparai dogs were screened for haemoparasitic diseases, out of which seven cases of *H. canis* were recorded. Among those seven cases, three were concurrent infection with transmissible venereal tumour, one with *Babesia canis* and three were solitary infections. The clinical signs were mostly non-specific with mild to moderate fever, anorexia and lethargy. The major haematological findings in these cases were severe anaemia in *B. canis* infection and rest of the cases showed neutrophilic leucocytosis. In one case, more the 30 per cent neutrophils were infected with gamonts of *H. canis*. The morphology of *H. canis* is altered from elliptical to round or oval due to chemotherapy with vincristine in Transmissible venereal tumour (TVT) affected dogs.

**Keywords:** Anaemia; Chippiparai Dogs; *H. canis*; Morphological Changes; Neutrophilic Leucocytosis

### Introduction

*Hepatozoon canis* is a haemoprotozoan pathogen, detected for the first time in the blood of dogs in India and determined as *Leukocytozoon canis* [1]. Canine hepatozoonosis is a tick-borne disease caused by apicomplexan protozoan parasite. In contrast to other tick borne pathogens, *H. canis* will be transmitted by the ingestion of tick or parts of ticks (*Rhipicephalus sanguineus*) containing the mature *Hepatozoon* oocysts [2].

The Chippiparai is a native hound group dog breed used mainly for hunting purpose of Southern parts of Tamil Nadu, India. The sturdy nature of the native dogs resists many parasitic diseases, but under stress due to excessive exercise, hunting, pregnancy and immune suppression due to steroidal drugs for other diseases,

therapy for transmissible venereal tumour made the mongrels susceptible to the haemoprotozoan infection when the dogs is having tick infection [3].

The canine hepatozoonosis could be asymptomatic with low level of parasitaemia or could be manifested as a severe life-threatening disease with non-specific clinical signs like fever, lethargy, anaemia, cachexia in dogs with high parasitaemia and concurrent infection [4]. Chhabra, *et al.* (2013) [5] recorded the haematological alterations mainly consisted of anaemia (73.53%), leukocytosis (44.11%), neutrophilia (44.11%) and eosinopenia (35.29%) in their study. The increase in blood urea nitrogen, creatinine level along with hypoproteinaemia was observed in mongrel dog hepatozoonosis [6]. The reports on the haematological and biochemi-

cal analysis showed no uniform changes in many studies of canine hepatozoonosis [7,8]. The study of *H. canis* in native dogs is skimpy and recondite information. Hence the study was proposed, to rule out the importance of concurrent infection, pathogenicity and morphological changes of the insidious *Hepatozoon canis* pathogen in Chippiparai dogs.

**Materials and Methods**

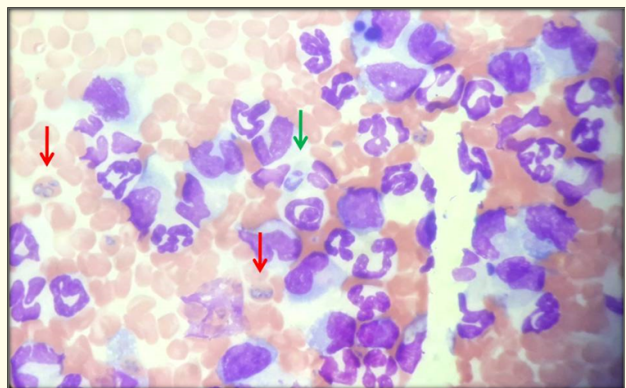
A total 275 Chippiparai dogs were screened for haemoparasitic infection from January 2020 to December 2020 in Veterinary Clinical Complex, VCRI, Tirunelveli. Out of 275 only seven diagnosed as positive for *Hepatozoon canis* with various clinical conditions like anorexia, pyrexia, myalgia, lethargy and genital masses. Among the seven cases, four were females and three were males, the age ranges from 1 to 5 years. The thorough physical examination in these animals was carried out. The cytological impression smears were taken from the genital mass in three cases. The thin blood smear and whole blood was collected to evaluate the haematobiochemical changes. The complete blood count was analysed in auto haematology analyzer (3part Celenium junior, Trivitron Pvt Ltd) and serum in semi-automated biochemical analyzer (Lab mate, Trivitron Pvt Ltd) by spectrophotometrically using standard diagnostic kits. Differential Leucocytes was counted manually in the monolayered area of the thin blood smear and *H. canis* gamont in neutrophils were identified by the stained with Leishman-Giemsa cocktail stain.

**Results and Discussion**

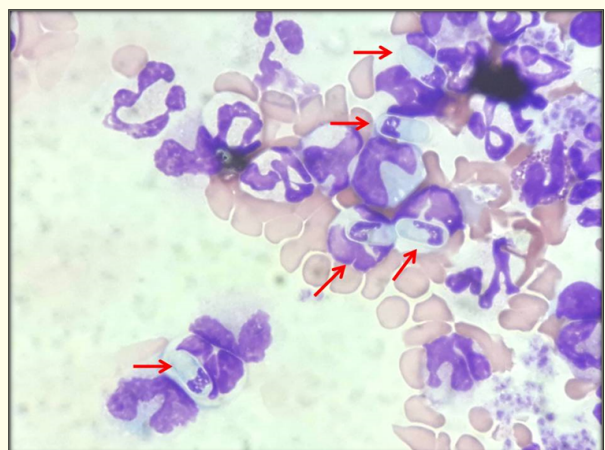
The presented dogs were showing non-specific clinical signs, asymptomatic to mild fever, tick infestation, anorexia and lethargy [9,10]. This was in concordance with the present study except the genital mass in TVT affected cases. The affected animal’s blood smear revealed the presence of gelatin capsule shaped *H. canis* gametocytes in the neutrophils as recorded in the other breeds of dogs. With regard to this study, breed was Chippiparai, age recorded was 1 to 5 years and sex of the animals was four females and three males were infected. The presence of *H. canis* in native dogs indicates that this haemoprotozoan infection had no breed, sex and age predilection. Current study observation corroborates with the earlier findings [5,11].

The genital masses were grossly small, fragile and multilobulated. The cytological study from the genital mass confirmed the presence of transmissible venereal tumour and the blood smear of the same dogs also had the gamonts in the neutrophils. In one *Babesia canis* co-infected anaemic dog and a dog with continuous bleeding

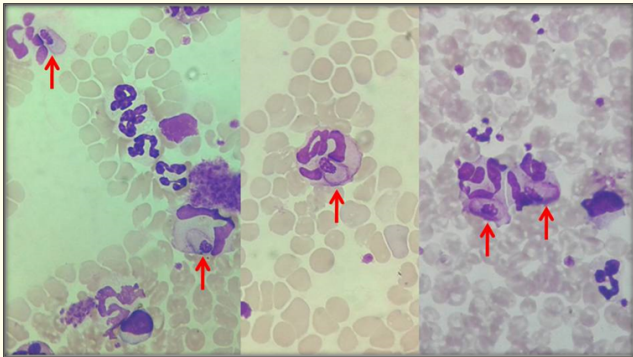
in genital mass had severe reduction in haematological parameters were observed viz., reduction in haemoglobin, packed cell volume and red blood cells [3]. The blood smears of that case were diagnosed as concomitant infection of *B. canis* and *H. canis* (Figure 1). Harbouring multiple parasites is due to sharing the same brown dog tick as vectors [12]. Among the other three solitary cases, one particular case blood smear had *H. canis* in more than 30 percent neutrophils (Figure 2). In most of these cases, neutrophilic leucocytosis is recorded due to the concurrent infections [13]. To the best of author’s knowledge, the morphological alterations of *H. canis* gamonts and the nucleus (Figure 3) were recorded first in this study for those native animals which underwent chemotherapy for the transmissible venereal tumour. Instead of ellipsoidal shape of the parasite, it was rounded or oval and the nucleus too changed from the normal horse-shoe appearance as centrally placed round nucleus. Eljadar, *et al.* 2012 [14] first time recorded abnormal gametocytes in neutrophils in the study of morphometric variations in gametocytes of *Hepatozoon canis* from naturally infected dogs.



**Figure 1:** Blood Smear- *B. canis* (Red arrow), *H. canis* (Green arrow) – Neutrophilia.



**Figure 2:** Blood smear – *H. canis*- Gelatin shaped gamonts (typical) in more number of neutrophils in single field – Neutrophilia.



**Figure 3:** Blood smear of 3 cases – *H. canis* (atypical) – Gametocytes and nuclear morphological alteration (Rounding of parasites with roundness of nucleus) of the gamont in neutrophils – in dogs (three cases) underwent chemotherapy for TVT

The protozoan concentration in the neutrophils is directly proportional to the severity of illness. In most dogs, haematological finding was mild to moderate leucocytosis and the blood picture revealed neutrophilia in all these cases probably due to the inflammation caused by the tumour and parasitic infection. The comorbid condition like *B. canis* with *H. canis*, TVT with *H. canis* or other mixed insidious infection could be due to low myeloperoxidase activity in *H. canis* infected neutrophils [15]. The serum biochemistry results in TVT co-infection dog revealed elevated alanine aminotransferase (ALT), alkaline phosphatase (ALP), blood urea nitrogen (BUN), creatinine and in the *B. canis* co-infection case, there was an elevation of ALT, ALP, hypoproteinaemia due to the concurrent chemotherapy and the liver damage caused by the piroplasms of *B. canis* and progression of schizogony of *H. canis* in bone marrow and spleen [3]. In the solitary cases, only in one case there was mild elevation of ALT and ALP might be due to more severity in concentration of parasites and its multiplication [13]. The increase in elevation of the biochemical values might be due to the inflammatory condition of the tumour and the parasitic proliferation in liver, bone marrow and causing damage to the glomeruli [4,10,13].

The identification of *H. canis* in most of these cases was incidental findings during the blood smear examination with no suspicion about the presence of this particular parasite. Even then *H. canis* contributes major changes in terms of clinical symptoms and clinical pathology of the infected animals. Henceforth *H. canis* should be considered as insidious pathogen which will cause severe threat

to the infected animals which addresses special attention while screening the blood smears.

### Conclusions

The canine hepatozoonosis caused by *H. canis* is less pathogenic, only thing pose threat is severe malady in dog is highly debilitating effect. The comorbid infection only makes high pathogenicity in affected dogs. Unfortunately, the native dog owners never notice the severity of ticks' infection on dogs until the animal shows severe clinical signs. Hence the prevention of exposure of dogs to ticks by the use of acaricides is warranted to control the spread of canine hepatozoonosis and can also safely guard the dogs from concurrent infection.

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