

Epidemiological studies of canine ehrlichiosis and babesiosis in Andhra Pradesh

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Abstract

The present study was aimed to report the prevalence of canine ehrlichiosis and babesiosis in dogs. Overall prevalence of canine ehrlichiosis and babesiosis was 28.23 per cent (35/124) based on polymerase chain reaction (PCR). Among 35 dogs, 20 (57.14%) were affected with canine ehrlichiosis, 11 (31.43%) with canine babesiosis and 4 (11.43%) with concurrent ehrlichiosis and babesiosis. Canine ehrlichiosis (45%) and babesiosis (63.64%) was found to be higher in dogs below 2 years of age. Breed wise prevalence of canine ehrlichiosis and canine babesiosis was highest in Labrador retriever. The occurrence was higher in males (68.57%) compared to females (31.43%) in canine ehrlichiosis and babesiosis.

Keywords: Epidemiology, Incidence, Canine Ehrlichiosis, Babesiosis

Introduction

Tick borne diseases are emerging problem worldwide in pet animal practice. Ticks are notorious vectors of various pathogenic protozoa, rickettsiae, bacteria, and viruses that cause serious and life-threatening illnesses in animals (Alekseev *et al.*, 2001). Hot and humid conditions favours the proliferation of tick population, tick borne diseases like ehrlichiosis and babesiosis are of significant concern in tropical countries like India., as they are transmitted mainly by the brown dog tick, *Rhipicephalus sanguineus* (Shaw *et al.*, 2001). *Rhipicephalus* is the most common tick present in urban India (Abd Rani *et al.*, 2011). *Rhipicephalus* (*Boophilus*) *microplus*, *R. sanguineus* and *Hyalomma anatolicum* species are reported in 24, 21 and 20 states of India, respectively (Ghosh and Nagar, 2014). Hence the incidence of tick-borne disease also varies according to abundance of parasitic burden. The variation in the occurrence might be attributed to sample size, geographical area, climatic conditions which directly influence the tick population, time of sample collection, sub clinical infection, immune status of the host and other managemental practices in different parts of India (Mahalingaiah *et al.*, 2017). Diagnosis of

the haemoparasitic diseases is challenging due to their different phases and multiple clinical manifestations. Traditional techniques like haematology, blood smear examination, serology and isolation were valuable tools for diagnosis of haemoparasites (Harrus and Waner, 2011). Molecular techniques like polymerase chain reaction was sensitive and specific diagnostic method in the diagnosis of canine ehrlichiosis and babesiosis. Hence, the present study was conducted to record the occurrence of canine ehrlichiosis and babesiosis in dogs by PCR.

Materials and Methods

During the study period that was carried out at Veterinary Clinical Complex, College of Veterinary Science, Tirupati and NTR College of Veterinary Science, Gannavaram, from february 2023 to november 2023. A total of 2,398 dogs were screened for canine ehrlichiosis and babesiosis. The age, gender and breed wise incidence was recorded during present study. Primary screening of dogs was done based on history, clinical signs, lymphadenopathy, tick infestation and blood smear examination. Blood was collected aseptically from cephalic and sephanous vein in EDTA vacutainers and stored at 4°C for DNA extraction. Thin peripheral blood smear was made from whole blood for microscopic examination. Rapid diagnosis was carried out by

using Rapid Ab test kit as per protocol recommended by the manufacturer (J&G BIOTECH LTD). Genomic DNA was extracted from 200µl of stored blood using HiPurA® Blood Genomic DNA Miniprep Purification kit as per manufacturer protocol. PCR was carried out with a reaction mixture of 20µl using (ECA-F): 5' AACACATGCAAGTCGAACGGA3' and (HE3-R): 5' TATAGGTACCGTCATTATCTTCCCTAT3' primers for *E. canis* (Wen *et al.*, 1997) targeting a 16S rRNA gene and (Ba103F): 5' CCAATCCTGACACAGGGAGGTAGTGACA 3' and (Ba721R): 5' CCCCAGAACCCAAAGACTTTGATTTCTCTCAAG 3' primers for *Babesia* sp. (Azhahianambi *et al.*, 2018) targeting a 18S rRNA gene.

Results

Out of 2,398 dogs, 124 were identified to have canine ehrlichiosis and babesiosis based on the history and clinical signs and blood smear examination. The identified cases were subjected to haemato-biochemical and urinalysis to assess the renal function and PCR analysis for further confirmation. In the present study, 35 dogs were confirmed with ehrlichiosis and babesiosis based on PCR, thus representing the overall incidence of 28.23 per cent. Out of 35 affected dogs, 20 (57.14%) had canine ehrlichiosis, 11 (31.43%) dogs affected with canine babesiosis and 4 (11.43%) dogs with concurrent ehrlichiosis and babesiosis (Table 1 and Figure 1).

The age wise incidence of canine ehrlichiosis and canine babesiosis was found to be higher in dogs less than 2 years of age (51.43%) followed by dogs aged in between 2-4 years (20%), 4-6 years (11.43%), 6-8 years (8.57%), 8-10 years (5.71%) and was less in dogs above 10 years of age (2.86%). The occurrence of canine ehrlichiosis was higher in dogs less than 2 years (45%) followed by 30 per cent in dogs aged between 2-4 years, 10 per cent in dogs aged between 4-6 years and 5 per cent each in dogs aged between 6-8 years, 8-10 years and 10-12 years. Higher occurrence of canine babesiosis was observed in dogs less than 2 years of age (63.64%) followed by 2-4 years, 4-6 years, 6-8 years and 8-10 years (9.09%) each. In concurrent infection, higher occurrence was noticed in dogs less than 2 years of age (50%) followed by 4-6 years and 8-10 years (25%) each (Table 2).

Out of 35 affected dogs with ehrlichiosis and babesiosis, 64.52 per cent (20/35) were male and 35.48 per cent (11/35) were female. Canine ehrlichiosis was recorded in 65 per cent (13/20) of the male dogs and 35 per cent (7/20) of female dogs, while canine babesiosis was more in males (63.34%) than in female dogs (36.36%) and in concurrent infection males (75%) were more affected than females (25%) (Table 3).

The breed wise incidence of canine ehrlichiosis and babesiosis was recorded high in Labrador retriever (48.57%) followed by Pomeranian (11.43%), Dobermann (8.57%), Golden retriever, Pug, German Shepherd, Mongrel (5.71%) each, followed by others. In our study, higher occurrence of canine ehrlichiosis was noticed in Labrador retriever (45%) followed by Pomeranian (15%), Mongrel (10%), Dobermann, Golden retriever, Pug, German Shepherd, Beagle and Belgian Shepherd (5%) each. Among the dogs with canine babesiosis, higher occurrence was observed in Labrador retriever (36.36%) followed by Dobermann (18.18%), Pomeranian, Golden retriever, Pug, German Shepherd and Shih Tzu (9.09%) each. All the concurrently infected dogs were Labrador retriever (Table 4).

Discussion

The incidence of canine ehrlichiosis and babesiosis was 28.23 per cent in the present study which might be due to hot and humid climate favouring the development, survival of tick population and increased urbanisation (Bayer, 2013). Among 35 dogs, 20 (57.14%) were affected with canine ehrlichiosis in the present study. Various studies had been carried out regarding the occurrence of *E. canis* and was reported to be 50 per cent (49/98) in Chennai (Lakshmanan *et al.*, 2007) and 20.6 per cent from four different regions of India (Abd Rani *et al.*, 2011). The variation in the occurrence might be attributed to sample size, geographical area, climatic conditions which directly influence the tick population and time of sample collection. Higher occurrence might be due to the fact that the study was carried out in dogs referred to the veterinary clinics presented with clinical manifestations consistent with canine ehrlichiosis (Milanjeet *et al.*, 2014).

Table I: Occurrence of canine ehrlichiosis and babesiosis

S.No	Total No. of dogs screened	No.of dogs affected	Percentage Affected	Canine Ehrlichiosis	Canine Babesiosis	Concurrent infection
1.	124	35	28.23%	20(57.14%)	11(31.43%)	4(11.43%)

*Figures in parenthesis indicates percentage

Table II: Age wise occurrence of canine ehrlichiosis and babesiosis

S.No	Age group	Number of dogs affected	Canine ehrlichiosis (n=20)	Canine babesiosis (n=11)	Concurrent infection (n=4)
1.	Up to 2 years	18(51.43%)	9(45%)	7(63.64%)	2(50%)
2.	2-4 years	7(20.00%)	6(30%)	1(09.09%)	-
3.	4-6 years	4(11.43%)	2(10%)	1(09.09%)	1(25%)
4.	6-8 years	3(08.57%)	1(5%)	1(09.09%)	1(25%)
5.	8-10 years	2(05.71%)	1(5%)	1(09.09%)	-
6.	10-12 years	1(02.86%)	1(5%)	-	-
	Total	35(100%)	20(100%)	11(100%)	4(100%)

*Figures in parenthesis indicates percentage

Table III: Gender wise occurrence of canine ehrlichiosis and babesiosis

S.No	Gender	Number affected	Canine ehrlichiosis (n=20)	Canine babesiosis (n=11)	Concurrent infection (n=4)
1.	Male	23(65.71%)	13(65%)	7(63.64%)	3(75%)
2.	Female	12(34.29%)	7(35%)	4(36.36%)	1(25%)
	Total	35(100%)	20(100%)	11(100%)	4(100%)

*Figures in parenthesis indicates percentage

Table IV: Breed wise occurrence of canine ehrlichiosis and babesiosis

S.No	Name of breed	Number affected	Canine ehrlichiosis (n=20)	Canine babesiosis (n=11)	Concurrent infection (n=4)
1.	Labrador retriever	17(48.57%)	9(45%)	4(36.36%)	4(100%)
2.	Pomeranian	4(11.43%)	3(15%)	1(09.09%)	-
3.	Dobermann	3(08.57%)	1(5%)	2(18.18%)	-
4.	Golden retriever	2(05.71%)	1(5%)	1(09.09%)	-
5.	Pug	2(05.71%)	1(5%)	1(09.09%)	-
6.	German Shepherd	2(05.71%)	1(5%)	1(09.09%)	-
7.	Mongrel	2(05.71%)	2(10%)	-	-
8.	Beagle	1(02.86%)	1(5%)	-	-
9.	Belgian shepherd	1(02.86%)	1(5%)	-	-
10.	Shih Tzu	1(02.86%)	-	1(09.09%)	-
	Total	35(100%)	20(100%)	11(100%)	4(100%)

*Figures in parenthesis indicates percentage

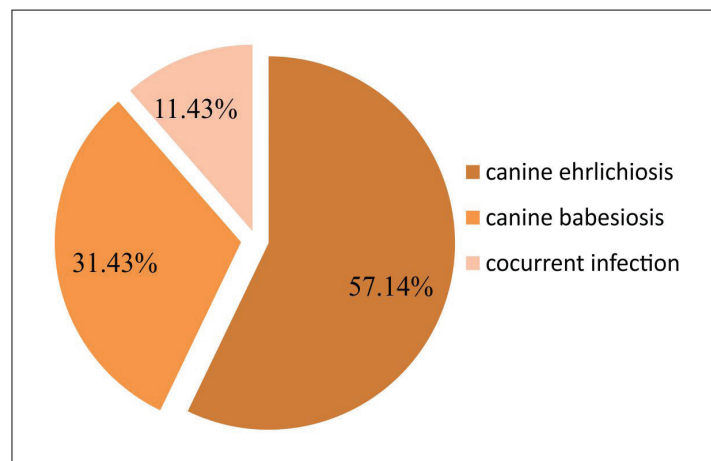


Figure 1: Pie diagram showing the occurrence of canine ehrlichiosis and canine babesiosis

The occurrence of babesiosis was 31.43 per cent (11/35) which was in congruence with the previous reports of Azhahianambi *et al.* (2018) and Muguiro *et al.* (2023) who recorded the occurrence of *Babesia* sp as 31 per cent and 28.8 per cent respectively. On the contrary, Gonde *et al.* (2017) and Sindhu *et al.* (2020) reported lower occurrence of canine babesiosis as 9.79 per cent and 4.25 per cent respectively. 11.43 (4/35) per cent dogs were positive for concurrent ehrlichiosis and babesiosis which was similar to the findings of Fonseca *et al.* (2017) and Azhahianambi *et al.* (2018) who reported the occurrence of concurrent infection of ehrlichiosis and babesiosis as 5.6 per cent and 5.92 per cent respectively and this might be due to *Rhipicephalus sanguineus* can cause co-infection of ehrlichiosis and babesiosis in dogs (Nava *et al.*, 2015). These variations in the occurrence of might be due to differences in sampling, testing methodology, background of study animals (Muguiro *et al.*, 2023) and sub clinical infection, immune status of the host and other managerial practices (Mahalingaiah *et al.*, 2017).

In the present study, the higher occurrence was observed in dogs less than 2 years of age in ehrlichiosis (45%), babesiosis (63.64%) and concurrent infection (50%) which corroborates with the findings of Milanjeet *et al.* (2014), Mahalingaiah *et al.* (2017), Rautenbach *et al.* (2018) and Preena *et al.* (2021). This might be ascribed to the low immunity of young animals compared to adults (Bhadesiya and Raval, 2015).

The gender wise occurrence of canine ehrlichiosis, canine babesiosis and concurrent infection was higher in male dogs (65% vs 63.64% vs 75%) while it was (35% vs 36.36% vs 25%) in female dogs. This

might be attributed to more liking of male dogs by pet owners which resulted in over presentation of male dogs, increased roaming behaviour, sex-linked genetic influences, temperament and hormonal status of the male dogs (Salem and Farag, 2014, Gonde *et al.*, 2017 and Rawangchue and Sungpradit, 2020). On the contrary, Opara *et al.* (2017) reported a higher occurrence in female dogs due to their sedentary nature during nursing periods and immunosuppressive effects of stressful reproductive activities. However, some authors reported that gender had no significant effect in the occurrence of canine ehrlichiosis and babesiosis (Milanjeet *et al.*, 2014 and Aktas *et al.*, 2015).

In the present study, breed wise occurrence was found to be higher in Labrador retriever in canine ehrlichiosis (47.05%), canine babesiosis (36.36%) and 100 per cent in concurrent infection which was in comparable with the study of Gonde *et al.* (2017) on contrary to the Milanjeet *et al.* (2014), Bhadesiya and Raval (2015) and Malik *et al.* (2018). The higher occurrence in Labrador retriever compared to other breeds might be due to poor managerial practices, health status and increased exposure to the tick vectors (Gonde *et al.*, 2017). In our study, all German Shepherd dogs died during the therapy suggesting that these breeds of dogs were more susceptible than the other breeds. The disease was more severe and had a poorer prognosis in our study and this could be due to low cellular immune response in this breed (Milanjeet *et al.*, 2014). The variation in the occurrence of ehrlichiosis among different breeds might be due to more distribution of particular breed in the geographical area and variations in managerial practices in and around Andhra Pradesh.

Conclusion

In the present study the overall occurrence of canine ehrlichiosis and babesiosis was 28.23 per cent and was higher in dogs below 2 years of age (51.43%). The gender wise incidence revealed that male (68.57%) dogs were more prone to canine ehrlichiosis and babesiosis. Similarly the incidence was higher in Labrador retriever (48.5%).

Acknowledgement

The authors are thankful to Sri Venkateswara Veterinary University, Tirupati, Andhra Pradesh for supporting my research work.

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