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Prevalence of protozoan parasitic infections in domestic dogs in Andhra Pradesh, India

C SREEDEVI, CH. JYOTHISREE* and J JAYALAKSHMI

NTR College of Veterinary Science, Gannavaram, Andhra Pradesh 521 101 India

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ABSTRACT

Faecal (2,250) and blood samples (1,350) of dogs received from Teaching Veterinary Clinical Complex (TVCC), Gannavaram, Veterinary Hospitals in and around Krishna district and nearby Krishna, Andhra Pradesh during last five years (January 2014 to December 2018) were examined to determine the prevalence of protozoan parasites. Faecal samples were analyzed by direct smear and floatation technique and blood samples after Giemsa's staining. The overall prevalence of protozoan parasitic infections including intestinal (15.60%) and haemoprotozoan (6.22%) infections was 12.10% (n=435). Six genera of protozoan parasites namely *Cystisospora*, *Entamoeba*, *Ehrlichia*, *Trypanosoma*, *Babesia* and *Hepatozoon* were identified. The most prevalent species was *Cystoisospora* (14.04%) and *Hepatozoan canis* (0.67%) was the least prevalent species. A significant relationship between age group, breed and prevalence of protozoan parasitism was observed. The prevalence of gastrointestinal and haemoprotozoan parasites was significant during winter and rainy seasons, respectively. Sex did not influence the overall prevalence of protozoan parasites.

Keywords: Dogs, Prevalence, Protozoan parasites, Season

Due to their zoonotic potential, endoparasites of dogs represent a great menace to human health in addition to their adverse effects on their primary hosts, canines. Dog population in India is estimated at 25 million and 17.00% of households in India were reported to own a pet dog (Sudarshan *et al.* 2006). Both intestinal protozoa and vector-borne protozoa are one of the important zoonotic endoparasites. The enteric protozoa, viz. *Cryptosporidium*, *Giardia* and *Entamoeba* species are of considerable importance in dogs due to their zoonotic implication especially in immunocompromised hosts (Sharma *et al.* 2017) and are endemic in India (Traub *et al.* 2005). In addition, vector-borne protozoa *Trypanosoma*, *Babesia*, *Ehrlichia*, *Anaplasma*, *Hepatozoon* and haemotropic *Mycoplasma* species have also been reported from dogs in India (Megat *et al.* 2010, Chhabra *et al.* 2013, Singla *et al.* 2016, Jain *et al.* 2017, Patra *et al.* 2018). Comprehensive knowledge on the prevalence of canine parasites is crucial to determine associated risk factors for humans as well as dogs and provides a substantial database to formulate control strategies. Regardless of climatic conditions that are frequently conducive for the transmission of enteric and vector-borne parasitic infections, knowledge on parasitic diseases specifically protozoan diseases of companion animals in India stand insufficient. Though prevalence of gastrointestinal (Das *et al.* 2009, Singh *et al.* 2011) and haemoprotozoan (Kumar *et al.* 2009, Shrivastava *et al.* 2014, Kottadamane *et al.* 2017) parasites has been published from different parts of India, authors' could not discover any published report on canine protozoan parasites from Andhra Pradesh except the age-old report on haemoprotozoa (Bhaskara *et al.* 1986). Owing to these gaps, the present study was aimed to investigate the level of protozoan parasitic infection over the last five years in domestic dogs that had limited access to public places in Andhra Pradesh.

MATERIALS AND METHODS

The study material comprised of 2,250 faecal and 1,350 blood samples of dogs that were received from TVCC, NTR College of Veterinary Science, Gannavaram, Veterinary Hospitals in and around Krishna district and nearby districts of Krishna, Andhra Pradesh during last five years (January 2014 to December 2018). Determination of the prevalence of infection was based both on faecal examination by direct smear and floatation using zinc sulphate solution and on blood smear examination after Giemsa staining (Taylor *et al.* 2016). Identification of protozoan cysts, oocysts and haemoprotozoa was carried out as per the description of Soulsby (1987). Faecal samples positive for coccidian oocysts were sporulated using 2.5% potassium dichromate solution for specific identification and sporulated oocysts were identified based on sporulation time and micrometry (Soulsby 1987). Percentage positivity was estimated from

*Corresponding author e-mail: jyothisreehari@gmail.com

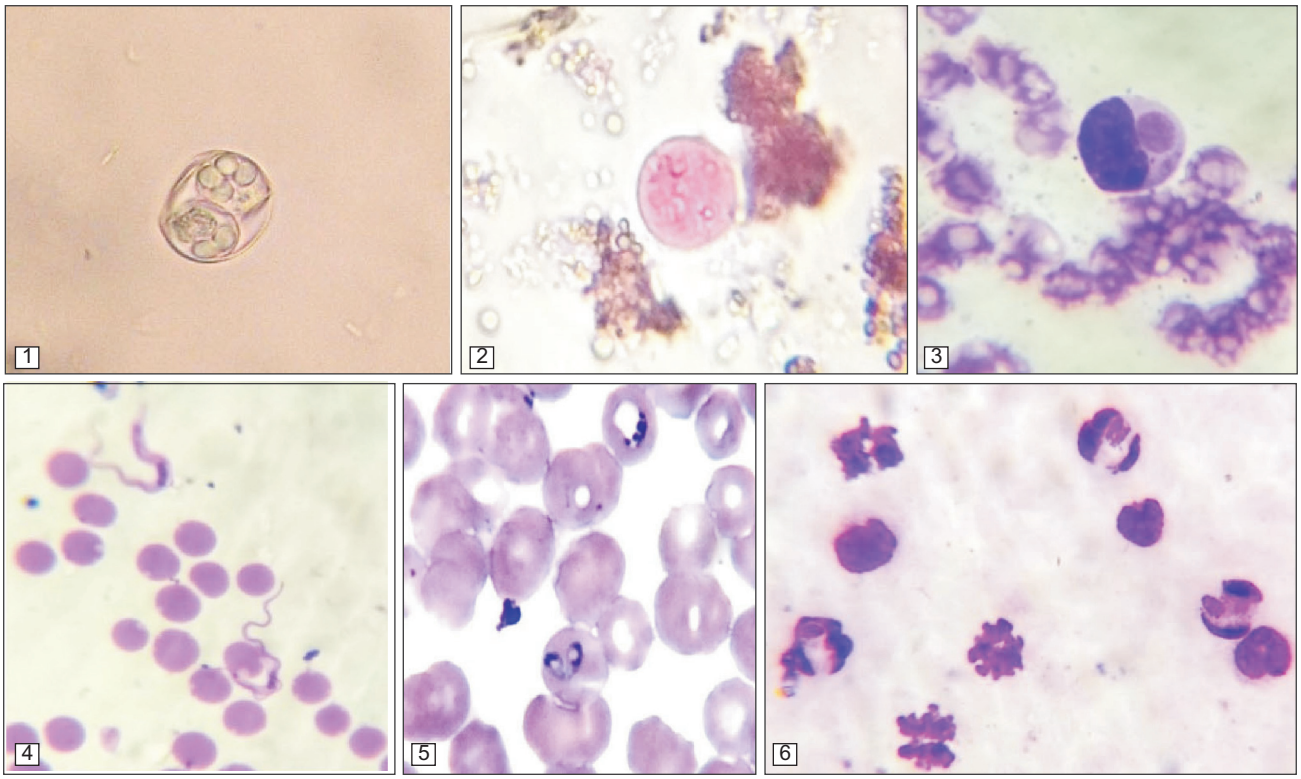
total number of samples (cases) examined. Data obtained was classified according to age (young: ≤ 1 year old and adult: ≥ 1 year old), sex, breed (pure breed; mongrels) and season (summer: March–June; monsoon or rainy: July–October and winter: November–February) and was analyzed as per the standard statistical technique (Petrie and Watson 2013).

RESULTS AND DISCUSSION

Six genera of protozoan parasites namely *Cystisospora*, *Entamoeba*, *Ehrlichia*, *Trypanosoma*, *Babesia* and *Hepatozoon* (Figs. 1–6) were identified in the studied area. On examination of 2250 faecal samples, 15.60% samples were found for intestinal protozoan infection, viz. cysts of *Entamoeba* (1.55%) and oocysts of *Cystoisospora* (14.04%). *Cystisospora* spp. were differentiated based on morphology, sporulation time and morphometry of sporulated oocysts as *C. wallacei* (13.83/8.33 µm; 14–16 h sporulation) and *C. ohioensis* (22.5/18.38 µm; 90–96 h sporulation). Out of 1350 blood smears, 2.74, 1.63, 1.18 and 0.67% were found for *Ehrlichia canis*, *Babesia* spp., *Trypanosoma evansi* and *Hepatozoon canis*, respectively with an overall prevalence of 6.22% haemoprotozoan infection which is low compared to the 11.6% prevalence reported in Tamil Nadu by Kumar *et al.* (2009). The overall prevalence of protozoan parasitic infections including intestinal and haemoprotozoan infections in domestic dogs in the study area was 12.10%. The relatively low prevalence observed in this study ensures that the available veterinary resources are well used by pet owners in the study area and above 60.00% of the dogs included in the study were adults (above one year age).

The results of prevalence according age, sex, breed and season are summarized in Table 1.

A significant (χ^2=222.59, 1 df, P<0.001) relationship between age group and prevalence of protozoan parasitism was observed in the present study. Overall, the prevalence of protozoan parasites was high (22.30%) in below one year age group than in above one year age group (5.61%) which is contrary to the findings of Mahmud *et al.* (2014) who reported highest prevalence in above one year age group. Though gastrointestinal protozoan parasites are generally observed in canine of all ages, the rate of infection is routinely high in young ones, due to the fact that young dogs have not yet acquired immunity to parasites (Ramirez-Barrios *et al.* 2004). The prevalence of gastrointestinal protozoan infection was significantly (χ^2=298.60, 1 df, P<0.001) high in young dogs compared to that of adults. Among two intestinal parasites identified, *Cystoisospora* spp. (30.09%) was the highest prevalent parasite in young dogs that is consistent with age patterns previously stated for this parasite in dogs (Buehl *et al.* 2006) and *Entamoeba* spp. were highly prevalent parasites in adult dogs. Surveys on GI parasitic infections in Pondicherry (Das *et al.* 2009) and Junagadh, Gujarat (Binod Kumar *et al.* 2015) revealed *Cystoisospora* spp. alone along with other intestinal helminths. Similarly, a significant (P<0.05) relationship between age group of dogs and prevalence of haemoprotozoa was observed. The results indicated significantly (χ^2=4.23, 1 df, P<0.05) higher prevalence rate



Figs 1–6. **1.** Oocyst of *Cystoisospora* spp. **2.** Cyst of *Entamoeba* spp. **3.** *Ehrlichia canis* morula stage in monocyte. **4.** *Trypanosoma evansi.* **5.** *Babesia* spp. in RBC. **6.** *Hepatozoon canis* gamonts in neutrophils.

Table 1. Frequency and prevalence of each individual parasite by age, sex, breed and season

	No of faecal samples examined	No. of faecal samples positive			No. of blood smears examined	No. of blood smears positive					No. of dogs positive for protozoa
		E	C	Total		Eh	B	T	H	Total	
Total	2250	35	316	351	1350	37	22	16	9	84	435
		–1.56	–14.04	–15.6		–2.74	–1.63	–1.18	–0.67	–6.22	–12.1
Age											
Young	947	14	285	**299	495	9	6	5	2	22	**321
		–1.48	–30.09	–31.57		–1.81	–1.21	–1.01	–0.4	–4.44	–22.3
Adult	1303	21	31	59	855	28	16	11	7	*62	121
		–1.61	–2.38	–4.53		–3.27	–1.87	–1.29	–0.82	–7.25	–5.61
Sex											
Male	1062	12	152	164	662	15	8	8	2	33	209
		–1.12	–12.7	–15.4		–2.26	–1.2	–1.2	–0.3	–4.98	–31.57
Female	1188	23	164	187	688	23	14	8	7	51	226
		–1.93	–15.44	–15.7		–3.34	–2.03	–1.16	–1.01	–7.41	–32.8
Breed											
Pure breed	1663	23	193	216	1039	24	14	10	6	54	270
		–1.39	–11.6	–12.99		–2.31	–1.35	–0.96	–0.58	–5.2	–10
Pomeranian	816	13	94	107	535	9	6	4	4	23	130
		–1.59	–11.5	–13.1		–1.68	–1.12	–0.74	–0.74	–4.29	–9.62
German Shepherd	539	6	71	77	326	11	5	4	2	22	**99
		–1.11	–13.17	–14.2		–3.37	–1.53	–1.22	–0.92	–6.74	–11.44
Labrador	133	1	16	17	103	2	2	1	0	5	22
		–0.75	–12	–12.7		–1.94	–1.94	–0.97	0	–4.85	–9.32
Others	175	3	12	15	75	2	1	1	0	4	19
		–1.71	–6.85	–8.5		–2.6	–1.3	((0.57)	0	–5.33	–7.6
Mongrel	587	12	123	**135	312	13	8	6	3	30	**165
		–2.04	–21	–23		–4.17	–2.56	–1.92	–0.96	–9.61	–18.4
Summer	477	0	12	12	465	15	4	1	1	21	33
Season			–2.52	–2.52		–3.23	–0.86	–0.22	–0.22	–4.52	–2.2
Rainy	972	26	33	59	381	13	11	11	5	**40	99
		–2.67	–3.39	–6.07		–3.41	–2.89	–2.89	–1.31	–10.49	–7.3
Winter	801	9	271	**280	504	9	7	4	3	23	**303
		–1.12	–33.83	–34.96		–1.79	–1.39	–0.79	–0.59	–4.56	–23.2

in adults than in young dogs (Kumar *et al.* 2009, Shrivastava *et al.* 2014) which could be due to the reverse age resistance phenomena observed in some haemoprotozoan infections and also can be presumed that vector exposure in young dogs could be minor due to better care by the pet owners at this age. Overall, *E. canis* was the highly prevalent haemoprotozoa and *H. canis* was the least among the haemoprotozoa identified. Sex did not influence (χ^2=0.005, 1 df, P>0.05) the overall prevalence of protozoan parasites, though haemoprotozoa infection which was more prevalent in female dogs (χ^2=3.41, 1 df, P>0.05) compared to male. There was significant (χ^2=44.40, 1 df, P<0.001) difference in prevalence between the breeds; the overall protozoan infection being highly prevalent in mongrels compared to that of purebred due to the poor management in former. Similar association was observed with regard to prevalence of intestinal protozoa and breeds (χ^2= 33.07, 1 df, P<0.001). However, though the prevalence of haemoprotozoa was more in mongrels than that of pure breeds the difference was not significant (χ^2=0.147, 1 df, P>0.05). Within pure breeds, the overall prevalence was significantly (χ^2= 19.45, 3 df, P<0.001) high in German Shepherd compared to other pure breed dogs that could be due to the breed susceptibility. Nevertheless, the prevalence of intestinal protozoa (χ^2=3.83, 3 df, P>0.05) and haemoprotozoa (χ^2=2.45, 3 df, P>0.05) between pure breeds was not significant.

Seasonal effect on prevalence of protozoan parasites in the present study revealed that the overall infection rate was significantly (χ^2=246.52, 2 df, P<0.001) higher during the winter season (23.2%) followed by the rainy and summer seasons which could be due to high prevalence of *Cystoisospora* spp. than other identified protozoa, that were frequently noticed during winter season (Table 1). Among different protozoa, the prevalence of gastrointestinal and haemoprotozoan parasites was significant during winter (χ^2=357.00, 2 df, P<0.001) and rainy (χ^2=16.64, 2 df, P<0.001) season respectively. Earlier, Bhaskara *et al.* (1986) observed common haemoprotozoan infections between May to September in Andhra Pradesh. The climate between July to September in Andhra Pradesh is more humid that in turn aid in perpetuation of vectors resulting significant prevalence of vector-borne infection. Sharma *et al.* (2011) and Shrivastava *et al.* (2014) also reported highest prevalence of haemoprotozoan during rainy season in

Mathura and Jabalpur respectively. *Entamoeba* spp. infection was observed during rainy and winter season and was not identified in summer. *Cystoisospora* spp. was observed throughout the year but was persistent in winter. In accordance to the present findings, Binod *et al.* (2015) also reported high prevalence of *Cystoisospora* spp. in dogs in Gujarat during winter.

Results indicated presence of protozoan parasites in the study area emphasizing the need of effective target selective chemo-prophylactic approaches. The data from this study will aid in the development of strategies for the prevention of parasites in pet dogs in study area that successively reduce the risks of environmental contamination and zoonotic transmission. Further, there is a dire need for large-scale studies involving different living conditions of dogs and advanced molecular techniques to get accurate epidemiological data on protozoan parasites.

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