Case Report

Concurrent Trypanosomosis, Babesiosis and Toxocarosis in a Dog: a Case Report

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Abstract

Background: Among the blood-borne parasitic infections of dogs, trypanosomosis and babesiosis are highly prevalent in the Indian subcontinent and highly pathogenic.

Methods: A six-month-old dog was presented with complaint of lethargy, anorexia and loss of body weight over the previous 15 days on October 11, 2021. A full clinical and targeted haematologic and parasitological examination was undertaken.

Results: Wet smears were positive for the motile trypanosomes while stained thin blood smears showed large number of extracellular trypanosomes and intra-erythrocytic ring-like inclusion bodies of *Babesia* spp. A high burden of *Toxo-cara canis* was identified on coprological examination.

Conclusion: This report describes the clinical manifestation of concurrent infection of haemoparasites and gastrointestinal nematodes in Pakistani dogs, and highlights the importance of prompt veterinary intervention.

Keywords: Trypanosoma; Babesia; Toxocara canis; Dog; Pakistan

Introduction

The haemoprotozoan parasites like *Babesia* (*B.*) *canis*, *B. gibsoni* and *Trypanosoma* (*T.*) *evansi* are common causes of life-threatening diseases in wide range of animals including dogs throughout the Indian subcontinent and in other parts of world (1-5). Salivarian trypanosomes are flagellate haemoprotozoa which belong to

family Trypanosomatidae and the genus *Tryp*anosoma (6), and are biologically transmitted by biting flies (7). *Trypanosoma evansi* is widely recognized as a cause of neuropathy, immune suppression and anemia in camels and horses, referred to as surra (8, 9); but adult dogs are also widely infected in Pakistan (10, 11). The

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clinical signs of trypanosomosis in dogs include fever, anemia, superficial lymph node enlargement, loss of appetite, lethargy and progressive loss of bodily condition, leading to fatal outcomes if untreated (12). Babesia spp. are intraerythrocytic parasites, biologically transmitted by Haemaphysalis or Rhipicephalus spp. ticks and infecting a wide variety of hosts including humans as well as domestic and wild animals (13). Many species of Babesia can infect dogs, causing anemia and hemoglobinuria due to haemolysis as the parasite replicates asexually in the red blood cells (14). The parasites complete their life history through sexual reproduction in the tick host mid gut and the formation of sporozoites in the salivary gland (15).

Canine gastrointestinal nematode (GIN) infections, especially Toxocara (T.) canis and Ancylostoma spp., are important causes of retarded growth and general malaise (16). Dogs and humans have a close association (17), and their GINs are of great concern due to their serious zoonotic potential (16). Toxocara canis is distributed worldwide, causing diarrhea, poor growth and death if present in large number (18). Environmental contamination with faeces containing vast number of eggs which shed during patent infections presents a serious public health risk (19), resulting in human disease referred to as visceral larval migrans. This case report presents the co-infection of Trypanosoma spp., and Babesia spp. haemoparasites along with the GIN, Toxocara canis, and describes the therapeutic response to treatment in a dog.

Case presentation

A six-month-old male pet dog (Pointer crossbred) was brought to the outdoor clinic at the Khan Bahadhar Chaudhry Mushtaq Ahmad (KBCMA) College of Veterinary and Animal Sciences, Narowal, sub-campus of University of Veterinary and Animal Sciences Lahore, Pakistan on October 11, 2021. The owner reported that the dog had been anorexic, dull and depressed for the past 15 days, and that the onset of clinical signs coincided with the dog having consumed a putrified dead lamb carcass. On examination, the dog was thin, weak and did not respond well to its surroundings (Fig. 1). The rectal temperature was normal (38.8 °C), oral mucous membranes were pale and the conjunctivae were pale and jaundiced (Fig. 2). There were no swollen lymph nodes, but signs of tachypnoea (38 breaths per minute), coughing and a frothy nasal discharge were observed.

With the consent of the owner, a 5ml blood sample was collected from the cephalic vein into an EDTA coated vacutainer to investigate hematologic parameters and detect any haemoparasites. Sample was collected for coprological examination and processed qualitatively by the faecal floatation technique and quantitatively by a McMaster egg counting method using three-chambered slide having threshold of 8 eggs per gram (EPG) (Chalex®, USA). Wet mounts were prepared for motile trypanosomes and the thin blood smears were stained with Field's stain and examined at 1,000x under oil immersion to detect haemoparasites.

The haematologic parameters (Table 1) showed significantly low packed cell volume (PCV) and haemoglobulin (Hb) values indicating severe anemia, and marked eosinophilia, lymphocytosis and neutropenia. The wet mount of blood sample showed the presence of large number of motile extracellular trypanosomes. The Field-stained thin smear confirmed their identity (Fig. 3) and showed the presence of multiple intracellular *Babesia* spp. with ring-like forms in the red blood cells (Fig. 4).

Toxocara canis eggs were identified on coprological examination, being about $75\mu m \ge 90$ μm in size and having a dark brown colour, a thick pitted shell and sub-globular shape (Fig. 5). An adult worm of *T. canis* was also found in the faeces measuring approximately 18cm in length. The *T. canis* faecal egg count was 2900 EPG, indicating a moderate to high level of infection.

The disease history and clinical signs were consistent with chronic haemoprotozoal infec-

tion and GIN infection (12); hence the dog was treated with intramuscular injection of diminazene aceturate (Pronil[®] Injection, Selmore) at a dose rate of 3.5mg/kg body weight on the first day and same dose was repeated after 72 hours. Doxycycline (Vibramycin[®], Pfizer) was given orally at a dose rate of 5mg/kg of body weight, along with a liver tonic (Iteaspoon per day,



Fig. 1. Dog showing emaciation and persistent recumbency



Fig. 2. Dog showing anaemic conjunctiva and an absence of corneal opacity



Fig. 3. *Trypanosoma* spp. (arrows) on a Field-stained thin blood smear (1000 x magnification) of an infected dog. The genus identity was confirmed by the morphology of extracellular protozoa with a flagellum, kinetoplast and undulating membrane

Hepa-Merz® Syrup, Brookes) and multivitamins (1 teaspoon per day, ViDayLin-L® Syrup, Abbott) for 5 days. A single treatment of anthelmintic 1 tablet per 10kg body weight (Drontal[®] Plus, Bayer) was given. The dog did not respond to supportive therapy and drug treatments, and died 20 days after clinical presentation.



Fig. 4. Erythrocytes showing ring-like forms of *Babesia* spp. (arrows) on a Field-stained thin blood smear (1000 x magnification) of an infected dog



Fig. 5. *Toxocara canis* eggs at 40 x identified on coprological examination of an infected dog

Hematologic parameters	Observed values	Reference values
Erythrocytes (10 ¹² /L)	1.69	5.5-8.5
Hemoglobin (g/dL)	3.70	12-18
Haematocrit (%)	14.0	37-55
Leucocytes (10 ⁹ /L)	17.0	6.0-17
Neutrophils (10 ⁹ /L)	1.50	2.9-12
Eosinophils (10 ⁹ /L)	2.0	0.0-1.3
Granulocytes (%)	41.1	60–70
Lymphocytes (%)	49.7	8.0-21
Platelets (10 ⁹ /L)	33.0	200-500

Table 1. Summary of haematologic parameters of the infected study dog

Discussion

Disruption of the erythrocytes by the release of *Babesia* spp. piroplasms and mechanical injury to erythrocytes by circulating trypanosomes would have accounted for the clinical signs that were observed (20, 21). Eosinophilia, and lymphocytosis are typical in case of trypanosomosis (22, 23).

Trypanosomal infections in dogs have been reported in different countries in Asia (10, 21, 24), southern Europe (25), Africa, north America (26) and South America (27). Salivarian trypanosomiasis in various animal hosts caused by T. evansi, T. brucei, T. vivax, T. congolense or T. theileri, is transmitted by biological vectors such as tsetse fly, and tabanids or *Stomoxys* spp. biting flies, depending upon the Trypanosoma spp. and geographical location (28). In Pakistan, the predominant species is T. evansi, which is considered to be the result of the bites of the flies of family Tabanidae and Stomoxidae, and the molecular studies should be performed to confirm the presence of species of this parasite in animals (29, 30). Babesia canis and B. gibsoni are important causes of disease in dogs with a similar global distribution to Trypanosoma spp. but dependent on the distribution of Dermacentor spp., Haemaphysalis spp. or Rhipicephalus tick vectors. The ring-like morphology of the piroplasms identified in this case are consistent with B. gibsoni, which is particularly pathogenic and prevalent in the Indian subcontinent, causing mainly chronic disease. The treatment response of these cases to antiprotozoal

drugs is generally poor (31). Male dogs are considered more susceptible to *Babesia* infection due to more exposure to tick infestation because of their roaming behaviour in search of mates (32), but the dog in the present case was a pet dog, and the development of infection of *Trypanosoma* and *Babesia* might be due to its being less trypanotolerant and having had less exposure to ticks at a young age (due to provision of more care and hygienic environment as compared to roaming dogs), resulting in less protective immunity. Babesiosis may also be predisposed to immunosuppression caused by *Trypanosoma* spp. (33, 34).

Toxocara canis infection is common in dogs worldwide, and particularly common in the Indian subcontinent (35, 36). Patent infections are usually seen in young puppies, following prenatal placental infection, lactogenic infection, ingestion of eggs or ingestion of a paratenic host. From about 3 months-old, second stage larvae ingested with eggs usually migrate to various tissues, where they become dormant. The high level of infection in a six month-old dog was unusual, and may have been predisposed to by immunosuppression due to trypanosomosis.

Conclusion

The poor response to diminazene aceturate and doxycycline was not unexpected, due to the advanced stage of disease in the dog, and highlights the importance of seeking veterinary advice and treatment immediately when signs of ill health are observed.

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Ethical considerations

With the consent of the owner, a 5ml blood sample was collected from the cephalic vein into an EDTA coated vacutainer to investigate hematologic parameters and detect any haemoparasites.

Conflict of interest statement

Authors declare that there is no conflict of interest.

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