

# GLOBAL JOURNAL OF BIO-SCIENCE AND BIOTECHNOLOGY

© 2004 - 2017 Society For Science and Nature (SFSN). All rights reserved

www.scienceandnature.org

# CLINICO-HAEMATOLOGICAL ALTERATIONS IN BABESIOSIS INFECTED DOGS IN AND AROUND BHUBANESWAR, ODISHA

Charchika Panda<sup>1\*</sup>, Manaswini Dehuri<sup>1</sup>, Mitra Ranjan Panda<sup>1</sup>, Sushen Kumar Panda<sup>2</sup> <sup>1</sup>Department of Veterinary Parasitology, <sup>2\*</sup>Department of Veterinary Pathology College of Veterinary Science and Animal Husbandry, OUAT, Bhubaneswar, Odisha \*Corresponding author email: richu.304@gmail.com

## ABSTRACT

Canine babesiosis is an important tick borne protozoan parasite belonging to *Babesia species* that occurs worldwide. In the current study, a total of 198 blood samples from both stray and pet dogs in and around Bhubaneswar with a history of fever (102<sup>0</sup>F to 106<sup>0</sup>F), anorexia, pale mucous membrane, enlarged lymph nodes and tick infestation were collected and examined during a period of one year. Examination of blood smears revealed an overall incidence of 8.07 %( 16/198). The most prominent clinical symptoms were fever, anorexia, lethargy, and pale mucus membrane. The hematological evaluation of the blood samples of the 16 dogs included Hemoglobin (Hb), Red Blood Cell (RBC), Platelet, Packed Cell Volume (PCV), Mean Corpuscular Volume (MCV), Mean Corpuscular Hemoglobin Concentration (MCHC) and differential counts. Anemia, thrombocytopenia, and monocytosis were the most common hematological alterations observed during the investigation.

KEY WORDS: Babesia sp, Anemia, Incidence, Monocytosis, Thrombocytopenia

### INTRODUCTION

Canine babesiosis is a clinically noteworthy and wellknown hemoprotozoan parasite of dogs worldwide. The commonly occurring Babesia species in dogs are B. canis and B. gibsoni. The life cycle involves gamogony, sporogony and schizogony. Schizogony occurs within the vertebrate host while gamogany and sporogany occur within the tick vector. Once inside the host babesia organisms are attached to the erythrocyte by endocytosis. Due to the parasites invading and replicating in the erythrocyte, babesiosis results in destruction of the erythrocyte. The destruction of the erythrocyte is multifactorial, including direct parasite damage to the infected erythrocyte and antibody-mediated cytotoxic destruction of erythrocytes. The common clinical signs include fever, anorexia, depression, oliguria, haemoglobinurea, vomiting, lethargy, dehydration,pale mucous membrane, icterus, splenomegaly and dyspnea (Irwin, 2005) The various clinical symptoms regularly depend on the severity of the strain involved and host factors, such as age and immunologic response generated against the parasite. In some cases many atypical signs are observed and they cannot be directly explain from hemolysis but appear to be the result of the host immune response.(Andoni et al,2012) Babesia normally causes massive injuries to the host depending on the virulence and pathogenicity of the parasite. The extent of the injuries also depends on the age and the hygiene of the host (Jacobson et al, 1994). The objectives of this study were to describe the clinical signs and hematological alterations in dogs affected with Babesia sp.

## **MATERIALS & METHODS**

The study was conducted in and around Bhubaneswar (Odisha) which is located between 20° 14' 0" North, 85° 50' 0" East and an average altitude of 45 m (148 ft) above sea level. The average relative humidity and annual rainfall are 70% and 1,542 mm (61 in) respectively. During the survey period of 1 year (Aug 2014 - June 2015), 2 ml of blood samples from a total of 98 stray dogs which brought to the Government Veterinary Dispensary, Saheed Nagar, Bhubaneswar for Animal Birth Control (ABC) programme (carried out jointly by the Fisheries and Animal Resources Development Department, Government of Odisha and Bhubaneswar Municipal Corporation under the direct supervision of the Animal Welfare Board of India and CPCSEA for controlling the population of stray dogs) and 100 pet dogs which was presented to the Teaching Veterinary Clinical Complex (TVCC) of the College of Veterinary Science and Animal Husbandry, OUAT, Bhubaneswar belonging to either sex and two different age groups ( below one year age and above one year age) showing clinical signs of progressive anemia, , tick infestation, pyrexia, anorexia or enlarged lymph nodes were collected and stored in EDTA vial. A drop of blood was placed on a clean glass slide drawn into a thin blood smear; air dried, fixed in methanol, stained with Giemsa strain and examined under light microscope by using the oil immersion objective (100X) to detect the parasitaemia. The blood of the positive cases were subjected to different haematological parameters like total leucocyte count  $(10^3/\text{mm}^3)$ , total erythrocyte count  $(10^6/\text{mm}^3)$ , total platelet count (10<sup>3</sup>/mm<sup>3</sup>), PCV (%), haemoglobin (gm/dl), differential count, red cell indices like MCH (pg), MCV(fL), MCHC(%) that were estimated within 4 to 6

hours according to the standard method described by Benjamin (2007).

### RESULTS

Out of 100 pet dogs and 98 stray dogs, overall incidence of canine babesiosis infections by microscopic method showed 16 (8.07%) *babesia* positive cases. The most consistent clinical signs observed during the examination



FIGURE 1. A dog showing heavily tick infested ear

On haematological evaluation, 10/16 of dogs (62.5%) showed RBCs count below reference values, while RBCs count was within reference values in 5/16 of dogs (31.25%). Haematocrit percentage was below reference values in 9/16 of dogs (56.25%), while 7/16 of dogs (43.75%) had haematocrit within reference range. The content of hemoglobin was below normal reference values in 9/16 of dogs (56.25%), while 7/16 (43.75%) of dogs came up with hemoglobin content within normal values. Red cell indices showed an increase in MCV in 6/16 (37.5%) of the dogs, and 3/16 (18.75%) of dogs showed MCV below the normal range with 7/16 (43.75%) of the dogs having MCV within normal reference values.

were elevated rectal temperature (above  $39.6^{\circ C}$ ) in 13/16 (81.25%) of dogs, pale mucous membrane in 10/16 (62.5%) of dogs, anorexia in 13/16 (81.25%) of dogs, Coughing in 6/16 (37.5%) of dogs, vomition in 1/16 (62.5%)of dogs, enlargement of lymph nodes in 11/16 (68.75%)of dogs, lethargy in 12/16 (75%) of dogs, presence of ticks in 9/16 (56.25%) of dogs and haematuria in 1/16 dogs (6.25%)(Fig. 1and 2).



FIGURE 2. A dog showing pale oral mucous membrane

Decrease in MCHC values was noted in 6/16 (37.5%) of the dogs, and an increase in MCHC values in 3/16(18.75%) of dogs with 7/16 (43.75%) of the dogs having MCHC within the normal reference values was noted. Decrease in WBCs count and neutrophils along with an increase in monocyte count were noted in infected dogs. Leukocytes abnormalities include neutropenia in 13/16 of dogs (80%), lymphocytosis in 10/16 (62.5%) of dogs and monocytosis in 10/16 (62.5%) of dogs. Thrombocytopenia is the most consistent hematologic abnormality observed in 100% of the affected dogs. (Table-1)

|                                      |                       | A               |
|--------------------------------------|-----------------------|-----------------|
| Haematological parameters            | Babesia positive dogs | Normal values   |
| RBCs(×10 <sup>6</sup> /              | $4.08\pm0.8$          | 5.5-8.5         |
| Hemoglobin(g/dL)                     | $11.385\pm0.61$       | 12.0-18.0       |
| PCV(%)                               | $32.37 \pm 7.00$      | 37– 55          |
| MCV(fL)                              | $74.71 \pm 13.2$      | 66–77           |
| MCH(pg)                              | $25.28 \pm 5.46$      | 21.0-26.2       |
| MCHC(%)                              | $32.19\pm3.72$        | 32.0-36.3       |
| WBCs( $\times 10^3$ /                | $9.655 \pm 1.4$       | 6,000-17,000    |
| Neutrophil( $\times 10^{3}/_{53/22}$ | $3.790 \pm 0.92$      | 3,000-11,500    |
| Lymphocyte ( $\times 10^3$ /2        | $4.587 \pm 1.33$      | 1,000-4,800     |
| Monocyte ( $\times 10^{3}$ /2003     | $1.069 \pm 0.196$     | 0.1–1.4         |
| Platelets $(\times 10^3/$            | $93.669 \pm 15.0$     | 200,000-500,000 |

TABLE. 1. Alterations of haematological parameters in Babesia positive case

(The references values are given according to Rizzi et al, 2010)

### DISCUSSION

Anemia, thrombocytopenia, and monocytosis were the most common hematological alterations observed in our study, which were in concurrence with reports from other parts of the country and abroad (Shah *et al.*, 2011; Andoni *et al.*, 2012). Anemia was observed in about 62.5% of the positive cases which may be due to the destruction of

circulating red cells by auto antibodies which are directed against infected and non-infected red cell membranes resulting in intravascular and extravascular haemolysis (Day, 1999; Pederson, 1999; Irwin *et al.*, 2005). However Taboada and Lobetti, 2006 proposed that direct parasitic damage contributes to anemia. Nevertheless, induction of serum haemolytic factors increased erythro-phagocytic activity of macrophages and damage induced by secondary immune system after the formation of anti-erythrocyte membrane antibodies are also important in the pathogenesis of anemia. A low RBC count, haematocrit and Hb concentration define anemia. In the present study these parameters were seen in 62.5%, 56.25%, and 56.25% of the positive cases. The reduction of Hb concentration below normal level in 56.25% of dogs in our study probably resulted from reticulocytosis and extravascular haemolysis (Brokus and Andreasen, 2003). Moderate to severe thrombocytopenia is common in canine babesiosis independent of sub species involved (Irwin, 2003). Our findings on consistent presence of thrombocytopenia in all patients corroborates with reports from previous studies (Furlanello *et al.*, 2005 and Zygner *et al.*, 2007).

The main clinical signs observed during the present study were fever, pale mucous membrane, lethargy, and anorexia; these signs were reported by other authors in different studies too (Macintire et al., 2003, Wadhwa et al, 2011). Though the clinical presentation of canine babesiosis can be highly variable, the classical presentation can be safely described as febrile illness with apparent anemia (Schetters et al., 2009). Babesia subspecies can cause different clinical presentations like acute renal failure, respiratory distress and fatal haemolytic anemia (Jacobson et al., 1994 and Gallego et al., 2008). Febrile illness was observed in canine babesiosis-infected dogs; this may be attributed to the release of endogenous pyrogens from erythrolysis, parasitic destruction, and activation of inflammatory mediators (Sakla et al., 1975). The less common signs like haematuria (6.25%) were in accordance to previous reports (Gupta et al., 2002, Varshney et al., 2008, Salem and Farag, 2014).

#### CONCLUSION

It can be concluded that due to its effect on various organs and systems, canine babesiosis shows a wide range of clinical symptoms. The clinical signs and haematological reports should not be neglected, so that the treatment and control of disease is better addressed.

#### ACKNOWLEDGEMENT

The authors acknowledge the co-operation extended by the In-charge of Animal Birth Control Program, Saheed Nagar, and Bhubaneswar for collection of samples, the Dean, C.V.Sc. & A.H., OUAT, Odhisa, India for providing necessary financial and administrative facilities to undertake the study.

### REFERENCES

Andoni, E., D. Rapti, R. Postoli and Zalla, P. (2012) Hematologic changes in dogs naturally infected with babesia. *Albanian Journal of Agricultural Sciences*. **11**(3), 156-158

Benjamin, M.M (2007) Outline of Veterinary Clinical Pathology. 5<sup>th</sup>Edn, pp: 26-37, Kalyani Publishers,New Delhi.

Brockus, C.W. and C.B. Andreasen (2003) Erythrocytes; In Clinical Pathology. Latimer, K.S., E.A. Mahaffey, K.W. Prasse (Eds.), Fourth ed., Ames, pp: 3–45, Blackwell Publishing Company.

Day, M.J.(1999) Antigen specificity in canine autoimmune haemolytic anemia. *Veterinary Immunology and Immunopathology*. **69**, 215–224.

Furlanello, T., Fiorio, F., Caldin, M., Lubas G. and Gallego L.S. (2005) Clinicopathlogical findings in naturally occurring cases of babesiosis caused by large form babesia from dogs of Northeastern Italy. *Veterinary Parasitology*. **134**, 77-85.

Gallego, S.M., M. Trotta , E. Carli ,B. Carcy , M. Caldin and T. Furlanello (2008) *Babesia canis canis and Babesia canis vogeli* clinic-pathological findings and DNA detection by means of PCR-RFLP in blood from Italian dogs suspected of tick-borne disease. *Veterinary Parasitology*. **157(3-4)**, 211–221.

Gupta, S.K., R.K. Bhardwaj, R. Singh, A.K. Raina and A. Kumar (2002) Management of canine babesiosis-A case report. *Indian J. Vet. Med.* **22**, 124.

Irwin, P.J. (2003) Babesiosis in Dogs and Cat. Proc. World Small Anim. Vety. Assoc. Bangkok, Thailand.

Irwin, P.J. (2005) *Babesiosis and Cytauxzoonsis. Arthropode-BorneInfectious Diseases of Dogs and Cats*, 1st edition Manson Publishing Ltd.,Barcelona, Spain,

Jacobson, L.S. and I.A. Clark (1994) The pathophysiology of canine babesiosis: new approaches to an old puzzle. *JS Afr Vet Assoc.* **65**, 134-145.

Macintire, D.K., M.K. Boudreaux, G.D.West, C. Bourne, J.C. Wright and P.A. Conrad (2002) *Babesia gibsoni* infection among dogs in the southeastern United States. *J Am Vet Med Assoc.***220** (3), 325-9.

Pederson, N.C. (1999) A review of immunologic diseases of the dog. *Veterinary Immunology and Immunopathology*. **69**, 251–342.

Rizzi, T.E., Meinkoth, J.H Clinkenbeard. (2010). Normal hematology of the dogs; in Schalm's Veterinary Hematology. Weiss, D.J. and K. J.Wardrop (Eds.), pp. 799–810, Weiley-Blackwell Publishing.

Sakla, AA (1975). Studies on Ticks in Assuit Governorate, with specialreference to their role in transmission of parasitic diseases. Ph.D.Thesis, Faculty of Medicine, Assuit University, Egypt.

Salem, N.Y and Farag H. S (2014) Clinical, Hematologic, and Molecular Findings in Naturally Occurring *Babesia canis vogeli* in Egyptian Dogs. *Veterinary Medicine International*.1.1-6.

Schetters, TP. (2005) Vaccination against canine babesiosis. *TrendsParasitol.* **21(4)**, 179–184.

Schetters, ThPM, J.A.G.M. Kleuskens ,J. Van De Cromment , P.W.J De Leeuw ,A.L. Finizio and A.

Gorenflot (2009) Systemic inflammatory responses in dogs experimentally infected with Babesia canis; a haemaotological study. *Vet Parasitol*.**162**, 7–15.

Shah,S.A., N.K Sood ,S.R. Tumati (2011). Hemato biochemical changes in natural cases of canine Babesiosis. *Asian Journal of Animal Sciences*, **5(6)**, 387-392.

Taboada, J. and R. Lobetti (2006) Babesiosis; in *Infectious Diseases of the Dog and Cat.* Greene, C.G. (Ed.), Elsevier, 3rd edition.

Taboada, J., J.W. Harvey, M.G. Levy and E.B. Breitschwerdt EB (1992) Seroprevalence of Babesiosis in Greyhounds in Florida. *Journal of the American Veterinary Medical Association*. **200**(1), 47-50.

Varshney, J.P., V.V. Deshmukh and P.S. Chaudhary (2008) Multisystemic effects of canine babesiosis and management of critical cases. *Intas polivet*. **9**,281-287.

Wadhwa, D.R., B. Pal, R.K. Mandial, A. Kumar and R.K. Agnihotri (2011) Clinical, haemato-biochemical and therapeutic studies on canine babesiosis in Kangra Valley of Himachal Pradesh. *Journal of Veterinary Parasitology*. **25(1)**, 39-41.

Zygner, W., O. Gójska , G. Rapacka , D. Jaros and H. W drychowicz(2007) Hematological changes during the course of canine babesiosis caused by large *Babesia* indomestic dogs in Warsaw (Poland). *Vet Parasitol*.**145**, 146–151.