Tehran University of Medical Sciences Publication
http://tums.ac.ir

Iran J Parasitol

Open access Journal at
http://ijpa.tums.ac.ir

Iranian Society of Parasitology
http://isp.tums.ac.ir

Case Report

Theileria annulata Induced Bilateral Ocular Signs in Cattle and Its Successful Therapeutic Management: A Case Report

***Anil Kumar [1], Pankaj Kumar [2], Sonam Bhatt [1], Ajeet Kumar [3], GD Singh [4], Ankesh Kumar [4]**

1. *Department of Veterinary Medicine, Bihar Veterinary College, BASU, Patna, Bihar, India*
2. *Department of Veterinary Parasitology, Bihar Veterinary College, BASU, Patna, Bihar, India*
3. *Department of Veterinary Biochemistry, Bihar Veterinary College, BASU, Patna, Bihar, India*
4. *Department of VCC, Bihar Veterinary College, BASU, Patna, Bihar, India*

Received 12 Feb 2023
Accepted 15 Apr 2023

Keywords:
Ocular signs;
Theleria annulata;
Buparvaquone

***Correspondence Email:**
dranilvet95@gmail.com

Abstract

Bovine tropical theileriosis is one of the potentially fatal disease of dairy cattle, which is caused by hemoparasite *Theleria annulata*. About seven years old cross-bred cow was presented with complaint of pyrexia, inappetance, lacrimation and ocular swelling since last 5 days. The clinical examination showed elevated rectal temperature (39.4 ^{0}C), mild enlarged pre-scapular lymph nodes, bilateral bulging of temporal fossa, protruded pale and icteric conjunctivae of the eyes with lacrimation and presence of ticks on body. The case was suspected for haemoprotozoan disease. Blood and serum sample were collected for hematological, blood smear examination and molecular examination (PCR), and biochemical analysis respectively. Microscopic examination of blood smear revealed intra-erythrocytic signet ring shaped periplasm of *Theileria annulata*. Hemato-biochemical examination revealed anemia, hypoproteinemia, hypoalbuminemia and jaundice. Further, PCR assay was done using *T. annulata*-specific primer pair, Cyto b1 gene targeting the amplicon of 312 bp showed specific band on Gel-electrophoresis. Therapeutic regimen was started with Buparvaquone @ 2.5 mg/kg body weight IM single dose followed by Oxytetracycline @ 10 mg/kg body weight IV in 500 ml of NS for 5 days and Prednisolone @ 0.25 mg/kg body weight IM for 3 days along with supportive therapy. The cattle well responded to the therapy and complete regression of ocular signs was observed within one week of treatment.

Introduction

Bovine tropical theileriosis is one of the potentially fatal diseases that frequently affect dairy cattle in India (1). It is caused by *Theileria annulata,* an apicomplexan hemo parasite, mainly transmitted by the bite of an ixodid tick, *Hyalomma anatolicum*. The clinical presentation of tropical theileriosis vary from sub-acute to chronic disease depending on the host susceptibility, the quantity of the inoculated sporozoites and the damaging effect of the pathogen on lymphoid tissue (2). The chief clinical signs are pyrexia, enlargement of superficial lymph nodes lacrimation; nasal discharge, conjunctival petechia, and anemia (3).

In addition to these classic signs, a rare form of *T. annulata* induced ocular signs have been reported in a very limited number of cases (4-6) and hardly recorded in regular practice. The present communication describes a rare case of molecular detection (PCR) and retrieval of ocular lesions in cattle infected with bovine tropical theileriosis.

Case Report

A crossbred cow aged about 7 years was examined on the request of farm owner at Asha Dairy Farm, Bihta, District Patna, Bihar (India) with complaint of pyrexia, inappetance, lacrimation and ocular swelling since last 5 days. The clinical examination revealed elevated rectal temperature (39.4 ^{0}C), mild enlarged pre-scapular lymph nodes, bilateral bulging of temporal fossa, protruded pale and icteric conjunctivae of the eyes with lacrimation (Fig. 1) and presence of ticks on body (Fig. 1). The case was suspected for haemoprotozoan diseases. 2.5 mL of blood was collected from the jugular vein into EDTA vial for hematology, blood smear preparation and PCR assay.

Blood samples were analyzed for hemoglobin (Hb) concentration, red blood cells count (RBCs), packed cell volume (PCV). To confirm the presence of *T. annulata*, PCR was performed. The primers (forward 5'- ACT TTG GCC GTA ATG TTA AAC and reverse 5'- CTC TGG ACC AAC TGT TTGG) were used for the amplification of 312 bp fragment of the cyto b1 gene in *T. annulata* (7). A total of 2.5 ml of blood were also collected in anticoagulant free tubes and sera were separated. The serum samples were subjected to biochemical analysis including total protein, albumin, alanine aminotransferase (ALT), and serum bilirubin in an auto analyzer using commercially available kit.

In the present study, examination of Giemsa stained blood smear under oil immersion objective of microscope showed signet ring shaped piroplasm of *Theileria* spp. in erythrocytes (Fig. 2). The amplification of targeting 312 bp amplicon confirmed the presence of *T. annulata* infection (Fig. 3). Hematological investigations revealed hemoglobin 7.4 g/dl, packed cell volume 21%, and red blood cells count 4.72x10^{6}/ µl, indicating anemia in the affected cow. Furthermore, the results of biochemical analysis revealed total protein 5.2 g/dl, albumin 2.12 g/dl, ALT 48 U/l, and total serum bilirubin 0.98 g/dl.

Based on the microscopic and molecular detection, the treatment was initiated with Buparvaquone @ 2.5 mg/kg body weight IM single dose followed by oxytetracycline @ 10 mg/kg body weight IV in 500 ml of NS for 5 days, and Prednisolone @ 0.25 mg/kg body weight IM for 3 days along with supportive therapy as antioxidants (E-care-Se), Hematinic (Sharkoferrol Vet) @ 50 gm daily for two weeks. The cattle responded to the therapy and clinical improvement was noticed on second day of treatment, like normal body temperature and appetite, and the complete regression of ocular signs was observed within one week of treatment (Fig. 4).

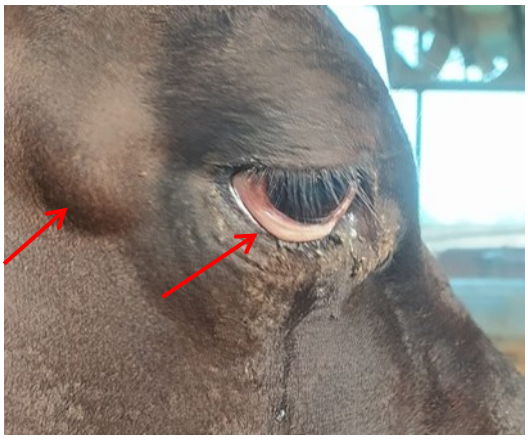

Fig.1: Bulging of temporal fossa and protruded pale and icteric conjunctivae of the eyes with lacrimation

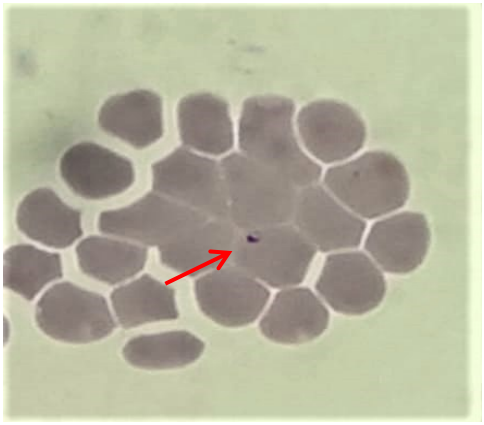

Fig. 2: A typical signet ring shaped *T. annulata* piroplasm in erythrocytes (x1000, Arrow)

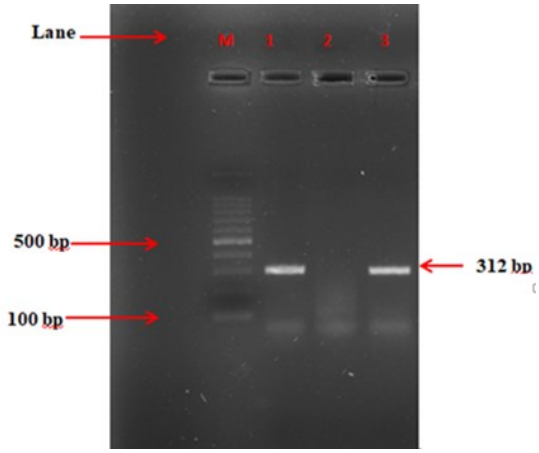

Fig. 3: Targeting 312 bp fragments of *T. annulata* Lane 1: Positive control; Lane 2: Negative control & Lane 3: Positive sample

Fig. 4: Complete regression of ocular signs

Discussion

Buparvaquone @2.5 mg/kg body weight intramuscularly is an effective compound for the therapy of all forms of theileriosis (2). Antibiotic, oxytetracycline was used as an adjunct because of its antitheilerial activity and to cure anaplasmosis, which may found concurrently (8). The excess pro-inflammatory cytokines are responsible for pathologies associated with tropical theileriosis (9). The good response of injection prednisolone might be due to its anti-inflammatory action and inhibitory effects on pro-inflammatory cytokines (10). The adjunction of antioxidants and or oral hematinic in the present case study and anti-theilerial agents could help in fast clinical recovery from theileriosis.

Ocular and nodular lesions are unusual clinical findings in tropical theileriosis (11), and most

frequently observed in young calves. Ocular signs in the present case study (Adult Cattle) might owing to accumulation of lymphoblastoid cells in periorbital tissues (4) and overproduction of TNF-a (2). The fall in hematological values was indicative of anemia, which might be due to destruction of erythrocytes by macrophages in lymph nodes, spleen and other organs of reticuloendothelial system (12). The anemia is considered associated with immune-mediated hemolytic anemia (13).The hyperproteinemia and hypoalbuminemia is possibly due to the harmful effect of toxic metabolites of *Theileria* and due to liver failure (14). The increased total serum bilirubin might result from the destruction of parasitized erythrocytes by erythrophagocytosis in the spleen, lymph nodes and other organs of the reticuloendothelial system (15).

The most common clinical signs of tropical theileriosis in cattle are pyrexia, enlargement of superficial lymph nodes, lacrimation, and nasal discharge, petechial of conjunctival mucous membranes, and anemia. However when encountering symptoms such as ocular signs (protrusion of eye conjunctiva) and bulging of temporal fossa, veterinarians should consider Tropical theileriosis in the differential diagnosis.

Acknowledgements

The authors wish to thanks Dean, Bihar Veterinary College, BASU, Patna, for the facilities provided.

Conflict of interest

The authors declare that there is no conflict of interest.

References

1. Tuli A, Singla LD, Sharma A, Bal MS, Filia G, Kaur P. Molecular epidemiology, risk factors and hematochemical alterations induced by *Theileria annulata* in bovines of Punjab (India). Acta Parasitol. 2015; 60:378–390.
2. Verma AK, Singh SK. A rare case of *Theileria annulata* induced corneal opacity in a calf. J Parasit Dis. 2016; 40: 208–21.
3. Aulakh GS, Singla LD. Clinico-haematobiochemical observations on bovines naturally infected with *Theileria annulata*. J Vet Parasitol. 2006; 20: 49–52.
4. Aslani MR, Mohri M. Ocular manifestations in cattle due to tropical theileriosis. J Vet Parasitol. 2002; 16: 53-54.
5. Sudan V, Sharma RL, Borah MK, Mishra R. Acute bilateral proptosis in a cross bred calf naturally infected with *Theileria annulata*. J Parasit Dis. 2012; 36: 215-9.
6. Sharma S, Kolte SW, Magar S. Exopthalmia in cross bred cattle naturally infected with *Theileria annulata*. Int J Curr Microbiol App Sci. 2020; 9(9): 493-497.
7. Bilgic HB, Karagenc T, Simuunzm A, et al. Development of a multiplex PCR assay for simultaneous detection of *Theileria annulata*, *Babesia bovis* and *Anaplasma marginale* in cattle. Exp Parasitol. 2013; 133:222-9.
8. Muhammad G, Saqib M, Athar M, Khan MZ, Asi MN. Clinico-epidemiological and therapeutic aspects of Bovine theileriosis. Pak Vet J. 1999; 19: 64–71.
9. Glass EJ, Craigmile SC, Springbett A, et al. The protozoan parasite, *Theileria annulata*, induces a distinct acute phase protein response in cattle that is associated with pathology. Int J Parasitol. 2003; 33:1409-18.
10. Aiello SE. The Merck Veterinary Manual. 11th edn. Merck & Co., Inc. Kenilworth, NJ, USA, 2016.
11. Tumer KC, Karabulut B, Ulucesme MC. Cutaneous and ocular signs in a calf infected with *Theileria annulata* -case report. Vet J KU. 2022; 1: 26-33.
12. Kaur R, Yadav A, Rafiqi SI, et al. *Theileria annulata* infection in a cross bred cattle with corneal opacity– a case report. Haryana Vet. 2020; 59: 305-307.
13. Alsaad KM, Suleiman EG, AL-obaidi QT. Theileriosis in newborn calves in Mosul, Iraq. Bas J Vet Res. 2013; 12: 265-274.
14. AL-emarah GHYA, Khudor MH., Daham HR. Clinical, haematological and biochemical study to cattle naturally infected with *Theileria annulata* in north of Basrah province Al-Qadisiya. J Vet Med Sci. 2012; 11, 54-62.
15. Khan IA, Khan A, Hussain A, Riaz A, Aziz A. Hemato-biochemical alterations in cross bred cattle affected with bovine theileriosis in semi-arid zone. Pak Vet J. 2011; 31:137-140.