

THEILERIA ANNULATA INFECTION IN A CROSS BRED CATTLE WITH CORNEAL OPACITY– A CASE REPORT

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SUMMARY

Three years old cross bred cow was attended during an animal health camp in Jammu with history of recent transportation from Gurdaspur district of Punjab. Clinical examination of the animal revealed high fever (104.5 °C), anorexia, pale mucous membrane, bilateral corneal opacity, lacrimation and swollen pre scapular lymph nodes, but no ticks were observed on the body. Haematological examination revealed significantly low count of red blood cells, haemoglobin and packed cell volume indicative of anaemia. Based on clinical signs and microscopic examination of blood smear after Giemsa staining, it was diagnosed as bovine tropical theileriosis. The infection was further confirmed as *Theileria annulata* by polymerase chain reaction (PCR) assay targeting Tams1 gene with amplification product of 751 bp.

Keywords: Corneal opacity, *Theileria annulata*, Theileriosis

In the Indian subcontinent, tropical theileriosis is one of the most prevalent diseases of bovine caused by *Theileria annulata*, involving *Hyalomma anatolicum* as vector (Mirzaei, 2007). *T. annulata* is very pathogenic parasite of dairy animals especially exotic dairy animals and their crosses with indigenous breeds (Hashemi-Fesharki, 1988) in which the disease accounts for high morbidity and mortality in cattle. In India, the annual loss reported due to tropical theileriosis is estimated to be US\$1295 million per annum (Narladkar, 2018).

Three years old cross bred cow was attended during an animal health camp at R.S. Pura, Jammu. Clinical examination revealed high fever (104.5 °C), laboured breathing, bilateral corneal opacity (Fig. 1), lacrimation, swollen lymph nodes and anaemia of the animal. However, no tick was observed on the body of the animal as acaricidal application was done by the animal owner one week earlier. The owner reported that animal was purchased 15 days back from Gurdaspur district of Punjab. Blood sample was collected from the jugular vein in vials containing EDTA for haematology, microscopic and molecular examination. Thin blood smear was prepared, air dried, fixed in methanol for 30 seconds and stained with Giemsa stain (1:10) and examined under oil immersion objective of microscope, where signet ring shaped piroplasm of *Theileria* spp. were recorded in erythrocytes (Fig. 2). Genomic DNA was extracted from 200 µl of the whole blood using DNA extraction kit (DNeasy blood kit, Qiagen) and 25 µl reaction mixture was subjected to PCR

assay targeting Tams 1 gene of *T. annulata*. The reaction mixture, primers (forward 5'- CCGTTAATGCTGCAAAT GAGGAGG and reverse 5'- GAGGCGAAGACTGC AAGGGGAG) and cycling conditions used for PCR were as described by Kundave *et al.* (2017). Amplification of 751 bp amplicon (Fig. 3) confirmed the presence of *T. annulata* infection. Haematological analysis using autoanalyser revealed red blood cells count $5.86 \times 10^3/\mu\text{l}$, white blood cell count $14.3 \times 10^3/\mu\text{l}$, haemoglobin 8.1 g/dl, mean corpuscular volume 54.3 fl, mean corpuscular haemoglobin 17.6 pg, mean corpuscular haemoglobin concentration 32.4 g/dl, platelet count $257 \times 10^3/\mu\text{l}$ and packed cell volume 22% indicating anaemia in the animal.

Based on the microscopic and molecular diagnosis, the animal was treated with buparvaquone @ 2.5 mg/kg body weight intramuscularly (i/m) once and meloxicam and paracetamol 25 ml i/m, chlphenarmine maleate 10 ml i/m and bolus containing Iron Sorbitol, Hydrocobalamin and Folic acid @ 1 bolus/day for 10 consecutive days as supportive therapy. The animal positively responded to above integrated therapy and recovered after 10 days post treatment. However, the corneal opacity did not resolve on the day of observation (10 days post treatment), which is not unlikely, as it takes 2-3 weeks to resolve completely (Mohan *et al.*, 2009). Further on subsequent visit, it was confirmed that corneal opacity had completely resolved by day 25 post treatment.

In the present case, corneal opacity was observed in *T. annulata* infected animal, although rarely occurs due to invasion of cornea and lens by leucocytes (Abdel-Rady *et*

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Fig. 1. Animal showing corneal opacity

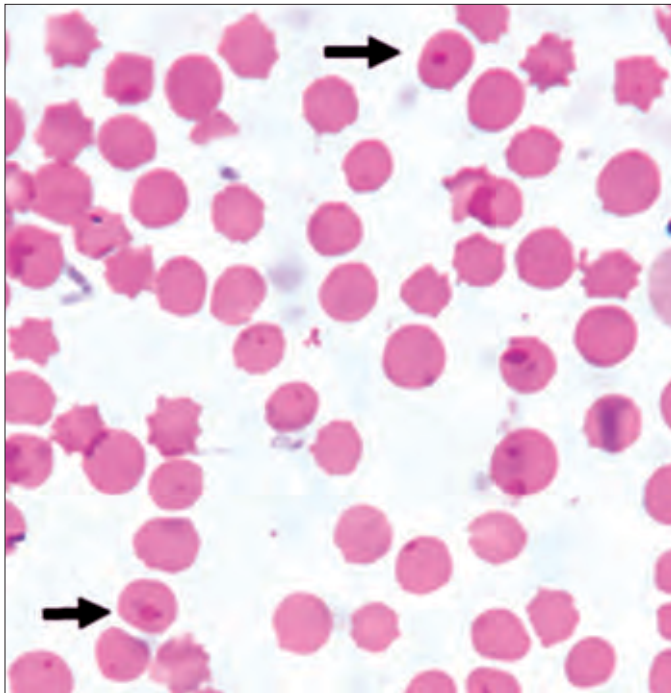


Fig. 2. *Theileria annulata* infected RBCs of cattle (x1000)

al., 2008) has been earlier reported (Joshi *et al.*, 2017). The fall in haematological values indicative of anaemia is mainly due to destruction of erythrocytes by macrophages in lymph nodes, spleen and other organs of reticuloendothelial system. The current case of *T. annulata* in cross bred cattle at Jammu, transported from Punjab indicates high probability of introducing a new infection to a geographical

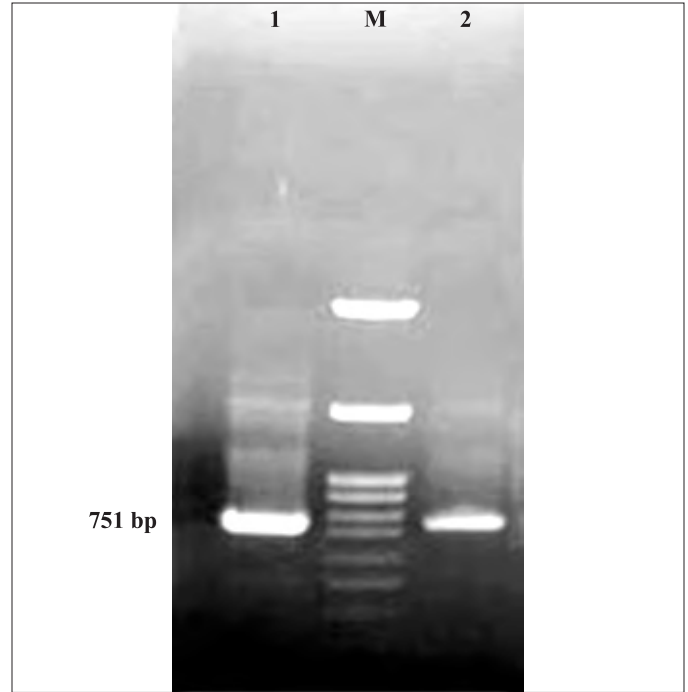


Fig. 3. Agarose gel electrophoresis of PCR amplified *T. annulata* DNA
Lane1: Positive Control. Lane2: Test Sample, M: 100bp DNA Ladder

region. Although sporadic cases (Gupta *et al.*, 2004) of *T. annulata* infection have been recorded in Jammu region, no established prevalence of theileriosis in bovines has been reported from Jammu region of Union Territory of Jammu and Kashmir till date. Livestock farmers of Jammu and Kashmir are procuring high yielder animals from neighbouring states like Punjab having high prevalence of *H. anatolicum* (Singh and Rath, 2013) and *T. annulata* in cattle (Tuli *et al.*, 2015). The translocation of animals from Punjab may inflict consequences which may be hard to predict. It is not known if the vector tick could adapt, survive and propagate in its new geographical environment. Thus, translocation of animals may carry pathogen to new susceptible population. Hence, an urgent need is advocated at government level and to educate farmers regarding awareness of theileriosis as well as cow-side diagnostic test for screening animals at entry point of Jammu and Kashmir for theileriosis prior to transboundary migration.

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