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## RISK factors associated with Hemato-biochemical changes in cross bred cows with *T. annulata* infection

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### Abstract

Few crossbred cows from an organized livestock farm in Jamdoli, Jaipur were reported with the clinical signs of anorexia for a period of 2 weeks, high fever, low milk yield, conjunctivitis and profuse nasal discharge. Clinical examinations of the above animals confirmed the above clinical signs in addition to lymphadenopathy, and blood smear examination of the animals confirmed *Theileria annulata* infection in 6 crossbred cows. The farms having 26 crossbred cows were screened for theileria infection since six animals were in the above clinical signs for more than 2 weeks period. Blood smear examination revealed Theileria infection. Hence these cows were evaluated for their hemato-biochemical parameters for *T. annulata* infection. In these crossbred cows significant low level of RBC count, Hb content, WBC, PLT, lymphocytes, granulocytes, and significant high levels of MCH were observed. Bio-chemical studies in crossbred cows revealed significant high values of ALT, AST values, total bilirubin, total protein, & globulin and insignificant low level of A: G ratio & calcium by the *T. annulata* infection in comparison with normal healthy crossbred cow values. The risk factors associated for hemato-biochemical changes in the *T. annulata* infected crossbred cows are discussed for further clinical management of the affected cows.

**Keywords:** Hematology-biochemical- Cross bred- Theileriosis

### 1. Introduction

Theileriae are obligate intracellular protozoan parasites that infect both wild and domestic bovidae throughout the world. The ticks playing major role in transmission of the disease (*OIE* 2014) [27]. There are a number of species of theileria that infect cattle; the two most pathogenic and economically important parasites are *T. parva* and *T. annulata* (Bhat nagar, C.S., *et al.* 2015; Demessie, Y and S. Derso 2015; Gul, N., *et al.* 2015; *OIE* 2014; Gebrekidan, H., *et al.* 2016) [5, 6, 17, 27, 13]. *T. annulata* occurs in Southern Europe as well as North Africa and Asia (*OIE* 2014) [27]. Traditional diagnosis of bovine theileriosis is mainly based on the microscopic examination of blood smears for the presence of the merozoites of theileria (Junlong, L., *et al.* 2015) [20]. The severity of infection in theileriosis is indicated by the changes in blood serum biochemical parameters (Mahmoud Rushdi Abd Allah *et al.* 2015) [23]. In ruminants, hematology and serum biochemical analysis are relevant for detecting hematological disorders.

### Breed differences in susceptibility to *T. annulata* infection

Exotic cattle breeds are extremely susceptible to tropical theileriosis caused by *T. annulata*. The infection induces severe, and often fatal, diseases in susceptible *Bos Taurus* and cross-bred cattle; (Spooner RL, *et al.* 1991) [38]. The rate of WBC reduction was significantly greater in the Friesian calves. Kenana cattle, 78% recovered spontaneously, and only 22% required treatment compared to 100% mortality in the Friesian controls. The macrophages in *B. indicus* breed are able to control the cytokine storm, where it is devastating in *B. Taurus* breeds. (Glass EJ *et al.* 2007) [14].

Different breeds of cattle are different in their susceptibility to theileriosis. Saeed, Z *et al.* (2016) [33] reported that the exotic and cross bred cattle are highly susceptible, while indigenous cattle are relatively resistant to tropical theileriosis. The prevalence studies on bovine theileriosis in indigenous animals and crossbred animals were 13.33% & 26.66% respectively for *T. annulata* infection in Rajasthan state.

Nejash A. *et al.* (2016) [25] reported that *Bos indicus* cattle are more resistant to ecto-parasites than *Bos Taurus* animals and claimed differences between these two breeds of cattle in regard to their susceptibility to parasitism by cattle ticks. Shashi Choudhary *et al.* (2022) [36] reported that the indigenous cattle were less prone than cross bred cattle and upgraded murrah buffaloes. Ayadi. Q *et al.* (2017) [3] reported that indigenous breeds are more resistant to *T. annulata* infection than exotic breeds such as Friesian and Holstein cattle (Gharbi *et al.* 2014; Saleem *et al.* 2014) [12, 35]. This difference in sensitivity is attributed to the difference in the immune response to produce pro-inflammatory cytokine which is higher in exotic breeds (Glass *et al.* 2005) [7]. Montbeliard reported *T. annulata* infection with normocytic normochromic anemia in cross bred cattle. Keeping in view of the above facts this study was carried out to determine the theileriosis in crossbred cows (*Bos taurus*) for hemato-biochemical parameter alterations under farm conditions.

### Materials and Methods.

The study was carried out in a livestock farm in Jamdoli, Jaipur. Few crossbred cows from this livestock farm were reported with the clinical signs of prolonged anorexia, fever, low milk yield conjunctivitis, profuse nasal discharge. Clinical examinations of the animals were carried out in these animals for confirmation of the above clinical signs and the infection of the *Theileria annulata* by blood smears examination. Blood smear examinations revealed *Theileria* infection in 6 crossbred cows, out of 29 crossbred cows in this livestock farm. Hence these cows were evaluated for their hemato-biochemical parameters for confirmation of the infection.

### Results and Discussion

**Table 1:** Hematological values in crossbred cows with theileriosis

Parameters	Crossbred cows mean values	Crossbred healthy cows range values
RBC (106/UL)	3.68	6.0-7.43
Hb (g/dl)	7.7	8.0-12.0
WBC (103/UL)	7.81	8.0-10.0
PLT (103/UL)	202	300-800
LYM (%)	52.91	55.0-60.0
Granulocytes (%)	13.50	15.0-45.0

### Hematological studies in cross bred cows with theileriosis. (Table. 1)

In this study the erythrocytes and the Hb content were in significant low levels. Anemia occurs in *T. annulata* infection due to destruction of erythrocytes (Uilenberg G *et al.* 1981) [40] in addition to pro inflammatory cytokines involvement (Graham SP *et al.* 2001) [16] Ganguly *et al.* (2015) [10] reported a normocytic hypochromic anemia in *T. annulata* infection in crossbred cattle. The anemia could be due to both an auto immune reaction (Hooshmand-Rad (1976) [18] and the effect of intra- erythrocytic piroplasms (Preston *et al.* 1992) [31]. Emad A.H *et al.* (2020) [8] reported significant reduction in RBCs count, Hb content, and MCH values in the affected cross bred theileriosis animals which indicates normocytic hypochromic anemia. Radostitis OM *et al.* (2008) [32] reported low level RBC, hemoglobin, PCV, TEC due to toxic metabolites of *Theileria* infection. Pandey V *et al.* (2017) [30] reported leukocytosis due to blood sucking ticks. Omer *et al.* (2002) [28] reported that the decreased level of erythrocytes

could be attributed to increased levels of activated complement products.

In this study a significant low level of lymphocytes count was observed Gaurav Charaya *et al.* (2021) [11] reported lymphocytosis in their studies in the crossbred cows in the initial days of infection (Somu *et al.* 2017; Abdel- hamied *et al.* 2020) [39, 1] followed by decreased lymphocytes in the later stages. (Ugalmugle *et al.* 2010; Modi *et al.* 2015) [41,24]. Nilima *et al.* (2022) [26] reported decreased level of lymphocytes due to RBCs damage and its multiplication. Feldman *et al.* (2000) [9] reported destruction of lymphocytes in lymphoid organs causes lymphopenia due to lysis of lymphocytes caused by release of protozoal merozoites in to blood stream. These observations endorse the present findings of this study. In this study the WBC count was insignificantly at low level in the crossbred theileria infected cows. Hussein *et al.* (2007) [19] opined that it could be due to large scale destruction of leucocytes by the protozoa and protozoa toxic metabolites. Bakheit and Latif (2002) [4] reported that the reduction of WBC was significantly greater in the Friesian calves, which is agreement with the present findings.

In this study the (platelet total) PLT count was at significant low level. Mahmoud Rushdi Abd Allah *et al.* (2015) [23] reported significant decreases in the PLT, & PCT values which indicated anemia and is attributed to the thrombocytopenia together with normocytic normochromic anemia and partially attributed to depression of the bone marrow. Ayadi .Q *et al.* (2017) [3] also reported normocytic normochromic anemia in cross bred animals. These observations are in the same line of this study. In this study significant decrease in the total RBC counts, hemoglobin concentration, platelet count, suggesting normocytic normochromic anemia in cross bred calves infected with *T.annulata*. This study revealed insignificant low level granulocytes count and the lymphocyte percentage was maintained at normal level. Hussein *et al.*, 2007 [19]; Abd el-Hamed *et al.* (2016) reported reduction in TLC and neutrophils with a significant elevation in lymphocytes and monocytes due to large scale destruction of leucocytes in theileriosis by toxic metabolites. Onyinyechukwu Ada Agina *et al.* (2021) [29] also reported that the granulocyte macrophage-colony stimulating factor was found to be present in high amounts in the serum of the blood parasite infected cattle groups with or without clinical signs of the disease. This indicates a possible response to low white blood cell count in the blood-infected cattle which was in the same line of observation of this study.

**Table 2:** Biochemical values of cross bred cows with theileriosis.

Parameters	Cross bred cows mean values	Healthy cross bred cows values
AST (SGOT)(UL)	122.19	55-60
Creatinine (mg/dl)	1.56	1.0-2.0
Urea (mg/dl)	36.47	20-30
Total Bilirubin (mg/dl)	0.925	0.01-0.15
Total protein (g/dl)	8.91	6.0-8.0
Albumin (g/dl)	3.15	3.0-3.5
Globulin (g/dl)	5.75	3.0-3.4
A: G ratio	0.601	0.80-0.94
Calcium (mg/dl)	6.47	9.0-12.0

### Bio-chemical studies in crossbred cows with theileriosis (Table.2)

The crossbred cows in this study showed significant high level changes in ALT values. Singh *et al.* (2001) [37] reported

high level AST in the animals' infected with Theileriosis. Omer *et al.* (2003) [28] and Kachhawa *et al.* (2016) [21] attributed the increased levels of AST due to hepatic injury of theileriosis infection. Latimer (2011) [22] attributed increased RBC lysis also a reason for elevated AST. These observations are in concurrence with the present study.

The level of creatinine was maintained and significantly altered urea level was observed in these cows which were endorsed by Somu *et al.* (2017) [39] who reported elevation of urea and creatinine in theileriosis infected cattle which were partially agreed by this study. The total bilirubin elevated to a significant level in these cows. Sandhu *et al.* (1998) [34] and Singh *et al.* (2001) [37] reported elevated indirect bilirubin values due to increased destruction of infected erythrocytes, and an elevated direct bilirubin values due to hepatic dysfunction and hemolysis in diseased cows which was in the same line of observations of this study.

Total protein was elevated in this infected crossbred cow to a significant level in this study. Elizabeth J. Glass *et al.* (2005) [7] reported prolonged production of acid glycoprotein in the Holstein cattle due to chronic production of circulating pro-inflammatory cytokines in tropical theileriosis. This observation coincides with this study. However decreased level of total protein values was reported by many authors. Emad A.H *et al.* (2020) [8] reported significant reduction in total protein and decreased serum albumin in cattle due to impaired liver function. In this study no alterations in albumin level noticed. Aulakh G S *et al.* 1998; Ghosh *et al.* 2007) [2], [15] reported that total protein and albumin becomes lower due to the harmful effective of toxic materials in *Theileria* infection due to hepatic deficiency.

Serum globulin levels were elevated in these cows which could be due to the body immune response for *Theileria* infection as suggested by Singh *et al.* (2001) [37]. The A: G ratio was insignificantly low in these *Theileria* infected crossbred cows. Onyinyechukwu Ada Agina *et al.* (2021) [29] also reported *T. sinensis* affected cattle with decreased A: G ratio due to alteration in liver function. This observation confirms the outcome of this work. Calcium level decreased to a significant high level in this study. Vural D *et al.* (2017) [42] reported decreased calcium level in *Theileria annulata* infected animals due to decreased dietary intake, intestinal malfunction, kidney damage which was in correlation with the finding of this work.

## Conclusions

*T. annulata* infection in crossbred cows creates changes in hematological and biochemical parameters. The changes varies from one region to another and the parameters getting influenced by the characteristics such as the innate immune response, the dose and virulence of the strain of *T. annulata* in each country. The observations presented in this paper can be taken as criteria for diagnosis and interpretation of results of *T. annulata* infection in crossbred cows of India for better management.

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