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Assessment of oxidative stress in cattle infected and subsequently recovered from theileriosis

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Abstract
The present study was conducted to elucidate the lipid peroxidation level, antioxidant status and haemato-biochemical alteration in cross bred suffering from tropical theileriosis. An organized farm of 40 cross bred cattle with a history of theileriosis was screened for disease on the basis of clinical signs and conventional microscopy. All the animals were clinically examined for mucous membrane, rectal temperature and swelling of lymph nodes. Seven animals found positive for characteristics piroplasm on microscopic examination of Geimsa stained smear and showing typical signs of disease were considered positive whereas fifteen other animals not showing signs and negative in blood smear was considered as control group. Lipid peroxidation was evaluated by calculating plasma Malondialdehyde levels and antioxidant status was evaluated by estimating plasma superoxide dismutase and plasma glutathione reductase. MDA level and SOD activity was found significantly increased whereas GSH activity was found to be significantly increased clearly indicating theileriosis impart oxidative stress. Following therapy with buaparvaquone non-significant changes were observed as no antioxidant was added in therapy. Therefore, it can be concluded that parasite impart oxidative stress and to decrease the level antioxidants must be added along with drug of choice.

Keywords: tropical theileriosis, malondialdehyde, superoxide dismutase, glutathione reductase

Introduction
Theileriosis is an important haemoprotozoan disease caused by intracellular haemoprotozoan *Theileria annulata*. In India, annual losses due to theileriosis are estimated to be Rs. 8426.7 crore (Narladkar, 2018) [6]. The main clinical signs shown by affected cattle are fever, pale mucous membrane and swollen lymph nodes which indicate about the disease. For confirmation of disease status, microscopic examination of Giemsa stained blood smear is considered as Gold standard.
During the part of life cycle inside the erythrocytes, merozoites metabolize hemoglobin and produces free radicals which enhance oxidative stress in the infected animals. Recently, many reports (Grewal *et al*., 2005, Rezaei and Dalir-Naghadeh 2006, Razavi *et al*. 2015) [3, 9, 10] have suggested that parasite enhances the oxidative stress by either leading to changes in erthrocytic membranes or by altering the anti-oxidative enzyme activity. Moreover, in past antioxidant used along with drug of choice (buparvaquone) has shown far better recovery than drug used alone (Nayak *et al*., 2018) [7]. Keeping the above in view, the present study has been planned to assess oxidative stress in animals infected and recovered of these diseases.

Material and Methods
Sample collection
A total of 40 blood samples collected from Tokas farm were screened for haemoprotozoan diseases on the basis of clinical signs and microscopic examination. Plasma values of MDA, GSH and SOD of seven cattle found positive for *Theileria annulata* on microscopic examination and showing clinical signs were compared with values of 15 apparently healthy animals values. These animals were given treatment with buparvaquone and plasma samples were again collected at day 3 to determine the impact of therapy on oxidative stress.

Calculation of oxidative stress parameters
Malondialdehyde (MDA) estimation: Malondialdehyde (MDA) in plasma was estimated following protocol explained by Shafiq-ur-Rahman *et al*. (1984) [12]. This test is based on the reaction of malondialdehyde (MDA) with thiobarbituric acid (TBA).

The MDA-TBA complex (TBARS) thus formed showed absorbtion strongly at 535 nm.

GSH activity: GSH activity in plasma was measured by method of Beutler *et al*. (1963) [1]. This method depends on the reaction of reduced glutathione with 5, 5-dithiobis, 2-nitrobenzoic acid (DTNB) to form yellow derivative 5'-thio-2-nitrobenzoic acid (TNB) that can be measured spectrophotometically at 412 nm.

Superoxide dismutase (SOD) activity: Superoxide dismutase (SOD) was estimated as per the method described by Madesh and Balsubramanian, (1998) [4]. It involves the generation of superoxide by pyrogallolautooxidation and inhibition of superoxide dependent reduction of the tetrazolium dye MTT [3-(4-5 dimethyl thiozol 2-yl) 2,5-diphenyl tetrazolium bromide] to its formazen, which is measured at 570 nm.

Statistical analysis

Comparison was done using SPSS 22.0 statistical software. Comparison between values of diseased to that of apparently healthy animals was done by applying Independent 't' test and to compare values of day 0 with day 3 of same samples paired 't' test was used. Statistical analysis was done using SPSS 22.0 statistical software.

Results

Impact of *Theileria annulata* parasite of oxidative stress parameters was estimated by calculating GSH, SOD and MDA in seven infected cattle as depicted in Table 1. In cattle suffering from theileriosis, mean reduced glutathione (GSH) value was found to be significantly (P<0.05) decreased as compared to mean GSH value of apparently healthy cattle (n = 15). Mean malondialdehyde levels of infected cattle were found to be significantly higher (P<0.05) as compared to mean values in apparently healthy cattle. Mean value of superoxide dismuatase in infected cattle was found to be significantly (P<0.05) increased in cattle suffering from clinical theileriosis as compared to mean values of protein calculated in apparently healthy cattle.

Table 1: Oxidative stress parameters alterations in cattle suffering from theileriosis

Parameter	Apparently healthy control (n = 15)	Infected cattle (n = 7)
GSH (µmol/ml)	1.29 ± .018	1.069 ± .015*
SOD (U/mg protein)	23.85 ± 2.04	36.77 ± 6.61*
MDA (nmole/ml)	2.03 ± 0.28	4.41 ± 1.19*

*significant at P<0.05

After treatment with buparvaquone, non-significant increase in GSH and SOD levels whereas non-significant decrease in MDA level was observed in cattle at day 3 of post treatment as shown in Table 2.

Table 2: Alteration in Oxidative stress parameters of theileriosis infected cattle treated with Buparvaquone at day 3

Parameter	Infected cattle (n = 7)	
	Day 0	Day 3
GSH (µmol/ml)	1.069 ± .015	1.09 ± .013
SOD (U/mg protein)	36.77 ± 6.61	38.03 ± 2.06
MDA (nmole/ml)	4.41 ± 1.19	2.84 ± .57

Discussion

Concerning lipid peroxidation, there was a significant increase in the malondialdehyde activity in theileriosis affected cows as compared to apparently healthy controls. MDA is a lipid peroxidation end product, produced from polyunsaturated fatty acids oxidation. It is a profound and commonly used marker of lipid peroxidation (Moore and Roberts, 1998) [5]. In the present study, the significant elevation in MDA indicated occurrence of lipid peroxidation and oxidative stress at significant high levels in theileriosis affected cows that could be attributed to inability of antioxidant system to neutralize excessive free radicles generated during the disease course (Grewal *et al*., 2005; Nayak *et al*., 2018; Singh *et al*., 2018) [3, 7]. Free radicals cause oxidative damage to target molecules, such as lipids, nucleic acids and proteins. The red blood cells membrane is rich in polyunsaturated fatty acids which are the primary targets for oxidative damage induced by reactive metabolites (Davies and Goldberg, 1987) [2] resulting in reduced membrane symmetry, increased membrane permeability, increased RBCs fragility and subsequently anemia (Saluja *et al*., 1999) [11]. These findings revealed that lipid peroxidation as consequence to oxidative stress in tropical theileriosis affected cattle plays a great role in RBCs destruction and accordingly anemia (Grewal *et al*., 2005) [3]. Significant increase in SOD level observed in present study is in conjuction with the results of Neelam *et al*. (2017) [8] which can be attributed to stimulatory effect of parasites. Non-significant changes were observed in day 3 post treatment values of oxidative stress parameters. Following therapy with buaparvaquone non-significant changes were observed as no antioxidant was added in therapy. Therefore, it can be concluded that parasite impart oxidative stress and to decrease the level antioxidants must be added along with drug of choice.

Ethical Matters: Consent from owner of farm was taken while conducting the study.

References

1. Beutler E, Duron O, Kelly BM. Improved Method for the Determination of Blood Glutathione. Journal of Laboratory and Clinical Medicine 1963;61:882-888.
2. Davies KJA, Goldberg AL. Oxygen radicals stimulate intracellular proteolysis and lipid peroxidation by independent mechanisms in erythrocytes. Journal of Biological Chemistry 1987;262:8220-8226.
3. Grewal A, Ahuja CS, Singh SP, Chaudhary KC. Status of lipid peroxidation, some antioxidant enzymes and erythrocytic fragility of crossbred cattle naturally infected with *Theileria annulata*. Veterinary Research Communications 2005;29:387-394.
4. Madesh M, Balsubramanian KA. Microtitre plate assay for superoxide dismutase using MTT reduction by superoxide. Indian Journal of Biochemistry and Biophysics 1998;35:184-188.
5. Moore K, Roberts LJ. Measurement of lipid peroxidation. Free Radical Research 1998;28:659-671.
6. Narladkar BW. Projected economic losses due to vector and vector-borne parasitic diseases in livestock of India and its significance in implementing the concept of integrated practices for vector management. Veterinary World 2018;11(2):151-160.
7. Nayak SM, Senapati SK, Samal P, Sethy K, Meher S,

Das MR *et al*. Therapeutic management of oxidative stress in cattle, naturally affected with bovine tropical theileriosis by vitamin e and selenium. Journal of Pharmaceutical Innovation 2018;7(4):1141-1145.
8. Neelam, Rakha NK, Jhambh R, Virmani M, Goel P, Charaya G. Investigation into haematobiochemical profile and oxidative stress indices in the cattle naturally infected with bovine tropical theileriosis. Haryana Veterinarian 2017;56(2):129-134.
9. Razavi SM, Nazifi S, Mokhtari, Rakhshandehroo E. Correlations among the level of homocysteine, antioxidant enzymes, antioxidant vitamins and lipid peroxidation of erythrocytes in malignant ovine theileriosis. Research Journal of Parasitology 2015;10:42-49.
10. Rezaei SA, Naghadeh BD. Evaluation of antioxidant status and oxidative stress in cattle naturally infected with *Theileria annulata.* Veterinary Parasitology 2006;142:179-186.
11. Saluja PS, Gupta SL, Malhotra DV, Ambawat HK. Plasma malondialdehyde in experimental *Theileria annulata* infected cross bred bovine calves. The Indian Veterinary Journal 1999;76:379-381.
12. Shafiq-ur-Rahman. Lead-induced regional lipid peroxidation in brain. Toxicology Letters 1984;21(3):333-337.
13. Singh SK, Singh VK, Ram PK, Yadav BK, Nakade UR. Assessment of non-corpuscular markers of protein oxidation, lipid peroxidation and antioxidant status of calves with natural tropical theileriosis. Indian Journal of Veterinary Medicine 2018;38(1&2):60-63.