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APPRAISAL OF ANTIOXIDATIVE STRATEGIES IN THE ERYTHROCYTES OF *Marwari* SHEEP DURING EXTREME AMBIENCES

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Abstract: The present investigation was carried out for appraisal of antioxidative strategies in the erythrocytes of *Marwari* sheep during moderate, hot and cold ambiences. Haemolysates were prepared to determine antioxidants. Antioxidants included vitamin A and vitamin C. Results revealed mean values of vitamin A and vitamin C in erythrocytes were significantly ($p \leq 0.05$) lower during hot and cold ambiences as compared to respective moderate mean value. The mean values were significantly ($p \leq 0.05$) higher in male animals in all the ambiences. Age effect showed a significant ($p \leq 0.05$) increase in the mean values of both indicators being highest in the sheep of 18-24 months of age. On the basis of pattern of observations obtained in the present study, it was appraised that definitive strategies are present in the animals and during adverse conditions modulations occur to combat the stress.

Keywords: *Marwari* Sheep, antioxidative strategies, antioxidants, vitamin A.

Introduction

Antioxidative strategies are present inside the cells in the form of various endogenous antioxidative enzymes, antioxidants and free radical scavengers. Erythrocytes are constantly subjected to oxidative stress, from their role as an oxygen transporter. Since oxidized erythrocyte components are antigenic in regards to the formation of auto antibodies, a long-term exposure to severe oxidative stress consequently causes an autoimmune response to oxidized erythrocytes that can be regarded as an oxidative modification (Iuchi *et al*., 2007). An important consequence of an imbalance between the erythrocytes's components is the poor ability to cope with oxidative stress, which can result in degenerative changes in haemoglobin, cell membrane and enzymes required for normal erythrocytic function (Edwards and Fuller, 1996). Oxidative stress induces impairment of energy metabolism of erythrocytes (Tavazzi *et al*., 2001). Heat stress modulates metabolic reactions through free radicals and produces oxidative stress (Kataria *et al*., 2010).Indicators of oxidative stress allow the assessment of real status of physiological defenses and prevention of the appearance of correlated pathologies (Piccione *et al*., 2007). Marwari breed of sheep

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constitutes a major portion of the sheep population in Western part of Rajasthan. Changes in enzymes necessary for physiological adjustments are brought about by great fluctuations in ambient temperatures during extreme ambiences. Timely detection of oxidative stress due to extreme ambiences is an appropriate field of investigation to explore adaptive physiological measures of the animals and their use in health management and clinical diagnosis.

MATERIALS AND METHODS

Three hundred and sixty blood samples of apparently healthy Marwari sheep of both sexes ageing 6 months to 24 months were collected from private slaughter houses in and around Bikaner district, Rajasthan during moderate, hot and cold ambiences. Blood samples were collected during slaughtering. In each ambience 120 blood samples were collected and the animals were grouped into male (60) and female (60). Moderate ambience was comprise of October-November; hot ambience of May and June and cold ambience of December-January. To assess the effect of hot and cold ambiences on the parameters of antioxidative strategies in the erythrocytes of Marwari sheep, the result of various parameters analysed was compared with those analysed during moderate months serving as control. Following indicators were analysed to appraise the antioxidative strategies in erythrocytes:

A. Antioxidants

1. Vitamin A
2. Vitamin C

Various computer programmes were used to determine means and standard error (http://www.miniwebtool.com) and analyses of variance (www.danielsoper.com) to test the significance of the effects of ambiences, sex and age groups and correlations (Kaps and Lamberson, 2004). The changes in the means were measured by using multiple mean comparison procedures. For this Duncan's new multiple range test was used (Duncan, 1955).

RESULT AND DISCUSSION

Antioxidants

1. Vitamin A

The mean overall value of vitamin A in erythrocytes of *Marwari* sheep during moderate ambience (control) was 0.45±0.05 µmol gHb^{-1}.It was obtained from 120 *Marwari* sheep irrespective of sex and age. The mean values of vitamin A in erythrocytes were significantly ($p \leq 0.05$) lower during hot and cold ambiences as compared to moderate overall mean value. A highly significant ($p \leq 0.01$) effect of variation in ambience was also observed by analysis of variance. Calamari *et al*. (1999) reported an increase of oxygen derived free radicals and

reduced blood antioxidants like carotene in summer. A decrease in antioxidant defense leads to oxidative damage of biomolecules (Beckman and Ames, 1998).Antioxidant effect of vitamin A is well documented as it is also used to recycle back the oxidised α-tocopheroxyl radicals to the active reduced form (Wang *et al*., 1999). The decreased levels of vitamin A indicated towards the presence of oxidative stress. The sex and age effects were significant ($p \leq 0.05$) in moderate, extreme hot and cold ambiences. The mean values were significantly ($p \leq 0.05$) higher in male animals than female animals in all the ambiences. Age effect showed a significant ($p \leq 0.05$) increase in the mean values being highest in the animals of 18-24 months of age. Highly significant ($p \leq 0.01$) sex and age effects were revealed by analysis of variance. Sex and age related changes in serum vitamin A values have been also reported in several animal species by earlier researchers (Joshi, 2012 and Kataria *et al*., 2012 and Pandey *et al*., 2012). Findings clearly indicated that decrease in vitamin A status in erythrocytes of both sexes and all age groups showed depletion of antioxidant status as vitamin A is one of the antioxidants of the body.

2. Vitamin C

The mean overall value of vitamin C in erythrocytes of *Marwari* sheep during moderate ambience (control) was 2.6±0.01 μmol gHb^{-1}.It was obtained from 120 *Marwari* sheep irrespective of sex and age. The mean values of vitamin C in erythrocytes were significantly ($p \leq 0.05$) lower during hot and cold ambiences as compared to moderate overall mean value. A highly significant ($p \leq 0.01$) effect of variation in ambience was also observed by analysis of variance. Vitamin C in erythrocytes is suggested to be essential in the detoxification of superoxide radicals and hydrogen peroxide formed during red cell metabolism (Fakhri, 1991). Role of vitamin C in preventing oxidative stress of erythrocytes was also studied by earlier workers (Vani *et al*., 2010). Depletion of vitamin C confirms the presence of oxidative stress because repletion is reported after supplementation of vitamin C (Weiss, 2001). Kataria *et al*. (2010) also recommended the use of antioxidants in the conditions causing oxidative stress. The sex and age effects were significant ($p \leq 0.05$) in moderate, extreme hot and cold ambiences. The mean values were significantly ($p \leq 0.05$) higher in male animals than female animals in all the ambiences. Age effect showed a significant ($p \leq 0.05$) increase in the mean values being highest in the sheep of 18-24 months of age. Highly significant ($p \leq 0.01$) sex and age effects were revealed by analysis of variance. Higher vitamin C level in male animals reflected towards its higher synthesis to combat free radicals (Long *et al*., 1963). De and Durad (1991) observed a decrease in vitamin C with the

advancement of age. Nazifi *et al*. (2009) discussed about the influence of age on free radical generation and consequently, the enzyme antioxidant defense.

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FIGURES

Fig. 1: Mean changes in vitamin A (µmol gHb-1) overall values in the erythrocytes of Marwari sheep during extreme ambiences.

Fig. 2: Mean changes in vitamin A (µmol gHb-1) values according to sex and age groups in the erythrocytes of Marwari sheep during extreme ambiences.

Fig. 3: Mean changes in vitamin C (µmol gHb-1) overall values in theerythrocytes of Marwari sheep during extreme ambiences.

Fig. 4: Mean changes in vitamin C (µmolgHb-1) values according to sex and age groups in the erythrocytes of Marwari sheep during extreme ambiences.

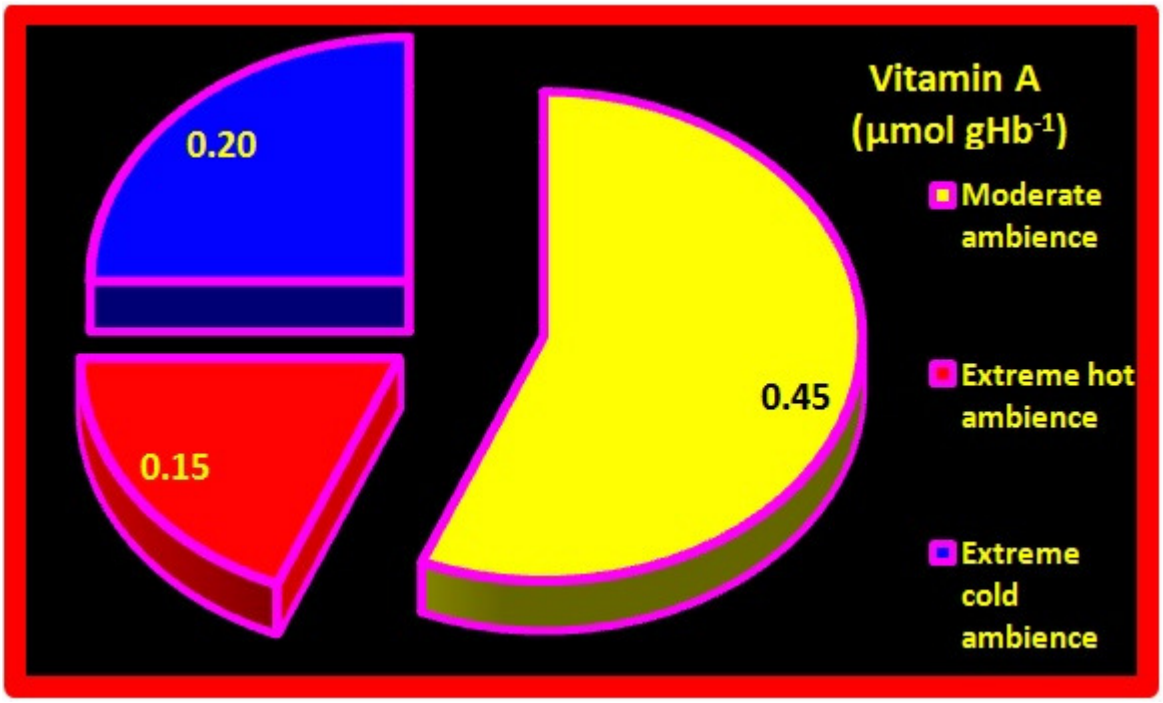
Vitamin A
(µmol gHb-1)
0.20
0.15
0.45
Moderate ambience
Extreme hot ambience
Extreme cold ambience

Fig. 1

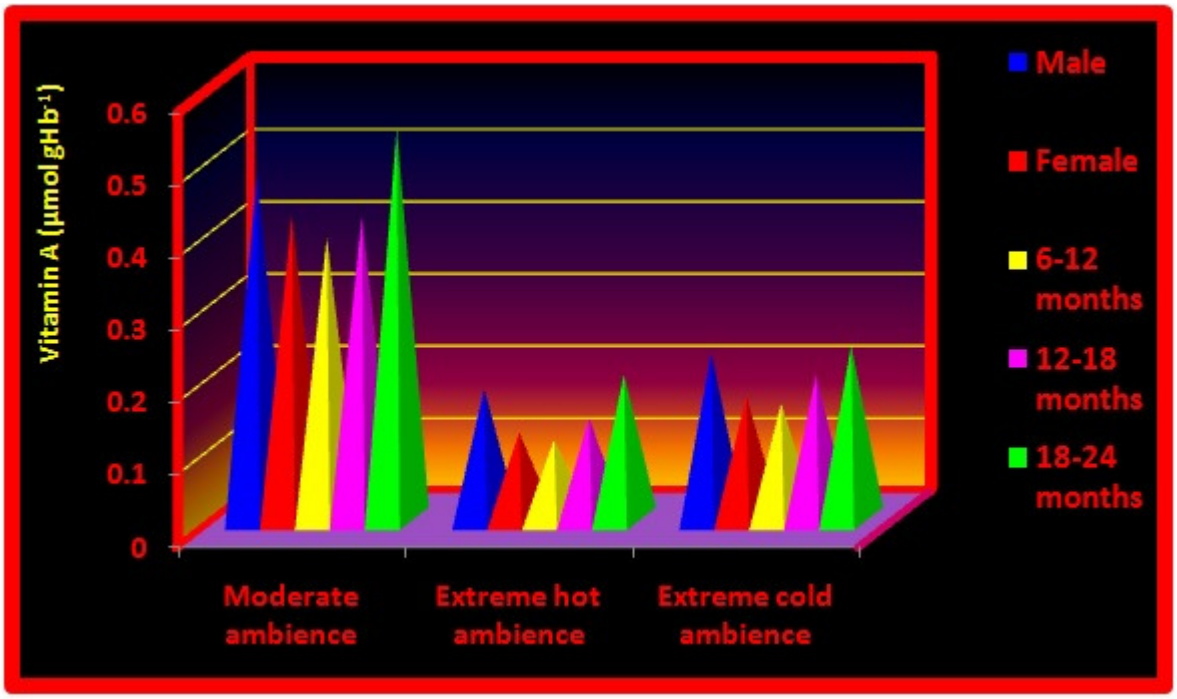
Vitamin A (µmol gHb-1)
0.6
0.5
0.4
0.3
0.2
0.1
0
Moderate ambience
Extreme hot ambience
Extreme cold ambience
Male
Female
6-12 months
12-18 months
18-24 months

Fig. 2

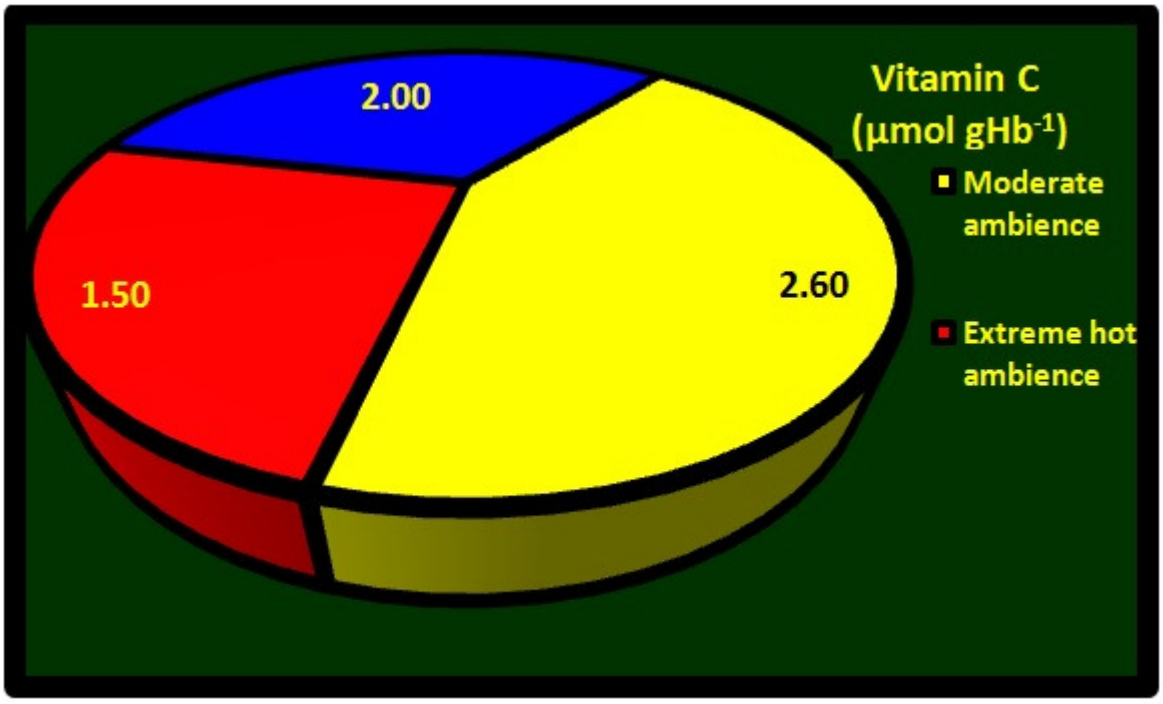
Vitamin C
(µmol gHb-1)
2.00
1.50
2.60
Moderate ambience
Extreme hot ambience

Fig. 3

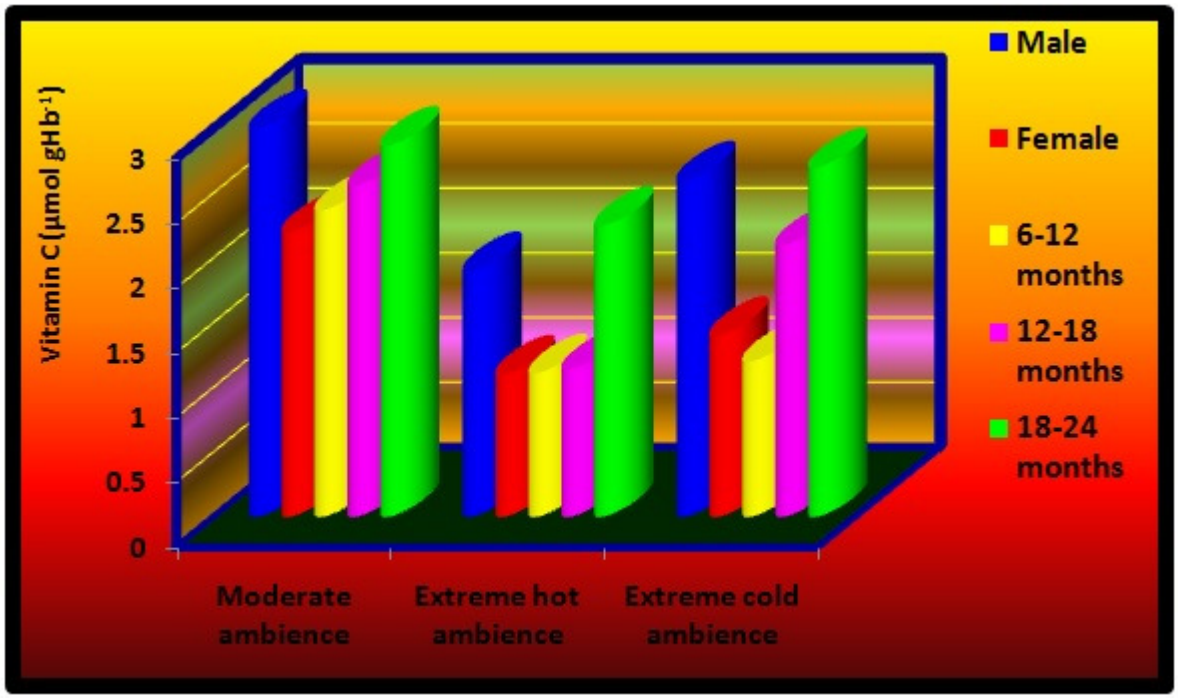
Vitamin C (µmol gHb-1)
3
2.5
2
1.5
1
0.5
0
Moderate ambience
Extreme hot ambience
Extreme cold ambience
Male
Female
6-12 months
12-18 months
18-24 months

Fig. 4