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Studies on haemato-biochemical profile of adult Jersey crossbred heifers during anestrus and non-hormonal induction of estrus

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ABSTRACT

In order to assess the haemato-biochemical profiles of adult Jersey crossbred heifers during anestrus and induced estrus condition, 12 adult heifers were selected for experimental studies. The age of the heifers were in between 2.5 - 3 years. The body weights were ranged between 185.5 - 252 kg. Selected animals were subjected to gynaecological examination per rectum and confirmed they were in anestrus condition. Heifers were maintained under standard managerial condition and feeding and they were treated with anthelmintic, mineral mixture, vitamin A injections and non-hormonal herbal preparation (*Sajani*,™ Sarabhai Zydus) for induction of heat. Blood samples were collected during anestrus and within 12 h after onset of heat symptoms (estrus). Eight animals exhibited estrus signs among treated animals. The results revealed that the total erythrocyte count, haemoglobin, packed cell volume, serum glucose, total serum protein, serum globulin, serum aspertate transaminase, serum alanine transaminase and serum alkaline phosphatase were significantly ($P < 0.01$) higher during estrus than in anestrus condition. The lymphocyte count was also significantly ($P < 0.05$) higher during estrus than anestrus condition. No significant differences were observed in mean corpuscular volume, mean corpuscular haemoglobin, mean corpuscular haemoglobin concentration, eosinophil count, basophil count, monocyte count, serum albumin and serum total cholesterol content between estrus and anestrus condition. Based on the observations, it may be concluded that good managerial practice along with treatment with non-hormonal herbal preparation improved physiological status of the anestrus animal.

Key words : Jersey crossbred heifer, haemato-biochemical profile, anestrus, estrus

Infertility among crossbred cattle is becoming burning problem and affecting economy of animal industry in rural area. Nutrients play an important role in fertility regulation. Deficiency or excess of certain haemato-biochemical constituents alter the proper function of reproductive organs during different reproductive phases (Rowlands, 1977). Deficiency of any nutrient affect enzymatic and cellular metabolism of reproductive tissue and thereby induce concomitant reproductive disorders, viz., anestrus and subestrus (Vhora *et al.*, 1995; Joe Arosh *et al.*, 1998). For assessing the reproductive status of dairy animals, the haematological and biochemical constituents can be used as precise guidelines. An attempt has been made to study the reproductive status of crossbred Jersey cattle through haematological and serum biochemical alteration during estrus and anestrus in Jersey crossbred heifers.

MATERIALS AND METHODS

Animals, feed, climate and management: Twelve Jersey crossbred heifers, aged between 2.5 - 3 years (as per record) and body weight ranged between 185.5-252 kg and apparently healthy were selected for experimental study from State Livestock Farm, Kalyani, Dept. of A.R.D., Govt. of West Bengal. The experiment was designed and work done from the month of April 2002 to August 2002 and the environmental temperature ranged from 19.4 - 38.9°C and relative humidity was 44-98% during experimental period. At start of experiment all the animals (12) were considered as anestrus heifer with the history of not showing any estrus signs previously with no palpable structure in both the ovaries and no detectable genital abnormalities on repeated careful rectal examination. The animals were maintained under standard

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managerial condition and feeding (NRC, 1989) and they were periodically treated with anthelmintic, mineral mixture and vitamin A injection. Ovary and reproductive tract were also managed at every 2 alternative days. The blood samples were collected from anestrus animals after maintaining the above mentioned managemental process. The anestrus animals were treated with a non-hormonal herbal heat inducing preparation (Sajani, Tm Sarabhai Zydus, each bolus consist of Mrigakshi-600 mg. Tikshna-60 mg, Krishna Pippali-60 mg and Sringaver 60 mg) @ 6 bolus orally per day per animal for two consecutive days. Out of 12 animals, 6 animals showed heat symptoms within 10 days. Nonresponded animals were treated in same way, out of which two animals were showed signs of estrus at 14th day of treatment. The blood samples were collected from estrus animals within 12 h after exhibiting estrus signs.

Haematological, serum biochemical and statistical analysis: Blood samples from individual animals were collected by Jugular vein puncture with and without anticoagulant (dipotassium salt of Ethylenediamine tetraacetate, EDTA) anestrus and estrus condition. The non-coagulated blood was used for haematological assessment viz., haemoglobin (Hb), packed cell volume (PCV), total erythrocyte count (TEC), total leucocyte count (TLC) and differential leucocyte count (DLC). The blood samples without anticoagulant were kept for separation of serum and were decanted into sterilized apendrop tube and stored at -20°C till further analysis. PCV, TEC, TLC and DLC (Schalm *et al.*, 1975) and Hb (Benjamin, 1985) were estimated using standard procedure as described. Serum samples were analyzed for glucose, total protein, albumin, globulin and cholesterol as described by Pattanaik *et al.* (1999). The serum enzymes namely, serum aspertate transaminase (AST) and serum alanine transaminase (ALT) were estimated colorimentrically (Reitman and Frankel, 1957) and serum alkaline phosphatase (ALP) by the method described by Walter and Schutte (1974). The erythrocyte indices viz., mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentration (MCHC) were calculated as per the formulae described by Schalm *et al.* (1975). All the data were subjected to unequal student `t' test as per Snedecor and Cochran (1989).

RESULTS AND DISCUSSION

The haematological and biochemical constituents of anestrus and estrus adult Jersey crossbred heifers are given in Table 1.

Haematological constituents: The estrus heifers were showed significantly ($P < 0.01$) higher level of TEC (10^6 cmm^{-1}), Hb (g%) and PCV (%) than that of anestrus heifers. The values were in corroboration with the findings of Kumar *et al.* (1986), Chetty and Rao (1986) and Ali *et al.* (1991). The TEC, PCV (%) and Hb (g%) were within normal range for cattle (Brar *et al.*, 2000) without any sign of anaemic condition (Feldman *et al.*, 2000). However, in the present study, these values were approaching higher range of normal value indicates that the animals were matured and in good health status. Lack of sufficient quantities of Hb in blood is responsible for reduced oxygen transport to the vital tissues, it causes reduced oxidation of nutrients, which in turn affect the whole cellular metabolism in gonadal cells which is metabolically more active (Swenson and Reece, 1993) and this might be one of the reason for heifers that were not showing estrus signs before proper management. The elevated TEC during estrus condition might be due to excitement, higher level of gonadal and other hormones, supplementation of micro minerals, higher feed intake and higher nutrients assimilation (Swenson and Reece, 1993). The erythrocyte indices (viz., MCV, MCH and MCHC) were not altered between estrus and anestrus condition in Jersey crossbred heifers. However, TLC significantly ($P < 0.05$) higher in anestrus than estrus condition. The similar observation was also reported by Kumar *et al.* (1985). Higher level of neutrophil count were observed during anestrus than estrus condition and this increase might be associated with mild degree of infection as suggested by Sharma *et al.* (1983). In contrary, lymphocyte counts were significantly higher in estrus than anestrus condition (Sharma *et al.*, 1983; Kumar *et al.*, 1985).

Biochemical constituents

Glucose : The serum glucose level was significantly ($P < 0.01$) higher in estrus than anestrus heifers. The values were within the normal ranges of cattle (Brar *et al.*, 2000). The present results were in agreement with the findings of Sharma *et al.* (1984), Chetty and Rao (1986), Dutta *et*

Table 1. Mean values of different haematological parameters during anestrus and estrus condition of Jersey crossbred heifers

Parameters		Anestrus	Estrus
Total erythrocyte count (10^6/cmm)	Mean	6.11±0.50^{a***}	8.49±0.39^{b***}
	Range	5.40 - 6.72	8.01 - 9.13
Haemoglobin (gm dl^{-1})	Mean	7.88±0.63^{a***}	11.54±1.11^{b***}
	Range	6.40 - 8.70	9.5 - 13.0
Packed cell volume (%)	Mean	30.93±1.22^{a***}	39.70±3.00^{b***}
	Range	28.40 - 35.20	33.6 - 44.8
Mean corpuscular volume (pg dl^{-1})	Mean	53.57±4.05	47.86±4.78
	Range	45.0 - 61.20	40.60 - 57.72
Mean corpuscular haemoglobin (pg dl^{-1})	Mean	6.11±0.50	8.49±0.39
	Range	4.82 - 8.24	7.04 - 9.62
Mean corpuscular haemoglobin concentration (g dl^{-1})	Mean	25.56±1.97	30.67±3.76
	Range	21.50 - 30.26	22.82 - 37.14
Total leucocyte counts (10^3 cmm^{-1})	Mean	8.44±0.81^{a**}	5.81±0.69^{b**}
	Range	5.72 - 11.25	4.86 - 8.27
Differential leucocyte count			
Neutrophil (%)	Mean	41.65±2.08^{a***}	31.76±1.56^{b***}
	Range	37 - 46	28 - 38
Eosinophil (%)	Mean	7.38±0.73	6.53±1.24
	Range	5 - 8	2 - 9
Basophil (%)	Mean	0.47±0.07	0.48±0.07
	Range	0 - 1	0 - 1
Lymphocyte (%)	Mean	43.51±2.39^{a**}	55.34±3.82^{b**}
	Range	36 - 52	45 - 66
Monocyte (%)	Mean	6.99±0.76	5.90±0.89
	Range	4 - 10	3 - 10

Means having different superscript of each parameter differ significantly
** $P < 0.01$; * $P < 0.05$

al. (1988) and Joe Arosh *et al.* (1998). The higher serum glucose level is affected the energy status and thereby reproductive status of the animal. The increased levels of serum glucose might have elevated the progesterone production directly by increasing LH production (Richards *et al.*, 1989). Hypoglycemia affects hypothalamus and release of gonadotrophins from hypophysis (Howland *et al.*, 1966) which reduces ovarian activity. Arthur *et al.* (1982) also reported that the temporary cessation of estrus in cows and delayed puberty in heifers occur when they exposed to negative energy balance.

Total serum protein albumin and globulin : The total serum protein and serum globulin concentrations were significantly ($P < 0.01$) higher during estrus than that of anestrus heifers. The results were in corroboration with the findings of Vhora *et al.* (1995), Joe Arosh *et al.* (1998). However, serum albumin level did not differ significantly, it might be due to amino acids necessary for albumin synthesis were in correct proportion at the same time globulin level decreased, because of different amino acid composition, which might be deficient. An optimum amount of total serum protein is necessary for the expression of estrus signs in cows. Bearcten and Fuquary (1992) suggested that deficiency of protein cause weak expression of estrus or temporary cessation of estrus. The low level of total serum protein in anestrus condition might cause deficiency of certain amino acids required for synthesis of gonadotrophins (Vhora *et al.*, 1995).

Serum and cholesterol : There was no significant difference observed between anestrus and estrus condition. The cholesterol levels in the serum of experimental animals were within the normal range (35-160 mg dl^{-1}) as described by Brar *et al.* (2000). The findings were in accordance with the results of Dutta *et al.* (1991) and Vhora *et al.* (1995). Cholesterol is an important

precursor for the synthesis of steroid hormone. Nonsignificant increased level of cholesterol in anestrus animals could be due to its non-utilization for production of steroid hormones (Zala *et al.*, 1972). However, these findings were in contrary to findings of Dutta *et al.* (1988) and Joe Arosh *et al.* (1998).

AST. ALT and AKP : The activities of AST, ALT and AKP were significantly ($P < 0.01$) low in anestrus animals when compared to estrus animals. The findings are in concurrence with the findings of Joe Arosh *et al.* (1998), Paul *et al.* (1991). Hormonal imbalance and deranged enzymatic activity affect the normal reproductive behaviour of the animal and cause physiological alteration (Paul *et al.*, 1991). The alterations of the different serum enzymatic activities are indicatives of physiological activity of the tissues. In this study, the reduced activities of these (AST, ALT and AKP) serum enzyme activities might be due to reduced physiological activities of reproductive organ in anestrus heifers.

It may be concluded that during anestrus condition animals have exhibited low levels of TEC, Hb, PCV, lymphocytes count than that of estrus condition. Similarly, concentration of serum glucose, total protein, globulin and activities of AST, ALT and AKP were also higher in estrus condition that that of anestrus condition. It indicates these haematological and serum biochemical constituents had been improved during estrus. It implies that without improvement of these constituents, animal can not exhibit heat symptoms. So these haemato-biochemical profiles can be used as a guideline for adult Jersey crossbred heifers for correcting nutritional related delayed puberty or anestrus condition.

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