

## Study of certain Haematological Parameter in Repeat Breeding Cows

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### Abstract

A study was conducted on certain haematological parameters in crossbred repeat breeder cows presented at outdoor of teaching veterinary clinical complex in Odisha Veterinary College, Bhubaneswar. Blood samples were collected from 30 crossbred repeat breeder cows (Group: I) and compared with normal cyclic cows (Group: II) at the stage of estrus. The blood parameters such as TWBC (Total White Blood Cells), TRBC (Total Red Blood Cells), HBG (Haemoglobin), HCT (Haematocrit Value), ESR (Erythrocyte sedimentation rate), TPLT (Total Platelet), MCV (Mean Corpuscular Volume), MCH (Mean Corpuscular Haemoglobin), MCHC (Mean Corpuscular Haemoglobin Concentration), RDW (Red Blood Cell Distribution Width), MPV (Mean Platelet Volume) and PDW (Platelet Distribution Width) were investigated. The mean values for repeat breeding cows were  $11.00 \pm 1.88 \times 10^3 \text{ mm}^{-3}$ ,  $7.43 \pm 0.95 \times 10^6 \text{ mm}^{-3}$ ,  $10.64 \pm 1.51 \text{ g dl}^{-1}$ ,  $32.78 \pm 5.71\%$ ,  $08.56 \pm 1.29 \text{ mm}^2 \text{ Hrs}^{-1}$ ,  $601.30 \pm 2.86 \times 10^3 \text{ mm}^{-3}$ ,  $44.20 \pm 3.15 \text{ } \mu\text{m}^3$ ,  $14.45 \pm 0.81 \text{ pg}$ ,  $32.82 \pm 0.82 \text{ g dl}^{-1}$ ,  $15.32 \pm 1.60\%$ ,  $11.67 \pm 2.64 \text{ } \mu\text{m}^3$  and  $1.69 \pm 0.37\%$  respectively. The corresponding values for the control group were  $11.73 \pm 1.24 \times 10^3 \text{ mm}^{-3}$ ,  $8.93 \pm 1.61 \times 10^6 \text{ mm}^{-3}$ ,  $11.66 \pm 2.88 \text{ g dl}^{-1}$ ,  $36.04 \pm 9.15\%$ ,  $11.87 \pm 1.34 \text{ mm}^2 \text{ Hrs}^{-1}$ ,  $2591.00 \pm 1.38 \times 10^3 \text{ mm}^{-3}$ ,  $53.00 \pm 5.34 \text{ } \mu\text{m}^3$ ,  $17.68 \pm 1.52 \text{ pg}$ ,  $32.88 \pm 1.53 \text{ g dl}^{-1}$ ,  $16.89 \pm 2.27\%$ ,  $7.90 \pm 1.72 \text{ } \mu\text{m}^3$  and  $0.97 \pm 0.09\%$  respectively. The TRBC, HBG, HCT, ESR, MCV, MCH, MPV, PDW values of normal cyclic cows significantly differed ( $p < 0.05$ ) from repeat breeder cows at oestrous stage. The TPLT count was highly significant ( $p < 0.01$ ) in normal cyclic cows as compared to repeat breeder cows. It was concluded that lower erythrocytic indices in the cyclic non breeding condition in crossbred cows has got to interfere in the normal conception and reproduction.

### 1. Introduction

Blood is one of the important constituents in the body, which maintains the physiological equilibrium. Thus any disturbance in this equilibrium maintenance will lead to disease condition or pathological condition which in turn can be known from the changed haematological parameters. So the knowledge of haematological values is useful in diagnosing various pathological and metabolic disorders, which can adversely affect the productive and reproductive performance of cows, which in turn leads to affect the economic status of the farmers (Dutta et al., 1988; Perumal et al., 2013a; Perumal et al., 2013b). Kumar et al. (1991) carried out haematological studies to correlate with reproductive performance of dairy cattle. The study of various haematological constituents or parameters during various states of reproductive stages is helpful for detection of various abnormalities in these parameters and

able to correlate with problems associated with reproductive dysfunction (Perumal et al., 2013a, Perumal et al., 2013b). These changes in haematological constituents are important indicators of the physiological or pathological state of the animal.

It has been reported that there is significant increase in RBC and WBC during estrus phase in cattle (Soliman and Zaki, 1963). Dhoble and Gupta (1981) and Nadiu and Rao (1982) reported a lower level of haemoglobin in anestrus cattle than in cycling buffaloes and cows. Kumar and Sharma (1991) has reported lower values of total erythrocyte count, haemoglobin concentration and PCV in anestrus and repeat breeder animals, while mean corpuscular volume and total leukocyte count were increased in these two groups. Information regarding the haematological parameters in the normal cyclic and repeat breeding condition at oestrous stage in crossbred cow

is meager. So the present study has been designed to analyze various haematological parameters in normal cyclic and repeat breeders at the stage of estrus in crossbred cows with an idea to understand the abnormal condition of reproduction and to rectify the same.

## 2. Materials and Methods

### 2.1. Experimental animals

This study was conducted at the Department of Animal Reproduction, Gynaecology and Obstetrics, Odisha University of Agriculture and Technology, Bhubaneswar, India. A total of 60 number of Jersey crossbred cows presented at the outdoor of Teaching Veterinary Clinical Complex of Orissa veterinary college, Bhubaneswar, India for artificial insemination and infertility treatment was selected. These experimental animals were grouped into two groups based on reproductive history, reproductive record, oestrous signs and rectal examination. Normal cyclic cows (n=30) were kept in group I, while cyclic non breeder cows (n=30) were retained in group II. All the experimental animals gynaeco-clinically examined per rectally for confirmation of the clinical status of the reproductive organs.

### 2.2. Collection and analysis of blood

Blood samples were collected aseptically by Jugular vein puncture from individual cows into collection tubes containing EDTA (Chauhan, 1995). The collected blood was used for haematological studies. The blood parameters such as TWBC (Total White Blood Cells), TRBC (Total Red Blood Cells), HBG (Haemoglobin), HCT (Haematocrit Value), TPLT (Total Platelet), MCV (Mean Corpuscular Volume), MCH (Mean Corpuscular Haemoglobin), MCHC (Mean Corpuscular Haemoglobin Concentration), RDW (Red Blood Cell Distribution Width), MPV (Mean Platelet Volume) and PDW (Platelet Distribution Width) were studied by standard methods described by Coles (1986) and Sastry (1989) and also by using automatic blood analyzer.

### 2.3. Statistical analysis

Mean values ( $\pm$ SE) of various haematological parameters for cows of two groups were computed. In order to observe the magnitude of variation in these parameters among cows of the two groups, the data were analyzed statistically using student t test described by Snedecor and Cochran (1980). The data are expressed as mean $\pm$ SEm. Differences were considered significant if  $p < 0.05$  and  $p < 0.01$ .

## 3. Results and Discussion

The mean values of blood parameters of normal cyclic oestrous and repeat breeding oestrous cows were presented in the Table 1. The total red blood cell count (TRBC) is higher

in normal cyclic oestrous ( $8.93 \pm 1.61 \times 10^6 \text{ mm}^{-3}$ ) and lower in repeat breeding oestrous cows ( $7.43 \pm 0.95 \times 10^6 \text{ mm}^{-3}$ ). Further the analysis of variance revealed that the TRBC count of normal cyclic oestrous cows significantly ( $p < 0.05$ ) differed from repeat breeding oestrous cows. The increased amount of TRBC may be due to hyperactivity and excitement, arising out of high level of estrogen from graffian follicle (Coles, 1986). Erythrocytes also play a part in the body's immune response: when lysed by pathogens such as bacteria, their hemoglobin releases free radicals that break down the pathogen's cell wall and membrane, killing it (Jiang et al., 2007). Kumar et al. (1991) observed significantly lower values of total erythrocyte count in estrous and repeat breeder animals in a study on clinico-pathological changes in normal, anestrus and repeat breeding buffaloes. Similarly in another study, Kumar et al. (1992) found the lower RBC count in non-cyclic heifers as compared to cyclic heifers. The values for haemoglobin concentration, haematocrit value, total platelet count, mean corpuscular volume were significantly ( $p < 0.05$ ) higher in normal cyclic oestrous cows as compared to repeat breeding oestrous cows. The MCH and MCHC values were influenced by variation in HBG synthesis (Benjamin, 1978). Since the MCHC is a measure of the quantity of HBG in each RBC and also relates to the weight of HBG and volume of cell, the cows having higher HBG concentration showed higher MCH and

Table 1: Haematological parameters (mean $\pm$ SE) at estrus in normal cyclic and cyclic non-breeder cows

Parameters	Normal cyclic cows (30)	Cyclic Non breeder cows (30)
TWBC ( $10^3 \text{ mm}^{-3}$ )	11.73 $\pm$ 1.24	11.00 $\pm$ 1.88
TRBC ( $10^6 \text{ mm}^{-3}$ )	8.93 $\pm$ 1.61 <sup>b</sup>	7.43 $\pm$ 0.95 <sup>a</sup>
HBG (g dl <sup>-1</sup> )	11.66 $\pm$ 2.88 <sup>b</sup>	10.64 $\pm$ 1.51 <sup>a</sup>
HCT (%)	36.04 $\pm$ 9.15 <sup>b</sup>	32.78 $\pm$ 5.71 <sup>a</sup>
ESR (mm 24 Hrs <sup>-1</sup> )	11.87 $\pm$ 1.34 <sup>b</sup>	08.56 $\pm$ 1.29 <sup>a</sup>
TPLT ( $10^3 \text{ mm}^{-3}$ )	2591.00 $\pm$ 1.38 <sup>b</sup>	601.30 $\pm$ 2.86 <sup>a**</sup>
MCV ( $\mu\text{m}^3$ )	53.00 $\pm$ 5.34 <sup>b</sup>	44.20 $\pm$ 3.15 <sup>a</sup>
MCH (pg)	17.68 $\pm$ 1.52 <sup>b</sup>	14.45 $\pm$ 0.81 <sup>a</sup>
MCHC (g dl <sup>-1</sup> )	32.88 $\pm$ 1.53	32.82 $\pm$ 0.82
RDW (%)	16.89 $\pm$ 2.27	15.32 $\pm$ 1.60
MPV ( $\mu\text{m}^3$ )	7.90 $\pm$ 1.72 <sup>b</sup>	11.67 $\pm$ 2.64 <sup>a</sup>
PDW (%)	0.97 $\pm$ 0.09 <sup>b</sup>	1.69 $\pm$ 0.37 <sup>a</sup>
Neutrophils (%)	27.53 $\pm$ 1.77 <sup>a</sup>	23.58 $\pm$ 1.36 <sup>b</sup>
Lymphocytes (%)	61.87 $\pm$ 1.66	62.56 $\pm$ 0.99
Monocytes (%)	3.98 $\pm$ 1.30	3.89 $\pm$ 1.20
Eosinophils (%)	8.16 $\pm$ 0.68 <sup>b</sup>	13.13 $\pm$ 1.23 <sup>a</sup>
Basophils (%)	0.08 $\pm$ 0.10	0.23 $\pm$ 0.11

<sup>\*\*</sup> $p < 0.01$

MCHC values. In the present study, the values for PCV were higher in normal cyclic oestrous cows than in repeat breeding oestrous cows. The values for MCV were higher in normal cyclic animals than in repeat breeding oestrous cows. Kumar and Sharma (1991) observed lower values of total erythrocyte count, haemoglobin concentration and PCV in anestrous and repeat breeder animals while mean corpuscular volume and total leukocyte count were increased in these two groups. Patil et al. (1992) noted that PCV was higher in cyclic heifers than other heifers. They also observed that the higher values of erythrocytes in heifers are responsible for higher PCV in them as compared to the adult buffaloes. The cyclic oestrous cows have higher metabolic rate, which causes increased production of higher number of RBC and this leads to increased values of other haematological parameters.

The TWBC concentration is non-significantly higher in normal cyclic animal ( $11.73 \pm 1.24 \times 10^3 \text{ mm}^{-3}$ ) as compared to repeat breeder animal and it may be due to movement of excessive amount of leucocytes in to uterus at the time of oestrous for phagocytosis process. In states of excitement, exercise and strange surroundings there is also leukocytosis (neutrophilia), since adrenaline liberated during these states mobilizes the marginal neutrophil pool cells (Sastry, 1989).

The MPV and PDW were significantly ( $p < 0.05$ ) higher in normal cyclic oestrous cows as compared to repeat breeder oestrous cows. This may be due to the fact that RBC is responsible for transport of  $\text{O}_2$  to the tissues that leads to more consumption of  $\text{O}_2$  to reproductive tract, which is helpful for better utilization of energy and contraction of smooth muscle that leads to successful fertilization and ultimate good conception rate and that leads to higher fertility rate. Ganong (2001) studied the high concentration of HBG, PCV and RBC were desirable physiological characters for efficient transport of oxygen and carbon-di-oxide. This is an essential requirement for maintaining the health of animals. Thus, cows with higher concentration of HBG, PCV and RBC were more economical as a result of improved reproductive efficiency. Ramakrishna (1996) reported a significantly lower level of HBG in anestrous cows as compared to normal cyclic cows. The platelet are responsible for production thrombocytokinin which is responsible for production of more amount of prostaglandins ( $\text{PGF}_2\alpha$  and  $\text{PGE}_2$ ) and PG helps for ovulation and subsequent contraction of reproductive tract and fertilization (Donna et al., 2008). PG induces platelet aggregation by combining with a specific (endoperoxide) receptor and further release of factors, which are helpful for induction of inflammatory like reaction and this helps for final ovulation and fertilization. The platelet number and mean platelet volume width determine the production of prostaglandins, histamine, serotonin and bradykinine. These all inflammatory factors are

responsible for final stage of ovum maturation and subsequent ovum release and corpus luteum formation. CL produces more progesterone, which will ultimately helpful for successful fertilization and pregnancy. Platelet aggregation via adhesion to (von Willebrand factor- interstitial collagen types VI) v WF-Col VI complexes leads to stimulate the triggering factor that stimulate the thrombotic cascade under high shear stress and in oestrous condition that stimulate release of arachidonic acid and PG secretion and the platelet dense granule and alpha granule constituents are responsible for arachidonate metabolism and is necessary for collagen-induced release of lysosomal enzymes that ultimately are useful for ovulation and fertilization (Macintyre, 1979).

The MCHC ( $\text{g dl}^{-1}$ ) value did not differ significantly between the normal cyclic and repeat breeder cows as compared to other haematological factors, but this value is higher in normal cyclic cows than the repeat breeder cows. The MCH and MCHC values are affected by variation in HBG synthesis (Benjamin, 1978). Since the MCHC is a measure of the quantity of HBG in each RBC and also relates to the weight of HBG and volume of cell, the cows having higher HBG concentration showed higher MCH and MCHC values. Kumar et al. (1991) found lower values of PCV in anoestrous and repeat breeder cows. In fertile oestrous, the level of HBG concentration ( $11.32 \pm 0.21 \text{ g dl}^{-1}$ ) was higher as compared to non-fertile oestrous (Kumar and Sharma, 1991; Perumal et al., 2013a; Perumal et al., 2013b) The ESR ( $24 \text{ Hrs mm}^{-1}$ ) value significantly differ ( $p < 0.05$ ) in normal cyclic oestrous as compared to repeat breeder estrous cows and this indicates the normal oestrous cows having higher RBC count as compared to repeat breeder oestrous cows and have very rapid sedimentation rate than repeat breeder cows. When erythrocytes undergo shear stress in constricted vessels, they release ATP which causes the vessel walls to relax and dilate so as to promote normal blood flow (Wan et al., 2008) When their haemoglobin molecules are deoxygenated, erythrocytes release S-nitrosothiols which also acts to dilate vessels (Diesen et al., 2008), thus directing more blood to areas of the body depleted of oxygen. It has been recently demonstrated that erythrocytes can also synthesize nitric oxide enzymatically, using L-arginine as substrate, just like endothelial cells (Kleinbongard et al., 2006) and this dilatation of blood vessels of uterus and other reproductive tract causes increase in the tonicity of uterine horn which favours the movement of gametes towards the site of fertilization and successful conception. Exposure of erythrocytes to physiological levels of shear stress activates nitric oxide synthetase and export of nitric oxide which may contribute to the regulation of vascular tonus (Ulker et al., 2009).

#### 4. Conclusion

Based on the results of the present study, it was concluded that



low leucocytic count low erythrocytic count, low concentration of haemoglobin and low platelet count in cyclic non-breeding repeat breeder crossbred cows might be contributing to the cause of repeat breeding.

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