

## CLINICAL, HAEMATOLOGICAL AND SERUM MACRO MINERAL CONTENTS IN BUFFALOES WITH GENITAL PROLAPSE

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### ABSTRACT

In the present study, 20 buffaloes suffering from genital prolapse and 10 normal pregnant (control group) buffaloes were used to investigate clinical and haematological changes and serum macro mineral contents. Blood samples of all animals were collected aseptically in two portions i.e. with and without anticoagulant. Haematological parameters were determined from samples containing anticoagulant. Serum was separated from the other portion for determination of calcium, phosphorus and magnesium concentrations with spectrophotometric kits. The results showed that there was a drastic decrease in serum calcium ( $6.42 \pm 1.05$  v  $10.96 \pm 0.95$  mg/dl), phosphorus ( $2.90 \pm 0.85$  v  $5.50 \pm 1.61$  mg/dl) and magnesium ( $1.50 \pm 0.53$  v  $2.40 \pm 0.53$  mg/dl) levels in prolapsed animals as compared to the controls ( $P < 0.01$ ). There was also a significant decrease in PCV, Hb concentration, lymphocytes and monocytes, while an increase in ESR, WBC counts and neutrophils was observed in prolapsed animals as compared to controls. However, there was no difference in haematological and serum macro mineral contents between vaginal prolapsed and uterine prolapsed buffaloes. It was concluded that deficiency of calcium, phosphorus or magnesium might be possible causes of genital prolapse in these buffaloes.

**Key words:** Haematology, serum biochemistry, genital prolapse, buffaloes.

### INTRODUCTION

Livestock play an important role in the economy of Pakistan by providing most essential items of human consumption like milk, meat, skins and wool. Nili-Ravi buffaloes occupy a key position in the rural economy of Pakistan, particularly for the landless farmers or those with smallholdings. Due to its importance in the small and landless farmer's economy, it is often called as the "Black gold of Pakistan". It has also been suggested that due to its large size, emphasis on selection of buffaloes for meat production can yield high returns (Cady *et al.*, 1983).

Genital prolapse is a common obstetrical problem, which adversely affects productive and reproductive performance of buffaloes by affecting postpartum return to oestrus, conception rate and calving interval. Samad *et al.* (1987) reported the incidence of genital prolapse as 42.9% among various obstetrical problems in buffaloes. The maximum incidence of prepartum vaginal prolapse was recorded in May, while postpartum uterine prolapse showed the highest incidence during July and September.

However, there is little information in the literature on haematological parameters and serum macro mineral levels in buffaloes with genital prolapse. Therefore, the present project was planned to investigate clinical, haematological and serum macro mineral contents in prolapsed animals and to compare them with normal pregnant animals of the same species.

### MATERIALS AND METHODS

#### Experimental animals

The present study was conducted on buffaloes brought to the clinic of the Department of Animal Reproduction, University of Agriculture, Faisalabad or maintained in and around Faisalabad city. For this purpose, a total of 30 animals were selected, out of which 20 were suffering from genital prolapse (10 with vaginal and other 10 with uterine prolapse), while the remaining 10 were normal pregnant (control). Complete history regarding the clinical status of the disease was obtained.

#### Blood collection and processing

About 20 ml of blood was collected in 2 parts, one with and other without anticoagulant, from each animal aseptically in clean, sterilized test tubes by jugular vene-puncture method. The blood samples containing anticoagulant were used for haematological parameters including red blood cells (RBC) count, packed cell volume (PCV), haemoglobin (Hb) concentration, mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH), mean corpuscular haemoglobin concentration (MCHC), erythrocyte sedimentation rate (ESR), white blood cell (WBC) count and differential leucocytic counts. Serum was separated from blood samples without anticoagulant and stored at  $-20^{\circ}\text{C}$  until analysed for the calcium, phosphorus and magnesium contents.

Serum calcium concentrations were estimated using the calcium kit (Human, Germany, Cat No. 10011). Serum phosphorus contents were determined using photometric UV Test kit (Cat No. 10027). Serum magnesium levels were studied by using the magnesium kit. (Crescent, Cat. No. CE 502).

### Statistical analysis

Mean values ( $\pm$  SE) of various parameters for control buffaloes and those suffering from vaginal prolapse including (vagino-cervical) and uterine prolapse, were computed. In order to ascertain magnitude of variation in various parameters between respective groups, the data were subjected to unpaired T-test, using computer programme M-STAT C (Freed, 1987).

## RESULTS AND DISCUSSION

### Haematology

Mean ( $\pm$  SE) value of red blood cell ( $10^6$ /ul) count in prolapsed animals ( $6.31 \pm 0.69$ ) was higher than the controls ( $6.10 \pm 0.43$ ), the difference was statistically non significant (Table 1). Similar results were reported by Tarjinder and Singh (1993), who observed RBC count of  $6.5 \pm 0.2 \times 10^6$ /ul in prolapsed animals.

**Table 1: Hematological parameters in prolapsed and control groups of buffaloes (mean  $\pm$  SE)**

Parameter	Prolapsed group	Control group	T-value
RBC ( $10^6$ /ul)	$6.31 \pm 0.69$	$6.10 \pm 0.43$	0.89 <sup>NS</sup>
PCV (%)	$35.70 \pm 2.20$	$42.10 \pm 2.23$	7.47**
Hb (gm/dl)	$11.26 \pm 1.17$	$14.05 \pm 0.84$	6.69**
MCV (fl)	$81.30 \pm 10.98$	$69.39 \pm 6.98$	0.49 <sup>NS</sup>
MCH (pg)	$18.00 \pm 2.78$	$23.04 \pm 2.32$	4.91**
MCHC (gm/dl)	$31.74 \pm 4.07$	$33.29 \pm 2.32$	1.11 <sup>NS</sup>
ESR (mm/hr)	$36.36 \pm 5.33$	$31.00 \pm 1.70$	4.10**
WBC ( $10^3$ /ul)	$12.82 \pm 1.10$	$6.63 \pm 0.56$	7.47**

\*\* = Highly significant ( $P < 0.01$ ), NS = Non significant.

A highly significant ( $P < 0.01$ ) decrease in PCV in prolapsed animals as compared to the control group was observed (Table 1). These results are in agreement with those of Kinney (1967), who stated that the decrease in PCV in prolapsed animals might be due to possible release of antidiuretic hormone as a result of stress. According to the Agarawal (1987), a significant decrease in PCV due to anorexia and toxemia was observed which may hold true for the present study also. However, no difference was observed in PCV when the vaginal prolapsed animals were compared with uterine prolapsed animals (Table 2).

In the present study, a significant ( $P < 0.01$ ) decrease in haemoglobin concentration was found in prolapsed animals as compared to the normal control group (Table 1). Tarjinder and Singh (1993) reported

similar results in buffaloes. Decrease in haemoglobin concentration in prolapsed animals may be due to loss of body fluid (Maxwell and Kleeman, 1972) or reduction in the size of erythrocyte rather than number of RBCs (Benjamin, 1978). This may be considered a possible explanation of decrease in haemoglobin concentration observed in prolapsed buffaloes in the present study. However, no difference was observed in haemoglobin concentration between vaginal prolapse and uterine prolapse groups (Table 2). There was a significant decrease ( $P < 0.01$ ) in MCH in buffaloes suffering from genital prolapse as compared to controls. However, there was no difference in MCV and MCHC between animals of the two groups (Table 1). Similarly, no difference was found in MCH, MCV and MCHC in buffaloes with vaginal or uterine prolapse (Table 2).

**Table 2: Haematological parameters in buffaloes with vaginal and uterine prolapse (mean  $\pm$  SE)**

Parameters	Vaginal prolapse	Uterine prolapse
RBC ( $10^6$ /ul)	$6.26 \pm 0.76$	$6.42 \pm 0.58$
PCV (%)	$35.54 \pm 2.26$	$36.00 \pm 2.24$
Haemoglobin (gm/dl)	$11.59 \pm 0.98$	$10.66 \pm 1.33$
MCV (fl)	$64.82 \pm 6.73$	$56.20 \pm 5.33$
MCH (pg)	$18.75 \pm 2.68$	$16.60 \pm 2.63$
MCHC (gm/dl)	$32.73 \pm 3.31$	$29.90 \pm 4.94$
ESR (mm/hr)	$36.34 \pm 6.06$	$36.39 \pm 4.07$
WBC ( $10^3$ /ul)	$12.83 \pm 0.75$	$12.80 \pm 1.65$

Values for each parameter differ non-significantly between the two groups.

A significant ( $P < 0.01$ ) increase in erythrocyte sedimentation rate (ESR) was observed in prolapsed animals as compared to controls in the present study (Table 1), but no difference was observed between vaginal and uterine prolapsed animals (Table 2). Similar results were found by Tarjinder and Singh (1993). According to Jain (1986), ESR increases in inflammatory conditions and in acute generalized infection, in addition to change in concentration of various proteins in blood. However, according to Benjamin (1978), change in ESR is a nonspecific reaction. The ESR in bovine is so variable that a normal value for use as a diagnostic tool for a specific condition cannot be estimated.

Higher WBCs counts (Table 1) in prolapsed group ( $P < 0.01$ ) compared to the control animals as observed in the present study are similar to those recorded by Verma *et al.* (1989), who stated that increase in WBC could be a result of higher level of cortisol due to stress in affected buffaloes. Singh *et al.* (1992) observed a progressive increase in WBC count with advancement of normal pregnancy, while Sindhu (1975) stated that this increase was due to stress. Mohy *et al.* (1985) reported that increase in WBC count in prolapsed animals was due to release of adrenocorticotrophic hormone from the adrenal gland.

According to Benjamin (1978), the degree of leukocytosis depends upon several factors such as nature of the causative agent, severity of the infection, resistance of the animal and localization of the inflammatory response. When vaginal prolapsed buffaloes were compared with uterine prolapsed animals, no difference was observed in WBC count between both the groups (Table 2). This may be due to the fact that degree of leukocytosis increases with the advancement of pregnancy, as both the conditions occur approximately at the same stage of parturition, hence no difference was observed.

In the present study, a significant increase in neutrophils ( $P < 0.01$ ), while a significant decrease in lymphocytes ( $P < 0.05$ ) and monocytes ( $P < 0.01$ ), was recorded in prolapsed animals as compared to control animals (Table 3). These findings agree with the results of Tarjinder and Singh (1993), who also reported neutrophilia in prolapsed animals. This may be due to increased level of cortisol due to stress (Verma *et al.*, 1989), or inflammatory process in the genital prolapse (Malik *et al.*, 1990). However, neutrophilia has also been reported during excitement, exercise and adrenaline release (Sastry, 1989). Basophils and eosinophils remained unchanged in animals of prolapsed and control groups (Table 3). Similarly, no difference in differential leukocytic count was observed between vaginal prolapsed and uterine prolapsed animals (Table 4). Prepartum vaginal and postpartum uterine prolapse both usually cause the same degree of inflammation in the genital tract, which may be a possible reason for similar differential leukocytic counts between animals of the two groups in this study.

**Table 3: Differential leukocyte counts (%) in prolapsed and control buffaloes (mean  $\pm$  SE)**

Parameter	Prolapsed group	Control group	T-value
Neutrophils	37.50 $\pm$ 4.95	31.20 $\pm$ 3.46	1.28**
Lymphocytes	50.15 $\pm$ 4.31	54.20 $\pm$ 4.57	2.29*
Monocytes	4.20 $\pm$ 4.58	9.20 $\pm$ 1.55	4.41**
Eosinophils	3.70 $\pm$ 1.84	4.60 $\pm$ 1.78	3.60 <sup>NS</sup>
Basophils	1.00 $\pm$ 1.30	0.80 $\pm$ 0.92	0.64 <sup>NS</sup>

\* = Significant ( $P < 0.05$ ), \*\* = Highly significant ( $P < 0.01$ )  
NS = Non significant.

**Table 4: Differential leukocytic counts (%) in buffaloes with vaginal and uterine prolapse**

Parameters	Vaginal prolapse	Uterine prolapse
Neutrophils	36.00 $\pm$ 4.67	40.29 $\pm$ 4.46
Lymphocytes	50.77 $\pm$ 4.42	49.00 $\pm$ 4.16
Monocytes	7.92 $\pm$ 1.80	7.42 $\pm$ 2.94
Eosinophils	4.08 $\pm$ 2.00	3.00 $\pm$ 1.41
Basophils	1.38 $\pm$ 1.50	0.57 $\pm$ 0.54

Values for each parameter differ non-significantly between the two groups.

## Serum macro minerals

### Calcium

Hypocalcemia was observed in prolapsed group as compared to the control group ( $P < 0.01$ ) in the present study (Table 5). Marques *et al.* (1996) and Salmanoglu and Salmanoglu (1998) found similar results in their studies, while the results of Paul *et al.* (2000) are contrary to the present findings. Risco *et al.* (1984) reported that hypocalcemia resulted in loss of muscular contractions and ultimately uterine prolapse in buffaloes. Goff *et al.* (1990) reported that cortisol might cause immuno-suppression, hypocalcaemia and loss of muscular contractions.

When comparison between vaginal prolapsed and uterine prolapsed groups was carried out, no significant difference was observed in serum calcium levels between the two groups (Table 6). This may be due to the reason that same factors like hypocalcaemia, inflammation and stress are responsible for both conditions.

**Table 5: Some biochemical parameters (mg/dl) in prolapsed and control buffaloes (mean  $\pm$  SE)**

Parameters	Prolapsed group	Control group	T-value
Calcium	6.42 $\pm$ 1.05	10.96 $\pm$ 0.95	11.55**
Phosphorus	2.90 $\pm$ 0.85	5.50 $\pm$ 1.61	4.79**
Magnesium	1.50 $\pm$ 0.53	2.40 $\pm$ 0.53	4.38**

\* = Highly significant ( $P < 0.01$ ), NS = Non significant.

**Table 6: Mean values ( $\pm$  SE) of minerals in buffaloes with vaginal and uterine prolapse (mg/dl)**

Parameters	Vaginal prolapse	Uterine prolapse
Calcium	6.48 $\pm$ 1.04	6.30 $\pm$ 1.12
Phosphorus	3.05 $\pm$ 0.90	2.61 $\pm$ 0.74
Magnesium	1.52 $\pm$ 0.61	1.46 $\pm$ 0.40
Ca : P	2.12 : 1	2.41 : 1

Values for each parameter differ non-significantly between the two groups.

### Phosphorus

There was a significant ( $P < 0.01$ ) decrease in serum phosphorus level in prolapsed animals as compared to the control group (Table 5) in the present study. These results are in agreement with those of Marques *et al.* (1996). Pathak and Janakiraman (1987) reported calcitonin and progesterone insufficiency as a cause of hypophosphoremia in cattle. Between vaginal and uterine prolapsed groups of animals, no difference was found in serum phosphorus levels (Table 6).

### Magnesium

In the present study, a significant ( $P < 0.01$ ) decrease in serum magnesium level in prolapsed animals was recorded as compared to the control animals (Table 5). These results are in agreement with the study of Marques *et al.* (1996). However, no

difference was observed in serum magnesium levels between vaginal prolapsed and uterine prolapsed groups of animals (Table 6). These results do not match with those of Paul *et al.* (2000). The variation in these results might be due to differences in diet fed to animals used in these two studies.

Based on the results of the present study, it may be concluded that deficiency of calcium, phosphorus and magnesium might be the possible factor that lead to prolapse of genital tract in buffaloes included in the present study.

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