VETERINARY SCIENCE RESEARCH JOURNAL
olume 7 | Issue 1 | April, 2016 | 9-15

DOI : 10.15740/HAS/VSRJ/7.1/9-15
Visit us - *www.researchjournal.co.in*
e ISSN-2230-942X

RESEARCH ARTICLE

Modulation of serum trace mineral profiles in post-partum suboestrous surti buffaloes with $PGF_2\sim$ alone and $PGF_2\sim$ along with vitamin A, D_3, E and toldimphos sodium preparation therapy at day 55

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Abstract : The serum profile of trace elements (Cu, Co, Zn, Fe and Mn) studied from 55 day to 120 day postpartum in 24 suboestrous surti buffaloes with $PGF_2\alpha$ alone (T_1), Vitamin A, D_3, E (T_2) and toldimphos sodium preparation and $PGF_2\alpha$ along with vitamin A, D_3, E and toldimphos sodium preparation treatment (T_3) and control (T_4) group revealed that the levels of most elements varied non-significantly between treatments T_1, T_2, T_3 and control groups and even within the group between different time intervals post-treatment. The overall mean serum copper, cobalt, zinc, iron and manganese values in T_1, T_2, T_3 and control groups at 0 hr, 24 hr, 48 hr and 72 hr post-treatment were 1.56±0.014, 1.49±0.012, 1.49±0.017 and 1.48±0.017 ppm; 0.61±0.016, 0.58±0.018, 0.60±0.019 and 0.62±0.016 ppm; 1.57±0.061, 1.66±0.062, 1.78±0.063 and 1.60±0.044 ppm; 3.48±0.04, 3.41±0.07, 3.31±0.05 and 3.33±0.08 ppm as well as 0.146±0.007, 0.155±0.022 , 0.139±0.007 and 0.153±0.008 ppm, respectively. In the study, we could not find differences in serum trace minerals levels between treated and control groups at different time intervals. Moreover, micronutrients can not be synthesized in the body. Hence, it is concluded that trace elements should be daily supplied in the field and in organized farms as mineral mixture to suffice the requirement of the trace elements.

Key words : Hormone therapy, Trace- minerals profile, Suboestrous, Surti buffaloes

How to cite this paper : Rede, A.S., Khasatiya, C.T., Soni, D.K.,Chaudhary, S.S. and Katkar, S.P. (2016). Modulation of serum trace mineral profiles in post-partum suboestrous surti buffaloes with $PGF_2\alpha$ alone and $PGF_2\alpha$ along with vitamin A, D_3, E and toldimphos sodium preparation therapy at day 55. *Vet. Sci. Res. J.*, **7**(1) : 9-15.

Paper History : Received : 15.12.2015; Revised : 17.02.2016; Accepted : 07.03.2016

INTRODUCTION

Various minerals (Cu, Co, Se, Mn, Zn and iodine) are the essential nutrients bearing a significant role in the reproductive performance of ruminants. Deficiency or excess of minerals like P, Cu and Zn have been associated

with subnormal fertility and anoestrus conditions. Trace elements including Cu, Co, Zn, Fe, Se, I, Mo, Mn and certain macro-elements like K, Ca, Na, Cl, P have been found to be very essential for normal livestock growth. Trace elements may function as cofactors, as activators of enzymes, or as stabilizers of secondary molecular structure. Hidiroglou (1979) suggested that reproductive failure may be induced by deficiencies of single or combined trace elements and by imbalances. There is lack of information on effect of postpartum hormonal therapy on mineral profile; hence the study was aimed to evaluate weekly serum trace minerals profile of $PGF_2\alpha$ and $PGF_2\alpha$ along with Vitamin A, D_3, E and Toldimphos sodium preparation treatments and control groups of postpartum sub-oestrous surti buffaloes.

RESEARCH METHODOLOGY

The study was conducted on twenty four suboestrous Surti buffaloes from 45 to 120 days post-partum. They were randomly divided into four groups (T_1, T_2, T_3 and T_4) comprising of six animals in each group. All these buffaloes had normal calving and subsequent normal genital health as assessed Gynaeco-clinically. Oestrus occurrence was detected daily in them with the help of teaser bull parading in morning and evening hours. The animals which were not exhibiting overt signs of oestrus during routine heat detection programme were segregated and subjected to rectal palpation. The animals with palpable structures either corpus luteum (CL) or follicle, on either of the ovaries were selected for another palpation after eleven days apart to ascertain their cyclic nature and considered as silent heat/subestrous buffaloes. The buffaloes in T_1 group were treated with 2 ml of Inj. Cloprostenol sodium (Inj. Cyclix) (500 µg, $PGF_2\alpha$ analogue, I/M route); the buffaloes in T_2 group were treated with [inj. Vit. AD_3E preparation (5 ml, I/M route) + inj. Toldimphos sodium preparation (15 ml, I/M route)]; the buffaloes in T_3 group were treated with 2 ml of Inj. Cloprostenol sodium (Inj. Cyclix) (500 µg, $PGF_2\alpha$ analogue, I/M route) + [inj. Vit. AD_3E preparation (5 ml, I/M route) + inj. Toldimphos sodium preparation (15 ml, I/M route)] and the buffaloes in group T_4 were kept as sub-oestrous control group. All these buffaloes were then followed for oestrus induction response, reproductive performance for upto 120 days post-partum. Approximately, 10 ml blood samples in serum clotting vaccutainers were collected from all those selected animals on 0 hr (prior to treatment), 24 hr, 48 and 72 hr post-treatment aseptically by jugular vein puncture. The vaccutainers containing blood samples were kept in slanting position at room temperature for 1-2 hours. Finally, serum was separated by centrifugation at 3000 rpm for 15 minutes and stored in properly labelled sterilized 4.5 ml plastic storage vials at –20°C in deep freezer until analysis. The levels of trace minerals *viz.*, copper, cobalt, zinc, iron and manganese were determined according to the method of Krishna and Ranjhan (1980). The blood serum samples (0.5 ml each) were digested with 4.5 ml volume of tri-acid mixture (perchloric acid: sulphuric acid: nitric acid; 1:2:1) on a hot plate. The clear transparent residues were diluted in double glass-distilled water and the final volume was made to 25 ml. These aliquots were then used for estimation of trace elements, *viz.*, copper, cobalt, zinc, iron and manganese on an Atomic Absorption Spectrophotometer. The data were analyzed using standard statistical procedures (Steel and Torrie, 1981).

RESULTS AND DISCUSSION

The mean of serum Cu, Co, Zn, Fe and Mn concentrations obtained in suboestrous treated (T_1, T_2 and T_3) and control (T_4) groups buffaloes during 55 postpartum to till 3 weeks post-treatment are depicted in respected Tables 1-5.

Serum copper concentration :

The mean serum copper levels of suboestrous surti buffaloes in T_1 group at 0 hr, 24 hr, 48 hr and 72 hr were 1.53±0.014, 1.54±0.014, 1.56±0.014 and 1.57±0.014 ppm, respectively. The corresponding values for T_2 group at 0 hr, 24 hr, 48 hr and 72 hr were 1.51±0.012, 1.45±0.012, 1.49±0.012 and 1.49±0.012 ppm; T_3 group were 1.47±0.017, 1.47±0.017, 1.51±0.017 and 1.51±0.017 ppm and T_4 group were 1.51±0.017, 1.45±0.017, 1.47±0.017 and 1.46±0.017

ppm, respectively. The overall serum copper values in T_1, T_2, T_3 and T_4 groups were 1.56±0.014 (ranging from 1.31 to 1.66) ppm, 1.49±0.012 (ranging from 1.33 to 1.66) ppm, 1.49±0.017 (ranging from 1.40 to 1.66) ppm and 1.48±0.017 (ranging from 1.39 to 1.64) ppm, respectively.

The mean serum copper concentration of suboestrous surti buffaloes did not differ significantly at 0 hr, 24 hr, 48 hr and 72 hr interval within and between all the treatment and control groups including overall means between the groups at different time intervals.

The levels of mean serum copper seen in various treatment and control groups of surti buffaloes compared well with the report of Khasatiya (2003), who recorded overall pooled mean copper concentration values did not vary significantly in $PGF_2\alpha$ treatment and control group (1.35±0.02 vs. 1.31±0.03 ppm) in suboestrous surti buffaloes. Similarly, Deshpande (2007) recorded non-significant difference among mean serum copper levels in suboestrous treated (0.47±0.07 µg/ml), control (0.41±0.06 µg/ml) and normally cyclic (0.43±0.06 µg/ml) crossbred cows. However, the mean serum copper values recorded by Khasatiya (2003) in surti buffaloes and Deshpande (2007) in crossbred cows were lower. In addition to this, Chauhan and Nderingo (1997) also recorded comparatively lower serum copper concentrations during cyclic, early postpartum and the late postpartum period as compared to present findings, in cattle as 0.72±0.06, 0.69±0.05 and 0.75±0.07 ppm, respectively. The lower values as compared to present findings found in respective animals may be due to species difference. Moreover, importance of copper in the animals feed stuff have been discussed by various workers as McDowell (1992) suggested that copper deficiency is the second most common mineral deficiency of cattle in the world, surpassed in prevalence only by phosphorus deficiency. Copper levels appear to be influenced by hormones of reproduction, the higher serum copper level indicated higher oestrogenic and lower FSH and LH activity in the serum and its concentration was found to be highest during peak breeding season (Desai *et al.*, 1978).

The overall mean serum copper concentration values in suboestrus buffaloes were found to be higher (1.48±0.017 µg/ml), when it compared with the anoestrus values 1.09±0.05µg/ml, 0.88±0.00 µg/ml and 0.73±0.032 µg/ml reported by Chandolia and Verma (1987) in anoestrous buffalo heifers, Khattab *et al.* (1995) in Egyptian buffaloes and Yassein *et al.* (1995) in anoestrous buffaloes, might be due to cyclic nature of suboestrous buffaloes and at the same time lower value might be prone to anoestrus condition. The critical level of copper (0.65 µg/ml) was suggested by McDowell (1992) below which the clinical signs of deficiency may occur. The blood copper level at 14 to 21 and 38 to 45 days postpartum was not related to any of the postpartum reproductive performance in non-suckled dairy cows (Larson *et al.*, 1980).

Serum cobalt concentration:

The mean serum cobalt levels of suboestrous surti buffaloes in T_1 group at 0 hr, 24 hr, 48 hr and 72 hr were 0.59±0.016, 0.62±0.016, 0.60±0.016 and 0.63±0.016 ppm, respectively. The corresponding values for T_2 group at 0 hr, 24 hr, 48 hr and 72 hr were 0.57±0.018, 0.59±0.018, 0.57±0.018 and 0.60±0.018 ppm; T_3 group at were 0.59±0.019, 0.60±0.019, 0.60±0.019 and 0.61±0.019 ppm and T_4 group were 0.60±0.016, 0.62±0.016, 0.62±0.016 and 0.65±0.016 ppm, respectively. The overall serum cobalt values in T_1, T_2, T_3 and T_4 groups were 0.61±0.016 (ranging from 0.49 to 0.73) ppm, 0.58±0.018 (ranging from 0.52 to 0.76) ppm, 0.60±0.019 (ranging from 0.48 to 0.71) ppm and 0.62±0.016

Table 1 : Serum copper concentration (ppm) pattern at different time intervals/days in sub-estrus treated and control groups of animals (Mean±SE)

Time intervals/ days	Groups (n=6)			
	$PGF_{2\alpha}$ (T_1)	Vit+ P (T_2)	$PGF_2\alpha$+vit+ P(T_3)	Control (T_4)
0 hr/0th Day	$1.53\pm0.014_a^w$	$1.51\pm0.012_a^w$	$1.47\pm0.017_a^w$	$1.51\pm0.017_a^w$
24 hr/1st Day	$1.54\pm0.014_a^w$	$1.45\pm0.012_a^w$	$1.47\pm0.017_a^w$	$1.45\pm0.017_a^w$
48 hr/2nd Day	$1.56\pm0.014_a^w$	$1.49\pm0.012_a^w$	$1.51\pm0.017_a^w$	$1.47\pm0.017_a^w$
72 hr/3rd Day	$1.57\pm0.014_a^w$	$1.49\pm0.012_a^w$	$1.51\pm0.017_a^w$	$1.46\pm0.017_a^w$
Overall	$1.56\pm0.014_a$	$1.49\pm0.012_a$	$1.49\pm0.017_a$	$1.48\pm0.017_a$

Means bearing common superscripts within a column (group) and means bearing common subscripts within a row (between the groups) do not differ significantly (p>0.05).

(ranging from 0.49 to 0.77) ppm, respectively.

The mean serum cobalt concentration of suboestrous surti buffaloes did not differ significantly at 0 hr, 24 hr, 48 hr and 72 hr within and between all the treatment and control groups including overall means between the groups at different time intervals.

The mean serum cobalt level are in agreement with the reports of Khasatiya (2003) in surti buffaloes, who reported overall pooled means did not vary significantly between groups and even within group between different weeks postpartum in $PGF_2\alpha$ treated and control groups (0.64±0.02 vs. 0.51±0.02 ppm). Similarly, Deshpande (2007) observed non-significant difference among mean serum cobalt levels in suboestrous treated (0.98±0.20 μg/ml), control (0.66±0.10 μg/ml) and normally cyclic (0.90±0.12 μg/ml) crossbred cows.

Moreover, the importance of cobalt in the animals feed stuff discussed by various workers as cobalt has been found to be required in the synthesis of vitamin B_{12} and its deficiency has been associated with non-functional ovaries (Wagner, 1963) and general infertility (Alderman, 1963). The perusal of literature available shows that most common manifestation of cobalt deficiency is marked reduction in conception rate with reduction in oestrus during normal breeding season.

Serum zinc concentration :

The mean serum zinc levels of suboestrous surti buffaloes in T_1 group at 0 hr, 24 hr, 48 hr and 72 hr were 1.58±0.061, 1.55±0.061, 1.57±0.061 and 1.56±0.061 ppm, respectively. The corresponding values for T_2 group at 0 hr, 24 hr, 48 hr and 72 hr were 1.69±0.062, 1.70±0.062, 1.70±0.062 and 1.55±0.062 ppm; T_3 group were 1.80±0.063, 1.74±0.063, 1.76±0.063 and 1.80±0.063 ppm and T_4 group were 1.64±0.044, 1.59±0.044, 1.58±0.044 and 1.56±0.044 ppm, respectively. The overall serum zinc values in T_1, T_2, T_3 and T_4 groups were 1.57±0.061 (ranging from 1.28 to 2.18) ppm, 1.66±0.062 (ranging from 1.23 to 2.16) ppm, 1.78±0.063 (ranging from 1.27 to 2.17) ppm and 1.60±0.044 (ranging from 1.14 to 2.11) ppm, respectively.

The mean serum zinc concentration of suboestrous surti buffaloes did not differ significantly at 0 hr, 24 hr, 48 hr and 72 hr within and between all the treatment and control groups including overall means between the groups at different time intervals.

Table 2 : Serum cobalt concentration (ppm) pattern at different time intervals/days in suboestrus treated and control groups of animals (Mean±SE)

Time intervals/ days	Groups (n=6)			
	$PGF_{2\alpha}$ (T_1)	Vit+ P (T_2)	$PGF_2\alpha$+vit+ P(T_3)	Control (T_4)
0 hr/0th Day	$0.59\pm0.016_a^w$	$0.57\pm0.018_a^w$	$0.59\pm0.019_a^w$	$0.60\pm0.016_a^w$
24 hr/1st Day	$0.62\pm0.016_a^w$	$0.59\pm0.018_a^w$	$0.60\pm0.019_a^w$	$0.62\pm0.016_a^w$
48 hr/2nd Day	$0.60\pm0.016_a^w$	$0.57\pm0.018_a^w$	$0.60\pm0.019_a^w$	$0.62\pm0.016_a^w$
72 hr/3rd Day	$0.63\pm0.016_a^w$	$0.60\pm0.018_a^w$	$0.61\pm0.019_a^w$	$0.65\pm0.016_a^w$
Overall	$0.61\pm0.016_a$	$0.58\pm0.018_a$	$0.60\pm0.019_a$	$0.62\pm0.016_a$

Means bearing common superscripts within a column (group) and means bearing common subscripts within a row (between the groups) do not differ significantly (p>0.05).

Table 3 : Serum zinc concentration (ppm) pattern at different time intervals/days in sub-estrus treated and control groups of animals (Mean±SE)

Time intervals/ days	Groups (n=6)			
	$PGF_{2\alpha}$ (T_1)	Vit+ P (T_2)	$PGF_2\alpha$+vit+ P (T_3)	Control (T_4)
0 hr/0th Day	$1.58\pm0.061_a^w$	$1.69\pm0.062_a^w$	$1.80\pm0.063_a^w$	$1.64\pm0.044_a^w$
24 hr/1st Day	$1.55\pm0.061_a^w$	$1.70\pm0.062_a^w$	$1.74\pm0.063_a^w$	$1.59\pm0.044_a^w$
48 hr/2nd Day	$1.57\pm0.061_a^w$	$1.70\pm0.062_a^w$	$1.76\pm0.063_a^w$	$1.58\pm0.044_a^w$
72 hr/3rd Day	$1.56\pm0.061_a^w$	$1.55\pm0.062_a^w$	$1.80\pm0.063_a^w$	$1.56\pm0.044_a^w$
Overall	$1.57\pm0.061_a$	$1.66\pm0.062_a$	$1.78\pm0.063_a$	$1.60\pm0.044_a$

Means bearing common superscripts within a column (group) and means bearing common subscripts within a row (between the groups) do not differ significantly (p>0.05).

These findings to some extent also corroborated with those of Khasatiya (2003), who found that the weekly-pooled mean plasma zinc levels in $PGF_2\alpha$ treatment and control groups of suboestrous surti buffaloes did not reveal any significant difference at any of the weeks postpartum including the overall means (1.87±0.04 vs. 1.73±0.07 ppm) and the values ranged from 1.62±0.15 to 2.11±0.13 and 1.57±0.26 to 1.98±0.31 ppm, respectively. Similarly, Deshpande (2007) recorded non-significant difference among mean serum zinc levels in suboestrous treated (1.95±0.51 μg/ml), control (1.55±0.14 μg/ml) and normally cyclic (1.42±0.11 μg/ml) crossbred cows. In addition to this, Chauhan and Nderingo (1997) also recorded comparatively lower serum zinc concentrations during cyclic, early postpartum and the late postpartum period as compared to present findings, in cattle as 0.66±0.05, 0.59±0.06 and 0.63±0.11 ppm, respectively. The overall mean serum zinc concentration values in suboestrus buffaloes were found to be higher (1.60±0.044 μg/ml), when it compared with the anoestrus values (1.21±6.80 μg/ml and 1.32±0.00 μg/ml) reported by Chandolia and Verma (1987) in buffalo heifers and Khattab *et al.* (1995) in Egyptian buffaloes, respectively, may be due to cyclic nature of suboestrous buffaloes and at the same time lower value may prone to anoestrus condition as it is true from the present comparison. The critical level of zinc (0.6 to 0.8 μg/ml) was suggested by McDowell (1992) below which the clinical signs of deficiency may occur. A reduction in zinc level might interfere with prostaglandin receptor mediated phase and consequently the luteolytic process which in turn causes some of the reproductive pathology (Carlson *et al.*, 1982). Optimum level of zinc is reported to be essential to maintain the activity of FSH and LH (Apagar, 1985).

Serum iron concentration :

The mean serum iron levels of suboestrous surti buffaloes in T_1 group at 0 hr, 24 hr, 48 hr and 72 hr were 3.46±0.04, 3.48±0.04, 3.48±0.04 and 3.51±0.04 ppm, respectively. The corresponding values for T_2 group at 0 hr, 24 hr, 48 hr and 72 hr were 3.43±0.07, 3.40±0.07, 3.41±0.07 and 3.41±0.07 ppm; T_3 group were 3.26±0.05, 3.32±0.05, 3.32±0.05 and 3.35±0.05 ppm and T_4 group were 3.32±0.08, 3.34±0.08, 3.33±0.08 and 3.35±0.08 ppm, respectively. The overall serum iron values in T_1, T_2, T_3 and T_4 groups were 3.48±0.04 (ranging from 2.83 to 3.48) ppm, 3.41±0.07 (ranging from 2.90 to 3.91) ppm, 3.31±0.05 (ranging from 2.91 to 3.88) ppm and 3.33±0.08 (ranging from 2.83 to 3.88) ppm, respectively.

The mean serum iron concentration of suboestrous Surti buffaloes did not differ significantly within all the treatment and control groups and also between all the treatment and control groups at 0 hr, 24 hr, 48 hr and 72 hr.

The mean serum iron level are in agreement with the reports of Deshpande (2007), who recorded non-significant difference among mean serum iron levels in suboestrous treated (4.70±0.34 μg/ml), control (3.68±0.34 μg/ml) and normally cyclic (3.72±0.66 μg/ml) crossbred cows; however, the values reported by Deshpande (2007) were comparatively little bit higher than present findings may attributed to breed and species differences. On the contrary, Khasatiya (2003) reported that the overall pooled mean iron concentration was significantly higher in $PGF_2\alpha$ control than its treatment group (3.76±0.11 vs. 3.34±0.09 ppm) as compared to the present findings in which we could not find difference between treatment control groups. Maynard and Loosli (1969) suggested that iron was of little importance in reproduction as compared to copper and zinc. However, low level of iron could possibly result in improper oxygenation of uterus resulting in impaired nutrition in the uterus for the conceptus causing death of the embryo (Reddy and

Table 4 : Serum iron concentration (ppm) pattern at different time intervals/days in sub-estrus treated and control groups of animals (Mean±SE)

Time intervals/ days	Groups (n=6)			
	$PGF_{2\alpha}(T_1)$	Vit+ P (T_2)	$PGF_2\alpha$+vit+ P(T_3)	Control(T_4)
0 hr/0th Day	$3.46\pm0.04_a^w$	$3.43\pm0.07_a^w$	$3.26\pm0.05_a^w$	$3.32\pm0.08_a^w$
24 hr/1st Day	$3.48\pm0.04_a^w$	$3.40\pm0.07_a^w$	$3.32\pm0.05_a^w$	$3.34\pm0.08_a^w$
48 hr/2nd Day	$3.48\pm0.04_a^w$	$3.41\pm0.07_a^w$	$3.32\pm0.05_a^w$	$3.33\pm0.08_a^w$
72 hr/3rd Day	$3.51\pm0.04_a^w$	$3.41\pm0.07_a^w$	$3.35\pm0.05_a^w$	$3.35\pm0.08_a^w$
Overall	$3.48\pm0.04_a$	$3.41\pm0.07_a$	$3.31\pm0.05_a$	$3.33\pm0.08_a$

Means bearing common superscripts within a column (group) and means bearing common subscripts within a row (between the groups) did not differ significantly (p>0.05).

Reddy, 1988).

The overall mean serum iron concentration values in suboestrus buffaloes were found to be higher (3.33±0.08 μg/ml), when it compared with the anoestrus values (3.05±0.35 μg/ml) reported by Chandolia and Verma (1987) in buffalo heifers and Khattab *et al.* (1995) in Egyptian buffaloes, respectively, might be due to cyclic nature of suboestrous buffaloes and at the same time lower value might be prone to anoestrus condition.

Serum manganese concentration :

The mean serum manganese levels of suboestrous surti buffaloes in T_1 group at 0 hr, 24 hr, 48 hr and 72 hr were 0.145±0.007, 0.147±0.007, 0.147±0.007 and 0.148±0.007 ppm, respectively. The corresponding values for T_2 group at 0 hr, 24 hr, 48 hr and 72 hr were 0.139±0.022, 0.204±0.022, 0.140±0.022 and 0.139±0.022 ppm; T_3 group were 0.139±0.007, 0.139±0.007, 0.139±0.007 and 0.139±0.007 ppm and T_4 group were 0.154±0.008, 0.153±0.008, 0.152±0.008 and 0.153±0.008 ppm, respectively. The overall serum manganese values in T_1, T_2, T_3 and T_4 groups were 0.146±0.007 (ranging from 0.073 to 0.238) ppm, 0.155±0.022 (ranging from 0.055 to 0.231) ppm, 0.139±0.007 (ranging from 0.079 to 0.241) ppm and 0.153±0.008 (ranging from 0.077 to 0.239) ppm, respectively.

The mean serum manganese concentration of suboestrous Surti buffaloes did not differ significantly at 0 hr, 24 hr, 48 hr and 72 hr within and between all the treatment and control groups including overall means between the groups at different time intervals.

The trend of circulatory levels of manganese in post-partum $PGF_2\alpha$ treated and control groups compared well with the observations of Khasatiya (2003), who reported that the pooled overall mean plasma manganese values did not vary significantly between $PGF_2\alpha$ treatment and control groups (0.12±0.01 vs. 0.11±0.01 ppm). Similarly, Deshpande (2007) recorded non-significant variation among mean serum manganese levels in suboestrous treated (0.15±0.02 μg/ml), control (0.13±0.04 μg/ml) and normally cyclic (0.22±0.04 μg/ml) crossbred cows.

From the present findings, the overall mean serum manganese concentration values in suboestrus buffaloes were found to be higher (0.153±0.008 μg/ml), when it compared with the anoestrus values (0.043±0.39 μg/ml) reported by Chandolia and Verma (1987) might be due to cyclic nature of suboestrous buffaloes and at the same time lower value might be prone to anoestrus condition as it is true from the present comparison.

Moreover, microelements can't be synthesizes in the body. Hence, it is concluded that trace elements should be daily supplied in the field and in organized farms as mineral mixture to suffice the requirement of the trace elements. From the available literature (Rogers, 1992), it has been found that dairy animals frequently affected with varying degree of trace element deficiencies, especially Cu, Co, Zn and Mn in various regions of the world and the imbalance leads to inactive ovaries with decreased progesterone production by corpus luteum.

Trace elements may function as cofactors, as activators of enzymes, or as stabilizers of secondary molecular structure. Hidiroglou (1979) suggested that reproductive failure may be induced by deficiencies of single or combined trace elements and by imbalances.

In the study, we could not find differences in various serum biochemical parameters between treated and control groups at different time intervals.

Table 5 : Serum manganese concentration (ppm) pattern at different time intervals/days in sub-estrus treated and control groups of animals (Mean±SE)

Time intervals/ days	Groups (n=6)			
	$PGF_{2\alpha}$ (T_1)	Vit+ P (T_2)	$PGF_2\alpha$+vit+ P (T_3)	Control (T_4)
0 hr/0th Day	$0.145\pm0.007_a^w$	$0.139\pm0.022_a^w$	$0.139\pm0.007_a^w$	$0.154\pm0.008_a^w$
24 hr/1st Day	$0.147\pm0.007_a^w$	$0.204\pm0.022_a^w$	$0.139\pm0.007_a^w$	$0.153\pm0.008_a^w$
48 hr/2nd Day	$0.147\pm0.007_a^w$	$0.140\pm0.022_a^w$	$0.139\pm0.007_a^w$	$0.152\pm0.008_a^w$
72 hr/3rd Day	$0.148\pm0.007_a^w$	$0.139\pm0.022_a^w$	$0.139\pm0.007_a^w$	$0.153\pm0.008_a^w$
Overall	$0.146\pm0.007_a$	$0.155\pm0.022_a$	$0.139\pm0.007_a$	$0.153\pm0.008_a$

Means bearing common superscripts within a column (group) and means bearing common subscripts within a row (between the groups) do not differ significantly (p>0.05)

Acknowledgement:

We thank Principal and Dean, Veterinary College and Research Scientist Livestock Research Station, for their permission and funds release for conduct the present research work as well as Professor and Head, Department of Physiology and Biochemistry, Veterinary College, Navsari for providing technical help in conducting biochemical analysis

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