



Efficacy of Hormonal Protocols on Haematological and Micro-minerals Parameters in Postpartum Anestrus Cows

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ABSTRACT

Thirty-six post partum anestrus cows were selected for the present research work. Group I (n=12) were treated with CIDR based protocols and Group II (n=12) were administered Ovsynch protocol and Group III (n=12) was kept as an untreated control. In Group I, II and III the values of Hematological parameters like Hb (9.00 ± 0.17 , 9.15 ± 0.15 and 8.65 ± 0.30 , 8.66 ± 0.20 , 8.73 ± 0.20 and 8.25 ± 0.20 , 7.76 ± 0.16 , 7.69 ± 0.32 and 8.18 ± 0.18 g/dl) RBC (7.28 ± 0.37 , 9.15 ± 0.15 and 8.65 ± 0.30 , 7.20 ± 0.38 , 8.73 ± 0.20 and 8.25 ± 0.33 , 5.77 ± 0.32 , 7.69 ± 0.32 and 8.18 ± 0.18 $\times 10^{12}/l$) and TLC (10.36 ± 0.26 , 10.49 ± 0.25 and 9.77 ± 0.40 , 9.67 ± 0.28 , 9.90 ± 0.47 and 8.08 ± 0.57 , 8.29 ± 0.53 , 7.98 ± 0.54 and 8.63 ± 0.44 $\times 10^9/l$) on Day 0, 7 and 25 were recorded in Group I, II and III, respectively. Microminerals parameters ($\mu g/dl$) like Iron (147.9 ± 4.44 , 147.9 ± 4.44 and 150.1 ± 7.84 , 143.4 ± 4.5 , 158.2 ± 4.65 and 149.4 ± 7.71 , 118.6 ± 3.23 , 118.6 ± 3.12 and 118.4 ± 3.04) Cobalt (1.8 ± 0.14 , 2.2 ± 0.10 and 1.99 ± 0.11 , 1.7 ± 0.10 , 2.1 ± 0.08 and 1.74 ± 0.13 , 1.6 ± 0.12 , 1.5 ± 0.10 and 1.6 ± 0.11) Copper (0.65 ± 0.02 , 0.78 ± 0.03 and 0.71 ± 0.04 , 0.63 ± 0.01 , 0.77 ± 0.03 and 0.69 ± 0.03 , 0.64 ± 0.02 , 0.63 ± 0.02 and 0.65 ± 0.02) and Zinc (87.6 ± 1.18 , 104.9 ± 2.00 and 96.4 ± 3.27 , 86.7 ± 1.22 , 104.7 ± 2.02 and 93.6 ± 2.51 , 85.5 ± 1.03 , 85.6 ± 1.02 and 86.5 ± 0.92) on Day 0, 7 and 25 were recorded in Group I, II and III, respectively. It can be concluded that the use of different hormonal protocols in postpartum anestrus cows has shown different effects on haematological and micro-minerals parameters.

Key words: Postpartum anestrus cows, CIDR, serum Progesterone, biochemical parameters, PGF₂ α and GnRH

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INTRODUCTION

Anestrus resulting from delayed postpartum conception in cows and delayed onset of cyclicity in heifers is the major

contributor to infertility in Indian cattle, which accounts for poor reproductive efficiency of Zebu cattle. It leads to increased calving interval and hinders milk and meat production (Roche et al., 1992).

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The presynchronization protocol based on PGF2 α developed by Moreira et al., (2001) has limited efficacy in anestrus cows, because response to injections of PGF2 α depends on the presence of a responsive corpus luteum (CL). Ramakrishna, (1997) noted as low level of Hb indicates long term protein deficiency and it influences tissue oxygenation of reproductive tract which affects the induction of cyclicity.

Deficiency of copper might be associated with delayed and depressed estrus. Copper deficiency might have effect on reproduction probably through an interaction between copper. (Hidiroglou, 1979). Considering the common problem of infertility in post partum cows due to deficient nutrients, the research programme was conducted.

MATERIALS AND METHODS

Thirty-six post partum anestrus cows were selected on the basis of reproductive history and twice gynaeco-clinical examinations with the interval of 7 days. The cows were selected Instructional Livestock Farm Complex, Cattle Breeding Farm, College of Veterinary and Animal Sciences, Udgir and adjoining villages. Cows were fed a ration consisting of concentrates, roughages, mineral mixture and salt. Hematological and microminerals studies were attempted before and after treatment. The selected cows were divided in to three equal groups, Group I (n=12) cows were subjected to intra vaginally CIDR implant on day 0, CIDR removal on day 7 and followed by intramus-

cular inj. Cloprostenol @ 2ml (250mcg/ml) on day 7. Inj. GnRH (Buserelin Acetate) @ 5ml (4mcg/ml) on day 9 was given intramuscularly. Group II (n=12) cows were treated with inj. GnRH (Buserelin Acetate) 4mcg/ml on day 0, inj. Cloprostenol @ 2ml (250mcg/ml) on day 7 and inj. GnRH (Buserelin Acetate) @ 2.5ml (4mcg/ml) on day 9. Group III (n=12) cows were kept as untreated control. All the treated cows were kept under observation. Haemoglobin was estimated by using Sahli's haemometer method (Sastry, 1983) and expressed in g/dl. RBC was estimated by Blood is diluted in a special pipette with whose osmotic pressure is great enough to prevent haemolysis of erythrocytes. TLC was estimated by diluted blood in a specific pipette with WBC diluting fluid which haemolizes erythrocytes but does not alter the leucocytes. Iron, Cobalt, Copper and Zinc was estimated from serum sample, which was diluted to 1:20 with the de-ionized water as per method described by Fernandez and Khan (1971) by using AAS-Atomic Absorption Spectrophotometer (Perkin Elmer A Analyst 100).The data generated was tabulated by using ANOVA One Way Factor Statistical Package for Social Sciences software (SPSS 21.0) and means were compared by Duncan's multiple range test and interpretations were drawn.

RESULTS AND DISCUSSION

Values of hematological and microminerals parameters are presented in Table 1 and 2.

Table 1: Serum progesterone and biochemical parameters in postpartum anestrus cows

Parameters	Group	Day 0	Day 7	Day 25	P Value
Serum Haemoglobin	I	9.00 ^b ±0.17	9.15 ^b ±0.15	8.65±0.30	0.27
	II	8.66 ^b ±0.20	8.73 ^{ab} ±0.20	8.25±0.20	0.32
	III	7.76 ^a ±0.16	7.69 ^a ±0.32	8.18±0.18	0.27
Serum Red blood cell	I	7.28 ^a ±0.37	9.15 ^b ±0.15	8.65±0.30	0.19
	II	7.20 ^a ±0.38	8.73 ^{ab} ±0.20	8.25±0.33	0.11
	III	5.77 ^a ±0.32	7.69 ^a ±0.32	8.18±0.18	0.27
Serum Total leucocyte count	I	10.36 ^b ±0.26	10.49 ^b ±0.25	9.77 ^a ±0.40	0.23
	II	9.67 ^{ab} ±0.28	9.90 ^{ab} ±0.47	8.08 ^a ±0.57	0.07
	III	8.29 ^a ±0.53	7.98 ^a ±0.54	8.63 ^{ab} ±0.44	0.66
Serum Iron	I	147.9 ^b ±4.44	147.9 ^b ±4.44	150.1 ^b ±7.84	0.35
	II	143.4 ^b ±4.5	158.2±4.65	149.4 ^b ±7.71	0.21
	III	118.6 ^a ±3.23	118.6±3.12	118.4 ^a ±3.04	0.9
Serum Cobalt	I	1.8±0.14	2.2 ^b ±0.10	1.99 ^a ±0.11	0.07
	II	1.7±0.10	2.1 ^b ±0.08	1.74 ^{ab} ±0.13	0.02
	III	1.6±0.12	1.5 ^a ±0.10	1.6 ^a ±0.11	0.96
Serum Copper	I	0.65±0.02	0.78 ^b ±0.03	0.71±0.04	0.01
	II	0.63±0.01	0.77 ^b ±0.03	0.69±0.03	0.04
	III	0.64±0.02	0.63 ^a ±0.02	0.65±0.02	0.85
Serum Zinc	I	87.6±1.18	104.9±2.00	96.4 ^b ±3.27	0.00
	II	86.7±1.22	104.7±2.02	93.6 ^{ab} ±2.51	1.43
	III	85.5±1.03	85.6±1.02	86.5 ^a ±0.92	0.72

*Means bearing different superscripts between the groups differ significantly

*Mean with same superscripts the overall effect significant, but within the group is not significant from each other

Table 2: Values of Parameter of serum progesterone and biochemical parameters in postpartum anestrus cows.

Parameters	Status	Group	Day 0	Day 7	Day 25	P Value
Serum Haemoglobin	Pregnant	I	9.1±0.21	9.3±0.20	9.36±0.22	0.82
		II	9.0±0.21	9.12±0.20	9.28±0.20	0.72
		III	--	--	--	--
	Non Pregnant	I	8.8 ^a ±0.32	9.0 ^a ±0.21	7.4 ^a ±0.23	0.00
		II	8.3 ^a ±0.26	8.4 ^a ±0.11	7.22 ^a ±0.16	0.00
		III	7.8±0.16	7.7±0.32	8.2±0.18	1.38
Serum Red blood cell	Pregnant	I	7.68±0.46	8.07±0.36	8.18±0.33	0.25
		II	7.72±0.61	7.80±0.61	7.80±0.64	0.99
		III	--	--	--	--
	Non Pregnant	I	6.5±0.45	6.7±0.31	5.5±0.34	0.06
		II	6.68±0.39	6.77±0.37	5.60±0.31	0.06
		III	5.77±0.32	5.55±0.28	6.28±0.36	0.27
Serum Total leucocyte count	Pregnant	I	10.42±0.36	10.38±0.35	10.61±0.25	0.86
		II	9.68±0.50	9.43±0.75	9.45±0.69	0.9
		III	--	--	--	--
	Non Pregnant	I	10.23 ^a ±0.40	10.72 ^a ±0.26	8.09 ^a ±0.14	0.00
		II	9.66 ^a ±0.32	9.36 ^a ±0.36	7.80 ^a ±0.30	0.00
		III	8.29±0.53	7.98±0.54	8.63±0.44	0.66
Serum Iron	Pregnant	I	149.8±5.94	166.3±4.77	165.5±4.88	0.06
		II	143.1 ^a ±6.84	164.2 ^b ±3.99	164.56 ^b ±4.10	0.01
		III	--	--	--	--
	Non Pregnant	I	144.1±6.69	145.7±6.69	119.5±9.80	0.07
		II	143.6 ^{ab} ±6.67	152.1 ^b ±8.01	126.2 ^a ±4.49	0.03
		III	118.6±3.23	118.6±3.12	118.4±3.04	0.9
Serum Cobalt	Pregnant	I	1.99±0.09	2.2±0.13	2.09±0.09	0.34
		II	1.88±0.11	2±0.13	2.05±0.12	0.59
		III	--	--	--	--
	Non Pregnant	I	1.9±0.32	2.1±0.19	1.8±0.25	0.59
		II	1.52 ^a ±0.13	1.95 ^b ±0.05	1.47 ^a ±0.12	0.01
		III	1.55±0.12	1.54±0.10	1.58±0.10	0.9
Serum Copper	Pregnant	I	0.63 ^a ±0.02	0.77 ^a ±0.04	0.77 ^a ±0.04	0.01
		II	0.61±0.02	0.70±0.03	0.7±0.04	0.07
		III	--	--	--	--
	Non Pregnant	I	0.62±0.03	0.66±0.03	0.62±0.03	0.6
		II	0.60±0.01	0.64±0.64	0.61±0.01	0.2
		III	0.64±0.02	0.63±0.02	0.65±0.02	0.85
Serum Zinc	Pregnant	I	87.4 ^a ±1.34	103.0 ^a ±2.71	103 ^a ±2.53	0.00
		II	87.1 ^a ±1.66	100.61 ^a ±1.48	100.63 ^a ±2.66	0.00
		III	--	--	--	--
	Non Pregnant	I	86.1 ^a ±2.30	91.1 ^a ±3.26	83.5 ^a ±2.05	0.00
		II	86.2±1.93	100±2.87	86.6±0.91	1.6
		III	85.5±1.03	85.57±1.02	86.5±0.92	0.72

*Mean with different superscripts between the groups differ significantly

*Mean with same superscripts the overall effect significant, but within the group is not significant from each other

Serum Haemoglobin

The mean Hb (g/dl) concentration in Group I on day 0, 7 and day 25 were found as 9.00 ± 0.17 , 9.15 ± 0.15 and 8.65 ± 0.30 , respectively. The difference between on day 0, 7 and 25 were non-significant within group ($P > 0.05$). The present findings are close proximity with Ramkrishana (1997) who recorded hemoglobin level in 35 normal cyclical and 35 cross bred anestrus Jersey cows and noted as 9.1 ± 0.80 and 10.43 ± 0.031 g/dl in anestrus and normal cycling cross bred cows, respectively whereas Jayanti et al. (2003) who observed 9.63 ± 0.9 gm/dl which is significantly lower Hb in anestrus cows.

In Group II mean Hb (g/dl) concentration on day 0, 7 and day 25 were recorded as 8.66 ± 0.20 , 8.73 ± 0.20 and 8.73 ± 0.20 , respectively. The difference between on day 0, 7 and 25 were found as non-significant in group ($P > 0.05$). The present findings are close agreement with Ahmad et al. (2003) who observed for haematological parameters including haemoglobin concentration (Hb). The results revealed that the values of Hb were recorded as significantly lower ($P < 0.05$) in the non-cyclic cows as compared to cyclic animals. The mean values of Hb, (g/dl) as significantly ($P < 0.05$) lower in the non-cyclic (9.26 ± 0.34) cows as compared to cyclic (12.41 ± 0.44) cows. In Group III, mean Hb (g/dl) concentration on day 0, 7 and day 25 were 7.76 ± 0.16 , 7.69 ± 0.32 and 8.18 ± 0.18 , respectively. The difference between on day 0, 7 and 25 were found as non-significant in group ($P > 0.05$).

Serum Red blood cell

In the present study, the mean Red blood cell ($\times 10^{12}/l$) concentration in Group I on day 0, 7 and day 25 were recorded as 7.28 ± 0.37 , 9.15 ± 0.15 and 8.65 ± 0.30 , respectively. The difference between on day 0, 7 and 25 were non-significant in group ($P > 0.05$). The present findings are close proximity with Kandasamy et al. (2018) who studied haematological and biochemical profile of lactating kangeyam cows and found red blood cells count ($10^6/\mu l$) as 7.68 ± 0.24 whereas Sarkar et al. (2016) who observed the mean values of total erythrocyte count in experimental and control group of cows as $6.16 \pm 0.79 \times 10^6/\mu l$ and $7.65 \pm 0.14 \times 10^6/\mu l$ and in case of non-descriptive and crossbred animals the count as $6.93 \pm 0.14 \times 10^6/\mu l$ and $6.88 \pm 0.08 \times 10^6/\mu l$, respectively. In Group II, mean Red blood cell ($\times 10^{12}/l$) concentration on day 0, 7 and day 25 were found to be 7.20 ± 0.38 , 8.73 ± 0.20 and 8.25 ± 0.33 , respectively. The difference between on day 0, 7 and 25 were non-significant in group ($P > 0.05$).

The present findings are close proximity with Sattar and Mirza (2009) who found the highest RBC count $6.18 \pm 0.47 (\times 10^6/\mu l)$ in non-pregnant heifers whereas,

Klinkon and Zadnik (1999) who observed mean erythrocyte (RBC) count $6.70 \pm 0.65 (\times 10^6/\mu l)$ at parturition in Black and White dairy cows.

In Group III, mean Red blood cell ($\times 10^{12}/l$) concentration in on day 0, 7 and day 25 were recorded as 5.77 ± 0.32 , 7.69 ± 0.32 and 8.25 ± 0.33 , respectively. The difference between on day 0, 7 and 25 were non significant on group ($P > 0.05$)

Serum total leucocyte count

In the present study the mean Total leucocyte count ($\times 10^9/l$) concentration in Group I, on day 0, 7 and day 25 were 10.36 ± 0.26 , 10.49 ± 0.25 and 9.77 ± 0.40 , respectively. The difference between on day 0, 7 and 25 were non-significant in group ($P > 0.05$).

The present findings are close proximity with Sarkar et al. (2016) who observed mean values of Total Leukocyte Count (TLC) in experimental and control group were found as $10.43 \pm 0.143 \times 10^3/\mu l$ and $8.75 \pm 0.261 \times 10^3 /\mu l$, respectively whereas Emad et al. (2019) who estimated hematological parameters and reported as significant increase in TLC (6.46 ± 0.56 and $10.34 \pm 0.87 \times 10^3/\mu l$) in normal cyclic and SIO groups respectively. The smooth inactive ovaries in buffalo-cows were characterized by an increase in TLC.

In Group II, mean TLC ($\times 10^9/l$) concentration on day 0, 7 and day 25 were recorded as 9.67 ± 0.28 , 10.49 ± 0.25 , and 8.08 ± 0.57 , respectively. The difference between on day 0, 7 and 25 were non-significant in group ($P < 0.05$).

The present findings of the present study are close proximity with Ahmad et al. (2003) who observed for haematological parameters total leukocytic count (TLC) and reported as significantly lower ($P < 0.05$) in the non-cyclic cows as compared to cyclic or endometritic animals. Also, they noted as TLC higher in cyclic cows ($10^3/\mu l$) 9.31 ± 0.38 than non-cyclic 8.96 ± 0.51 whereas Devipriya et al. (2016) who studied haematological parameters as total leukocytes count (TLC $\times 10^3/\text{mm}^3$) values 7.64 ± 0.26 vs. 7.92 ± 0.91 in cyclic ($n=10$) and summer anestrus buffaloes respectively, which values are lesser than the present study.

In Group III, mean TLC ($\times 10^9/l$) concentration on day 0, 7 and day 25 were 8.29 ± 0.53 , 7.98 ± 0.54 and 8.63 ± 0.44 , respectively. The difference between on day 0, 7 and 25 were non-significant in group ($P > 0.05$).

Serum Iron

In the present study, mean Iron ($\mu\text{g}/\text{dl}$) concentration in Group I on day 0, 7 and 25 were recorded as 147.9 ± 4.44 ,

147.9±4.44 and 150.1±7.84, respectively. The difference between on day 0, 7 and 25 were non-significant in group. ($P>0.05$).

The present findings are close agreement with Vadnere and Singh (1989) who observed 21 postpartum anestrus crossbred cows the serum iron levels as 194.30±8.35 µg/dl in anestrus cows, which were significantly lower than normal cyclic cows.

In Group II, mean Iron (µg/dl) concentration on day 0, 7 and day 25 were investigated as 143.4±4.5, 158.2±4.65 and 118.4±3.04, respectively. The difference between on day 0, 7 and 25 were non-significant in group ($P>0.05$).

The present findings are close agreement with Ramakrishna (1997) who observed significantly lower serum iron concentration in the anestrus crossbred rural cows (86.33±6.27µg/dl) than the normal cycling group as 129.83±9.73µg/dl.

In Group III, mean Iron (µg/dl) concentration on day 0, 7 and day 25 were 118.6±3.23, 118.6±3.12 and 8.18±0.18, respectively. The difference between on day 0, 7 and 25 were non-significant in group ($P>0.05$).

Seum Cobalt

In the present study, the mean Cobalt (µg/dl) concentration in Group I on day 0, 7 and day 25 were found to be 1.8±0.14, 2.2±0.10 and 1.99±0.11, respectively. The difference between on day 0, 7 and 25 were non-significant in group ($P>0.05$).

The present findings are close results with Prasad et al. (1989) who found serum cobalt level in 50 crossbred cows as 1.05±0.3µg/ml in prolonged postpartum anestrus cows and 0.5±0.1µg/ml in acyclic cows. Jayachandran et al. (2013) recorded anestrus and regular cyclic values as Cobalt (µg/ml) (0.480 ± 0.03 and 0.551 ± 0.15), respectively.

In Group III, mean Cobalt (µg/dl) concentration on day 0, 7 and day 25 were recorded as 1.6±0.12, 1.5±0.10 and 1.6±0.11, respectively. The difference between on day 0, 7 and 25 were non-significant in group ($P>0.05$).

Serum Copper

In the present study, the mean Copper (µg/dl) concentration in Group I on day 0, 7 and day 25 were recorded as 0.65±0.02, 0.78±0.03 and 0.71±0.04, respectively. The difference between on day 0, 7 and 25 were significant in group ($P<0.05$).

The present findings are close proximity with Prasad and Rao, (1997) who found subnormal serum copper level in anestrus cows and in that of normal cows to be 0.87µg/ml.

In Group II, mean Copper (µg/dl) concentration on day 0, 7 and day 25 were found to be 0.63±0.01, 0.77±0.03 and 0.69±0.03, respectively. The difference between on day 0, 7 and 25 were significant in group ($P<0.05$).

The present findings are close agreement with Chandrakar et al. (2002) observed serum copper level in normal fertile cows as 0.88±0.10µg/ml, while in repeat breeding cows as 0.53±0.04µg/ml.

In Group III, mean Copper (µg/dl) concentration on day 0, 7 and day 25 were observed as 0.64±0.02, 0.63±0.02 and 0.65±0.02, respectively. The difference between on day 0, 7 and 25 were non-significant in group ($P>0.05$).

Serum Zinc

In the present study, the mean zinc (µg/dl) concentration in Group I on day 0, 7 and 25 were recorded as 87.6±1.18, 104.9±2.00 and 96.4±3.27, respectively. The difference between on day 0, 7 and 25 were significant in group ($P<0.01$).

The present findings are close agreement with Underwood (1977) who reported zinc level content between 0.9 to 2.5ppm, 0.8 to 1.2ppm as a normal range in cattle whereas Saxena and Gupta (1995) who observed significantly lower levels of serum zinc (115.55±12.17µg/dl) in postpartum anestrus cows than the cows returning to estrus after calving (203.75±22.26µg/dl).

In Group II, mean Zinc (µg/dl) concentration on day 0, 7 and 25 were found to be 86.7±1.22, 104.7±2.02 and 93.6±2.51, respectively. The difference between on day 0, 7 and 25 were non-significant in group ($P>0.05$).

The present findings are close proximity with Underwood, (1977) who reported zinc levels between 0.9 to 2.5ppm, 0.8 to 1.2ppm as a normal range in cattle.

In Group III, mean Zinc (µg/dl) concentration on day 0, 7 and 25 were recorded to be 85.5±1.03, 85.6±1.02 and 86.5±0.92, respectively. The difference between on day 0, 7 and 25 were non-significant in group ($P>0.05$).

CONCLUSION

Haematological parameters observed significant difference between treatment and control group was observed in hematological parameters on day 0 and 25.

However, significant difference in values of haematological parameters was observed between pregnant and non pregnant animals on day 25. Micro minerals such as Fe, Cu, Co and Zn no significant alteration in levels observed with the administration of hormonal protocols in postpartum anestrus cows. However, significant difference in values of macro minerals was observed between pregnant and non pregnant animals on day 25.

CONFLICT OF INTERESTS

There is no conflict of interest.

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