Haryana Vet. (December, 2023) **62(2),** 78-83 ISSN 0033-4359 (P) **Research Article**

EFFECT OF HORMONAL THERAPY AND BLOOD CONSTITUENTS IN REPEAT BREEDER DAIRY COWS

C.K. LAKDE*, S.K. SAHATPURE, A.D. PATIL, S.V. UPADHYE and A.P. GAWANDE
Department of Animal Reproduction Gynaecology and Obstetrics,
Nagpur Veterinary College, Maharashtra Animal & Fishery Sciences University, Seminary Hills, Nagpur-440 006, India

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ABSTRACT

The study was conducted to evaluate the effect of hormonal therapies and blood constituents in repeat breeder dairy cows. Cows were randomly divided into six groups. Group 1 (CIDR 5-9 days post AI intravaginally n=10), Group 2 (CIDR 5-13 days post AI intravaginally; n=10), Group 3 (GnRH at 20 µg IM, n=10), Group 4 (hCG at 2000 IU IM; n=10), Group 5 (Dextrose 5% IV 2 liters; n=10) and Group 6 (Control; n=10). The treatments were given on 5th day post breeding in Group 1-4 and in Group 5 on day of AI. The level of serum Progesterone and Serum Glucose, Calcium, Phosphorus, Magnesium, Cholesterol, Total protein, Copper, Cobalt, Zinc, Iron and Manganese were estimated. It was concluded that the level of progesterone in pregnant cows was nearly same on day 21 after AI in GnRH and hCG treated cows.

Keywords: CIDR, Gonadotropin-releasing hormone, Human chorionic gonadotropin, Repeat-breeder

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Most of the embryonic mortality about 70-80% occurs between days 8 and 16 after insemination and luteal insufficiency is a known cause of repeat breeding. Early stages of pregnancy, luteal deficiency and low level of plasma progesterone may cause reproductive failure before and/or after insemination. A delayed rise in progesterone level and lower total progesterone concentrations are the most prominent endocrine alternations in repeat-breeding cow. Cows with high progesterone concentrations have larger embryos than those of cows with lower progesterone levels on day 16. Progesterone is essential for orchestrating the histotrophic environment for the nourishment of the conceptus (Santos *et al.*, 2004). Low progesterone concentrations during early pregnancy may affect embryonic development and maternal recognition of pregnancy. Lower circulating progesterone concentrations from day 3-8 post-ovulation are associated with smaller embryos on Day 16, and consequently, may result in ineffective interferon-tau secretion to block the luteolytic process and to maintain pregnancy (Mann and Lamming, 2001).

High incidence of repeat-breeding and anestrous are associated with the deficiencies of cholesterol, glucose, albumin etc. The use of the hormonal substances is still one of the useful treatments to decrease the delay of the ovulation together with AI, thus increases the conception rate in dairy cows (Turmalaj *et al.*, 2014).

MATERIALS AND METHODS

Repeat-breeder dairy cows (3-6 years) were maintained at individual owner place or private dairy farm (n=60). All animals were thoroughly examined per-rectally to rule out any anatomical defect of genitalia and ovarian abnormalities. Dairy animals were randomly divided into six groups: Group 1 (CIDR 5-9 days Post AI intravaginally; n=10), Group 2 (CIDR 5-13 days Post AI intravaginally; n=10), Group 3 (GnRH at 20 µg IM; n=10), Group 4 (hCG at 2000 IU IM; n=10), Group 5 (Dextrose 5% IV 2 liters; n=10) and Group 6 (Control; n=10). The treatments were given on 5th day post breeding in Group 1-4 and in Group 5 on day of AI. Blood samples were collected on day of AI and 5, 13, 21 of post A.I. Serum samples progesterone hormone and Serum Glucose, Calcium, Phosphorus, Magnesium, Cholesterol, Total protein, Copper, Cobalt, Zinc, Iron and Manganese were estimated.

*Corresponding author: dr.chetanlakde@gmail.com

RESULT AND DISCUSSION

On day 0 and 5th, the difference in variation was found to be non-significant. But on day 13 and 21, significant difference was observed in pregnant animals. Higher level of progesterone on day 21 as compared to non-pregnant animal within each group. In group III and IV, mean serum progesterone level was found to higher than rest of groups in pregnant animal on day-21. In non-pregnant animal, non-significant changes were observed in all treated animals. It was found that in group I on day 13 and 21, mean serum progesterone increase significantly as compared with day 5. Similarly, in group II, III, IV and V was observed same pattern of increasing progesterone level on day 13 and 21 as compared day 0 and 5. Significant variation was observed in each group between day 0 and 21. It was also found on day 13, significantly increased progesterone level than day 0, 5 and 21 in non-pregnant experimental cows (Table 1). The findings are in

Table 1. Mean Serum Progesterone level (ng/dl)

Days/ Groups		Group I (CIDR D 5-9)	Group II (CIDR D 5-13)	Group III (GnRH)	Group IV (hCG)	Group V (Dextrose)	Group VI (Control)	
0 day	Pregnant	0.27^{B}±0.12	0.27^{B}±0.04	0.31±0.07	0.34^{C}±0.04	0.42^{B}±0.02	0.25±0.00	NS
	Non Pregnant	0.37^{B}±0.03	0.36^{C}±07	0.30±0.03	0.30^{C}±0.03	0.35^{C}±0.03	0.32^{C}±0.03	NS
5 day	Pregnant	1.90^{B}±0.08	2.40^{B}±0.24	2.04±0.10	1.92^{C}±0.06	2.1^{B}±0.14	1.96±0.00	NS
	Non Pregnant	1.34^{B}±0.03	2.21^{B}±0.21	1.55±0.12	1.93^{B}±0.13	1.83^{B}±0.09	1.68^{B}±0.08	NS
13 day	Pregnant	5.81^{A}±0.52^{a}	8.12^{A}±0.38^{a}	7.15^{B}±0.39^{b}	7.97^{B}±0.25^{b}	5.08^{A}±0.3^{a}	4.42±0.00^{b}	1.98
	Non Pregnant	4.27^{A}±0.24	8.16^{A}±0.28	5.53^{A}±0.36	4.81^{A}±0.31	3.69^{A}±0.19	3.55^{A}±0.20	NS
21 day	Pregnant	6.87^{A}±0.44^{b}	9.13^{A}±0.41^{a}	10.82^{A}±0.56^{a}	10.50^{A}±0.44^{a}	6.14^{A}±0.50^{b}	5.04±0.00^{b}	2.64
	Non Pregnant	0.43^{B}±0.03	2.23^{B}±0.38	0.99^{B}±0.14	1.27^{B}±0.28	0.90^{B}±0.17	0.69^{B}±0.10	NS

Table 2. Mean Serum Glucose level (mg/dl)

Days/ Groups		Group I (CIDR D 5-9)	Group II (CIDR D 5-13)	Group III (GnRH)	Group IV (hCG)	Group V (Dextrose)	Group VI (Control)	
0 day	Pregnant	82.60±5.06	69.80±2.77	70.45±15.05	69.94±2.62	72.00±4.00	60.2±0.00	NS
	Non Pregnant	73.97±0.95	69.55±2.55	60.29±1.60	62.67±5.11	76.10±1.30	62.46±1.19	6.60
5 day	Pregnant	77.33±1.96	73.32±3.81	69.5±11.5	71.73*±1.19	70.40±8.00	65.0±0.00	NS
	Non Pregnant	74.46±1.48	70.32±1.80	61.11±1.87	62.53±2.43	73.31±2.05	61.36±2.39	7.39
13 day	Pregnant	75.00±0.95	71.93±1.62	74.2±10.2	70.80±2.44	66.40±9.60	69.7±0.00	NS
	Non Pregnant	74.23±2.54	68.68±1.47	63.14±1.48	64.60±2.21	72.55±1.24	64.52±1.51	7.15
21 day	Pregnant	80.57±0.90	72.20±1.13	77.1*±4.3	71.83*±1.62	74.40±3.80	59.4±0.00	7.43
	Non Pregnant	77.40±1.71	72.88±2.38	64.53±1.31	64.60±2.95	71.53±1.48	66.24±1.43	6.67

Table 3. Mean Serum Calcium level (mg/dl)

Days/ Groups		Group I (CIDR D 5-9)	Group II (CIDR D 5-13)	Group III (GnRH)	Group IV (hCG)	Group V (Dextrose)	Group VI (Control)	
0 day	Pregnant	11.43 ±0.68	10.50±0.61	11.82±1.48	10.63±0.63	8.58±0.32	13.0±0.00	NS
	Non Pregnant	9.90±0.37	9.95±0.43	10.99±0.58	9.66±0.34	10.70±0.40	10.70±0.41	NS
5 day	Pregnant	11.93 ± 1.20	10.00±0.50	9.95±1.65	9.44±0.34	8.46±0.46	12.6±0.00	NS
	Non Pregnant	10.19±0.45	10.79±0.45	11.07±0.46	9.92±0.92	9.90±0.37	10.70±0.39	NS
13 day	Pregnant	10.40 ±0.87	11.13±0.59	9.60±1.22	11.05±0.64	9.24±0.36	12.0±0.00	NS
	Non Pregnant	10.02±0.47	10.70±0.71	10.82±0.30	9.80±0.31	11.44±0.61	10.88±0.34	NS
21 day	Pregnant	10.80± 1.14	10.87±0.57	11.30±1.50	10.44±0.51	8.75±0.65	11.6±0.00	NS
	Non Pregnant	10.04±0.39	11.30±0.69	10.65±0.44	11.37±0.67	11.07±0.48	11.42±0.47	NS

Table 4. Mean Serum Phosphorus level (mg/dl)

Days/ Groups		Group I (CIDR D 5-9)	Group II (CIDR D 5-13)	Group III (GnRH)	Group IV (hCG)	Group V (Dextrose)	Group VI (Control)	
0 day	Pregnant	5.13±0.05	5.59±0.27	6.15± 0.65	5.32±0.16	4.51±0.99	5.77± 0.00	NS
	Non Pregnant	5.66±0.19	5.84±0.21	5.40±0.20	5.35±0.43	5.74±0.13	5.39±0.16	NS
5 day	Pregnant	5.86±0.11	5.29±0.30	6.7± 0.30	5.47±0.14	5.02±1.06	5.95± 0.00	NS
	Non Pregnant	5.73±0.12	5.39±0.25	5.27±0.16	4.77±0.59	5.65±0.13	5.62±0.20	NS
13 day	Pregnant	5.35±0.23	5.61±0.23	5.66 ±0.34	5.47±0.12	4.87±0.33	5.89± 0.00	NS
	Non Pregnant	5.81±0.13	5.38±0.17	5.36±0.17	4.88±0.24	5.75±0.14	5.60±0.24	NS
21 day	Pregnant	5.76±0.12	5.79±0.13	5.96± 0.06	5.49±0.16	5.08±0.89	5.68± 0.00	NS
	Non Pregnant	5.69±0.11	5.26±0.08	5.69± 0.24	4.89±0.18	5.72±0.11	5.60±0.22	NS

Table 5. Mean Serum Magnesium level (mg/dl)

Days/ Groups		Group I (CIDR D 5-9)	Group II (CIDR D 5-13)	Group III (GnRH)	Group IV (hCG)	Group V (Dextrose)	Group VI (Control)	
0 day	Pregnant	2.00±0.10	1.95±0.05	2.10±0.10	1.80±0.06	1.94±0.04	1.92±0.00	NS
	Non Pregnant	1.88±0.07	2.18±0.07	2.08±0.07	1.82±0.07	1.96±0.05	2.07±0.05	NS
5 day	Pregnant	2.06±0.24	1.98±0.14	2.01±0.20	1.88±0.07	1.88±0.04	2.1±0.00	NS
	Non Pregnant	1.85±0.04	2.03±0.04	1.94±0.04	1.85±0.08	1.94±0.03	2.01±0.04	NS
13 day	Pregnant	1.88±0.06	1.85±0.13	1.90±0.05	1.83±0.07	2.02±0.12	2.12±0.00	NS
	Non Pregnant	1.91±0.05	1.98±0.13	1.96±0.06	1.91±0.04	1.91±0.04	2.00±0.07	NS
21 day	Pregnant	2.06±0.11	2.19±0.12	1.85±0.15	1.94±0.05	1.89±0.02	1.93±0.00	NS
	Non Pregnant	1.88±0.07	2.03±0.11	2.05±0.03	1.93±0.17	1.88±0.06	1.96±0.07	NS

Table 6. Mean Serum Total Protein level (mg/dl)

Days/ Groups		Group I (CIDR D 5-9)	Group II (CIDR D 5-13)	Group III (GnRH)	Group IV (hCG)	Group V (Dextrose)	Group VI (Control)	
0 day	Pregnant	8.19±0.17	7.86±0.37	8.55±0.05	7.62±0.19	7.62±0.06	7.4±0.00	NS
	Non Pregnant	8.16±0.35	7.65±0.31	7.71±0.32	7.21±0.34	7.51±0.14	7.99±0.13	NS
5 day	Pregnant	7.82±0.35	6.00±0.37	7.75±0.27	6.55±0.20	5.81±0.20	7.6±0.00	1.31
	Non Pregnant	7.96±0.45	7.61*±0.55	7.25±0.26	7.06±0.47	6.90*±0.17	7.49±0.11	NS
13 day	Pregnant	8.14±0.46	7.51±0.57	8.08±0.47	7.11±0.35	6.80±0.40	7.9±0.00	NS
	Non Pregnant	7.90±0.27	7.75±0.52	7.30±0.26	7.30±0.43	7.19±0.14	7.30±0.19	NS
21 day	Pregnant	7.46±0.19	7.44±0.38	7.77±0.31	7.18±0.29	6.72±0.38	7.93±0.00	NS
	Non Pregnant	7.61±0.29	8.06±0.09	7.48±0.23	7.34±0.56	7.35±0.12	7.54±0.18	NS

Table 7. Mean Serum Cholesterol level (mg/dl)

Days/ Groups		Group I (CIDR D 5-9)	Group II (CIDR D 5-13)	Group III (GnRH)	Group IV (hCG)	Group V (Dextrose)	Group VI (Control)	
0 day	Pregnant	153.01±13.20	164.05± 9.88	147.56±28.89	126.03±6.76	114.37±30.75	158.19±0.00	NS
	Non Pregnant	146.53±5.87^{ab}	169.46±7.03^{a}	163.33±6.52^{a}	110.67±12.97^{b}	143.02±9.64^{ab}	166.35±12.53^{a}	40.06
5 day	Pregnant	157.15±7.94	185.52±16.17	134.33±17.77	134.61±14.51	110.47±28.22	161.81±0.00	NS
	Non Pregnant	150.30±7.29	154.99±21.02	165.78±5.87	118.02±18.96	140.09 ±11.54	166.82±12.95	NS
13 day	Pregnant	153.19±6.92	176.03±17.01	156.17±20.73	127.31±5.61	134.41±14.67	163.00±0.00	NS
	Non Pregnant	151.08±6.98	148.55±6.66	175.52±10.13	128.25±8.91	152.36±9.91	161.56±8.46	NS
21 day	Pregnant	166.70±11.15	175.23 ±19.30	168.91±5.89	138.56±7.82	154.26±21.56	178.4±0.00	NS
	Non Pregnant	154.02±7.28	170.17±15.70	159.55±10.17	122.85±0.88	150.61±9.10	154.21±6.22	NS

Table 8. Mean Serum Zinc level (µg/dl)

Days/ Groups		Group I (CIDR D 5-9)	Group II (CIDR D 5-13)	Group III (GnRH)	Group IV (hCG)	Group V (Dextrose)	Group VI (Control)	
0 day	Pregnant	141.21±2.09	136.14±1.17	137.00±0.26	131.91±0.45	136.18±3.44	132.83±0.00	5.01
	Non Pregnant	140.52±1.51	136.01±1.39	134.60±0.96	132.57±0.40	132.35±0.99	128.91±1.37	5.14
5 day	Pregnant	140.11*±1.50	133.84±1.60	132.20±0.53	131.01±1.01	124.25±2.38	129.23±0.00	6.05
	Non Pregnant	132.42±1.54	127.92±2.91	127.25±1.51	126.37±2.20	127.40±1.75	124.83±1.87	NS
13 day	Pregnant	134.23±2.94	134.36±1.67	133.47±0.69	131.54±1.61	122.80±4.26	127.48±0.00	NS
	Non Pregnant	129.26±2.05	127.28±3.13	128.64±1.28	126.84±1.20	123.45±1.04	124.02±1.40	NS
21 day	Pregnant	137.88*±0.44	135.09±2.38	129.26±0.11	131.94*±1.55	124.71±4.43	129.23±0.00	NS
	Non Pregnant	131.17±1.13	129.48±2.71	131.90±0.88	125.05±1.16	124.68±1.68	127.05±1.23	5.79

Table 9. Mean Serum Copper level (µg/dl)

Days/ Groups		Group I (CIDR D 5-9)	Group II (CIDR D 5-13)	Group III (GnRH)	Group IV (hCG)	Group V (Dextrose)	Group VI (Control)	
0 day	Pregnant	77.66±1.51	78.46±0.91	76.64±2.49	77.83±0.93	76.66±2.50	78.07±0.00	NS
	Non Pregnant	77.78±0.59	77.61±1.32	78.00±0.88	77.60±1.43	77.30±0.84	77.75±0.63	NS
5 day	Pregnant	76.36±0.31	77.52±0.61	77.89±1.98	78.38±0.31	77.84±1.00	76.95±0.00	NS
	Non Pregnant	74.02±0.90	73.15±1.37	74.17±0.80	71.87±2.45	75.49±1.03	75.52±0.52	NS
13 day	Pregnant	75.64±1.72	77.63±0.98	73.84±2.00	76.19±0.29	77.35±0.49	73.86±0.00	NS
	Non Pregnant	73.15±0.81	73.82±0.81	72.62±0.71	71.61±1.29	72.82±0.99	73.72±1.06	NS
21 day	Pregnant	77.43±0.30	76.98±0.96	74.44±4.40	77.68±0.53	77.78±1.14	79.93±0.00	NS
	Non Pregnant	73.58±1.46	72.60±2.21	73.13±1.15	74.84±1.52	73.80±1.38	73.87±0.92	NS

Table 10. Mean Serum Iron level(µg/dl)

Days/ Groups		Group I (CIDR D 5-9)	Group II (CIDR D 5-13)	Group III (GnRH)	Group IV (hCG)	Group V (Dextrose)	Group VI (Control)	
0 day	Pregnant	168.09±1.25	162.80±1.84	167.00±0.27	161.80±1.85	160.97±1.77	162.3±0.00	NS
	Non Pregnant	163.83±1.58	163.51±2.41	163.35±1.20	162.59±0.39	163.56±1.39	162.98±1.23	NS
5 day	Pregnant	156.74±5.19	159.26±2.56	132.20±0.53	166.51*±1.15	164.25±2.38	169.23±0.00	10.37
	Non Pregnant	163.94±1.96	162.18±1.60	127.25±1.51	159.13±2.33	161.11±1.61	162.42±1.63	7.09
13 day	Pregnant	160.92±0.38	159.36±2.57	133.47±0.69	161.40±1.56	167.80*±0.74	167.48±0.00	8.76
	Non Pregnant	160.70±1.34	162.31±1.51	128.64±1.28	166.78±1.15	162.20±0.63	160.46±1.87	5.90
21 day	Pregnant	167.80±0.40	163.42±1.64	129.26±0.11	164.63±1.69	164.71±4.43	159.23±0.00	7.81
	Non Pregnant	163.95±1.22	161.98±2.69	131.90±0.88	164.95±1.33	162.18±1.55	163.55±1.02	5.50

Table 11. Mean Serum Manganese level (ppm)

Days/ Groups		Group I (CIDR D 5-9)	Group II (CIDR D 5-13)	Group III (GnRH)	Group IV (hCG)	Group V (Dextrose)	Group VI (Control)	
0 day	Pregnant	0.32±0.01	0.34±0.01	0.32±0.02	0.34±0.01	0.31±0.01	0.31±0.00	NS
	Non Pregnant	0.34±0.01	0.32±0.01	0.33±0.01	0.30±0.01	0.31±0.01	0.32±0.01	NS
5 day	Pregnant	0.33±0.01	0.33±0.01	0.32±0.03	0.30±0.01	0.32±0.02	0.34±0.00	NS
	Non Pregnant	0.30±0.01	0.31±0.01	0.33±0.01	0.32±0.01	0.30±0.01	0.31±0.01	NS
13 day	Pregnant	0.33±0.01	0.31±0.01	0.31±0.01	0.33±0.01	0.33±0.01	0.31±0.00	NS
	Non Pregnant	0.30±0.01	0.33±0.01	0.30±0.01	0.30±0.01	0.32±0.01	0.31±0.01	NS
21 day	Pregnant	0.33±0.01	0.30±0.01	0.31±0.01	0.31±0.01	0.32±0.03	0.32±0.00	NS
	Non Pregnant	0.32±0.01	0.30±0.01	0.31±0.01	0.31±0.01	0.31±0.01	0.32±0.01	NS

Table 12. Mean Serum cobalt level (ug/ml)

Days/ Groups		Group I (CIDR D 5-9)	Group II (CIDR D 5-13)	Group III (GnRH)	Group IV (hCG)	Group V (Dextrose)	Group VI (Control)	
0 day	Pregnant	0.66±0.02	0.66±0.03	0.64±0.01	0.62±0.04	0.65±0.04	0.62±0.00	NS
	Non Pregnant	0.69±0.01	0.63±0.04	0.68±0.01	0.65±0.03	0.68±0.01	0.61±0.01	NS
5 day	Pregnant	0.65±0.03	0.62±0.03	0.64±0.02	0.62±0.06	0.65±0.03	0.64±0.00	NS
	Non Pregnant	0.64±0.02	0.64±0.03	0.64±0.05	0.61±0.01	0.63±0.03	0.62±0.04	NS
13 day	Pregnant	0.66±0.05	0.68±0.02	0.63±0.04	0.63±0.04	0.69±0.04	0.62±0.00	NS
	Non Pregnant	0.69±0.03	0.69±0.01	0.63±0.04	0.63±0.03	0.68±0.01	0.64±0.02	NS
21 day	Pregnant	0.65±0.06	0.65±0.03	0.64±0.01	0.68±0.01	0.62±0.03	0.62±0.00	NS
	Non Pregnant	0.60±0.02	0.65±0.05	0.62±0.03	0.66±0.04	0.63±0.03	0.67±0.01	NS

close with Pandey *et al.* (2015) who treated repeat breeder crossbred cows and recorded the plasma level of progesterone.

Significant variation between pregnant and non-pregnant animals within groups recorded except on day 5 for group IV. Significant difference in serum glucose level between the groups I to VI during experimental period for non-pregnant animals. Serum glucose level was higher in group I and V as compared to group II, III, IV and VI for non-pregnant animals. Higher serum glucose level noted in group I to V as compared to group VI for pregnant animals on day 21 (Table 2). These findings closely related with Pandey *et al.* (2015) found significantly higher in pregnant animals in PRID treated group.

No significant difference in serum calcium was observed between pregnant and non-pregnant animals in different groups on different days of estrus cycle (Table 3). The present findings are closely related with the study of Pandey *et al.* (2009), Kalita and Sarmah (2006), Kumar (2014) and Das *et al.* (2009) reported level of serum calcium in repeat breeder cow as 9.40±0.60,9.87±0.29, 9.98±0.04 and 10.045±0.327 mg/dl, respectively.

No significant difference in phosphorus between pregnant and non pregnant animal within each group (Table 4). Ceylan *et al.* (2008) who recorded mean serum phosphorus concentration in the repeat breeders 5.19± 0.22 which is closely agreed.

No significant difference in magnesium between pregnant and non-pregnant animals in all evaluated days within all groups (P 0.05) (Table 5). Kumar *et al.* (2009) reported in serum magnesium in repeat breeder as 3.65± 0.20 mg/dl.

No significant difference between groups for pregnant as well as non-pregnant animals during the experimental period except on day 5, Group I, III and VI has significantly higher total protein level than Group II, IV and V for pregnant animals (Table 6). Similar results were recorded by Modi *et al.* (2013).

No significant change in cholesterol between pregnant and non pregnant animals as well as between 6 different groups for experimental period (Table 7). It was found to be significant difference between different groups of non-pregnant animal on day 0. In group II, III and VI was found more significant serum cholesterol level than Group I, IV and VI on day 0 only. Similar level of cholesterol level were reported by Barson *et al.* (2019) and Modi *et al.* (2013).

Significant change during the treatment period in mean serum zinc concentration was observed in the present study (Table 8). The Group I had more significant ($P<0.05$) increase in zinc level than the other groups. After 13 days, the group I, II, III and IV had similar zinc level but significantly increase the group V and VI. On the 21st day, there was no significant increase of zinc level in Group I, II, III and IV but significantly differed from V and VI. In Group I, V and VI, statistically mean serum zinc concentrations were observed lower on day 5, 13 and 21 than day 0. In Group II, significantly more high concentration recorded on day 0. Barui *et al.* (2015) evaluated high significant ($p<0.01$) variations between cyclic (20.43± 0.45 and repeat breeder cow (16.89±0.34).

No significant difference in Copper between pregnant and nonpregnant animals in all evaluated days within all group for 5% level of significance (P 0.05) (Table 9). Comparatively lower level of Copper level was recorded by Ceylan *et al.* (2008) as 0.59 ± 0.04 mg/L.

No significant difference in mean serum iron between pregnant and non-pregnant animals within groups during the experimental period (Table 10). Except on day 5 for group IV and day 13 for group V where higher iron level recorded in pregnant animal as compared to non-pregnant animal. After comparing the groups for serum iron level, it was found that on day 5 to 21 group III having lowest iron level as compared to other groups in pregnant and non-pregnant animals. Khasatiya *et al.* (2005) estimated micro-minerals in fertile and infertile buffaloes at weekly intervals for first 15 weeks post-partum and reported the overall mean plasma Fe was 3.48±0.11 vs. 3.69 ± 0.11 ppm.

It was found that no significant difference between pregnant and non pregnant animals in all evaluated days within all group for 5% level of significance (P 0.05) (Table 11). Similar results were reported by Kalita and Sarmah (2006) who recorded the mean concentration of serum Mn as 0.34±0.04 ppm in repeat breeder cows. Ahmed *et al.* (2017) who recorded level as 0.18±0.05 g/ml, 0.1±0.04 g/dl in repeat breeder cows.

No significant difference in cobalt between pregnant and non-pregnant animals in all evaluated days within all group for 5% level of significance (P 0.05) (Table 12) and agrees with the finding of Jayachandran *et al.* (2013) who compared the blood biochemical and mineral profile of anestrus buffaloes with those of regular cyclic animals and recorded anestrus and regular cyclic values as cobalt (g/ml) (0.480±0.03 and 0.551±0.15, respectively).

CONCLUSION

It was concluded that in GnRH and hCG treated repeat breeder cows, progesterone level in pregnant dairy cows was nearly same animal on day 21 after AI but

conception rate was higher in hCG treated cows. Serum glucose, calcium, cholesterole and zinc recorded to be higher in pregnant than non-pregnant cows. Serum magnesium was higher in non-pregnant than pregnant animals. No variation was observed in other estimated parameters.

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