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Effect of various hormonal protocols on biochemical profile in post-partum anestrus Murrah buffaloes

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Abstract

In this study, Forty eight anestrus postpartum Murrah buffaloes (over 90 days) were randomly selected and equally divided into four groups. Animals were subjected to treatment with CIDR implant+PGF2 α (Lutalyse)+ PMSG, Crestar + PGF2 α (Lutalyse)+PMSG, Intravaginal progesterone impregnated sponge implant+PGF2 α (Lutalyse)+ PMSG and Intravaginal progesterone impregnated sponge implant with CMC+PGF2 α (Lutalyse)+ PMSG as group I, II, III and IV respectively. Biochemical profile of serum were analysed after collection of blood on day 0, 3rd, 6th and 9th of hormonal treatment. The level of glucose (61.05 \pm 2.33 to 70.53 \pm 2.40 mg/dl), total protein (7.15 \pm 0.17 to 7.90 \pm 0.28 g/dl), cholesterol (95.99 \pm 4.48 to 116.48 \pm 5.88 mg/dl), calcium (10.12 \pm 0.27 to 10.37 \pm 0.11 mg/dl) were within the normal range in all groups. However, the level of phosphorus (3.26 \pm 0.08 to 3.59 \pm 0.09 mg/dl) was lower compared to reference value (4.39-7.85mg/dl) in all groups, may be one of contributing factor for postpartum anestrus in buffaloes.

Keywords: CIDR, crestar, polyurethane sponge, biochemical profile, calcium, phosphorus, and buffalo

Introduction

Buffaloes are well known for efficient conversion of low grade fibrous food in to high value milk. Buffalo milk is preferred over cow milk by dairy product manufacturers not only for its higher total solids but also for higher fat content. Due to its superior whitening property, buffalo milk is more suitable for infant milk powder. Fertility is the main economic factor of a successful dairy industry. Post-partum anestrus is the prevalent, frustrating and challenging problem encountered in dairy animals. Field survey on reproductive disorder have revealed that anestrus is most common single cause of infertility in buffaloes among which inactive or non-functional ovaries are single cause of anestrus (EL-Wishy, 2007) [4]. Incidence of anestrus is more in buffaloes than cattle and problem is more severe in summers (Singh *et al.*, 2010) [17]. Higher Incidences of anestrus are due to inactive ovaries in buffaloes than in cows as reported by Tanwar *et al.* (2003) [19]. Increased partitioning of energy towards milk production can result in anestrus by delaying the resumption of follicular activity (Bauman and Currie, 1980) [3]. Deficiency of protein (hypoproteinemia) retards the development of sex organs and body growth in young animals and affects the subsequent reproductive performance (Herrick, 1977) [5]. Low intake of protein therefore, will put the animal in negative nitrogen balance and continued status of such nitrogen deficiency obviously will affect the phenomenon of reproduction. Cholesterol, is an essential precursor for steroidogenesis in gonads (Martin, 2012) [10] and could be a good probe to clinicians towards improving postpartum fertility in bovines. The role of calcium in steroid biosynthesis has been well documented in ovaries (Veldhnius and Klase, 1982) [20]. One of the classical manifestations of phosphorus deficiency on reproduction is alternation of estrus (Morrow, 1980) [11]. This experiment was planned to study the biochemical status of animal which may help in understanding the anestrus condition in Murrah buffalo.

Materials and Methods

Forty eight anestrus postpartum Murrah buffaloes (over 90 days) located in different villages of R.S. Pura region of Jammu district were randomly selected and equally divided into four groups. Animals were subjected to treatment with CIDR implant+PGF2 α (Lutalyse)+ PMSG, Crestar + PGF2 α (Lutalyse)+PMSG, Intravaginal progesterone impregnated sponge implant+PGF2 α (Lutalyse) + PMSG and Intravaginal progesterone impregnated sponge implant with CMC+PGF2 α (Lutalyse)+ PMSG as group I, II, III and IV respectively. Blood samples were collected from postpartum anestrus buffalo for biochemical from jugular vein.

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About 10 ml of blood were collected on Day 0, 3rd, 6th and day of removal of implants (9th day) by venipuncture of jugular vein under aseptic conditions in dry vials. The blood was centrifuged for 10-15 minutes @3000 rpm and serum was harvested and collected in sterilized vials and stored at -20 °C for biochemical analysis.

Results and Discussion

The concentration of biochemical parameters was recorded on day 0, 3, 6 and 9th of treatment under CIDR, Crestar, Intra-vaginal progesterone impregnated sponge and Intra-vaginal progesterone impregnated sponge with CMC are presented in Table 1.0. Analysis of serum glucose levels revealed that in group IV, mean concentration of serum glucose had increased significantly ($P < 0.05$) on day 6th (65.17±1.76 mg/dl) and 9th (70.53±2.40 mg/dl) as compared to day 0 (61.37±1.87 mg/dl) with non-significant increase ($P > 0.05$) on day 3rd. The mean concentration of glucose did not differ significantly with each other in group I, II and III. However, mean concentration of glucose was significantly higher ($P < 0.05$) in Group IV (70.53±2.40 mg/dl) compared to Group I (63.52±1.78 mg/dl), II (62.67±1.92 mg/dl) and III (61.93±1.92 mg/dl) on day 9th. The mean level of glucose did not differ significantly during treatment period (Hormonal implant insertion to withdrawal of implant) in all the group except Group IV, where mean level of glucose had increased significantly ($P < 0.05$) on day 6 and 9th compared to day 0 and 3rd (Table 4.4). Increased glucose level on 6 and 9th day may be indicative of bacterial growth induced by adding CMC (Swidsinski *et al.*, 2009) [18] which increases the level of cortisol to fight the infection leading to side effect of raising serum glucose level (Aronson, 2009) [2]. The same reason also holds true for increase in level of glucose on day 9 in group IV compared to other groups. In the present study, the mean serum glucose concentration in all the groups were within range as reported by Jerome *et al.*, 2016 [6]. In Group II, III and IV, the mean serum glucose concentration on day 0 to 9th varied between 61.05±2.33 to 70.53±2.40 mg/dl. Similar concentration of Glucose in post-partum anestrus buffaloes has been reported by Kalsotra *et al.*, 2016 [7]. The normal serum glucose in buffaloes ranged from 40-60 mg/dl (Neama, 2015) [13]. Analysis for serum protein levels revealed that mean concentration were increased non-significantly ($P < 0.05$) from day 0 (7.63±0.31, 7.67±0.21, 7.67±0.18 and 7.84±0.15 g/dl) today 9th (7.79±0.17, 7.87±0.25, 7.70±0.13 and 7.86±0.24 g/dl) in group I, II, III and IV, respectively. The differences, however in the mean concentration of total protein within and among the groups on sampling days were non-significant. Similar concentration of total protein in post-partum anestrus buffaloes has been reported by several workers (Kalsotra *et al.*, 2016) [7]. In contrary to above Jerome *et al.* (2016) [6] reported lower concentration of total protein in anestrus buffaloes. Significantly higher protein in normal cyler than postpartum anestrus in buffaloes has been reported by Kumar *et al.* (2010) [8]. The reference value of serum total protein of buffaloes has been reported as 5.63-8.10g/dl (Abid Ellah *et al.*, 2014) [1] and 6.00 – 8.00 g/dl (Neama, 2015) [13]. In our study, the mean concentration of total protein concentration in all groups varied between 7.15±0.17 to 7.90±0.28g/dl, which falls under the reference value. Analysis of data for serum

cholesterol levels revealed that the mean concentration were increased non- significantly from day 0 (95.99±4.48, 99.11±4.94, 98.44±6.19 and 101.26±4.28 mg/dl) to day 9th (110.46±4.41, 116.48±5.88, 105.94±5.30 and 110.15±3.88 mg/dl) in group I, II, III and IV respectively. The differences, however, in the mean concentration of serum cholesterol within and among the groups on sampling days were non-significant ($P > 0.05$). This finding are supported by findings of Nakrani *et al.* (2014) [12] and Savalia *et al.* (2014) [15]. Analysis of serum calcium level revealed that the mean concentration increased non-significantly from day 0 (10.21±0.10, 10.13±0.07, 10.12±0.27 and 10.24±0.05 mg/dl) to day 9th (10.30±0.17, 10.37±0.11, 10.27±0.28 and 10.34±0.08 mg/dl) in group I, II, III and IV, respectively. The differences, however, in the mean concentration of serum calcium within and among the groups on sampling days were non-significant ($P > 0.05$). Similar observations have been made by Nakrani *et al.* (2014) [12] using CIDR, Ovsynch and Crestar protocol, where plasma calcium level did not vary significantly in buffaloes between day/period of study of treatment. Parmar (2013) [14] and Savalia *et al.* (2014) [15] also made similar observation using CIDR and Ovsynch protocol in buffaloes. Similarly, Kumar *et al.* (2015) [9] reported non-significant increase in level of calcium on day of induced estrus in buffaloes treated with Ovsynch, Ovsynch + CIDR and Heat sync protocol. In the present study, the mean calcium concentration in all groups varied between 10.12±0.27 to 10.37±0.011 mg/dl on day 0 to 9. The level of calcium in present study are in line with Shahzad *et al.* (2016) [16] in buffaloes. The reference value of serum calcium of buffaloes has been reported as 8.11-12.46 mg/dl (Abid Ellah *et al.*, 2014) [1]. In present study, the mean concentration of calcium concentration in all groups varied between 10.13±0.31 to 10.34±0.08mg/dl, which fall under the reference value. Analysis of data for serum phosphorus level revealed that the mean concentration increased non-significantly from day 0 (3.39±0.11, 3.26±0.08, 3.27±0.08 and 3.31±0.07 mg/dl) to day 9th (3.59±0.09, 3.56±0.13, 3.51±0.09 and 3.58±0.11 mg/dl) in group I, II, III and IV, respectively. The differences, however, in the mean concentration of serum phosphorus within and among the groups on sampling days were non-significant ($P > 0.05$). Similar observations have been made by Nakrani *et al.* (2014) [12] using CIDR, Ovsynch and Crestar protocol, where plasma phosphorus level did not vary significantly in buffaloes between day/period of study of treatment. Parmar (2013) [14] and Savalia *et al.* (2014) [15] also had similar observation using CIDR and Ovsynch protocol in buffaloes. Similarly, Kumar *et al.* (2015) [9] reported non-significant increase in level of phosphorus on day of induced estrus in buffaloes treated with Ovsynch, Ovsynch + CIDR and Heat sync protocol. In the present study, the mean phosphorus concentration in all groups varied between 3.26±0.08 to 3.59±0.09 mg/dl on day 0 to 9. The reference value of serum phosphorus of buffaloes has been reported as 4.39-7.85 mg/dl (Abid Ellah *et al.*, 2014) [1]. In present study, the mean concentration of phosphorus concentration in all groups varied between 3.26±0.08 to 3.59±0.09 mg/dl, which are lower compared to reference value. This may be one of contributing factor of postpartum anestrus in buffaloes.

Table 1: Effect of various treatments on Biochemical attributes in post-partum anestrus buffaloes

Attributes	Group I				Group I				Group III				Group IV			
	Day 0	Day 3	Day 6	Day 9	Day 0	Day 3	Day 6	Day 9	Day 0	Day 3	Day 6	Day 9	Day 0	Day 3	Day 6	Day 9
Serum Glucose (mg/dl)	62.74 ± 3.06	62.83 ± 1.84	63.13 ± 1.78	63.52 ± 1.78A	62.31 ± 2.84	62.33 ± 1.73	62.52 ± 1.76	62.67 ± 1.92A	61.05 ± 2.33	61.30 ± 1.59	61.65 ± 1.57	61.93 ± 1.92A	61.37 ± 1.83a	62.38 ± 1.65a	65.17 ± 1.76b	70.53 ± 2.40Bb
Total Protein (g/dl)	7.63 ± 0.31	7.55 ± 0.16	7.90 ± 0.28	7.79 ± 0.17	7.67 ± 0.21	7.59 ± 0.18	7.61 ± 0.18	7.87 ± 0.25	7.67 ± 0.18	7.65 ± 0.16	7.64 ± 0.20	7.70 ± 0.13	7.84 ± 0.15	7.15 ± 0.17	7.58 ± 0.19	7.86 ± 0.24
Cholesterol (mg/dl)	95.99 ± 4.48	101.11 ± 4.70	106.73 ± 4.89	110.46 ± 4.41	99.11 ± 4.94	103.26 ± 4.46	103.68 ± 6.88	116.48 ± 5.88	98.44 ± 6.19	99.35 ± 6.10	99.79 ± 4.69	105.94 ± 5.30	98.44 ± 6.19	99.35 ± 6.10	99.79 ± 4.69	105.94 ± 5.30
Calcium (mg/dl)	10.21 ± 0.10	10.25 ± 0.56	10.27 ± 0.10	10.30 ± 0.17	10.13 ± 0.07	10.21 ± 0.17	10.31 ± 0.12	10.37 ± 0.11	10.12 ± 0.27	10.13 ± 0.31	10.22 ± 0.33	10.27 ± 0.28	10.24 ± 0.05	10.24 ± 0.06	10.28 ± 0.06	10.34 ± 0.08
Phosphorus (mg/dl)	3.39 ± 0.11	3.41 ± 0.12	3.47 ± 0.13	3.59 ± 0.09	3.26 ± 0.08	3.38 ± 0.11	3.43 ± 0.09	3.56 ± 0.13	3.27 ± 0.08	3.37 ± 0.13	3.42 ± 0.09	3.51 ± 0.09	3.31 ± 0.07	3.47 ± 0.10	3.50 ± 0.08	3.58 ± 0.11

^{AB}Means bearing different superscript within the row differ significantly ($P < 0.05$) between the groups

^{ab}Means bearing different superscript within the row differ significantly ($P < 0.05$) within the groups

Conclusion

In this experiment, biochemical profiles were studied and their status in response to hormonal treatment in postpartum anestrus buffaloes correlated. It is concluded that levels of serum glucose, total protein, albumin, cholesterol and calcium and phosphorus increased on the day of induced estrus in post-partum anestrus buffaloes and their concentration could be responsible for anestrus condition in field buffaloes. The level of phosphorus was lower in buffaloes of all groups, which may be one of contributing factor of postpartum anestrus in buffaloes.

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