www.ThePharmaJournal.com

The Pharma Innovation

ISSN (E): 2277-7695
ISSN (P): 2349-8242
NAAS Rating: 5.23
TPI 2022; SP-11(10): 1155-1159

www.thepharmajournal.com
Received: 16-08-2022
Accepted: 19-09-2022

Yogesh
M.V.Sc Scholar, Department of Veterinary Physiology and Biochemistry, Veterinary College, KVAFSU, Bidar, Karnataka, India

Shrikant Kulkarni
Professor and Head, Department of Veterinary Physiology and Biochemistry, Veterinary College, Bidar, Karnataka, India

Srinivas Reddy B
Associate Professor, Department of VPB, Veterinary College, Bidar, Karnataka, India

Hiremath SV
Associate Professor, Department of AGB, Veterinary College, Gadag, Karnataka, India

Ravindra BG
Associate Professor, Department of VCC, Veterinary College, Shivamogga, Karnataka, India

Kartikesh SM
Associate Professor, Department of VPB, Veterinary College, Bidar, Karnataka, India

Gangadhar Kapase
Assistant Professor, Department of VPB, Veterinary College, Bidar, Karnataka, India

Corresponding Author:
Yogesh
M.V.Sc Scholar, Department of Veterinary Physiology and Biochemistry, Veterinary College, KVAFSU, Bidar, Karnataka, India

Mineral profile of repeat breeding buffalo in relation to different phases of oestrus cycle

Yogesh, Shrikant Kulkarni, Srinivas Reddy B, Hiremath SV, Ravindra BG, Kartikesh SM and Gangadhar Kapase

Abstract
Reproductive activity is influenced by several nutrients which are involved in the synthesis of hormones that are important for reproduction. Macro-minerals like calcium, phosphorus influence the ability of animal to utilize other micro-minerals. Micro-minerals like zinc, copper are involved in the regulation of enzymatic activity at subcellular level which inturn affects reproduction. In the present study eight clinically healthy normal breeding and eight repeat breeding buffaloes presented to the Veterinary Dispensaries in and around Bidar were selected for the study. Mineral parameters viz., calcium, phosphorus, zinc, copper, iron, manganese and electrolytes like sodium, potassium and chloride were estimated during different phases viz. 0 day (day of oestrus), 11th day (mid luteal phase) and 16th day (early follicular phase) of the oestrus cycle. The mean calcium, copper and zinc concentrations were significantly lower in repeat breeders. There was no significant difference in iron, manganese, sodium, potassium and chloride levels between normal and repeat breeding buffaloes during all the phases of oestrus cycle. It may be concluded that the altered mineral profile may predispose the buffaloes to repeat breeding.

Keywords: Buffalo, repeat breeder, phases of oestrus cycle, mineral profile

Introduction
The repeat breeding syndrome is defined as a condition in which dairy animal have regular oestrus cycle and appear normal on superficial clinical examination but fails to become pregnant following three or more breeding (Bartlett *et al.*, 1986) [5]. The incidence of repeat breeding condition in buffaloes varies; variations in incidences may be attributed to the heterogeneity or multifactorial causes of the repeat-breeder syndrome as well as the effect of locality and season (Ali *et al.*, 2009) [4]. Incidence varies from as low as 0.61 percent (Pandit, 2004) [24] to as high as 55.7 percent (Singh *et al.*, 1984) [31]. Hence, exploration of possible causes and measures to improve fertility in repeat breeding buffaloes necessitates a comprehensive study on mineral determinants.

Materials and Methods
For the estimation of various mineral parameters blood samples were collected from jugular vein on day 0 (day of estrus), 11th day (mid luteal phase) and 16th day (Early follicular phase) of the oestrus cycle from normal breeding as well as repeat breeding buffaloes. Total of 10ml blood was collected and stored in vials coated with clot-activator and serum was separated after one hour and stored at -20 ^{o}C until analysis of mineral profile. Serum concentrations of electrolytes viz., sodium, potassium and chloride were estimated by automated ion selective electrode based electrolyte analyzer (LABLYTETM). Macro-minerals viz., calcium and phosphorus were analyzed by semi automated biochemistry analyzer (ARTOS®, SWEMED BIOMEDIALS PVT.LTD) using SWEMED® Diagnostic Kits. Serum concentrations of micro-minerals viz., zinc, copper, iron and manganese were estimated by atomic absorption spectrophotometer (A ANALYST 400®, Perkin Elmer). The data obtained from the present study was tabulated, and subjected to statistical analysis using one way ANOVA and Student 't' test as per the standard procedure described by (Snedecor and Cochran, 1994) [33].

Result and Discussion
Calcium (Ca)
The mean calcium concentration was significantly ($p \leq 0.05$) lower in repeat breeding group as compared to normal breeding group in all the phases of oestrus cycle (Table 1).

These findings are in agreement with the reports of (Chaurasia *et al.*, 2010, Akhtar *et al.*, 2014) [10, 3] who documented lower calcium concentration in repeat breeding buffaloes. Similar results were noticed in buffaloes by (Pandey *et al.*, 2009; Kumar, 2014) [25, 21]. Calcium plays an important role in relation to oestrus and ovulation by regulation of the membrane potential of oocytes, as calcium is involved in the disruption of cumulus cell cohesiveness by regulating the number of gap junctions between the cells (Peracchia, 1978) [27], which contributes to the process of ovulation. Optimum level of calcium is also required for sensitization of reproductive tract for actions of steroid hormones (Kumar *et al.*,1986) [22] as well as for improving number and size of ovarian preovulatory follicle (El-Shahata, 2010) [14]. Hence, the lower levels of calcium in the repeat breeding buffaloes could be one of the potential causes for repeat breeding syndrome in the present study. Contrary to the present findings, (Kalita and Sarmah, 2006; Jaychandran *et al.*, 2007 and Barui *et al.*, 2015) [18, 17, 06] have reported no significant difference in the mean calcium concentration of normal and repeat breeding bovines. The comparison within group revealed no significant ($p \leq 0.05$) difference in calcium concentration among oestrus, mid luteal and early follicular phase of oestrus cycle in both normal and repeat breeding groups. These results are in conformation with the observations made by (Sharma *et al.*, 1999) [30]. While, (Burle *et al.*, 1995) [07] reported significantly different calcium values between different stages of oestrus cycle, being highest on the day of estrus.

Phosphorus (P)

The phosphorus concentration (mg/dl) in normal and repeat breeding group on 0, 11th and 16th day were 5.13±0.09, 5.27±0.23 and 5.25±0.21; 4.55±0.25, 4.67±0.20 and 4.86±0.25 respectively (Table 1). Though the statistical analysis of the data revealed that there was no significant ($p \leq 0.05$) difference in the phosphorous level but repeat breeding buffaloes had apparently lower phosphorous concentration than the normal breeders in all the phases of oestrus cycle. (Chaurasia *et al.*, 2010 Akhtar *et al.*, 2014) [10, 03] have also reported lower phosphorus levels in repeat breeding buffaloes. Similarly in cows, low phosphorus values in repeat breeding cows is also reported by (Das *et al.*, 2002; Chandrahar *et al.*, 2003; Jaychandran *et al.*, 2007; Pandey *et al.*, 2009; Das *et al.*,2012; Kumar, 2014) [11, 09, 17, 25, 12, 21]. Phosphorus is essential for energy transformation at cellular level, particularly through ATP and hence, the deficiency of phosphorus may arrest the phenomenon of fertilization and cause early embryonic death and repeat breeding (Kumar *et al.*, 2011) [20]. The results of this study indicate that the significantly low calcium and marginally low phosphorus levels might be related to the ovulatory disturbances in buffalo as disturbed calcium-phosphorus ratio has a blocking action on the pituitary gland and consequently on the ovarian function (Herrick and John 1977) [16] resulting in disturbances of the pituitary-ovarian axis leading to repeat breeding. In the present study, no significant ($p \leq 0.05$) difference was observed in phosphorus levels on day of oestrus, mid luteal and early follicular phase of oestrus cycle in both normal and repeat breeding. Whereas, (Sharma *et al.*, 1999) [30] has reported significantly lower inorganic phosphorus concentration during proestrus phase compared to other phases of the oestrus cycle in repeat breeder and normal cycling buffaloes.

Copper (Cu)

Mean copper concentration was significantly ($p \leq 0.05$) lower in repeat breeding buffaloes as compared to normal breeding buffaloes in all the phases of oestrus cycle (Table 2). In agreement with the present study (Singh *et al.*, 2006 and Ahmed *et al.*, 2010) [32, 02] have documented significantly lower copper levels in repeat breeding buffaloes. Similarly (Modi *et al.*, 2013) [23] have observed lower copper values in repeat breeding cows. In contrast, (Barui *et al.*, 2015) [06] have reported no significant difference in the mean copper concentration between normal and repeat breeding buffaloes and cows respectively. Copper has a significant role in maintaining optimum fertility as it is involved in FSH, LH, estrogen activity (Desai *et al.*, 1982) [13] and its levels are positively correlated to progesterone concentration. Deficiencies of phosphorus and zinc are linked to low levels of progesterone, which could be the cause of failures in fertilization or early embryonic death. (Prasad *et al.*, 1989) [28] opined that Cu and Zn either singly or in combination play a major role in maintaining reproductive rhythm. Copper and zinc play an important role in regulating progesterone production by luteal cells via involvement of superoxide dismutase (Sales *et al.*, 2011) [29]. Positive correlation was reported between serum progesterone level and copper-zinc in cows by (Yildiz and Akar 2001) [34]. Hence, significantly lower levels of copper as in the present study can be attributed to repeat breeding syndrome as asynchrony of sex hormones is evident in repeat breeders.

When the mean copper values were compared within the group, there was no significant ($p \leq 0.05$) difference among different phases of oestrus cycle in both normal and repeat breeding groups. These findings are in agreement with the reports of (Kapadiya and Siddiquee 2012) [19] and disagreement with (Sharma *et al.*, 1999) [30] who reported significant variation during different phases of oestrus cycle.

Zinc (Zn)

The mean zinc concentration was significantly ($p \leq 0.05$) lower in repeat breeding group as compared to normal breeding group (Table 2). Similar result was reported by (Akhtar *et al.*, 2014 and Barui *et al.*, 2015) [03, 06]. However, (Butani *et al.*, 2011) [08] reported no significant difference between normal and repeat breeder groups. Zinc act as cofactor in number of enzyme system that assists in maintenance of the activity of labile hypophyseal hormones in circulation and it is also involved in the progesterone production by luteal cell (Sales *et al.*, 2011) [29]. Hence, significantly lower zinc as observed in the present study might cause reduction in GnRH secretion by hypothalamus and eventually leading to decreased levels of GnRH dependent hormones downstream resulting into repeat breeding syndrome. Zinc also plays an important role in maintenance of uterine epithelium necessary for embryo implantation.

Iron (Fe)

In the present study no significant ($p \leq 0.05$) difference in iron levels was observed between repeat and normal breeding groups (Table 3). Similar results were reported by (Abhilash *et al.*, 2008 and Butani *et al.*, 2011) [01, 08]. Contrary to the present results, (Akhtar *et al.*, 2014) [03] have reported significantly lower iron levels in repeat breeding animals. When the mean iron values were compared within the group, there was no significant ($p \leq 0.05$) difference among different phases of oestrus cycle in both normal and repeat breeding

groups. These findings are in agreement with the reports of (Kapadiya and Siddiquee, 2012) [19] and disagreement with (Parmar *et al.*, 1986) [26] who reported significant variation during different phases of oestrus cycle.

Manganese (Mn)

The comparison between two groups revealed that there was no significant ($p \leq 0.05$) difference in manganese levels between the repeat breeder and normal breeder groups (Table 3). Results are in accordance with the similar reports in buffaloes by (Butani *et al.*, 2011) [08]. In contrary (Abhilash *et al.*, 2008 and Modi *et al.*, 2013) [01, 23] have reported significantly higher manganese levels in normal breeding cows as compared to repeat breeding cows.

Sodium (Na)

In the present study there was no significant ($p \leq 0.05$) difference in sodium levels between repeat and normal breeding groups (Table 4). Similar results were reported by (Chaurasia *et al.*, 2010 and Akhtar *et al.*, 2014) [10, 3]. However, contrary to the present results, (Elthomy *et al.*, 1989) [15] have reported lower sodium levels in repeat breeder buffaloes and (Pandey *et al.*, 2009) [25] have reported higher sodium levels in repeat breeder cows. No significant ($p \leq 0.05$) difference was observed in sodium levels among different phases of oestrus cycle within normal and repeat breeding groups of the present study.

Potassium (K)

In the present study there was no significant ($p \leq 0.05$) difference in potassium levels between repeat and normal breeding groups (Table 4). Similar results were reported by (Chaurasia *et al.*, 2010) [10]. In contrast, (Pandey *et al.*, 2009) [25] have reported higher levels of potassium in repeat breeding cows. There was no significant ($p \leq 0.05$) difference observed in potassium levels among different phases of oestrus cycle within normal and repeat breeding groups of the present study.

Chloride (Cl)

In the present study there was no significant ($p \leq 0.05$) difference in chloride levels between repeat and normal breeding groups (Table 4). Similar results were reported by (Sharma *et al.*, 1999) [30] in cows and buffaloes respectively. In contrast, (Elthomy *et al.*, 1989) [15] have reported lower chloride levels in repeat breeding buffaloes. No significant ($p \leq 0.05$) difference was observed in chloride levels among different phases of oestrus cycle within normal and repeat breeding groups of the present study.

Table 1: Serum concentrations (Mean± SE) of calcium and phosphorus at different phases of oestrus cycle in normal and repeat breeding buffaloes

Day of oestrus cycle	Calcium (mg %)		Phosphorus (mg %)	
	Normal breeder	Repeat breeder	Normal breeder	Repeat breeder
0 day	9.24±0.33	8.35±0.22*	5.13±0.09	4.55±0.25
11th day	9.40±0.22	8.57±0.27*	5.27±0.23	4.67±0.20
16th day	9.56±0.25	8.42±0.19*	5.25±0.21	4.86±0.25

* Significant difference ($p \leq 0.05$) between groups for each parameter on respective day of oestrus cycle.

Table 2: Serum concentrations (Mean± SE) of copper and zinc at different phases of oestrus cycle in normal and repeat breeding buffaloes

Day of Oestrus cycle	Copper (µg/dl)		Zinc (µg/dl)	
	Normal breeder	Repeat breeder	Normal breeder	Repeat breeder
0 day	75.54±0.23	68.91±0.24*	140.30±0.60	137.10±1.20*
11th day	75.70±0.25	68.67±0.18*	139.00±0.49	135.60±1.40*
16th day	76.60±0.41	68.77±0.24*	140.00±0.43	135.70±0.87*

*Significant difference ($p \leq 0.05$) between groups for each parameter on respective day of oestrus cycle.

Table 3: Serum concentrations (Mean± SE) of iron and manganese at different phases of oestrus cycle in normal and repeat breeding buffaloes

Day of oestrus cycle	Iron (µg/dl)		Manganese (µg/dl)	
	Normal breeder	Repeat breeder	Normal breeder	Repeat breeder
0 day	146.80±3.02	145.80±2.14	51.75±1.96	49.38±1.30
11th day	146.90±0.37	145.30±1.88	51.25±1.41	49.63±1.05
16th day	146.10±0.30	145.20±2.27	51.13±1.45	49.50±1.03

Table 4: Serum concentrations (Mean± SE) of sodium, potassium and chloride at different phases of oestrus cycle in normal and repeat breeding buffaloes

Day of oestrus cycle	Sodium (mmol/L)		Potassium (mmol/L)		Chloride (mmol/L)	
	Normal breeder	Repeat breeder	Normal breeder	Repeat breeder	Normal breeder	Repeat breeder
0 day	138.20±1.70	138.30±1.49	4.76±0.19	4.73±0.07	101.8±0.70	102.50±1.54
11th day	137.00±1.01	137.20±1.49	4.75±0.19	4.70±0.15	104.10±2.93	102.50±1.35
16th day	138.80±2.63	138.20±2.37	4.79±0.09	4.69±0.43	102.30±1.96	102.00±0.67

Conclusion

From the present study it may be concluded that the mean calcium, copper and zinc concentrations were significantly lower in repeat breeders. There was no significant difference in iron, manganese, sodium, potassium and chloride levels between normal and repeat breeding buffaloes during all the phases of oestrus cycle. As found in this study altered mineral profile viz. significantly lower mean calcium, copper and zinc concentrations might have predisposed the buffaloes to repeat breeding.

Reference

1. Abhilash RS, Rao AS, Krishna KM, Naidu KV. Trace mineral profile and incidence of repeat breeding in crossbred cows. Indian Veterinary Journal. 2008;85(11):1166.
2. Ahmed WM, Zaabal MM, Abdel Hameed AR. Relationship between ovarian activity and blood lead concentration in cows and buffaloes with emphases on gene frequencies of hemoglobin. Global Journal of Biotechnology & Biochemistry. 2010;5(1):1-5.
3. Akhtar MS, Asim Farooq A, Akbar Lodhi L, Muhammad S, Mazhar Ayaz M, Lashari MH, *et al*. Studies on serum macro and micro minerals status in repeat breeder and normal cyclic Nili-Ravi buffaloes and their treatment strategies. African Journal of Biotechnology. 2014;13(10):1143-1146.
4. Ali A, Abdel-Razek AK, Derar R, Abdel-Rheem HA, Shehata SH. Forms of reproductive disorders in cattle and buffaloes in middle Egypt. Reproduction in domestic animals. 2009;44(4):580-586.
5. Bartlett PC, Kirk JH, Mather EC. Repeated insemination in Michigan Holstein-Friesian cattle: Incidence, descriptive epidemiology and estimated economic impact. Theriogenology. 1986;26(3):309-322.
6. Barui A, Batabyal S, Ghosh S, Saha D, Chattopadhyay S. Plasma mineral profiles and hormonal activities of normal cycling and repeat breeding crossbred cows: A comparative study. Veterinary world. 2015;8(1):42.
7. Burle PM, Mangle NS, Kothekar MD, Kalorey DR. Blood biochemical profiles during various reproductive states of Sahiwal and Jersey x Sahiwal cattle. Livestock Adviser. 1995;20:13-20.
8. Butani MG, Dhami AJ, Kumar R. Comparative blood profile of progesterone, metabolites and minerals in anoestrus, suboestrus, repeat breeding and normal cyclic buffaloes. Indian Journal of Field Veterinarians (The). 2011;7(2):20-24.
9. Chandrahar D, Tiwari RP, Awasthi MK, Dutta GK. Serum biochemical profile of repeat breeder cows. Indian J Anim. Reprod. 2003;24:125-127.
10. Chaurasia R, Kushwaha HS, Chaurasia D, Gendley MK, Santra AK. Comparative studies of certain macro minerals during various reproductive states in buffaloes. Buffalo Bull. 2010;29(4):291-298.
11. Das S, Bandopadhya SK, Basu S, Ghosh BB, Dattagupta R. Blood mineral profile of normal cyclic and repeat breeder crossbred cows under rural condition. Indian J Anim. Reprod. 2002;23(2):167-169.
12. Das S, Mishra SK, Swain RK, Mohanty DN, Mishra SR. Comparative study of certain serum biochemical parameters in anoestrus and repeat breeding cows of Bhadrak district of Orissa. Indian Journal of Field Veterinarians (The). 2012;7(4):62-65.
13. Desai MC, Thakkar TP, Dharshana RA, Janakiraman K. Note on serum copper and iron in Surti buffalo in relation to reproduction and gonadotropins. Indian journal of animal sciences; c1982.
14. El-Shahat KH. The effect of dietary supplementation with calcium salts of long chain fatty acids and/or l-carnitine on ovarian activity of Rahmani ewes. Animal reproduction science. 2010;117(1-2):78-82.
15. Eltohamy MM, Younis M, Salem HA, Azouz A, Shawky H, Farahat AA. Role of some micro-elements and macro-elements in inducing repeat breeding in buffalos. Indian journal of Animal Sciences. 1989;59(11):1406-1409.
16. Herrick JB. External factors affecting reproduction in dairy cattle. In1st All India Symposium on Anim. Reprod; c1977. p. 17-19.
17. Jayachandran S, Selvaraj P, Visha P. Blood biochemical profile in repeat breeding buffaloes. Tamil Nadu Journal of Veterinary and Animal Science. 2007;3(2):70-73.
18. Kalita DJ, Sarmah BC. Mineral profile and serum enzyme activities of normal cycling and repeat breeding cows. Indian J Anim. Res. 2006;40(1):49-51.
19. Kapadiya FM, Siddiquee GM. Blood plasma trace elements in repeat breeding Mehsana buffaloes. Indian Journal of Animal Reproduction. 2012;33(1):21-24.
20. Kumar S, Pandey AK, AbdulRazzaque WA, Dwivedi DK. Importance of micro minerals in reproductive performance of livestock. Veterinary world. 2011;4(5):230.
21. Kumar AS. Blood biochemical profile in repeat breeding crossbred dairy cows. International Journal of Veterinary Science. 2014;3(4):172-173.
22. Kumar S, Shrma MC, Dwivedi SK. 5th National Congress on Animal Reproduction – proceedings, Guwahati; 1986, 23-30
23. Modi LC, Suthar BN, Chaudhari CF, Chaudhari NF, Nakhashi HC, Modi F. Trace minerals profile of blood serum and estrual mucus in repeat breeder Kankrej cows. Veterinary World. 2013;6(3):143.
24. Pandit RK. Incidence of different kinds of reproductive disorders in livestock. Indian J Anim. Reprod; 2004;25:35-36.
25. Pandey V, Singh AK, Sharma N. Blood biochemical profile in fertile and repeat breeding crossbred cows under field conditions. Veterinary Practitioner. 2009;10(1):45-48.
26. Parmar KS, Mehta VM, Patel JM. Biochemical profile of repeat breeding crossbred cattle in relation to different phase of oestrous cycle. Indian. J Anim. Reprod. 1986;7(2):31-35.
27. Peracchia C. Calcium effects on gap junction structure and cell coupling. Nature. 1978;271(5646):669-671.
28. Prasad CS, Sharma PV, Obireddy A, Chinnaiya GP. Trace elements and ovarian hormonal levels during different reproductive conditions in crossbred cattle. Indian J Dairy Sci. 1989;42(3):489-492.
29. Sales JN, Pereira RV, Bicalho RC, Baruselli PS. Effect of injectable copper, selenium, zinc and manganese on the pregnancy rate of crossbred heifers (*Bos indicus*× *Bos taurus*) synchronized for timed embryo transfer. Livestock Science. 2011;142(1-3):59-62.
30. Sharma KB, Nayyar S, Malik VS, Singh R, Sodhi SP. Levels of hormones and minerals in cyclic, anestrus and subestrus buffalo heifers. Indian journal of animal sciences. 1999;69(4):214-216.
31. Singh CS, Sharma DB, Verma SK, Chauhan HV. Comparative studies on Friesian X Sahiwal crossbred cows and buffaloes. I. Reproductive disorders birth weight and early growth. Indian journal of dairy science. 1984;37(2):145-149.
32. Singh B, Rawal CV, Singh JP. Serum zinc, copper and cobalt level in normal cyclic, anestrous and repeat breeder buffaloes. Indian Journal of Animal Reproduction. 2006;27:34-36.
33. Snedecor GW, Cochran WG. Statistical methods. Edn 8,

Oxford and IBH Publications company, Calcutta, 1994.
34. Yildiz H, Akar Y. Relationships between serum progesterone and some mineral levels during the oestrous cycles in cows. Saglik-Bilimleri-Dergisi-Firat universities. 2001;15(77):84.