

METABOLIC AND HORMONAL CHANGES IN WATER BUFFALOES DURING POST PARTURIENT PEAK LACTATION

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ABSTRACT

Present study was undertaken to appraise dynamic changes in metabolic and hormonal profile in water buffaloes during post parturient peak lactation. Total 30 early lactating buffaloes were studied and compared at two points, first at 15 to 20 days and second at 40 to 45 days post calving. The parameters investigated were milk yield, milk composition, blood glucose, β -hydroxy butyrate (BHBA), glucagon, Triiodothyronine (T_3) and Thyroxin (T_4). Farm history, milk samples and blood samples were collected at two time points from all the buffaloes for analysis. Buffaloes during post parturient peak lactation showed significant ($P<0.05$) changes in milk yield and milk fat and non-significant ($P<0.05$) changes in milk SNF percentage, glucose, BHBA, glucagon T_3 , and T_4 , respectively. In the conclusion, study recorded deviation in metabolic and hormonal profile during early peak lactation.

Keywords: *Bubalus bubalis*, buffaloes, post parturient period, peak lactation, glucose, β -hydroxy butyrate (BHBA), glucagon and thyroid hormones

INTRODUCTION

Post-parturient period is challenging in dairy animals as they have to cope altered physiological, metabolic, endocrine, environmental and management changes (Sepulveda *et al.*, 2013). These animals are at higher risk for many disease and disorders during early lactation (Drackley *et al.*, 1999) as lactation burst feed intake is low due to transforming microbial flora in rumen. To maintain peak lactation during post parturient period, excess mobilization of fats, ketone associated energy breakdown, succeeding production of ketone bodies *viz.* acetone, acetoacetate and β -hydroxybutyrate (BHBA) are well documented in dairy animals (Radostits *et al.*, 2010). The role of thyroid hormone in milk synthesis is not yet understood, however, Huszenicza *et al.* (2002) suggested that thyroid hormones maintain the homeostasis of energy and protein metabolism, thermoregulation and productivity parameter during lactation. Glucagon (catabolic hormone) works to raise the concentration of glucose and fatty acids in the bloodstream (Voet *et al.*, 2011). Studies on investigation of deviation in complete blood count and biochemical profile during early lactation were investigated by many authors in buffaloes in India (Hagawane *et al.*, 2009;

Reddy *et al.*, 2014 and Reddy and Sivajothi, 2020). However, as far as Indian water buffaloes are concern, information about metabolic and hormonal profile during early peak lactation is not available. Therefore, present study was undertaken to evaluate dynamic changes in metabolic and hormonal profile in water buffaloes during post parturient peak lactation.

MATERIALS AND METHODS

Ethical statutory approval

Present study was initiated after permission from Institutional Ethics Committee for Veterinary Clinical Research (Project Approval No: IAEC-VCR/Subcommittee/08/2018, Dated: 12.12.2018) and Institutional Bio-safety Committee (Project meeting dated 25.04.2019, project No. 3).

Selection of animals

Present study was undertaken in 6 different buffalo farm in and around Mumbai city (Maharashtra-India). Total 30 early lactating buffaloes selected and compared at two points first at 15 to 20 days and second at 40 to 45 days of post calving. The observations pertaining to parity of animal, feed intake and feed composition were recorded.

Sampling and laboratory investigations

Animals were hand milked. Milk yield was recorded after complete milking, after through mixing of the milk, 10 ml milk sample was collected in 50 ml vial and subjected for milk analyser (Lactoscan SL30, Vers:42 00-11-14,Ser.N:9417) to estimate % fat and SNF. After proper restraining of buffaloes, blood samples were collected from

jugular venipuncture using 18 G needle and 10 ml syringe. Total 7 ml blood was collected from each animal then 2 ml of total blood transferred Sodium Fluoride vial for glucose estimation and remaining 5 ml blood transferred to serum vacationer vial to harvest serum samples. The aliquots of serum sample were stored at -20°C until estimations.

Random blood sugar was estimated by using GOD-POD (Glucose oxidase and peroxidase) method. Estimation of BHBA and Glucagon were undertaken by using “Bovine BHBA ELISA Kit” and ‘Bovine glucagon ELISA Kit’ (commercial ELISA kits manufactured by Biospes, Jiulongpo District, Chongqing, 400039, China and supplied by infobio, New Delhi-India).

Estimation of thyroid hormones using Radio immune Assay kits was carried out at Department of Veterinary Nuclear Medicine, including Radioisotope laboratory (Type II-Research Purpose), Mumbai Veterinary College, Mumbai. Concentration of thyroid hormones T_3 and T_4 in serum was determined by using ready to use RIA kits supplied by Board of Radiation and Isotope Technology (BRIT) Mumbai. The assay procedure for estimation of T_3 and T_4 was followed as per methods described by manufacturer. Results of hormonal concentration were extracted from standard graph after satisfactory quality control parameters viz. observed value of controls supplied with kits and recovery percentage outlined by Galdhar and Gaikwad (2017).

Statistical analysis

Mean and standard error of collected data was calculated and analysed for comparison as per standard methods outlined by Snedecor and Cochran (2009).

RESULTS AND DISCUSSION

The results of deviation in milk yield, milk composition (% fat and SNF), blood glucose, BHBA, glucagon T₃ and T₄ at two points, 15 to 20 days and at 40 to 45 days post calving are presented in Table 1.

Average per day milk yield was 8.49±0.11 kg and 9.23±0.19 kg, average milk % fat was 5.88±0.15 and 6.86±0.18 and average milk % SNF was 8.28±0.13 and 8.68±0.16 at 15 to 20 days and 40 to 45 days of post calving, respectively.

A significant (P<0.05) increase in magnitude of daily milk yield and milk % fat and non-significant (P<0.05) decrease in magnitude of SNF % was recorded from 15 to 20 days to 40 to 45 days of post calving. The results obtained in the present study of increased milk yield and % fat from 15 to 20 days to 40 to 45 days of post calving are in accordance with earlier reports of Djokovic *et al.* (2010); Enrico *et al.* (2018). Enrico *et al.* (2018) studied metabolic and hormonal adaptation in buffaloes around calving and early lactation. They reported that an increased trend in milk yield was recorded throughout postpartum period with the highest values at 50 days post-partum. They also reported significant increase in % fat from 30th day to 50th day.

Average estimated blood glucose was 66.43±1.19 mg/dl and 64.16±1.25 mg/dl, at 15 to 20 days and 40 to 45 days of calving respectively. This suggests non-significant (P<0.05) decrease in values of glucose from 15 to 20 days and 40 to 45 days of calving. These findings are in agreement with findings of Greenfield *et al.*, (2000); Enrico *et al.* (2018). They elucidated that the values of glucose remained stable throughout the post-partum period. In the healthy dairy animals, glucose is not reduced before calving but it reduced only after

calving because at the stage of pregnancy glucose need is very low and after first week of postpartum high demand of glucose for lactose synthesis. Block *et al.* (2001) also estimated that glucose level were 50 mg/dl, 47 mg/dl and 55 mg/dl at 1 week, 3 week and 8 week after parturition and concluded non-significant difference.

The Mean concentration of BHBA in present study were 515.50±43.31 µmol/l and 543.16±39.64 µmol/l at 15 to 20 days and 40 to 45 days of calving respectively. This suggest a gradual increase in value of BHBA from 15 to 20 days and 40 to 45 days respectively, but the deviation was non-significant (P<0.05). There was no overproduction of BHBA recorded and the concentration of BHBA (µmol/l) obtained from each animal throughout the study period were within the physiological range established for buffalo indicating an appropriate fat metabolism. The BHBA results in present study suggest that early lactating buffaloes selected in the study did not prone to ketosis. Youssef *et al.* (2010) estimated that average concentration of BHBA in normal buffaloes after parturition was 8.6±2.2 mg/dl or 826.04±211.31 µmol/l. Xia *et al.* (2012) also noticed concentration of Plasma BHBA as 0.60±0.12 mmol/l during lactation in normal buffaloes.

The mean serum concentration of glucagon was 78.87±8.27 ng/l and 68.07±6.14 ng/l at 15 to 20 days and 40 to 45 days of calving, respectively. The present study demonstrated non-significant (P<0.05) deviation. Brockman (1978) observed that the plasma concentration of insulin and glucagon increases significantly after feeding, and peak level at four hours after a meal. He also suggested that plasma insulin and glucagon concentrations correlate poorly with changes in blood glucose. Sartin *et al.* (1985) observed that basal glucagon level at day 5 postpartum was 156.9±24.7 pg/ml

Table 1. Deviation in milk yield, milk composition, Glucose, BHBA, Thyroid profile and Glucagon during early peak lactation.

| Sr. No. | Parameters | Animals after calving (n=30) | Mean \pm SE | t cal |
|---------|-------------------------|------------------------------|--------------------|-------|
| 1 | Milk Yield (Kg) | 15 to 20 Days | 8.49 \pm 0.11 | 4.24* |
| | | 40 to 45 Days | 9.23 \pm 0.19 | |
| 2 | Fat (%) | 15 to 20 Days | 5.88 \pm 0.15 | 4.15* |
| | | 40 to 45 Days | 6.86 \pm 0.18 | |
| 3 | SNF (%) | 15 to 20 Days | 8.28 \pm 0.13 | 1.94 |
| | | 40 to 45 Days | 8.68 \pm 0.16 | |
| 4 | Glucose (mg/dl) | 15 to 20 Days | 66.43 \pm 1.19 | 1.31 |
| | | 40 to 45 Days | 64.16 \pm 1.25 | |
| 5 | BHBA (μ mol/l) | 15 to 20 Days | 515.50 \pm 43.31 | 0.47 |
| | | 40 to 45 Days | 543.16 \pm 39.64 | |
| 6 | Glucagon (ng/ml) | 15 to 20 Days | 78.87 \pm 8.27 | 1.05 |
| | | 40 to 45 Days | 68.07 \pm 6.14 | |
| 7 | T ₃ (nmol/l) | 15 to 20 Days | 1.13 \pm 0.10 | 1.58 |
| | | 40 to 45 Days | 0.87 \pm 0.08 | |
| 8 | T ₄ (nmol/l) | 15 to 20 Days | 33.68 \pm 0.99 | 0.82 |
| | | 40 to 45 Days | 35.37 \pm 1.77 | |

*P<0.05 (significant at 5% level).

and at 30 day postpartum 193.3 \pm 17.8 pg/ml. Xia *et al.* (2012) reported the values of glucagon as 180 \pm 39 (pg/ml) in ketotic and 180 \pm 45 pg/ml in non-ketotic cow, with non-significant difference in blood glucagon between ketotic and non ketotic cows. The present research work also showed non-significant difference between glucagon levels in buffaloes during early lactation.

Mean concentration of T₃ was 1.13 \pm 0.10 nmol/l and 0.87 \pm 0.08 nmol/l at 15 to 20 days and 40 to 45 days of calving respectively. The observed value of T₃ in present research work was decreased significantly (P<0.05). Enrico *et al.* (2018), observed similar pattern. They noticed that after calving in buffalo the T₃ level increases up to approx. 20 days and after that level of T₃ decreases. The decreasing

pattern of T₃ during post calving period may be due to reduced thyroidal hormone secretion rate, possibly, due to increase of the number of receptors for this hormone in the mammary gland (Wilson and Gorewit, 1980).

The mean concentration of T₄ observed in present study was 33.68 \pm 0.99 nmol/l and 35.37 \pm 1.77 nmol/l at 15 to 20 days and 40 to 45 days of calving, respectively. This suggests non-significant (P<0.05) increase in serum concentration of Thyroxin after calving. Enrico *et al.* (2018) found that T₄ value was gradually decreases throughout the post-partum period. Kunj (1985) concluded that blood Thyroxin (T₄) level slowly increases up to 20 days before parturition, and then rapidly decreases towards calving i.e., T₄ concentration

was higher before than after parturition. The observed concentration of T_4 , showing that normal thyroidal secretion rate and may be less secretion of T_4 through milk (Akasha and Anderson, 1984).

Thus, present study recorded deviation in metabolic and hormonal profile in water buffaloes during post-parturient peak lactation.

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REFERENCES

- Akasha, M. and R.R. Anderson. 1984. Thyroxine and triiodothyronine in milk of cows, goats, sheep, and guinea pigs. *In Proceedings of the Society for Experimental Biology and Medicine*, **177**(2): 360-371. DOI: 10.3181/00379727-177-41957
- Block, S.S., W.R. Butler, R.A. Ehrhardt, A.W. Bell, M.E.V. Amburgh and Y.R. Boisclair. 2001. Decreased concentration of plasma leptin in periparturient dairy cows is caused by negative energy balance. *J. Endocrinol.*, **171**(2): 339-348. DOI: 10.1677/joe.0.1710339
- Brockman, R.P. and M.R. Johnson. 1978. Evidence of a role for glucagon in regulating glucose and B-hydroxybutyrate metabolism in sheep. *Can. J. Anim. Sci.*, **57**(1): 177-180. 10.4141/cjas77-021
- Djokovic, R., H. Samanc, J. Bojkovski and N. Fratric. 2010. Blood concentrations of thyroid hormones and lipids of dairy cows in transitional period. *Lucrări Științifice Medicină Veterinară*, **43**(2): 34-40. Available on: https://www.usab-tm.ro/vol10bMV/5_vol10b.pdf
- Drackley, J.K. 1999. Biology of dairy cows during the transition period: The final frontier. *J. Dairy Sci.*, **82**(11): 2259-2273. DOI: 10.3168/jds.S0022-0302(99)75474-3
- Fiore, E., F. Arfuso, M. Gianesella, D. Vecchio, M. Morgante, E. Mazzotta, T. Badon, P. Rossi, S. Bedin and G. Piccione. 2018. Metabolic and hormonal adaptation in *Bubalus bubalis* around calving and early lactation. *PLoS ONE*, **13**(4): e0193803. DOI: 10.1371/journal.pone.0193803.
- Galdhar, C.N. and R.V. Gaikwad. 2017. Presented lead paper on “Radio Immune Assay (RIA): Concept and application in veterinary clinical practice and research, p. 321-324. *In 35th Annual Convention of ISVM and National Symposium on Innovative Techniques, Emerging Issues and Advancement in Veterinary Medicine to Meet the Challenges*, Veterinary College and Research Institute, Tirunelveli, India.
- Greenfield, R.B., M.J. Cecava and S.S. Donkin. 2000. Changes in mRNA expression for gluconeogenic enzymes in liver of dairy cattle during the transition to lactation. *J. Dairy. Sci.*, **83**(6): 1228-1236. DOI: 10.3168/jds.S0022-0302(00)74989-7
- Hagawane, S.D., S.B. Shinde and D.N. Rajguru. 2009. Haematological and blood biochemical profile in lactating buffaloes in and around Parbhani city. *Vet. World*, **2**(12): 467-469. Available on: <http://citeseerx.ist.psu.edu/viewdoc/download?doi=10.1.1.302.4558&rep=rep1&type=pdf>

- Huszenicza, G.Y., M. Kulcsar and P. Rudas. 2002. Clinical endocrinology of thyroid gland function in ruminants. *Vet. Med. Czech.*, **47**(7): 199-210. DOI: 10.17221/5824-VETMED
- Kunj, P.L., J.W. Blum, I.C. Hart, H. Bicket and J. Landis. 2010. Effects of different energy intakes before and after calving on food intake, performance and blood hormones and metabolites in dairy cows. *Anim. Prod.*, **40**(2): 219-231. DOI: 10.1017/S0003356100025320
- Radostits, O.M., C.C. Gay, K.W. Hinchcliff and P.D. Constable. 2010. *Veterinary Medicine-A Textbook of the Diseases of Cattle, Horses, Sheep, Pigs and Goats*, Elsevier Saunders, USA. **10**: 1664-1671.
- Reddy, B.S. and S. Sivajothi. 2020. Assessemnet of haemato - Biochemical and stress parameters during the transition period in buffaloes. *Buffalo Bull.*, **39**(2): 161-165. Available on: <https://kuojs.lib.ku.ac.th/index.php/BufBu/article/view/2227>
- Reddy, B.S.S., K.N. Kumari, B.S. Reddy and Y.R. Reddy. 2014. Therapeutic management of ketosis associated with sub-clinical mastitis in transition cows. *Intas Polivet*, **15**(2): 504-506.
- Sartin, J.L., K.A. Cummins, R.J. Kemppainen, D.N. Marple, C.H. Rahe and J.C. Williams. 1985. Glucagon, insulin, and growth hormone responses to glucose infusion in lactating dairy cows. *Journal American Physiological Society*, **248**(1): E108-E114. DOI: 10.1152/ajpendo.1985.248.1.E108
- Sepulveda, P.V., J.M. Huzzey, D.M. Weary and M.A.G.V. Keyserling. 2013. Behaviour, illness and management during the periparturient period in dairy cows. *Anim. Pro. Sci.*, **53**(9): 988-999. DOI: 10.1071/AN12286
- Snedecor, G.W. and W.G. Cochran. 2009. *The Principles and Practices of Statistics in Biological Research*, 8th ed. State University Press, Iowa, USA. Voet, D. and J.G. Voet. 2011. *A Text Book of Biochemistry*, 4th ed. Willey, New York, USA.
- Wilson, D.B. and R.C. Gorewit. 1980. Specific thyroxine receptors in mammary cytosol from lactating cattle. *Biochemi. Bioph. Res. Co.*, **95**(2): 807-815. DOI: 10.1016/0006-291x(80)90859-1
- Xia, C., Z. Wang, C. Xu and H.Y. Zhang. 2012. Concentrations of plasma metabolites, hormones, and mRNA abundance of adipose leptin and hormone-sensitive Lipase in Ketotic and Nonketotic dairy cows. *J. Vet. Intern. Med.*, **26**(2): 415-417. DOI: 10.1111/j.1939-1676.2011.00863.x
- Youssef, M.A., M.R. El-Ashker and M.S. Younis. 2010. The effect of subclinical ketosis on indices of insulin sensitivity and selected metabolic variables in transition dairy cattle. *Comp. Clin. Pathol.*, **26**(2): 329-334. DOI: 10.1007/s00580-016-2377-z