



## Metabolic profile of Purnathadi buffaloes during transition period

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Buffaloes, also known as “the black gold of India,” are favoured by Indian farmers for their high milk yield, draught power, transportation, and disease resistance. The Purnathadi buffalo, the 20<sup>th</sup> registered breed in India, is notable for its good reproductive efficiency, low maintenance cost, high milk fat percentage, and adaptability to harsh climates (Ali *et al.* 2019). The transition period, from three weeks before to three weeks after calving, is the most stressful for dairy animals due to physiological and metabolic changes. This period sees a decline in nutrient intake and an increased energy demand, leading to negative energy balance and mobilization of body reserves. Non-esterified fatty acids (NEFAs) increase due to fat mobilization, potentially causing metabolic and reproductive disorders. Changes in haemodynamics, hepatic function, and hormonal status during pregnancy and lactation require a balance between anabolism and catabolism. Blood metabolic profiles are useful to assess health and nutritional status, while haematology aids in diagnosing and monitoring diseases. Understanding these changes is crucial for optimizing milk productivity and managing health during the transition period. The present study focusses on evaluating haematological and biochemical changes in Purnathadi buffaloes during this critical phase.

Healthy pregnant Purnathadi buffaloes (9) maintained at Instructional Livestock Farm, PGIVAS, Akola were selected for present study. The pregnancy was confirmed using ultrasonography. All the experimental animals were maintained under similar management and feeding conditions. These animals were fed as per Kearn (1982) feeding standards to meet out their requirements for maintenance and pregnancy. They were provided with *ad lib.* clean and fresh supply of water. All the buffaloes were offered concentrate, twice a day, i.e. 8.00 AM and 6.30 PM daily. All animals were apparently healthy throughout the

experimental period. The blood samples were collected aseptically from all the experimental animals from the external jugular vein using vacutainers (EDTA vacutainer for whole blood and plain vacutainer for serum preparation), 21 days before parturition, on the day of parturition, and 21 days after parturition. The non-esterified fatty acid was analysed with the chemical reagents and other biochemical parameters were analysed with reagent kits on PC-Based Double Beam Spectrophotometer 2201 and AGD 2020 Clinical Chemistry Analyzer, respectively. Haematology was performed on the same day within 3 h with the help of a Haematological Auto Analyzer (ABAXIS Vetscan HM5), and differential leucocyte count was done manually. According to Snedecor and Cochran (1994), the data gathered during this inquiry were treated to a one-way analysis of variance using a completely randomised design (CRD) with treatment as a factor (1994). Means were compared using Tukey’s test and Duncan’s multiple range test for certain parameters, and data were processed for statistical analysis using SPSS software version (29.0) (IBM 2022).

**Biochemical parameters:** The study observed significant metabolic changes in Purnathadi buffaloes during the transition period (Table 1). On the day of parturition, non-esterified fatty acids (NEFA) levels showed significant increase ( $p < 0.05$ ) and continued to rise 21 days postpartum. Total cholesterol levels were lower on the day of parturition compared to 21 days before, but significantly ( $p < 0.01$ ) increased to peak at 21 days postpartum. Triglycerides, albumin and globulin, alanine transaminase (ALT), and aspartate transaminase (AST) levels showed non-significant changes. Glucose levels increased non-significantly on the day of parturition, but significantly decreased 21 days later ( $p < 0.01$ ). Throughout the transition period, total protein concentrations increased considerably ( $p < 0.05$ ), reaching a high at 21 days postpartum. Calcium levels declined non-significantly, and phosphorus levels, highest on the day of parturition, decreased significantly ( $p < 0.01$ ) by 21 days postpartum.

NEFA concentration is influenced by the energy balance of the animal, the onset of parturition stress, and its

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Table 1. Mean±SE values of biochemical parameters during transition period

| Parameter                          | Transition period | 21 days before parturition | On the day of parturition | 21 days after parturition |
|------------------------------------|-------------------|----------------------------|---------------------------|---------------------------|
| Non-esterified fatty acid (mmol/L) |                   | 0.39 ± 0.07 <sup>b</sup>   | 0.64 ± 0.09 <sup>a</sup>  | 0.72 ± 0.08 <sup>a</sup>  |
| Total cholesterol (mg/dL)          |                   | 87.31 ± 1.3 <sup>b</sup>   | 84.33 ± 0.68 <sup>b</sup> | 92.78 ± 2.28 <sup>a</sup> |
| Triglyceride (mg/dL)               |                   | 160.81 ± 2.51              | 154.31 ± 3.70             | 151.46 ± 2.92             |
| Glucose (mg/dL)                    |                   | 41.14 ± 1.38 <sup>a</sup>  | 43.87 ± 1.55 <sup>a</sup> | 36.33 ± 0.89 <sup>b</sup> |
| Total protein (g/dL)               |                   | 7.56 ± 0.22 <sup>b</sup>   | 8.04 ± 0.28 <sup>ab</sup> | 8.61 ± 0.25 <sup>a</sup>  |
| Albumin (g/dL)                     |                   | 2.43 ± 0.15                | 2.81 ± 0.24               | 2.73 ± 0.12               |
| Globulin (g/dL)                    |                   | 5.13 ± 0.25                | 5.23 ± 0.31               | 5.88 ± 0.24               |
| Alanine transaminase (IU/L)        |                   | 32.78 ± 4.39               | 28.28 ± 3.01              | 33.14 ± 5.75              |
| Aspartate transaminase (IU/L)      |                   | 116.39 ± 8.11              | 109.96 ± 12.84            | 108.59 ± 5.75             |
| Calcium (mg/dL)                    |                   | 24.85 ± 1.56               | 23.25 ± 1.15              | 23.12 ± 0.96              |
| Phosphorous (mg/dL)                |                   | 4.67 ± 0.36 <sup>a</sup>   | 5.59 ± 0.62 <sup>a</sup>  | 2.83 ± 0.20 <sup>b</sup>  |

Mean bearing same superscript in row did not differ significantly.

previous nutritional status (Khan *et al.* 2011). In this study, NEFA concentration showed a gradual increase during the transition period. During this period, reduced feed intake (Fiore *et al.* 2017) and elevated energy demands for lactation create a negative energy balance, leading to adipose tissue mobilization (Abdelrazek *et al.* 2018). The NEFA concentration in blood reflects the degree of adipose tissue mobilization. Thus, a more pronounced negative energy balance leads to increased release of NEFA from body fat, resulting in higher blood NEFA concentrations (Pande *et al.* 2016). El-Maghraby *et al.* (2016) reported that lipid catabolism from reserves and a physiological shift to satisfy lactation requirements were responsible for the increase in total cholesterol content during the postpartum weeks. The level of blood triglycerides serves as a crucial source of long-chain fatty acids for milk production (Tharwat *et al.* 2023). The high initial values observed here were associated with milk production that has not yet reached its peak (Ashmawy 2015, Jyani *et al.* 2018) and the further decrease in the concentration could be ascribed to the upregulation of mammary gland lipoprotein lipase (Tharwat *et al.* 2023).

The blood glucose level is regarded as a key indicator

reflecting the energy status of buffaloes (Tanmaie *et al.* 2022). According to the current study, hyperglycemia on the day of parturition is associated with the gluconeogenic effect of cortisol and adrenaline due to the excitement and stress associated with parturition, whereas decreased glucose concentration 21 days postpartum is caused by the mammary gland's significant glucose withdrawal for milk lactose synthesis (Aswal *et al.* 2021). A significant increase in total protein concentration in the postpartum phase may be due to haemoconcentration and water losses during parturition (Das *et al.* 2017) and maternal protein requirements for lactation and the provision of immunoglobulins (Tharwat *et al.* 2023). The postpartum decline in calcium concentration is due to its significant excretion in colostrum during intense lactation (Tharwat *et al.* 2023), while the decrease in phosphorus concentration is linked to its role in colostrum synthesis and increased carbohydrate metabolism (Reddy and Sivajothi 2020). Also Singh *et al.* (2021) indicated that the decrease in Ca level causes higher PTH levels leading to drainage of phosphorus through urine.

*Haematological parameters:* In this study, several haematological parameters exhibited significant changes

Table 2. Mean±SE values of haematological parameters during transition period

| Parameter                 | Transition period | 21 days before parturition | On the day of parturition | 21 days after parturition  |
|---------------------------|-------------------|----------------------------|---------------------------|----------------------------|
| TEC (10 <sup>6</sup> /μL) |                   | 6.59 ± 0.27 <sup>b</sup>   | 7.61 ± 0.44 <sup>a</sup>  | 6.40 ± 0.26 <sup>b</sup>   |
| Hb (g/dL)                 |                   | 12.39 ± 0.72               | 12.02 ± 0.36              | 11.70 ± 0.43               |
| PCV (%)                   |                   | 41.14 ± 2.39               | 38.31 ± 1.39              | 36.89 ± 1.65               |
| MCV (fL)                  |                   | 62.38 ± 2.00 <sup>a</sup>  | 51.58 ± 3.3 <sup>b</sup>  | 58.3 ± 3.09 <sup>ab</sup>  |
| MCH (pg)                  |                   | 18.7 ± 0.58                | 16.12 ± 0.97              | 18.46 ± 0.81               |
| MCHC (g/dL)               |                   | 30.09 ± 0.35 <sup>b</sup>  | 31.39 ± 0.33 <sup>a</sup> | 31.79 ± 0.54 <sup>a</sup>  |
| TLC (10 <sup>3</sup> /uL) |                   | 11.5 ± 0.92                | 12.41 ± 0.73              | 13.24 ± 0.90               |
| Neutrophil %              |                   | 31.22 ± 2.62 <sup>b</sup>  | 41.89 ± 4.03 <sup>a</sup> | 45.11 ± 3.53 <sup>a</sup>  |
| Eosinophil %              |                   | 2.67 ± 0.41                | 2.78 ± 0.52               | 2.56 ± 0.78                |
| Basophil %                |                   | 0.44 ± 0.34                | 0.22 ± 0.22               | 00                         |
| Lymphocyte %              |                   | 51.8 ± 1.92 <sup>a</sup>   | 40.52 ± 3.15 <sup>b</sup> | 45.16 ± 2.53 <sup>ab</sup> |
| Monocyte %                |                   | 6.19 ± 0.51                | 5.61 ± 0.48               | 6.27 ± 0.55                |

Mean bearing same superscript in row did not differ significantly.

during the peripartum period (Table 2). On the day of parturition, the total erythrocyte count (TEC) became significantly ( $p < 0.05$ ) higher, but by day 21 postpartum, it had significantly dropped. Haemoglobin (Hb) concentration and packed cell volume (PCV) showed a non-significant decline over the peripartum period. Mean corpuscular volume (MCV) declined significantly ( $p < 0.05$ ) on the day of parturition and increased notably by day 21 postpartum, while mean corpuscular haemoglobin concentration (MCHC) improved significantly ( $p < 0.05$ ) on the day of parturition and non-significantly by day 21 postpartum. The mean corpuscular haemoglobin (MCH), total leukocyte count (TLC), eosinophil %, basophil %, and monocyte % showed non-significant changes. On the day of parturition, the neutrophil % became significantly ( $p < 0.05$ ) high, whereas the lymphocytes % significantly ( $p < 0.05$ ) declined and then significantly rose by day 21 postpartum.

According to Gomaa *et al.* (2021), the present study's findings regarding the decline in TEC during the postpartum period could be explained by modifications in nutritional status that impact erythropoiesis and lower blood erythrocyte concentration. Meanwhile, Mohamed *et al.* (2015) attributed the decrease in haemoglobin concentration during the transition period to the stress of pregnancy and parturition in addition to reduced erythropoiesis. The stress of parturition and lactation may be the cause of the decrease in packed cell volume (PCV) values seen during the study's transition period (Kour *et al.* 2023). The decrease in MCV observed in this research may be ascribed to increased cell division resulting from insufficient haemoglobin concentration (Mohamed *et al.* 2015). The increase in TLC at parturition and during the postpartum period observed in this research is attributed to elevated cortisol levels, which stimulate the bone marrow and cause a rise in TLC (Abdelrazek *et al.* 2018). Elevated cortisol levels during stressful conditions also explain the increased neutrophil % and the non-significant reduction in eosinophil % due to cortisol's immunosuppressive effects. The decline in basophil and monocyte levels, as well as the significant decrease in lymphocyte % on the day of parturition, are also linked to the immunosuppressive effects of elevated cortisol (Abdelrazek *et al.* 2018).

Non-esterified fatty acids and triglycerides are crucial biomarkers for evaluating energy balance during the transition period. This study identified these markers as significant, indicating that although standard management practices were adhered to at the farm, special attention was needed for pregnant buffaloes. Elevated triglyceride levels may suggest increased mobilization of fat reserves to meet the animals' metabolic demands, ensuring adequate energy for fetal development, milk production, and postpartum recovery. The study's findings imply that the animals experienced metabolic stress, offering insights into the physiological changes in Purnathadi buffaloes during critical stages such as pre-parturition, parturition, and post-parturition. This information could

aid in refining management practices to optimize the health and productivity of these buffaloes in similar climatic conditions. However, since no prior research has been conducted on Purnathadi buffaloes, further studies are required to confirm these findings.

## SUMMARY

This study investigated the impact of the transition period on the blood metabolic profile of Purnathadi buffaloes in Vidarbha's tropical climate. Blood samples were taken from nine pregnant buffaloes 21 days before parturition, at parturition, and 21 days after parturition. Triglycerides, albumin, globulin, alanine transaminase, aspartate transaminase, calcium, haemoglobin, packed cell volume, mean corpuscular haemoglobin, total leukocyte count, eosinophils, basophils and monocytes were non-significantly affected. However, non-esterified fatty acids, total cholesterol, glucose, total protein, phosphorus, total erythrocyte count, mean corpuscular volume, mean corpuscular haemoglobin concentration, neutrophils and lymphocytes were significantly affected during transition period. Elevated triglyceride levels indicated increased fat mobilization for fetal development and postpartum recovery. The study highlights the metabolic stress during this period, offering insights for improved management practices to support buffalo health and productivity.

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