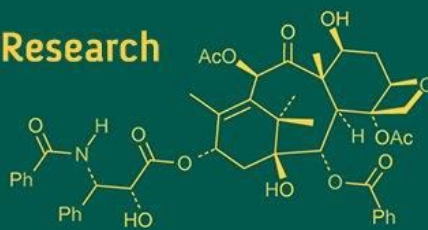


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**Dr. Neha Tomar**  
Department of Livestock  
Production Management, College  
of Veterinary Science and A.H.,  
(NDVSU), Mhow, Indore,  
Madhya Pradesh, India

**Dr. DS Yadav**  
Department of Livestock  
Production Management, College  
of Veterinary Science and A.H.,  
(NDVSU), Mhow, Indore,  
Madhya Pradesh, India

**Dr. S Nanavati**  
Department of Livestock  
Production Management, College  
of Veterinary Science and A.H.,  
(NDVSU), Mhow, Indore,  
Madhya Pradesh, India

**Dr. MS Jamra**  
Department of Livestock  
Production Management, College  
of Veterinary Science and A.H.,  
(NDVSU), Mhow, Indore,  
Madhya Pradesh, India

**Dr. N Kurechiya**  
Department of Animal Nutrition,  
College of Veterinary Science and  
A.H., (NDVSU), Mhow, Indore,  
Madhya Pradesh, India

**Dr. NS Rawat**  
Department of Livestock  
Production Management, College  
of Veterinary Science and A.H.,  
(NDVSU), Mhow, Indore,  
Madhya Pradesh, India

**Dr. Nirmala Jamra**  
Department of veterinary  
Parasitology, College of  
Veterinary Science and A.H.,  
(NDVSU), Mhow, Indore,  
Madhya Pradesh, India

**Dr. KP Saini**  
Department of Livestock  
Production Management, College  
of Veterinary Science and A.H.,  
(NDVSU), Mhow, Indore,  
Madhya Pradesh, India

**Corresponding Author:**  
**Dr. Neha Tomar**  
Department of Livestock  
Production Management, College  
of Veterinary Science and A.H.,  
(NDVSU), Mhow, Indore,  
Madhya Pradesh, India

## Effect of *Moringa oleifera* leaves supplementation on blood profile of murrah buffalo calves

**Neha Tomar, DS Yadav, S Nanavati, MS Jamra, NS Rawat, N Kurechiya, Nirmal Jamra and KPS Saini**

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### Abstract

The present study was undertaken to evaluate the effect of *Moringa oleifera* leaves supplementation on haematological profile of Murrah buffalo calves. For haematological evaluation, 2mL of blood was collected aseptically from each calf by jugular vein puncture into EDTA-containing vials at every monthly intervals during morning hours prior to feeding and watering for a period of three months. Haematological analysis revealed a significant increase in total erythrocyte count, packed cell volume, mean corpuscular volume, and mean corpuscular haemoglobin in the supplemented groups compared to the control, while total leucocyte count, haemoglobin concentration, and mean corpuscular haemoglobin concentration showed an increasing trend but the differences were statistically non-significant. Overall, the findings indicated that *Moringa oleifera* leaves supplementation positively influenced the haematological profile of Murrah buffalo calves without exerting any adverse effects.

**Keywords:** *Moringa oleifera* leaves, Murrah buffalo calves and haematological

### Introduction

Buffaloes are an integral part of the Indian agricultural economy, providing substantial support to smallholder farmers and landless labours and contributing more than 67% of the total dairy animal population in the country. According to the 20<sup>th</sup> Livestock Census, India possesses the largest buffalo population in the world, with an estimated 109.85 million animals. Buffaloes play a crucial role in the rural economy of India by contributing significantly to milk production, providing meat, and supplying draft power for agricultural operations. Adequate nutrition and proper management during the early stages of life are critical for optimal growth, physiological development, future productivity, and overall health of buffalo calves. Nevertheless, calf rearing is frequently overlooked due to high rearing costs and delayed economic returns, which may negatively impact long-term performance. *Moringa oleifera*, commonly known as the drumstick tree or horseradish tree and locally referred to as “Saijhan” or “Mungaa,” is a fast-growing, deciduous, multipurpose leguminous tree widely recognized for its nutritional and functional properties reported by Mishra *et al.* (2012) [9]. For centuries, Moringa has been valued for its diverse benefits and is often described as the “Miracle Tree” or “Magic Tree” by Falowo *et al.* (2018) [4]. Its leaves are particularly rich in nutrients and are considered to provide a more complete nutritional profile compared to many conventional plant-based feed resources by Jattan *et al.* (2021) [5]. Dried Moringa leaves contain high levels of crude protein (30.3%), essential amino acids (19%), vitamins, dietary fiber and bioactive compounds such as tannins and polyphenols, along with approximately 17% fatty acids, including palmitic, capric, alpha-linolenic, and gamma-linolenic acids reported by Kumar *et al.* (2025) [6]. Owing to this rich nutrient composition, *Moringa oleifera* leaves hold considerable promise as a sustainable and cost-effective feed supplement in buffalo nutrition.

### Materials and Methods

The study was conducted at the Livestock Farm Complex, Department of Livestock Production and Management, College of Veterinary Science and Animal Husbandry, NDVSU, Mhow (M.P.). A total of eighteen healthy male Murrah buffalo calves, aged 6-7 months and with comparable initial body weights, were selected for the experiment.

All animals were clinically examined prior to the study to ensure they were free from diseases and parasitic infestations. The experiment was carried out over a six-month period, including a 10 days acclimatization phase followed by a 90 days feeding trial. The calves were randomly allocated into three groups of six animals each. The control group (T<sub>0</sub>) was fed a basal diet of roughage and concentrate, while in treatment groups T<sub>1</sub> and T<sub>2</sub>, 5% and 10% of the concentrate mixture, respectively, were replaced with *Moringa oleifera* leaves powder.

### Haematological parameters

About 2 mL of blood was collected aseptically from each calf by jugular vein into clean, dry, sterilized vials containing EDTA as an anticoagulant. Blood sampling was carried out at monthly intervals on days 0, 30, 60, and 90 of the experimental period. Haematological parameters (Total erythrocyte count, total leucocytes count, Haemoglobin, packed cell volume, mean corpuscular volume, mean corpuscular haemoglobin concentration and mean corpuscular haemoglobin in growing Murrah buffalo calves at monthly interval.

### Statistical analysis

Data were subjected to statistical analysis using analysis of variance (one-way ANOVA) Snedecor and Cochran (1994). Duncan's multiple range test was used to compare differences among groups and period means for different parameters by using SPSS 20.0 software.

## Results and Discussion

### Haematological parameters in growing Murrah buffalo calves

#### Total erythrocyte count

The total erythrocyte count (TEC) of Murrah buffalo calves increased progressively over the 90 days study duration in all treatment groups (Table 01).

**Table 1:** Effect of *Moringa oleifera* leaves supplementation on total erythrocyte count (million/ $\mu$ L) in Murrah buffalo calves

| Months                | T <sub>0</sub>               | T <sub>1</sub>                | T <sub>2</sub>                | p value |
|-----------------------|------------------------------|-------------------------------|-------------------------------|---------|
| Initial               | 7.17 $\pm$ 0.16              | 8.03 $\pm$ 0.11               | 8.11 $\pm$ 0.13               | 0.11    |
| 1                     | 7.22 $\pm$ 0.17              | 8.03 $\pm$ 0.15               | 8.36 $\pm$ 0.13               | 0.11    |
| 2                     | 7.47 <sup>b</sup> $\pm$ 0.15 | 8.68 <sup>a</sup> $\pm$ 0.16  | 9.37 <sup>a</sup> $\pm$ 0.17  | 0.01    |
| 3                     | 8.24 <sup>b</sup> $\pm$ 0.15 | 9.41 <sup>ab</sup> $\pm$ 0.18 | 10.06 <sup>a</sup> $\pm$ 0.20 | 0.03    |
| Overall mean $\pm$ SE | 7.52 <sup>b</sup> $\pm$ 0.16 | 8.54 <sup>ab</sup> $\pm$ 0.15 | 8.98 <sup>a</sup> $\pm$ 0.16  | 0.04    |

a, b, c = Mean bearing different superscript in row differ significantly ( $p \leq 0.05$ )

The overall mean TEC value were greatest in T<sub>2</sub> (8.98 $\pm$ 0.16 million/ $\mu$ L, with T<sub>1</sub> (8.54 $\pm$ 0.15 million/ $\mu$ L) and T<sub>0</sub> (7.52 $\pm$ 0.16 million/ $\mu$ L) showing slightly lower count. At the 2<sup>nd</sup> and 3<sup>rd</sup> month, TEC in T<sub>1</sub> and T<sub>2</sub> was significantly higher than in T<sub>0</sub>. Analysis showed that treatment groups differed significantly at the 2<sup>nd</sup> and 3<sup>rd</sup> month as well as for the overall mean ( $p \leq 0.05$ ).

Similar results were observed in the studies by Wafa *et al.* (2017) [13] reported that supplementation of *Moringa oleifera* leaves in heat-stressed buffalo bulls significantly increased red blood cell (RBC) counts ( $\times 10^6/\text{mm}^3$ ), with values of 7.05  $\pm$  0.13 (G<sub>1</sub>, control), 7.49  $\pm$  0.11 (G<sub>2</sub>, 120 g/d) and 7.94  $\pm$  0.15 (G<sub>3</sub>, 240 g/d). Anwar *et al.* (2024) reported that heifers supplemented with 10% *Moringa oleifera* leaf

meal showed increased red blood cell (RBC) counts compared to the control group.

In contrast, Chandra (2023) [1] reported that replacing 20% of concentrate with *Moringa oleifera* in lactating Sahiwal cows had no significant effect on total erythrocyte count (TEC), indicating that the dietary inclusion did not adversely affect erythrocyte production.

### Total leucocyte count

At the beginning of the trial (Initial month), the mean TLC values were 11.57 $\pm$ 0.25  $\times 10^3/\mu\text{L}$ , 13.07 $\pm$ 0.19  $\times 10^3/\mu\text{L}$  and 13.19 $\pm$ 0.22  $\times 10^3/\mu\text{L}$  for the T<sub>0</sub>, T<sub>1</sub> and T<sub>2</sub> groups, respectively. A consistent rise was seen across all groups over successive months, reaching 13.75 $\pm$ 0.46  $\times 10^3/\mu\text{L}$ , 14.26 $\pm$ 0.31  $\times 10^3/\mu\text{L}$  and 15.28 $\pm$ 0.30  $\times 10^3/\mu\text{L}$  by the fourth month in T<sub>0</sub>, T<sub>1</sub> and T<sub>2</sub>, respectively. Among the groups, T<sub>2</sub> exhibited the greatest overall mean TLC (14.18 $\pm$ 0.24  $\times 10^3/\mu\text{L}$ ), with progressively lower values in T<sub>1</sub> (13.80 $\pm$ 0.19  $\times 10^3/\mu\text{L}$ ) and T<sub>0</sub> (12.96 $\pm$ 0.30  $\times 10^3/\mu\text{L}$ ).

Similarly, Sharma *et al.* (2024) [10] reported that replacing 50% of crude protein with *Moringa oleifera* in crossbred calves had no significant effect on total leukocyte count (WBC), indicating that immune cell levels remained stable. Contrast, Mandal *et al.* (2015) [7] reported that feeding 25 g of dried *Moringa oleifera* fruit powder to fluorosis-affected calves significantly increased total leukocyte count (TLC), along with other leukocyte fractions.

### Haemoglobin

At the beginning of the trial, the mean Hb values were 10.43 $\pm$ 0.15, 11.60 $\pm$ 0.16 and 11.43 $\pm$ 0.12 g/dL for the control (T<sub>0</sub>) and the *Moringa oleifera* supplemented groups (T<sub>1</sub> and T<sub>2</sub>), respectively. By the end of the third month, Hb values had increased to 12.08 $\pm$ 0.25 g/dL in T<sub>0</sub>, 13.32 $\pm$ 0.21 g/dL in T<sub>1</sub> and 13.50 $\pm$ 0.14 g/dL in T<sub>2</sub>. The highest overall mean Hb concentration was observed in T<sub>2</sub> (12.46 $\pm$ 0.13 g/dL), subsequently T<sub>1</sub> (12.31 $\pm$ 0.20 g/dL) and T<sub>0</sub> (11.17 $\pm$ 0.19 g/dL).

**Table 2:** Effect of *Moringa oleifera* leaves supplementation on haemoglobin (g/dL) in Murrah buffalo calves

| Months                | T <sub>0</sub>   | T <sub>1</sub>   | T <sub>2</sub>   | p value |
|-----------------------|------------------|------------------|------------------|---------|
| Initial               | 10.43 $\pm$ 0.15 | 11.60 $\pm$ 0.16 | 11.43 $\pm$ 0.12 | 0.07    |
| 1                     | 10.70 $\pm$ 0.13 | 11.80 $\pm$ 0.23 | 11.75 $\pm$ 0.11 | 0.12    |
| 2                     | 11.73 $\pm$ 0.20 | 12.53 $\pm$ 0.19 | 13.17 $\pm$ 0.16 | 0.11    |
| 3                     | 12.08 $\pm$ 0.25 | 13.32 $\pm$ 0.21 | 13.50 $\pm$ 0.14 | 0.13    |
| Overall mean $\pm$ SE | 11.17 $\pm$ 0.19 | 12.31 $\pm$ 0.20 | 12.46 $\pm$ 0.13 | 0.14    |

During the experimental period, Hb levels increased gradually, but the differences were not statistically significant ( $p > 0.05$ ). The results suggest that *Moringa oleifera* leaf supplementation at the T<sub>2</sub> level improved haemoglobin concentration relative to the control, reflecting beneficial effects on the haematological profile of Murrah buffalo calves

Likewise, Mandal *et al.* (2015) [7] reported that feeding fluorosis-affected calves with 25 g of dried *Moringa oleifera* fruit powder significantly increased haemoglobin (Hb) levels. Foidl and Paull (2014) reported a slight increase in haemoglobin in moringa-fed groups.

In contrast, Elaidy *et al.* (2017) [3] reported that replacing calf starter with up to 20% *Moringa oleifera* leaf meal had no significant effect on haemoglobin (Hb), indicating that

the dietary inclusion did not adversely affect the haematological status of suckling buffalo calves.

### Packed cell volume

At the start of the trial (Initial month), the mean PCV values were  $33.72 \pm 0.25\%$ ,  $34.38 \pm 0.29\%$  and  $35.96 \pm 0.20\%$  for T<sub>0</sub>,

T<sub>1</sub> and T<sub>2</sub>, respectively, where differences among the groups were significant ( $p \leq 0.05$ ). This increase became highly significant in the later stages of the experiment, with PCV values reaching  $36.32 \pm 0.32\%$ ,  $39.80 \pm 0.39\%$  and  $42.03 \pm 0.14\%$  in T<sub>0</sub>, T<sub>1</sub> and T<sub>2</sub>, respectively, by the third month.

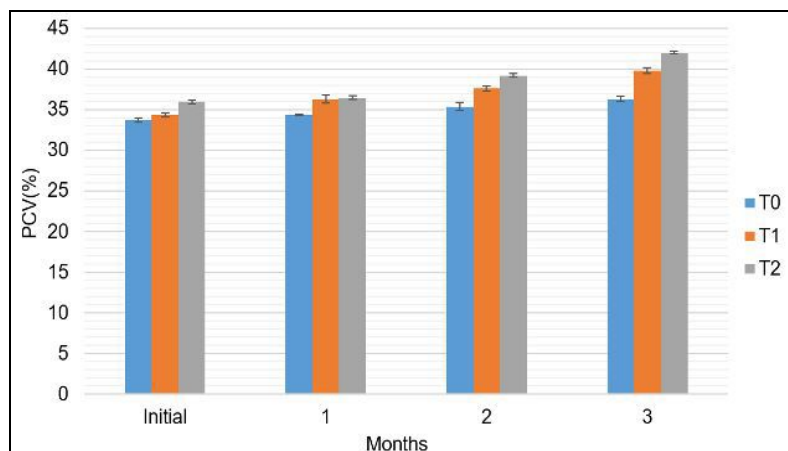


**Fig 1:** Effect of *Moringa oleifera* leaves supplementation on packed cell volume (%) in Murrah buffalo calves

Overall, the mean PCV was greatest in the T<sub>2</sub> group ( $38.42 \pm 0.20\%$ ), followed by T<sub>1</sub> ( $37.03 \pm 0.38\%$ ) and T<sub>0</sub> ( $34.94 \pm 0.30\%$ ). Statistical analysis indicated that supplementation with *Moringa oleifera* leaves had a highly significant positive effect on PCV, suggesting enhanced erythropoiesis and improved haematological status in Murrah buffalo calves during the 90 days study period.

Likewise, Wafa *et al.* (2017) [13] reported that supplementation of *Moringa oleifera* leaves in heat-stressed buffalo bulls significantly increased packed cell volume (PCV), with values of  $31.39 \pm 0.35$  (G<sub>1</sub>, control),  $33.69 \pm 0.19$  (G<sub>2</sub>, 120 g/d) and  $34.84 \pm 0.15$  (G<sub>3</sub>, 240 g/d) indicating improved oxygen-carrying capacity and physiological status. Ehshan *et al.* (2025) reported that supplementation of *Moringa oleifera* leaves through urea molasses blocks significantly increased packed cell volume (PCV) in Azikheli buffaloes, with the 30% inclusion group showing the highest values.

In contrast, Elaidy *et al.* (2017) [3] observed that replacing calf starter with up to 20% *Moringa oleifera* leaf meal had no significant effect on PCV in suckling buffalo calves.

### Mean corpuscular volume

During the initial stage and throughout the first, second, and third months, MCV values showed no significant difference among the treatment groups ( $p > 0.05$ ). In contrast, the overall mean MCV was significantly greater in non-supplemented group (T<sub>0</sub>:  $47.08 \pm 0.93$  fL) compared to the *Moringa oleifera* supplemented groups T<sub>1</sub> ( $43.67 \pm 0.56$  fL) and T<sub>2</sub> ( $43.21 \pm 0.63$  fL) ( $p < 0.01$ ).

**Table 4:** Effect of *Moringa oleifera* leaves supplementation on mean corpuscular volume (fL) in Murrah buffalo calves

| Months            | T <sub>0</sub>            | T <sub>1</sub>            | T <sub>2</sub>            | p value |
|-------------------|---------------------------|---------------------------|---------------------------|---------|
| Initial           | 47.86 ± 1.27              | 43.01 ± 0.57              | 44.58 ± 0.54              | 0.28    |
| 1                 | 48.37 ± 1.15              | 45.41 ± 0.42              | 43.86 ± 0.54              | 0.26    |
| 2                 | 47.69 ± 0.73              | 43.61 ± 0.57              | 42.15 ± 0.64              | 0.07    |
| 3                 | 44.37 ± 0.56              | 42.66 ± 0.68              | 42.27 ± 0.80              | 0.65    |
| Overall mean ± SE | 47.08 <sup>a</sup> ± 0.93 | 43.67 <sup>b</sup> ± 0.56 | 43.21 <sup>b</sup> ± 0.63 | 0.01    |

a, b, c = Mean bearing different superscript in row differ significantly ( $p \leq 0.05$ )

Monthly analysis revealed that differences in MCV was observed across the treatment groups, and the differences were not significant ( $p > 0.05$ ) during individual months. However, when the overall mean of the 90 days period was considered, a significant difference was detected. The mean MCV in T<sub>0</sub> was significantly higher than in T<sub>1</sub> and T<sub>2</sub>, indicating that while monthly variations were minor, MOL supplementation had a cumulative impact on the average red blood cell volume over the entire experimental period.

Present results are supported with the result of Wafa *et al.* (2017) [13], who reported significantly higher MCV values in heat-stressed buffalo bulls supplemented with 4% and 8% *Moringa oleifera* leaves.

On the contrary, Mishra (2008) [8] who also reported non-significant difference in MCV concentration in lactating animals received MOLM and shatavari. Elaidy *et al.* (2017) [3] observed no significant change in mean corpuscular volume when calf starter was replaced with up to 20% *Moringa oleifera* leaf meal.

### Mean corpuscular haemoglobin concentration

The mean corpuscular haemoglobin concentration of Murrah buffalo calves varied slightly during the experimental period (Table 05). At the start of the study, MCHC values were  $30.53 \pm 0.67$  g/dL,  $33.83 \pm 0.59$  g/dL and  $31.78 \pm 0.24$  g/dL for T<sub>0</sub>, T<sub>1</sub> and T<sub>2</sub>, respectively. The overall mean MCHC was highest in T<sub>1</sub> ( $33.26 \pm 0.42$  g/dL), succeeded by T<sub>2</sub> ( $32.43 \pm 0.29$  g/dL) and T<sub>0</sub> ( $31.96 \pm 0.52$  g/dL).

**Table 5:** Effect of *Moringa oleifera* leaves supplementation on mean corpuscular haemoglobin concentration (g/dL) in Murrah buffalo calves

| Months            | T <sub>0</sub> | T <sub>1</sub> | T <sub>2</sub> | p value |
|-------------------|----------------|----------------|----------------|---------|
| Initial           | 30.53 ± 0.67   | 33.83 ± 0.59   | 31.78 ± 0.24   | 0.23    |
| 1                 | 30.85 ± 0.39   | 32.46 ± 0.34   | 32.21 ± 0.24   | 0.34    |
| 2                 | 33.24 ± 0.54   | 33.27 ± 0.29   | 33.59 ± 0.35   | 0.96    |
| 3                 | 33.21 ± 0.48   | 33.46 ± 0.45   | 32.13 ± 0.33   | 0.64    |
| Overall mean ± SE | 31.96 ± 0.52   | 33.26 ± 0.42   | 32.43 ± 0.29   | 0.24    |

MCHC values were comparable among the treatment groups, with no significant differences detected ( $p>0.05$ ) at any time point or for the overall mean. This indicates that supplementation with *Moringa oleifera* leaves did not substantially affect the haemoglobin concentration of red blood cells in Murrah buffalo calves and all values remained within the normal physiological range.

Likewise, Mandal *et al.* (2015) [7] reported that administration of 25 g of dried *Moringa oleifera* fruit powder to calves affected by fluorosis did not produce any significant changes in the erythrocyte indices MCHC.

In contrast, Sharma *et al.* (2024) [10] reported that replacing 50% of crude protein with *Moringa oleifera* in crossbred calves significantly increased mean corpuscular haemoglobin concentration (MCHC), indicating improved red blood cell quality.

### Mean corpuscular haemoglobin

**Table 6:** Effect of *Moringa oleifera* leaves supplementation on mean corpuscular haemoglobin (pg) in Murrah buffalo calves

| Months            | T <sub>0</sub>           | T <sub>1</sub>            | T <sub>2</sub>           | p value |
|-------------------|--------------------------|---------------------------|--------------------------|---------|
| Initial           | 14.40±0.18               | 14.48±0.17                | 14.16±0.17               | 0.86    |
| 1                 | 14.80±0.17               | 14.71±0.11                | 14.10±0.11               | 0.29    |
| 2                 | 15.75 <sup>a</sup> ±0.12 | 14.49 <sup>b</sup> ±0.18  | 14.09 <sup>b</sup> ±0.10 | 0.01    |
| 3                 | 14.73±0.28               | 14.20±0.13                | 13.51±0.15               | 0.23    |
| Overall mean ± SE | 14.92 <sup>a</sup> ±0.19 | 14.47 <sup>ab</sup> ±0.15 | 13.96 <sup>b</sup> ±0.14 | 0.02    |

a, b, c = Mean bearing different superscript in row differ significantly ( $p\leq 0.05$ )

The overall mean MCH was highest in T<sub>0</sub> (14.92±0.19 pg), followed by T<sub>1</sub> (14.47±0.15 pg) and T<sub>2</sub> (13.96±0.14 pg), with the differences being statistically significant. This suggests that supplementation with *Moringa oleifera* leaves resulted in slightly lower MCH values in T<sub>1</sub> and T<sub>2</sub>, though all values remained within the normal physiological range. Present result is corroborated with Umar *et al.* (2017) [12] who also reported similar MCH concentration between the control group and the group received *M. oleifera*.

### Conclusion

*Moringa oleifera* supplementation improved key blood parameters in Murrah buffalo calves, with significant increases in TEC, Hb and PCV-greatest in T<sub>2</sub>-indicating enhanced erythropoiesis. Other indices remained within normal limits.

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