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Seasonal and Spatial Variation in Selected Mineral Concentrations of Dairy Cows in the Rift Valley of Ethiopia: A Case Study of Bishoftu and Wonji Shewa

Wondewsen Bekele

Department of Animal and Range Sciences, College of Agriculture & Natural Resources, Dilla University, Ethiopia.
Email: wondewsen19@gmail.com

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Abstract

Mineral nutrition is a key determinant of dairy cow productivity and health in Ethiopia's Rift Valley, where seasonal fluctuations and location-specific feeding systems influence mineral availability. Despite its importance, evidence on seasonal and spatial variations in the mineral status of lactating dairy cows in Ethiopia's major dairy-producing areas remains limited. This knowledge gap constrains the development of effective, location-specific mineral supplementation strategies. This study assessed seasonal and spatial variations in calcium, phosphorus, magnesium, copper, iron, zinc, and cobalt in lactating dairy cows and their feeds in the dairy production areas of Bishoftu and Wonji. A total of 80 blood and 78 feed samples collected during the dry and wet seasons were analyzed using standard laboratory procedures. Significant location × season interactions were observed for Ca and Fe ($p < 0.05$). Calcium was marginally adequate, phosphorus remained close to deficiency thresholds, and magnesium and zinc were generally adequate, particularly during the dry season. Copper concentrations were below critical levels and were likely worsened by elevated iron concentrations. Mineral deficiencies were more prevalent in Wonji Shewa (72.5%) than Bishoftu (60%) and were associated with season and physiological status. Lactating cows sampled during the dry season showed the highest deficiency rates. In addition, farmers had limited awareness of mineral deficiency symptoms, especially subclinical reproductive effects. Overall, mineral status varied significantly across locations and seasons, with copper deficiency emerging as the major nutritional constraint. Region- and season-specific mineral supplementation programs, along with farmer education on mineral nutrition and deficiency recognition, should be implemented to improve dairy productivity and cow health.

Keywords/Phrases: Dairy cow nutrition, Trace mineral deficiency, Seasonal dynamics, Spatial variability, Rift Valley

1 Introduction

Mineral nutrition is essential for dairy cow health, reproduction, immune function, and productivity because minerals play key roles in metabolic processes, skeletal development, enzyme activation, and hormonal regulation (McDowell, 2003; Suttle, 2010). In Ethiopia's Rift Valley, where livestock farming supports millions of livelihoods, mineral deficiencies —especially calcium (*Ca*), phosphorus (*P*), copper (*Cu*), and cobalt (*Co*) —pose major challenges to sustainable dairy production. The region's diverse agro-ecological zones and distinct wet and dry seasons influence the mineral composition of soils and forages, thereby affecting the mineral intake of cattle. Variations in soil characteristics, rainfall patterns, vegetation types, and forage species across agro-ecological zones can change the concentration, bioavailability, and uptake of minerals by plants, ultimately influencing the mineral status of grazing animals (McDowell & Arthington, 2005; Underwood & Suttle, 1999).

Studies have shown that Ethiopian forages often contain inadequate levels of essential minerals, particularly during the dry season when pasture quality and biomass availability decline (Gizachew & Smit, 2012). Seasonal rainfall fluctuations not only affect forage quantity but also influence nutrient cycling, plant maturity, and mineral accumulation in pasture species. During prolonged dry periods, reduced forage growth and increased lignification may decrease mineral concentrations and mineral bioavailability to ruminants (Suttle, 2010; McDowell, 2003). As a result, serum mineral levels in dairy cows from areas such as East Shoa are frequently below critical thresholds, leading to suboptimal health, reduced milk production, impaired fertility, weakened immune responses, and increased susceptibility to metabolic disorders (Wondewsen *et al*., 2020).

Despite the recognized importance of mineral nutrition, detailed information on the seasonal and spatial variation of mineral concentrations in dairy cows —particularly comparisons between lactating and dry cows—remains limited in Ethiopia's Rift Valley (Gizachew & Smit, 2012). Physiological differences associated with lactation, including increased mineral demands for milk synthesis, may further affect mineral status and worsen deficiencies when dietary mineral supply is inadequate (Suttle, 2010). This knowledge gap limits the development of evidence-based, region-specific, and season-specific mineral supplementation strategies to improve animal health and productivity (Khakurel *et al*., 2021).

Therefore, this study investigates the macro- and micro-mineral status of dairy cows in Bishoftu and Wonji Shewa across different seasons and physiological stages. The study aims to generate baseline information that can guide targeted nutritional interventions and improve dairy production efficiency in Ethiopia's Rift Valley.

2 Materials and Methods

2.1 Study Area

The study was conducted in Bishoftu and Wonji Shewa, two districts in the East Shewa Zone of the Oromia Region, Ethiopia, located in the central Rift Valley. Bishoftu lies at an elevation of approximately 1,850 – 1,900m above sea level and has a subtropical highland climate with bimodal rainfall patterns and a rapidly expanding peri-urban dairy sector. Wonji Shewa is located at an elevation of 1,500 – 1,700m above sea level, has a semi-humid climate, and benefits from irrigation-based agriculture associated with the Awash River basin. Both areas have distinct agro-ecological characteristics and support mixed crop–livestock production systems (Abebe *et al*., 2020; CSA, 2022).

2.2 Study Population

The study included a total of 80 dairy cows, comprising 40 from Bishoftu and 40 from Wonji Shewa. In each district, 20 lactating and 20 dry cows were selected to ensure balanced representation of physiological status. Cows were chosen based on age (2 – 5 years) and good general health, which was confirmed through clinical examination by a veterinarian. These selection criteria were used to minimize potential confounding effects related to age and health status.

2.3 Blood Sample Collection

Blood samples were aseptically collected from the jugular vein of each cow using sterile vacutainer tubes according to standard veterinary procedures (Radostits *et al*., 2007). For plasma mineral analysis, blood was drawn into tubes containing ethylenediaminetetraacetic acid (EDTA) as an anticoagulant. Immediately after collection, samples were labeled and transported to the laboratory in an ice-cooled container. Upon arrival, plasma samples were stored at -20°C until laboratory analysis.

2.4 Plasma Mineral Estimation

Plasma concentrations of calcium (Ca), phosphorus (P), magnesium (Mg), copper (Cu), iron (Fe), zinc (Zn), and cobalt (Co) were determined using Atomic Absorption Spectrophotometry (AAS) following standard analytical procedures (Underwood and Suttle, 1999). Prior to analysis, plasma samples were thawed and prepared according to laboratory protocols. Calibration standards and quality-control samples were included throughout the analytical procedure.

The concentration of each mineral was calculated using the following equation:

Mineral concentration ($mg\ L^{-1}$) =

$$\frac{(Absorbance \times Standard\,Concentration) \times Dilution\,Factor}{Sample\,Volume}$$

Where:

- Absorbance = instrument reading of the sample
- Standard Concentration = concentration of calibration standard
- Dilution Factor = dilution applied during sample preparation
- Sample Volume = volume of plasma analyzed

2.5 Trace mineral concentrations

Trace mineral concentrations of copper (Cu), iron (Fe), zinc (Zn), and cobalt (Co) were determined using an ECIL 4141 Atomic Absorption Spectrophotometer (Electronics Corporation of India Ltd., India) equipped with a double-beam optical system operating over a wavelength range of 190 – 900nm (Reddy *et al.*, 2021). Each mineral was analyzed using its corresponding hollow cathode lamp with an air–acetylene flame. Calibration curves were prepared using at least three standard solutions. Mineral concentrations were recorded in parts per million (ppm) using a computer-integrated data acquisition system. To minimize cross-contamination, the instrument was flushed with distilled water for at least 10 minutes between successive samples.

2.6 Feed Sample Collection Preparation

A total of 78 feed samples, consisting of 30 roughage, 36 concentrate, and 12 non-conventional feed samples, were collected from dairy farms in Bishoftu and Wonji Shewa during both wet and dry seasons using random sampling procedures. Approximately 500 g of each sample was collected according to established protocols (FAO, 2013; NRC, 2001). Samples were placed in labelled polyethylene bags and transported to the laboratory for analysis. Upon arrival, samples were air-dried, ground to pass through a 1mm sieve, and stored in airtight containers until analysis (AOAC, 2005). Macro-minerals (Ca, P, and Mg) and trace minerals (Cu, Fe, Zn, and Co) were determined using Atomic Absorption Spectrophotometry following AOAC (2005) procedures.

2.7 Digestion of Plasma Samples

Before mineral determination, plasma samples were digested using a mixture of concentrated nitric acid (HNO_3) and perchloric acid ($HClO_4$) in a 4:1 ratio following standard wet-digestion procedures (Gent *et al.*, 2011). Samples were heated gradually until a clear solution was obtained, indicating complete oxidation of the organic matter. The digested samples were then cooled and diluted to a known volume with deionized water prior to mineral analysis.

Statistical Analysis

Data were analyzed using two-way analysis of variance (ANOVA) to evaluate the effects of season, district, and their interaction on mineral concentrations. The model included fixed effects for season, district, and the season × district interaction. Post-hoc pairwise comparisons were performed using Tukey's Honest Significant Difference (HSD) test to identify significant differences between group means. Statistical analyses were conducted using SPSS version 26.0, and significance was determined at $p < 0.05$ (Field, 2013).

2.8 Ethical Clearance

This study was conducted in accordance with ethical standards and procedures for animal research. Ethical clearance was obtained from the Addis Ababa University of Debrezeit Veterinary and Agricultural College (protocol approval number VM/ERC/07/11/018). Verbal consent was also obtained from all participating dairy farm owners and personnel prior to sample collection. No invasive procedures were performed on the animals; all sampling and handling were carried out with minimal stress to ensure animal welfare.

3 Results and Discussions

3.1 Prevalence Patterns by Risk Factors

The overall prevalence of mineral deficiency was 60% in cows from Bishoftu and 72.5% in cows from Wonji Shewa (Table 1). A significant association was observed between location and mineral status ($\chi^2 = 10.20$, $p < 0.005$). This difference may reflect variations in soil mineral content, feeding practices, and forage availability. Cows in Wonji Shewa were

more likely to experience deficiencies, possibly due to differences in grazing or mineral supplementation. In a study conducted in the East Shoa Zone, Oromia Region, phosphorus (*P*) deficiency was reported in both lactating and dry cows, which was attributed to low *P* levels in native pastures (Wondewsen *et al*., 2020). This finding supports the notion that regional soil and forage characteristics influence cattle mineral status.

Table 1. Prevalence of the condition under study by origin, physiological state, body condition and season

Variables	Categories	No sampled	Positive	Prevalence %	χ^2	P-value
Origin	Bisheftu	40	24	60.00	10.20	0.005
	Wonji Shewa	40	29	72.50		
	Total	80	53	66.30		
Physiological state	Dry cow	40	22	55.00	25.48	0.000
	Lactating cow	40	31	77.50		
	Total	80	53	25.48		
Body condition	Good	43	26	60.50	0.00	0.517
	Medium	21	14	66.70		
	Poor	16	13	81.30		
	Total	80	53	66.30		
Season	Wet	40	20	50.00	2.48	0.000
	Dry	40	33	82.50		
	Total	80	53	66.30		

3.2 Physiological State

Lactating cows had a much higher rate of mineral deficiency (77.5%) compared to dry cows (55%). As a result, there was a strong association between physiological state and mineral status ($\chi^2 = 25.48$, $p < 0.001$). Notably, this finding highlight that lactating cows are more prone to mineral depletion. This suggests that the increased metabolic demands for milk production place additional stress on mineral reserves. Consistent with this, Kumar *et al*. (2018) also reported higher requirements for calcium, phosphorus, and magnesium during lactation. However, Mummed *et al*. (2023) found sufficient magnesium levels in both lactating and dry cows in the same area. This indicates that not all minerals are equally influenced by physiological state, and some may be buffered despite increased demands.

Mineral deficiency was most common in cows with poor body condition (81.3%), followed by those with medium (66.7%) and good condition (60.5%). Overall, this pattern suggests greater nutritional stress in animals with low body reserves. However, the differences were not statistically significant ($\chi^2 = 0.00$, $p > 0.05$). This implies that, although body condition may influence mineral status, the effect may not always be detectable statistically. Similarly, Mummed *et al*. (2023) reported irregular mineral levels in cows with poor body condition in nutrient-deficient environments. In contrast to earlier studies that found a strong association between body condition and mineral balance, the present findings did not confirm such a relationship, possibly due to variations in local feed quality and herd management practices.

3.3 Seasonal Variation

Season had a significant impact on mineral deficiency ($\chi^2 = 29.45$, $p < 0.001$), with a higher prevalence in the dry season (82.5%) than in the wet season (66.3%). This suggests reduced pasture quality and lower forage mineral content during dry periods. Consistent with previous studies, Ndoti *et al*. (2020) reported severe mineral and protein deficiencies during the dry season in Senegal, particularly

in lactating cows. In addition, Kumar *et al*. (2018) observed marked seasonal variation in feed mineral content in Ethiopian dairy systems. Unlike earlier research that focused mainly on protein, the present study emphasizes the need for seasonal mineral supplementation.

3.4 Dairy Owners' Knowledge of Mineral Deficiency

This study assessed the seasonal occurrence of mineral deficiencies and related symptoms in dairy cows in the two locations —Bishoftu and Wonji Shewa (Table 2).

Table 2. Dairy owners' knowledge of seasonal occurrence of mineral deficiency in the study area

Variables	Bishoftu (n = 40)	Wonji Shewa (n = 40)	Overall Total (N = 80)	Chi-Square (χ^2)
Dairy owners' knowledge about Mineral Deficiency (%)				
Yes	24 (60.0)	26 (65.0)	50 (62.5)	0.23, p > 0.05
No	16 (40.0)	14 (35.0)	30 (37.5)	
Dairy owners' opinion about seasonal occurrence of Mineral Deficiency (%)				
Dry Season	28 (70.0)	29 (72.5)	57 (71.3)	0.04, p > 0.05
Wet Season	1 (2.5)	1 (2.5)	2 (2.5)	
Both Seasons	2 (5.0)	1 (2.5)	3 (3.8)	
No Idea	9 (22.5)	9 (22.5)	18 (22.5)	
Total	40(100)	40(100)	80(100)	

Mineral deficiencies were most common during the dry season; therefore, fodder scarcity likely reduced mineral availability, particularly for lactating cows (Ndoti *et al*., 2020). This highlights the role of seasonal changes in driving mineral imbalances through reduced pasture quality and forage intake. Dairy farmers in both areas showed comparable awareness of mineral deficiencies, consistent with Alemayehu *et al*. (2012), who reported that access to extension services —rather than location —shapes knowledge. In contrast, studies from northern Ethiopia found that cultural beliefs strongly influenced awareness (Gizaw *et al*., 2019). Hence, the relatively uniform knowledge observed in the present study is likely associated with stronger veterinary outreach in these urban-adjacent regions.

3.5 Recognition of Mineral Deficiency Symptoms

The study assessed dairy owners' recognition of clinical signs of mineral deficiency (Table 3) and revealed overall low awareness, with no significant difference between Bishoftu and Wonji Shewa (χ^2 = 0.16, p > 0.05). Milk fever was the most recognized symptom, reported by 22.5% of farmers in Bishoftu and 20.0% in Wonji Shewa, likely reflecting its acute and visible presentation (McDowell, 2003). Notably, awareness of subclinical signs was poor; farmers mostly identified severe symptoms such as recumbency or milk loss, consistent with Girma (2016). However, unlike Suttle (2010), who reported that emaciation, lameness, and soil licking were relatively commonly recognized, the present study found significantly lower acknowledgment of soil licking (3.8%) and emaciation (1.3%).

Knowledge of reproductive signs (e.g., silent heat and delayed puberty) was also limited, suggesting that subclinical effects on fertility often go unnoticed, which may contribute to reproductive inefficiencies. The present study, consistent with Alemayehu *et al*. (2012), indicates that mineral deficiencies are frequently overlooked because their symptoms overlap with other conditions. In contrast to studies from highland areas, where lameness and emaciation were commonly reported (Tadesse *et al*., 2017), this study observed fewer such cases, possibly due to differences in management practices or breed susceptibility.

Table 3. Recognition of mineral deficiency symptoms among dairy owners in the study area

Variables	Bishoftu (n = 40)	Wonji Shewa (n = 40)	Overall Total (N = 80)	
Dairy owners observation about mineral deficiency symptoms (%)				
Loss of Hair Coat	4 (10.0)	3 (7.5)	7 (8.8)	$\chi^2 = 0.16$, $p > 0.05$
Licking of Human Urine	5 (12.5)	6 (15.0)	11 (13.8)	
Lameness	1 (2.5)	1 (2.5)	2 (2.5)	
Soil Licking	1 (2.5)	2 (5.0)	3 (3.8)	
Delayed Onset of Puberty	0 (0.0)	1 (2.5)	1 (1.3)	
Silent Heat	1 (2.5)	1 (2.5)	2 (2.5)	
Irregular Estrous	1 (2.5)	2 (5.0)	3 (3.8)	
Failure of Estrous	1 (2.5)	1 (2.5)	2 (2.5)	
Long Period of Calving Interval	1 (2.5)	1 (2.5)	2 (2.5)	
Embryonic Death	1 (2.5)	0 (0.0)	1 (1.3)	
Increased Incidence of Dystocia	1 (2.5)	1 (2.5)	2 (2.5)	
Retention of Placenta	1 (2.5)	1 (2.5)	2 (2.5)	
Blindness in Newborns	0 (0.0)	1 (2.5)	1 (1.3)	
Emaciation	0 (0.0)	1 (2.5)	1 (1.3)	
Milk Fever Incidence	9 (22.5)	8 (20.0)	17 (21.3)	
Milk Quality and Quantity Affected	3 (7.5)	3 (7.5)	6 (7.5)	

Notably, this study determined the concentrations of key macro-minerals (*Ca*, *P*, and *Mg* in $g\ L^{-1}$) and micro-minerals (*Cu*, *Fe*, *Zn*, and *Co* in $mg\ L^{-1}$) in lactating and dry cows across wet and dry seasons in the peri-urban dairy regions of Bishoftu and Wonji Shewa in the East Shoa Rift Valley, Ethiopia (Table 4). The results were compared with established critical thresholds for dairy cattle and interpreted in relation to recent literature, considering seasonal, spatial, and physiological variations that may contribute to mineral imbalances and affect dairy health and productivity.

Table 4. Mean concentrations of macro ($g\ L^{-1}$) and micro-minerals ($mg\ L^{-1}$) in the blood serum of lactating and dry dairy cows across dry and wet seasons in the study area

Physiological status	Study area	Macro Minerals ($g\ L^{-1}$)			Micro Minerals ($mg\ L^{-1}$)			
		Ca	*P*	*Mg*	*Cu*	*Fe*	*Zn*	*Co*
Lactating n=40	Wonji shewa	0.085	0.040	0.155	0.590	2.281	1.980^{b}	ND
	Bisheftu	0.080	0.040	0.135	0.675	2.590	1.890^{c}	ND
Mean		0.083	0.040	0.145	0.633	2.436	1.935	
Dry Cow n=40	Wonji shewa	0.090	0.040	0.135	0.521	2.481	2.406^{a}	ND
	Bisheftu	0.085	0.045	0.125	0.641	2.420	2.170^{ab}	ND
Mean		0.086	0.043	0.130	0.581	2.451	2.288	
Overall Mean		0.085	0.042	0.138	0.607	2.443	2.112	
SE		0.002	0.0011	0.006	0.0013	0.5103	0.118	
Interaction effect								
Phy × study area		0.8833	0.7645	0.1827	0.6745	0.0758	0.8369	
Critical level		$< 0.08^{g}$	< 0.04	< 0.012	< 0.65	< 1.0	< 0.6	ND

Values are means per location and season.
"ND" indicates not detected. Superscript letters within columns denote statistically significant differences at $p < 0.05$.
Critical levels indicate the minimum concentrations required to avoid deficiency according to McDowell (2003) and Puls (1994).

3.6 Blood Serum Mineral Concentrations

Plasma calcium concentrations ranged from 0.075g L^{-1} in Wonji Shewa during the dry season to 0.085g L^{-1} in Bishoftu, with an overall mean of 0.080g L^{-1} (Table 5). A significant location $\times$ season interaction ($p < 0.05$) indicated that both spatial and seasonal factors influenced calcium status. Calcium levels in Bishoftu were slightly above the critical threshold of 0.08g L^{-1}, suggesting adequate status, consistent with Tadesse *et al.* (2017), who reported similar values (0.081g L^{-1}) in well-managed herds in Ethiopia's central highlands. In contrast to Nzioka *et al.* (2020), who reported stable calcium concentrations throughout the year due to consistent supplementation in southern Ethiopia, this study showed seasonal declines, particularly in Wonji Shewa during the dry season, suggesting uneven supplementation practices across regions.

In Kenya and Uganda, calcium concentrations in herds fed fortified diets were reported to exceed 0.09g L^{-1} (Fall-Toure *et al.*, 2014). Conversely, calcium levels in Wonji Shewa during the dry season fell below the threshold, which may be linked to poorer forage quality in lowland, semi-arid areas. Likewise, although Mekonnen *et al.* (2022) reported no seasonal variation in calcium under intensive feeding, the present findings suggest that reliance on natural forage and extensive grazing may predispose cows to marginal deficiencies. These results align with Goff (2008), who associated forage mineral decline with prolonged dry periods in Ethiopian rangelands, and with Chibsa and Gure (2020), who reported that suboptimal calcium during early lactation can impair productivity and reproduction.

Plasma phosphorus concentrations ranged from 0.035 to 0.040g L^{-1}, with a mean of 0.0395g L^{-1}, which was slightly but significantly below the critical threshold of 0.04g L^{-1}", indicating a marginal deficiency. No significant interaction between study area and season was detected ($p > 0.05$), suggesting phosphorus levels were relatively consistent across locations and seasons. Phosphorus is important for energy metabolism, bone development, and reproduction. The borderline deficiency observed —particularly in Bishoftu during the dry season —may be related to reduced forage phosphorus due to declining soil fertility and increased plant maturity. These findings are consistent with Tadesse *et al.* (2023), who reported suboptimal phosphorus in central Ethiopian dairy herds linked to reduced reproductive performance and milk yield, and with Abay *et al.* (2021), who associated soil phosphorus limitations and low-quality forage with mineral imbalances, especially during dry seasons. By comparison, Crnkic and Hodzic (2012) in South Africa reported more severe phosphorus deficiency in cattle grazing phosphorus-depleted pastures; however, the present study found only a marginal shortfall. Similarly, while Orjales *et al.* (2018) reported lower phosphorus in indigenous Zebu cows near parturition in South Africa, the current study showed no significant difference between lactating and dry cows, suggesting a broadly uniform deficiency across physiological states. The lack of seasonal variation may indicate sustained year-round dietary insufficiency regardless of environmental conditions or lactation stage.

Plasma magnesium concentrations ranged from 0.105 to 0.170g L^{-1} (mean: 0.151g L^{-1}), well above the deficiency threshold of 0.012g L^{-1}. No significant season $\times$ location interaction was observed ($p > 0.05$), indicating stable magnesium status across sites and seasons. All values fell within physiologically adequate ranges, suggesting sufficient intake and absorption under current management. These results agree with Suttle (2010), who reported elevated magnesium levels in Rift Valley livestock, likely due to mineral-rich volcanic soils and groundwater. Magnesium also plays a key role in preventing hypomagnesemia (grass tetany) (Musa *et al.*, 2022). Nevertheless, high potassium or nitrogen in lush pastures may reduce magnesium absorption without necessarily lowering plasma concentrations (Suttle, 2010), so continued monitoring—particularly after pasture flushes following rains—is warranted.

Plasma copper concentrations ranged from 0.570 to 0.610mg L^{-1} (mean: 0.588mg L^{-1}), consistently below the critical threshold of 0.65mg L^{-1} for dairy cattle. No significant season $\times$ location interaction was found ($p > 0.05$), indicating uniformly low copper status across sites. Although the deficiency was marginal, these concentrations suggest the possibility of subclinical copper deficiency. Copper is essential for hemoglobin synthesis, immune function, reproduction, and antioxidant defense, and requirements

increase in lactating cows due to greater metabolic activity (Suttle, 2010). The marginal deficiency may be aggravated by mineral antagonism, particularly the elevated iron levels observed in this study, which can impair copper absorption. Similar subclinical copper deficiencies have been reported in central Ethiopia (Orjales *et al*., 2018) and Kenya (Musa *et al*., 2022).

Plasma iron concentrations ranged from 2.281$mg\ L^{-1}$ (dry season, Wonji Shewa) to 3.381$mg\ L^{-1}$ (wet season, Wonji Shewa), with a mean of 2.784$mg\ L^{-1}$, which was well above the critical threshold of 1.0$mg\ L^{-1}$ for dairy cattle. A significant location × season interaction was reported ($p > 0.05$). Although iron toxicity is uncommon in cattle, excessive iron can interfere with the absorption of other trace elements, particularly copper (*Cu*) and zinc (*Zn*). Therefore, this antagonism may have contributed to the marginal copper and zinc deficiencies observed, despite adequate or near-threshold intakes.

Plasma zinc concentrations ranged from 2.041 to 2.280$mg\ L^{-1}$ (mean: 2.163$mg\ L^{-1}$), which was well above the critical threshold of 0.6$mg\ L^{-1}$. No significant interaction between season and location was detected ($p > 0.05$), indicating consistent zinc status across districts and seasons. These adequate levels are encouraging because zinc supports immune function, wound healing, and reproduction. However, even with adequate plasma zinc concentrations, zinc bioavailability may still be reduced by elevated iron and the marginal copper levels observed in this study.

3.7 Seasonal Mineral Concentrations

3.7.1 Macro Mineral Concentrations

Calcium concentrations ranged from 0.075 to 0.08$g\ L^{-1}$, with the highest level in Bishoftu during the dry season (0.085$g\ L^{-1}$) and the lowest in Wonji Shewa (0.075$g\ L^{-1}$). The mean *Ca* level (0.080$g\ L^{-1}$) was slightly but significantly above the critical threshold (< 0.08$g\ L^{-1}$). A significant interaction between location and season ($p > 0.05$; SEM = 0.0037) indicates that calcium availability is influenced by both geographic and seasonal factors. These findings are in line with Mekonnen *et al*. (2022), who highlighted the role of soil type and rainfall in regional mineral variation. However, in contrast to the current study, Crnkic and Hodzic (2021) reported no seasonal fluctuation in plasma calcium, likely due to consistent mineral supplementation.

Phosphorus levels ranged from 0.035 to 0.040$g\ L^{-1}$, with a mean of 0.0395$g\ L^{-1}$, notably just below the critical threshold (< 0.04$g\ L^{-1}$). No significant interaction between season and location was observed ($p > 0.05$; SEM = 0.0012), therefore indicating stable *P* status across sites and seasons. These marginal values are consistent with Garamu (2019). who reported borderline *P* levels in Ethiopian smallholder dairy systems. The lack of seasonal variation contrasts with Suttle, N.F. (2010), who noted higher *P* levels during the wet season due to improved forage quality.

3.7.2 Micro Mineral Concentrations

Copper concentrations ranged from 0.570 to 0.610$mg\ L^{-1}$, just below the critical threshold (< 0.65$mg\ L^{-1}$), with no significant location-season interaction ($p > 0.05$; SEM = 0.0025). Although marginally deficient, these values align with those reported by NRC (2001) for tropical grazing systems. The absence of seasonal variation mirrors findings from Miller *et al*. (2021), therefore suggesting stable *Cu* levels in mixed feeding systems.

Iron concentrations showed the most pronounced variability. Levels ranged from 2.281$mg\ L^{-1}$ at Wanji Shewa (dry season) to 3.381$mg\ L^{-1}$ at Wanji Shewa (wet season), with an overall mean of 2.784$mg\ L^{-1}$. The interaction between season and location was significant ($p < 0.05$), indicating that *Fe* content was strongly influenced by both factors. Importantly, all values were well above the critical requirement of 1.0$mg\ L^{-1}$, highlighting that drinking water is a major *Fe* source in both study areas. However, excessive *Fe* can impair *Cu* and *Zn* absorption (Crnkic & Hodzic, 2012), potentially leading to secondary deficiencies even when absolute dietary levels appear adequate. The elevated wet-season values may reflect soil erosion and runoff during rains, as previously reported in volcanic soil areas of East Africa (Musa *et al*., 2022).

Zinc concentrations varied between 2.041 – 2.280$mg\ L^{-1}$, with an overall mean of 2.163$mg\ L^{-1}$. No significant differences were observed across seasons or

study areas ($p > 0.05$). All values were well above the critical requirement of $0.6 mg\ L^{-1}$, indicating that water makes a meaningful contribution to *Zn* nutrition. Adequate *Zn* is crucial for reproductive performance and immune competence (Garamu, 2019). However, given the high *Fe* levels in these samples, *Zn* bioavailability may be partially reduced through mineral antagonism (Miller *et al*., 2021).

Table 5. Mean concentrations of macro-minerals ($g\ L^{-1}$) and micro-minerals ($mg\ L^{-1}$) in the blood serum of lactating and dry dairy cows across dry and wet seasons in study area

Study area	Season	Macro Minerals ($g\ L^{-1}$)			Micro Minerals ($mg\ L^{-1}$)			
		Ca	*P*	*Mg*	*Cu*	*Fe*	*Zn*	*Co*
Bishoftu	Dry	0.085^{a}	0.035	0.170	0.570	2.681^{ab}	2.041	ND
	Wet	0.080^{ab}	0.040	0.105	0.580	2.690^{ab}	2.190	ND
Mean		0.083	0.041	0.138	0.575	2.686^{ab}	2.116	
Wanji Shewa	Dry	0.075^{b}	0.040	0.170	0.610	2.281^{b}	2.140	ND
	Wet	0.080^{ab}	0.035	0.155	0.592	3.381^{a}	2.280	ND
Mean		0.076	0.038	0.163	0.601	2.883	2.210	
Overall Mean		0.080	0.0395	0.151	0.588	2.784	2.163	
SE		0.0037	0.0012	0.0036	0.0025	0.1123	0.1301	
Interaction effect								
Study area × Season		0.0182	0.6974	0.0836	0.914	0.0199	0.2387	
Critical level		$< 0.08^{g}$	< 0.04	< 0.012	< 0.65	< 1.0	< 0.6	ND

Values are presented as means per location and season.
Superscripts indicate statistically significant differences within minerals ($p < 0.05$). "ND" denotes minerals not detected.
Critical levels represent deficiency thresholds based on Puls (1994) and McDowell (2003).

Table 6 shows that roughages had the highest calcium (*Ca*) content, averaging 0.63% during the wet season, which was significantly greater than concentrates (0.36%) and non-conventional feeds (0.54%) ($p < 0.0001$). Both feed type and the feed × season interaction significantly affected *Ca* levels ($p < 0.0001$), whereas season alone had no significant effect ($p > 0.05$). Roughage *Ca* ranged from 0.61% (dry season) to 0.63% (wet season), while concentrates and non-conventional feeds had lower, relatively stable values.

These results align with previous studies indicating that green and leguminous forages generally contain more *Ca*, likely because their deeper root systems access mineral-rich soil (Suttle, 2010; Gizaw *et al*., 2019). Overall, roughages appear to be the main contributors to dietary calcium in this production system, whereas diets relying primarily on concentrates or non-conventional feeds may increase the risk of marginal *Ca* deficiency.

The seasonal increase in roughage *Ca* corresponds with higher nutrient uptake during wetter periods, as reported in the Ethiopian highlands by (Kumar, 2018; Ndoti *et al*., 2020). Unlike studies that attribute *Ca* variation primarily to season alone, the present study emphasizes the importance of the feed × season interaction, indicating that mineral uptake is jointly influenced by both factors. The lower *Ca* concentration in concentrates highlights the need for dietary supplementation, particularly for lactating cows, to reduce the risk of hypocalcaemia. Non-conventional feeds contained moderate *Ca* levels; however, their variability may limit their reliability as a consistent *Ca* source. Overall, these findings are consistent with existing literature, while underscoring site-specific and seasonal dynamics in feed mineral composition.

Table 6. Seasonal variations in macro- and micro-mineral concentrations (mean ± SEM) in different feed types (roughages, concentrates, and non-conventional feeds) sampled in study area

Feed type	Season	Macro minerals (%)			Micro minerals ($mg\ kg^{-1}$)			
		Ca	*P*	*Mg*	*Cu*	*Fe*	*Zn*	*Co*
Roughage n=30	Dry	0.61^{a}	0.50^{ab}	0.86^{a}	8.47	103.25	32.46^{b}	ND
	Wet	0.63^{a}	0.64^{b}	1.09^{a}	8.49	115.26	33.41^{b}	ND
Concentration n=36	Dry	0.35^{b}	0.59^{ab}	0.45^{b}	7.71	106.24	38.38^{ab}	ND
	Wet	0.38^{b}	0.64^{b}	0.58^{b}	8.01	122.15	41.46^{a}	ND
Non-conventional n=12	Dry	0.50^{ab}	0.65^{b}	0.34^{b}	7.54	95.27	30.15^{ab}	ND
	Wet	0.59^{a}	0.89^{a}	0.54^{b}	7.68	106.13	33.00^{b}	ND
Overall mean (n= 78)		0.51	0.65	0.807	7.98	108.05	34.81	
SEM		0.034	0.033	0.047	0.264	6.543	1.139	
P-value								
Feed type		0.0001	0.0003	0.0001	0.079	0.408	0.0001	
Season		0.702	0.754	0.0009	0.084	0.094	0.857	
Feed × Season		0.0001	0.0001	0.0001	0.124	0.357	0.0001	

Different superscripts (a, b) within columns indicate significant differences ($p < 0.05$) between means; ND = Not Detected.

Phosphorus (*P*) concentrations were highest in non-conventional feeds (0.89%) during the wet season, followed by roughages and concentrates, both at approximately 0.64%. *P* varied significantly with feed type ($p < 0.001$) and the feed × season interaction ($p < 0.001$), but not with season alone ($p > 0.05$; SEM = 0.033). These results are consistent with findings from East Shoa (Ndoti *et al.*, 2020), where non-conventional feeds —such as brewery residues and agro-industrial by-products —contained elevated *P* due to mineral enrichment during processing. Although roughages and concentrates showed moderate and similar *P* levels, the higher *P* content in non-conventional feeds during the wet season highlights their potential value as *P* supplements. The observed seasonal increase also agrees with (Ndoti *et al.*, 2020), who reported enhanced plant phosphorus uptake under moist and biologically active conditions. While some studies report larger seasonal variation in traditional forages, this study demonstrates that both feed type and seasonal dynamics strongly shape *P* availability (Underwood & Suttle, 1999). Consequently, incorporating non-conventional feeds may help mitigate *P* deficiencies; however, supplementation should be balanced to avoid mineral imbalances.

Magnesium (*Mg*) concentrations were highest in roughages (1.09%) during the wet season, followed by concentrates (0.58%) and non-conventional feeds (0.54%). *Mg* levels were significantly affected by feed type ($p < 0.0001$), season ($p < 0.001$), and their interaction ($p < 0.0001$; SEM = 0.047). Roughages consistently contributed more *Mg*, particularly during the wet season, likely due to improved soil solubility and greater plant uptake. These findings are consistent with (Field, 2013; Gizaw *et al.*, 2019), who reported higher *Mg* in leguminous and green forages, especially during rainy periods (Ndoti *et al.*, 2020). Concentrates, which are largely grain- and oilseed meal-based, are typically lower in *Mg*, whereas non-conventional feeds provide moderate *Mg* depending on their composition. Therefore, even with lower *Mg* than roughages, non-conventional feeds can still support adequate intake when used within balanced rations. Magnesium is essential for neuromuscular function, enzyme activity, and calcium regulation; thus, *Mg* deficiency can contribute to grass tetany, particularly in early spring or during dry seasons (Suttle, 2010). The high *Mg* levels in roughages during the wet season suggest a lower risk, but supplementation may still be necessary during dry periods or under high-potassium conditions that inhibit *Mg* absorption.

3.8 Micro-Minerals in Dairy Feeds

Copper (*Cu*) concentrations were highest in roughages ($8.49 mg\ kg^{-1}$) during the wet season, followed by concentrates ($8.01 mg\ kg^{-1}$) and non-conventional feeds ($7.68 mg\ kg^{-1}$). Differences among feed types ($p < 0.001$), across seasons ($p = 0.084$), and for the feed × season interaction ($p > 0.05$) were not statistically significant (SEM = 0.264). Overall, *Cu* levels remained close to the minimum dietary requirement of $8 – 10 mg\ kg^{-1}$ for ruminants. The slightly higher *Cu* in roughages is consistent with findings from mineral-rich or volcanic soils (Field, 2013; Gizaw *et al*., 2019). Seasonal increases may also reflect greater microbial activity and improved solubility in moist soils (Ndoti *et al*., 2020). However, *Cu* bioavailability can be reduced by antagonistic minerals, particularly iron, which was also elevated in these feeds and is known to interfere with *Cu* absorption (Crnkic & Hodzic, 2012). Although differences were statistically non-significant, the marginal *Cu* levels in concentrates and non-conventional feeds agree with previous reports from the Ethiopian highlands (Chibsa & Gure, 2020; Tadesse *et al*., 2023).

Iron (*Fe*) concentrations peaked in concentrates ($122.15 mg\ kg^{-1}$) during the wet season, followed by roughages ($115.26 mg\ kg^{-1}$) and non-conventional feeds ($106.13 mg\ kg^{-1}$). Feed type ($p > 0.05$), season ($p < 0.05$), and their interaction ($p > 0.05$) showed no significant overall effects (SEM = 6.543). Nonetheless, all feeds exceeded the ruminant *Fe* requirement of $50 mg\ kg^{-1}$ DM (Abule *et al*., 2021). Elevated *Fe* in concentrates may be attributed to the presence of iron-rich ingredients (e.g., wheat bran or molasses) or potential contamination during processing (Hu *et al*., 2021). In addition, volcanic soils and mineral-rich water can contribute to elevated *Fe* in forages (Orjales *et al*., 2018). While *Fe* toxicity is uncommon, excessive *Fe* can inhibit the absorption of *Cu* and *Zn*, thereby increasing the risk of secondary deficiencies (Crnkic & Hodzic, 2012). The wet-season increase in *Fe* likely reflects erosion and runoff processes, as well as the use of mineral-laden water during feed mixing, as also reported by Orjales *et al*. (2021).

Zinc (*Zn*) concentrations were highest in concentrates ($41.46 mg\ kg^{-1}$), followed by roughages ($33.41 mg\ kg^{-1}$) and non-conventional feeds ($33.00 mg\ kg^{-1}$). *Zn* varied significantly with feed type ($p < 0.0001$) and the feed × season interaction ($p < 0.0001$), but not with season alone ($p > 0.05$; SEM = 1.139). All feeds exceeded the critical *Zn* requirement ($30 – 40 mg\ kg^{-1}$ DM) (Abule *et al*., 2021). Higher *Zn* in concentrates likely reflects the inclusion of mineral premixes and more consistent diet formulation (NRC, 2001). The seasonal rise in *Zn* —particularly in concentrates —may be related to improved feed quality and supplementation during the wet season. Despite overall adequate *Zn* concentrations, bioavailability may be reduced by elevated dietary *Fe* and *Ca*, both of which were present in excess (Crnkic & Hodzic, 2012). Therefore, *Zn* uptake may be influenced by seasonal factors, plant maturity, and soil characteristics such as p*H* and organic matter (Miller *et al*., 2021). Even in forage-based systems, seasonal *Zn* limitations may occur; hence, routine mineral profiling and targeted supplementation are recommended to support reproductive and immune functions.

4 Conclusion and Recommendations

This study demonstrated variations in the mineral status of dairy cows across locations, seasons, and physiological states in Bishoftu and Wonji Shewa, Ethiopia. The results showed that mineral deficiencies were more prevalent in Wonji Shewa, particularly during the dry season and among lactating cows. These findings indicate that location, season, and physiological status are key risk factors influencing mineral balance. Among the minerals assessed, calcium and phosphorus were marginally deficient, especially during the dry season. In contrast, copper concentrations remained consistently below critical levels across the study areas. Magnesium and zinc were generally within adequate ranges. Cobalt was not detected in most samples, whereas iron concentrations exceeded critical requirements, suggesting a potential risk of antagonistic interactions that may reduce the bioavailability of copper and zinc. The study also showed that seasonal changes in feed mineral composition contribute to variations in animal mineral status, emphasizing the importance of feed quality for sustaining optimal productivity and animal health. These mineral imbalances may negatively affect milk production, re-

productive performance, immune competence, and overall herd productivity, particularly in lactating and high-producing dairy cows.

Based on these findings, location- and season-specific mineral supplementation programs are recommended, with particular emphasis on calcium, phosphorus, copper, and cobalt. Routine monitoring of mineral status in both dairy cows and feeds should be strengthened to enable early detection and correction of deficiencies. In addition, dairy producers should receive support through targeted extension services and farmer training programs focused on mineral nutrition, appropriate supplementation practices, and the recognition of health problems associated with mineral deficiencies. Such interventions will contribute to improved dairy productivity, better animal health, and greater sustainability of dairy production systems in the Ethiopian Rift Valley.

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Data availability statement

The data that support the findings of this study are available from the corresponding author on reasonable request.

Declaration of interests' statement

The authors declare no conflict of interest

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