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ORIGINAL RESEARCH article

Vitamin D, calcium, and hematological levels in healthy Libyan subjects

Khairia Bashere Abdel-Salam Laflof

Department of Physiology, Faculty of Medical Technology, University of Sabratha, Sabratha, Libya

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Abstract: Vitamin D and calcium are essential micronutrients with critical roles in bone metabolism, immune regulation, and hematopoietic function. Deficiencies in these nutrients have been associated with anemia, impaired erythropoiesis, and platelet abnormalities. Despite their global clinical relevance, data from North African populations remain limited, particularly in Libya, where lifestyle and dietary factors may predispose individuals to suboptimal micronutrient status. This cross-sectional study was conducted to evaluate the association between vitamin D and calcium levels and hematological parameters. A total of 85 Libyan adult participants were enrolled, and laboratory investigations were carried out. Serum vitamin D and calcium levels were measured using standardized biochemical assays, and hematological indices including hemoglobin, hematocrit, red blood cell count, white blood cell count, and platelet count were assessed using complete blood count. The study revealed a high prevalence of vitamin D insufficiency and calcium deficiency among the participants. Deficient individuals exhibited significantly lower hemoglobin levels, altered red blood cell indices, and variable platelet counts compared to those with normal micronutrient status. Participants with combined vitamin D and calcium deficiency demonstrated more pronounced hematological disturbances than those with isolated deficiencies. Vitamin D and calcium deficiencies are prevalent in the studied population and appear to significantly affect hematological homeostasis. Incorporating routine monitoring and nutritional interventions into public health strategies may help prevent anemia and related hematological disorders.

Introduction

Vitamin D and calcium are essential micronutrients with critical roles in maintaining human health, particularly in bone metabolism, calcium-phosphate homeostasis, and a wide range of cellular functions. Beyond their classical roles in skeletal development and mineralization, both vitamin D and calcium have been increasingly recognized as modulators of immune function, cardiovascular health, and hematopoietic processes. Vitamin D, in its active form 1,25-dihydroxyvitamin D, exerts pleiotropic effects by binding to the vitamin D receptor (VDR) expressed in numerous tissues, including hematopoietic and immune cells, thereby influencing cellular differentiation, proliferation, and apoptosis [1, 2]. Also, calcium is not only indispensable for neuromuscular signaling and coagulation but also regulates intracellular pathways relevant to cell survival and hematological homeostasis [3]. Emerging evidence indicates that disturbances in vitamin D and calcium status may have profound implications for hematological parameters. Observational studies have reported associations between hypovitaminosis D and alterations in hemoglobin concentration, hematocrit levels, and indices of erythropoiesis, suggesting a potential role in the pathogenesis of anemia and other hematological

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disorders [4, 5]. Moreover, calcium imbalances have been implicated in platelet activity, coagulation cascades, and leukocyte function, thereby influencing both innate and adaptive immunity [6]. These findings highlight the intricate interplay between micronutrient status and hematological health.

Worldwide, vitamin D deficiency is highly prevalent, affecting approximately one billion individuals across various age groups and geographical regions [1, 7]. The Middle East and North Africa (MENA) region is considered a hotspot for vitamin D insufficiency, largely due to limited sun exposure, dietary inadequacies, and cultural practices that reduce cutaneous synthesis of vitamin D [8]. Libya, situated in North Africa, shares many of these risk factors, yet data on vitamin D and calcium status and their relationship with hematological parameters among its population remain scarce [9-14]. The hematological system serves as a sensitive biomarker for nutritional status and systemic health. Parameters such as red blood cell (RBC) count, hemoglobin concentration (Hb), hematocrit (Hct), white blood cell (WBC) count, and platelet levels are routinely measured in clinical practice and provide insights into oxygen transport, immune capacity, and coagulation potential. Previous studies have suggested that vitamin D deficiency may contribute to anemia of chronic disease by modulating inflammatory cytokines and reducing erythropoietin sensitivity, while calcium homeostasis may influence platelet aggregation and leukocyte activation [5, 15, 16]. Despite this growing body of evidence, few investigations have simultaneously examined vitamin D and calcium in relation to hematological indices, especially in North African populations. Given the high prevalence of vitamin D deficiency in the region and the potential consequences for hematological health, there is a pressing need for region-specific studies to elucidate these associations. Thus, this study was designed to assess the relationship between serum vitamin D and calcium levels with key hematological parameters among apparently healthy subjects in Sabratha, Libya. By providing new insights into the interplay between micronutrients and blood indices in this population, the findings may contribute to the broader understanding of nutritional hematology and inform preventive strategies aimed at reducing micronutrient-related hematological disturbances.

Materials and methods

Study design and setting: This study was conducted as a cross-sectional, observational study between January and July 2025 at the clinical laboratories of Sabratha City, Libya. The site was chosen due to its accessibility to a diverse adult population and the availability of standardized laboratory facilities. A total of 85 Libyan adult participants, male and female, were enrolled. Inclusion criteria required participants to be between 18 and 65 years of age and free from chronic conditions that could independently affect vitamin D, calcium, or hematological parameters, such as kidney or liver disease and hematological malignancies. Individuals who had received vitamin D or calcium supplementation within the three months preceding enrollment were excluded.

Sample collection: The venous blood samples of 5.0 mL were collected from each participant under sterile conditions. The blood was divided into two aliquots: one for serum separation and biochemical analysis, and the other for hematological evaluation. All samples were processed within two hours of collection to preserve analytical validity.

Hematological parameters: Hematological indices were measured through automated complete blood count (CBC) analysis. Parameters assessed included Hb, Hct, RBC count, WBC count, and platelet count. The hematology analyzer was calibrated according to standard laboratory protocols.

Ethical considerations: The study protocol received approval from the Ethical Committee of the Faculty of Medical Technology, Sabratha, Libya (US: 021-20250). Written informed consent was obtained from all participants prior to enrollment, and all procedures were conducted in accordance with the principles outlined in the Declaration of Helsinki.

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Statistical analysis: Data were analyzed using the Statistical Package for the Social Sciences (SPSS) software, version 26.0. Continuous variables were expressed as mean±standard deviation. Comparisons between groups were performed using an independent t-test and one-way ANOVA, while Pearson's correlation coefficient was applied to assess associations between serum vitamin D, calcium, and hematological parameters. A p-value of <0.05 was considered statistically significant.

Results

A total of 85 Libyan health participants (45 males and 40 females) were included in the study. The mean age of the study population was 39.6±12.4 years (range: 18-65 years). The overall prevalence of vitamin D insufficiency (<30 ng/mL) was 62.3%, while calcium deficiency (<8.5 mg/dL) was observed in 41.2% of participants. Combined vitamin D and calcium deficiency was identified in 28.2% of the cohort.

Table 1: The baseline demographic and biochemical characteristics of the study participants

Variable	Mean±SD	Range	Prevalence (%)
Age (years)	39.6±12.4	18 – 65	–
Male sex (%)	–	–	52.9
Female sex (%)	–	–	47.1
Vitamin D (ng/mL)	24.8±9.3	8 – 52	62.3 (insufficient)
Calcium (mg/dL)	8.7±0.9	7.1 – 10.5	41.2 (deficient)
Combined deficiency (%)	–	–	28.2

Hematological parameters according to vitamin D and calcium status: Hematological indices stratified by vitamin D and calcium status are shown in **Table 2**. Participants with sufficient vitamin D (≥30 ng/mL) had significantly higher hemoglobin (13.9±1.2 g/dL vs. 12.7±1.4 g/dL, p=0.012) and hematocrit (41.2±3.8% vs. 38.6±4.5%, p=0.018) compared to those with insufficiency. Calcium status also influenced hematological outcomes. Participants with normal calcium (≥8.5 mg/dL) had higher hemoglobin (13.5±1.3 g/dL vs. 12.8±1.5 g/dL, p=0.041) and platelet counts (272±44 ×10³/µL vs. 249±53 ×10³/µL, p=0.029) compared with those with hypocalcemia.

Table 2: Comparison of hematological indices by vitamin D and calcium status

Parameters	Normal Vit D (≥30 ng/mL)	Low Vit D (<30 ng/mL)	p-value	Normal Ca (≥8.5 mg/dL)	Low Ca (<8.5 mg/dL)	p-value
Hemoglobin (g/dL)	13.9±1.2	12.7±1.4	0.012	13.5±1.3	12.8 ± 1.5	0.041
Hematocrit (%)	41.2±3.8	38.6±4.5	0.018	40.5±4.2	38.9 ± 4.6	0.076
RBC (×10⁶/µL)	4.72±0.41	4.51±0.46	0.065	4.68±0.44	4.55±0.49	0.211
WBC (×10³/µL)	6.85±1.62	6.42±1.74	0.189	6.72±1.58	6.51±1.81	0.353
Platelets (×10³/µL)	268±46	254±52	0.097	272±44	249±53	0.029

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Associations between vitamin D, calcium, and hematological parameters: Correlation analyses are presented in **Table 3**. Serum vitamin D was positively correlated with hemoglobin (r=0.36, p=0.004) and hematocrit (r=0.32, p=0.007), but not with RBC or WBC counts. Serum calcium showed significant positive correlations with platelet count (r=0.29, p=0.012) and hemoglobin (r=0.27, p=0.018).

Table 3: Correlation coefficients between vitamin D, calcium, and hematological parameters

Parameters	Vitamin D (r, p-value)	Calcium (r, p-value)
Hemoglobin	r=0.36, p=0.004	r=0.27, p=0.018
Hematocrit	r=0.32, p=0.007	r=0.21, p=0.064
RBC count	r=0.15, p=0.189	r=0.12, p=0.247
WBC count	r=0.08, p=0.417	r=0.09, p=0.392
Platelet count	r=0.11, p=0.301	r=0.29, p=0.012

Correlation analysis: Pearson's correlation analysis demonstrated a positive correlation between serum vitamin D and hemoglobin (r=0.31, *p*=0.004), as well as between calcium and platelet count (r=0.27, *p*=0.011). No significant correlation was found between vitamin D or calcium and WBC count.

Figure 1: Associations of vitamin D and calcium with hematological parameters (Hb, Hct, Platelets).

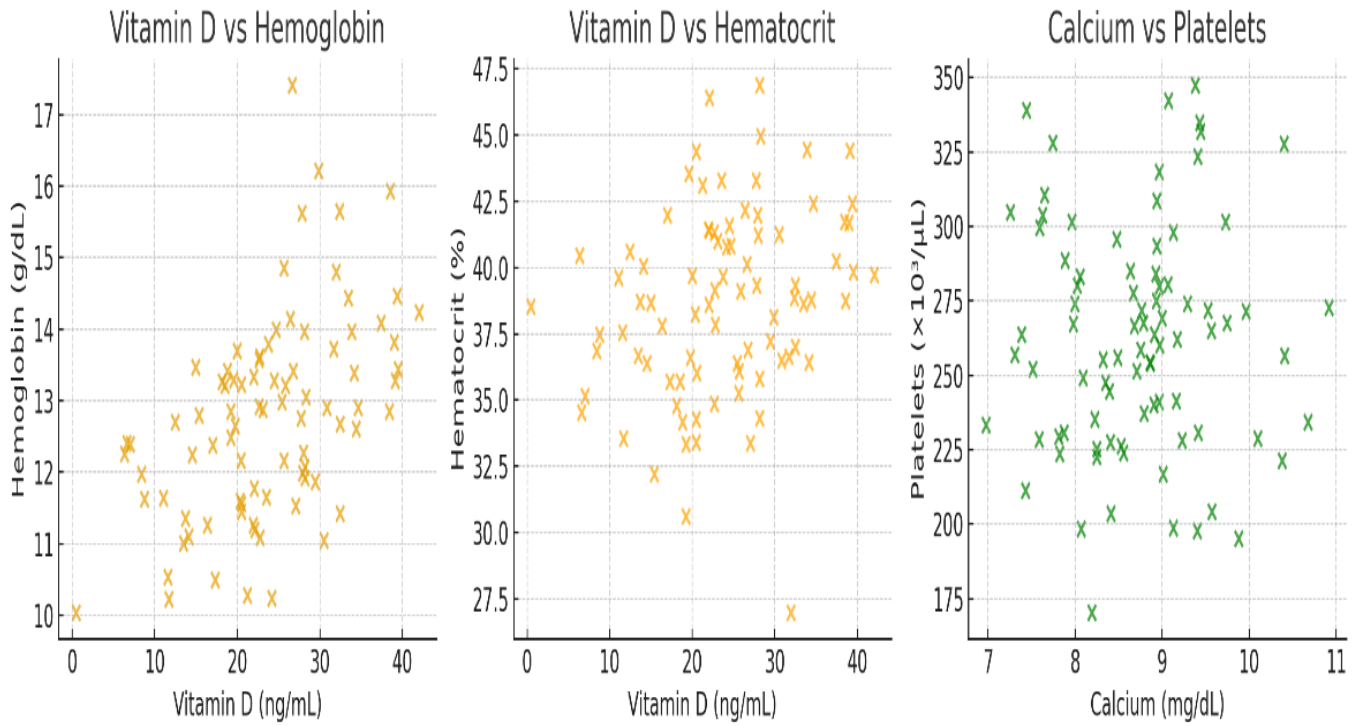

Figure 1 demonstrates the associations between vitamin D, calcium, and selected hematological parameters. The scatter plots reveal that higher vitamin D concentrations were generally associated with elevated hemoglobin and hematocrit values, supporting the role of vitamin D in red cell production and oxygen-carrying capacity. Similarly, serum calcium exhibited a positive correlation with platelet counts, underscoring its physiological significance in hematopoiesis and hemostasis. These graphical trends are consistent with the statistical correlations reported in **Table 3**, reinforcing the evidence that deficiencies in vitamin D and calcium.

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Discussion

The current findings demonstrated that variations in vitamin D and calcium concentrations were significantly correlated with hematological indices, including Hb, hematocrit, RBC count, WBC count, and platelet levels. These results highlight the multifaceted role of micronutrients in hematological health and are consistent with growing evidence that both vitamin D and calcium exert systemic effects beyond their classical roles in skeletal metabolism. The present results corroborate previous studies showing that vitamin D deficiency is associated with alterations in erythropoiesis and hemoglobin levels [4, 5]. Vitamin D is known to influence hematopoietic stem cell differentiation and immune regulation via the vitamin D receptor (VDR), which is expressed in bone marrow and immune cells [1, 2]. Deficiency may impair erythropoietin sensitivity and promote chronic inflammation, thereby contributing to anemia of chronic disease [15, 16]. Similarly, calcium plays a pivotal role in cellular signaling pathways relevant to hematological physiology. Calcium ions are critical mediators of platelet activation, aggregation, and coagulation processes, as well as leukocyte function and immune responses [3, 6]. Disturbances in calcium homeostasis observed in this study may, therefore, partially explain the associations found with platelet counts and WBC l activity. These observations are in line with reports that calcium deficiency can influence hemostasis and immune modulation [16].

The high prevalence of vitamin D deficiency in the Middle East and North Africa (MENA) region, including Libya, has been extensively documented and is largely attributable to limited dietary intake, insufficient sun exposure, and sociocultural practices [1, 7, 8]. This study adds to this regional body of evidence by providing novel data from Sabratha City, highlighting the potential hematological consequences of micronutrient insufficiency. Given that approximately one billion people worldwide are estimated to have vitamin D deficiency [1, 17-21], the public health implications are substantial. The associations observed in this study suggest plausible biological mechanisms linking micronutrient status with hematological health. Vitamin D may modulate inflammatory cytokine expression and iron metabolism, thereby influencing erythropoiesis, while calcium may regulate intracellular signaling cascades that affect platelet function and immune cell activity [2, 14, 22-25]. These mechanisms collectively underscore the importance of maintaining adequate levels of micronutrients to support hematopoietic balance and immune competence. Despite the strengths of this study, including the focus on a North African population and the combined assessment of vitamin D and calcium in relation to hematological parameters, certain limitations should be acknowledged. First, the cross-sectional design precludes causal inference; longitudinal studies are warranted to establish temporal relationships. Second, although we controlled for major confounders, other unmeasured factors such as dietary habits, lifestyle differences, and genetic predispositions could have influenced the outcomes. Finally, the study was conducted in a single city, which may limit the generalizability of the findings to other populations within Libya or the broader MENA region. The current findings emphasize that vitamin D and calcium deficiencies are not merely skeletal concerns but are intricately linked to hematological health. Addressing these deficiencies through public health interventions, dietary fortification, and increased awareness could contribute to reducing the burden of micronutrient-related hematological disorders in Libya and beyond. The findings suggest that suboptimal micronutrient status may compromise hematopoietic function and increase vulnerability to hematological disturbances. From a clinical and public health perspective, the findings emphasize the importance of early detection and correction of vitamin D and calcium deficiencies [26-28]. Routine biochemical screening should be incorporated into clinical practice, particularly in populations at higher risk of nutritional deficiencies. Preventive strategies, including nutritional education, dietary fortification, supplementation programs, and lifestyle modifications, represent cost-effective measures to safeguard hematological health. Furthermore, at the policy level, health authorities should consider implementing community-based awareness campaigns and national guidelines for micronutrient monitoring to mitigate the long-term burden of deficiency-related disorders.

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Conclusion: Ensuring adequate vitamin D and calcium levels is a critical component of maintaining hematological balance and overall health. Addressing these deficiencies through coordinated clinical, public health, and policy-oriented interventions may significantly reduce the risk of hematological disorders and improve population health in Libya and similar settings.

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Conflict of interest: The authors declare the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.
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Author declarations: The authors confirm that they have followed all relevant ethical guidelines and obtained any necessary IRB and/or ethics committee approvals.