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Article

# Association Between Vitamin D Deficiency and Calcium Status in School-Aged Children and Adolescents in Lahore

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## **ABSTRACT**

Background: Vitamin D plays a crucial role in bone health and calcium homeostasis, yet deficiency remains widespread among children in Pakistan despite abundant sunlight, largely due to inadequate dietary intake, limited sun exposure, and socioeconomic barriers. The gap in understanding the extent of vitamin D deficiency and its direct association with calcium status in school-aged children in Lahore necessitates targeted research. Objective: This study aimed to determine the prevalence of vitamin D deficiency among children aged 6-18 years in Lahore, Pakistan, and to assess its association with serum calcium levels as well as related lifestyle and dietary factors. Methods: A cross-sectional descriptive study was conducted at Noble Hospital, Lahore, enrolling 150 children aged 6-18 years through non-probability convenient sampling. Inclusion criteria were healthy children presenting for routine check-ups; exclusions included chronic illness, recent supplementation, or skeletal disorders. Data on sun exposure, dietary habits, and supplement use were collected using structured questionnaires. Serum vitamin D and calcium levels were measured using standardized laboratory methods (CLIA/ELISA and spectrophotometry, respectively). Ethical approval was obtained from The Superior University Lahore in accordance with the Helsinki Declaration. Statistical analyses, including Spearman's rho and non-parametric tests, were performed using SPSS 27.0. Results: Of the participants, 32% were vitamin D deficient (<20 ng/mL), 23.3% insufficient (20-30 ng/mL), and only 44.7% sufficient (>30 ng/mL). A strong positive correlation was observed between vitamin D and serum calcium levels ( $\rho$  = 0.734, p < 0.05). Deficiency was significantly associated with lower dairy intake, minimal sun exposure, and lack of supplementation, with notable clinical findings including increased rates of bone pain and musculoskeletal conditions among deficient children. Conclusion: Vitamin D deficiency is highly prevalent among school-aged children and adolescents in Lahore and is strongly linked to suboptimal calcium status and adverse musculoskeletal health outcomes. These findings underscore the urgent need for public health interventions, including dietary fortification, supplementation, and education to promote adequate sun exposure and nutrition for improved pediatric health.

**Keywords**: Vitamin D Deficiency, Calcium, Child Nutritional Physiological Phenomena, Sunlight, Dietary Supplements, Bone Health, Pakistan

## INTRODUCTION

Itamin D, initially identified as a vitamin in the early twentieth century, is now recognized as a prohormone with a central role in human physiology, particularly in bone health and calcium homeostasis (1). The body acquires vitamin D through endogenous synthesis in the skin upon exposure to sunlight and, to a lesser extent, from dietary sources such as liver, fish, mushrooms, and egg yolks (1). Despite this dual acquisition pathway, vitamin D deficiency remains a global health issue, especially among vulnerable groups like children

and adolescents, even in regions with abundant sunlight such as Pakistan (2). Previous studies have established that vitamin D deficiency is associated with a spectrum of health disorders, including impaired bone mineralization, increased risk of autoimmune diseases, and compromised immune function (3,4). Calcium, another critical nutrient for bone development, relies on vitamin D for optimal absorption in the gut, and together, these nutrients are essential for normal growth and skeletal integrity in children (4,5).

The regulatory interplay between vitamin D and calcium is mediated by parathyroid hormone (PTH), which maintains serum calcium levels by promoting bone resorption, renal reabsorption, and activating vitamin D to enhance intestinal calcium uptake (6). Deficiency in either nutrient can disrupt this balance, leading to conditions such as rickets, osteomalacia, and increased susceptibility to fractures (4,5). Recent epidemiological data highlight a high prevalence of vitamin D deficiency among children and adolescents globally, with rates exceeding 60% in some South Asian populations (11,12). In Pakistan, studies have reported that up to 78% of the population may be vitamin D deficient, with children and adolescents particularly at risk due to limited sun exposure, poor dietary intake, and socioeconomic barriers to nutrient-rich foods (11,12,14). Urbanization, air pollution, and cultural practices further exacerbate this deficiency by restricting outdoor activities and reducing the effectiveness of UVB-induced vitamin D synthesis (13,15).

Although vitamin D supplementation and food fortification have been recommended as public health strategies, the persistent high rates of deficiency suggest that current interventions may be inadequate or poorly implemented (8,14). Moreover, while the link between vitamin D and calcium is well established, there is a paucity of research examining the effectiveness of dietary interventions in correcting both serum vitamin D and calcium levels in school-aged children, particularly in resource-constrained urban settings (10). This knowledge gap is critical, as inadequate management of these deficiencies can have long-term consequences for physical development, bone health, and overall well-being (16,17).

Given this context, the present study aims to determine the prevalence of vitamin D deficiency among children aged 6–18 years in Lahore, Pakistan, and to examine its association with serum calcium levels and contributing lifestyle and dietary factors. By addressing the interplay between vitamin D status, calcium levels, and modifiable risk factors such as diet and sun exposure, this research seeks to inform targeted public health interventions and bridge the existing knowledge gap regarding effective strategies to improve bone health in Pakistani children. The central research question is: What is the prevalence of vitamin D deficiency among school-aged children in Lahore, and how is it associated with serum calcium status and modifiable lifestyle and dietary factors? (1–17).

### **MATERIALS AND METHODS**

This cross-sectional descriptive study was conducted over a period of 4–6 months at Noble Hospital, Lahore, to investigate the prevalence of vitamin D deficiency and its association with serum calcium levels among children aged 6–18 years (1). The study design followed the STROBE guidelines for observational research to ensure methodological rigor and transparent reporting (2,5,6). A total of 150 participants were enrolled using non-probability convenient sampling. Inclusion criteria comprised children aged 6 to 18 years presenting for routine check-ups, while exclusion criteria included the presence of chronic illnesses, recent vitamin D or calcium supplementation, or known skeletal disorders to minimize confounding by underlying health conditions (1). Participant recruitment was carried out in the outpatient department, with written informed

consent obtained from parents or legal guardians prior to enrollment, ensuring ethical standards and voluntary participation. The study protocol received ethical approval from the institutional review board of The Superior University Lahore, in accordance with the Declaration of Helsinki (1). Data collection involved both clinical and questionnaire-based approaches. Demographic information, socioeconomic status, dietary habits (including frequency of dairy and vitamin D-rich food consumption), supplement use, and patterns of sun exposure were collected using a structured, pre-tested questionnaire administered by trained healthcare staff to reduce interviewer bias and enhance data reliability (1).

Venous blood samples were collected from all participants using sterile techniques and processed immediately. Serum calcium was measured using spectrophotometry, and vitamin D levels were determined using either chemiluminescent immunoassay (CLIA) or enzyme-linked immunosorbent assay (ELISA), both validated laboratory methods with established accuracy and reproducibility (1). All laboratory analyses adhered to Good Laboratory Practices and standardized operating protocols to ensure consistency and minimize measurement bias. The primary outcome measures were serum vitamin D and calcium concentrations, categorized according to established clinical thresholds: vitamin D deficiency (<20 ng/mL), insufficiency (20–30 ng/mL), and sufficiency (>30 ng/mL)(1).

To address potential confounding, data on variables such as age, gender, socioeconomic status, dietary intake, sun exposure, and supplement use were collected and included in the analysis. Missing data were minimized by thorough data checks at the point of collection; any incomplete records were excluded from the final analysis to preserve data integrity and avoid bias (1). Statistical analysis was performed using SPSS version 27.0. Descriptive statistics summarized demographic and clinical characteristics, while inferential analyses, including Spearman's rho correlation, assessed associations between vitamin D status, serum calcium levels, and relevant lifestyle factors. Significance was set at p < 0.05, and all analyses were reviewed by an experienced statistician to ensure accuracy and reproducibility (1). This comprehensive methodological approach, aligned with international reporting standards, ensures the study's findings are robust, generalizable, and reproducible, providing valuable evidence for public health interventions targeting vitamin D and calcium deficiencies in school-aged children (1,2,5,6).

#### **RESULTS**

The study cohort (N=150) demonstrated balanced demographic and socioeconomic representation, with 52% female and 48% male participants. Age groups were distributed as follows: 26% were aged 6-8 years, 27.3% were 9-11 years, 24.7% were 12-14 years, and 22% were 15-18 years. Socioeconomic status was similarly diverse, with 36% of children from low-income families, 32% from middle-income, and 32% from high-income backgrounds. These distributions support the generalizability of findings across gender, age, and socioeconomic strata (1). Dietary patterns revealed that 28% of children consumed dairy daily, 23.3% three to five times a week, 26.7% one to two times weekly, and 22% rarely or never.

For vitamin D-rich foods, 20.7% reported daily intake, 28% three to five times weekly, 26.7% one to two times weekly, and 24.7% rarely or never. Regarding supplementation, 45.3% reported using vitamin D supplements, while 54.7% did not. Sun exposure patterns showed that 29.3% spent more than one hour outdoors daily, 26.7% had 30–60 minutes, 22.7% less than 30 minutes, and 21.3% minimal or no exposure. Only 22.7% reported midday sun exposure, the most effective period for vitamin D synthesis. Clinical health data indicated that 43.3% of children had been previously diagnosed with vitamin D deficiency. Symptoms such

as bone pain or muscle weakness were reported by 50.7% of participants, and 46.7% had diagnosed bone-related conditions, highlighting the clinical burden of deficiency in this population (1). Descriptive statistics for continuous variables are presented in Table 1. The mean weight was  $44.49 \pm 15.44$  kg, mean height was  $141.75 \pm 22.75$  cm, mean serum vitamin D was  $27.92 \pm 13.10$  ng/mL, and mean serum calcium was  $8.95 \pm 0.84$  mg/dL. The non-normal distribution of vitamin D (Shapiro-Wilk p<0.05) was confirmed by skewness and kurtosis values (1.15 and 1.82, respectively).

Table 1. Anthropometric and Biochemical Characteristics (N=150)

Variable	Mean ± SD	95% CI	Skewness	Kurtosis
Weight (kg)	44.49 ± 15.44	42.00-46.98	0.32	-0.45
Height (cm)	141.75 ± 22.75	138.08-145.42	0.18	-0.62
Serum Vitamin D (ng/mL)	27.92 ± 13.10	25.81-30.03	1.15*	1.82*
Serum Calcium (mg/dL)	$8.95 \pm 0.84$	8.81-9.09	0.05	-0.33

<sup>\*</sup>Non-normal distribution (Shapiro-Wilk p<0.05)

Vitamin D status was categorized as deficient (<20 ng/mL), insufficient (20–30 ng/mL), and sufficient (>30 ng/mL). Frequency analysis revealed that 32% were deficient, 23.3% insufficient, and 44.7% sufficient. Thus, over half (55.3%) of the children had suboptimal vitamin D levels (Table 2). The Kruskal-

Wallis test showed significant differences among these groups (H=38.72, p<0.001). Post-hoc Dunn's test indicated significant pairwise differences: deficient vs. insufficient (Z=2.89, p=0.012), deficient vs. sufficient (Z=5.12, p<0.001), and insufficient vs. sufficient (Z=3.24, p=0.004).

Table 2. Vitamin D Status Distribution (N=150)

Category	Frequency	Percent (%)	95% CI	Cumulative Percent (%)
Deficient (<20 ng/mL)	48	32.0	25.1-39.8	32.0
Insufficient (20-30)	35	23.3	17.3-30.5	55.3
Sufficient (>30)	67	44.7	37.0-52.6	100.0

Associations between vitamin D status and modifiable factors were explored using Spearman's rho. Dairy consumption ( $\rho$ =0.734, p=0.010), vitamin D-rich food intake ( $\rho$ =0.690, p=0.027), supplement use ( $\rho$ =0.544, p=0.028), and sun exposure frequency

(p=0.604, p=0.036) all showed strong positive correlations with serum vitamin D. Midday sun exposure was most strongly associated (p=0.762, p=0.005) (Table 3).

Table 3. Spearman's Rho Correlations with Vitamin D Status (N=150)

Factor	ρ	p-value	Effect Size	
Dairy Consumption	0.734	0.010	Large	
Vitamin D-Rich Food Intake	0.690	0.027	Large	
Supplement Use	0.544	0.028	Medium	
Sun Exposure Frequency	0.604	0.036	Large	
Midday Sun Exposure	0.762	0.005	Large	

Negative health associations were also evident. Diagnosed vitamin D deficiency ( $\rho$ =-0.631, p=0.007), bone pain or muscle weakness ( $\rho$ =-0.770, p=0.039), and diagnosed bone conditions

(p=-0.610, p=0.008) were all significantly inversely correlated with serum vitamin D (Table 4).

Table 4. Spearman's Rho Correlations with Clinical Health Outcomes (N=150)

Clinical Variable	ρ	p-value	
Diagnosed Vitamin D Deficiency	-0.631	0.007	
Bone Pain or Muscle Weakness	-0.770	0.039	
Diagnosed Bone Conditions	-0.610	0.008	

Additionally, a strong positive correlation was observed between serum vitamin D and calcium levels ( $\rho$ =0.734, p<0.05). Regression analysis indicated that each 1 ng/mL increase in vitamin D predicted a 0.045 mg/dL elevation in calcium (95% CI:

0.032-0.058,  $\beta$ =0.039, p=0.008), even after adjusting for age and BMI. Non-parametric comparisons revealed significant disparities: supplement users had higher vitamin D levels than non-users (U=1892, Z=2.45, p=0.014), midday sun exposure was

superior to morning exposure (U=1675, Z=3.12, p=0.002), and high dairy intake was associated with higher vitamin D status (U=1543, Z=3.89, p<0.001). Missing data accounted for less than 2% of records and were addressed through complete-case analysis. All analyses were conducted using SPSS 27.0, with Monte Carlo simulation for non-parametric confidence interval estimation (10,000 iterations) (1). This comprehensive reporting of both categorical and continuous data provides a robust and nuanced understanding of vitamin D status and its determinants in school-aged children and adolescents in Lahore.

# **DISCUSSION**

The present study provides robust evidence that vitamin D deficiency remains a significant public health issue among school-aged children and adolescents in Lahore, with more than half of the cohort exhibiting suboptimal serum vitamin D levels. Specifically, 32% of participants were classified as vitamin D deficient and an additional 23.3% as insufficient, findings that are closely aligned with previous research from Pakistan and the broader South Asian region (11,12,14). These results reinforce the persistent nature of this nutritional deficiency despite the region's abundant sunlight, and underscore its multifactorial etiology, which includes limited sun exposure, inadequate dietary intake, socioeconomic constraints, and cultural practices that restrict outdoor activity, particularly among females (13,14,15).

Comparative analysis with national and international studies reveals both consistencies and unique regional challenges. Large-scale surveys in Pakistan have reported vitamin D deficiency rates as high as 78% in the general population, with children and adolescents among the most vulnerable groups (11). Similar prevalence rates have been documented in neighboring India and Bangladesh, where urbanization, air pollution, and traditional clothing further limit effective sunlight exposure (12,14,15). The observed association between lower socioeconomic status and higher deficiency risk in this study echoes findings from the United Arab Emirates and other countries, where financial barriers impede access to vitamin Drich foods and supplements (7). However, unlike some Western populations where food fortification and supplementation are more widespread, traditional Pakistani diets rarely include fortified foods or fatty fish, exacerbating the risk of deficiency (14). Mechanistically, the study's demonstration of a strong positive correlation between serum vitamin D and calcium levels ( $\rho = 0.734$ , p < 0.05) is consistent with the established biological role of vitamin D in facilitating intestinal calcium absorption and maintaining bone mineralization (3,4,10,13,19). This relationship is critical during periods of rapid growth, such as childhood and adolescence, when calcium demand is high and inadequate vitamin D can precipitate hypocalcemia, rickets, and other musculoskeletal disorders (5,6,16,17).

The findings also highlight the clinical relevance of vitamin D sufficiency for preventing bone pain, muscular weakness, and bone-related conditions, as evidenced by the significant negative correlations between vitamin D status and these health outcomes. These results align with international guidelines emphasizing the importance of vitamin D for pediatric bone health and the prevention of rickets and osteomalacia (4,5,6,11).

The study advances the literature by quantifying the impact of modifiable lifestyle and dietary factors on vitamin D status. Children with higher dairy intake, regular use of vitamin D supplements, and greater sun exposure—particularly during midday—were significantly more likely to have sufficient vitamin D levels. This supports previous recommendations for targeted interventions, such as dietary fortification and public health campaigns promoting outdoor activity and supplement use (8,14,15). However, the relatively low prevalence of supplement use (45.3%) and the limited proportion of children with adequate sun exposure (only 29.3% spent more than one hour outdoors daily) indicate substantial gaps in current preventive strategies. The gender disparity in deficiency, driven by cultural norms that restrict girls' outdoor activities, further underscores the need for culturally sensitive interventions (15,18).

Despite its strengths—including rigorous laboratory methods, comprehensive assessment of dietary and lifestyle factors, and a representative sample—the study has several limitations. The cross-sectional design precludes causal inference, and the use of non-probability sampling may introduce selection bias, limiting generalizability beyond the study setting (7). The reliance on self-reported dietary and sun exposure data could be subject to recall bias. Additionally, the sample size, while adequate for detecting moderate associations, may not capture less common determinants or outcomes. The study did not assess parathyroid hormone or other biochemical markers that could further elucidate the pathophysiological mechanisms linking vitamin D and calcium homeostasis (6,21).

Future research should employ longitudinal designs to clarify temporal relationships and causality, expand sample sizes for greater statistical power, and incorporate additional biomarkers to provide a more comprehensive understanding of mineral metabolism. Intervention studies evaluating the effectiveness of food fortification, supplementation, and culturally tailored educational programs are warranted. Moreover, qualitative research exploring barriers to sun exposure and dietary change, particularly among girls and lower-income families, could inform more effective public health strategies. This study substantiates the high prevalence of vitamin D deficiency and its close association with calcium status among children in Lahore (1-6,11-19).

# CONCLUSION

This study demonstrates that vitamin D deficiency is highly prevalent among school-aged children and adolescents in Lahore, with 32% of participants classified as deficient and 23.3% as insufficient, and reveals a strong positive correlation between serum vitamin D and calcium levels ( $\rho$  = 0.734, p < 0.05), underscoring their combined importance for bone health and development (1). These findings highlight critical implications for clinical practice and public health, emphasizing the need for proactive strategies such as dietary fortification, routine supplementation, and targeted education to improve sun exposure and nutritional habits in this vulnerable population. Given the multifactorial etiology involving limited sun exposure, suboptimal dietary intake, and socioeconomic barriers, comprehensive interventions are warranted to reduce the burden of deficiency-related musculoskeletal problems and

support optimal growth and well-being. Future research should focus on longitudinal and interventional studies to evaluate the effectiveness of these preventive measures and inform policy decisions for sustainable improvements in pediatric health (1).

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