RESEARCH ARTICLE

Serum Vitamin D Status Among Schoolchildren in Rural and Urban Areas of Tripura, Northeast India: A Comparative Study

Dr. Manidip Chakraborty[1], Dr. Debina Sarkar[2*], Dr. Soumyamoy Das[3], Dr. Sankar Roy[4], Dr. Elvia Jamatia[5]

[1]Assistant Professor, Department of Biochemistry, Tripura Medical College, Agartala.
[2]Assistant Professor, Department of Biochemistry, Dr. B.S. Kushwah Institute of Medical Sciences, Kanpur.
[3]Assistant Professor, Department of Biochemistry, Tripura Medical College, Agartala.
[4]Professor, Department of Biochemistry, Tripura Medical College, Agartala.
[5]Associate Professor, Department of Biochemistry, Dr. Baba Saheb Ambedkar Medical College, New Delhi
***Corresponding Author:** Dr. Debina Sarkar, Email: sarkardebina@gmail.com

Abstract

Background: Vitamin D is required for normal skeletal growth and mineral homeostasis. Differences in living environments may be associated with variation in vitamin D status among children. Data comparing rural and urban schoolchildren in Northeast India remain relatively limited.

Objective: To compare serum 25-hydroxyvitamin D [25(OH)D] concentrations and vitamin D status among schoolchildren from rural and urban areas of Tripura.

Methods: The analysis included laboratory test records for 101 children: 59 classified as rural residents and 42 as urban residents. Serum 25(OH)D was measured using a Mindray VD-T 25-OH Vitamin D Total chemiluminescence immunoassay (CLIA) on a Mindray CL-series analyser. Values were non-normally distributed and are summarised as median and interquartile range (IQR). Rural and urban concentrations were compared using the Mann–Whitney U test. Vitamin D status was categorized using the Indian Academy of Pediatrics thresholds: deficient (<12 ng/mL), insufficient (12–20 ng/mL) and sufficient (>20 ng/mL).

Results: The median serum 25(OH)D concentration was 22.97 ng/mL (IQR 18.27–29.83) overall, 26.22 ng/mL (IQR 19.17–30.42) among rural children and 20.55 ng/mL (IQR 15.78–26.45) among urban children. The rural–urban difference was statistically significant (Mann–Whitney U=937.5, p=0.038).

Deficiency was present in 1/59 (1.7%) rural and 5/42 (11.9%) urban children. The three-category distribution by residence was not statistically significant (Fisher's exact p=0.083).

Conclusion: Rural children had higher median serum 25(OH)D concentrations than urban children in this sample. However, the difference in the distribution of vitamin D status categories by residence did not reach statistical significance. The findings should be interpreted in light of the modest sample size and the absence of data on potential behavioral and environmental determinants.

Keywords: Vitamin D; 25-hydroxyvitamin D; schoolchildren; rural; urban; Tripura; India.

Introduction

Vitamin D has an established role in calcium and phosphate homeostasis and in the mineralisation of growing bone. Childhood and adolescence are important periods for skeletal development, making adequate vitamin D status relevant to normal growth. Serum 25-hydroxyvitamin D [25(OH)D] is used to assess vitamin D status because it reflects vitamin D derived from both cutaneous synthesis and dietary sources.

Low vitamin D concentrations have been described in children living in countries with abundant sunlight, including India. Sunlight availability alone does not determine vitamin D status: ultraviolet-B exposure, time spent outdoors, clothing, skin pigmentation, season and dietary intake may all influence an individual's vitamin D level [8,9]. The Indian Academy of Pediatrics (IAP) revised its paediatric guidance in 2021 and recommended classifying serum 25(OH)D as deficient below 12 ng/mL, insufficient at 12–20 ng/mL and sufficient above 20 ng/mL [1]. These are the categories used in the present analysis.

Evidence from Northeast India includes a cross-sectional study of 500 schoolchildren in and around Guwahati, Assam. Sarma and colleagues reported no significant rural–urban difference in mean 25(OH)D concentrations, although sunlight exposure and calcium intake were associated with vitamin D measures [2]. In a semi-rural Indian school population, Mandlik and colleagues also described a high proportion of children with suboptimal vitamin D status and identified sunlight exposure and body fat as relevant correlates [3]. A multicentre study of Indian children and adolescents reported higher vitamin D concentrations among rural participants and boys, while urban residence was among the factors associated with deficiency [4]. National analyses of the Comprehensive National Nutrition Survey have further shown variation by age, sex, residence and region [5,6]. Differences in sample selection, age, assay methods and classification thresholds need to be considered when comparing results across studies.

The present study compares serum 25(OH)D concentrations and vitamin D categories in schoolchildren recorded as residing in rural and urban areas of Tripura. By examining the two groups within the same regional setting,

the analysis adds local observations to the evidence on vitamin D status in children, an area in which estimates vary across Indian populations and study methods [7,8].

Materials And Methods

Study design and setting

This was a laboratory record-based, cross-sectional comparative study conducted over six months, from January 2025 to June 2025, in Tripura, Northeast India. Children aged 3–17 years who had resided in the selected study area for at least one year and had no known metabolic or endocrine disorders were considered eligible for inclusion. Children receiving vitamin D supplementation or medication, as well as those with a history of chronic illness or hospitalization during the preceding six months, were excluded from the study. Data were obtained from records maintained at a single diagnostic laboratory for children whose recorded residence was classified as rural or urban. A total of 101 eligible records were included: 59 rural and 42 urban.

Study participants and variables

Laboratory records from the single diagnostic laboratory were reviewed for children with an available serum vitamin D result and the demographic information required for the analysis. The variables extracted were age, sex, religion, recorded residence and serum vitamin D concentration. The dataset included 101 records, comprising 59 rural and 42 urban residents. For subgroup analysis, age was grouped as 3–5, 6–11 and 12–17 years; the corresponding numbers were 10, 59 and 32.

Laboratory measurement

Serum total 25-hydroxyvitamin D [25(OH)D] was measured using the Mindray VD-T 25-OH Vitamin D Total (CLIA) reagent kit (Shenzhen Mindray Bio-Medical Electronics Co., Ltd., Shenzhen, China) on a compatible Mindray CL-series chemiluminescence immunoassay analyser. According to the manufacturer's instructions for use, the assay is a competitive-binding chemiluminescence immunoassay intended for quantitative determination of total 25(OH)D in human serum. The required sample volume is 40 µL per test, and results are reported in ng/mL. The manufacturer-stated reportable range is 3.0–150 ng/mL. The reagent kit is stored at 2–8°C; opened reagent may remain on board for up to 28 days under the specified storage conditions.

Definition of vitamin D status

Vitamin D status was categorised according to the revised Indian Academy of Pediatrics recommendations: deficiency, <12 ng/mL; insufficiency, 12–20 ng/mL; and sufficiency, >20 ng/mL [1]. IAP paediatric thresholds were retained to maintain consistency with the study's predefined statistical analysis. Values below 20 ng/mL were also combined to describe low vitamin D in Figure 1.

Statistical analysis

Serum vitamin D values were assessed for normality using the Shapiro–Wilk test and were non-normally distributed. Continuous variables are therefore summarised as median and IQR. Rural and urban serum vitamin D concentrations were compared using the Mann–Whitney U test. Categorical variables are presented as frequencies and percentages. The statistical document reports use of chi-square and Fisher's exact tests where appropriate. A p-value <0.05 was considered statistically significant.

Results

A total of 101 children were included: 59 (58.4%) from rural areas and 42 (41.6%) from urban areas. The median age in the full sample was 10 years (IQR 6–12). Boys accounted for 57 (56.4%) participants and girls for 44 (43.6%). The age-group distribution was 10 (9.9%) children aged 3–5 years, 59 (58.4%) aged 6–11 years and 32 (31.7%) aged 12–17 years. Age and sex distributions did not differ significantly between the rural and urban groups (Table 1).

The median serum 25(OH)D concentration in the full sample was 22.97 ng/mL (IQR 18.27–29.83). Rural children had a higher median concentration than urban children: 26.22 ng/mL (IQR 19.17–30.42) compared with 20.55 ng/mL (IQR 15.78–26.45), respectively. This difference was statistically significant (Mann– Whitney U=937.5, p=0.038; Table 2).

According to the study categories, 6 children (5.9%) were deficient, 31 (30.7%) were insufficient and 64 (63.4%) were sufficient. Deficiency was observed in 1 rural child (1.7%) and 5 urban children (11.9%). The proportion with vitamin D below 20 ng/mL was 30.5% in the rural group and 45.2% in the urban group.

However, the overall distribution across the three vitamin D categories did not differ significantly by residence (Fisher's exact p=0.083; Table 3).

No statistically significant association was observed between vitamin D category and age group (Pearson χ^2=5.885, df=4, p=0.208) or sex (Fisher's exact p=0.315; Table 4). Religion was recorded as Hindu in 78 participants, Muslim in 21 and Christian in 2. The Christian subgroup was too small for a meaningful comparison; religion-specific findings are therefore presented descriptively.

Table 1. Sociodemographic characteristics of participants by residence

Characteristic	Total (N=101)	Rural (n=59)	Urban (n=42)	p-value
Age, years, median (IQR)	10 (6–12)	10 (6–12)	9 (7–13)	0.948*
Age group, n (%)				0.851†
3–5 years	10 (9.9)	5 (8.5)	5 (11.9)	
6–11 years	59 (58.4)	35 (59.3)	24 (57.1)	
12–17 years	32 (31.7)	19 (32.2)	13 (31.0)	
Sex, n (%)				0.488†
Male	57 (56.4)	35 (59.3)	22 (52.4)	
Female	44 (43.6)	24 (40.7)	20 (47.6)	
Religion, n (%)				Not tested
Hindu	78 (77.2)	38 (64.4)	40 (95.2)	
Muslim	21 (20.8)	20 (33.9)	1 (2.4)	
Christian	2 (2.0)	1 (1.7)	1 (2.4)	

Values are n (%) unless otherwise specified. *Mann–Whitney U test calculated from the master chart.
†Pearson chi-square test calculated from the master chart.

Table 2. Serum 25(OH)D concentrations according to residence

Measure	Overall (N=101)	Rural (n=59)	Urban (n=42)
Median (IQR), ng/mL	22.97 (18.27–29.83)	26.22 (19.17–30.42)	20.55 (15.78–26.45)
Minimum–maximum, ng/mL	7.86–75.91	9.26–75.91	7.86–59.96
Mann–Whitney U	937.5	—	—
p-value	0.038	—	—

Table 3. Distribution of vitamin D status by residence

Vitamin D status	Overall (N=101), n (%)	Rural (n=59), n (%)	Urban (n=42), n (%)	p-value
Deficiency (<12 ng/mL)	6 (5.9)	1 (1.7)	5 (11.9)	0.083
Insufficiency (12–20 ng/mL)	31 (30.7)	17 (28.8)	14 (33.3)	
Sufficiency (>20 ng/mL)	64 (63.4)	41 (69.5)	23 (54.8)	
Total	101 (100)	59 (100)	42 (100)	

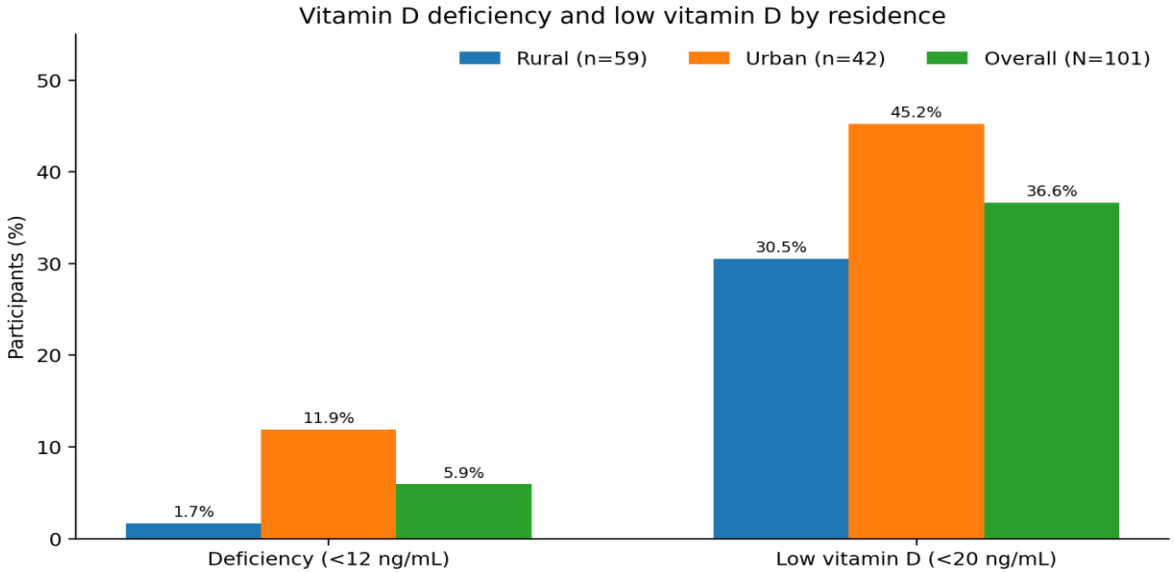


Figure 1. Prevalence of vitamin D deficiency and low vitamin D by residence

Deficiency was defined as serum 25(OH)D <12 ng/mL; low vitamin D was defined as <20 ng/mL. Fisher's exact p-values reported in the statistical document were 0.079 for deficiency and 0.147 for low vitamin D.

Table 4. Vitamin D status according to demographic characteristics

Characteristic	Category	Total n	Deficient n (%)	Insufficient n (%)	Sufficient n (%)	p-value
Age group	3–5 years	10	0 (0.0)	1 (10.0)	9 (90.0)	0.208*
	6–11 years	59	3 (5.1)	17 (28.8)	39 (66.1)	
	12–17 years	32	3 (9.4)	13 (40.6)	16 (50.0)	
Sex	Male	57	4 (7.0)	14 (24.6)	39 (68.4)	0.315†
	Female	44	2 (4.5)	17 (38.6)	25 (56.8)	
Religion	Hindu	78	5 (6.4)	26 (33.3)	47 (60.3)	Not tested
	Muslim	21	1 (4.8)	4 (19.0)	16 (76.2)	
	Christian	2	0 (0.0)	1 (50.0)	1 (50.0)	

*Pearson chi-square test recalculated from the master chart for the three age groups. †Fisher's exact p-value as reported in the statistical document; Pearson chi-square from the master chart was p=0.303.

Discussion

The present study provides valuable regional data on serum 25-hydroxyvitamin D [25(OH)D] concentrations among schoolchildren from Tripura, a region where evidence on vitamin D status in children remains relatively limited. An important finding was the significantly higher median serum 25(OH)D concentration among rural children compared with urban children. This observation highlights the potential influence of residential environment and associated lifestyle patterns on vitamin D status. It also adds to the growing evidence that geographical and residential differences may contribute to variations in vitamin D concentrations among Indian children, even in regions with abundant sunlight.

An interesting aspect of the findings was the difference between the continuous and categorical analyses. Although rural children had significantly higher serum 25(OH)D concentrations, the distribution across deficient, insufficient and sufficient categories did not differ significantly between the two groups. This distinction is important because categorisation converts a continuous biological measurement into broader ranges and may obscure smaller differences in the underlying concentrations. The higher median concentration in rural children therefore represents a meaningful observation, even though the categorical comparison did not demonstrate statistical significance. Similarly, the higher proportion of urban children with concentrations below 20 ng/mL should be interpreted as a numerical trend rather than a confirmed difference in prevalence.

The findings can be viewed in the context of previous studies from Northeast India. Sarma and colleagues, in their study of 500 schoolchildren from rural and urban areas in and around Guwahati, did not observe a statistically significant difference in mean serum 25(OH)D concentrations between the two groups [2]. In contrast, a multicentre study involving Indian children and adolescents reported higher vitamin D concentrations among rural participants than among urban participants [4]. National survey analyses have also demonstrated variations in vitamin D status according to residential setting, although the pattern may differ across age groups and geographical regions [5,6]. A multicentre study of urban school-going children from ten Indian cities further documented a considerable burden of vitamin D deficiency among children and adolescents [10]. The findings from Tripura add to this body of evidence and support the importance of examining vitamin D status across different residential settings rather than assuming that abundant sunlight ensures adequate vitamin D concentrations in all children.

Using the revised Indian Academy of Pediatrics classification, which defines deficiency as serum 25(OH)D below 12 ng/mL, insufficiency as 12–20 ng/mL and sufficiency as above 20 ng/mL [1], 36.6% of the study participants had concentrations below 20 ng/mL. The proportion was 30.5% among rural children and 45.2% among urban children. Although the difference was not statistically significant, the findings indicate that suboptimal vitamin D concentrations remain relevant in both residential groups. This is particularly noteworthy because vitamin D plays an important role in skeletal growth, bone mineralisation and calcium homeostasis during childhood and adolescence. The observations reinforce the importance of maintaining awareness of vitamin D status during these critical stages of growth.

The study also examined variations in vitamin D status across age groups and between sexes. No statistically significant associations were observed, suggesting that the rural–urban difference in serum concentrations was the more prominent finding in this population. However, the relatively small subgroup sizes should be

considered when interpreting these results. The findings provide a useful basis for further investigation into the relationship between residential environment and vitamin D status among children in Tripura.

The study was limited by its relatively small sample size and unequal representation of rural and urban participants, which may have reduced the ability to detect modest differences in categorical vitamin D status and subgroup analyses. As the study was cross-sectional, the observed rural–urban difference cannot establish a causal relationship between residential setting and vitamin D concentrations. In addition, factors such as sunlight exposure, dietary habits, physical activity, screen time, body composition and socioeconomic circumstances may influence vitamin D status and warrant more detailed assessment in future research. Despite these limitations, the study offers useful baseline evidence from Tripura and highlights the need for larger, well-designed studies to better understand regional patterns and determinants of vitamin D status among schoolchildren.

Overall, the present study contributes to the limited regional evidence on vitamin D status among children in Tripura. The significantly higher serum 25(OH)D concentrations among rural participants provide an important observation for future research, while the presence of suboptimal concentrations in both groups underscores the relevance of childhood vitamin D health across residential settings.

Conclusion

Rural children in this study had significantly higher median serum 25(OH)D concentrations than urban children, highlighting a notable variation in vitamin D levels across residential settings in Tripura.

Although the distribution across deficient, insufficient and sufficient categories did not differ significantly, low vitamin D concentrations were observed in both groups. These findings provide useful regional evidence on childhood vitamin D status and underscore the importance of promoting adequate vitamin D health among all schoolchildren. The study also offers a foundation for larger investigations to explore the influence of dietary, environmental and lifestyle factors on vitamin D status and to guide appropriate preventive strategies in the region.

References

1. Gupta P, Dabas A, Seth A, Bhatia VL, Khadgawat R, Kumar P, et al. Indian Academy of Pediatrics Revised (2021) Guidelines on Prevention and Treatment of Vitamin D Deficiency and Rickets. Indian Pediatr. 2022;59(2):142–158.
2. Sarma D, Saikia UK, Baro A. Vitamin D Status of School Children in and around Guwahati. Indian J Endocrinol Metab. 2019;23(1):81–85. doi:10.4103/ijem.IJEM_552_18.
3. Mandlik R, Kajale N, Ekbote V, Patwardhan V, Khadilkar V, Chiplonkar S, et al. Determinants of Vitamin D Status in Indian School-children. Indian J Endocrinol Metab. 2018;22(2):244–248. doi:10.4103/ijem.IJEM_622_17.
4. Khadilkar A, Kajale N, Oza C, Oke R, Gondhalekar K, Patwardhan V, et al. Vitamin D status and determinants in Indian children and adolescents: a multicentre study. Sci Rep. 2022;12:16790. doi:10.1038/s41598-022-21279-0.
5. Ministry of Health and Family Welfare, Government of India; UNICEF; Population Council. Comprehensive National Nutrition Survey (CNNS) National Report 2016–2018. New Delhi: Ministry of Health and Family Welfare; 2019.
6. Mustafa A, Shekhar C. Concentration levels of serum 25-Hydroxyvitamin-D and vitamin D deficiency among children and adolescents of India: a descriptive cross-sectional study. BMC Pediatr. 2021;21:334. doi:10.1186/s12887-021-02803-z.
7. Kamboj P, Dwivedi S, Toteja GS. Prevalence of hypovitaminosis D in India & way forward. Indian J Med Res. 2018;148(5):548–556. doi:10.4103/ijmr.IJMR_1807_18.
8. Munns CF, Shaw N, Kiely M, Specker BL, Thacher TD, Ozono K, et al. Global Consensus Recommendations on Prevention and Management of Nutritional Rickets. J Clin Endocrinol Metab. 2016;101(2):394–415. doi:10.1210/jc.2015-2175.
9. Phinney KW, Sempos CT, Betz JM, Camara JE, Carter GD, Clarke MW, et al. General steps to standardize the laboratory measurement of serum total 25-hydroxyvitamin D. J AOAC Int. 2017;100(5):1230–1233. doi:10.5740/jaoacint.17-0259.
10. Awasthi S, Kumar D, Mahdi AA, Agarwal GG, Pandey AK, Parveen H, et al. Prevalence of specific micronutrient deficiencies in urban school going children and adolescence of India: a multicenter cross- sectional study. PLoS One. 2022;17(5):e0267003. doi:10.1371/journal.pone.0267003.
11. Shenzhen Mindray Bio-Medical Electronics Co., Ltd. 25-OH-Vitamin D Total (CLIA), VD-T: Instructions for Use. Shenzhen, China: Mindray; 2024-04. Reagent kit, catalogue number VD-T113 (50 tests).