VITAMIN D STATUS AMONG CHILDREN AGED 2 MONTHS TO 13 YEARS-A HOSPITAL BASED STUDY

Rimal HS,[1] Walavalkar S[2]*

Affiliation

1. *Associate Professor, Department of Paediatrics, Birat Medical College & Teaching Hospital.*
2. *Senior Consultant, Department of Pathology, Biratnagar Aspatal Pvt. Ltd., Biratnagar*

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**** Corresponding Author***

Dr Hem Sagar Rimal
Associate Professor
Birat Medical College & Teaching Hospital
Tankisinuwari, Morang
Email: hemsagarr@yahoo.com
https://orcid.org/0000-0002-5060-2611

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ABSTRACT

Introduction

Vitamin D is an important micronutrient having crucial role in calcium and phosphate homeostasis but there is emerging evidence to suggest its role in prevention of infectious, inflammatory and neoplastic diseases. The vitamin D insufficiency/deficiency is a widely recognized problem among children in developed as well as developing countries.

Objective

To find out the prevalence of vitamin D deficiency / insufficiency among children aged 2 months to 13 yrs and its correlation with serum calcium, phosphate and demographic factors.

Methodology

Prospective, cross sectional, hospital based study conducted at Birat Medical College Teaching Hospital for a period of one year among children presented to pediatric OPD in whom pediatrician had a suspicion of Vitamin D Insufficiency /Deficiency. Venous blood Samples were taken for the estimation of 25(OH) Vitamin D, Calcium and Phosphate. Statistical analysis was done using SPSS software version 16.

Results

Among 174 children aged 2 months to 13 years, the prevalence of vitamin D insufficiency at the cutoff of <30ng/ml was 82.75% where as no cases of vitamin D deficiency was noted at cut off level of < 10 ng/ml. Children residing in urban area, samples taken in the winter season had higher prevalence of vitamin D insufficiency (p value <0.05). Serum calcium and phosphate level were found positively correlated with Vitamin D level (p<.05). However the relationship was very strong between Vitamin D and Calcium (r=0.402, p-value <0.001) and weak positive correlation was noted with phosphate(r=0.155, p value <0.05).

Conclusion

There is very high prevalence of Vitamin D insufficiency among children. The vitamin D insufficiency was significantly higher in Winter season as compared to summer. Children living in the urban area are more prone to Vitamin D insufficiency. Moderately strong positive correlation was observed between Vitamin D level and calcium but weak correlation existed with phosphate .

KEYWORDS

Calcium, children, phosphate, vitamin D insufficiency

INTRODUCTION

The fat soluble micro nutrient, Vitamin D is known best for its role in the metabolism and absorption of calcium and phosphate from the gastrointestinal tract and it helps in mineralization of bone.[1,2] In Humans, vitamin D is primarily synthesized by the skin through exposure to sunlight (UV-B radiation, wavelength 290–315nm) while only a small fraction (5–10%) comes from the diet.[3-5] In vitamin D deficient condition there is decreased serum calcium level that leads to increased parathyroid hormone secretion which then raises the level of 1,25 dihydroxy vitamin D. With the increasing severity of vitamin D deficiency there is increasing levels of parathyroid hormone, alkaline phosphatase and slightly increased or normal level of 1,25 Dihydroxy Vitamin D but the serum phosphate level remains low. Vitamin D deficiency/insufficiency causes osteomalacia in children and osteoporosis in adults due to ineffective mineralization of bone.

Emerging research has revealed the role of vitamin D beyond bone health including protection from infectious, inflammatory and neoplastic diseases.[6-8] There is wide variation of Vit D levels between different countries across the globe.[9] Common causal factors of vitamin D deficiency have been identified as dark skin pigmentation, low cow's milk intake and breastfeeding without vitamin D supplementation.

Vitamin D level can be measured optimally by examining an individual's 25(OH)D blood level.[10]Although 1,25(OH)D is the biologically active form, it provides no information about vitamin D status as it is often normal or even raised in both children and adults who have deficiency or insufficiency of vitamin D. There has been a lot of controversy regarding the definition of Vitamin D deficiency and insufficiency but the Canadian pediatric society, has defined vitamin D deficiency as having serum 25(OH)D level of <10ng/ml(<25 nmol/L).[11] This cut off level will be taken for this study too. Similarly, Priemel M, von Domarus C, Klatte TO, et al, found in their study that no-one having serum 25(OH)D level of >30 ng/ml (i.e 75 nmol/L) had any signs of osteomalacia.[12] Maxmen A, in the article 'The Vitamin D-lemma' also found a small percentage of subjects (8.5%) having serum 25(OH)D level 21-29 ng/ml showing signs of osteomalacia, which also supports the cutoff for Vitamin D insufficiency being <30 ng/ml(75 nmol/L and more than 10 ng/ml.[8] This cut off level has been used in the course of this study to define insufficiency. There are only a few studies aiming to look at the vitamin D status of the Children residing in Nepal. One of the studies done by Diana et al reported that nearly 91% of preschool Nepali children had vitamin D level <50 nmol/L.[14]

We hypothesize that the prevalence of Vitamin D deficiency and insufficiency is very high among children in the Eastern part of Nepal. Hence this study is designed to find out the Vitamin D status among children and its correlation with demographic factors and calcium and phosphtate levels.

METHODOLOGY

This was a prospective, hospital based cross sectional study, conducted for a period of One year commencing from Jan 2017 to Dec 2017 at Birat Medical College and Teaching Hospital, Biratnagar.

The children aged 2 months to 13 years presenting to Pediatric OPD for various reasons were included in the study whom we suspected to have Vitamin D insufficiency/ deficiency. The criteria to suspect were poor weight and height gain, limb pain, back pain, musculoskeletal pain fatiguability and parental concern about bowed legs. The conventional northern hemispherical seasonal system was used for the purpose of the study. The seasons were categorized as December – February (Winter), March – May (Spring), June–August (Summer) and September– November (Autumn).

The children with diagnosed renal disease and endocrine problems were excluded from the study. Informed consents were obtained from the parents and permission was obtained from the institute to carry out this study.

Sample Collection and Serum preparation:

Venipuncture was performed to collect blood samples taking universal precaution from antecubital vein. Blood samples were allowed to clot for five minutes, followed by centrifugation at 3000 RPM for 15 minutes to separate the serum. All of these steps were carried out under sterile conditions and precautions were taken to prevent blood from hemolysis.

Estimation of Vitamin D:

Competitive immunoassay was performed to determine serum levels of vitamin D using direct Chemiluminescent technology (ADVIA Centaur Vitamin D assay kit). The ADVIA Centaur XP system was employed while performing assay. The system was prepared after loading reagent packs (containing anti- vitamin D anti body). Instrument automatically performed certain actions that include dispensing 100 UL serum into cuvette, adding 115 UL Tithiothreitol, 200 UL solid phase reagent followed by incubation at 37 degrees for 2.5 minutes. Then the machine had separated, aspirated and washed the cuvettes with reagent water. Afterwards instruments had dispensed 300 UL each of acid and base reagents according to the selection of options in system. System had reported serum levels of Vitamin D in ng/ml. Similarly the normal range for calcium and phosphate were considered 8.5-10 mg/dl and 2.5-4.5 mg/dl respectively. The season in which the Vitamin D levels were checked were noted.

Data was entered in excel and transferred to SPSS version 16. The descriptive and inferential statistics were used for data analysis. The test of significance was done by using chi square test. Pearson's coefficient was calculated to see the correlation between vitamin D and serum calcium and phosphate level. The prevalence of vitamin D deficiency/ insufficiency was calculated .

RESULTS

One hundred and seventy four children aged 2 months to 13 years were recruited in the study in which 91(52%)were male and 83(48%) were female. The median value for the age cohort was 68 months (range 2-156 months). (see table 1)

Table 1 : Social demographics of the study population:

Age group (Yrs)	N	Percentage
Infant (<1)	22	12.64
1-5	46	26.44
5-13	106	60.92
Sex		
Male	91	52.3
Female	83	47.7
Residence		
Rural	68	39.08
Urban	106	60.92

Of the 174 children in the study, no one had vitamin D deficiency at cut off of < 10 ng/ml and 144(82.75%) had vitamin D insufficiency at cut off level of >10 ng/ml and <75ng/mL. and 30 (17.25%) had sufficient level of Vitamin D of >30 ng/L. The vitamin D insufficiency was highest (86.36%) in the age group(2months-<1 year) followed by 76.08%, 84.91% in the age group (1-5 years) and more than 5 yrs age group respectively. Among the children studied, those residing in rural area were 68 (39.08%) and rest 106 (60.92%) were from urban area. There was statistically significant (p-value of 0.03) number of vitamin D insufficiency reported in children from urban areas as compared to rural areas. There was no significant difference in vitamin levels in different age group and genders (p value - 0.19, 0.06) respectively. (see table 2)

Table 2 : Vitamin D status of the study population based on Age group, Sex and Residence:

Age group (Yrs)	Normal (> 30 ng/ml) n(%)	Insufficient (10-30 ng/ml) n(%)	Deficiency (<10ng/ml) n(%)	P Value
Infant(<1)	3(13.64)	19 (86.36)	0	**0.06**
1-5	11(23.92)	35 (76.08)	0	
5-13	16(15.09)	90(84.91)	0	**0.19**
Sex				
Male	14 (15.4)	77(84.6)	0	**0.49**
Female	16 (19.3)	67(80.7)	0	
Residence				
Rural	17 (25)	51(75)	0	**0.03***
Urban	13 (12.26)	93(87.74)	0	
Total	30 (17.25)	144 (82.75)	0	

*the test result is significant at p<0.05

The mean serum Vitamin D concentration of the study population was 24.23ng/mL where as it was 24.98 ng/mL in < 1year, 23.72ng/mL in (1-5 years) and 24.25ng/mL in (5-13 years) age group respectively. The Highest percentage of vitamin D insufficiency was noted in the winter season 55(94.8%) followed by autumn19 (90.4%), spring 36(80%) and summer season 34(68%). In the winter the insufficiency of vit D was noted to be higher than that in the summer and the test result was statistically significant (p-value of <0.05). (See table 3)

Table 3 : Season wise Vitamin D status of study population

Season	Normal (> 30 ng/ml)	Insufficient (10 – 30 ng/ml)	P Value
Winter	3(5.2)	55(94.8)	0.001*
Summer	2(9.6)	19(90.4)	
Spring	9(20)	36(80)	0.3
Autumn	14(32)	34(68)	

*the test result is significant at p<0.05

The correlation between Vitamin D and Calcium as well as Vitamin D and phosphate were examined. The mean Calcium and phosphate concentration among study population were 9.13 mg/dl(SD 1.01) and 3.67 mg/dl(SD 0.76) respectively. Vitamin D status of the study population has moderately strong positive correlation (r=0.402, p-value of <0.0001) with serum calcium level however the Vit D level in the study population had weak positive correlation (r=0.155, P-value 0.04). (See table 4)

Table 4: Correlation of Vitamin D with Calcium and Phosphate.

Vitamin D	Calcium	Phosphate
r	0.402	0.155
P-value	<0.001	<0.05

DISCUSSION

Vitamin D deficiency/insufficiency has already been considered as an important health related problem throughout the world with its implication in various diseases/conditions.[6]

We noted very high prevalence of Vitamin D insufficiency in our study (82.7%) with cut off of <30 ng/ml which is comparable to the study of Diana et al where the prevalence was 91%.[14] Even higher prevalence of 95%was noted in a large prevalence study (1-11 yrs) done in USA keeping cut off level of <30 ng/ml whereas18% had<20 ng/mL and only <1% had level <10 ng/mL. We had no cases of Vitamin D deficiency, at a cut off of < 10 ng/ml which is different from the study in USA.[15]

In our study higher percentage of vitamin D deficiency was noted in the age group (2 months- 1 yr) and this finding is higher than the finding from a study done by A.M. Molla, et al.in Kuwait in which 60% of new borns and infants had < 20

ng/ml of 25(OH) vitamin D.[16]This needs to be cautiously interpreted as the result was statistically insignificant at p value < 0.05 and also the sample size was small in this category.

This study noted that children from Urban area had higher percentage of vitamin Insufficiency as compared to children from rural area and the test result was significant at p value of < 0.05. This is probably due to greater sun exposure time for rural children. There are evidences to support the fact that decreasing outdoor activities are linked to reduction in the vitamin D levels as there will be less exposure to the sunlight.[17]

This study has further supported the fact that sun exposure is an important determinant of Vitamin D level as we noted much higher percentage of Vitamin D insufficiency in the winter season as compared to summer (p value at < 0.05). [18] When we looked at the correlation of Vitamin D with calcium by calculating Pearson's coefficient, Serum calcium showed moderate correlation with Vitamin D level with Pearson value of r being 0.402 and the test was statistically significant at p- value of < 0.05. There have been other studies to show this positive correlation.[19] At the same time the correlation of Phosphate with Vitamin D was weak . This could be due to better intake of phosphate in the study population.

CONCLUSION

There is high prevalence of vitamin D across all age groups in Nepal. There is higher prevalence of Vitamin D insufficiency among children residing in urban as compared to the rural area. Vitamin D insufficiency is more pronounced during winter season. Moderately strong positive correlation was observed between Vitamin D level and calcium but weak correlation existed with phosphate level.

RECOMMENDATIONS

In view of higher prevalence of Vitamin D insufficiency in < 1 year age group children, vitamin D level of pregnant mother can be moniterod and early intervention for mother as well as newborns and infants can be instituted by supplementing them with vitamin D. There is a need of nationally representative translational research to know the vitamin D status of infants and children.

LIMITATION OF THE STUDY

This study would have been more informative if we had looked at dietary and sun exposure pattern in the study population. Biochemical parameters like alkaline phosphatase should have been evaluated to understand the correlation better.

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CONFLICT OF INTEREST

None declared.

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