

Correlation Of Vitamin D Status With Pediatric Leg (Growing) Pains

Shah Nawaz¹, Irfan Khan², Arshad Khan³, Saifoor Ahmad Khan⁴, Zahid Irfan Marwat⁵,
Alamzeb Jadoon⁶

¹. Professor of Biochemistry, Nowshera Medical College Nowshera, Khyber Pakhtunkhwa (KP)

². Associate Professor of Pediatrics Medicine, Nowshera Medical College/ QHAMC, Nowshera KP

³. Assistant Professor of Pediatrics Medicine, Nowshera Medical College/ QHAMC, Nowshera KP

⁴. Assistant Professor of Community Medicine, Nowshera Medical College Nowshera, KP

saifoorahmadkhan@yahoo.com

⁵. Professor of Biochemistry, Nowshera Medical College Nowshera KP

⁶. Associate Professor of Physiology, Nowshera Medical College Nowshera KP

Cite this paper as: Shah Nawaz, Irfan Khan, Arshad Khan, Saifoor Ahmad Khan, Zahid Irfan Marwat, Alamzeb Jadoon (2024) Correlation Of Vitamin D Status With Pediatric Leg (Growing) Pains. *Frontiers in Health Informatics*, 13(3), 525-533.

Abstract

Background: Vitamin D plays an important role in calcium and bone metabolism. It helps in the mineralization of bone and calcification of growth plate in children. Its deficiency has been associated with various conditions of chronic pain in adults in certain studies. Objective was to determine serum 25(OH)D levels & Vitamin D status in pediatric population aged 5-12 years presenting with growing pains (GP)/leg pains, belonging to Nowshera, Khyber Pakhtunkhwa & to find any correlation between vitamin D status of children and GP.

Methods: Hundred children presenting with GP and hundred children without GP were randomly selected in a cross-sectional study. Their blood samples were taken for the measurement of 25(OH) D levels and to determine their vitamin D status. Intensity of growing pains was measured using Visual Analog Scale (VAS). The correlation of vitamin D status with GP was determined with Pearson's Correlation.

Results: Of the 200 children included in the study, 68% (n=170) were found to have low vitamin D status. Among these, children with GP (n=100) had a mean VAS score of 6.5. Children with GP when compared to healthy children without GP showed significantly low levels of serum 25(OH) D (ng/mL) (13.4 ± 8.6 vs 16.25 ± 7.2 , $p < 0.05$) and Hemoglobin (g/dL) (10.8 ± 2.3 vs 11.7 ± 2.6) while significantly high level of ALP (U/L) (318.17 ± 40.14 vs 268.24 ± 34.92 , $p < 0.05$). Serum 25 (OH) D level showed a statistically significant negative correlation with VAS score ($r = -0.387$, $p = 0.0001$). Pearson correlation analysis confirmed low vitamin D status as an independent predictor of GP.

Conclusion:

Serum 25(OH) D concentration is significantly decreased in children with GP which concludes that Hypovitaminosis D and low Vitamin D status is related with greater intensity of pain in children with GP. Moreover, Vitamin D is associated negatively with ALP in the studied population. Routine screening and management of vitamin D deficiency may help reduce the incidence and severity of GP, thereby improving the quality of life in affected children.

INTRODUCTION

Vitamin D is a sterol that has a hormone-like function. 1, 25-dihydroxycholecalciferol (1, 25-diOH-D3), the active molecule, binds to intracellular receptor proteins. The 1, 25-diOH-D3–receptor-complex interacts with DNA in the nucleus of target cells and either selectively stimulates gene expression or specifically represses gene transcription. The most prominent action of 1, 25-diOH-D3 is to regulate the plasma levels of calcium and phosphorus. It performs this function by increasing uptake of calcium by the intestine, minimizing loss of calcium by the kidney and stimulating resorption of bone when necessary. The normal homeostasis of calcium and phosphorus is vital for bone mineralization, normal growth-plate calcification, skeletal growth and overall bone health¹⁻³.

Normal serum Vitamin D level is likely to be protective against various musculoskeletal disorders like fractures and weakness of muscles^{4,5}. Serum level of 25(OH) D is believed to be the best test to determine or indicate vitamin D status of an individual. It is reported in both nanograms per milliliter (ng/mL) and nanomoles per liter (nmol/L). One ng/mL equals 2.5 nmol/L & One (1) nmol/L equals 0.4 ng/mL. Serum level of <30 nmol/L of 25(OH) D is regarded as Vitamin D deficiency whereas a level of 30 to <50 nmol/L as Vitamin D insufficiency⁶⁻⁸.

The available assays used by laboratories to measure serum 25(OH) D are considerably variable & hence complicate the analysis. Falsely low or falsely high levels are common depending on the laboratory & the assay used. Procedures have been developed by the International Vitamin D Standardization Program to standardize the measurement of serum 25(OH) D in the laboratories⁹⁻¹².

An association between chronic pain conditions in adult patients and hypovitaminosis D has been described^{13,14}. It was hypothesized that an association might also exist between vitamin D deficiency & the intensity of pediatric leg pains, long described as “growing pains”.

GP refers to the most common recurrent lower limb pain condition in children¹⁵. The term was first coined in medical literature in 19th century as *Maladies de la Croissance* (pains of growth) by Marcel Duchamp, a French physician¹⁶. Since that time, it has been a topic of many studies¹⁷⁻²⁷. Though the term seems to be a misnomer as there is no evidence that growth *per se* is reason for pain, it enjoys acceptance and popularity worldwide.

Mainly affecting children aged 3–12 years, GP are located in the muscles, predominantly arising in the anterior thighs, calves, backs of the knees or shins, precipitated by increased activities and daily exercise like playing, running etc. and lasting from minutes to hours. GP are typically non-articular, intermittent & bilateral, and usually occur during the night to the extent that may awaken the child from sleep. Older children often describe GP as cramping & severe; parent/s of younger children report that their children have crying episodes due to the pain intensity. On examination there are no objective signs of inflammation, and laboratory tests generally reveal normal results²⁸⁻²⁹.

The prevalence of GP is estimated to range from 2.6% to 36.9%; the wide range being mainly due to unspecified & different sizes of samples, different age ranges in the literature, and lack of objective diagnostic criteria adopted in different studies³⁰⁻³². The intensity of pain in GP can be quantified with the help of Visual Analogue Scale (VAS) which is widely used for older children for self-reporting³³⁻³⁵.

Our country receives a high level of sunlight, and Hypovitaminosis D is thought to be unusual here; however, in one study³⁶, the prevalence of Vitamin D deficiency in this country has been reported as 53.5%.

In this study, we compared the vitamin D status of children with and without growing pains by checking their serum 25-hydroxyvitamin D [25(OH) D] level and investigated whether such limb pains were associated with a low vitamin D status.

MATERIAL AND METHODS

This cross-sectional analytical study was conducted at Medical Teaching Institution (MTI) Nowshera, Khyber Pakhtunkhwa, for a period of six months. This study was conducted on randomly selected otherwise healthy children, both male & female, with age ranging 4 to 10 years, who were brought to the pediatric outpatient department of the institution's attached hospital with the complaint of leg pains. The study consisted of two groups: Group I consisted of 100 control healthy children without leg (growing) pains and group II included children with leg (growing) pains.

The inclusion criteria for the group II were children with complaint of intermittent, bilateral, nocturnal, leg pains with no limitation of activity and normal physical examination, as defined by Evan. VAS was used for assessing level of pain intensity in these children. The children who had adequate cognitive development for understanding the scale and for others, their parents/attendants, were taught in detail, how to use the scale. The children and/or the parents/attendants were asked to mark the point on the scale as a level of pain intensity in the most recent attack. VAS consisted of a linear scale, a 10 centimeters (cm) long horizontal line. The number of 'cm' marked was recorded as a level of pain intensity; mild to moderate pain was defined as a score of ≤ 5 and moderate to severe as 6 to 10.

Children who had persistent pain still occurring the following morning with increasing intensity, pains located in the joints, unilateral pains or those associated with any abnormal physical examination related to the musculoskeletal system, and/or if they had underlying systemic illnesses such as rheumatologic disorders, protein calorie malnutrition, rickets or celiac disease were excluded from the study.

A written informed consent was obtained from the children's parent/s or caregivers/attendants after fully explaining the aim of the study to them before their child being enrolled for the study. Approval of the study was obtained from the Institutional Ethical Review Board.

After enrollment, anthropometric measurements like weight & height of all subject children were measured and the body mass index (BMI) calculated for each. Venous blood sample of 6 ml was collected from each child included in the study. Full blood count (FBC) for the purpose of Hemoglobin (Hb), the erythrocyte sedimentation rate (ESR) and serum levels of Calcium (Ca^{++}), Phosphorus (P), Alkaline phosphatase (ALP) were measured on 3 ml of the fresh samples. The remaining blood sample was centrifuged at 3000 revolutions per minute (rpm) to get serum which, for later analysis of 25(OH) D, was stored at -70°C .

Serum Ca^{++} was measured colorimetrically using kit provided by Labtech. Serum ALP & P (inorganic) were measured colorimetrically using kit provided by DiaTech Plus, Germany.

A chemiluminescence enzyme assay was used to determine 25(OH) D level using method (AccuLite CLIA Microwells, Monobind Inc. USA). Operational definition for low vitamin D status included both Vitamin D deficiency (VDD) (serum level of < 30 nmol/l (12 ng/ml) & Vitamin D insufficiency (VDI) (serum level of 30 to < 50 nmol/l (12 to < 20 ng/ml), whereas a level of ≥ 50 nmol/l (≥ 20 ng/ml) was believed to be sufficient.

For the statistical analysis of data, the statistical program, Statistical Package for the Social Sciences (SPSS) Version 22 was used. Results were expressed as Mean \pm SD. Distribution of categorical variables was presented

as percentages & frequencies A probability (p) value of < 0.05 was accepted as statistically significant. Comparison of parameters between groups I & II was done by using independent sample t-test. The correlation of vitamin D status in the form of 25 (OH) D level with growing pains in the form of VAS scores was determined using Pearson correlation coefficient.

RESULTS:

Children without GP in the control group I, comprised of 62 boys and 38 girls and in the GP group II, 58 boys & 42 girls. No statistically significant difference was found between groups I & II in terms of gender (p>0.05).

From the VAS scores among the children with GP, group II, 63 children experienced mild to moderate and 37 children, moderate to severe pain (Table 1).

Mean and standard deviation of the age & anthropometric parameters of the studied population are summarized in Table 2 and the hematological/biochemical parameters in Table 3. Group I included 100 healthy children without GP having a mean age (in years) of 6.9±2.9 while group II included 100 children with GP having a mean age (in years) of 7.4±2.7. A p-value > 0.05 shows that there is no statistically significant difference between both the groups. Similarly, though the inter group comparison for weight and height as independent measurements are not significant (p > 0.05), that for B.M.I (kg/m²) was statistically significant (p < 0.05). The comparison of variables between the GP group (II) and the control group (I), showed significantly high levels of ALP & low level of Hb in the former than the latter (p < 0.05), but ESR, Serum Ca & P levels of no statistical significance between the two groups. Serum (25 OH) D level (ng/mL) was significantly lower in children with GP with 13.4 ± 8.6 as compared to 16.25 ± 7.2 in normal healthy children without GP (p value < 0.05).

The Pearson correlation coefficient (r) between serum Vitamin D levels in children with GP and VAS scores was found to be -0.387 with a p-value of far less than < 0.05 indicating a strong negative correlation which is statistically significant (Figure). This suggests that with higher Vitamin D levels, the intensity of pain (as VAS score for growing pains) decreases; in other words, lower vitamin D status is associated with GP and higher level of pain intensity.

Table 1: Evaluation of intensity of pain among Group II (n=100) children with growing pain by VAS Score

Severity of Pain	Score	% age
Mild to Moderate	1-≤5	63
Moderate to severe	6-10	37

Table 2: Comparison of age & anthropometric parameters among children (N=100) with GP (Group II) and children (N=100) without GP (Group I)

Parameters	Group I	Group II	p value*
Age (yr.)	6.9 ± 2.9	7.4 ± 2.7	0.208
Weight (kg)	21.4±5.6	22.8±6.2	0.095
Height (cm)	122.4± 5.3	123.5 ± 5.9	0.167

Body Mass Index (kg/m ²)	15.8 ± 1.3	16.3± 1.6	0.016
--------------------------------------	------------	-----------	-------

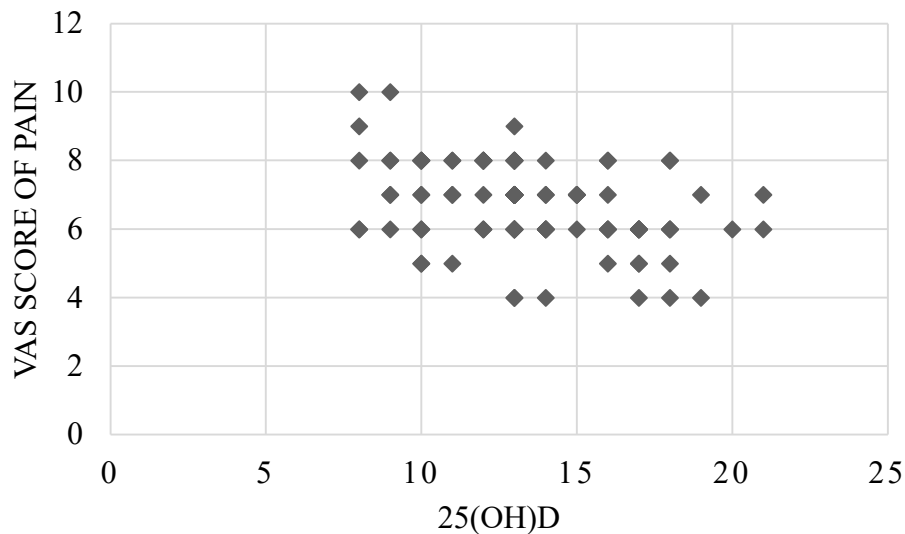
*p-value < 0.05 considered significant

Table 3: Comparison of hematological & biochemical parameters among children (n=100) with GP (Group II) and children (N=100) without GP (Group I)

Parameters	Group I	Group II	p value*
Hemoglobin (g/dL)	11.7±2.6	10.8±2.3	0.010
ESR (mm/1 st hour)	9.1±2.2	10.1±2.1	0.320
Serum Calcium (mg/dL)	9.2±1.2	8.9±1.3	0.091
Serum Phosphorus (mg/dL)	4.6±1.3	4.8±1.2	0.259
Serum Alkaline Phosphatase (U/L)	268.24±34.92	318.17±40.14	0.001
Serum 25 (OH) D (ng/ml)	16.25 ± 7.2	13.4 ± 8.6	0.011

*p-value < 0.05 considered significant

Figure: Correlation between serum Vitamin D levels in children with GP (n=100) and their VAS scores



D status. We also hypothesized that an association might exist between low vitamin D status & the intensity of pediatric GP.

The mean age of 7.4 yrs in children with GP in our study is comparable to mean age of 7.8 yrs in a study conducted by Vehapoglu A et al. in Turkey³⁷ and in another study by Asadi Pooya et al.²⁹ who noted a mean age of 7.88 yrs. A study from India³⁸ reported a mean age of 9.6 years; the reason for higher age in that study may be due to lesser sample of subject children taken (n= 45). A mean age of 5.2 yrs was reported in another study from Korea³⁹; lower mean age in that study may be explained by the fact that children in age ranging 2-15 yrs were included in that study.

The predominance of boys over girls in GP group in our study is comparable to the studies by Joghee et al⁴⁰ & Haq MI et al³² whereas El Metwaly et al⁴¹ reported in a study that leg pains were experienced by girls more frequently than boys. The predominance of boys with GP can be attributed to more outdoor physical activities by boys than girls and also to our cultural & social trend where parents seek medical attention for boys more frequently than girls. No such relationship was found between gender and musculoskeletal pains by Paladino et al.⁴²

GP were not related to age, sex, weight & height in our study but its relationship with BMI was statistically significant. Our results are comparable to the findings in the studies of Sunil Joghee et al⁴⁰ & Vehapoglu et al³⁷ who too did not find any relationship between GP and parameters of gender, age and anthropometric indices of height and weight.

In our study, hemoglobin was significantly lower in children in the GP group as compared to that in children without GP (the control group). This fact is similar to a case-control study carried out on 77 children by Evans et al³⁰ in which low Hb and anemia was found in 10.7% of the children with growing pains.

The parameter indicating inflammation i.e. ESR was evaluated in children in the GP and those in (otherwise healthy) control groups. It is matter of concern if elevated; ⁴³ it was found to be normal in our study, similar to other studies^{44,45}.

We found normal levels of serum Ca & P in our study but statistically significant higher serum ALP in subject children with GP. Marwaha et al⁴⁶ showed a low levels of serum Ca and higher levels of Alkaline Phosphatase comparable to our study. Our findings are also comparable to the results of Haq MI et al³² & Peacey et al⁴⁷. who found in their studies that the levels of calcium, phosphate and Alkaline Phosphatase were normal in all children they studied.

The mean value of 25(OH) D level in group II children with GP in our study was found to be low; this fact is consistent with various other studies⁴⁸⁻⁵¹. In a study carried out by Park et al.⁴, the mean value of 25(OH)D level was found to be more (19.1 ng/mL) in children with GP compared to that in our study (13.4 ng/mL). However, this former study didn't include a control group. Although the mean values of 25(OH) D levels have been reported differently, VDD/VDI or low vitamin D status is generally found in children with GP, as was found in our study as well.

The authors agree with the proposed mechanism described by Morandi G et al.⁵² in that the pain in children with low Vitamin D status might be due to their less thick bones in turn first due to deposition by osteoblasts and then of expansion of rubbery matrix on the periosteal & endosteal both surfaces of the affected bone, thus putting an outward pressure and anomalous weight on sensory pain fibers of the bone, bringing on pain.

REFERENCES:

1. Christakos S, Dhawan P, Verstuyf A, Verlinden L, Carmeliet G. Vitamin D: Metabolism, molecular mechanism of action, and pleiotropic effects. *Physiol. Rev.* 2016; 96:365–408
2. Ferrier DR. Vitamins (Vitamin D). In; Harvey RA, series editor. *Biochemistry; Lippincott's Illustrated Reviews*. 6th ed. Philadelphia, PA: Lippincott Williams & Wilkins; 2014. p. 396-399
3. Palermo NE, Holick MF: Vitamin D, bone health, and other health benefits in pediatric patients. *J Pediatr Rehabil Med* 2014; 7: 179–192.
4. Pludowski P, Holick MF, Pilz S, et al. Vitamin D effects on musculoskeletal health, immunity, autoimmunity, cardiovascular disease, cancer, fertility, pregnancy, dementia and mortality – a review of recent evidence. *Autoimmun Rev* 2013; 12: 976–989
5. Wintermeyer E, Ihle C, Ehnert S, Stockle U, Ochs G, de Zwart P, Flesch I, Bahrs C, Nussler AK. Crucial role of vitamin D in the musculoskeletal system. *Nutrients*. 2016; 8(6): 319.
6. Misra M, Pacaud D, Petryk A, et al: Vitamin D deficiency in children and its management: review of current knowledge and recommendations. *Pediatrics* 2008; 122: 398–417.
7. Giustina A, Adler RA, Binkley N, Bouillon R, Ebeling PR, Lazaretti-Castro M, Marcocci C, Rizzoli R, Sempos CT, Bilezikian JP. Controversies in Vitamin D: Summary Statement from an International Conference. *J Clin Endocrinol Metab.* 2019; 104 (2):234-240.
8. Herrmann M, Farrell CL, Pusceddu I, Fabregat-Cabello N, Cavalier E. Assessment of vitamin D status—a changing landscape. *Clin Chem Lab Med.* 2017;55(1):3–26.
9. Binkley N, Sempos CT; Vitamin D Standardization Program (VDSP). Standardizing vitamin D assays: the way forward. *J Bone Miner Res.* 2014; 29(8):1709–1714.
10. Carter GD, Jones JC, Shannon J, Williams EL, Jones G, Kaufmann M, Sempos C. 25-Hydroxyvitamin D assays: potential interference from other circulating vitamin D metabolites. *J Steroid Biochem Mol Biol.* 2016; 164:134–138.
11. Máčová L, Bičíková M. Vitamin D: Current Challenges between the Laboratory and Clinical Practice. *Nutrients*. 2021; 13(6): 1758.
12. Tsuprykov O., Chen X., Hoche C.F., Skoblo R., Lianghong Y., Hoche B. Why should we measure free 25(OH) vitamin D? *J. Steroid Biochem. Mol. Biol.* 2018; 180:87–104
13. de Rezende Pena C, Grillo LP, das Chagas Medeiros MM: Evaluation of 25-hydroxyvitamin D serum levels in patients with fibromyalgia. *J Clin Rheumatol* 2010; 16: 365–369.
14. Tandeter H, Grynbaum M, Zuili I, et al: Serum 25-OH vitamin D levels in patients with fibromyalgia. *Isr Med Assoc J* 2009; 11: 339– 342
15. Evans AM: Growing pains: contemporary knowledge and recommended practice. *J Foot Ankle Res* 2008; 28: 4
16. Duchamp M. Maladies de la Croissance. In: Levrault FG, editor. *Mémoires de médecine pratique*. Paris: Jean- Frédéric Lobstein; 1823.

17. Leung A, Robson W. Growing pains. *Can Fam Physician*. 1991; 37:1463-7
18. Al-Khattat A, Campbell J: Recurrent limb pain in childhood ('growing pains'). *Foot*. 2000; 10:117-123.
19. Apley J: Clinical canutes: A philosophy of pediatrics. *Proc Royal Soc Med*. 1970; 63(5):479-484.
20. Bennie P: Growing pains. *Arch Pediatr*. 1894; 11(5):10.
21. Brady M, Grey M: Growing pains: a myth or reality. *J Pediatr Health Care*. 1989; 3(4): 219-220.
22. Cullen K, Macdonald W: The periodic syndrome: its nature and prevalence. *Med J Aust*. 1963; 2(5):167-73.
23. Hawksley J: Race, Rheumatism and Growing Pains. *Arch Dis Child*. 1931; 6:303-306.
24. Oster J, Nielson A: Growing pain: a clinical investigation of a school population. *Acta Paediatr Scand*. 1972; 61:329-334.
25. Sherry D: Limb pain in childhood. *Pediatr Rev*. 1990; 12(2):39-46.
26. Weiner SR: Growing pains. *Am Fam Physician*. 1983; 27:189-191.
27. Williams M: Rheumatic conditions in school children. *Lancet*. 1928; 6:720-721.
28. Pavone V, Vescio A, Valenti F, Sapienza M, Sessa G. Growing pains: What do we know about etiology? A systematic review. *Gianluca Testa World J Orthop* 2019 April 18; 10(4): 176-218
29. Asadi-Pooya AA, Bordbar MR: Are laboratory tests necessary in making the diagnosis of limb pains typical for growing pains in children? *Pediatr Int* 2007; 49: 833–835.
30. Evans AM, Scutter SD: Prevalence of 'growing pains' in young children. *J Pediatr* 2004; 145: 255–258.
31. Khuntadar BK, Mondal S, Naik S, Mohanta MP. Prevalence of growing pains in a general paediatric OPD: A descriptive, observational and cross-sectional study. *Journal of Family Medicine and Primary Care*. 2023 ;12(1):117-22.
32. Haq MI, Ahmed A. A prospective study for causal relationship of growing pains and vitamin D. *International Journal of Contemporary Medical Research*. 2021; 8(7): G5-G9.
33. Insaf AI. Growing pains in children and Vitamin D Deficiency, The impact of Vit D treatment for resolution of symptoms. *Journal of Health, Medicine and Nursing*. 2017; 39: 80-85.
34. Wehby GL, Naderi H, Robbins JM, et al: Comparing the Visual Analogue Scale and the Pediatric Quality of Life Inventory for measuring health-related quality of life in children with oral clefts. *Int J Environ Res Public Health* 2014; 16: 4280–4291.
35. Dhanani S, Quenneville J, Perron M, et al. Minimal difference in pain associated with change in quality of life in children with rheumatic disease. *Arthritis Rheum* 2002; 15: 501–505.
36. Riaz H, Finlayson AE, Bashir S, Hussain S, Mahmood S, Malik F, Godman B. Prevalence of Vitamin D deficiency in Pakistan and implications for the future. *Expert Rev Clin Pharmacol*. 2016; 9(2): 329-38.
37. Vehapoglu A, Turel O, Turkmen S, Inal BB, Aksoy T, Ozgurhan G, Ersoy M. Are Growing Pains

- Related to Vitamin D Deficiency? Efficacy of Vitamin D Therapy for Resolution of Symptoms. *Med Princ Pract.* 2015; 24(4):332-8
38. Mohanta MP: Growing pains: practitioners' dilemma. *Indian Pediatr* 2014; 51: 379–383
 39. Park MJ, Lee J, Lee JK et al. Prevalence of vitamin D deficiency in Korean children presenting with nonspecific lower-extremity pain. *Yonsei Med J* 2015; 56:1384-8.
 40. Joghee; *International Journal of Basics and Applied Science.* /dept. Of Pediatrics, Dr. RML Hospital and PGIMER 2012; 2:221-234.
 41. El-Metwally A, Salminen JJ, Auvinen A, Kautiainen H, Mikkelsen M. Prognosis of non-specific musculoskeletal pain in preadolescents: a prospective 4-year follow-up study till adolescence. *Pain.* 2004 ; 110(3): 550-559
 42. Paladino C, Eymann A, Llera J, De Cunto CL. Estimated prevalence of musculoskeletal pain in children who attend a community hospital. *Arch Argent Pediatr.* 2009; 107:515-9.
 43. Lehman PJ, Carl RL. Growing pains: when to be concerned. *Sports Health.* 2017 Mar;9(2):132-8.
 44. Liao CY, Wang LC, Lee JH et al. Clinical, laboratory characteristics and growth outcomes of children with growing pains. *Sci Rep.* 2022; 12: 14835
 45. Vito P, Elena L, Valerio G, Francesco RE, Luciano C, Giuseppe S. Growing Pains: A Study of 30 Cases and a Review of the Literature. *Journal of Pediatric Orthopaedics.* 2011; 31(5): 606-609
 46. Marwaha RK, Khadgawat R, Tandon N, Kanwar R, Narang A, Sastry A, Bhadra K, Kalaivani M. Reference intervals of serum calcium, ionized calcium, phosphate and alkaline phosphatase in healthy Indian school children and adolescents. *Clinical Biochemistry.* 2010; 43 (15): 1216-1219,
 47. Peacey SR. Routine biochemistry in suspected vitamin D deficiency. *J R Soc Med* 2004; 97:322-5.
 48. Ali MA, Haque M, Islam MI, Khan MZ, Rahman SA. Serum Vitamin D and Bone Mineral Density in Children with Growing Pain in a Tertiary Hospital of Bangladesh. *Open Journal of Pediatrics.* 2022 Oct 26;12(5):815-26.
 49. Plotnikoff GA, Quigley JM. Prevalence of severe hypovitaminosis D in patients with persistent, non-specific musculoskeletal pain. *Mayo Clin Proc* 2003; 78:1463-70. Comment in: p. 1457-9.
 50. Günbey Ö, Gürgöze MK, Günbey FB. Vitamin D Levels in Growth-Paining Children: Vitamin D Treatment in Growth Pain. *The Journal of Pediatric Academy.* 2024.
 51. Morandi G, Maines E, Piona C, Monti E, Sandri M, Gaudino R, Boner A, Antoniazzi F. Significant association among growing pains, vitamin D supplementation, and bone mineral status: results from a pilot cohort study. *J Bone Miner Metab* 2015; 33: 201-206