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ORIGINAL RESEARCH

Serum Vitamin D, Calcium and Phosphate among children with pneumonia

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Abstract

Background: Vitamin D is partly responsible for maintaining calcium and phosphate homeostasis but has been shown to have immune modulatory functions.
Objective: To measure serum levels of vitamin D, and plasma levels of calcium and phosphate in children with pneumonia and compare with those of apparently healthy controls.
Methods: A cross-sectional study involving seventy-six children with pneumonia, matched with 76 apparently healthy controls.
Results: There was no statistically significant difference in the mean serum vitamin D levels between the cases and the controls (t = 1.190, p = 0.236). The mean serum vitamin D level was significantly higher in children with non-severe pneumonia than in those with severe pneumonia (t = 3.299, p = .002). The mean serum vitamin D level was higher among the controls than those with severe pneumonia (t = 2.674, p = 0.009). The mean plasma calcium and phosphate levels in the controls were significantly higher than in the cases (t = 2.528, p = .013 and t = 4.594, p < .001 respectively). Plasma calcium and phosphate levels did not vary with the severity of pneumonia. Pneumonia was independently associated with the occurrence of hypocalcaemia and hypophosphataemia (OR = 4.366, 95% CI = 1.851-10.295, p = 0.001; OR = 7.355, 95% CI = 1.545-35.027, p = 0.01 respectively).
Conclusion: Children with severe pneumonia had lower levels of vitamin D than those with the non-severe disease. Derangements in plasma levels of calcium and phosphate are common in children with pneumonia, and these abnormalities occur independently of low vitamin D levels.

Keywords: Calcium, Children, Phosphate, Pneumonia, Vitamin D.

Introduction

Pneumonia is the inflammation of the lung parenchyma caused by micro-organisms. [1] Pneumonia is the leading cause of under-five

mortality globally, and the significant death burden lies within Asia and Sub-Saharan Africa. [2] Nigeria ranks first in Africa and second globally among the fifteen countries with the highest number of deaths from childhood pneumonia. [2]

Studies have shown that children with severe pneumonia have lower serum vitamin D levels than children without the disease. [3-5] On the contrary, the results of the few studies available in Nigeria showed no difference in the serum vitamin D levels of children with pneumonia and controls. [6, 7] The controls in one of these studies were very ill children, who had severe inflammatory diseases like meningitis and severe malaria. [6] The study conducted by Oduwole *et al*., [6] also showed that children who had complications like pleural effusion had lower serum vitamin D levels than those without similar complications. Ahmed *et al*., [7], on the other hand, did not find this dissimilarity between the patients with and those without complications. Vitamin D enhances expression of cathelicidin at barrier sites, including the respiratory epithelium.[8] Cathelicidin is an endogenous antimicrobial agent which is active against bacteria, viruses and fungi.[8,9] Children with low serum levels of vitamin D may be predisposed to developing pneumonia due to impaired innate immunity at the respiratory epithelium.

Vitamin D deficiency leads to reduced intestinal absorption of calcium. Low serum calcium level stimulates the release of parathyroid hormone (PTH), which increases bone resorption to maintain calcium levels. [10] Parathyroid hormone also reduces urinary calcium losses while promoting phosphaturia with eventual hypophosphataemia. [10] Blood calcium may, thus, be normal or reduced in vitamin D deficiency states. Haider *et al*., [11] showed that 74% of patients with severe pneumonia had a combination of low or normal calcium with hypophosphataemia. However, in the study by Oduwole *et al*., [6] calcium and phosphate levels were higher while vitamin D was lower in patients with pneumonia than in children with whom they were compared. Due to the disparities in the results between the few studies from Nigeria and those from other countries, there was a need to determine the serum levels of vitamin D, calcium and phosphate in children with pneumonia and compare with apparently healthy controls. It is hypothesized that vitamin D insufficiency (serum vitamin D less than 50 nmol/L) or vitamin D deficiency (serum vitamin D less than 30 nmol/L) may be more common among children with pneumonia. It is also hypothesized that hypocalcaemia and hypophosphataemia would be more common in children with pneumonia since calcium and phosphate metabolism are affected by blood levels of vitamin D. Therefore, this study was aimed at investigating the patterns of derangements of serum vitamin D, plasma calcium and plasma phosphate levels in children with pneumonia, determine whether these derangements vary according to the severity of pneumonia and also determine if serum calcium and serum phosphate levels occur as a result of low serum vitamin D levels.

Methods

A cross-sectional study was conducted, and subjects were recruited between November 2014 and September 2015. The study was approved by the Ethics and Research Committee of the Obafemi Awolowo University Teaching Hospitals Complex, Ile-Ife, Nigeria, where the study was conducted. Informed consent was obtained from the caregivers of the study participants. Children aged two months to five years, who presented at the Children's Emergency Room with clinical features of pneumonia were recruited into the study. Age- and sex-matched controls who did not have

history or physical examination findings to suggest respiratory tract infections were also recruited. The inclusion criteria for children with pneumonia included cough, tachypnoea, difficulty with breathing and fever. Pneumonia was confirmed by the presence of abnormal auscultatory chest signs or radiologic features of dense or fluffy opacities, patchy infiltrates or pleural effusion. Children with pneumonia were classified into those with non-severe and severe pneumonia based on the new WHO classification of severity of pneumonia. [12] Those with non-severe pneumonia had tachypnoea with or without chest in-drawing while those with severe pneumonia had in addition, one or more danger signs, which included refusal to drink or suck, persistent vomiting, severe malnutrition, convulsions, lethargy or loss of consciousness. The controls were recruited among children who came for follow-up care at the Out-patient Clinics of the Hospital, having recovered from previous acute illnesses like severe malaria, cellulitis, urinary tract infections, acute gastroenteritis or sepsis. The controls were included only if they had been discharged for more than one month at the time they were being recruited for the study and were found to be apparently healthy after history has been obtained, and physical examination conducted. The controls were also recruited among children who presented to the hospital with minor physical injuries. The controls were excluded if they had history to suggest pneumonia in the last two months. Study participants with clinical features of chronic liver diseases, chronic kidney diseases or any evidence of infection were excluded.

The diagnosis of pneumonia was made in all patients by one investigator. The principal investigator filled a proforma containing information on bio-data and symptoms of participants. All the patients and controls were examined, and anthropometric measurements, including weight and height, were taken. Chest X-rays were cross-checked by a radiologist to confirm radiological pneumonia. Children with complications of congestive cardiac failure (CCF) and pleural effusion were identified. Serum vitamin D levels were determined using GENTAUR 25-OH Vitamin D Total ELISA Kit with catalogue number GDMSKAP1791, manufactured by GENTAUR, Voortstraat, Kampenhout, Belgium. [13] Plasma calcium, phosphate and albumin were determined by endpoint spectrophotometric analysis using the Cecil CE2502 UV/Visible Spectrophotometer. Corrected (adjusted) plasma calcium was derived for study participants who had hypoalbuminaemia.[14]

The data were analyzed using version 20 of the Statistical Package for Social Sciences (SPSS) software. Some of the known risk factors for pneumonia were analyzed in the cases and controls using the Chi-Square test. The status of serum vitamin D, plasma calcium and phosphate levels were compared between subjects and controls using the Chi-Square test. A binary logistic regression analysis was done to identify the factors associated with vitamin D insufficiency, low plasma calcium, and low plasma phosphate status. Independent variables for the logistic regression analysis were some of the risk factors for pneumonia; and the patient/control status.

Results

Seventy-six patients and an equal number of age- and sex-matched controls were recruited for the study. There were 40 males and 36 females in each arm of the study. The mean age of the cases was 15.3 ± 12.4 months, while the mean age of the controls was 15.4 ± 12.6 months. The difference in the mean ages was not statistically significant ($t = 0.032$, $p = 0.974$). One (1.32%) of the patients with pneumonia died. Table I compares some of the known risk factors

for pneumonia between the cases and controls. Incomplete immunization and wasting were significantly more common among the cases than controls (χ^2 = 6.036, p = 0.01 and χ^2 = 11.527, p = 0.001 respectively).

There was no statistically significant difference between the mean of serum vitamin D in controls (57.27 ± 23.30nmol/L) and in the cases (52.72 ± 23.04nmol/L) (t = 1.190, p = 0.236). The mean values of plasma calcium in controls (2.42 ± 0.23 mmol/L) was significantly higher than the mean of 2.32 ± 0.25 mmol/L in patients (t = 2.528, p = 0.013). The mean of plasma phosphate in controls (1.80 ± 0.42 mmol/L) was significantly higher than the mean of 1.49 ± 0.42 mmol/L in patients (t = 4.594, p< 0.001).

Table I: Comparison of Risk Factors for Pneumonia between Patients and Controls

Risk Factors	Category	Cases (%)	Controls (%)	χ^2	P value
EBF	≥ 4 months	59 (48)	64 (52)	1.065	0.30
	< 4 months	17 (59)	12 (41)		
Immunization	Adequate	59 (46)	69 (54)	6.036	0.01
	Inadequate	17 (74)	6 (26)		
Cooking fuel	Gas	12 (43)	16 (57)	0.700	0.40
	Biomass	64 (52)	60 (48)		
Overcrowding	Absent	47 (46)	56 (54)	2.439	0.12
	Present	29 (59)	20 (41)		
Socioeconomic class	High	18 (38)	29 (62)	3.727	0.05
	Low	58 (55)	47 (45)		
Nutritional status	Normal	57 (44)	72 (56)	11.527	0.001
	Wasting	19 (83)	4 (17)		

EBF: exclusive breastfeeding

The mean of serum vitamin D in children with non-severe pneumonia was 62.4 nmol/L while it was 45.6 nmol/L in those with severe pneumonia. The difference in means of vitamin D between the two groups was statistically significant (t = 3.229, p = 0.002). The difference in mean values between the severe pneumonia group and controls was also statistically significant (t = 2.674, p = 0.009) while that between the non-severe pneumonia group and controls was not statistically significant (t = 1.030, p = 0.305). There was no statistically significant difference in the means of calcium and phosphate between non-severe and severe pneumonia groups.

Table II compares the status of serum vitamin D, plasma calcium and phosphate levels between the cases and controls. None of the cases or controls had hypervitaminosis D taken to be vitamin D level greater than 250 nmol/L. One case and two controls had hypercalcaemia while two cases and five controls had hyperphosphataemia. Out of the 12 cases with vitamin D deficiency (vitamin D level less than 30 nmol/L), 11 (91.7%) had severe pneumonia while only one (8.3%) had non-severe pneumonia. There were statistically significantly higher proportions of the cases than controls with low plasma calcium and low plasma phosphate (χ^2 = 11.368, p = 0.001; χ^2 = 10.059, p = 0.002 respectively). Vitamin D levels were correlated with calcium and phosphate levels. There was no significant correlation between serum vitamin D and either plasma calcium (r = 0.041, p = 0.624) or phosphate levels (r = -0.035, p = 0.672).

Table II: Comparison of the Status of Calcium, Phosphate and ALP between Patients and Controls

Laboratory parameter	Status	Cases (%)	Controls (%)	χ^2	P value
Vitamin D status	Sufficiency (≥50nmol/L)	37 (49)	36 (47)	0.026	.871
	Insufficiency (<50nm0l/L)	39 (51)	40 (53)		
	Level ≥ 30nmol/L	64 (84)	76 (0)	13.029	<.001
	Deficiency	12 (16)	0 (100)		
Calcium status	High / normal	48 (63)	66 (87)	11.368	.001
	Low	28 (37)	10 (13)		
Phosphate status	High / normal	62 (82)	74 (97)	10.059	.002
	Low	14 (18)	2 (3)		

Table III shows the binary logistic regression analysis to determine the likelihood of having vitamin D insufficiency, low plasma calcium, or low plasma phosphate. Binary logistic regression analyses were performed with the nutritional status, social class, immunization status and study status (whether a study participant is a case or control), as the variables to determine the likelihood of a subject having low calcium, low phosphate or low vitamin D levels. The risks of having low plasma calcium or low plasma phosphate levels were 4.366 and 7.355 times, respectively higher in the cases than in controls. Therefore, pneumonia was independently associated with low plasma calcium and low plasma phosphate, whereas pneumonia was not independently associated with vitamin D insufficiency.

Factors such as the age group, sex, social class and nutritional status were compared between children who had normal levels of serum vitamin D, plasma calcium or plasma phosphate and those with low levels. There was no statistically significant association among these variables except for the age group and plasma calcium levels. Fifteen (18.5%) of the 81 infants had hypocalcaemia compared with 23 (32.4%) of the 71 older children ($\chi^2 = 3.885$, $p = 0.049$).

Table III: Logistic Regression Analysis to Determine Factors Associated with Low Calcium, Low Phosphate, and Low Alkaline Phosphatase

	Vitamin D insufficiency			Low Plasma Calcium			Low Plasma Phosphate		
Parameter	Odds ratio	95% C.I.	p-value	Odds ratio	95% C.I.	p-value	Odds ratio	95% C.I.	p-value
Study status									
Control	Ref			Ref			Ref		
Patient	0.860	0.426-1.735	.67	4.366	1.851-10.295	**.001**	7.355	1.545-35.027	**.01**
Nutritional status									
Normal	Ref			Ref			Ref		
Undernourished	0.862	0.333-2.233	.76	1.457	0.496-4.279	.49	0.925	0.252-3.396	.91
Immunization									
Complete	Ref			Ref			Ref		
None/incomplete	1.434	0.557-3.696	.46	0.810	0.276-2.383	.70	2.052	0.592-7.105	.26
Social class									
High	Ref			Ref			Ref		
Low	1.508	0.729-	.27	0.795	0.338-	.60	0.922	0.260-	.90

	3.119	1.871	3.275

Ref = Reference Category

Discussion

The present study investigated derangements in serum vitamin D, plasma calcium and plasma phosphate levels in childhood pneumonia. The present study did not find a significant difference in the mean of serum vitamin D between the cases and controls. Neither was low serum vitamin D a risk factor for the development of pneumonia. However, there was a significant difference in the mean serum levels of vitamin D between the cases with non-severe and those with severe pneumonia. There was a statistically significant difference in the mean plasma calcium and phosphate levels between the cases and controls.

The trend in the serum vitamin D levels among the cases and controls is similar to what other investigators from Nigeria have found. [6, 7] Similar to what was found in the present study, Oduwole *et al.* [6] and Ahmed *et al.* [7] did not find any significant difference between serum vitamin D levels of the cases and controls. The results of a study done in Canada and another conducted in China were also similar to that of the present study. On the contrary, a study conducted in Egypt by Albanna *et al.* [15] showed a significantly higher mean serum level of vitamin D levels among controls than in the cases. Therefore, the results of the present study are in support of the findings of many other studies in which the overall mean serum vitamin D levels of children with various categories of severity of pneumonia did not differ significantly from the mean serum level of the controls. This is because serum vitamin D values of many of the cases with less severe pneumonia were comparable with serum vitamin D values of the controls. Vitamin D deficiency has been reported to be uncommon in Nigeria due to the abundance of sunlight exposure which is the major source of vitamin D. [16] It was reported in the study by Albanna *et al.* [15] in Egypt, that children with pneumonia had little exposure to sunlight.

Children with severe pneumonia had been shown to have significantly lower levels of serum vitamin D when compared those with non-severe pneumonia. [3, 4, 17] Investigators in Canada [3] and Japan [17] found that the mean serum vitamin D levels in children with acute lower respiratory tract infections (ALRTI) who required intensive care, was lower than the mean levels in the controls and among the cases who did not require intensive care. Wayse *et al.* [4] studied only children with severe pneumonia and reported that the mean serum vitamin D levels in them were significantly lower than the mean serum levels in the controls. The association of low serum vitamin D levels with severe pneumonia may be related to the impairment of air flow and difficulty with clearing of secretions along the airways due to soft ribs, which occurs in children with low levels of vitamin D. [18] The impairment of diaphragmatic movement, which can occur due to soft ribs in children with vitamin D deficiency, may also predispose children to severe forms of pneumonia. [18] Therefore, adequate levels of serum vitamin D may be a protective factor against the development of severe pneumonia.

The finding of a significant difference in the mean serum vitamin D levels between children with non-severe and those with severe pneumonia in the present study was in contrast to what was reported by Ahmed *et al* [7] in Abuja, in which there was no significant

difference in the means of serum vitamin D levels between children with severe and less severe pneumonia. The difference between the findings in the present study and the Abuja study may be partly explained by the differences in the methods used in assessing the severity of pneumonia in the two studies. While the present study assessed the severity of pneumonia using the WHO new classification of the severity of pneumonia, the study in Abuja identified only four children with complications as those with severe pneumonia. The larger number of patients with severe pneumonia in the present study, when compared with the study in Abuja, might have also contributed to the detection of the significant difference in the mean serum vitamin D levels between the two categories of severity of pneumonia.

The present study showed that children with pneumonia had lower levels of plasma calcium and phosphate when compared with the controls. Haider *et al.* [11] showed that hypophosphataemia with low plasma calcium levels was common in children with pneumonia. In contrast, a study by Oduwole *et al.* [6] showed that children with pneumonia had higher plasma calcium and phosphate levels than ill patients, with other diagnoses other than pneumonia, with whom they were compared. Since there was no correlation between serum vitamin D levels and either plasma calcium or plasma phosphate levels in the present study, other factors apart from low serum vitamin D levels probably accounted for the lower levels of plasma calcium and phosphate in the children. Reduced dietary intake in children with pneumonia could reduce calcium or phosphate levels, and this may occur even when serum vitamin D levels are normal. The depletion of plasma calcium and phosphate in pneumonia may, therefore, be due to impairment in any of the multi-systemic homeostatic mechanisms regulating the levels of the elements in the blood. Ill children, including those with pneumonia, may have reduced blood levels of calcium and phosphate regardless of their vitamin D levels. The presence of high phytates in the diets of Nigerian children has been associated with a higher prevalence of hypocalcaemia in them. [19] Dietary intake of high phytate content might have contributed to the degree of hypocalcaemia among children with pneumonia, though the controls would have been expectedly exposed to similar diets.

Conclusion

There was no statistically significant difference in the mean serum vitamin D levels between children with pneumonia and the controls. However, those with severe pneumonia had significantly lower mean serum vitamin D levels than those with non-severe pneumonia. Hypocalcaemia and hypophosphataemia occur commonly in some Nigerian children with pneumonia, independent of their vitamin D levels. The evidence for calcium and phosphate derangements should be sought in children with pneumonia. Further studies are required to determine if vitamin D supplementation, especially in children with vitamin D insufficiency, can prevent the occurrence of severe pneumonia.

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References

1. Johnson AW, Abdulkarim AA. Childhood pneumonia in developing countries. Afr J Resp Med 2013; 8: 4-9.

2. Liu L, Oza S, Hogan D, Perin J, Rudan I, Lawn JE, et al. Global, regional, and national causes of child mortality in 2000-13, with projections to inform post-2015 priorities: an updated systematic analysis. Lancet 2014; 385: 430-440.

3. McNally J, Leis K, Matheson L, Karuananyake C, Sankaran K, Rosenberg A. Vitamin D deficiency in young children with severe acute lower respiratory infection. Pediatr Pulmonol 2009; 44: 981-988.

4. Wayse V, Yousafzai A, Mogale K, Filteau S. Association of subclinical vitamin D deficiency with severe acute lower respiratory infections in Indian children under-5 y. Eur J Clin Nutr 2004; 58: 563-567.

5. Ren J, Sun B, Miao P, Feng X. Correlation between serum vitamin D level and severity of community-acquired pneumonia in young children. Zhongguo Dang Dai Er Ke Za Zhi 2013; 15: 519-521.

6. Oduwole AO, Renner JK, Disu E, Ibitoye E, Emokpae E. Relationship between vitamin D levels and outcome of pneumonia in children. West Afr J Med 2010; 29: 373-378.

7. Ahmed P, Babaniyi IB, Yusuf KK, Dodd C, Langdon G, Steinhoff M, *et al*. Vitamin D status and hospitalisation for childhood acute lower respiratory tract infections in Nigeria. Paediatr Int Child Health 2015; 35: 151-156.

8. Adams JS, Hewison M. Unexpected actions of vitamin D: new perspectives on the regulation of innate and adaptive immunity. Nat Clin Pract Endocrinol Metab 2008; 4: 80-90.

9. Durr H, Sudheendra US, Ramamoorthy A. LL-37, the only human member of the cathelicidin family of antimicrobial peptides. Biochim Biophys Acta 2006; 1758: 1408-1425.

10. Ulasi TO, Ebenebe J. Nutritional disorders in childhood. In: Azubuike JC, Nkanginieme KE (Editors). Paediatrics and Child Health in a Tropical Region. 2nd ed. Owerri: African Educational Services; 2007. p. 250-67.

11. Haider N, Nagi AM, Khan KM. Frequency of nutritional rickets in children admitted with severe pneumonia. J Pak Med Assoc 2010; 60: 729-732.

12. World Health Organization. Revised WHO classification and treatment of childhood pneumonia at health facilities. Evidence summaries. In: https://apps.who.int/iris/handle/10665/137319. (Accessed on April 8, 2019)

13. Vitamin D ELISA kit. In: www.gentaur.com (Accessed on January 15, 2015).

14. Payne RB, Little AJ, Williams RB, Milner JR. Interpretation of serum calcium in patients with abnormal serum proteins. Br Med J 1973; 4: 643-646.

15. Albanna EAM, Ali YF, Reayid AM. Vitamin D and LL-37 in children with pneumonia. Egypt J Paediatr Allergy Immunol 2010; 8: 81-86.

16. Pfitzner MA, Thacher TD, Pettifor JM, Zoakah AI, Lawson JO, Isichei CO, *et al*. Absence of vitamin D deficiency in young Nigerian children. J Pediatr 1998; 133: 740-744.

17. Inamo Y, Hasegawa M, Saito K, Hayashi R, Ishikawa T, Yoshino Y, *et al*. Serum vitamin D concentrations and associated severity of acute lower respiratory tract infections in

Japanese hospitalized children. Pediatr Int 2011; 53: 199-201.

18. Akpede GO, Ekanem EE, Thacher TD. Nutritional and non-nutritional rickets in the tropics and subtropics. In: Azubuike JC, Nkanginieme KE (Editors). Paediatrics and Child Health in a Tropical Region. 2nd ed. Owerri: African Educational Services; 2007. p. 684-96.

19. Thacher TD, Aliu O, Griffin IJ, Pam SD, O'Brien KO, Imade GE, *et al*. Meals and dephytinization affect calcium and zinc absorption in Nigerian children with rickets. J Nutr 2009; 139: 926-932.