ORIGINAL ARTICLE

Evaluation of the relationship between vitamin D levels and prevalence of urinary tract infections in children

H. Sherkatolabbasieh[1], M. Firouzi[1], S. Shafizadeh[2] and M. Nekohid[3]

1) *Department of Pediatrics,* 2) *Internal Medicine Department, Faculty of Medicine and* 3) *Student Research Committee, Lorestan University of Medical Sciences, Khorramabad, Iran*

Abstract

Urinary tract infection (UTI) is associated with increased health burden among paediatric patients. Vitamin D is known for its immunoregulatory effects, particularly antimicrobial activity. The aim of this study was to determine the relationship between vitamin D levels and UTIs in children referred to a hospital in Khorramabad, Iran. This case–control study was conducted on 258 children aged between 2 and 14 years; 44 children with UTI and 214 healthy children were enrolled. Patients were tested for UTI on the basis of signs and symptoms, and urine culture and analysis. Vitamin D levels were measured in children in both groups. According to the results, the two groups were significantly different in terms of sex (p 0.007). There was no significant difference between the mean vitamin D among the two groups. Vitamin D levels were not related to UTI by multivariate logistic regression. The relationship between the level of vitamin D and the incidence of UTI in children in accordance with age and sex had an odds ratio of 0.99, indicating that for a 1-unit increase in vitamin D, the odds of having a UTI decreased by 1, a correlation which was not statistically significant. According to the findings of this study, no significant correlation was found between UTI and vitamin D levels. However, the prevalence of UTI was higher in girls than boys.

Keywords: Antimicrobial activity, children, immunoregulator, paediatric, urinary tract infection, vitamin D
Original Submission: 5 May 2020; **Revised Submission:** 29 June 2020; **Accepted:** 1 July 2020
Article published online: 7 July 2020

Corresponding author: M. Firouzi, Department of Pediatrics, Faculty of Medicine, Lorestan University of Medical Sciences, Khorramabad, 6813833946, Iran.
E-mail: dr.maj.firouzi@gmail.com

Introduction

Urinary tract infection (UTI) is the most common genitourinary tract disease and the second most common bacterial infection (after respiratory tract infections) in childhood. Gram-negative bacteria are the most common cause of UTIs that may affect the upper or lower urinary tract [1].

UTIs occur in 3% to 5% of girls and 1% of boys [2]. Studies have shown that the prevalence of UTIs is greater than that of bacterial meningitis, bacterial pneumonia, middle ear infection and bacteraemia [3]. During infancy, 5% of febrile girls and 20% of uncircumcised febrile boys present with UTI. Additionally, 80% to 90% of UTI are characterized by *Escherichia coli* infection [1].

Vitamin D plays an important role in regulating inflammation and chemokine production, and it has long been known for its antimicrobial properties [4]. Vitamin D receptors are widely expressed in immune cells such as B and T lymphocytes, monocytes and dendritic cells, where it exerts immunomodulatory effects. Circulating vitamin D has a direct effect on macrophages, enhancing its oxidative ability, including the synthesis and production of cytokines, phosphatase and hydrogen peroxide [5]. Vitamin D also accelerates neutrophil motility and phagocytic activity [6,7].

During bacterial infections, macrophages convert 25-hydroxyvitamin D (25(OH)D) into circulating vitamin D3 (1,25(OH)2D3), where it modulates the gene expression of antimicrobial peptides [8]. These peptides play a key role in the

https://doi.org/10.1016/j.nmni.2020.100728

body's defense against the microbial pathogens [9]. It enhances both immune response and bacteria clearance.

Several studies have revealed a link between vitamin D deficiency and UTIs. Nonetheless, numerous controversies are reported in this regard [10–12]. Therefore, we aimed to determine the relationship between vitamin D levels and UTIs in children referred to Shahid Madani and Shahid Rahimi Hospital in Khorramabad.

Methods

This case–control study was performed on 258 children, aged between 2 and 14 years. It included 44 children with UTI and 214 healthy children who were referred to our centre for routine checkups. Patients with no risk of UTI, clinical signs and symptoms (presenting signs and symptoms such as dysuria, haematuria, abdominal and back pain, increased frequency of urination with rectal temperature >39°C), positive urine culture, pyuria, no malnutrition or obesity, not receiving multivitamins or vitamin D and no renal dysfunction were included in the study.

Patients with immunodeficiency, urinary anomalies, recurrent UTIs and interfering factors of vitamin D were excluded. Patients underwent urinalysis and urine culture for the diagnosis of infection. Urine samples were collected by clean catch (for toilet-trained children) and sterile urine bag or urethral catheterization (for small children). Urine samples were refrigerated if the analysis was not performed within 10 minutes of receipt.

All procedures performed in this study involving human participants were in accordance with the ethical standards of the institutional and/or national research committee, and with the 1964 Helsinki Declaration and its later amendments or comparable ethical standards. Consent to participate for those under 16 years old was provided by a parent or legal guardian.

Cultures with more than 100 000 colonies of per millilitre, along with the presence of symptoms in the patient, were considered indicative of a UTI (in cases of midstream sampling). On the basis of the diagnostic outcomes, patients were divided into two groups: patients and controls. Vitamin D levels were measured by high-performance liquid chromatography (Agilent 1100 series HPLC; Agilent Technologies, Santa Clara, CA, USA). The data of each patient were recorded on the questionnaire, and the relationship between the incidence of UTI and levels of vitamin D was evaluated.

Relevant interfering factors include severe haemolysis, which leads to a false increase in vitamin D; dihydroxylated metabolites of vitamin D, which interfere with test results (cross-reaction); corticosteroids, which decrease vitamin D absorption by reducing absorption; and cholesterol-lowering drugs such as cholestyramine, which can reduce vitamin D and other fat-soluble vitamins.

The data were collected and recorded in SPSS 22 (IBM, Armonk, NY, USA) for statistical analysis. Central indexes and distribution were determined for the data and independent *t* test performed; logistic regression and Mann-Whitney test were used for the analysis and comparison of nonnormalized parameters between the two groups. A significance level of 5% was considered statistically meaningful ($p < 0.05$).

Results

Of 258 subjects enrolled onto the study, 44 were diagnosed with UTI. The mean age of the children in the patient group was 6.58 ± 1.57 years and in the healthy control group was 6.7 ± 1.58 years. The difference between the mean age of the two groups was not statistically significant ($p = 0.64$) according to independent *t* tests. Therefore, these two groups are similar in age, which was important in the study process.

In the patient group, 15 patients (34.1%) were male and 29 female (65.9%), and in the healthy controls, 123 (57.5%) were male and 91 female (42.5%). Significant differences were observed between the two groups in terms of sex (p 0.007) (Table 1). The prevalence of UTI was higher in female subjects than male subjects, such that female sex can be seen as a risk factor of infection. (Fig. 1)

TABLE 1. Descriptive statistics for variables

		Case[a] (n=44)	Control (n=214)	P-value
Age (Years)		6.580 (1.570)	6.700 (1.580)	0.64
Vitamin D3 (ng/ml)		29.20 (12.27)	32.04 (15.04)	0.24
Sex	Male	34.10	57.47	0.007
	Female	65.90	42.53	

[a]mean (SD) for continuous and % for categorical variables.

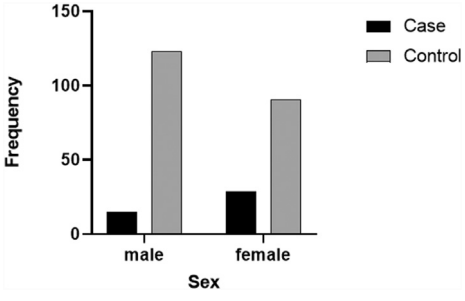

FIG 1. The prevalence of urinary tract infection was higher in females than males.

The mean vitamin D level in the patient group was reported to be 29.2 ± 12.27 nmol/L, whereas, in the control group it was 32.04 ± 15.04 nmol/L. Using independent T-test, there was no significant difference between the mean levels of vitamin D among the two groups, p = 0.24

The levels of vitamin D were further assessed based on sufficient level (30 $\leq$ 25(OH) Vit D3 $\leq$ 50 nmol/L) and insufficient level (20 << 30 nmol/L). In the patient group, 19 children (43.2%) had sufficient levels of vitamin D and in control group 101 (47.2%) children had vitamin D sufficiency. Whereas, 25 (56.8%) patients in the patient group and 113 (52.8%) in healthy control group had low levels of vitamin D. According to Pearson Chi-square test, there was no significant relationship between urinary tract infection and vitamin D level (p = 0.74).

Multivariate logistic regression analysis of the association between vitamin D level, age and sex with UTI showed that children with variable age and sex had an odds ratio of 0.99 (Table 2). The results showed that with a 1-unit increase in vitamin D level, the risk of UTI decreases by 1%. These findings were also not statistically significant (p 0.384).

Given age as a constant variable, the incidence of UTI was 2.57 times higher in girls than boys, which was statistically significant (p 0.008).

Discussion

Vitamin D is an important immunomodulator and is known to provide defense against foreign invasion, which is hypothesized to be a result of system-wide expression of vitamin D receptor. Vitamin D is reported to play an important role in infectious disease and provides protective effects against drug-resistant hosts [13].

Studies conducted in recent years have shown no evidence of the association between vitamin D deficiency and increased risk of infection [14]. Apart from the role of vitamin D in bone health, vitamin D is an important factor in the effective immune system and in antimicrobial activity [15]. One issue that has been seriously considered recently is the resistance of microorganisms to common antibiotics, which appears to be an important factor in community health and treatment [16].

TABLE 2. Relationship between vitamin D level, age and sex with UTI infection by multivariate logistic regression

Variable	OR (UTI)	95% CI for OR	p
Sex			
Boy	Ref		
Girl	2.57	1.27, 5.19	0.008
Age	0.95	0.86, 1.05	0.334
Vitamin D level	0.99	0.96, 1.01	0.384

CI, confidence interval; OR, odds ratio; UTI, urinary tract infection.

Shalaby et al. [17] showed a significant association between UTI and vitamin D deficiency. Their study also found that there was a significant relationship between serum vitamin D level in the lower urinary tract and upper UTI. In a meta-analysis, Deng et al. [18] reported that vitamin D insufficiency is associated with UTI in children, with an odds ratio of 3.01. Tekin et al. [19] showed that vitamin D levels of <20 ng/mL are associated with increased risk of UTI, with an odds ratio of 3.503. Furthermore, reduction in vitamin D levels is greater in female than male patients. Vitamin D deficiency is also marked by UTI in renal transplant recipients, compared to renal transplant recipients with normal vitamin D. The antimicrobial role of vitamin D modulates the production of cathelicidin (LL-37) and β-defensin against UTI [20]. Yang et al. [21] showed that vitamin D supplementation can significantly reduce the risk of UTI among children aged 1 to 12 months.

Owing to the importance of LL-37 as an antimicrobial peptide in UTI [22], Ovunc Hacihamdioglu et al. [23] demonstrated that UTI children with low levels of vitamin D might not be able to increase urinary cathelicidin levels against *Escherichia coli*–mediated UTI. LL-37 mediates the release of chemokines and cytokines during the infection to provide immunity. With vitamin D deficiency, macrophages are unable to produce enough antimicrobial peptide for the defense and can be thereby be associated with disease pathology and severity [24,25].

However, in contrast, a study conducted by Mahyar et al. [26] in Qazvin, Iran, enrolling children aged 1 month to 12 years who presented with UTI reported that mean serum 25(OH)D levels in children with UTI is significantly higher than the control group. They concluded that vitamin D is likely to play an important role in the pathogenesis of the infection. Similarly, Katikaneni et al. [27] reported that in infants < 3 months of age receiving formula feeding, the incidence of UTI can increase up to 76% with vitamin D supplementation. It is hypothesized that excess vitamin D can lead to Albright calcinosis (nephrocalcinosis), which favours the growth of bacteria. Excessive production of vitamin D products can suppress the immune response and exacerbate UTIs [26].

Our study failed to find any meaningful association between vitamin D and UTI. Aydogmus and Demirdal [28] reported that among women with lower UTI, vitamin D deficiency was significantly associated with pelvic floor muscle strength. However, no correlation was found between UTI and vitamin D deficiency. Merrikhi et al. [12] also reported that vitamin D supplementation does not play a protective role against recurrent UTI in paediatric patients. In our study, the sample size of the two groups of our study varied greatly; discrepancies

in the findings may be a result of this limitation. Furthermore, we do not provide data on the effects of vitamin D supplementation on UTI.

Shalaby et al. [17] found that mean age was not significantly different among the UTI patient group and the control group; the two groups were matched for age, which was consistent with our study. They also reported that the statistical distribution of the two groups was not homogeneous, indicating a significant difference, which is consistent with the higher prevalence of UTIs in girls in our study.

According to Langley [29], the sex ratio for UTIs varies according to age group, with boys at a fivefold greater risk to acquire UTI in the first 3 months of infancy. The outcomes of our study indicated female sex to be a risk factor of infection.

Conclusion

According to the findings of this study, there was no significant relationship between UTI and vitamin D. The prevalence of UTI was higher in girls than boys, which is a known risk factor. In light of previous studies, clinically applicable conclusions cannot be drawn regarding the association of vitamin D and UTI, which could also be the result of unmeasured confounders in terms of seasonal variation or sex that could influence vitamin D levels, or of the fact that the number of children with UTIs in the study was not sufficiently powered to detect a difference. Future studies that are based on molecular evidence may provide better insight.

Financial disclosure

The remaining author have no financial relationships relevant to this article to disclose.

Conflict of interest

The authors deny any conflict of interest in any terms or by any means during the study.

References

[1] Leung AK, Wong AH, Leung AA, Hon KL. Urinary tract infection in children. Recent Pat Inflamm Allergy Drug Discov 2019;13:2–18.
[2] Sood A, Penna FJ, Eleswarapu S, Pucheril D, Weaver J, Wagner JC, et al. Incidence, admission rates, and economic burden of pediatric emergency department visits for urinary tract infection: data from the nationwide emergency department sample, 2006 to 2011. J Pediatr Urol 2015;11:246. e1–8.
[3] Zendehdel A, Roham M. Role of *Helicobacter pylori* infection in the manifestation of old age–related diseases. Mol Genet GenomIC Med 2020;8:e1157.
[4] Zendehdel A, Roham M. Biological evidence of the relationship between *Helicobacter pylori* and associated extragastric diseases. J Cell Biochem 2019;120:12128–40.
[5] Zendehdel A, Arefi M. Molecular evidence of role of vitamin D deficiency in various extraskeletal diseases. J Cell Biochem 2019;120:8829–40.
[6] Aghsaeifard Z, Alizadeh R, Bagheri N. Association between neutrophil gelatinase–associated lipocalin (NGAL) and iron profile in chronic renal disease. Arch Physiol Biochem 2020.
[7] Maleki-Sadeghi N, Rahmani P, Aghsaeifard Z, Heidari G. Effects of aminophylline on the levels of neutrophil gelatinase–associated lipocalin (NGAL) in asphyxiated term neonates. Arch Physiol Biochem 2020.
[8] Moradniani Mosayeb, Mirbeik-Sabzevari Zohre, Jaferian Soleiman, Shafiezadeh Shiva, Javad Ehsani Ardakani Mohammad, Mirzaee Roozbahany Mehrdad, et al. A randomized clinical trial; co levofloxacin based sequential based sequential versus triple *Helicobacter pylori* eradication. Gastroenterol Hepatol Bed Bench 2018;11:19–26.
[9] Anbari K, Firouzi M, Abbaszadeh S. Probiotics and gastrointestinal diseases: a promising complementary medicine resource for treatment of gastrointestinal disorders and diseases. J Pharm Pharmacog Res 2019;7:193–9.
[10] Hertting Olof, Holm Åsa, Lüthje Petra, Hanna Brauner, Dyrdak Robert, Fianu Jonasson Aino, et al. Vitamin D induction of the human antimicrobial peptide cathelicidin in the urinary bladder. PLoS One 2010;5:e15580.
[11] Rolf Jorde, Therese Sollid Stina, Svartberg Johan, Joakimsen Ragnar M, Grimnes Guri, Moira Y, et al. Prevention of urinary tract infections with vitamin D supplementation 20,000 IU per week for five years. Results from an RCT including 511 subjects. Infect Dis 2016;48:823–8.
[12] Merrikhi Alireza, Ziaei Elahe, Shahsanai Armindokht, Kelishadi Roya, Maghami-Mehr Asieh. Is vitamin D supplementation effective in prevention of recurrent urinary tract infections in the pediatrics? A randomized triple-masked controlled trial. Adv Biomed Res 2018;7:150.
[13] Lang PO, Aspinall R. Vitamin D status and the host resistance to infections: what it is currently (not) understood. Clin Ther 2017;39:930–45.
[14] Kearns Malcolm D, Alvarez Jessica A, Seidel Natan, Tangpricha Vin. Impact of vitamin D on infectious disease. Am J Med Sci 2015;349:245–62.
[15] Sassi F, Tamone C, D'Amelio P. Vitamin D: nutrient, hormone, and immunomodulator. Nutrients 2018;10:1656.
[16] Cantas L, Shah Syed QA, Cavaco LM, Manaia CM, Walsh F, Popowska M, et al. A brief multi-disciplinary review on antimicrobial resistance in medicine and its linkage to the global environmental microbiota. Front Microbiol 2013;4:96.
[17] Shalaby S, Handoka N, Amin R. Vitamin D deficiency is associated with urinary tract infection in children. Arch Med Sci 2018;14:115–21.
[18] Deng Qi-Fei, Han Chu, Zhu Wen, Cao Yong-Sheng. Vitamin D and urinary tract infection: a systematic review and meta-analysis. Ann Clin Lab Sci 2019;49:134–42.
[19] Tekin Mehmet, Konca Capan, Celik Velat, Almis Habip, Kahramaner Zelal, Erdemir Aydin, et al. The association between vitamin D levels and urinary tract infection in children. Horm Res Paediatr 2015;83:198–203.

[20] Young Eun Kwon, Kim Hyunwook, Jung Oh Hyung, Jung Tak Park, Han Seung Hyeok, Ryu Dong-Ryeol, et al. Vitamin D deficiency is an independent risk factor for urinary tract infections after renal transplants. Medicine 2015;94:e594.

[21] Yang Jianhuan, Chen Guangdao, Wang Dexuan, Chen Minguang, Xing Chao, Wang Bin. Low serum 25-hydroxyvitamin D level and risk of urinary tract infection in infants. Medicine (Baltimore) 2016;95: e4137.

[22] Nielsen Karen L, Dynesen Pia, Larsen Preben, Jakobsen Lotte, Andersen Paal S, Frimodt-Møller Niels. Role of urinary cathelicidin LL-37 and human beta-defensin 1 in uncomplicated *Escherichia coli* urinary tract infections. Infect Immun 2014;82:1572–8.

[23] Övünç Hacıhamdioğlu Duygu, Altun Demet, Hacıhamdioğlu Bülent, Çekmez Ferhat, Aydemir Gökhan, Kul Mustafa, et al. The association between serum 25-hydroxy vitamin D level and urine cathelicidin in children with a urinary tract infection. J Clin Res Pediatr Endocrinol 2016;8:325–9.

[24] White JH. Vitamin D signaling, infectious diseases, and regulation of innate immunity. Infect Immun 2008;76:3837–43.

[25] White JH. Vitamin D as an inducer of cathelicidin antimicrobial peptide expression: past, present and future. J Steroid Biochem Mol Biol 2010;121:234–8.

[26] Mahyar Abolfazl, Ayazi Parviz, Safari Sara, Dalirani Reza, Javadi Amir, Esmaeily Shiva. Association between vitamin D and urinary tract infection in children. Korean J Pediatr 2018;61:90–4.

[27] Katikaneni Ranjitha, Ponnapakkam Tulasi, Ponnapakkam Adharsh, Gensure Robert. Breastfeeding does not protect against urinary tract infection in the first 3 months of life, but vitamin D supplementation increases the risk by 76%. Clin Pediatr (Phila) 2009;48:750–5.

[28] Aydogmus H, Demirdal US. Vitamin D deficiency and lower urinary tract symptoms in women. Eur J Obstet Gynecol Reprod Biol 2018;228:48–52.

[29] Langley JM. Defining urinary tract infection in the critically ill child. Pediatr Crit Care Med 2005;6:S25–9.