The Egyptian Journal of Hospital Medicine (July 2022) Vol. 88, Page 3063-3068

Association of Vitamin D with Inflammatory Bowel Diseases Activity in Pediatric Patients

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ABSTRACT

Background: Inflammatory bowel disease (IBD) is a chronic inflammatory disease of the gastrointestinal tract and is divided into Crohn's disease and ulcerative colitis.

Objective: This study aimed to find the correlation between vitamin D deficiency and IBD activity in pediatric patients.

Patients and Methods: A case-control study was conducted at the Gastroenterology Clinic, Pediatric Department, Zagazig University Hospital performed on 36 subjects divided into two equal groups; (group A) was a comprehensive sample, and (group B) contained apparently healthy participants as a control group of the same age, sex, and ethnically matched to the cases in the period between March 2021 to September 2021.

Results: The Mean 1,25 dihydroxycholecalciferol (ng/l) levels in the remission and active phases were 28.18 ± 3.42 and 14.06 ± 3.92 respectively and the fecal calprotectin ranged from 50 to 257 with a mean of 118.28 ± 55.06. There was a non-statically significant correlation between Vit D and 1,25 dihydroxycholecalciferol levels in the remission and active phases. **Conclusion**: This study found that 1, 25 dihydroxycholecalciferol level was lower in patients with IBD than in healthy people.

Keywords: Vitamin D, Hypovitaminosis D, Inflammatory bowel disease.

INTRODUCTION

Inflammatory bowel diseases (IBDs), such as ulcerative colitis (UC) and Crohn's disease (CD), are chronic inflammatory diseases that involve the inflammation of the gastrointestinal tract[(1)]**.** The underlying causes of IBD are not well understood, although the current hypothesis of pathogenesis involves the interaction between environmental triggers**,** gut microbiota**,** and genetic risk factors[(2)]**.**

Vitamin D is an important micronutrient that plays a critical biological role in various processes in human tissues. Vitamin D can be obtained from dermal synthesis following ultraviolet B (UVB) exposure, dietary intake, and supplementation[(3)]**.**

Vitamin D has a role not only in bone health and calcium homeostasis but also a significant role in immune system function[(4)].

However, both intestinal epithelial cells (IEC) and a wide variety of immune cells also possess the vitamin D receptor (VDR), suggesting a role for vitamin D in the regulation of IECs and the immune system. The VDR has been identified in multiple types of immune cells including T-helper cells (CD4+), cytotoxic T-cells (CD8+), B cells, neutrophils, dendritic cells, and macrophages[(2)]**.**

Many studies have revealed a relationship between vitamin D status with the development and progression of different chronic autoimmune disorders [(5)]**.** Vitamin D is even proposed as a treatment for autoimmune diseases such as multiple sclerosis, systemic lupus erythematosus, and IBD[(6)]**.**

Recent studies have reported that vitamin D deficiency may play a role in the increased risk of malignancies in IBD. However, limited studies have been conducted about the relationship between vitamin D and IBD activity, especially in pediatric patients[(7)]**.**

This study aimed to determine the association between vitamin D levels and IBD activity in pediatric patients.

PATIENTS AND METHODS

This study included 36 children (their mean age was 9.39 ±3.03), they were divided into equally two groups; Group (A) (Control) which included 18 apparently healthy participants. Group (B) (Patients) included 18 children with inflammatory bowel disease.

Inclusion criteria: Children with documented IBD diagnosis. Age below 18 years. Not on any VD supplements.

Exclusion criteria: Children > 18 years. Missing medical records. Missing informed consent. Written Informed consent was taken from patients' parents and/or their caregivers.

Ethical consent:

The permission for the study was received from the Pediatrics Departments of Zagazig University Hospitals after the permission of the Institutional Review Board (IRB). The research was carried out in compliance with the Code of Ethics of the World Medical Association (Declaration of Helsinki) for studies involving humans.

METHODS

The medical records of the enrolled patients were retrospectively analyzed. Clinical records and laboratory test results during the active and remission phases were collected for each patient. We recorded the highest disease activity score and longest duration of remission in cases when there were multiple active phases or remission

Received: 06/02/2022
Accepted: 05/04/2022

phases in one patient. In this study, the active phase was defined as when the disease activity score (Pediatric Crohn's Disease Activity Index [PCDAI] or Pediatric Ulcerative Colitis Activity Index [PUCAI]) was 10 or more. The remission phase was defined as the period when the patient's clinical symptoms were relieved, and the disease active score remained below 10 points for more than 3 months without the use of steroids. Exacerbation of the disease and Seasonal variation were documented by direct assessment of the patients or by information from the referring physicians.

Our registry includes demographic, careful clinical examination, anthropometric measurements referred to growth chart and complete blood count (CBC), C-reactive protein (CRP) level, erythrocyte sedimentation rate (ESR), as well as levels of serum Ca and Po4, albumin, PANCA and ASCA tests. To evaluate patients' vitamin D status, the serum level of 25-hydroxyvitamin D 25[OH]D was measured using the liquid chromatography-tandem mass spectrometry in the clinical laboratory.

Patients were classified based on their serum 25[OH]D level. Vitamin D deficiency was defined as a 25[OH]D level < 20 ng/mL, insufficiency as 21–29 ng/mL, and normal or sufficient as > 30 ng/mL.10,11,12.

Statistical Analysis:

Data were collected, revised, coded, and entered into the Statistical Package for Social Science (IBM SPSS) version 20. The comparison between the two groups with qualitative data was done by using the Chi-square test and/or Fisher exact test was used instead of the Chi-square test when the expected count in any cell was found less than 5. The comparison between two independent groups with quantitative data and parametric distribution was done by using an independent t-test. The confidence interval was set to 95% and the margin of error accepted was set to 5%. So, the p-value P < 0.001 = highly significant (HS).

RESULTS

Table 1: showed that there was no statistically significant difference between the control group and the patient group regarding Age (Year), Sex, and Height (cm), and there was a highly statistically significant difference found between the control group and patient group regarding Weight (Kg) and BMI.

Table (1): Comparison between the control group (no. =18) and the patient Group (no. =18) regarding Age (Year), Sex, Weight (Kg), Height (cm), and BMI

		Control Group	Patient Group	Test value	P-value	Sig.
		No.= 18	No.= 18			
Age (Year)	Mean±SD	9.44 ± 2.81	9.33 ± 3.33	0.108•	0.914	NS
	Range	4 – 14	1 – 14			
Sex	Female	8 (44.4%)	10 (55.6%)	0.444	0.505	NS
	Male	10 (55.6%)	8 (44.4%)			
Weight (Kg)	Mean±SD	17.28 ± 4.28	31.44 ± 15.44	-3.752•	0.001	HS
	Range	12 – 25	10 – 55			
Height (Cm)	Mean±SD	137.72 ± 20.01	138.11 ± 19.23	-0.059•	0.953	NS
	Range	100 – 165	110 – 165			
BMI	Mean±SD	12.75 ± 3.49	22.04 ± 8.68	-4.209•	0.000	HS
	Range	7.88 – 20	7.41 – 33.95			

P-value >0.05: Non significant(NS); P-value <0.05: Significant(S); P-value< 0.01: highly significant(HS), *: Chi-square test, •: Independent t-test.

Table 2: showed that there was no statistically significant difference found between the control group and patient group regarding ESR (mm/hr) First and ESR (mm/hr) Second, and there was a statistically significant difference found between the control group and patient group regarding WBCS (10^3/Ul), Platelet (10^3u/L), Ca (mg/dl) and albumin (g/dl), and there was highly statistically significant difference found between the control group and patient group regarding HB (g/dl), Po4 (mg/dl) and CRP (mg/l). The Previous table shows that there was a highly statistically significant difference found between the control group and patient group regarding Vit D.

Table (2): Comparison between the control group (no. =18) and patient group (no. =18) regarding HB (g/dl), WBCS (10^3/Ul), platelets (10^3u/L), Ca (mg/dl), Po4 (mg/dl), albumin (g/dl), ESR (mm/hr) First, ESR (mm/hr) Second, CRP (mg/l) and Vit D

		Control Group	Patient Group	Test value•	P-value	Sig.
		No.= 18	No.= 18			
HB (g/dl)	**Mean ± SD**	14.85 ± 1.06	11.39 ± 1.39	8.404	0.001	HS
WBCS (10^3/Ul)	**Mean ± SD**	7.46 ± 1.29	8.99 ± 1.32	16.123	0.001	HS
Platelet (10^3u/L)	**Mean ± SD**	269.38 ± 9.33	228.50 ± 6.37	11.983	0.001	HS
Ca (mg/dl)	**Mean ± SD**	9.11 ± 0.91	8.32 ± 1.31	+2.092	0.044	S
Po4 (mg/dl)	**Mean ± SD**	3.63 ± 0.68	5.58 ± 1.26	-5.741	0.001	HS
Albumin (g/dl)	**Mean ± SD**	4.23 ± 0.74	3.61 ± 0.65	2.676	0.011	S
ESR (mm/hr) First	**Mean ± SD**	6.61 ± 1.33	8.17 ± 2.33	-2.002	0.063	NS
ESR (mm/hr) Second	**Mean ± SD**	12.72 ± 2.26	11.33 ± 2.92	0.744	0.462	NS
CRP (mg/l)	**Mean ± SD**	3.99 ± 0.99	7.82 ± 0.35	6.745	0.001	HS
Vit D	**Mean±SD**	29.61 ± 4.84	12.44 ± 3.23	9.051	0.001	HS

P-value >0.05: Non significant(NS); P-value <0.05: Significant(S); P-value< 0.01: highly significant(HS), *: Chi-square test, •: Independent t-test

Table 3: showed that the mean 1,25 dihydroxycholecalciferol level in the remission phase (ng/l) was 28.18 ± 3.42. The Mean 1,25 dihydroxycholecalciferol level in the active phase (ng/l) was 14.06 ± 3.92 and the Fecal calprotectin ranged from 50 to 257 with a mean of 118.28±55.06.

Table (3): Distribution of the studied cases according to 1,25 dihydroxycholecalciferol level in remission phase (ng/l), 1,25 dihydroxycholecalciferol level in active phase (ng/l), and Fecal calprotectin

		Patient Group
		No.= 18
1,25 dihydroxycholecalciferol level in remission phase (ng/l)	**Mean ± SD**	28.18± 3.42
1,25 dihydroxycholecalciferol level in active phase (ng/l)	**Mean ± SD**	14.06± 3.92
Fecal protectin	**Mean ± SD**	118.28±5.06

Table 4: showed that there was Non statically significant correlation between Vit D With Age (Year), Weight (Kg), Height (Cm), BMI, Highest disease activity score, longest remission duration (months), HB (g/dl), WBCS (10^3/Ul), platelets (10^3u/L), Ca (mg/dl), Po4 (mg/dl), albumin (g/dl), ESR (mm/hr) First, ESR (mm/hr) Second, CRP (mg/l), 1,25 dihydroxy cholecalciferol level in remission phase (ng/l), 1,25 dihydroxycholecalciferol level in active phase (ng/l), ASCA (IgG) and ASCA (IgA), and there was a statically significant positive correlation between Vit D and Fecal calprotectin, and there was a statically significant negative correlation between Vit D and hospitalization days (Weeks).

Table (4): Correlation between Vit D with age (Year), weight (Kg), height (Cm), BMI, highest disease activity score, longest remission duration (months), HB (g/dl), WBCS (10^3/Ul), platelet (10^3u/L), Ca (mg/dl), Po4 (mg/dl), Albumin (g/dl), ESR (mm/hr) First, ESR (mm/hr) second, CRP (mg/l), 1,25 dihydroxycholecalceferol level in remission phase (ng/l), 1,25 dihydroxycholecalceferol level in active phase (ng/l), fecal calprotectin, ASCA (IgG), ASCA (IgA) and histological data

	Vit D	
	r	**P- value**
Age (Year)	0.046	0.857
Weight (Kg)	0.090	0.724
Height (Cm)	0.331	0.180
BMI	0.100	0.694
Highest disease activity score	-0.031	0.902
Longest remission duration (months)	0.202	0.422
HB (g/dl)	0.126	0.618
WBCS (10^3/Ul)	-0.331	0.180
Platelet (10^3u/L)	0.378	0.121
Ca (mg/dl)	-0.066	0.795
Po4 (mg/dl)	0.286	0.250
Albumin (g/dl)	0.237	0.345
ESR (mm/hr) First	-0.210	0.403
ESR (mm/hr) Second	0.025	0.922
CRP (mg/l)	-0.178	0.480
1,25 dihydroxycholecalciferol level in remission phase (ng/l)	-0.019	0.940
1,25 dihydroxycholecalciferol level in active phase (ng/l)	0.179	0.478
Fecal calprotectin	**0.532***	**0.023**
ASCA (IgG)	0.140	0.579
ASCA (IgA)	0.185	0.462
Hospitalization days (Weeks)	**-0.509***	**0.031**

DISCUSSION

In our study, females were more predominant than males (55.6%; 44.4% respectively). Studies showed that although there is no distinction between genders for disease involvement, the higher occurrence among females may be due to hormonal factors, which may interfere with the expression of the disease[8]. There was a highly statistically significant difference found between the control and patient groups regarding weight (Kg) and BMI, weight loss, and non-bloody diarrhea are suggestive of Crohn's disease flare-up. Fistula formation may result in fecaluria, pneumaturia, and rectovaginal fistulas. Masses in the right lower quadrant suggest an abscess. Affected children may present with growth retardation[9].

The present study showed that there was a statistically significant decrease in Albumin level (g/dl) in the patient group than that found in the control group.

Similarly, **Pappa *et al.*[10]** reported that low serum albumin concentration was a significant independent predictor of low serum 25OHD concentration, a finding that has not been reported previously in patients with IBD, to our knowledge. The present work also showed that there was no statistically significant difference between the control and patient groups regarding ESR, there was a statistically significant difference between the control and patient groups regarding WBCS and platelets and there was a highly statistically significant difference between the control and patient groups regarding HB, Po4, and CRP as diagnosing IBD requires a combination of clinical findings, inflammatory laboratory markers, imaging findings, and endoscopic biopsies. Hematologic findings include microcytic anemia, leukocytosis, and thrombocytosis, inflammatory markers such as the erythrocyte sedimentation rate (ESR), and high-sensitivity C-reactive protein (hsCRP) are commonly elevated. In some patients, the diagnosis may require ruling out a parasitic disease like giardiasis, amebiasis, and strongyloidiasis as well as tuberculosis. [11] Complete blood picture will identify anemia and leukocytosis. Also, albumin levels can be detected.

In the current study, there was a highly statistically significant difference between the control and the patient groups regarding Vit D.

This comes in agreement with previous studies that reported a high prevalence of vitamin D deficiency in IBD[12,13].

As shown in **Table (3),** Vitamin D deficiency may be associated with an increased IBD activity score, lower sun exposure, and decreased absorption from inflammatory mucosa[14].

Ziv *et al.* [15] showed that vitamin D is involved in the immune system and inflammatory response by inhibiting the regulation of inflammatory cytokines and the differentiation of pro-inflammatory cells.

Cantorna and Mahon[16] demonstrated an experimental link between vitamin D status and severity of IBD in animals: vitamin D deficiency exacerbated the symptoms and severity of enterocolitis in interleukin-10 knockout mice, whereas supplementation with vitamin D ameliorated IBD symptoms, reduced inflammation, and improved histologic scores and mortality.

Pappa *et al.* [10] identified vitamin D deficiency in 34.6% of children and young adults with IBD. This prevalence of vitamin D deficiency is higher than has been reported previously by **Sentongo *et al.*** [12] who found that its level was higher than that among healthy New England adolescents (24.1% <15 ng/mL) [18].

Li *et al.* [19] found that patients with CD and UC had mean lower levels of 25(OH)D than did healthy populations; however, there was no significant difference in serum 25(OH)D levels between CD and UC patients. This can be explained by insufficient intake, insufficient absorption, or excessive loss of VitD in patients with IBD[20].

Li *et al.* [19] reported that the average concentration of 1,25(OH)2D3 in CD patients was significantly higher than in patients with UC.

In children, **El-Matary *et al.*** [21] found that VitD levels were lower (though not statistically significant) in UC patients than in a CD group. 25(OH)D was significantly higher in children with CD than in children with UC[22].

The mean highest disease activity score was 44.39 ± 5.93 and the mean longest remission duration (months) was 8.89 ± 2.59.

In our study, the Mean 1,25 dihydroxycholecalciferol level in the remission phase (ng/l) was 28.18 ± 3.42, and the Mean 1,25 dihydroxycholecalciferol level in the active phase (ng/l) was 14.06 ± 3.92 and the fecal calprotectin ranged from 50 to 257 with a mean of 118.28 ± 55.06.

Kim *et al.* [23] reported that the vitamin D level was higher in the remission phase than in the active phase in UC and CD. The reason for the increased vitamin D level in the remission is unclear but is probably a result of a combination of increased nutritional intake, increased outdoor activity, and reduced need for steroid therapy[24].

These results indicate that vitamin D deficiency in IBD is mostly related to disease activity rather than decreased absorption due to inflammation of the small intestine[23].

According to **Garg *et al.*** [24] fecal leukocytes (e.g., calprotectin) more closely reflect intestinal inflammation than systemic markers (e.g., CRP, ESR, and WBC). They also reported an inverse correlation between the levels of vitamin D and calprotectin in patients with IBD. These findings support the theory that serum 25(OH)D may affect local tissue inflammation.

Kim *et al.* [23] reported that patients in the active phase showed seasonal variation, with decreasing vitamin D levels in spring and winter. However, there was no significant difference by season in the remission phase, suggesting that patients in the active phase are more sensitive to seasonal factors.

Middleton *et al.* [25] found that CD patients reported taking VitD supplements in winter, and their levels of 25(OH)D were significantly higher than non-users. This further confirms the views of **Pappa *et al.*** [26] and **Grunbaum *et al.*** [27] who suggested that higher doses may yield better results.

CONCLUSION

This study found that 1, 25 dihydroxy cholecalciferol level was lower in patients with IBD than in healthy people. There was a non-statically significant correlation between Vit D with 1,25 dihydroxy cholecalciferol level in the remission phase and 1,25 dihydroxy cholecalciferol level in the active phase.

Further studies on large scale are necessary to detect the relationship between Vit D levels and IBD morbidity and mortality due to steroid use and other immunomodulator therapy.

Conflict of interest: The authors declare no conflict of interest.

Sources of funding: This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

Author contribution: Authors contributed equally to the study.

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