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Original Article

Impact of vitamin D supplementation on rotavirus disease severity and duration in Iraqi children

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ABSTRACT

In young children, rotavirus is a major cause of severe gastroenteritis. Vitamin D (calciferol) may affect the course of gastroenteritis and is essential for gut health and immunological control. To evaluate how vitamin D affects inflammation and clinical results in kids with rotavirus gastroenteritis. A prospective, non-randomized study was conducted at Al-Kadhimiya Pediatric Hospital on 60 children under 5 years with vitamin D deficiency and confirmed rotavirus infection. Participants were divided into two groups: Group II received standard treatment plus a single dosage of vitamin D, whereas Group I (control) received regular treatment. Serum vitamin D levels were assessed both at baseline and seven days later. Clinical outcomes and laboratory parameters were assessed on days 1, 4, and 7. Data were analyzed using SPSS v24. Demographic, clinical, and baseline laboratory parameters were comparable between groups. Following intervention, Group II showed significant improvements in temperature and dehydration ($P < 0.001$), while the control group showed no change. Both groups exhibited time-dependent changes in laboratory markers (CRP, MPV, NLR, PLR) ($P < 0.05$), with the vitamin D group showing more pronounced improvements ($P < 0.001$). Vitamin D supplementation in children with rotavirus infection significantly improved clinical outcomes and inflammatory markers, highlighting its potential as an adjunct therapy in pediatric rotavirus gastroenteritis.

Keywords: Clinical outcomes, Gastroenteritis, Pediatrics, Rotavirus, Vitamin D

Introduction

Rotavirus gastroenteritis (RG) is a leading cause of severe gastroenteritis in newborns and young children. They also infect the young of numerous other mammalian and bird species [1]. It has been demonstrated that rotaviral gastroenteritis accounts for about 40% of all outpatient visits to pediatric primary care practices for acute gastroenteritis in babies and young [2]. Diarrhea and its aftermath remain one of the top causes of morbidity and mortality among children, especially in developing countries. For children under the age of five, it is the second most common cause of death [3]. Compared to other viral infections that cause gastroenteritis, rotavirus produces more severe illness; children are more likely to arrive with fever, dehydration, and metabolic acidosis and need to be admitted or readmitted to the hospital [4]. Making a diagnosis with the exception of symptoms of dehydration, physical examination results for rotavirus infection are frequently normal [5].

As the severity of rotavirus gastroenteritis worsened, the mean platelet volume-to-platelet (MPV/P) decreased and the neutrophil-to-lymphocyte ratio (NLR) rose [6]. Children with acute gastroenteritis may have a high NLR, which might be a helpful indicator for the diagnosis and differential diagnosis of RV infection [7]. Additionally, it was discovered that patients with rotavirus-associated gastroenteritis have statistically significantly higher NLR and platelets to lymphocytes ratio (PLR) values [8]. When it came to the differential diagnosis of infantile diarrhea,

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serum C-reactive protein (CRP) levels were highly significant [9].

There is no specific treatment for a rotavirus infection [10-13]. Antibiotics and antivirals cannot be used to treat a rotavirus infection. The ailment often clears up in three to seven days, but staying hydrated is crucial [14]. Vitamin D (calciferol) is a fat-soluble vitamin that can be bought as a dietary supplement, added to other foods, and found naturally in some foods. Furthermore, the skin produces it naturally when ultraviolet (UV) rays from the sun begin the synthesis of vitamin D [15]. The assessment of vitamin D deficiency entails determining serum levels of the 25-hydroxy form; levels between 20 and 30 ng/mL suggest insufficiency, while values below 20 ng/mL indicate deficiency [16]. Vitamin D is most likely safe when taken at the recommended dosages [17]. Despite being uncommon, vitamin D overdose can have serious consequences that impact both long-term renal function and the short-term vital prognosis. In developing nations, vitamin D poisoning is still a concern [18]. As early as one day following 600,000 IU of D3 and 540,000 IU of D3, serum 25(OH)D concentrations greater than 30 ng/mL were noted; the greatest increases in serum 25(OH)D consistently occurred between days 1 and 30. Three and seven days after the dose, peak values were observed [19].

This study aims to assess the impact of vitamin D supplementation on the clinical outcomes, such as the severity and duration of gastroenteritis in children caused by the rotavirus. Additionally, to assess how well vitamin D reduces inflammation in children with rotavirus gastroenteritis.

Materials and Methods

Study design and setting

Currently, Al-Kadhimiya Pediatric Hospital carried out a foreseeable, non-randomized clinical study between December 2024 and May 2025.

Study population

The study included 60 patients who met the inclusion criteria and had been diagnosed with RG. Participants were chosen from the Al-Kadhimiya Pediatric Hospital's patient population. Conveniently chosen patients with a confirmed rotavirus infection were included in the current investigation. Thirty RG patients were assigned to group I, and thirty RG patients were assigned to group II of the study population:

Group I (control): received standard treatment which include rehydration therapy and antipyretic therapy [20].

Group II (Treatment) group: received vitamin D single oral dose 300000 IU for age < 1 year and dose of 600000IU for age> 1 year in addition to standard treatment [21]. The study lasted 6 months, with follow-ups at 7-10 days

Inclusion criteria

The following were the study's inclusion criteria:

1-Children with confirmed rotavirus infection by rotavirus stool examination by rotavirus rapid test from CerTest BIOTEC company in Spain.

2-Serum vitamin D levels indicating deficiency (below 20 ng/mL) [22].

Exclusion criteria

The study's exclusion criteria were as follows:

1- Kids suffering from acute dehydration, other serious illnesses, or underlying medical issues.

2- Kids who are currently taking vitamin D supplements.

Outcome measures

Primary outcome

Clinical outcomes were measured includes severity and duration of diarrhea, severity and duration of vomiting, hydration state and body temperature.

Secondary outcomes

1. Markers of inflammation and hematology: C-reactive protein (CRP), neutrophil-to-lymphocyte ratio (NLR), platelet-to-lymphocyte ratio (PLR), and mean platelet volume (MPV) were assessed in both groups on admission (day 1), day 4, and day 7. Complete blood count (CBC) data acquired at the hospital laboratory using an automated hematology analyzer were used to compute MPV, NLR, and PLR. CRP levels were determined using an agglutination method with the CRP Latex Kit (Atlas Medical Company, China).
2. Serum Vitamin D Levels: Serum vitamin D was measured at admission (day 1) and day 7. The measurements were performed using the Electrochemiluminescence (ECL) method with the serum vitamin D kit from Roche Diagnostics (Germany).

Data analysis

Data were analyzed using IBM SPSS Statistics (Version 24). Continuous variables were presented as mean ± SD and categorical variables as numbers (percentages). Normality was assessed using the Shapiro-Wilk test. Categorical variables were compared across groups using the chi-square test and within groups using McNemar's test. An independent t-test or Mann-Whitney test was used for between-group comparisons, and a paired t-test or Wilcoxon rank-sum test was used for within-group comparisons of laboratory data, severity, and duration. Changes in vitamin D levels were compared to clinical/laboratory indicators using Pearson or Spearman correlation analysis.

Results and Discussion

Demographic data of patients

The study recruited 60 pediatric patients, 30 for group I (control) and 30 for group II (treatment). 21 males (70%) and 9 females (30%) make up group I, whereas 22 males (73.3%) and 8 females (26.6%) make up group II. The patients in group I were 6.23± 6.39 months old on average. Group II patients were 1.3±0.46 months old on average. Regarding age and sex, there was no discernible difference between the two groups.

Baseline clinical characteristics of patients

According to the patients' baseline clinical parameters, there was no discernible change in the degree of dehydration (P = 0.295) or temperature status (P = 0.218) between groups I (control) and II (treatment).

Baseline lab data of the patients

Prior to the intervention, no statistically noteworthy variations were seen in any of the baseline laboratory measures between groups I (control) and II (treatment) (P > 0.05).

Effect of vitamin D supplementation on clinical characteristics.

The change of clinical characteristics between the two groups between day 1 and day 4.

In **Table 1**, the change of clinical characteristics between the two groups was compared across four clinical parameters.

Study findings showed that treatment had a statistically significant effect on all parameters (P < 0.001), including temperature and dehydration, which were not significantly changed in the control group.

Table 1. The change of clinical characteristics between the two groups between day 1 and day 4

Parameter	Group I (Control)					Group II (Treatment)				
	Day 1		Day4		P value	Day 1		Day4		P value
Temperature n (%)	Afebrile Febrile	3 (10%) 27 (90%)	Afebrile Febrile	23 (76.6%) 7 (23.3%)	0.219*	Afebrile Febrile	2 (6.66%) 28 (93.33%)	Afebrile Febrile	(26) (86.6%) (4) (13.3%)	<0.001*
Dehydration n (%)	Well Mild Moderate Severe	1 (3.3%) 4 (13.3%) 18 (60%) 7 (23.33%)	Well Mild Moderate Severe	3 (10%) 15 (50%) 11 (36.66%) 1 (2.55%)	0.5*	Well Mild Moderate Severe	1 (3.3%) 5 (16.6%) 16 (53.33%) 8 (26.66%)	Well Mild Moderate Severe	25 (83.3%) 5 (16.66%) 0 (0%) 0 (0%)	<0.001*
Diarrhea frequency (Mean± SD)	11.96± 5.76		9± 3.08		0.002#	15.13± 6.8		3.36± 2.09		<0.001#
Vomiting frequency (Mean ± SD)	3.36± 2.6		2.1± 2.3		0.001#	5.43± 4.81		0.40± 0.77		<0.001#

*Statistical analysis was done using the McNemar's test

#Statistical analysis was done using the Wilcoxon rank sum test

Association between vitamin D level change and the clinical characteristics change between day 1 and day 7

Statistical analysis revealed significant differences in all evaluated clinical parameters between the two groups [23-26]. The statistical test yielded p-values of <0.001, indicating a strong association between the intervention and improvement in these parameters. The statistical test also demonstrated significant differences with p-values of <0.001 as shown in **Table 2**.

Table 2. Association between Vitamin D Level Change and the Clinical Characteristics Change between day 1 and day 7

Parameter	Group I (Control)					Group II (Treatment)				
	Day 1		Day7		P value	Day 1		Day7		P value
Temperature n (%)	Afebrile Febrile	3 (10%) 27 (90%)	Afebrile Febrile	27 (90%) 3 (10%)	<0.001*	Afebrile Febrile	2 (6.66%) 28 (93.33%)	Afebrile Febrile	30 (100%) 0 (0%)	<0.001*
Dehydration n (%)	Well Mild Moderate Severe	1 (3.3%) 4 (13.3%) 18 (60%) 7 (23.33%)	Well Mild Moderate Severe	22 (73.33%) 8 (26.66%) 0 (0%) 0 (0%)	<0.001*	Well Mild Moderate Severe	1 (3.3%) 5 (16.6%) 16 (53.33%) 8 (26.66%)	Well Mild Moderate Severe	29 (97.45%) 0 1 (2.55%) (0%) 0 (0%)	<0.001*
Diarrhea frequency (Mean± SD)	11.96± 5.76		3.86± 2.68		<0.001#	15.13± 6.8		3.36± 2.09		<0.001#
Vomiting frequency (Mean ± SD)	3.36± 2.6		0.4± 1.32		<0.001#	5.43± 4.81		0± 0		<0.001#

*Statistical analysis was done using the McNemar's test

#Statistical analysis was done using the Wilcoxon rank sum test

Effect of vitamin D supplementation on laboratory data of the patients

Association between vitamin D level change and the laboratory change between day 1 and day 4

The results include statistical comparisons between groups I (control) and II (treatment) across four lab measures (CRP, LMR, MPV, and NLR), demonstrating the correlation between changes in vitamin D levels and changes in laboratory measures, as shown in **Table 3** [27, 28]. This is an organized interpretation:

These findings revealed that all four parameters have statistically significant changes in both control and treatment groups ($P < 0.05$), indicating strong associations with vitamin D level changes.

Table 3. Association between Vitamin D Level Change and the laboratory Change between day 1 and day 4

Parameter	Group I (Control)			Group II (Treatment)		
	Day 1	Day4	P value	Day 1	Day4	P value
CRP (Mean± SD)	32.8± 13.15	32.8± 13.15	<0.001*	32.4± 10.40	12± 5.45	<0.001#
MPV (Mean± SD)	7.29± 0.30	7.42± 0.27	<0.001*	7.24± 0.27	8.52± 0.35	<0.001#
NLR (Mean ± SD)	3.45± 0.35	3.36± 0.33	<0.001*	3.53± 0.51	2.04± 0.33	<0.001#
PLR (Mean ± SD)	297.03±74.14	296.83± 67.03	<0.001*	295.4± 87.08	198.46± 59.55	<0.001#

*Statistical analysis was done using the Paired T-test test

#Statistical analysis was done using the Wilcoxon rank sum test

The change of laboratory data between the two groups between day 1 and day 7

Table 4 shows statistical comparisons of various laboratory parameters between control and treatment groups between day 1 and day 7. Here is a summary interpretation**:**

CRP, MPV, NLR, and PLR results indicated statistically noteworthy variations ($P < 0.001$) between the control and treatment groups, indicating that both groups underwent changes over time [29-32]. Vitamin D levels showed a non-significant change in the control group ($P = 0.161$). Significant increase in the treatment group ($P < 0.001$).

Table 4. The Change of Laboratory Data between the Two Groups between day 1 and day 7

Parameter	Group I (Control)			Group II (Treatment)		
	Day 1	Day7	P value	Day 1	Day7	P value
CRP (Mean± SD)	32.8± 13.15	10.4±7.37	<0.001*	32.4± 10.40	12±10.4	<0.001#
MPV (Mean± SD)	7.29± 0.30	7.56±0.28	<0.001*	7.24± 0.27	9.72±0.28	<0.001#
NLR (Mean ± SD)	3.45± 0.35	3.25±0.35	<0.001*	3.53± 0.51	1.18±0.14	<0.001#
PLR (Mean ± SD)	297.03±74.14	291.7±67.7	<0.001*	295.4± 87.08	129.± 40.8	<0.001#
Vit D (Mean ± SD)	15.43±2.47	15.0±2.47	0.161#	15.5±2.16	39.73±8.45	<0.001#

*Statistical analysis was done using the Paired T-test test

#Statistical analysis was done using the Wilcoxon rank sum test

The correlation between clinical/laboratory variables and vitamin D level change

According to the correlation study, as shown in **Table 5**, improvements in a number of clinical and laboratory measures were substantially correlated with higher increases in vitamin D levels. Specifically, moderate negative correlations were found with diarrhea severity and duration, vomiting duration, and CRP levels, while strong negative correlations were observed with NLR and PLR, indicating reduced inflammation. Positive correlations were seen with MPV, suggesting favorable hematological changes. Vomiting frequency showed a weak, non-significant negative correlation.

Table 5. The correlation between clinical/laboratory variables and vitamin D level change

Variable	Correlation Coefficient	P-value
Diarrhea	-0.464	<0.001*
Vomiting	–0.202	0.122*
Diarrhea duration	–0.727	<0.001*
Vomit duration	–0.479	<0.001*

CRP	–0.453	<0.001*
MPV	0.748	<0.001*
NLR	–0.767	<0.001*
PLR	–0.758	<0.001*

*Statistical analysis was done using the Spearman correlation test

The study's internal validity is strengthened by the absence of significant differences in baseline demographic characteristics, ensuring that observed variations in clinical outcomes can be attributed to the intervention rather than demographic factors. The findings indicate that vitamin D supplementation reduces hospital stays and improves clinical outcomes, including severity and duration of diarrhea and vomiting, as well as degree of dehydration, independently of age or sex. These results are consistent with a 2018 randomized clinical trial in Iran reporting similar benefits of vitamin D supplementation [33].

Additionally, the current study found that vitamin D significantly affects plasma CRP and complete blood picture results which means that giving Vitamin D affects body inflammatory response. Also, it is found that giving single dose of vitamin D significantly reduces plasma CRP in participants with rotavirus gastroenteritis faster than participants that were not receiving Vitamin D. The results of a 2014 study conducted in China on the impact of vitamin D supplementation on the amount of circulating high-sensitivity C-reactive protein were consistent with these findings. Vitamin D supplements have been shown to have a significant favorable impact on circulating hs-CRP levels, especially in those whose baseline hs-CRP levels were ≥5 mg/L [34]. Pro-inflammatory cytokines like IL-6 and TNF-α, which cause the liver to produce more CRP, are downregulated, while anti-inflammatory T-regulatory cell activity is increased to lessen excessive immune responses [35].

According to the current study, vitamin D dramatically lowers NLR. This is similar to a 2015 study conducted in Iran on the effects of high-dose vitamin D supplementation on the distribution of the neutrophil-to-lymphocyte ratio (NLR) and the level of high-sensitivity C-reactive protein, which discovered that both NLR and hs-CRP sharply dropped after high-dose vitamin D treatment [36].

According to our research, vitamin D dramatically lowers NLR in kids who have rotavirus infections. A 2014 study in Turkey evaluated the association between vitamin D and inflammation using the neutrophil-to-lymphocyte ratio and the platelet-to-lymphocyte ratio. In this study, it was shown that the platelets-to-lymphocytes ratio (PLR) was an independent predictor of 25(OH)D levels. Patients with lower 25(OH)D levels also had considerably greater PLR and NLR. Furthermore, the study used these widely accessible and reasonably priced markers to show a negative relationship between inflammation and vitamin D levels [8]. This effect happens by directly influencing platelet behaviour via vitamin D receptors, which can decrease platelet activation and aggregation in inflammatory states [37]. This was consistent with a 2022 investigation on the connection between vitamin D deficiency and inflammatory biomarkers determined from hemograms in children that was carried out in Turkey [38]. Pro-inflammatory cytokines like IL-6 and TNF-α, which cause the liver to produce more CRP, are downregulated, while anti-inflammatory T-regulatory cell activity is increased to lessen excessive immune responses [35].

This study demonstrated that rotavirus infection decreases MPV in pediatric patients, and Vitamin D has a significant positive effect on MPV.

Vitamin D may play a role in raising MPV through its anti-inflammatory effect. This agrees with a study conducted in Romania in 2022 on the relationship between vitamin D levels and platelet parameters in children with viral respiratory infections [39].

The elevation in mean platelet volume (MPV) noted during recovery from rotavirus gastroenteritis likely indicates the resolution of inflammation and the restoration of bone marrow function. In the acute phase of infection, elevated inflammatory cytokines and oxidative stress can inhibit megakaryocyte maturation, leading to the production of smaller platelets and lower MPV values. As the infection diminishes and systemic inflammation abates, megakaryocyte function is restored, leading to the release of larger, more mature platelets into the bloodstream. The increase in MPV throughout the recovery period can thus be seen as an indicator of enhanced immunological modulation and hematological restoration in children recuperating from rotavirus gastroenteritis [40].

The current study has several limitations, such as being a single-center study, having a small sample size because of the study time limit, only looking at one vitamin D regimen (stoss therapy), not evaluating the different dosing methods (daily, weekly, IM vs. oral), possibly not capturing the long-term effects of vitamin D on immunity or rotavirus infection recurrence, and not being able to measure vitamin D levels at day 4.

Conclusion

The current study concluded that vitamin D improved clinical outcomes of rotavirus patients including severity of diarrhea and vomiting, duration of diarrhea and vomiting, temperature, and severity of dehydration. Additionally, vitamin D improved laboratory investigation including CRP, MPV, NLR, and PLR.

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Conflict of interest: None

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Ethics statement: Under clearance number REC06202507R, the current study's proposal was submitted to the "College of Pharmacy, University of Baghdad" and approved by the "Scientific and Ethical Committee" on May 1, 2025. In addition, approval of the Ministry of Health was obtained according to approval number (120624) in 31-12-2024. While patients' consent was obtained verbally. This study was registered in clinical library of medicine in (clicicaltrialtrials.gov) according to approval number (NCT07167797) in 2025-09-19, also signed informed consent had been taken from the parents of all participants.

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