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The Link between Vitamin D and Irritable Bowel Syndrome

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Abstract

Background: Irritable bowel syndrome (IBS) and other functional gastrointestinal disorders are common and can significantly affect quality of life. This study aimed to evaluate the efficacy of vitamin D3 supplementation in treating IBS symptoms using the IBS Symptom Severity Score (SSS), and to assess the relationship between serum 25-hydroxyvitamin D (25-OH vitamin D) levels and IBS. **Methods:** This observational cross-sectional study included 130 participants aged ≥18 years attending the Internal Medicine outpatient clinic at Benha University Hospitals. They were divided into two equal groups: Group A (65 IBS patients) and Group B (65 healthy controls). Blood samples were collected to measure serum 25-OH vitamin D levels. IBS patients were also assessed using the IBS-SSS. **Results:** Vitamin D deficiency was significantly more prevalent in the IBS group (38.46%) compared to the control group (21.54%) (P = 0.012). Among IBS subtypes, the irritable bowel syndrome with constipation (IBS-C) showed the highest prevalence of deficiency (78.46%) compared to irritable bowel syndrome with diarrhea (IBS-D) (13.48%) and irritable bowel syndrome with mixed bowel habits (IBS-M) (7.59%). Following vitamin D supplementation, the IBS group demonstrated a significant increase in vitamin D levels and a marked decrease in SSS scores (P < 0.001 for both). No significant correlations were found between vitamin D levels and age, BMI, hemoglobin, WBCs, platelets, liver enzymes, creatinine, or urea. However, vitamin D levels were inversely correlated with SSS (r = -0.196, P = 0.025). **Conclusion:** Vitamin D supplementation alleviates IBS symptoms. IBS patients tend to have lower serum vitamin D levels, with an inverse relationship between vitamin D levels and symptom severity.

Keywords: Irritable Bowel Syndrome; Link; Symptoms Severity Score; Vitamin D

Introduction

The most prevalent functional gastrointestinal disorder is irritable bowel syndrome (IBS). IBS is prevalent in 5-10% of the global population. Diarrhea, constipation, bloating, and stomach pain are just some of the symptoms that these patients may encounter. Irritable bowel syndrome has a major influence on patients' quality of life [(1)].

There are four primary forms of irritable bowel syndrome: discomfort, mixed bowel habits, unclear bowel habits, and irritable bowel syndrome unclassified or unsubtyped (IBS-U). Several factors contribute to the development of irritable bowel syndrome, including the gut-brain axis, visceral hypersensitivity, stress, and the gastrointestinal habitat. Factors that increase the likelihood of developing IBS include inflammation, antibiotic use, and bacterial infections. Research is now showing that the gut microbiota plays a significant role in the development of irritable bowel syndrome (2).

The pathophysiology is yet not fully understood. Treatment options for IBS are limited and targeted toward symptomatic relief. The available treatment options are rarely successful in long-term control of symptoms and quality of life improvement (3).

Most individuals suffering from IBS also have vitamin D deficiency, according to several research. Parathyroid, calcium, and phosphate hormone pathways all include vitamin D as a hormone contributor. The type of vitamin D that is most abundant in the blood is 25-hydroxyvitamin D, or 25(OH)D. Here is the format for evaluating vitamin D levels in clinical practice [(4).]

Sub-clinical vitamin D deficiency affects about 80-85% of the population of Arab countries (5). A variety of diseases, such as inflammatory bowel diseases (IBD), diabetes, cardiovascular diseases, cancer, and obesity, have been strongly associated with vitamin D deficiency in numerous recent studies [(6).] A diet high in fatty foods like fatty fish and dairy products is one way to acquire vitamin D, but body can also make it when exposed to sunlight (7).

Many studies have demonstrated that a considerable number of individuals with IBS are vitamin D deficient, despite the absence of a plethora of evidence connecting the two. The primary objectives of this investigation are to determine the vitamin D levels of IBS patients and evaluate the effectiveness of vitamin D supplementation in ameliorating symptoms three months after the commencement of treatments (1, 8).

Patients and methods

This observational cross-sectional study included 130 individuals aged ≥ 18 years, 65 of them meeting Rome IV criteria for IBS and the other 65 are

control group with no IBS symptoms, attending the Internal Medicine outpatient clinic- done at Benha University Hospitals over a period from January 2023 to October 2023.

Each patient provided written consent that was informed. Each patient was informed of the purposes of the study and allocated a secret code number. The investigation was conducted with the consent of the investigation Ethics Committee at the Faculty of Medicine at Benha University.

Rome IV defined IBS as a functional bowel disorder in which recurrent abdominal pain on average at least 1 day/week in the last 3 months, associated with two or more of the following criteria; related to defecation, associated with a change in the frequency of stool and associated with a change in the form (appearance) of stool (9).

Exclusion criteria were patient < 18 years, patients with known organic gastrointestinal disorders, patients with systemic diseases associated with any treatment with steroids, vitamin D or calcium and pregnant or lactating females, chronic inflammatory diseases, parathyroid disorders, parasitic infestations, anxiety, and neurosis.

Grouping: Patients were divided into two equal groups: **Group A (n = 65):** IBS group and **group B (n = 65):** control group without IBS symptoms.

All participants were subjected to assessment of serum vitamin D levels by obtaining CBC, ALT, AST, serum creatinine, urea, and blood samples. The enzyme-linked immunoassay technique was employed to measure the 25-hydroxyvitamin D (25(OH)D) level at the commencement of the trial. Vitamin D levels are explicated as follows: (Deficient: < 20 ng/ml, Insufficient: 20 - < 30 ng/ml, Sufficient: 30-100 ng/ml) (10).

The Irritable Bowel Severity Scoring System (IBS-SSS), developed by Francis et al. (11), it is a 5-item self-administered questionnaire. The first three questions ask patients with IBS to self-assess the frequency and intensity of abdominal pain and abdominal distension experienced in the last 10 days preceding the completion of the questionnaire. The other two questions require patients to self-assess their overall satisfaction with their bowel habits and the degree to which IBS interferes in their daily routine. The score for each question ranges from 0-100, except for the question that refers to the number of days that patients experience abdominal pain which is scored from 0-10. However, for the calculation of the final score, the score of the abovementioned question is also converted to a 0–100-point score, by multiplying the answer given by ten. In all questions, extreme values indicate the absence (0) or the highest value (one hundred) of the variable investigated. The final score of IBS-SSS ranges from 0-500. According to the creators of the

questionnaire, IBS-SSS can discriminate IBS patients from healthy volunteers, and concurrently stratify IBS patients into four severity classes, based on their final score. The first class indicates quiescent IBS and includes total IBS-SSS scores <75; the second class indicates mild IBS and includes scores that range from 75-175; the third class indicates moderate IBS and includes scores that range from 175-300; and the fourth class indicates severe IBS including scores that exceed 300 (12).

Blood collection and storage

A screened tube was used to prevent the influence of light, while a conventional tube was used to collect blood samples. The serum aliquots were prepared and stored at −20 °C in screened and standard containers for use in subsequent analyses. The samples were centrifuged immediately after collection. Each patient acquired a vitamin D supplement (50,000 IU/week) that was prescribed for individuals with inadequate vitamin D levels. A follow-up visit was conducted on these subjects three months after the initial diagnosis.

Enzyme-Linked Immunosorbent Assay technique:

In order to evaluate 25(OH) vitamin D levels, human sera or plasma (50 µl) were subjected to immuno-purification and subsequently quantified using the Enzyme-Linked Immunosorbent Assay (Human Vitamin D3,VD3 ELISA KIT unit 96 T and 48T Bioassay England/China Cat# catalogue number (E1546Hu)). Briefly, delipidated samples were obtained by treating sera or plasma samples with magnesium chloride and dextran sulfate solutions. The vitamin D-binding protein inhibitors and monoclonal antibodies to 25(OH)D attached to solid-phase particles were used to extract the analytes from immunocapsules (50 µl sample/capsule). The samples were then spun in a centrifuge.

Following three rinses with deionized water, the immunocapsules were subjected to 90 minutes of agitation on a rocker-shaker at temperatures ranging from 18 to 25 °C. Following the incorporation of 25(OH)D biotin solution, the samples were subjected to 60 minutes of incubation on an orbital shaker set to 500-750 rpm and maintained at 18-25 °C. After 30 minutes, the samples were analyzed at 450 nm using the Synergy H1-BioTek microplate reader.

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Statistical analysis

The statistical analyses were conducted using SPSS v27, a program produced by IBM (Armonk, NY, USA). Histograms and the Shapiro-Wilks test were used to see if the data had a normal distribution. An unpaired student t-test was used to assess the quantitative parametric data that was acquired by employing the standard deviation (SD) and mean. The Mann Whitney test was utilized to assess

the quantitative non-parametric data points of the median and interquartile range (IQR) (13). When comparing the means of two populations, particularly those that are linked, the paired sample t-test is the way to go. The findings of the study of qualitative variables were presented using percentages and frequencies and were analyzed by chi-squared tests and Fisher's exact tests (14). Statistical significance was determined using a two-tailed p-value (P value) less than 0.05. Pearson correlation analysis was used to ascertain the extent of correlation between two numerical variables (15).

Results

The baseline characteristics, which encompassed age, sex, weight, height, body mass index (BMI), marital status, and occupation, as well as the laboratory investigations, which included hemoglobin (Hb), white blood cells (WBCs), platelets, ALT, AST, creatinine, and urea, did not exhibit any significant differences between the two groups. **Table 1**

After treatment, the vitamin D level had significantly increased in comparison to the pretreatment level (P<0.001). Vit D levels were significantly different between the two groups prior to treatment. The Vit D level was significantly lower in the IBS group than in the patients without IBS symptoms group.

The IBS group exhibited a higher prevalence of vitamin D deficiency (38.46%) than the control group (21.54%) (P = 0.012).

IBS-C subtype (78.46%) was more prevalent in terms of vitamin D deficiency than IBS-D subtype (13.48%) and IBS-M subtype (7.59%). **Table 2**

The level of vitamin D in the IBS group was significantly higher after treatment than it was prior to treatment (P<0.001). The level of IBS-SSS after treatment was markedly improved (decreased) in comparison to the level before treatment (P<0.001). **Table 3**

The symptoms severity score after treatment was significantly improved (decreased) compared to the level before treatment (P<0.001). **Table 4**

A significant negative correlation was observed between Vitamin D levels and IBS-SSS (r=-0.196, P=0.025). Vitamin D levels were not significantly correlated with other parameters, such as age, BMI, Hb, WBCs, platelets, ALT, AST, creatinine, and urea. **Table 5**

The multivariate logistic regression analysis showed that only symptoms severity score and vitamin D level before treatment were significant predictors of IBS.

Table 1: Baseline characteristics and laboratory investigations of the studied patients

		Total (n=130)	IBS group (n = 65)	Control group without IBS symptoms (n = 65)	P value
Age (years)	**Mean± SD**	41.62± 10.95	42.85± 11.04	40.4± 10.8	0.204
	Range	25-60	25-60	26-58	
Sex	**Male**	67 (51.54%)	37 (56.92%)	30 (46.15%)	0.219
	Female	63 (48.46%)	28 (43.08%)	35 (53.85%)	
Weight (Kg)	**Mean± SD**	80.35± 11.05	79.34± 10.9	81.35± 11.18	0.300
	Range	59-98	60-97	59-98	
Height (m)	**Mean± SD**	1.67± 0.05	1.66± 0.05	1.67± 0.04	0.124
	Range	1.59-1.74	1.6-1.75	1.59-1.74	
BMI (Kg/m^2)	**Mean± SD**	28.94± 4.14	28.78± 4.04	29.11± 4.27	0.651
	Range	20.05-37.89	20.76-36.96	20.05-37.89	
Marital status	**Single**	59 (45.38%)	32 (49.23%)	27 (41.54%)	0.378
	Married	71 (54.62%)	33 (50.77%)	38 (58.46%)	
Occupation	**Working**	90 (69.23%)	44 (67.69%)	46 (70.77%)	0.703
	Not working	40 (30.77%)	21 (32.31%)	19 (29.23%)	
Hb (gm/dL)	**Mean± SD**	13.09± 0.93	13.11± 0.94	13.07± 0.91	0.828
	Range	11.5-14.7	11.5-14.7	11.6-14.5	
WBCs ($\times 10^9$/L)	**Mean± SD**	7.85± 1.95	7.75± 1.94	7.95± 1.98	0.571
	Range	4.5-11.2	4.6-11.1	4.5-11.2	
Platelets ($\times 10^9$/L)	**Mean± SD**	297.11± 27.51	294.43± 26.96	299.78± 28.01	0.269
	Range	250-340	256-340	250-339	
ALT (U/L)	**Mean± SD**	29.2± 6.54	28.92± 6.1	29.48± 6.99	0.631
	Range	18-41	19-41	18-40	
AST (U/L)	**Mean± SD**	29.38± 6.85	28.83± 6.7	29.92± 7	0.365
	Range	18-42	18-42	19-40	
Creatinine (mg/dL)	**Mean± SD**	1.09± 0.18	1.07± 0.17	1.11± 0.19	0.249
	Range	0.8-1.4	0.9-1.4	0.8-1.4	
Urea (mg/dL)	**Mean± SD**	37.52± 11.77	37.55± 11.25	37.49± 12.36	0.976
	Range	20-60	20-60	20-59	

Data presents as mean ± SD or frequency (%). BMI: body mass index, Hb: Hemoglobin, WBCs: white blood cells, ALT: Alanine transaminase, AST: aspartate aminotransferase.

Table 2: Vit D level and distribution of different IBS types of the studied patients

		Total (n=130)		IBS group (n = 65)	Control group Without IBS symptoms group. (n = 65)	P value
Vit D before treatment	**Deficient**	39 (30%)		25 (38.46%)	14 (21.54%)	**0.012***
	Insufficient	50 (38.46%)		27 (41.54%)	23 (35.38%)	
	Sufficient	41 (31.54%)		13 (20.0%)	28 (43.08%)	
		Before	**After**	--	--	--
Vit D (ng/mL)	**Mean± SD**	35.37± 25.32	41.44± 21.95	--	--	--
	Range	10-100	16-100	--	--	--
	Median (IQR)	25 (18 - 44.5)	34 (27 - 46.75)	--	--	--
	P value	**<0.001***		--	--	--
IBS	**IBS-C**	51(78.46%)		--	--	--
	IBS-D	9(13.48%)		--	--	--
	IBS-M	5 (7.69%)		--	--	--

Data presents as mean ± SD, range, or frequency (%). IQR: interquartile range, IBS: Irritable bowel syndrome, *: statistically significant as p value <0.05.

Table 3: Vit D, symptoms severity score of the studied patients regarding IBS symptoms

		IBS group (n = 65)		P value
		Before	**After**	
Vit D (ng/mL)	**Mean± SD**	31.52± 23.91	37.6± 20.14	**<0.001***
	Range	10-96	16-96	
	Median (IQR)	24 (17 – 29)	31 (24 – 39)	
SSS	**Mean± SD**	323.95± 112.9	136.85± 66.41	**<0.001***
	Range	109-490	13-319	
	Median (IQR)	342 (240- 424)	126 (93 – 184)	

Data presents as mean ± SD, range, or frequency (%). IBS: Irritable bowel syndrome, IBS-QOL: Irritable bowel syndrome-quality of life. SSS: Symptoms severity score, *: statistically significant as p value <0.05.

Table 4: Correlation between Vit D level and the other variables

	Vit D (ng/mL)	
	r	**p**
Symptoms severity score	-0.196	**0.025***
Age (years)	-0.018	0.835
BMI (Kg/m^2)	-0.031	0.728
Hb (gm/dL)	-0.033	0.705
WBCs (× 10^9/L)	0.089	0.309
Platelets (× 10^9/L)	-0.069	0.432
ALT (U/L)	0.011	0.896
AST (U/L)	0.032	0.716
Creatinine (mg/dL)	-0.092	0.298
Urea (mg/dL)	-0.017	0.850

Data presents as mean ± SD or frequency (%). BMI: body mass index, Hb: Haemoglobin, WBCs: white blood cells, ALT: Alanine transaminase, AST: aspartate aminotransferase. *: statistically significant as p value <0.05.

Table 5: Multivariate logistic regression analysis for prediction of IBS

Variable	Odds ratio	95% CI	P
Age (years)	1.0269	0.9892 to 1.0660	0.165
Sex	1.4416	0.6508 to 3.1936	0.367
BMI (Kg/m2)	0.9837	0.8908 to 1.0864	0.746
Hb (gm/dL)	0.9472	0.6101 to 1.4703	0.809
WBCs (× 10^9/L)	0.9226	0.7469 to 1.1396	0.455
Platelets (× 10^9/L)	0.9903	0.9751 to 1.0056	0.213
ALT (U/L)	0.9814	0.9211 to 1.0456	0.561
AST (U/L)	0.9755	0.9194 to 1.0350	0.411
Creatinine (mg/dL)	0.193	0.0197 to 1.8878	0.157
Urea (mg/dL)	0.9915	0.9579 to 1.0263	0.628
Symptoms severity score before treatment	1.0056	1.0018 to 1.0095	**0.004***
Vit D level before treatment (ng/mL)	0.9769	0.9594 to 0.9947	**0.011***

BMI: body mass index, Hb: Hemoglobin, WBCs: white blood cells, ALT: Alanine transaminase, AST: aspartate aminotransferase.

Discussion

At present, researchers are conducting research to ascertain the precise role of vitamin D deficiency in IBS, although this role has not yet been fully established. New evidence has aroused scientists' interest in whether high-dose oral vitamin D supplementation can alleviate diarrhea-predominant irritable bowel syndrome. The treatment not only resolved the IBS symptoms but also the anxiety and depression that were associated with them. As a result, there has been a surge of medical blogs [16].

The current study showed that, before treatment, 39 (30%) patients had vit D deficiency, 50 (38.46%) patients had vit D insufficiency and 41 (31.54%) patients had vit D sufficiency, and the level ranged from 10 to 100 ng/mL with a mean of 35.37± 25.32 ng/mL. After treatment, vit D ranged from 16 to 100 ng/mL with a mean of 41.44± 21.95 ng/mL. Vit D level had significantly increased after treatment compared to before treatment (P<0.001). Vitamin D was significantly different between both groups. Vit D was significantly lower in IBS group compared to control group without IBS symptoms group.

These results are supported by Khayyat and Attar al., (17) who revealed results with statistical significance. At the outset, the average serum 25(OH)D level for the control group was 31+16 nmol/L, while the average for the IBS group was 21+12 nmol/L. Consequently, vitamin D insufficiency was common (82% in the IBS group).

On explanation of the current findings, Sprake et al. (18), the psychosocial behavioral patterns of individuals with irritable bowel syndrome (IBS) were revealed, including their frequent avoidance of vitamin D and calcium-rich foods like fortified milk. Based on these findings, it was concluded that the observed social behaviors could be caused by an altered vitamin D absorption mechanism. The study also found a correlation between active diarrhea symptoms and IBS, suggesting a potential treatment response.

Vitamin D enhances the activity of natural killer cells while inhibiting the

immune response by type-1 helper T cells, as shown experimentally; this is in addition to the fact that the immune response is altered in irritable bowel syndrome, which is a major contributing factor (19).

Vitamin D deficiency is common among people who suffer from irritable bowel syndrome, according to a large body of research. Irritable bowel syndrome symptoms like these could have something to do with vitamin D levels, how they develop, or both. Irritable bowel syndrome is impacted by inflammation.

These results underscore the necessity of conducting additional research on vitamin D supplementation as a potential treatment for IBS. Further, there is increasing evidence that vitamin D may affect a variety of pathways associated with IBS, such as the regulation of intestinal permeability, the release of antimicrobial peptides, the regulation of microbiota, the regulation of immune response, inflammation, and the interference of gut-brain communication. IBS is primarily caused by intestinal inflammation; however, vitamin D can alleviate this inflammation by increasing T regulatory cells and suppressing Th1/Th17 cells. The immunomodulatory effect of vitamin D is demonstrated by its regulation of intestinal epithelial cell integrity and enhancement of antimicrobial peptide secretion.

Gut health is dependent on antimicrobial peptides, which are essential for the regulation of microbiota, the preservation of barrier integrity, and the regulation of immune responses. Inflammation is exacerbated by dysbiosis and impaired intestinal barrier function in IBS. Adenosine monophosphates (AMPs) engage in the regulation of microbial homeostasis and the reduction of pathogenic proliferation, and cathelicidins and defensins are examples of their activity. Through the VDR, vitamin D encourages the production of AMP, which in turn enhances gastrointestinal health. Achieving adequate vitamin D intake may alleviate symptoms of IBS by reestablishing intestinal barrier function, reducing inflammation, and rebalancing the microbiota [(19).]

In the current study, 51(78.46%) patients had IBS-C, 9(13.48%) patients had IBS-D, and 5 (7.69%) patients had IBS-M.

Further, this agreed with Elhosseiny et al., [(20)] the results indicated that 26.6% of the participants had IBS-D, while 73.4% had IBS-C, which was characterized by constipation.

On the contrary with Dorn et al., (21) IBS-D was the most prevalent IBS subtype, with 46% of cases, followed by IBS-C (32%), and IBS-M (29%).

The current study revealed that the vitamin D level in the IBS group was significantly higher after therapy than it was prior to it ($P<0.001$). When comparing the level of IBS-SSS before and after treatment, a significant improvement (reduction) was noted ($P<0.001$). The current study also discovered a negative link between vitamin D levels and IBS-SSS, with a p-value of 0.025 and a correlation coefficient of -0.196.

In accordance with the current study, Huang et al., (22) it was emphasized that the use of supplemental vitamin D significantly enhances the quality of life and alleviates the symptoms of IBS.

Additionally, Yan et al., [23] Vitamin D supplementation may be advantageous for individuals with IBS in order to mitigate gastrointestinal symptoms, as was declared.

Further, Jalili et al., [24] in terms of IBS-SSS, reported a significant difference ($P < 0.05$).

Furthermore, a significant reduction in IBS-SSS scores ($P < 0.001$) was reported in Abbasnezhad et al. (25) in Iran, participants were divided into two groups and given 50,000 IU twice a week for six months as part of the intervention.

Moreover, Amrousy et al. (26) Once again, after six months of delivering a lower daily dose of 2000 IU, there was a notable improvement in IBS-SSS ($P < 0.001$).

Also, Bin et al., [27] They supported the current study by identifying data from twelve clinical trials that included IBS patients. Vitamin D levels in the serum of patients with IBS are low. The quality of life, or QOL score, is improved by vitamin D supplementation.

On the other hand, Williams et al., (8) asserted that the quality of life and the severity of symptoms in individuals with IBS are not influenced by vitamin D supplementation.

Alongside the current study, Qi et al., [28] Nine randomized controlled trials were conducted to investigate the hypothesis that vitamin D supplementation enhances the IBS-SSS in adolescents and young adults with irritable bowel syndrome. The total number of participants in this study was 780. Irritable bowel syndrome symptoms and elevated serum 25(OH)D levels were alleviated in young adults and adolescents with IBS-SSS when vitamin D supplementation was administered. To enhance the IBS severity scoring system (IBS-SSS), a statistically significant shift in blood 25(OH)D levels was utilized, with a means difference of 11.29 and a confidence interval of 7.13–15.45. The hazards ratio was 2.34 and the confidence interval was 1.56-3.50. There was a negligible probability of adverse events (RR0.49, 95% CI 0.35-0.69). The p-value is less than 0.05, which means that the results cannot be considered non-significant.

However, these findings are in inconsistent with Abboud et al., (29) using a cross-sectional design, researchers in Lebanon looked at a sample of adults to see if their serum vitamin D levels were associated with IBS symptoms. On average, out of 230 participants, they found a serum vitamin D level of 17.53 (12.40) ng/mL and a Birmingham IBS Symptom Questionnaire score of 16.98 (15.16). In the case of constipation and diarrhea, the scores on the pain subscale were 20.75 (23.63), 25.06 (29.99), and 9.88 (13.37), respectively. For all four regression analyses, the total score and subscales were unrelated to serum vitamin D levels ($p > 0.05$). It is possible that ethnic and genetic factors explain the variation from the present study.

A small sample size, a single-center design, a brief follow-up period, and a hot climate all contributed to a lack of statistical power in the analysis. The findings were also less applicable to a broader population. As a result, people tend to wear cultural garments that shield them from direct sunlight.

Conclusion

Individuals with irritable bowel syndrome have inadequate vitamin D levels. Significant in the development of irritable bowel syndrome. IBS is predicted by vitamin D levels, which are inversely correlated. Irritable bowel syndrome symptoms may be alleviated by taking a vitamin D supplement.

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