# **Original Article**

# Evaluation of Clinical Significance of Vitamin D3 Status in Hospitalized Cases of Acute Febrile Illness with Thrombocytopenia in North-West Zone of Rajasthan

Bal Kishan Gupta<sup>1</sup>, Nitesh Kumar Garwa<sup>2</sup>, Shyam Lal Meena<sup>3</sup>, Jigyasa Gupta<sup>4</sup>, Gurmeet Singh<sup>5</sup>, Suryalakshmi Kala C Thankom<sup>6</sup>

<sup>1</sup>Senior Professor, <sup>2,5,6</sup>Senior Resident, <sup>3</sup>Associate Professor, <sup>4</sup>Junior Medical Officer, Department of General Medicine, SP Medical College and Associated Group of Hospitals, Bikaner, Rajasthan, India DOI:10.37821/ruhsjhs.8.3.2023.612

## **ABSTRACT**

**Introduction:** Thrombocytopenia is an important haematological manifestation of acute febrile illness caused by a variety of infections. Besides the role of vitamin D3 in bone homeostasis, its significance in infections, inflammation, and immune response has also been documented. This study aimed to evaluate clinical significance of vitamin D3 status in patients of acute febrile illness with thrombocytopenia.

**Methodology:** The present cross-sectional study was conducted on 400 patients of fever with thrombocytopenia admitted between May 2022 to October 2022. Laboratory evaluation including vitamin D3, complete blood count (CBC), renal function test (RFT), liver function test (LFT), blood sugar, lactate dehydragenase (LDH), rapid diagnostic test for malaria, peripheral blood smear (PBS) for malaria parasite (MP), test for dengue, HBsAg, *HCV*, *HIV*, blood culture, D dimer, test for scrub typhus and leptospirosis, ultrasonography, and chest X-ray was also done in all the cases.

**Results:** Out of 400 patients, 227 were males (mean age 5.46±15.63 years) and 173 females (mean age 39.75±17.36 years). Dengue fever was found to be the commonest cause (29.5%) followed by malaria, COVID-19 infection (0.75%), and one case each of *HIV* and *Hepatitis B*. Low level of vitamin D3 was found in 73.5% cases as compared to 17.5% in control subjects (p <0.001). Vitamin D3 status was found low in most of the patients irrespective of age group (p>0.2089), residence (p>0.97), comorbidities (p>0.248), and etiology (27.70±14.0, 27.45±11.96, 34.13±6.36, 28.79±26.04, respectively for dengue, malaria, COVID-19, and undiagnosed; p>0.609). Longer

duration of illness at the time of hospitalization (p<0.02) and longer duration of hospital stay (p<0.04) was associated with lower vitamin D3 status. Low D3 status was associated with increased incidence of bleeding manifestations (p<0.05) and more requirement of platelet transfusion (p<0.017).

**Conclusion:** Acute febrile illness patients should be investigated for platelet count irrespective of the bleeding manifestations as decreased platelet count could be severe without external manifestations and could be an indicator of bad prognosis.

**Keywords:** Acute febrile illness, Dengue, Malaria, Thrombocytopenia.

#### INTRODUCTION

Thrombocytopenia is an important haematological manifestation of acute febrile illness. The normal range of platelet count is 1,50,000-4,00,000/µl. Thrombocytopenia is defined as a platelet count below the lower normal limit (<1,50,000/μl). This may be due to decreased production, increased destruction (immunogenic or non-immunogenic), and increased sequestration by spleen. Infections of varying types (viral, parasitic, bacterial) like dengue, malaria, leptospirosis, typhoid, miliary tuberculosis, HIV, and others have been associated with thrombocytopenia. 1-3 Acute febrile illness associated with thrombocytopenia accounts for most outpatient visits and in-patient admissions in India and the causes for the same are variable and need a systematic approach to identify the etiology and appropriate therapy. This can be potentially fatal if the etiology is not recognized and if not appropriately treated early.

Vitamin D is synthesized from cholesterol and has hormonal activity. Vitamin D has some important

metabolic effects such as regulation of calcium and phosphorus homeostasis, bone mineralization, enhancing immune system, regulating cell division and differentiation, regulating coagulation, and decreasing inflammation.4 Vitamin D3 enhances antimicrobial activities of macrophages and monocytes in an autocrine fashion via VDR-RXR signalling, which in turn stimulates the production of endogenous antimicrobial Cathelicidin LL-37.5 Cathelicidin acts against invading bacteria and fungi by destabilizing microbial membranes. Autophagy induced by vitamin D signalling in macrophages is dependent on 1,25 D-stimulated expression of branchedchain aminotransferase (BCAT1)<sup>7</sup>, the enzyme that initiates the catabolism of BCAAs; the effect of 1,25 D on autophagy was eliminated in cells in which BCAT1 had been ablated<sup>8</sup>, thereby implicating enhanced BCAA catabolism in 1,25 D-regulated autophagy in macrophages. Collectively, the aforementioned results provide a mechanistic rationale for adequate vitamin D levels promoting widespread antimicrobial defence. Although few studies and case reports related with significance of vitamin D in malaria, dengue, and acute and chronic immune thrombocytopenia have been undertaken but we could not find any study earlier in the literature reporting significance of vitamin D3 in cases of acute febrile illness with thrombocytopenia. Therefore, this study was planned to evaluate clinical correlation of vitamin D3 status in admitted patients of acute febrile illness with thrombocytopenia.

## **METHODS**

This cross-sectional study was carried out on 400 cases of acute febrile illness with thrombocytopenia admitted in the Department of Medicine between May 2022 to October 2022 to evaluate clinical significance of vitamin D3 status. Ethics committee approval was taken before starting the study (letter no. EC/SPM/ECA/07), all participating subjects were explained about the study, and informed consent was taken. Age more than 15 years presenting with fever and thrombocytopenia were included. Patients with thrombocytopenia without fever, chronic liver disease, hematological disorders, idiopathic and drug induced thrombocytopenia were excluded.

All patients were evaluated as per proforma including detailed clinical history and physical examination. Laboratory evaluation including complete blood count (CBC), renal function test (RFT), liver function test (LFT), blood sugar, lactate dehydraogenase (LDH), rapid

diagnostic test for malaria, peripheral blood smear (PBS) for malaria parasite (MP), test for dengue, HBsAg, Anti *HCV*, *HIV*, blood culture, D dimer, test for scrub typhus and leptospirosis, ultrasonography, and chest X-ray was done in all cases. Vitamin D3 assay was done in all the cases by immunoassay method using Unicel dxI 800 machine (Beckman Coulter). Vitamin D3 deficiency is classified according to vitamin D3 levels as per the following recent criteria: deficient (<20 ng/ml), insufficient (20-30 ng/ml), and sufficient (>30 ng/ml). Levels of 25(OH)D were classified into three categories as per US Endocrine Society (2011)<sup>9</sup> criteria as deficient (≤20 ng/ml), insufficient (21-30 ng/ml), and sufficient (>30 ng/ml).

In the present study, low level of 25(OH)D (Hypovitaminosis D) indicates levels ≤30 ng/ml. Complete blood count was carried out using Mindray 6-part cell counter. Thrombocytopenia was defined as total platelet count <1.5 lac/mm³ and severity were graded as per NCICTCAE grading¹⁰ into mild (<1,50,000-75,000/mm³), moderate (<75,000-50,000/mm³), severe (<50,000-25,000/mm³), and life threatening (<25,000/mm³). All the patients were treated as per guidelines. Platelet transfusion was done when indicated. All patients were followed up till discharge.

Statistical analysis was performed using statistical package for the social sciences (IBM SPSS Statistics for Windows, Version 22.0. Armonk, NY: IBM Corp.). Chi-square test was done for qualitative variables and Student t-test was used for quantitative variables. Pearson's correlation test was done to evaluate correlation between D3 level and platelet count and multiple linear regression analysis was done to predict the effect of vitamin D3 in relation to various variables. p<0.05 was considered as statistically significant.

# **RESULTS**

Out of 400 patients, 227 were males (age ranging 15-87 years, mean  $35.46 \pm 15.63$ ) and 173 females (age ranging 15-92 years, mean  $39.75\pm 17.36$ ). On etiological differential diagnosis, dengue fever was found to be the commonest cause in this study (118, 29.5%) followed by malaria (15%, 59 vivax and one falciparum), 3 patients (0.75%) were diagnosed with COVID-19 infection, and one each with *HIV* and *Hepatitis B* while in 54.25% of the cases (n=217) definite etiological diagnosis could not be ascertained.

Table 1: Vitamin D3 status in relation to different epidemiological parameters

Variable	No. of		Vitamin D3 status, n (%)						
	cases	< 20	20-30	>30	<30	Mean±SD	•		
	(%)	ng/ml	ng/ml	ng/ml	ng/ml				
Age (years)									
15-30	171 (42.7)	65 (38.01)	63 (36.84)	43 (25.15)	128 (74.85)	26.89±11.5	p>0.2089		
31-40	87 (21.75)	32 (36.78)	28 (32.18)	27 (31.03)	60 (68.97)	$27.42\pm12.6$			
41-50	56 (14.00)	20 (35.71)	21 (37.5)	15 (26.79)	41 (73.21)	$27.55 \pm 12.1$			
51-60	45 (11.25)	16 (35.56)	16 (35.56)	13 (29.89)	32 (71.11)	27.84±13.9			
>60	41 (10.25)	17 (41.46)	16 (39.02)	8 (19.51)	33 (80.49)	$27.25 \pm 12.1$			
Gender									
Male	227 (56.7)	64 (28.19)	87 (38.33)	76 (33.48)	151 (66.52)	26.56±10.6	p<0.0004		
Female	173 (43.2)	86 (49.71)	57 (32.95)	30 (17.34)	143 (83.24)	21.56±10.5			
Residence									
Urban	182 (45.5)	65 (35.71)	68 (37.36)	49 (26.92)	133 (73.08)	25.20±11.1	p>0.97		
Rural	218 (54.5)	85 (38.99)	76 (34.86)	57 (26.15)	161 (73.85)	25.24±11.0			
Comorbidities									
DM	28 (7.00)	10 (35.71)	6 (21.43)	12 (42.86)	16 (57.14)	26.80±11.2	p>0.248		
HT	38 (9.05)	9 (23.68)	12 (31.58)	17 (44.74)	21 (55.26)	26.54±11.2			
COPD	8 (2.00)	4 (50)	1 (12.5)	3 (37.5)	5 (62.5)	26.26±11.0			
None	337 (84.2)	127 (37.69)	125 (37.0)	74 (21.96)	252 (74.78)	27.26±11.1			
Etiology									
Dengue	118 (29.5)	50 (42.37)	43 (36.44)	26 (22.03)	93 (78.81)	27.70±14.0	p>0.609		
Malaria	60 (15.00)	26 (43.33)	23 (38.33)	11 (18.33)	49 (81.67)	27.45±11.9			
COVID-19	3 (0.75)	1 (33.33)	1 (33.33)	1 (33.33)	2 (66.67)	34.13±6.36			
Undiagnosed	219 (54.7)	73 (33.33)	77 (35.16)	68 (31.05)	151 (68.95)	$28.79\pm26.0$			
Total	400 (100)	150 (37.5)	144 (36)	106 (26.5)	294 (73.5)	25.19±11.1			

p value<.05: Significant, p value<0.001: Highly significant; DM: Diabetes mellitus; COPD: Chronic obstructive pulmonary disease.

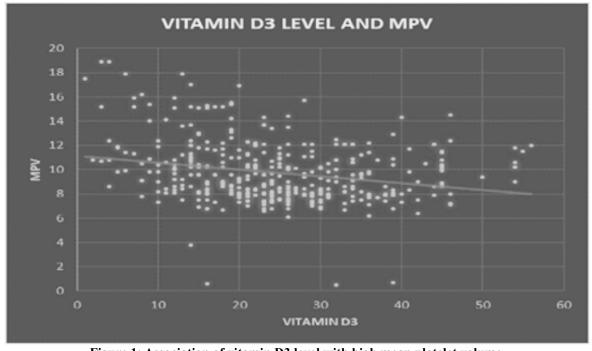


Figure 1: Association of vitamin D3 level with high mean platelet volume.

Table 2: Vitamin D3 status in relation to different laboratory parameters

Variable	No of	Vitamin D3 status, n (%)							
	cases (%)	<20 ng/ml	20-30 ng/ml	>30 ng/ml	<30 ng/ml	Mean±SD			
Anemia									
Yes (M)	146 (64.30)	41 (28.08)	53 (36.30)	52 (35.62)	94 (64.38)	$25.19\pm11.21$	p>0.943		
No (M)	81 (35.70)	18 (22.22)	31 (38.27)	32 (39.50)	49 (60.50)	$25.30 \pm 11.20$			
Yes (F)	100 (57.80)	43 (43.00)	33 (33.00)	24 (24.00)	76 (76.00)	$25.26 \pm 11.60$	p>0.9458		
No (F)	73 (42.20)	39 (53.42)	23 (31.50)	11 (15.1)	62 (84.9)	$25.14 \pm 11.24$			
TLC									
<4000	209 (52.25)	81 (38.75)	70 (33.49)	58 (27.7)	151 (72.3)	25.21±11.18	p>0.692		
4000-11000	24 (6.0)	56 (33.8)	59 (35.32)	52 (31.14)	115 (68.86)	25.22±11.23	•		
>11000	93 (23.25)	4 (16.67)	12 (50.0)	8 (33.33)	16 (66.67)	25.00±11.21			
MPV									
<8	107 (26.75)	24 (22.43)	51 (47.66)	32 (29.91)	75 (70.09)	26.26±9.13	p<.00001		
8-15	271 (67.75)	111 (40.96)	88 (32.47)	72 (26.57)	199 (73.43)	24.16±11.29	1		
>15	22 (5.5)	15 (68.18)	5 (22.73)	2 (9.09)	20 (90.91)	18.23±10.99			
Blood urea	. ,	, ,	, ,	,	, ,				
<45	323 (80.75)	124 (38.39)	114 (35.29)	85 (26.32)	238 (73.68)	25.26±11.60	p>0.876		
>45	77 (19.25)	26 (33.77)	30 (38.96)	21(27.27)	56 (72.73)	25.14±11.24	p 0.070		
Mean±SD	,, (13,120)	33.26±18.44	37.05±20.45	34.79±17.65	35.12±19.51	2011 11,2			
Serum creatin	ine								
<1.4	339 (84.75)	129 (38.05)	122 (35.99)	88 (25.96)	251 (74.04)	25.20±11.17	p>0.928		
>1.4	61 (15.25)	21 (34.43)	22 (36.07)	18 (29.51)	43 (70.49)	25.03±11.25	p* 0.520		
Mean±SD	01 (13.23)	1.03±0.72	1.09±1.11	1.01±0.45	1.1.06±0.93	25.05-11.25			
SGOT		1.03-0.72	1.07=1.11	1.01-0.12	1.1.00=0.75				
>40	81 (17.50)	25 (30.86)	32 (39.51)	24 (29.63)	57 (70.37)	25.54±11.43	p>.252		
>40	319 (82.50)	125 (39.18)	112 (35.11)	82 (25.71)	237 (74.29)	24.11±10.71	p>.232		
Mean±SD	319 (82.30)	124.57±173.28	145.46±238.18	110.08±130.46	134.8±207.52	24.11±10.71			
		124.57±175.26	143.40±236.16	110.06±130.40	134.6±207.32				
SGPT	(0 (17)	22 (22 25)	27 (20 71)	10 (27 04)	40 (72 06)	26 20 - 11 05	0. 422		
<35	68 (17)	22 (32.35)	27 (39.71)	19 (27.94)	49 (72.06)	26.38±11.95	p>0.423		
>35	332 (83)	128 (38.55)	117 (35.24)	87 (26.20)	245 (73.80)	23.99±10.59			
Mean±SD		110.51±221.87	125.48±473.21	92.22±106.56	117.82±366.59				
LDH									
<420	230 (57.5)	86 (37.39)	85 (36.70)	59 (25.65)	171 (74.35)	27.27±11.19	p>0.620		
>420	170 (42.5)	64 (37.65)	59 (34.71)	47 (27.65))	123 (72.35)	27.03±11.06			
Mean±SD		409.14±239.29	393.55±182.08	413.89±177.12	402.42±212.95				

M: Male; F: Female; MPV: Mean platelet volume; SGOT: Serum glutamic oxaloacetic transaminase; SGPT: Serum glutamic pyruvic transaminase; LDH: Lactate dehydrogenase; TLC: Total leucocyte count; p value<0.05: Significant; p value<0.001: Highly significant.

Vitamin D3 status in relation to different epidemiological parameters are shown in table 1. A significantly low status of vitamin D3 was found among study participants. Mean level of vitamin D3 in male patients was  $26.56\pm10.64$  ng/ml (ranging 1-56 ng/ml) while in female patients it was  $21.56\pm10.51$  ng/ml (ranging 2-54 ng/ml) (p <0.0004). 83.24% females had low levels of vitamin D3 (< 30 ng/ml) as compared to 66.52% males. Vitamin D3 status was

found low in most of the patients irrespective of age group (p>0.2089), residence (p>0.97), comorbidities (p>0.248), and etiology (27.70±14.0 ng/ml, 27.45±11.96 ng/ml, 34.13±6.36 ng/ml, 28.79±26.04 ng/ml, respectively for dengue, malaria, COVID-19, and undiagnosed; p>0.609).

Although the mean level of vitamin D3 was not significantly different in abnormal laboratory parameters like renal function test, liver function test, haemoglobin,

Table 3: Vitamin D3 status in relation to different clinical parameters

Variable	No. of		Vitamin D3 st	atus, n (%)		p value	
	cases (%)	< 20 ng/ml	20-30 ng/ml		<30 ng/ml	Mean±SD	•
<b>Duration of</b>							
illness (days)							
1-5	282 (70.50)	110 (39.01)	94 (33.33)	78 (28.66)	204 (72.34)	25.58±11.11	p<0.024
6-10	114 (28.50)	38 (33.33)	49 (42.98)	27 (35.96)	87 (76.32)	$24.06\pm10.19$	-
>10	4 (01.00)	2 (50)	1(25)	1 (25)	3 (75)	21.25±13.33	
Bleeding	` ′	` /	` /	` '	` '		
manifestation							
Yes	17 (4.25)	8 (47.06)	8 (47.06)	1 (5.88)	16 (94.12)	$19.52\pm7.67$	p<0.04
No	383 (95.75)	142 (37.08)	136 (35.51)	105 (27.42)	278 (72.58)	25.11±11.36	1
Severity of	, ,	,	,	` /	` /		
thrombocytopenia							
Mild	76 (19.00)	18 (23.68)	22 (28.95)	36 (47.37)	40 (52.63)	$26.38\pm9.73$	p<0.007
Moderate	87 (21.75)	26 (29.89)	30 (34.48)	31 (35.63)	56 (64.37)	$26.51\pm11.48$	•
Severe	180 (45.00)	46 (25.56)	63 (35.00)	71 (39.44)	109 (60.56)	23.87±10.47	
Life	57 (14.25)	15 (26.32)	34 (59.65)	8 (14.04)	49 (85.96)	21.33±11.57	
threatening	` /	` ′	` ,	, ,	, ,		
Duration of hospital							
stay day							
<3	205 (51.25)	63 (30.73)	81(39.51)	61 (29.76)	144 (70.24)	$25.71\pm10.80$ .	p<0.04
4-5	175 (43.75)	74 (42.29)	57 (32.57)	44 (25.14)	131 (74.86)	24.97±11.57	1
>5	20 (5.00)	13 (65)	6 (30)	1 (05)	19 (95)	$19.32\pm8.90$	
Need for platelet	` ,	, ,	` /	` /	` /		
transfusion							
Yes	49 (12.25)	21 (42.86)	21(42.86)	7 (14.28)	42 (85.72)	21.93±8.87	p<0.017
No	351(87.75)	129 (36.75)	123 (35.04)	99 (28.21)	252 (71.79)	25.54±11.37	1

p value <.05: Significant; p value <0.001: Highly significant.

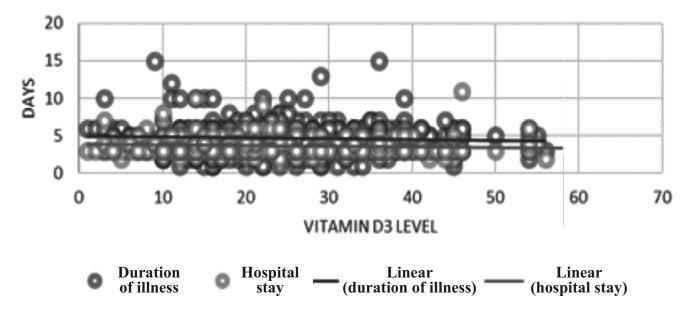


Figure 2: Correlation analysis of vitamin D3 with duration of illness and hospital stay.

total leucocyte count, and LDH, their incidences were high in low vitamin D3 status (Table 2). Notably a significant negative correlation of vitamin D3 level was found with mean platelet volume (MPV), higher MPV were associated with lower vitamin D3 levels (p<0.00001) (Figure 1).

Vitamin D3 status in relation to different clinical parameters are shown in table 3. Longer duration of illness at the time of hospitalization was associated with lower vitamin D3 status (p<0.02) (Figure 2).

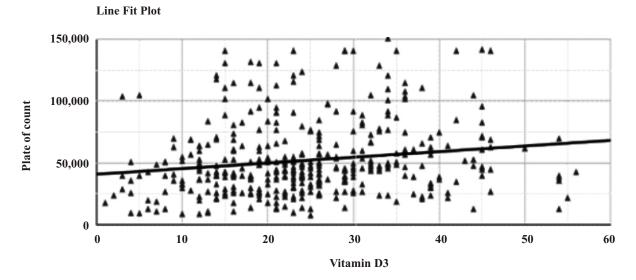


Figure 3: Pearson correlation analysis of Vitamin D3 and platelet count.

Table 4: Pearson correlation analysis of different parameters

Variable	Vitamin D3	Age	Gender	Duration of Illness	Duration of hospital stay	Total platelet count	Bleeding manifestation	Requirement of RDP
Vitamin D3	1	0.0	-0.22	-0.05	-0.09	0.16	-0.03	0.04
Age	0.01	1	0.13	-0.05	0.01	-0.00	-0.09	-0.05
Gender	-0.23	0.1	1	-0.00	-0.02	0.04	0.1	0.00
Duration of illness	-0.06	-0	-0.00	1	-0.02	0.02	-0.02	-0.04
Duration of hospital stay	-0.09	0.0	-0.03	-0.01	1	-0.03	-0.13	0.03
Total platelet count	0.16	0.0	0.04	0.02	-0.031	1	0.08	0.022
Bleeding manifestation	-0.03	0.1	0.15	-0.02	-0.13	0.08	1	0.18
Requirement of RDP	0.04	0.05	0.002	-0.036	0.033	0.022	0.185	1

RDP: Random donor platelet

Mean duration of illness was  $4.73\pm2.05$  days v/s  $4.19\pm1$  days in patients with low D3 status and sufficient D3 level, respectively. Mean platelet count was significantly low in patients with vitamin D3 deficiency  $(45,509.08\pm28,183.22$  per  $\mu$ l) and insufficiency  $(49,032.35\pm29,299.87$  per  $\mu$ l) as compared to patients with normal D3 level  $(60539.36\pm30115.37$  per  $\mu$ l) (p<0.05). Pearson correlation test results found significant small positive relationship between

vitamin D3 and platelet count (Table 4, figure 3).

Low D3 status was also found to be associated with increased incidence of bleeding manifestations (p<0.05) (Figure 4) and more requirement of platelet transfusion (p<0.017) (Figure 5).

Results of the multiple linear regression indicated that there was a very strong collective significant effect

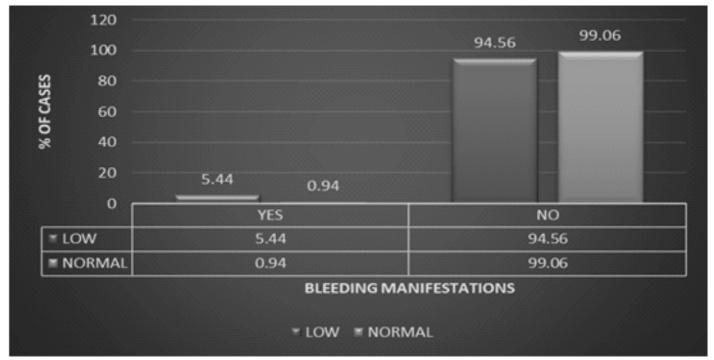


Figure 4: Association of vitamin D3 status and bleeding manifestations.

Table 5: Linear regression analysis of different parameters

Variable	Coefficient	SE	t-stat	lower t0.025	upper t0.975	Standard coefficient	p value	VIF
Age	0.0681	0.031	2.137	0.0054	0.130	0	0.033	1.03
Gender	-4.80	1.103	-4.34	-6.97	-2.63	-0.234	0.000	1.04
Duration of illness	-0.031	0.259	-0.12	-0.542	0.47	-0.065	0.903	1.00
Duration of hospital stay	-0.07	0.48	-0.16	-1.033	0.874	-0.100	0.870	1.022
Total platelet count	0.00	0.00	3.88	0.000	0.00	0.173	0.0001	1.007
Bleeding manifestation	8.72	1.85	4.70	5.07	12.37	-0.031	0.000	1.09
Requirement of RDP	4.40	1.58	2.78	1.29	7.51	0.0535	0.005	1.042

RDP: Random donor platelet; p value <.05: Significant; p value <0.001: Highly significant; VIF: Variance inflation factor

between the age, gender, duration of illness, duration of hospital stay, total platelet count, bleeding manifestation, requirement of RDP, and vitamin D3 (F(5, 395)=420.28, p<0.001, R2=0.84, R2adj=0.84). The individual predictors were examined further and indicated that age (t=-4.37, p<0.001) and gender (t=3.891, p<0.001) were significant predictors in the model while duration of illness (t = 4.946, p < .001) and duration of hospital stay (t=2.816, p=0.005) were non-significant predictors in the model (Table 5).

# **DISCUSSION**

This study was conducted on 400 patients admitted with acute febrile illness and thrombocytopenia at a tertiary care centre. Common acute febrile illnesses associated with thrombocytopenia are dengue, malaria, leptospirosis, typhoid, scrub typhus, miliary tuberculosis, *HIV* etc; prevalence depending on epidemiology and seasonal variation.<sup>1</sup>

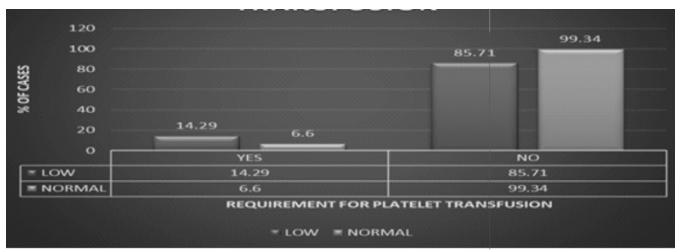


Figure 5: Association of vitamin D3 status and requirement of platelet transfusion.

This study found a high prevalence of vitamin D3 deficiency (73.5%) among cases as compared to control (17.5%) living in the same vicinity and similar environmental exposure. Low levels of vitamin D3 have been reported by other workers in dengue<sup>11</sup> and malaria.<sup>12</sup> Recent evidence has indicated that vitamin D also plays an essential role in the immune response against viral infections and suggested that vitamin D deficiency increases susceptibility to viral infections as well as the risk of recurrent infections. 13 Vitamin D has been shown to exert multiple effects on the immune system.<sup>14</sup> It acts in an autocrine and paracrine fashion to modulate the innate and adaptive immune systems. There is also some evidence that vitamin D itself may modulate immune function in a nongenomic manner by stabilizing endothelial membranes.<sup>15</sup> Gupta et al<sup>16</sup> also found high prevalence of hypovitaminosis D3 (\le 30 ng/ml) status in 70.9% of the cases of swine flu as compared to 44.4% in control group. Vitamin D3 level was significantly low in female patients in current study as compared to male, however Zargar et al<sup>17</sup> reported higher prevalence of vitamin D3 in males as compared to females. This may be because of changing lifestyle and urbanization leading to lesser exposure to sun by females in present study.

In the present study, it was found that severity of thrombocytopenia was associated with lower levels of vitamin D3. Alanli et al<sup>18</sup> also found significant negative correlation between 25-hydroxyvitamin D3 levels and platelet counts. It was observed that a low vitamin D level was correlated with a high MPV in the present study. Apart from their classical roles, both platelets and vitamin D play an important role in inflammation and infectious diseases. Vitamin D deficiency increases the release of proin-

flammatory cytokines such as IL-6 and TNF-α that may lead to a high MPV. 19 Feketea et al<sup>20</sup> found higher MPV in children with influenza respiratory tract infection (RTI) than in those with other RTI, suggesting that the platelets are activated. MPV acted as a positive acute phase reactant which reacted differently compared with other acute phase markers that were not different in the two groups. Activation of platelets is associated with increased MPV.<sup>21</sup> A higher MPV occurs as a result of increased platelet activity and thus of more intense inflammation as a result of a certain infection. Various inflammatory conditions cause an increase in thrombopoiesis, with an increase in the number and the size of platelets. The platelets migrate to the area of inflammation where they are consumed, leading to thrombocytopenia. Cumhur et al<sup>22</sup> also showed in linear regression analysis that low level of vitamin D was independently associated with increased MPV.

Duration of illness and hospital stay was longer in the study participants with low status of vitamin D3. In a study on swine flu patients by Gupta et al<sup>16</sup>, low status of vitamin D was shown to require longer duration for recovery. The current study observed that increased incidence of bleeding manifestations and the need for platelet transfusion in patients was associated with lower vitamin D3 level. Similarly, Iqtadar et al<sup>23</sup> found an association between vitamin D3 deficiency and susceptibility towards severe dengue illness including dengue haemorrhagic fever.

Limitation: The control group was not included in the study as the study was focused on clinical correlation of vitamin D3 status in admitted patients of acute febrile illness with thrombocytopenia. Large case control and interventional studies are required to assess clinical significance of vitamin D3 in such cases.

#### **CONCLUSION**

High prevalence of vitamin D3 deficiency was found in patients admitted with acute febrile illness and thrombocytopenia. Low level of vitamin D3 is found to be associated with increased susceptibility to get ill because of exposure to infectious agents, more severe thrombocytopenia, higher incidence of bleeding manifestation, more requirement for platelet transfusion, and longer duration for recovery. Early recognition of vitamin D3 status may be helpful for better management of such cases in reducing morbidity and mortality.

Funding source: Nil

Conflict of Interest: None

#### REFERENCES

- 1. Parikh F. Infections and thrombocytopenia. *J Assoc Physicians India*. 2016;64(2):11-12.
- 2. Raadsen M, Du Toit J, Langerak T, Van Bussel B, Van Gorp E, Goeijenbier M. Thrombocytopenia in virus infections. *J Clin Med.* 2021;20;10(4):877. doi:10.3390/jcm10040877.
- 3. Agarwal M, Rodgers GM. Miscellaneous causes of thrombocytopenia In: Wintrobe's Clinical Hematology 13<sup>th</sup> edition. 2014;1097-105.
- 4. Bikle DD. Vitamin D metabolism, mechanism of action, and clinical applications. *Chem Biol.* 2014;20;21(3):319-29. doi: 10.1016/j.chembiol.2013.12.016.
- 5. Adams JS, Ren S, Liu PT, Chun RF, Lagishetty V, Gombart AF, et al. Vitamin D directed rheostatic regulation of monocyte antibacterial responses. *J Immunol*. 2009;182:4289-95. doi: 10.4049/jimmunol.0803736.
- Shahmiri M, Enciso M, Adda CG, Smith BJ, Perugini MA, Mechler A. Membrane core-specific antimicrobial action of Cathelicidin LL-37 peptide switches between pore and nanofibre formation. *Sci Rep.* 2016;6:381-84. doi: 10.1038/ srep38184.
- 7. Carlberg C, Molnár F. Vitamin D receptor signaling and its therapeutic implications: Genome-wide and structural view. *Can J Physiol Pharmacol.* 2015;93:311-18.
- 8. Abada A, Elazar Z. Getting ready for building: Signaling and autophagosome biogenesis. *EMBO Rep.* 2014;15(8): 839-52. doi: 10.15252/embr.201439076.
- 9. Holick MF, Binkley NC, Bischoff-Ferrari HA, Gordon CM, Hanley DA. Evaluation, treatment, and prevention of vitamin D deficiency: An endocrine society clinical practice guideline. *J Clin Endocrinol Metab*. 2011;96:1911-30.
- Common Terminology Criteria for Adverse Events (CTCAE) Version 5.0 Published: November 27, 2017 U.S. department of health and human services national institutes of health.

- Dissanayake S, Tennekoon S, Gaffoor S, Liyanage G. Vitamin D deficiency in dengue hemorrhagic fever and dengue shock syndrome among Sri Lankan children: A case-control study. *J Trop Med.* 2021;4173303. doi:10. 1155/2021/4173303.
- 12. Setto JM, Libonati RMF, Ventura AMRDS, Chaves TDSS, Sequeira CG, Martins Filho, et al. Association between vitamin D serum levels and clinical, laboratory, and parasitological parameters in patients with malaria from an endemic area of the Amazon. *Rev Soc Bras Med Trop.* 2022; 8:55:e00772021. doi:10.1590/0037-8682-0077-2021.
- 13. Siddiqui M, Manansala JS, Abdulrahman HA, Nasrallah GK, Smatti MK, Younes N, et al. Immune modulatory effects of vitamin D on viral infections. *Nutrients*. 2020;12 (9):2879. doi: 10.3390/nu12092879.
- 14. Charoenngam N, Holick MF. Immunologic effects of vitamin D on human health and disease. *Nutrients*. 2020;12 (7):2097. doi: 10.3390/nu12072097.
- Kim DH, Meza CA, Clarke H, Kim JS, Hickner RC. Vitamin D and endothelial Function. *Nutrients*. 2020;12:575. doi: 10.3390/nu12020575.
- Gupta BK, Narula K, Saini ML, Baberwal RK, Gupta J, Rulaniya S. Clinical evaluation of significance of 25(OH)D (Vitamin D) status in swine flu (H1N1). *IJN*. 2020;6(2):1-17. doi: 10.14302/issn.2379-7835.ijn-20-3369.
- 17. Zargar AH, Ahmad S, Masoodi SR, Wani AI, Bashir MI, Laway BA, et al. Vitamin D status in apparently healthy adults in Kashmir Valley of Indian subcontinent. *Postgrad Med J.* 2007;83:713-16.
- 18. Alanlı R, Küçükay MB, Yalçın KS. Relationship between vitamin D levels and platelet count: A retrospective study. *Gulhane Med J.* 2020;62:174-48.
- Di Rosa M, Malaguarnera G, De Gregorio C, Palumbo M, Nunnari G, Malaguarnera L. Immuno-modulatory effects of vitamin D3 in human monocyte and macrophages. *Cell Immunol*. 2012;280:36-43.
- Feketea G, Vlacha V, Pop RM, Bocsan IC, Stanciu LA, Buzoianu AD, et al. Relationship between vitamin D level and platelet parameters in children with viral respiratory infections. *Front Pediatr*. 2022;10:8249-59. doi:10.3389/ fped.2022.824959.
- 21. Afsar N, Afroze I, Tahniath H, Abid Z. Role of mean platelet volume as an adjunct in evaluation of acute inflammation. *Ann Pathol Lab Med.* 2017;4:A466-69. doi:10.21276/APALM.1486.
- 22. Cumhur CM, Cure E, Yuce S, Yazici T, Karakoyun I, Efe H. Mean platelet volume and vitamin D level. *Ann Lab Med*. 2014;34:98-103.
- 23. Iqtadar S, Khan A, Mumtaz SU, Livingstone S, Chaudhry MNA, Raza N, Zahra M, et al. Vitamin D deficiency (VDD) and susceptibility towards severe dengue fever: A

prospective cross-sectional study of hospitalized dengue fever patients from Lahore, Pakistan. *Tropical Medicine and Infectious Disease*. 2023;8(1):43.

#### **Article Information**

Received: 13 Jun 2023; Revised: 06 Jul 2023; Accepted: 25 Jul 2023.

# **Corresponding Author**

Dr Bal Kishan Gupta, Senior Professor, Department of General Medicine, SP Medical College and Associated Group of Hospitals, Bikaner, Rajasthan, India.

email: bkgbkn@rediffmail.com

ORCID ID: https://orcid.org/0000-0002-3222-0011