Original Article

Correlation between Serum Vitamin D3 Levels and Severity of COVID-19, Experience from a COVID-19-Dedicated Tertiary Care Hospital from Western India

Prudwiraj Sanamandra, Jugal V. Gada, Sukirti Misra, Sagar A. Barasara, Premlata K. Varthakavi, Nikhil M. Bhagwat

Department of Endocrinology, Topiwala National Medical College and BYL Nair Charitable Hospital, Mumbai, Maharashtra, India

Abstract

Context: It is postulated that 25(OH)D deficiency is associated with a worse prognosis of COVID-19. **Aims:** We aimed to find out whether baseline serum 25-hydroxy vitamin D levels were correlated with COVID-19 disease severity or not in Indian population. **Settings and Design:** It is a prospective observational study. **Methods and Material:** We prospectively recruited 200 COVID-19-positive adult patients and measured their baseline vitamin D levels on admission and prospectively followed their clinical course for their outcome and correlated the association. **Statistical Analysis Used:** The continuous data were represented as mean (±SD) or median (IQR), while the categorical data were represented as proportions. Parametric data were analysed using unpaired *T*-test and ANOVA for two and more than two groups, and for categorical, nonparametric data, Chi-square test were applied. A two-sided *P* value of <0.05 was considered as statistically significant with 95% confidence interval. **Results:** Eighty-six per cent (172/200) of patients had hypovitaminosis D (<30 ng/mL). The prevalence of 25(OH) severe deficiency, deficiency and vitamin D insufficiency was 23%, 41% and 22%, respectively. Clinical severity was graded as asymptomatic (11%), mild (14%), moderate (14.5%), severe (37.5%) and critical (22%). Sixty per cent of patients had clinically severe or critical disease requiring oxygen support with eleven per cent (n = 22) mortality overall. Age (*P*: 0.001), HTN (*P*: 0.049) and DM (*P*: 0.018) were negatively associated with clinical severity. No linear association was found between vitamin D levels and clinical severity. Low vitamin D levels had a significant inverse association with inflammatory markers like neutrophil–lymphocyte ratio (NLR, *P*: 0.012) and IL-6 (*P*: 0.002). **Conclusions:** Vitamin D deficiency was not associated with worse outcomes of COVID-19 infection in Indian population.

Keywords: COVID-19, inflammation, neutrophil–lymphocyte ratio, vitamin D deficiency

Introduction

The COVID-19 pandemic caused by a novel coronavirus (SARS-CoV-2) is characterized by significant morbidity and mortality with no current specific treatment. Preclinical research suggests that the SARS-CoV-2 virus enters cells via the angiotensin-converting enzyme 2 (ACE2).[1]

Coronavirus replication downregulates ACE2, causing the renin–angiotensin system (RAS) to become dysfunctional and producing a cytokine storm with increased production of pro-inflammatory cytokines. This increases the risk of pneumonia, sepsis, acute respiratory distress syndrome and heart failure.[2] Research shows that vitamin D plays a role in balancing RAS, in reducing lung damage through the anti-inflammatory effect which is seen with higher vitamin D levels ranging from 20 to 60 ng/mL.[3-5] Vitamin D deficiency is one of India's most underdiagnosed and undertreated nutritional deficiencies.[6] Many studies have found that lacking vitamin D or vitamin D receptors causes altered innate and adaptive immune functions.[7,8] Vitamin D supplementation is a potentially exciting treatment for COVID-19 infection but scientifically, with a low level of evidence until now.[9] Some studies have confirmed the

Access this article online

Quick Response Code:

Website: www.ijem.in

DOI: 10.4103/ijem.ijem_383_22

Address for correspondence: Dr. Sukirti Misra, Department of Endocrinology, Room No. 419, 4th Floor, College Building, Topiwala National Medical College and BYL Nair Charitable Hospital, AR Nair Road, Mumbai Central, Mumbai - 400 008, Maharashtra, India.
E-mail: sukirtimisra09@gmail.com

Submitted: 07-Oct-2022 **Revised:** 16-Jan-2023
Accepted: 15-Feb-2023 **Published:** 14-Apr-2023

This is an open access journal, and articles are distributed under the terms of the Creative Commons Attribution-NonCommercial-ShareAlike 4.0 License, which allows others to remix, tweak, and build upon the work non-commercially, as long as appropriate credit is given and the new creations are licensed under the identical terms.

For reprints contact: WKHLRPMedknow_reprints@wolterskluwer.com

How to cite this article: Sanamandra P, Gada JV, Misra S, Barasara SA, Varthakavi PK, Bhagwat NM. Correlation between serum vitamin D3 levels and severity of COVID-19, experience from a COVID-19-dedicated tertiary care hospital from Western India. Indian J Endocr Metab 2023;27:170-6.

increased risk of COVID-19 infection in vitamin D–deficient patients.[10,11] Nevertheless, there is insufficient evidence of the association between 25(OH) vitamin D (25(OH) D) levels and COVID-19 severity or outcomes. Therefore, the present study aimed to investigate the association between 25(OH) vitamin D levels and the severity or outcomes of COVID-19 disease.

Material and Methods

The study was conducted after Institutional Ethical Committee approval (ECARP/2020/74). After written informed consent, we prospectively recruited 200 COVID-19-positive adult patients from the wards and ICUs of the COVID-19-dedicated tertiary care centre from August 2020 to March 2021.

Sample selection

Each ward was visited by the primary investigator (PI) to screen for eligible patients. For logistical and safety reasons, only one building with a maximum number of COVID-19 wards, including a high-dependency unit and intensive care units, was selected. The PI visited 3–4 wards daily (one floor per day) to assess eligibility and enrol patients. The PI returned to the wards every 7 days to look for more suitable patients and to follow up on already recruited patients. The parent internal medicine units used the standard hospital treatment regimen to treat hospitalized patients. Vitamin D levels were checked on the first or second day of admission. All the patients were followed up till their outcomes. Disease severity was categorized based on their maximum clinical severity during hospital stay according to NIH COVID-19 severity.[12] All the routine blood investigations and inflammatory markers (IL-6, Ferritin, ESR, CRP etc.) were recorded during the hospital stay. Radiology findings (CT lung involvement) were also recorded whenever available [Figure 1].

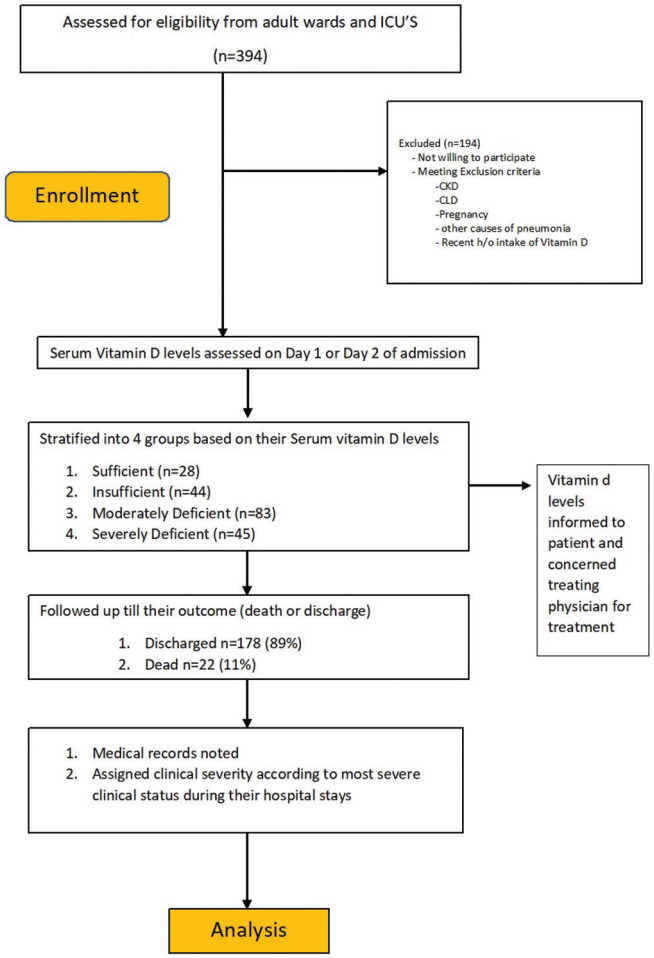

Figure 1: Methodology

Vitamin D assessment

Vitamin D was assessed by chemiluminescent immunoassay method on access immunoassay system (Beckman Coulter) in the endocrinology lab with an interassay and intraassay CV of 5.9–7.2% and 1.5–2.2%, respectively. Serum 25(OH) D levels were stratified into four categories: <10 ng/mL, 10–20 ng/mL, 20–30 ng/mL and >30 ng/ml. Vitamin D deficiency was defined as serum 25(OH) D levels <20 ng/mL (50 nmol/L) following endocrine society guidelines.[13]

HRCT lung

CT scan was done on Philips Brilliance 64 slice machine per hospital protocol and radiologists reported it. Each of the five lobes of both lungs was looked at for the presence of inflammatory abnormalities, including ground-glass opacities, mixed ground-glass opacities, or consolidation. Each lobe was awarded 0–4 points, depending on the percentage of the involved lobe: 0 (0%), 1 (1–25%), 2 (26–50%), 3 (51–75%), or 4 (76–100%) and total involvement of the lung parenchyma was graded as grade 1 having <25%, grade 2 having 26–50%, grade 3 having 51–75%, or grade 4 having 76–100% lung parenchymal involvement.

Sample size

There were no data on the prevalence of vitamin D deficiency in COVID-19 patients from India and worldwide at the time of study planning (August 2020) to calculate a statistically powerful sample size. Hence, we started it as a pilot project with a sample size of 100 patients. During the sample collection of our study, two studies were published in September and October 2020, one from China with a sample size of 62 and another from Iran with a sample size of 235.[14,15] Hence, to increase the power of our study, we increased our sample size to 200 to get a meaningful data.

Statistical analysis

The continuous data were represented as mean (±SD) or median (IQR), while the categorical data were represented as proportions. Parametric data were analysed using unpaired *T*-test and ANOVA for two or more groups, and for categorical, nonparametric data, Chi-square test was applied. The Fisher exact test or χ^2 test and the Kruskal–Wallis test were employed to determine the statistical significance of differences in

proportions and medians, respectively. Pearson *r* correlation was used to measure the degree of the relationship between linearly related variables. A two-sided *P* value of <0.05 was considered statistically significant with a 95% confidence interval.

Results

Baseline characteristics

Two hundred patients were enrolled, including 139 males and 61 females. One hundred and seventy-eight patients (89%) were discharged and 22 (11%) expired during the hospital stay. Eighty-six per cent of patients had hypovitaminosis D (<30 ng/mL). The prevalence of 25(OH)D insufficiency, deficiency and severe deficiency was 22%, 41% and 23%, respectively. The proportion of clinically asymptomatic, mild, moderate, severe and critical patients was 11.5%, 13%, 12%, 42%, and 22.5%, respectively [Figure 2]. Young adults (<40 years of age) in our cohort had slightly low mean vitamin D levels (16.5 vs. 19.2 ng/mL) compared to older adults (>40 years of age). No difference in mean vitamin D levels was seen between males and females (18.79 vs. 18.47 ng/mL). The vitamin D deficient group had a high NLR, IL-6 and CK-MB, explaining the high percentage of mechanical ventilation and death.

Vitamin D and clinical severity [Tables 1 and 2]

Oxygen-requiring patients had a significantly high proportion of HTN (43.4% vs. 21.1%) and DM (41.08% vs. 23.9%). They also had a slightly more proportion of severely vitamin D deficient population (24.03% vs. 19.71%); however, that was not statistically significant (*P*: 0.484).

Vitamin D and lung involvement [Table 2]

One hundred and twelve patients underwent HRCT chest during their hospital stay. Vitamin D deficiency patients had more proportion of patients with severe lung involvement involving >50% lung parenchyma (43.7% vs. 38.2%). Mean vitamin D levels in patients with severe lung involvement (>50%) were also low when compared to patients with mild–moderate lung involvement (0–50%) (20.21 vs. 16.06 ng/mL).

Vitamin D and mortality

Low 25(OH) vitamin D levels were not associated with severity or mortality. On subgroup analysis of patients with 25(OH) D levels ≥ and <20 ng/mL, those with higher levels had a slightly better outcome in terms of decreased mechanical ventilation (18% vs. 21%) and death (9.7% vs. 11.7%); however, these results were not statistically significant [Table 3].

Mortality in our cohort was significantly associated with advancing age (mean 65.9 vs. 53 years, *P*: 0.001). Even though there were a high proportion of severely vitamin D-deficient patients in the deceased cohort (27.2% vs. 21.9%), it was not statistically significant.

A multinominal logistic regression model was used to determine the independent association of age, vitamin D deficiency, HTN, DM and IHD with clinical severity and

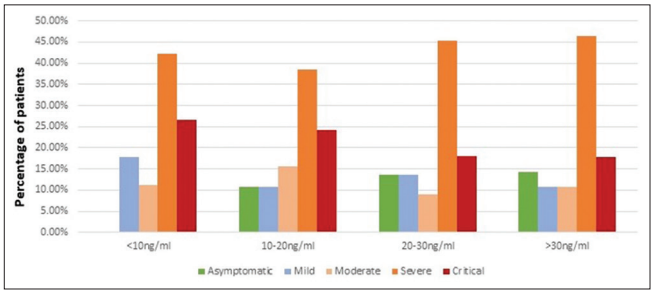

Figure 2: Frequency of various clinical severity according to vitamin D levels

Table 1: Comparison of comorbidities and vitamin D levels among various clinical severity groups and outcomes

	Severity					*P*	Mortality		*P*
	Asymptomatic	Mild	Moderate	Severe	Critical		Discharged	Deceased	
Total	20	26	25	84	45		178	22	
Mean age (SD)	46.2 (13.6)	42.6 (16.4)	52.5 (15.6)	56.9 (13.1)	60.7 (13.2)	0.001*	52.8 (15)	65.9 (10.1)	0.001*
Female:Male	6:14	6:20	10:15	25:59	14:31				
Hypertension	3	4	8	36	20	0.049*	61	10	0.344
Type 2 DM	6	5	6	39	14	0.018*	64	6	0.485
IHD	2	2	0	5	3	0.675	10	6	0.518
Smoking	6	3	2	11	3	0.111	23	2	0.608
Alcohol	4	3	3	7	51	0.681	21	1	0.305
Mean vitamin D in ng/mL (SD)	22.8 (10.9)	17 (10.6)	16.9 (8.8)	19.8 (13)	16.6 (10.8)	0.207	16.8 (12.5)	18.9 (11.5)	0.445
Vitamin D levels									
<10 ng/mL	1	8	5	19	12	0.795	39	6	0.936
10-20 ng/mL	9	9	13	32	20		74	9	
20-30 ng/mL	6	6	4	20	8		40	4	
>30 ng/mL	4	3	3	13	5		25	3	

*P<0.05 is significant. IHD=Ischemic heart disease, T2DM=type 2 diabetes mellitus

Table 2: Comparison of comorbidities, lung involvement, and laboratory parameters between COVID-19 groups based on vitamin D levels

	Vitamin D levels (ng/mL)				*P*
	<10 (*n*=45)	10.1-20 (*n*=83)	20.1-30 (*n*=44)	30.1-60 (*n*=28)	
Comorbidities					
Hypertension	17 (37.7%)	30 (36.1%)	14 (31.8%)	10 (12%)	0.945+
Type 2 diabetes	18 (40%)	28 (33.7%)	13 (29.5%)	11 (39.2%)	0.716+
IHD	6 (13.3%)	0	2 (4.5%)	4 (14.2%)	0.001+
Lung involvement on HRCT chest					
	(*n*=32)	(*n*=39)	(*n*=25)	(*n*=16)	
<25%	15 (46.8%)	14 (35.8%)	8 (32%)	8 (50%)	0.179+
25-50%	4 (12.5%)	7 (17.9%)	9 (36%)	4 (25%)	
50-75%	8 (25%)	11 (28.2%)	6 (24%)	1 (6.25%)	
>75%	5 (15.6%)	7 (17.9%)	2 (8%)	3 (18.75%)	
Inflammatory markers					
Median NLR+ (IQR)	7.8 (3-15.1)	5.6 (2.9-11)	2.9 (2-9.9)	3.8 (2-6.1)	0.012*
ESR	69.55±30.95 (*n*=19)	56±25.9 (*n*=39)	66.25±37.6 (*n*=16)	64±32.23 (*n*=14)	0.330*
CRP	35.23±43.04 (*n*=17)	116.71±330 (*n*=24)	85.9±118 (*n*=16)	70.9±52.8 (*n*=12)	0.107*
Ferritin	643±838 (*n*=22)	741±762.1 (*n*=32)	693.7±523 (*n*=24)	928±491 (*n*=6)	0.320*
LDH	744±415.6	811±438.8	923±356	749±190	0.237*
Median IL-6 (IQR)	3455.4±4004.05 (*n*=24)	2672±3707 (*n*=28)	550±1387 (*n*=20)	1111±2179 (*n*=11)	0.002*
d-Dimer	10.3±20.81 (*n*=24)	3.38±5.05 (*n*=43)	6.73±8.44 (*n*=23)	2.63±2.39 (*n*=7)	0.430*

Mean and SD values were mentioned for all except for NLR and IL-6. **P*-values were calculated by the Kruskal-Wallis test. +*P*-values were calculated by the Chi-square test. NLR=Neutrophil-lymphocyte ratio, ESR=erythrocyte sedimentation rate, IL=interleukin-6, CRP=C-reactive protein, LDH=lactate dehydrogenase

mortality. Only age was found to be significantly associated with both clinical severity and mortality [Table 4].

Discussion

This was one of the few prospective studies done in India, assessing the prevalence of 25(OH)D deficiency among hospitalized COVID-19 patients and its association with various clinical severity groups. Because there were few publications at the time of the study that linked vitamin D insufficiency to COVID-19 severity, we systemically planned our methodology such that the PI prospectively screened 394 patients from COVID-19 wards and enrolled eligible 200 COVID-19 patients.

A recent meta-analysis showed that the prevalence of vitamin D deficiency in India was approximately 56–90%, significantly higher in hospitalized patients.[6] This prevalence was similar to what we found in our population cohort suggesting that vitamin D deficiency is still an under-corrected problem in our country. Like our study, most of the studies published till now have also found a higher proportion of hypovitaminosis D (<30 ng/mL) (86%) among hospitalized COVID-19 patients.[11,15-18]

Our study did not find any correlation between baseline vitamin D levels and O_2 requirements, CT lung involvement, invasive mechanical ventilation and mortality [Table 2]. Few other prospective studies also showed similar findings.[11,19-22] However, some of the epidemiological and prospective studies showed a significant association between vitamin D deficiency with higher O_2 requirements, invasive mechanical ventilation and/or mortality.[15,18] To date, two major meta-analyses found a higher odd ratio of developing the disease, severity and death.[23,24] However, both these meta-analyses mentioned the following biases, that is, the timing of vitamin D testing in relation to the illness, criteria in diagnosing vitamin D deficiency, the severity of illness, and methodological heterogeneity. Comparatively, we used standard definitions to classify vitamin D deficiency and clinical severity of illness and collected vitamin D samples during early admission thereby avoiding heterogeneity.

Most studies that have found a significantly negative association with COVID-19 outcomes used vitamin D cut-off of less than 12 ng/mL. We found an increased proportion of O_2 support (68.8% vs. 63.2%) and mortality (27% vs. 21.9%) in severely vitamin-deficient groups (<10 ng/mL vs. >10 ng/mL); however, they were not statistically significant.

Two Indian studies looked at the relationship between vitamin D and COVID-19 severity.[18,25] One study ascertained a link between vitamin D insufficiency (20 ng/mL) and invasive mechanical ventilation and death, whereas the other did not. The distinction between asymptomatic and severely ill, on the other hand, appeared less validated in the first study. The other study's methodology and results were comparable to ours, but they also examined the effect of vitamin D supplementation, which revealed no further improvement in outcomes. These

Table 3: Clinical, biochemical parameters and disease outcomes in patients with vitamin D deficiency compared to patients without vitamin D deficiency

	Vitamin D <20 ng/mL (*n*=128)	Vitamin D ≥20 ng/mL (*n*=72)	*P*
Male:Female	92:36	47:25	
Mean age in years (SD)	52.8 (15.5)	57.2 (14.1)	0.169
Hypertension	47 (36.7%)	24 (33.3%)	0.639
DM	46 (35.9%)	24 (33.3%)	0.711
IHD	6 (4.6%)	6 (8.3%)	0.297
COAD	7 (5.4%)	3 (4.1%)	1
Median NLR (IQR)	6.19 (2.9-12.8)	3.19 (2-7)	0.001
ESR	60.1 (28.6)	65.1 (34.6)	0.484
Median CRP (IQR)	20.1 (11.7-46)	64.3 (24.8-100)	0.014
Ferritin	702.2 (802.8)	740.7 (517.9)	0.233
LDH	777 (431.7)	854.6 (309.8)	0.093
Median IL-6 (IQR)	1218.5 (54.1-5500)	22.7 (13.5-324)	0.001
Median d-dimer (IQR)	1.3 (0.5-5.3)	2.6 (0.7-6.3)	0.260
Clinical severity			
Asymptomatic	10	10	0.440
Mild	17	9	
Moderate	18	7	
Severe	51	33	
Critical	32	13	
HRCT chest			
<25%	29 (39.7%)	16 (41%)	0.228
25-50%	12 (16.4%)	12 (30.7%)	
50-75%	20 (27.3%)	6 (15.4%)	
>75%	12 (16.4%)	5 (12.8%)	
Oxygen support	83 (64.8%)	46 (63.9%)	1
Mechanical ventilation (ICU)	27 (21.1%)	13 (18%)	0.713
Death	15 (11.7%)	7 (9.7%)	0.815

NLR=Neutrophil-lymphocyte ratio, ESR=erythrocyte sedimentation rate, IL=interleukin-6, CRP=C-reactive protein, LDH=lactate dehydrogenase. IHD=ischemic heart disease, T2DM=type 2 diabetes mellitus, COAD=chronic obstructive airway disease

Table 4: Multinomial logistic regression

	Clinical severity				Outcome			
	Severe		Critical		Mechanical ventilation		Death	
	OR (95% CI)	*P*	OR (95% CI)	*P*	OR (95% CI)	*P*	OR (95% CI)	*P*
Age	1.05 (1.01-1.1)	0.01	1.08 (1.03-1.13)	0.001*	0.96 (0.94-0.99)	0.015*	0.9 (0.89-0.96)	0.001*
Gender (female)	0.60 (0.18-2.03)	0.10	0.64 (0.17-2.3)	0.028	0.88 (0.41-1.9)	0.76	2.7 (0.8-8.9)	0.101
HTN	0.37 (0.79-1.7)	0.212	0.29 (0.05-1.8)	0.141	1.20 (0.52-2.7)	0.665	0.960 (0.32-2.8)	0.942
DM	1.03 (0.28-3.7)	0.965	2.5 (0.6-10.5)	0.203	0.68 (0.3-1.8)	0.37	0.56 (0.18-1.7)	0.31
IHD	2.5 (0.297-21.1)	0.393	1.8 (0.17-20)	0.60	0.60 (0.11-3.04)	0.53	1.32 (0.23-7.4)	0.74
Vitamin D <30 ng/mL	0.975 (0.23-4.1)	0.973	0.937 (0.17-5)	0.940	0.648 (0.179-2.3)	0.7	0.833 (0.172-4.03)	0.821
Vitamin D <20 ng/mL	0.937 (0.29-3)	0.914	0.600 (0.16-2.2)	0.448	0.866 (0.35-2.11)	0.75	1.216 (0.35-4.18)	0.75
Vitamin D <10 ng/mL	0.187 (0.02-1.58)	0.125	0.185 (0.02-1.6)	0.130	1.18 (0.48-2.89)	0.5	1.265 (0.42-3.8)	0.67

**P*<0.05 is significant. Multinomial logistic regression was used to calculate OR, CI and *P*. OR=Odds ratio, CI=confidence interval

disparities point to a flaw in the intrinsic relationship between vitamin D level and COVID-19 severity.

It is now established that the outcome of COVID-19 infection largely depends on the host's immune response. Previous studies showed an association between vitamin D deficiency and increased levels of IL-6 in patients with HIV infections.[26] There is also evidence indicating that vitamin D supplementation can reduce excess IL-6 levels in diabetic mice.[27] Our cohort showed a significant negative correlation between vitamin D levels and IL-6 (*P*: 0.002). This suggests that vitamin D plays a role in modulating the production of interleukins, and vitamin D deficiency may exaggerate inflammatory response by IL-6 production. However, the therapeutic efficacy of vitamin D in suppressing IL-6 in humans needs to be studied by randomized control trials.

Several studies found a relationship between vitamin D deficiency and high neutrophil–lymphocyte ratio in type 2 diabetes patients and other chronic illnesses.[28,29] Recently NLR is found to be an early-stage predictive factor for developing critical illness in COVID-19-infected patients.[29] Our study reciprocated these findings and found high-baseline NLR association with developing severe inflammation and oxygen requirement at presentation or during their hospital stay. We also found inverse association between baseline NLR and vitamin D levels same as Mardani *et al*.[16] Additionally, it has been shown that NLR levels decrease after taking high doses of vitamin D supplements; however, similar therapeutic response in COVID patients is yet to be studied.[30]

25(OH)D and CRP have an antagonistic relationship, according to previous research.[31] This effect is more pronounced in patients with inflammatory diseases than in non-inflammatory diseases. Studies done on COVID patients showed contrasting evidence with no strong association for this correlation till now. Our cohort did not find any significant correlation between CRP and vitamin D levels; however, we had values for 69 patients in our study. Similarly, we did not find any correlation between 25(OH)D and ferritin, LDH and d-dimer levels.

Our understanding is that vitamin D deficiency, by exaggerating the immune response, appears to be an accessory aggravating factor for COVID-19 outcomes in the high-risk cohort while providing no evidence that it is the primary cause in the low-risk population (i.e. young population with less than two comorbidities). Whether 25(OH)D adequacy may prevent COVID-19-related morbidity and mortality needs to be assessed by adequately sized and designed population-based studies, which is essential in the current scenario.

The prospective nature of our study from the western (unstudied) Indian population with an adequate sample size is one of its strengths. However, because our institution was a dedicated COVID-19 tertiary hospital, the study was monocentric, observational and had a selection bias of including more severe/critical cases.

Conclusion

Hypovitaminosis D was prevalent among our hospitalized COVID-19 patients, but low 25(OH) vitamin D levels were not associated with severity or mortality. Vitamin D is negatively associated with inflammatory markers like NLR and IL-6, thus ascertaining its immunomodulatory actions.

Financial support and sponsorship

The study was supported by Department Development Fund.

Conflicts of interest

There are no conflicts of interest.

References

1. Gheblawi M, Wang K, Viveiros A, Nguyen Q, Zhong JC, Turner AJ, *et al*. Angiotensin-converting enzyme 2: SARS-CoV-2 receptor and regulator of the renin-angiotensin system: Celebrating the 20th anniversary of the discovery of ACE2. Circ Res 2020;126:1456-74.
2. Ni W, Yang X, Yang D, Bao J, Li R, Xiao Y, *et al*. Role of angiotensin-converting enzyme 2 (ACE2) in COVID-19. Crit Care 2020;24:422. doi: 10.1186/S13054-020-03120-0.
3. Shi Y, Liu T, Yao L, Xing Y, Zhao X, Fu J, *et al*. Chronic vitamin D deficiency induces lung fibrosis through activation of the renin-angiotensin system. Sci Rep 2017;7:3312. doi: 10.1038/s41598-017-03474-6.
4. Munger KL, Levin LI, Hollis BW, Howard NS, Ascherio A. Serum 25-hydroxyvitamin D levels and risk of multiple sclerosis. JAMA 2006;296:2832-8.
5. Khazai N, Judd SE, Tangpricha V. Calcium and vitamin D: Skeletal and extraskeletal health. Curr Rheumatol Rep 2008;10:110-7.
6. Aparna P, Muthathal S, Nongkynrih B, Gupta SK. Vitamin D deficiency in India. J Family Med Prim Care 2018;7:324-30.
7. Hansdottir S, Monick MM. Vitamin D effects on lung immunity and respiratory diseases. Vitam Horm 2011;86:217-37.
8. di Rosa M, Malaguarnera M, Nicoletti F, Malaguarnera L. Vitamin D3: A helpful immuno-modulator. Immunology 2011;134:123.
9. Vitamin D | COVID-19 Treatment Guidelines. Available from: https://www.covid19treatmentguidelines.nih.gov/therapies/supplements/vitamin-d/. [Last accessed on 2022 Apr 04].
10. D'avolio A, Avataneo V, Manca A, Cusato J, De Nicolò A, Lucchini R, *et al*. 25-hydroxyvitamin D concentrations are lower in patients with positive PCR for SARS-CoV-2. Nutrients 2020;12:1359. doi: 10.3390/nu12051359.
11. Hernández JL, Nan D, Fernandez-Ayala M, García-Unzueta M, Hernández-Hernández MA, López-Hoyos M, *et al*. Vitamin D status in hospitalized patients with SARS-CoV-2 infection. J Clin Endocrinol Metab 2021;106:E1343-53.
12. Clinical Spectrum | COVID-19 Treatment Guidelines. Available from: https://www.covid19treatmentguidelines.nih.gov/overview/clinical-spectrum/. [Last accessed on 2022 Oct 01].
13. Holick MF, Binkley NC, Bischoff-Ferrari HA, Gordon CM, Hanley DA, Heaney RP, *et al*. Evaluation, treatment, and prevention of vitamin D deficiency: An endocrine society clinical practice guideline. J Clin Endocrinol Metab 2011;96:1911-30.
14. Ye K, Tang F, Liao X, Shaw BA, Deng M, Huang G, *et al*. Does serum vitamin D level affect COVID-19 infection and its severity?-A case-control study. J Am Coll Nutr 2021;40:724-31.
15. Maghbooli Z, Sahraian MA, Ebrahimi M, Pazoki M, Kafan S, Tabriz HM, *et al*. Vitamin D sufficiency, a serum 25-hydroxyvitamin D at least 30 ng/mL reduced risk for adverse clinical outcomes in patients with COVID-19 infection. PLoS One 15:e0239799. doi: 10.1371/journal.pone.0239799.
16. Mardani R, Alamdary A, Mousavi Nasab SD, Gholami R, Ahmadi N, Gholami A. Association of vitamin D with the modulation of the disease severity in COVID-19. Virus Res 2020;289:198148. doi: 10.1016/J.VIRUSRES.2020.198148.
17. Radujkovic A, Hippchen T, Tiwari-Heckler S, Dreher S, Boxberger M, Merle U. Vitamin D deficiency and outcome of COVID-19 patients. Nutrients 2020;12:1-13.
18. Jain A, Chaurasia R, Sengar NS, Singh M, Mahor S, Narain S. Analysis of vitamin D level among asymptomatic and critically ill COVID-19 patients and its correlation with inflammatory markers. Sci Rep 2020;10:1-8.
19. Alkhafaji D, al Argan R, Albaker W, Al Elq A, Al-Hariri M, AlSaid A, *et al*. The impact of vitamin D level on the severity and outcome of hospitalized patients with COVID-19 disease. Int J Gen Med 2022;15:343-52.
20. Nasiri M, Khodadadi J, Molaei S. Does vitamin D serum level affect prognosis of COVID-19 patients? Int J Infect Dis 2021;107:264-7.
21. Walk J, Dofferhoff ASM, van den Ouweland JMW, van Daal H, Janssen R. Vitamin D – contrary to vitamin K – does not associate with clinical outcome in hospitalized COVID-19 patients. medRxiv 2020;2020.11.07.20227512. doi: 10.1101/2020.11.07.20227512.
22. Pizzini A, Aichner M, Sahanic S, Böhm A, Egger A, Hoermann G, *et al*. Impact of vitamin D deficiency on COVID-19-A prospective analysis from the CovILD registry. Nutrients 2020;12:1-9. doi: 10.3390/NU12092775.

23. Dissanayake HA, de Silva NL, Sumanatilleke M, de Silva SDN, Gamage KKK, Dematapitiya C, *et al*. Prognostic and therapeutic role of vitamin D in COVID-19: Systematic review and meta-analysis. J Clin Endocrinol Metab 2022;107:1484-502.
24. al Kiyumi M, Kalra S, Davies J, Kalhan A. The impact of vitamin D deficiency on the severity of symptoms and mortality rate among adult patients with Covid-19: A systematic review and meta-analysis. Indian J Endocrinol Metab 2021;25:261-82.
25. Jevalikar G, Mithal A, Singh A, Sharma R, Farooqui KJ, Mahendru S, *et al*. Lack of association of baseline 25-hydroxyvitamin D levels with disease severity and mortality in Indian patients hospitalized for COVID-19. Sci Rep 2021;11:1-8. doi: 10.1038/s41598-021-85809-y.
26. Liu Q, Zhou Y. The cytokine storm of severe influenza and development of immunomodulatory therapy. Cell Mol Immunol 2016;13:3-10.
27. Labudzynskyi D, Shymanskyy I, Veliky M. Role of vitamin D3 in regulation of interleukin-6 and osteopontin expression in liver of diabetic mice. Eur Rev Med Pharmacol Sci 2016;20:2916-9.
28. Wang SY, Shen TT, Xi BL, Shen Z, Zhang X. Vitamin D affects the neutrophil-to-lymphocyte ratio in patients with type 2 diabetes mellitus. J Diabetes Investig 2021;12:254-65.
29. Akbas EM, Gungor A, Ozcicek A, Akbas N, Askin S, Polat M. Vitamin D and inflammation: Evaluation with neutrophil-to-lymphocyte ratio and platelet-to-lymphocyte ratio. Arch Med Sci 2016;12:721-7.
30. Tabatabaeizadeh SA, Avan A, Bahrami A, Khodashenas E, Esmaeili H, Ferns GA, *et al*. High dose supplementation of vitamin D affects measures of systemic inflammation: Reductions in high sensitivity C-reactive protein level and neutrophil to lymphocyte ratio (NLR) distribution. J Cell Biochem 2017;118:4317-22.
31. Daneshkhah A, Agrawal V, Eshein A, Subramanian H, Roy HK, Backman V. Evidence for possible association of vitamin D status with cytokine storm and unregulated inflammation in COVID-19 patients. Aging Clin Exp Res 2020;32:2141. doi: 10.1007/S40520-020-01677-Y.