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Prevalence and Clinical Outcomes of Vitamin D Deficiency in COVID-19 Hospitalized Patients: A Retrospective Single-Center Analysis

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Vitamin D attenuates inflammatory responses to viral respiratory infections. Hence, vitamin D deficiency may be a highly significant prognostic factor for severity and mortality in COVID-19 patients. To evaluate the complications and mortality in different vitamin D status groups in COVID-19 hospitalized patients, we conducted this retrospective study on 646 laboratory-confirmed COVID-19 patients who were hospitalized in Shahid Modarres Hospital, Tehran, Iran from 16th March 2020 until 25th February 2021. Overall, patients with vitamin D deficiency, insufficiency and sufficiency were 16.9%, 43.6% and 39.5%, respectively. The presence of comorbidity, length of hospitalization, ICU admission, and invasive mechanical ventilation requirement and overall complications were significantly more in patients with vitamin D deficiency (p-value < 0.001). 46.8% (51/109) of vitamin D deficient patients died due to the disease, whilst the mortality rate among insufficient and sufficient vitamin D groups was 29.4% (83/282) and 5.5% (14/255), respectively. In univariate analysis, age > 60 years (odds ratio (OR) = 6.1), presence of comorbidity (OR = 10.7), insufficient vitamin D status (OR = 7.2), and deficient vitamin D status (OR = 15.1) were associated with increase in COVID-19 mortality (p-value < 0.001). Finally, the multivariate analysis adjusted for age, sex, and comorbidities indicated vitamin D deficiency as an independent risk factor for mortality (OR = 3.3, p-value = 0.002). Vitamin D deficiency is a strong risk factor for mortality and severity of SARS-CoV-2 infection. Vitamin D supplementation may be able to prevent or improve the prognosis of COVID-19 during this pandemic.

Keywords: COVID-19; prognosis; vitamin D; vitamin D deficiency
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Introduction

The Coronavirus-2019 (COVID-19) pandemic, even though more than 5 months have passed from its emerging time, remains an unsolved global issue. Whilst more European countries could control the spread of COVID-19 to the extent of success, some countries, including Iran, have entered a concerning second phase of the disease spread. Exceeding 30 different mutations of Severe Acute Respiratory Syndrome-Coronavirus (SARS-CoV2), the causative agent of COVID-19, have been recently revealed (Yao et al. 2020a). By rising discoveries in the number of new strains, the worldwide enthusiasm has been remarkably increased for studies evaluating the effects of antiviral drugs

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affecting COVID-19 infection as well as finding an effective vaccine to prevent the disease. Unfortunately, the findings of these trials still have not rendered humans any success in the battle against the novel coronavirus (Thanh Le et al. 2020; Wu et al. 2020). It has been revealed that the complex of hormonally active vitamin D with its receptor (VDR) on T cells has a direct impact on the development and differentiation of T cells, which improves T cell-mediated immunity. On the other hand, it has an immunomodulatory effect through reduction of interleukin-17 (IL-17) production (Joshi et al. 2011). It also has an effect on the expression of anti-oxidation related genes (Aranow 2011; Fisher et al. 2019; Wimalawansa 2019). Considering the noteworthy potential roles of vitamin D in immunity, multiple studies suggested that supplements of vitamin D may be conceivably effective in COVID-19 treatment or at least decrease the disease severity (Grant et al. 2020; Ilie et al. 2020; Jakovac 2020; Marik et al. 2020). In fact, the current pieces of evidence propose that serum concentrations of 25-hydroxyvitamin D (25(OH)D) can be related to the final outcomes of viral infection diseases (Yamshchikov et al. 2009; Gunville et al. 2013; Teymoori-Rad et al. 2019). A cross-sectional analysis on 6,789 individuals in a national birth cohort study in the United Kingdom, with their 25(OH)D measurements revealed that levels of vitamin D were related to lung functions and respiratory tract infection in a linear trend (Berry et al. 2011). Furthermore, it has been reported that vitamin D deficiency upon hospitalization of patients aggravates both short-term and long-term fatality rates among viral respiratory diseases (Watkins et al. 2011; Laaksi 2012; Watkins et al. 2015). During this global pandemic, some studies have confidently suggested the use of vitamin D supplements for cases at higher risks of vitamin D deficiency to maintain the circulating 25(OH) D at the ideal levels (Ali 2020), and some other studies have not shown the effectiveness of it. Even though the effect of vitamin D on improving the prognosis of viral infection has not been thoroughly established yet, confirmed data show that vitamin D could perform its anti-viral role by up-regulating human β defensin and LL-37 antimicrobial peptide (Beard et al. 2011). This study aims to indicate the status of the disease in hospitalized COVID-19 patients and the probable correlation between 25(OH)D serum levels and complications of COVID-19. To provide better insight into the function of vitamin D, we also analyzed the prognostic functions of comorbidities, age, and sex as possible factors contributing to the prognosis of the disease in our COVID-19 cases.

Methods

Study Design and Participants

The present retrospective cohort study has been performed on confirmed COVID-19 patients hospitalized from 16th March 2020 until 25th February 2021 in Shahid Modarres Hospital, Tehran, Iran, which is a tertiary hospital considered as a referral center during the outbreak of Influenza H1N1 2009 and the recent pandemic of COVID-19. All the patients were admitted according to the WHO confirmation guideline (World Health Organization 2020). Nasopharyngeal swab samples and RT-PCR tests were utilized for all admitted patients as COVID-19 laboratorial confirmation. A total of 646 patients including 484 discharged and 162 expired cases were included in the study. Of note, the patients who were still hospitalized and did not have a determined final status (thoroughly recovered or expired) were not included in the study (n = 109). The requirement for written informed consent has been waived by the Ethics Commission of Shahid Beheshti University of Medical Sciences. In order to ensure anonymity, all names were preserved throughout the research.

Data Collection

Data regarding the patients' age, sex, comorbidities, level of vitamin D, complications (length of hospitalization, ICU admission, and invasive mechanical ventilation requirement) and final status, were all gathered with specifically designed data collection forms. Two experienced physicians separately collected data using electronic medical records and double-checked the obtained data. Missing information was obtained by medical researchers or family medicine physicians to have direct contact with the family member of the patients.

Vitamin D evaluation

Vitamin D status has been assessed by laboratorial measuring of patient's serum 25(OH) D levels upon the hospital admission. According to the most recent published guideline (Sizar et al. 2021), we considered 25(OH)D serum concentrations of less than 20 ng/ml as 'deficient status'. Based on the same document, 25(OH) D concentrations between 20-30 ng/ml were described as 'insufficient status'; and the 25(OH)D more than 30 ng/ml was considered as 'sufficient status'.

Statistical Analysis

Continuous, and categorical variables were indicated by mean ± standard deviation (SD) and number (percentage), respectively. To compare the differences between the results, chi-square and Mann-Whitney U tests have been employed as necessary. Univariate logistics regression was used to assess the relation of predictor variables with COVID-19 mortality. The odds ratio (OR) concerning the effect of a one-standard-deviation increase in the predicting factor was used in the interpretation of data. To appraise the correlation between vitamin D status and COVID-19 mortality, all ORs were adjusted for sex, age, and comorbidities by the generalized linear model. P-value < 0.05 has been considered as statistically significant in all comparisons. The statistical analyses of this study were conducted utilizing SPSS statistics software (version: 26.0).

Results

As indicated in Table 1, different demographic and clinical features of the cases have been assessed. The mean age of all cases, in general, was 53.7 (± 15.8). The mean age was 65.4 (± 13.3) years in the non-survived group, which was significantly more than 50.3 (± 14.9) years, reported in the survived group (p-value < 0.001). Also, the percentage of patients with more than 60 years of age was 26.3% and 68.9% in the survived and non-survived groups respectively, marking a significant difference between the two groups (p-value < 0.001). Regarding the sex distribution of cases, male patients were markedly more prevalent in the non-survived group (p-value = 0.004); having formed 71.3% and 83.1% of the survived and non-survived groups, respectively. Besides, male patients in this study were overall more than female patients (74.0% vs. 26.0%).

Fig. 1 illustrates that as the age increases, the overall vitamin D levels decrease; in fact, it can be seen how the maximum, Q3, median, and Q1 of vitamin D in each age group have reduced as the age is increased. It is also demonstrated that between the two sexes, the range in which vitamin D levels have been dispersed is approximately the same. But as the age increases above 60 years old, this range becomes wider for the male patients.

In terms of having any comorbidity, it is shown in Table 1 that 41.8% of the survived cases were reported to have at least one comorbidity, while 88.5% of the non-survived cases were reported with that (p-value < 0.001). Of all the comorbidities that were assessed in the patients, diabetes mellitus and ischemic heart disease were the ones showing statistically significant results. The former was observed in 14.4% and 27.7% of the survived and non-survived groups, respectively (p-value < 0.001), and the latter was reported in 17.5% and 31.7% of the survived and non-survived groups, respectively (p-value < 0.001). The other comorbid conditions evaluated, including hypertension, chronic respiratory disease, and malignancy, did not yield significant results; with hypertension in 30.1% and 33.1% of the survived and non-survived cases respectively (p-value = 0.615); chronic respiratory disease in 16.9% of the both survived and non-survived groups (p-value = 0.994), and malignancy in 4.4% and 8.1% of the survived and non-survived cases respectively (p-value = 0.782). Regarding the vitamin D status of the patients, the mean levels of it were 27.6 (± 5.5) and 22.7 (± 4.8) ng/ml in the survived and non-survived cases, respectively, highlighting a considerable difference between the two groups (p-value < 0.001). Additionally, cases with insufficient vitamin D levels (20-30 ng/ml) formed 40.0% and 56.1% of the survived and non-survived groups respectively, underlining a marked difference between the two cohorts (p-value < 0.001). Furthermore, cases with deficient Vitamin D levels (< 20 ng/ml) formed 11.6% and 34.4% of the survived and non-survived groups respectively, showing how remarkably different the two groups are (p-value < 0.001).

Table 2 shows the status of multiple demographic and clinical features among patients categorized by their vitamin D levels. Generally, 39.5% of all cases had sufficient vitamin D levels (> 30 ng/ml), whereas 43.6% and 16.9% of the patients were respectively insufficient (20-30 ng/ml) and deficient (< 20 ng/ml) in terms of vitamin D. Also, the mean levels of vitamin D were 32.6 (± 1.1), 24.4 (± 2.4), and 17.6 (± 1.2) in patients with sufficient, insufficient, and deficient vitamin D levels, respectively. The mean age of cases with sufficient vitamin D level was 41.7 (± 3.4) years, while it was 60.4 (± 14.8) and 64.7 (± 12.0) years for the cases with insufficient and deficient vitamin D levels, respectively; marking a significant difference between the deficient and non-deficient vitamin D cases (p-value < 0.001). The percentage of male patients was 63.1%, 82.6%, and 77.1% for the cases with sufficient, insufficient, and deficient vitamin D levels, respectively (p-value = 0.423). Of note, only 22.7% of cases with normal vitamin D had

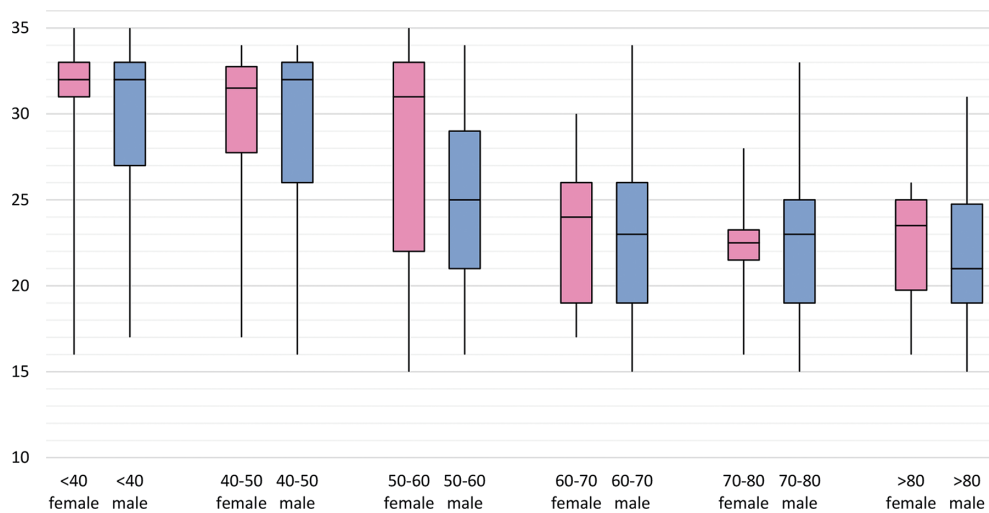

Fig. 1. Vitamin D levels in COVID-19 patients categorized by age (box plot). Vitamin D levels are shown in ng/ml.

Table 1. Demographic and clinical characteristics of the COVID-19 patients.

Variables	Total n = 646	Survived n = 498	Non-survived n = 148	p-value
Age[a]	53.7 ± 15.8	50.3 ± 14.9	65.4 ± 13.3	< 0.001*
< 60 years[b]	413 (63.9%)	367 (73.7%)	46 (31.1%)	--
≥ 60 years[b]	233 (36.1%)	131 (26.3%)	102 (68.9%)	< 0.001*
Sex[b]				
Female	168 (26.0%)	143 (28.7%)	25 (16.9%)	--
Male	478 (74.0%)	355 (71.3%)	123 (83.1%)	0.004*
Comorbidity[b]				
No	307 (47.5%)	290 (58.2%)	17 (11.5%)	--
Yes	339 (52.5%)	208 (41.8%)	131 (88.5%)	< 0.001*
Hypertension	203 (31.4%)	154 (30.1%)	49 (33.1%)	0.615
Diabetes	113 (17.5%)	72 (14.4%)	41 (27.7%)	< 0.001*
Ischemic heart disease	134 (20.7%)	87 (17.5%)	47 (31.7%)	< 0.001*
Chronic respiratory disease	109 (16.9%)	84 (16.9%)	25 (16.9%)	0.994
Malignancy	56 (8.7 %)	22 (4.4%)	12 (8.1%)	0.782
Vitamin D status[a]	26.5 ± 5.7	27.6 ± 5.5	22.7 ± 4.8	< 0.001*
Sufficient[b]	255 (39.5 %)	241 (48.4%)	14 (9.5%)	--
Insufficient[b]	282 (43.6 %)	199 (40.0%)	83 (56.1%)	< 0.001*
Deficient[b]	109 (16.9 %)	58 (11.6%)	51 (34.4%)	< 0.001*

[a]mean ± SD.
[b]number (%).
*indicates the statistically significant p-values ($p < 0.05$).

Table 2. Demographic and clinical characteristics of COVID-19 patients according to vitamin D status.

Variables	Vitamin D < 20 ng/ml (Deficient status)	Vitamin D 20-30 ng/ml (Insufficient status)	Vitamin D > 30 ng/ml (Sufficient status)	P-value[c]
25(OH)D serum levels[a]	17.6 ± 1.2	24.4 ± 2.4	32.6 ± 1.1	--
Number of patients	109	282	255	--
Age[a]	64.7 ± 12.0	60.4 ± 14.8	41.7 ± 3.4	< 0.001*
Sex (male)[b]	84 (77.1 %)	233 (82.6%)	161 (63.1%)	0.423
Presence of comorbidity[b]	86 (78.9 %)	195 (69.1%)	58 (22.7%)	< 0.001*
ICU admission[b]	65 (59.6 %)	117 (41.5%)	94 (36.9%)	< 0.001*
Invasive mechanical ventilation[b]	41 (37.6 %)	60 (21.3%)	29 (11.4 %)	< 0.001*
Length of hospitalization (days)[a]	11.2 ± 4.1	9.2 ± 4.8	6.2 ± 3.4	< 0.001*
Death[b]	51 (46.8 %)	83 (29.4%)	14 (5.5%)	< 0.001*

[a]mean ± SD.
[b]number (%).
[c]This column shows the p-values for the statistical comparison of vitamin D deficient patients with non-deficient patients (those with both insufficient vitamin D levels and sufficient vitamin D levels).
*indicates the statistically significant p-values ($p < 0.05$).
25(OH)D, 25-hydroxyvitamin D.

comorbidities, whereas 69.1% and 78.9% of the cases with insufficient and deficient vitamin D, respectively, had comorbidities. The need for ICU admission, as an important clinical characteristic, was reported in 36.9% of patients with sufficient vitamin D level, while 41.5% and 59.6% of the patients with insufficient and deficient vitamin D levels, respectively, required ICU admission; underlining a significant requirement of ICU admission among vitamin D deficient patients compared to non-deficient vitamin D cases (p-value < 0.001). As another prominent clinical feature, the necessity for utilizing invasive mechanical ventilation for patients had been raised as the levels of vitamin D decreased; accordingly, 11.4% of the cases with normal vitamin D needed invasive ventilation, whilst it was recorded in 21.3% and 37.6% of the cases with insufficient and deficient vitamin D, respectively. Furthermore, the length of hospitalization was reported to be 6.2 (± 3.4) days in the cases with normal vitamin D level, whereas it was

Table 3. Univariate analysis of factors associated with COVID-19 mortality.

Variables	Confidence Interval 95 %	Odds ratio (OR)	p-value
Mean age ≥ 60 years	4.159-9.278	6.212	< 0.001*
Male sex	1.236-3.177	1.982	0.004*
Presence of comorbidity	6.288-18.358	10.744	< 0.001*
Diabetes mellitus	1.463-3.514	2.267	< 0.001*
Ischemic heart disease	1.450-3.333	2.198	< 0.001*
Vitamin D status			
Sufficient	--	--	--
Insufficient	3.954-13.038	7.180	< 0.001*
Deficient	7.846-29.204	15.137	< 0.001*

*indicates the statistically significant p-values ($p < 0.05$).

reported to be 9.2 (± 4.8) and 11.2 (± 4.1) days for cases with insufficient and deficient vitamin D levels, respectively; spotlighting a marked difference between the deficient and non-deficient cases (p-value < 0.001). Moreover, only 5.5% of patients with normal vitamin D levels died, while 29.4% and 46.8% of the patients with insufficient and deficient vitamin D levels, respectively, expired; marking how significantly the mortality rate of the patients was associated with levels of vitamin D (p-value < 0.001).

As demonstrated in Table 3, factors associated with COVID-19 mortality have been evaluated. Each predictor has been separately analyzed using univariate logistic regression; odds ratio of mortality for patients with significantly prominent risk factors have been obtained. Cases with more than 60 years of age have been significantly 6.2 times more likely to die due to COVID-19, compared to younger cases (OR = 6.212; p-value < 0.001). Regarding sex, as a predictor of mortality, it was shown that male patients were approximately 2.0 times more probable to die from the disease than female patients (OR = 1.982; p-value = 0.004). Accordingly, the odds ratio of mortality for patients who have comorbidities is 10.744, meaning that cases who had at least one comorbidity were roughly 10.7 times more likely to expire (p-value < 0.001). As previously mentioned, the two comorbidities which had shown a significant difference in the initial statistical analysis between the two groups were diabetes mellitus and ischemic heart disease. The odds ratio of mortality for these two showed that patients who had the former as comorbidity were 2.3 times more likely to expire (p-value < 0.001), and the patients who had the latter as comorbidity were 2.2 times more likely to expire (p-value < 0.001). Evaluating the low level of vitamin D, as another important predictor of mortality in this study, indicated that compared to normal cases, patients with insufficient vitamin D were approximately 7.2 times more likely to die (OR odds ratio = 7.180, confidence interval: 3.954-13.038; p-value < 0.001) while patients with deficient vitamin D were roughly 15.1 times more likely to die from the disease (OR = 15.137, confidence interval: 7.846-29.204; p-value < 0.001).

Table 4. Association between vitamin D status and COVID-19 mortality (adjusted for sex, age, and comorbidities).

Vitamin D status	Confidence interval 95 %	Odds ratio (OR)	p-value
Sufficient	--	--	--
Insufficient	0.935-3.688	1.857	0.077
Deficient	1.540-6.994	3.284	0.002*

*indicates the statistically significant p-values (OR with cross-tabulation analysis).

In order to control for the possible confounding effect of age, sex, and comorbidity on the correlation of vitamin D status with mortality, a generalized linear model was employed, the results of which are shown in Table 4. After accounting for these possibly confounding variables in the model, a significant link has been established between vitamin D status and mortality; the odds ratio of death for patients with deficient vitamin D was significantly 3.284 (p-value = 0.002), meaning that when compared with normal cases, vitamin D deficient patients were approximately 3.3 times more likely to die from CVOID-19. Meanwhile, cases with insufficient vitamin D were obtained to be 1.9 more probable to die from the disease (odds ratio = 3.284, confidence Interval: 0.935-3.688). Although, the results concerning the insufficient vitamin D were not significant (p-value = 0.77).

Discussion

In this study, we highlighted the connection of vitamin D status with complications and mortality due to SARS-CO-2 infection in 646 laboratory-confirmed patients. Our study is the first to indicate such a profound and detailed link between 25(OH)D low serum levels and increased risk of advanced respiratory complications, along with secondary infection during hospitalization of the patients. We also revealed that vitamin D, even when being adjusted for confounding variables including sex, age, and comorbidity, independently increased the mortality risk in

hospitalized COVID-19 patients.

This research is compatible with the recent research conducted by Raharusun et al. (2020) in Indonesia which evaluated 780 COVID-19 patients. Similar to what we have discovered, they obtained that vitamin D insufficiency and deficiency elevate the risk of fatality with odds of 7.63 and 10.12, respectively. The present study is also aligned with an observational study carried out by Bychinin et al. (2021), who showed that all 40 COVID-19 cases had a low median (IQR) serum 25(OH)D concentration at admission to the ICU; underlining the median (IQR) serum 25(OH)D concentration to be greater in survivors (13.3 ng/mL) than in non-survivors (9.6 ng/mL).

Of note, we also quite expectedly spotted that vitamin D deficiency, as a neglected health issue in Iran, was a considerable finding even among our younger adult patients; in fact, the percentage of the hospitalized patients who did not meet the efficient 25(OH)D serum concentrations of more than 30 ng/ml, exceeded 60 percent. Formerly, in a 2-year prospective cohort study in 2016 (Talebi et al. 2019), it has been revealed in Iran that community-acquired pneumonia (CAP) infected patients who were vitamin D deficient, had greater severity of the disease and mortality as against those having higher vitamin D levels. It can be assumed that these numbers can be representative of the total Iranian population that showed much higher all-cause excess mortality during the SARS-CoV-2 peak infection phase than neighboring countries; despite the sufficient capacity of hospitals and ICUs.

The first step to comprehend the possible effect of vitamin D on COVID-19 patients is to fully understand the physiologic roles of vitamin D. Vitamin D could both increase and decrease the gene expression. The gene promotor is affected by the interaction of VDR and vitamin D responsive element (VDRE) which is the mechanism of increased expression. On the other hand, the decrease in the gene expression or downregulation is considered to be due to corepressors. Therefore, vitamin D can either positively or negatively affect the target genes' expression (Wimalawansa 2019). The non-genomic role of vitamin D concerned activating some signaling factors in the promoter parts of the genes, associated with the vitamin D receptive factor (VDRE) (Hii and Ferrante 2016). Additionally, vitamin D is of huge significance in the function of the immune system (Prietl et al. 2013). As a matter of fact, Cathelicidin, a polypeptide developed by the stimulated expression of vitamin D, has demonstrated antibacterial effects on fungi, bacteria, and enveloped viruses, like the members of the coronavirus family (Liu et al. 2006; Adams et al. 2009). The active vitamin D metabolite in dendritic cells and macrophages, produced from predecessor 25(OH)D, contributes to VDR activity, resulting in the production of multiple peptides important in the adaptive and innate immune response (Chun et al. 2014).

Previously, multiple well-established pieces of evidence demonstrated that the anti-viral role of vitamin D is attributed to not only affecting the viral replication directly but also participating in immunomodulation and anti-inflammation processes (Teymoori-Rad et al. 2019). These characteristics are mainly shaped by the cell junction's maintenance, strengthening cellular immunity by impacting tumor necrosis factor-alpha and interferon-gamma (Grant et al. 2020), as well as regulating adaptive immunity via suppression of T helper cell type 1 (Th1) and increasing the stimulation of T type regulatory cells (Cantorna et al. 2015). These effects of vitamin D could justify our findings that ARDS complication had occurred significantly more among vitamin D deficient patients: it appears that SARS-CoV-2 firstly escape immune responses, which then might be accompanied with cytokine storming and the immune hyperreactivity (Gattinoni et al. 2020), as a usual pathogenic mechanism in the development of systemic inflammatory response syndrome (SIRS) and ARDS, disregarding the etiologic factor.

In fact, a substantial body of evidence explains a causal pathophysiological function of vitamin D in the severity of respiratory viral infections. The immunological response to SARS-CoV-2 indicates many similarities to the response to SARS-CoV-1v (Channappanavar and Perlman 2017): in cases with belated viral clearance and overdue type I/III interferon response, increasing recruiting of pro-inflammatory Th1/M1-polarized immune cells and neutrophils contributes to alveolar and endothelial cell necrosis, triggering a cytokine storm in some patients that enhances diffuse alveolar damage. In this regard, the protecting role of vitamin D is observed in countless situations concerning ARDS, cytokine hyperproduction, and pneumonia (Tsujino et al. 2019; Hong et al. 2020). Also vitamin D has been recently suggested as a redeployed therapeutic agent for the lung injuries associated with influenza A H5N1 virus (Huang et al. 2020).

Furthermore, vitamin deficiency seems to be simply a surrogate marker for underlying confounding comorbidity or general indicator of poor nutrition and ill health. The chronic obstructive pulmonary disease has been shown to alter 25(OH)D metabolism or sequestration (Jolliffe et al. 2013) with lower increments in circulating 25(OH)D status after controlled repletion. Notwithstanding, chronic respiratory diseases were not observed significantly more in our expired cases in contrast to survived ones.

Vitamin D deficiency has also been correlated to known COVID-19 risk factors such as cardiovascular diseases and diabetes, and subsequent risk of fatality (Gouni-Berthold et al. 2009). Similarly, our analysis of comorbidities strongly supported such confounding effects, observed in non-survived COVID-19 cases that demonstrated the significantly greater prevalence of recognized vitamin D-impacted diseases such as diabetes, and coronary artery disease. Moreover, vitamin D deficiency has been shown to impact the promotion of the renin-angiotensin system (RAS), persistent stimulation of which might contribute to chronic cardiovascular disease (CVD) (Shi et al. 2017).

Subsequently, as we revealed in our cases, individuals with these comorbid conditions make up a greater percentage of critically ill COVID-19 patients.

All in all, because of the scarcity of specific therapeutics, and the urgent need to act, what we have found is inferred to be applicable for SARS-CoV-2 infection, warranting vitamin D usage as a practicable auxiliary treatment in COVID-19 patients.

Our study has several limitations. First, because a significant number of patients did not have a definitive final status during this study, they were excluded. Therefore, the mortality rate of patients cannot be definitively assessed. Second, another prominent and new-discussed characteristic of COVID-19 which we found in some of our critically ill patients, was coagulopathy: a greater concentration of D-dimer, as an indicator of a predominantly pro-thrombotic disseminated intravascular coagulation (DIC), has been found in our patients, particularly in those with lower levels of vitamin D. However, we refrained to evaluate and discuss the mentioned characteristic due to the lack of efficient data for analyzing and comparing such complications by the methods of this study. Of note, low vitamin D has been recently verified in the pathology findings, suggesting that microvascular thrombosis is present in the lung tissue of COVID-19 patients (Yao et al. 2020b). Another issue that was more of a challenge rather than a limitation was the process of compensating for the unwanted effects of confounding factors. In fact, the influence of confounders such as age, sex, and comorbidities, were all acknowledged and taken into consideration. In this study we tried to omit the undesired effect of these confounders by the proper matching measures between two study groups, and also by employing the right statistical modeling tools and analytical methods. However, some other confounding factors such as sunlight exposure-time might have not been completely compensated for in our study. Last but not least, the majority of our study population was Iranian patients. Due to possible racial, geographical, economic, and cultural conditions, the Iranian population is revealed to be highly prevalent in terms of vitamin D deficiency and related complications in multiple evidence. Similar studies are much needed to be conducted for other nationalities and ethnicities, to confirm the findings of our study.

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