The Role of Vitamin D in the Age of COVID-19: A Systematic Review and Meta-Analysis

Roya Ghasemian[1], Amir Shamshirian[2,3]*, Keyvan Heydari[3,4], Mohammad Malekan[4], Reza Alizadeh-Navaei[3], Mohammad Ali Ebrahimzadeh[5], Majid Ebrahimi Warkiani[6,7], Hamed Jafarpour[4], Sajad Razavi Bazaz[6], Arash Rezaei Shahmirzadi[8], Mehrdad Khodabandeh[9], Benyamin Seyfari[10], Alireza Motamedzadeh[11], Ehsan Dadgostar[12], Marzieh Aalinezhad[13], Meghdad Sedaghat[14], Nazanin Razzaghi[8], Bahman Zarandi[15], Anahita Asadi[5], Vahid Yaghoubi Naei[16], Reza Beheshti[5], Amirhossein Hessami[2], Soheil Azizi[17], Ali Reza Mohseni[17,18], Danial Shamshirian[19]*

1. Antimicrobial Resistance Research Center, Department of Infectious Diseases, Mazandaran University of Medical Sciences, Sari, Iran.

2. Department of Medical Laboratory Sciences, Student Research Committee, School of Allied Medical Science, Mazandaran University of Medical Sciences, Sari, Iran.

3. Gastrointestinal Cancer Research Center, Mazandaran University of Medical Sciences, Sari, Iran.

4. Student Research Committee, School of Medicine, Mazandaran University of Medical Sciences, Sari, Iran.

5. Pharmaceutical Sciences Research Center, Department of Medicinal Chemistry, School of Pharmacy, Mazandaran University of Medical Science, Sari, Iran.

6. School of Biomedical Engineering, University of Technology Sydney, Sydney, Ultimo, NSW, 2007, Australia.

7. Institute of Molecular Medicine, Sechenov First Moscow State University, Moscow, 119991, Russia.

8. Student Research Committee, Golestan University of Medical Sciences, Gorgan, Iran.

9. Neuromusculoskeletal Research Center, Department of Physical Medicine and Rehabilitation, Iran University of Medical Sciences, Tehran, Iran.

10. Department of Surgery, Faculty of Medicine, Kashan University of Medical Sciences, Kashan, Iran.

11. Department of Internal Medicine, Faculty of Medicine, Kashan University of Medical Sciences, Kashan, Iran.

12. Halal Research Center of IRI, FDA, Tehran, Iran.

13. Department of Radiology, Isfahan University of Medical Sciences, Isfahan, I.R. Iran.

14. Department of Internal Medicine, Imam Hossein Hospital, Shahid Beheshti University of Medical Sciences, Tehran, Iran.

15. Student Research Committee, Iran University of Medical Sciences, Tehran, Iran.

16. Immunology Research Center, Mashhad University of Medical Sciences, Mashhad, Iran.

17. Department of Medical Laboratory Sciences, School of Allied Medical Science, Mazandaran University of Medical Sciences, Sari, Iran.

18. Thalassemia Research Center, Hemoglobinopathy Institute, Mazandaran University of Medical Sciences, Sari, Iran.

19. Chronic Respiratory Diseases Research Center, National Research Institute of Tuberculosis and Lung Diseases (NRITLD), Shahid Beheshti University of Medical Sciences, Tehran, Iran.

Corresponding authors:

1. Danial Shamshirian, Chronic Respiratory Diseases Research Center, National Research Institute of Tuberculosis and Lung Diseases (NRITLD), Shahid Beheshti University of Medical Sciences, Tehran, Iran.

E-Mail: shamshirian@sbmu.ac.ir

Tel: +98 912 640 6146

2. Amir Shamshirian; Gastrointestinal Cancer Research Center, Mazandaran University of Medical Sciences, Sari, Iran.

E-Mail: shamshirian.amir@gmail.com

Tel: +98 915 187 2961

DR AMIR SHAMSHIRIAN (Orcid ID : 0000-0002-2735-0209)

The Role of Vitamin D in the Age of COVID-19: A Systematic Review and Meta-Analysis

Abstract

Background: Evidence recommends that vitamin D might be a crucial supportive agent for the immune system, mainly in cytokine response regulation against COVID-19. Hence, we carried out a systematic review and meta-analysis in order to maximize the use of everything that exists about the role of vitamin D in the COVID-19.

Methods: A systematic search was performed in PubMed, Scopus, Embase, and Web of Science up to December 18, 2020. Studies focused on the role of vitamin D in confirmed COVID-19 patients were entered into the systematic review.

Results: Twenty-three studies containing 11901participants entered into the meta-analysis. The meta-analysis indicated that 41% of COVID-19 patients were suffering from vitamin D deficiency (95% CI, 29%-55%), and in 42% of patients, levels of vitamin D were insufficient (95% CI, 24%-63%). The serum 25-hydroxyvitamin D concentration was 20.3 ng/mL among all COVID-19 patients (95% CI, 12.1-19.8). The odds of getting infected with SARS-CoV-2 is 3.3 times higher among individuals with vitamin D deficiency (95% CI, 2.5-4.3). The chance of developing severe COVID-19 is about five times higher in patients with vitamin D deficiency (OR: 5.1, 95% CI, 2.6-10.3). There is no significant association between vitamin D status and higher mortality rates (OR: 1.6, 95% CI, 0.5-4.4).

Conclusion:

This study found that most of the COVID-19 patients were suffering from vitamin D deficiency/insufficiency. Also, there is about three times higher chance of getting infected with SARS-CoV-2 among vitamin D deficient individuals and about 5 times higher probability of developing the severe disease in vitamin D deficient patients. Vitamin D deficiency showed no significant association with mortality rates in this population.

Keywords: Pandemic, 2019-nCoV, Coronavirus Outbreaks, SARS-CoV-2, Vitamin D, 25-hydroxyvitamin D, 25(OH)D.

What's already known about this topic?

- Evidence recommended that vitamin D might be a crucial supportive agent for the immune system, mainly in cytokine response regulation against COVID-19

- Several functions, such as modulating the adaptive immune system and cell-mediated immunity, as well as an increase of antioxidative-related genes expression, have been proven for Vitamin D as an adjuvant in the prevention and treatment of acute respiratory infections

What does this article add?

- Most of the COVID-19 patients were suffering from vitamin D deficiency/insufficiency.

- There is about three times higher chance of getting infected with SARS-CoV-2 among vitamin D deficient individuals.

- There is about five times higher probability of developing the severe disease in vitamin D deficient patients.

Introduction:

Following the emergence of a novel coronavirus from Wuhan, China, in December 2019, the respiratory syndrome coronavirus 2 (SARS-CoV-2) has affected the whole world and is declared a pandemic by World Health Organization (WHO) on March 26, 2020[1]. According to Worldometer metrics, this novel virus has been responsible for approximately 83,848,186 infections, of which 59,355,654 cases are recovered, and 1,826,530 patients have died worldwide up to January 01, 2021.

After months of medical communities' efforts, one of the hottest topics is still the role of Vitamin D in the prevention or treatment of COVID-19. Several functions, such as modulating the adaptive immune system and cell-mediated immunity, as well as an increase of antioxidative-related genes expression, have been proven for Vitamin D as an adjuvant in the prevention and treatment of acute respiratory infections [2-4]. According to available investigations, it seems that such functions lead to cytokine storm suppression and avoid Acute Respiratory Distress Syndrome (ARDS), which has been studied on other pandemics and infectious diseases in recent years [4-7].

To the best of our knowledge, unfortunately, after several months, there is no adequate high-quality data on different treatment regimens, which raises questions about gaps in scientific works. On this occasion, when there is an essential need for controlled randomized trials, it is surprising to see only observational studies without a control group or non-randomized controlled studies with retrospective nature covering a small number of patients. The same issue is debatable for 25-hydroxyvitamin D (25(OH)D); hence, concerning all of the limitations and analyze difficulties, we carried out a systematic review and meta-analysis to try for maximizing the use of everything that exists about the role of this vitamin in the COVID-19.

Methods:

Search Strategy

The Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guideline was considered for the study plan. A systematic search through databases of PubMed, Scopus, Embase, and Web of Science was done up to December 18, 2020. Moreover, to obtain more data, we considered gray literature and references of eligible papers. The

search strategy included all MeSH terms and free keywords found for COVID-19, SARS-CoV-2, and Vitamin D (Supplementary Table). There was no time/location/ language limitation in this search.

Criteria study selection

Four researchers have screened and selected the papers independently, and the supervisor solved the disagreements. Studies met the following criteria included in the meta-analysis: 1) comparative or non-comparative studies with retrospective or prospective nature; and 2) studies reported the role of vitamin D in confirmed COVID-19 patients. Studies were excluded if they were: 1) *in vitro* studies, experimental studies, reviews, 2) duplicate publications.

Data extraction & quality assessment

Two researchers (H.J and M.M) have evaluated the papers' quality assessment and extracted data from selected papers. The supervisor (D.Sh) resolved any disagreements in this step. The data extraction checklist included the name of the first author, publication year, region of study, number of patients, co-morbidity, vitamin D Status, serum 25-hydroxyvitamin D levels, ethnicity, mean age, medication dosage, treatment duration, adverse effects, radiological results, and mortality. The Newcastle-Ottawa Scale (NOS) checklist [8] and its modified version for cross-sectional studies [9] and Jadad scale [10] for randomized clinical trials were used to value the studies concerning various aspects of the methodology and study process.

Vitamin D cut-off [11]

In this case, according to most of the studies, vitamin D cut-off points were considered as follows:

- Vitamin D sufficiency: 25(OH)D concentration greater than 30 ng/mL
- Vitamin D insufficiency: 25(OH)D concentration of 20 to 30 ng/mL
- Vitamin D deficiency: 25(OH)D level less than 20 ng/mL

Targeted outcomes

1) Frequency of Vitamin D status in COVID-19 patients; 2) Mean 25(OH)D concentration; 3) Association between Vitamin D Deficiency and SARS-CoV-2 infection; 4) Association

between Vitamin D Deficiency and COVID-19 severity; 5) Association between Vitamin D Deficiency and COVID-19 mortality; 6) Co-morbidity frequency; 7) Ethnicity frequency.

Heterogeneity assessment

I-square (I^2) statistic was used for heterogeneity evaluation. Following Cochrane Handbook for Systematic Reviews of Interventions [12], the I^2 was interpreted as follows: "*0% to 40%: might not be important; 30% to 60%: may represent moderate heterogeneity; 50% to 90%: may represent substantial heterogeneity; 75% to 100%: considerable heterogeneity. The importance of the observed value of I^2 depends on (i) magnitude and direction of effects and (ii) strength of evidence for heterogeneity (e.g., P-value from the chi-squared test, or a confidence interval for I^2).*" Thus, the random-effects model was used for pooling the outcomes in case of heterogeneity; otherwise, the inverse variance fixed-effect model was used. Forest plots were presented to visualize the degree of variation between studies.

Data analysis

Meta-analysis was performed using Comprehensive Meta-Analysis (CMA) *v*. 2.2.064 software. The pooling of effect sizes was done with 95% Confident Interval (CI). The fixed/random-effects model was used according to heterogeneities. In the case of zero frequency, the correction value of 0.1 was used.

Publication bias

Begg's and Egger's tests were used for publication bias evaluation. A *P*-value of less than 0.05 was considered as statistically significant.

Results

Study selection process

The first search through databases resulted in 1382 papers. After removing duplicated papers and first step screening based on title and abstract, 121 papers were assessed for eligibility. Finally, 23 articles were entered into the meta-analysis. PRISMA flow diagram for the study selection process is presented in Figure 1.

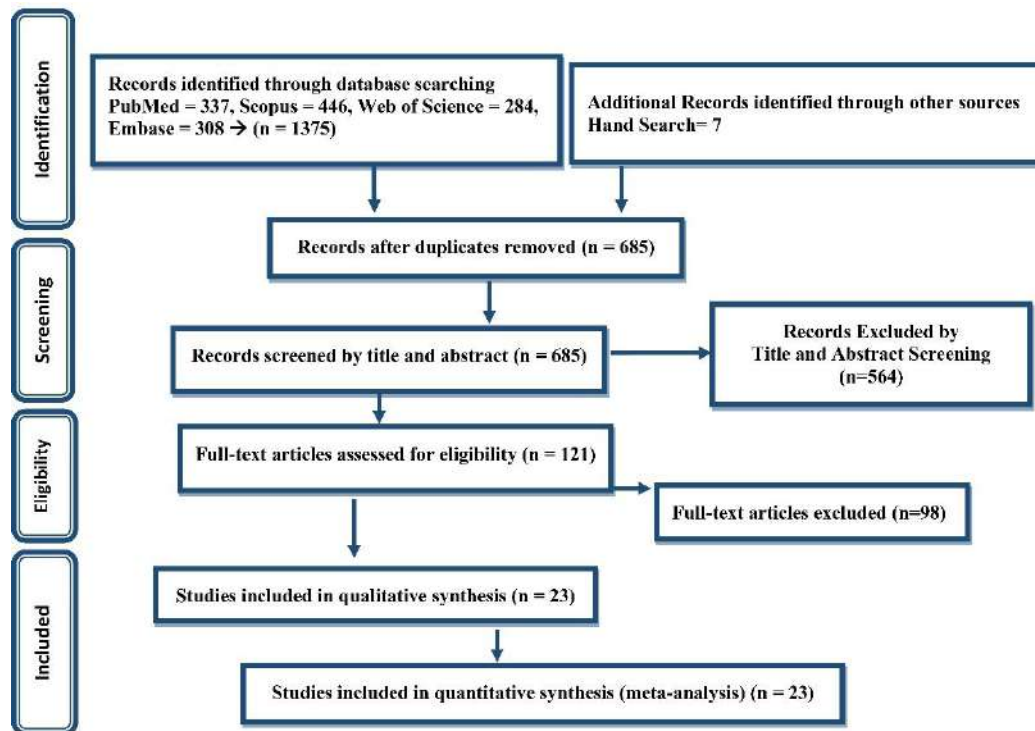*Figure 1.PRISMA flow diagram for the study selection process*

Study characteristics

Among the 23 studies included in the meta-analysis, all were designed in retrospective nature, except for five study in prospective nature. The studies' sample size ranged from 19 to 7807, including 11901 participants. Characteristics of studies entered into the systematic review presented in Table 1.

Table 1. Characteristics of studies entered into the systematic review

Study	Country	Study design	No. of Patients (Cases) (male/female)	Controls (male/female)	Mean (± SD) Median (IQR) Age of Patients (Cases)	Comorbidity of Patients (Cases)	Vitamin D Status of Patients (Cases)			Ethnicity of Patients (Cases)			Quality score**
							N	I	D	CS	AC	O	
J. H. Im et al. 2020 [13]	South Korea	Case-control study	50	150	57.5 (34.5-68.0)	-	13	-	37	-	-	-	7/9
Z. Maghbooli et al. 2020 [14]	Iran	Retrospective cross sectional	235	-	58.72 (±15.2) *mean	Diabetes: 86 Hypertension: 104 Respiratory disease: 72 Cancer: 2	77	-	-	-	-	-	7/10
V. Baktash et al. 2020 [15]	UK	Prospective cohort study	70 (42/28)	-	≥65	Hypertension: 34 Diabetes mellitus: 26 Ischemic heart disease: 15 Chronic respiratory disease: 13 Heart failure: 12 Stroke: 9 Dementia: 6 CKD: 16 Atrial fibrillation: 14 Cancer: 3 Endocrinological disease: 3	31	-	39	50	-	20	9/10
D. O. Meltzer et al. 2020 [16]	U.S.	Retrospective cohort study	71	-	-	Hypertension:261 Diabetes:137 COPD:117 Pulmonary circulation disorders: 20 Depression :119 CKD:116 Liver disease :56 Comorbidities with immunosuppression: 105	39	-	32	-	-	-	9/10
J. L. Faul et al. 2020 [17]	Ireland	Retrospective cross sectional	33 (33/0)	-	≥40	-	21	-	12	33	-	-	5/10

Study	Country	Study design	Cases (M/F)	Controls	Age	Comorbidities							Quality
E. Merzon et al. 2020 [18]	Israel	Case-control study	782 (385/397)	7,025 (2,849, 4,176)	35.58	Depression/Anxiety: 73 Schizophrenia: 15 Dementia: 27 Diabetes mellitus: 154 Hypertension: 174 Cardiovascular disease: 78 Chronic lung disorders: 66 Obesity: 235	79	598	105	-	-	-	6/9
G. Panagiotou et al. 2020 [19]	UK	Retrospective cross sectional	134 (73/61)	-	-	Hypertension: 56 Diabetes: 38 Obesity: 14 Malignancy: 15 Respiratory: 42 Cardiovascular disease: 20 Kidney and Liver diseases: 19	-	-	44	132	1	1	6/10
G. E. Carpagnano et al. 2020 [20]	Italy	Retrospective cohort study	42 (30/12)	-	65 (±13) *mean	Hypertension: 26 Cardiovascular disease: 16 CKD: 16 Diabetes type II: 11 Cerebrovascular disease: 5 Psychosis, depression, anxiety: 10 Malignancy: 5 COPD: 5 Asthma: 2	8	11	23	-	-	-	8/9
M. D. Nicola et al. 2020 [21]	Italy	Retrospective cohort study	112 (52/60)	-	47.2 (± 16.4)	-	-	-	-	-	-	-	6/9
F. Macaya et al. 2020 [22]	Spain	Retrospective cohort study	80 (35/45)	-	67.65 (50-84)	Hypertension: 50 Diabetes mellitus: 32 Cardiac disease: 19	-	-	45	-	-	-	7/9
S. Karahan et al. 2020 [23]	Turkey	Retrospective cohort study	149 (81/68)	-	63.5 (± 15.3)	Coronary artery disease: 32 Hypertension: 85 Dyslipidemia: 39 Diabetes mellitus: 61 Cerebrovascular accident: 9 COPD: 15 Malignancy: 23	12	34	103	-	-	-	8/9

Author	Country	Study design	Cases (M/F)	Controls (M/F)	Age	Comorbidities							Quality
		----				CKD: 29 Chronic atrial fibrillation: 15 Congestive heart failure: 18 Acute kidney injury: 16							
A. Abdollahi *et al.* 2020 [24]	Iran	Case-control study	201 (66/135)	201 (66/135)	48 (±16.95)	Hypothyroidism: 15 Diabetes mellitus: 42 Splenectomy: 1 Heart failure and hypertension: 20 Respiratory infections: 14 Autoimmune diseases: 11 AIDS: 4	39	161	1	-	-	-	7/9
C. Arvinte *et al.* 2020 [25]	U.S.	Prospective cohort study (pilot study)	21 (15/6)	-	60.2 (±17.4) 61 (20-94)	-	-	-	-	4	-	17	6/9
E. Cereda *et al.* 2020 [26]	Italy	Prospective cohort study	129 (70/59)	-	77 (65.0-85.0)	COPD: 16 Diabetes: 39 Hypertension: 89 Ischemic heart disease: 52 Cancer: 27 CKD: 24	-	30 #	99	-	-	-	7/9
A. Hamza *et al.* 2020 [27]	Pakistan	Randomized controlled trial study	168 (94/74)	-	42.26 (±13.69)	-	22	47	98	-	-	-	3/5
J. L. Hernandez *et al.* 2020 [28]	Spain	Case-control study	19 (7/12)	197 (123/74)	60.0 (59.0-75.0)	Hypertension: 12 Diabetes: 0 Cardiovascular disease: 3 COPD: 2 Active cancer: 0 Immunosuppression: 6	-	-	-	-	-	-	7/9
A. Jain *et al.* 2020 [29]	India	Prospective cohort study	154 (95/69)	-	46.05 (±8.8)	-	-	-	90	-	-	-	8/9
S. F. Ling *et al.* 2020 [30]	UK	Retrospective cohort study	444 (245/199)	-	74 (63-83)	Diabetes mellitus: 129 COPD: 100 Asthma: 52 IHD: 73 ACS: 48 Heart failure: 54	63	80	87	386	5	53	8/9

Study	Country	Study design			Age	Comorbidities							Quality
						Hypertension: 197							
						TIA: 40							
						Dementia: 59							
						Obesity: 20							
						Malignancy of solid organ: 71							
						Malignancy of skin: 8							
						Hematological malignancy: 8							
						Solid organ transplant: 4							
						Inflammatory arthritis: 16							
						Inflammatory bowel disease: 5							
X. Luo *et al.* 2020 [31]	China	Retrospective cross-sectional study	335 (148/187)	560 (257/303)	56.0 (43.0-64.0)	Comorbidity status: 147	-	-	218	-	-	-	7/10
A. Radujkovic *et al.* 2020 [32]	Germany	Retrospective cohort study	185 (95/90)	93 (59/34)	60 (49–70)	Cardiovascular disease: 58 Diabetes: 19 Chronic kidney disease: 8 Chronic lung disease: 15 Active or history of malignancy: 17	-	-	41	-	-	-	7/9
A. G. Vassiliou *et al.* 2020 [33]	Greece	Retrospective cohort study	39 (31/8)	-	61.17 (±13)	Hypertension: 18 COPD: 1 Hyperlipidemia: 9 Diabetes: 6 CAD: 4 Asthma: 1	-	7	32	-	-	-	6/9
K. Ye *et al.* 2020 [34]	China	Case-control study	62 (23/39)	80 (32/48)	43 (32–59)	Diabetes: 5 Hypertension; 6 Liver injury: 1 COPD: 1 Asthma: 0 Renal failure: 16	-	-	26	-	-	-	6/9
T. L. Karonova *et al.* 2020 [35]	Russia	Retrospective cohort study	80 (43/37)	-	53.2 (±15.7)	Obesity: 18 Ischemic heart disease: 21 Diabetes: 12	7	16	57	-	-	-	6/9

SD: Standard deviation, IQR: Interquartile range, U.S.: United States, UK: United Kingdom, N: Normal, I: Insufficient, D: Deficient, CS: Caucasian, AC: Afro-Caribbean, O: Other, COPD: Chronic obstructive pulmonary disease, CKD: Chronic Kidney Disease, AIDS: Acquired immunodeficiency syndrome, ACS: Acute coronary syndrome (Current or previous), TIA: Transient ischemic attack

In the study defined as patients with 25(OH)Vitamin D > 20 ng/mL

Quality assessment

Results of quality assessment for studies entered into meta-analysis were fair.

Publication bias

Begg's and Egger's tests findings was as follows for publication bias in main analysis: frequency of vitamin D status (P_B=0.38; P_E=0.02); mean 25(OH)D concentration (P_B=0.80; P_E=0.76); vitamin D deficiency and SARS-CoV-2 infection (P_B=1.00; P_E=0.55); Vitamin D deficiency and COVID-19 severity (P_B=0.12; P_E=0.14); and vitamin D deficiency and COVID-19 mortality (P_B=0.54; P_E=0.62).

Meta-analysis findings

Frequency of Vitamin D status in COVID-19 patients

The meta-analysis of event rates in peer reviewed papers showed that 41% of COVID-19 patients were suffering from vitamin D deficiency (95% CI, 29%-55%), in 42% of patients, levels of vitamin D were lower than the normal range (95% CI, 24%-63%), and only 19% of patients had normal vitamin D levels (95% CI, 11%-32%) (Fig. 2).
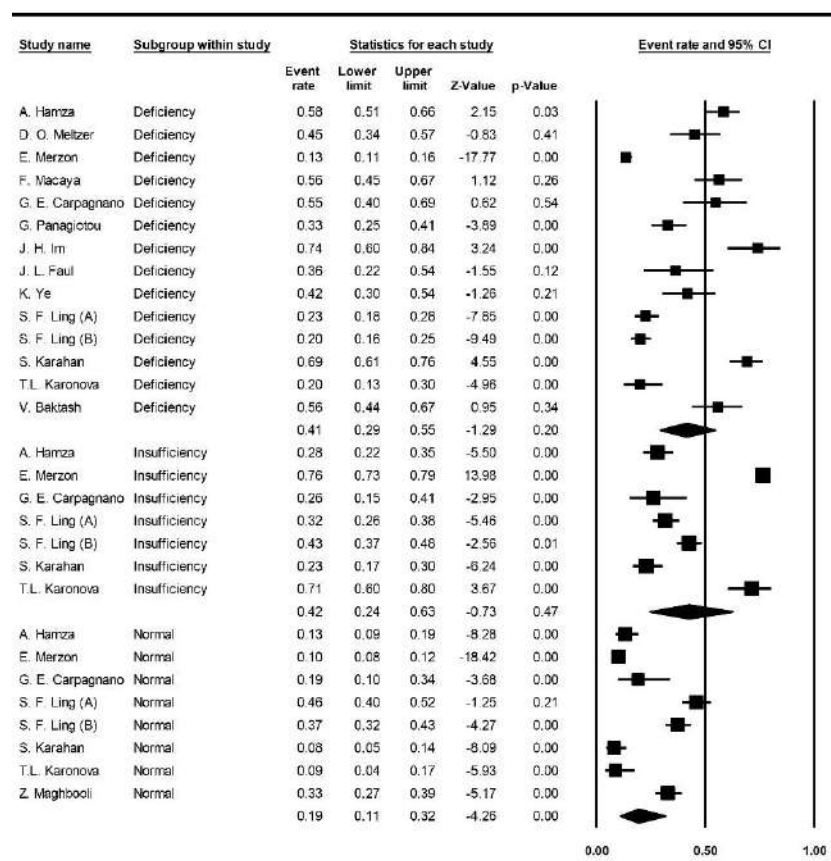
Figure 2. Forest plot for pooling events of vitamin D status

Mean serum 25-hydroxyvitamin D concentration

The meta-analysis of mean 25(OH)D concentration was 20.3 ng/mL among all COVID-19 patients (95% CI, 11.5-28.1), 16.0 ng/mL in severe cases (95% CI, 12.1-19.8) and 24.5 ng/mL in non-severe cases (95% CI, 20.0-29.0) (Fig. 3).
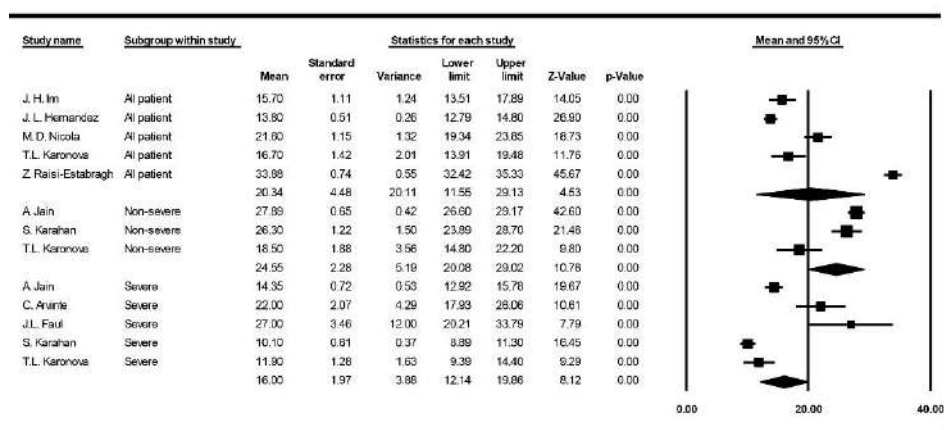

Figure 3. Forest plot for pooling mean 25(OH)D concentrations

Vitamin D Deficiency and SARS-CoV-2 infection

The meta-analysis indicated that odds of getting infected with SARS-CoV-2 increases by 3.3 times in individuals with vitamin D deficiency (95% CI, 2.5-4.3) (Fig. 4).
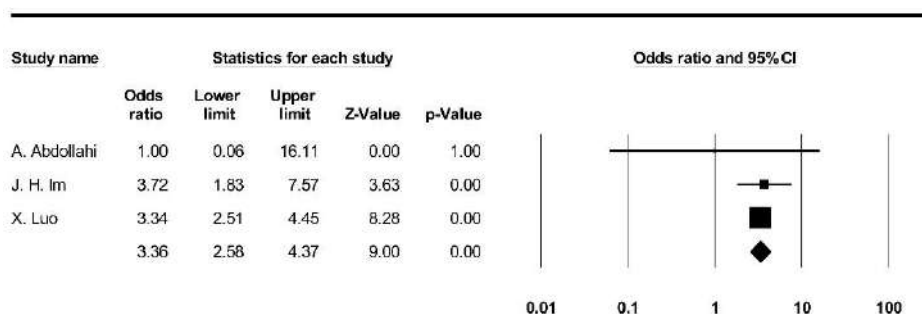

Figure 4. Forest plot for pooling odds ratios of vitamin D deficiency and SARS-CoV-2 infection

Vitamin D Deficiency and COVID-19 severity

The meta-analysis showed that the probability of developing severe stages of COVID-19 is 5.1 times higher in patients with vitamin D deficiency (95% CI, 2.6-10.3) (Fig. 5).
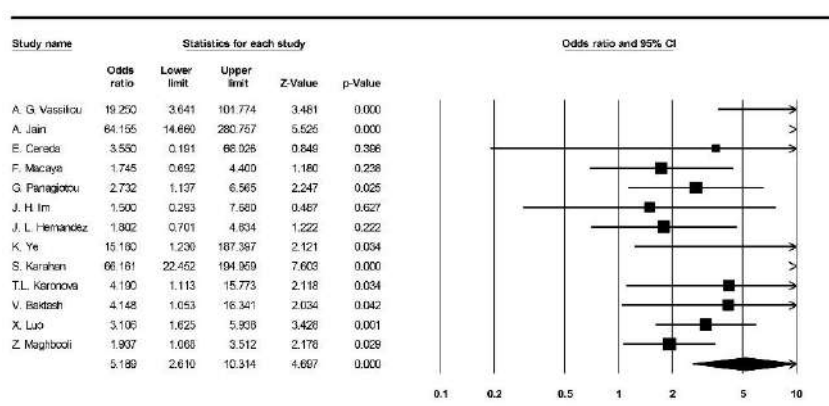

Figure 5. Forest plot for pooling odds ratios of vitamin D deficiency and COVID-19 severity

Vitamin D Deficiency and COVID-19 mortality

The meta-analysis indicated no significant higher COVID-19 mortality related to vitamin D deficient patients (OR: 1.6, 95% CI, 0.5-4.4) (Fig. 6).

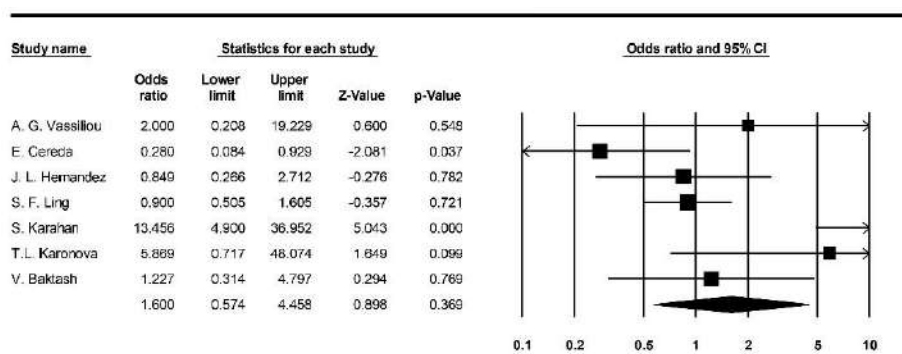| Study name	Statistics for each study					Odds ratio and 95% CI
	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value	
A. G. Vassiliou	2.000	0.208	19.229	0.600	0.549	
E. Cereda	0.280	0.084	0.929	-2.081	0.037	
J. L. Hernandez	0.849	0.266	2.712	-0.276	0.782	
S. F. Ling	0.900	0.505	1.605	-0.357	0.721	
S. Karahan	13.456	4.900	36.952	5.043	0.000	
T.L. Karonova	5.869	0.717	48.074	1.649	0.099	
V. Baktash	1.227	0.314	4.797	0.294	0.769	
	1.600	0.574	4.458	0.898	0.369	

Figure 6. Forest plot for pooling odds ratios of vitamin D deficiency and COVID-19 mortality

Co-morbidities

Meta-analysis of available data on co-morbidities frequency in COVID-19 patients were as follows: in non-severe cases, 13% cancer, 12% chronic kidney disease (CKD), 18% cardiovascular diseases (CVD), 21% diabetes, 29% hypertension (HTN), 12% obesity, and 13% respiratory diseases (Supplementary Figure 1); in severe cases, 13% cancer, 34% CKD, 31% CVD, 35% diabetes, 64% HTN, 33% obesity, and 17% respiratory diseases (Supplementary Figure 2); in overall, 8% cancer, 20% CKD, 26% CVD, 5% dementia, 15% depression/anxiety, 22% obesity, 26% diabetes, 49% HTN, and 15% respiratory diseases. (Supplementary Figure 3)

Ethnicity frequency

Pooling available data regarding ethnicity distribution among COVID-19 patients resulted in 2% Afro-Caribbean, 13% Asian, and 87% Caucasian (Supplementary Figure 4). The results for severe cases were as follows: 2% Asian, 68% Caucasian and 81% Hispanic (Supplementary Figure 5).

Discussion

Epidemiological & clinical aspects

Although comparing global statistics of COVID-19 outcomes is difficult, it is clear that the mortality rate is higher in several countries. It seems that among various factors such as age, healthcare system quality, general health status, socioeconomic status, etc. One of the underestimated factors that might be associated with COVID-19 outcome is the vitamin D status in every population. In recent years, vitamin D deficiency/insufficiency has become a global health issue, and its impact has been studied on respiratory viral infections. Most of the epidemiological studies have been reported a higher risk of developing the infection to the

severe stages and death in patients with low levels of vitamin D [36-39]. Besides, vitamin D clinical interventions have demonstrated a significantly reduced risk of respiratory tract infection (RTI), further proposed as a prophylactic or treatment approach against RTIs by WHO in 2017 [40-42].

Concerning all of the limitations and lack of high-quality data about the relation of vitamin D status and COVID-19 after several months, we have conducted this systematic review and meta-analysis to maximize the use of every available data, which would give us an overview toward further studies like what we have done recently on the effectiveness of hydroxychloroquine in COVID-19 patients [43], which have underestimated first, but the value was revealed after a while.

According to available data entered into our meta-analysis, we could find that approximately 43% of the patients infected with SARS-CoV-2 were suffering from vitamin D deficiency, and this vitamin was insufficient in about 42% of them. We have also found that mean 25(OH)D levels were low (~20 ng/mL) in all COVID-19 patients. More importantly, our analysis showed that the chance of infecting with SARS-CoV-2 is about three times higher in individuals with vitamin D deficiency and the probability of developing the severe disease in such patients is about 5 times higher than others. However, vitamin D deficiency did not substantially affected mortality rates in such patients.

These findings are in the same line with studies that have debated the association of vitamin D and COVID-19 [44-48]. Recently, Kaufman *et al.* [49], studied the relation of SARS-CoV-2 positivity rates with circulation 25(OH)D among 191,779 patients retrospectively. They found the highest SARS-CoV-2 positivity rate among patients with vitamin D deficiency (12.5%, 95% CI, 12.2-12.8%). Overall, the study indicated a significant inverse relation between SARS-CoV-2 positivity and circulating 25(OH)D levels in COVID-19 patients.

Along with all observational studies, a pilot randomized clinical trial performed by Castillo *et al.* [50] on 76 hospitalized COVID-19 patients indicated a promising result for calcifediol therapy in these individuals. In this study, high dose oral calcifediol significantly reduced the need for intensive care unit (ICU) treatment. However, due to the small sample size, more extensive, well-organized clinical trials are needed to robust and confirm this study's findings.

Additionally, in the case of vitamin D supplements' benefits against acute respiratory tract infections, Martineau *et al.* conducted a meta-analysis of randomized controlled on 10.933

participants and resulted in an inverse association between vitamin D levels and risk of acute respiratory tract infections. Thus, it can be concluded that patients with lower vitamin D levels or patients with vitamin D deficiency are at higher risk of developing the disease to the severe form [40].

Co-morbidities

After months of investigation on COVID-19, several factors such as male sex, older age, CVD, HTN, chronic lung disease, obesity, and CKD are proposed to be risk factors toward deteriorating COVID-19 patients' outcomes [51-54]. Interestingly, one of the conditions that lead to most of the considered risk factors is vitamin D deficiency. Studies indicated that malignancies, diabetes, HTN, and CVDs are significantly related to vitamin D deficiency. Also, studies reported the important role of vitamin D deficiency in older males [55-57]. Evidence shows that aging, physical activity, obesity, seasonal variation, less vitamin D absorption, pregnancy, thyroid disorders, prolonged use of corticosteroids, ethnicity/race can substantially affect the circulating 25(OH)D levels [58-64].

Hence, although studies reported vitamin D deficiency as one of the critical risk factors in clinical outcomes of COVID-19 patients, it seems that it can also be in a strong relationship with basic underlying risk factors and diseases in such patients.

In this case, our analyses indicated that HTN, CVDs, CKDs, diabetes, obesity, and respiratory diseases were the most frequent co-morbidities in COVID-19 patients. According to the facts mentioned above and our findings, it is plausible that both vitamin D deficiency and underlying diseases, which affect each other, may worsen the condition of these patients more than others.

Ethnicity

From the beginning of the COVID-19 pandemic, different studies have been reported probable associations between COVID-19 and the ethnicity of these patients. Most studies found that the mortality rate among black people is higher than the other ethnic groups [65-69]. However, other challenges such as human resources, health care systems budgetary, poor management, etc. have to be considered among such people and low-income countries [70-72], which unavoidably affects the subject significantly. In recent years, many studies have focused on vitamin D mechanisms and status among various ethnic groups to find the roles of vitamin D and its relationships with any factors or disorders in various ethnicities [73-76].

Herein, our findings demonstrated that the most frequent ethnic group has belonged to Caucasians, followed by Hispanic, Asian and Afro-Caribbean. Although there is some evidence on the role of genetic variants in COVID-19 patients, the subject is still not clear enough [77,78].

In contrast to many studies about vitamin D status in different ethnicities, Aloia *et al.* have reported that serum 25(OH)D concentration is the same in cross-racial comparison. They found an inconsistency between monoclonal and polyclonal assays for detecting vitamin D binding protein [79]. Hence, the approach for considering serum 25(OH)D concentration is much important.

Vitamin D mechanisms & COVID-19

Vitamin D metabolism has been well studied throughout history. Numerous investigations indicate vitamin D's roles in reducing microbial infections through a physical barrier, natural immunity, and adaptive immunity [2,80-85]. For example, investigations on respiratory infections indicated that 25(OH)D could effectively induce the host defense peptides against bacterial or viral agents. Vitamin D insufficiency/deficiency can lead to non-communicable and infectious diseases [2,86,87]. The other potential role of vitamin D is reducing inflammatory induced following SARS-CoV-2 infection by suppressing inflammatory cytokines, reducing leukocytes' infiltration, interaction with polymorphonuclear leukocytes, and inhibiting complement component C3 [36,88-92]. Also, according to the available evidence for infections and malignancies [93,94], vitamin D may enhance the serological response and $CD8^+$ T lymphocytes performance against COVID-19 when the T cells' exhaustion is related to the critical stages of the disease [95-97].

Besides, according to the revealed association of SARS-CoV-2 and angiotensin-converting enzyme 2 (ACE2), this virus can substantially downregulate the ACE2 expression, which seems to leads the COVID-19 patients to deterioration [98-100]. In contrast, vitamin D affects the renin-angiotensin system pathway and promotes the expression of ACE2 [101,102]. However, since the high expression of ACE2 can be a risk factor for the severity of the disease [103], it is not yet clear enough to conclude how much vitamin D helps the condition. Hence, more evidence and trials are needed to design a treatment plan for three groups of mild, moderate, and severe patients.

It is worth noticing that the current meta-analysis includes the following limitations:1) most of studies entered into the meta-analysis were retrospective in nature; 2) There are inevitable

challenges with the reliability of data due to different strategies in a testing (e.g., vitamin D measurement, COVID-19 test, etc.), various subpopulations, etc.; 3) other immunomodulatory factors (e.g., vitamin C, zinc, selenium, etc.), which might be influential in the outcome of COVID-19 patients, have not considered in included studies; and 4) type *II* statistical errors following studies with small sample size. Eventually, to overcome the limitations and bias, the study's results should be confirmed by robustly large multicentral randomized clinical trials.

Conclusion

The conditional evidence recommends that vitamin D might be a critical supportive agent for the immune system, mainly in cytokine response regulation against pathogens. In this systematic review and meta-analysis, we found that mean serum 25(OH)D level was low (~20 ng/mL) in all COVID-19 patients and most of them were suffering from vitamin D deficiency/insufficiency. Also, there is about three times higher chance of getting infected with SARS-CoV-2 among vitamin D deficient individuals and 5 times higher probability of developing the severe disease in such patients. Vitamin D deficiency showed no significant association with mortality rates in these population. The Caucasian was the dominant ethnic group, and the most frequent co-morbidities in COVID-19 patients were HTN, CVDs, CKDs, diabetes, obesity, and respiratory diseases, which might be affected by vitamin D deficiency directly or indirectly. However, further large clinical trials following comprehensive meta-analysis should be taken into account to achieve more reliable findings.

Declarations

Ethics approval and consent to participate

Not applicable.

Consent for publication

Not applicable.

Availability of data and material

Not applicable.

Competing interests

The authors declare that they have no competing interests.

References:

1. Organization WH. Coronavirus disease (COVID-2019) situation reports. In: Geneva: WHO; 2020.

2. Greiller CL, Martineau AR. Modulation of the immune response to respiratory viruses by vitamin D. *Nutrients.* 2015;7(6):4240-4270.

3. Zdrenghea MT, Makrinioti H, Bagacean C, Bush A, Johnston SL, Stanciu LA. Vitamin D modulation of innate immune responses to respiratory viral infections. *Reviews in medical virology.* 2017;27(1).

4. Mercola J, Grant WB, Wagner CL. Evidence Regarding Vitamin D and Risk of COVID-19 and Its Severity. *Nutrients.* 2020;12(11):3361.

5. Aranow C. Vitamin D and the immune system. *Journal of investigative medicine : the official publication of the American Federation for Clinical Research.* 2011;59(6):881-886.

6. Goncalves-Mendes N, Talvas J, Dualé C, et al. Impact of Vitamin D Supplementation on Influenza Vaccine Response and Immune Functions in Deficient Elderly Persons: A Randomized Placebo-Controlled Trial. *Frontiers in immunology.* 2019;10:65.

7. Grant WB, Giovannucci E. The possible roles of solar ultraviolet-B radiation and vitamin D in reducing case-fatality rates from the 1918-1919 influenza pandemic in the United States. *Dermato-endocrinology.* 2009;1(4):215-219.

8. Wells GA, Shea B, O'Connell D, et al. The Newcastle-Ottawa Scale (NOS) for assessing the quality of nonrandomised studies in meta-analyses. In: Oxford; 2000.

9. Modesti PA, Reboldi G, Cappuccio FP, et al. Panethnic differences in blood pressure in Europe: a systematic review and meta-analysis. *PloS one.* 2016;11(1):e0147601.

10. Halpern SH, Douglas MJ. Appendix: Jadad scale for reporting randomized controlled trials. *Evidence-based Obstetric Anesthesia Oxford, UK: Blackwell Publishing Ltd.* 2005:237-238.

11. Giustina A, Adler RA, Binkley N, et al. Controversies in vitamin D: summary statement from an international conference. *The Journal of Clinical Endocrinology & Metabolism.* 2019;104(2):234-240.

12. Higgins JPT, Thomas J, Chandler J, et al. *Cochrane handbook for systematic reviews of interventions version 6.0.* 2nd Edition ed: John Wiley & Sons; 2019.

13. Im JH, Je YS, Baek J, Chung MH, Kwon HY, Lee JS. Nutritional status of patients with COVID-19. *International journal of infectious diseases : IJID : official publication of the International Society for Infectious Diseases.* 2020;100:390-393.

14. Maghbooli Z, Sahraian MA, Ebrahimi M, 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.* 2020;15(9):e0239799.

15. Baktash V, Hosack T, Patel N, et al. Vitamin D status and outcomes for hospitalised older patients with COVID-19. *Postgraduate medical journal.* 2020.

16. Meltzer DO, Best TJ, Zhang H, Vokes T, Arora V, Solway J. Association of Vitamin D Status and Other Clinical Characteristics With COVID-19 Test Results. *JAMA network open.* 2020;3(9):e2019722.

17. Faul JL, Kerley CP, Love B, et al. Vitamin D Deficiency and ARDS after SARS-CoV-2 Infection. *Ir Med J.* 2020;113(5):84.

18. Merzon E, Tworowski D, Gorohovski A, et al. Low plasma 25(OH) vitamin D level is associated with increased risk of COVID-19 infection: an Israeli population-based study. *The FEBS journal.* 2020;287(17):3693-3702.

19. Panagiotou G, Tee SA, Ihsan Y, et al. Low serum 25-hydroxyvitamin D (25[OH]D) levels in patients hospitalized with COVID-19 are associated with greater disease severity. *Clinical endocrinology.* 2020;93(4):508-511.

20. Carpagnano GE, Di Lecce V, Quaranta VN, et al. Vitamin D deficiency as a predictor of poor prognosis in patients with acute respiratory failure due to COVID-19. *Journal of endocrinological investigation.* 2020:1-7.

21. Di Nicola M, Dattoli L, Moccia L, et al. Serum 25-hydroxyvitamin D levels and psychological distress symptoms in patients with affective disorders during the COVID-19 pandemic. *Psychoneuroendocrinology.* 2020;122:104869.

22. Macaya F, Espejo Paeres C, Valls A, et al. Interaction between age and vitamin D deficiency in severe COVID-19 infection. *Nutricion hospitalaria.* 2020;37(5):1039-1042.

23. Karahan S, Katkat F. Impact of Serum 25(OH) Vitamin D Level on Mortality in Patients with COVID-19 in Turkey. *Journal of Nutrition, Health and Aging.* 2020.

24. Abdollahi A, Sarvestani HK, Rafat Z, et al. The Association Between the Level of Serum 25(OH) Vitamin D, Obesity, and underlying Diseases with the risk of Developing COVID-19 Infection: A case-control study of hospitalized patients in Tehran, Iran. *Journal of medical virology.* 2020.

25. Arvinte C, Singh M, Marik PE. Serum Levels of Vitamin C and Vitamin D in a Cohort of Critically Ill COVID-19 Patients of a North American Community Hospital Intensive Care Unit in May 2020: A Pilot Study. *Medicine in Drug Discovery.* 2020;8.

26. Cereda E, Bogliolo L, Klersy C, et al. Vitamin D 25OH deficiency in COVID-19 patients admitted to a tertiary referral hospital. *Clinical nutrition (Edinburgh, Scotland).* 2020.

27. Hamza A, Ahmed M, Ahmed K, Durrani AB. Role of Vitamin D in Pathogenesis and Severity of Coronavirus Disease 2019 (COVID-19) Infection. *Pakistan Journal of Medical and Health Sciences.* 2020;14(2):462-465.

28. Hernández JL, Nan D, Fernandez-Ayala M, et al. Vitamin D Status in Hospitalized Patients with SARS-CoV-2 Infection. *The Journal of clinical endocrinology and metabolism.* 2020.

29. 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. *Scientific reports.* 2020;10(1):20191.

30. Ling SF, Broad E, Murphy R, et al. High-Dose Cholecalciferol Booster Therapy is Associated with a Reduced Risk of Mortality in Patients with COVID-19: A Cross-Sectional Multi-Centre Observational Study. *Nutrients.* 2020;12(12).

31. Luo X, Liao Q, Shen Y, Li H, Cheng L. Vitamin D Deficiency Is Inversely Associated with COVID-19 Incidence and Disease Severity in Chinese People. *The Journal of nutrition.* 2020.

32. 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(9).

33. Vassiliou AG, Jahaj E, Pratikaki M, et al. Vitamin D deficiency correlates with a reduced number of natural killer cells in intensive care unit (ICU) and non-ICU

patients with COVID-19 pneumonia. *Hellenic journal of cardiology : HJC = Hellenike kardiologike epitheorese.* 2020.

34. Ye K, Tang F, Liao X, et al. Does Serum Vitamin D Level Affect COVID-19 Infection and Its Severity?-A Case-Control Study. *Journal of the American College of Nutrition.* 2020.

35. Karonova T, Andreeva A, Vashukova M. Serum 25 (OH) D level in patients with COVID-19. *Journal Infectology.* 2020;12(3):21-27.

36. Teymoori-Rad M, Shokri F, Salimi V, Marashi SM. The interplay between vitamin D and viral infections. *Reviews in medical virology.* 2019;29(2):e2032.

37. McNally JD, Leis K, Matheson LA, Karuananyake C, Sankaran K, Rosenberg AM. Vitamin D deficiency in young children with severe acute lower respiratory infection. *Pediatric pulmonology.* 2009;44(10):981-988.

38. Belderbos ME, Houben ML, Wilbrink B, et al. Cord blood vitamin D deficiency is associated with respiratory syncytial virus bronchiolitis. *Pediatrics.* 2011;127(6):e1513-e1520.

39. Inamo Y, Hasegawa M, Saito K, et al. Serum vitamin D concentrations and associated severity of acute lower respiratory tract infections in Japanese hospitalized children. *Pediatrics international : official journal of the Japan Pediatric Society.* 2011;53(2):199-201.

40. Martineau AR, Jolliffe DA, Hooper RL, et al. Vitamin D supplementation to prevent acute respiratory tract infections: systematic review and meta-analysis of individual participant data. *BMJ (Clinical research ed).* 2017;356:i6583.

41. Bergman P, Lindh AU, Björkhem-Bergman L, Lindh JD. Vitamin D and Respiratory Tract Infections: A Systematic Review and Meta-Analysis of Randomized Controlled Trials. *PloS one.* 2013;8(6):e65835.

42. Aponte R, MSHN, Palacios C. Vitamin D for prevention of respiratory tract infections. *WHO; e-Library of Evidence for Nutrition Actions (eLENA).* 2017.

43. Shamshirian A, Hessami A, Heydari K, et al. The Role of Hydroxychloroquine in COVID-19: A Systematic Review and Meta-Analysis. *Annals of the Academy of Medicine, Singapore.* 2020;49(10).

44. Carter SJ, Baranauskas MN, Fly AD. Considerations for Obesity, Vitamin D, and Physical Activity Amid the COVID-19 Pandemic. *Obesity.* 2020;28(7):1176-1177.

45.   Ilie PC, Stefanescu S, Smith L. The role of vitamin D in the prevention of coronavirus disease 2019 infection and mortality. *Aging Clinical and Experimental Research.* 2020;32(7):1195-1198.

46.   Jakovac H. COVID-19 and vitamin D—Is there a link and an opportunity for intervention? *American Journal of Physiology-Endocrinology and Metabolism.* 2020;318(5):E589-E589.

47.   Molloy E, Murphy. Vitamin D, Covid-19 and Children. *Ir Med J.* 2020;113:64.

48.   Zemb P, Bergman P, Camargo CA, Jr., et al. Vitamin D deficiency and the COVID-19 pandemic. *Journal of global antimicrobial resistance.* 2020;22:133-134.

49.   Kaufman HW, Niles JK, Kroll MH, Bi C, Holick MF. SARS-CoV-2 positivity rates associated with circulating 25-hydroxyvitamin D levels. *PloS one.* 2020;15(9):e0239252.

50.   Entrenas Castillo M, Entrenas Costa LM, Vaquero Barrios JM, et al. "Effect of calcifediol treatment and best available therapy versus best available therapy on intensive care unit admission and mortality among patients hospitalized for COVID-19: A pilot randomized clinical study". *J Steroid Biochem Mol Biol.* 2020;203:105751-105751.

51.   Holman N, Knighton P, Kar P, et al. Risk factors for COVID-19-related mortality in people with type 1 and type 2 diabetes in England: a population-based cohort study. *The lancet Diabetes & endocrinology.* 2020;8(10):823-833.

52.   Centers for Disease C, Prevention. People with certain medical conditions. 2020.

53.   Preliminary Estimates of the Prevalence of Selected Underlying Health Conditions Among Patients with Coronavirus Disease 2019 - United States, February 12-March 28, 2020. *MMWR Morbidity and mortality weekly report.* 2020;69(13):382-386.

54.   Hessami A, Shamshirian A, Heydari K, et al. Cardiovascular diseases burden in COVID-19: Systematic review and meta-analysis. *The American journal of emergency medicine.* 2020.

55.   Orwoll E, Nielson CM, Marshall LM, et al. Vitamin D deficiency in older men. *The Journal of clinical endocrinology and metabolism.* 2009;94(4):1214-1222.

56.   Mosekilde L. Vitamin D and the elderly. *Clin Endocrinol (Oxf).* 2005;62(3):265-281.

57.   La Vignera S, Cannarella R, Condorelli RA, Torre F, Aversa A, Calogero AE. Sex-Specific SARS-CoV-2 Mortality: Among Hormone-Modulated ACE2 Expression, Risk of Venous Thromboembolism and Hypovitaminosis D. *International journal of molecular sciences.* 2020;21(8).

58.   Yang C-Y, Leung PSC, Adamopoulos IE, Gershwin ME. The implication of vitamin D and autoimmunity: a comprehensive review. *Clin Rev Allergy Immunol.* 2013;45(2):217-226.

59.   Vranić L, Mikolašević I, Milić S. Vitamin D Deficiency: Consequence or Cause of Obesity? *Medicina (Kaunas).* 2019;55(9):541.

60.   Kim D. The Role of Vitamin D in Thyroid Diseases. *International journal of molecular sciences.* 2017;18(9):1949.

61.   Kdekian A, Alssema M, Van Der Beek EM, et al. Impact of isocaloric exchanges of carbohydrate for fat on postprandial glucose, insulin, triglycerides, and free fatty acid responses-a systematic review and meta-analysis. *European journal of clinical nutrition.* 2020;74(1):1-8.

62.   Cashman KD, Dowling KG, Škrabáková Z, et al. Vitamin D deficiency in Europe: pandemic? *The American journal of clinical nutrition.* 2016;103(4):1033-1044.

63.   Weishaar T, Rajan S, Keller B. Probability of Vitamin D Deficiency by Body Weight and Race/Ethnicity. *Journal of the American Board of Family Medicine : JABFM.* 2016;29(2):226-232.

64.   Correia A, Azevedo Mdo S, Gondim F, Bandeira F. Ethnic aspects of vitamin D deficiency. *Arquivos brasileiros de endocrinologia e metabologia.* 2014;58(5):540-544.

65.   Yancy CW. COVID-19 and African Americans. *Jama.* 2020.

66.   Milam AJ, Furr-Holden D, Edwards-Johnson J, et al. Are Clinicians Contributing to Excess African American COVID-19 Deaths? Unbeknownst to Them, They May Be. *Health equity.* 2020;4(1):139-141.

67.   Laurencin CT, McClinton A. The COVID-19 Pandemic: a Call to Action to Identify and Address Racial and Ethnic Disparities. *Journal of racial and ethnic health disparities.* 2020;7(3):398-402.

68.   Khunti K, Singh AK, Pareek M, Hanif W. Is ethnicity linked to incidence or outcomes of covid-19? *BMJ (Clinical research ed).* 2020;369:m1548.

69.   Hastie CE, Mackay DF, Ho F, et al. Vitamin D concentrations and COVID-19 infection in UK Biobank. *Diabetes & metabolic syndrome.* 2020;14(4):561-565.

70.   Moszynski P. WHO report highlights Africa's health challenges. *BMJ (Clinical research ed).* 2006;333(7578):1088.

71.   Oleribe OO, Momoh J, Uzochukwu BS, et al. Identifying Key Challenges Facing Healthcare Systems In Africa And Potential Solutions. *International journal of general medicine.* 2019;12:395-403.

72.   Organization WH. The African regional health report 2014: The health of the people. 2014.

73.   Yuen AW, Jablonski NG. Vitamin D: in the evolution of human skin colour. *Medical hypotheses.* 2010;74(1):39-44.

74.   Word AP, Perese F, Tseng LC, Adams-Huet B, Olsen NJ, Chong BF. 25-Hydroxyvitamin D levels in African-American and Caucasian/Hispanic subjects with cutaneous lupus erythematosus. *The British journal of dermatology.* 2012;166(2):372-379.

75.   Powe CE, Evans MK, Wenger J, et al. Vitamin D-binding protein and vitamin D status of black Americans and white Americans. *The New England journal of medicine.* 2013;369(21):1991-2000.

76.   Bikle D, Christakos S. New aspects of vitamin D metabolism and action - addressing the skin as source and target. *Nature reviews Endocrinology.* 2020;16(4):234-252.

77.   Darbeheshti F, Rezaei N. Genetic predisposition models to COVID-19 infection. *Medical hypotheses.* 2020;142:109818.

78.   The COVID-19 Host Genetics Initiative, a global initiative to elucidate the role of host genetic factors in susceptibility and severity of the SARS-CoV-2 virus pandemic. *European journal of human genetics : EJHG.* 2020;28(6):715-718.

79.   Aloia J, Mikhail M, Dhaliwal R, et al. Free 25(OH)D and the Vitamin D Paradox in African Americans. *The Journal of clinical endocrinology and metabolism.* 2015;100(9):3356-3363.

80.   Holick MF. Vitamin D deficiency. *The New England journal of medicine.* 2007;357(3):266-281.

81.   Hewison M. An update on vitamin D and human immunity. *Clin Endocrinol (Oxf).* 2012;76(3):315-325.

82.   Beard JA, Bearden A, Striker R. Vitamin D and the anti-viral state. *Journal of clinical virology : the official publication of the Pan American Society for Clinical Virology.* 2011;50(3):194-200.

83.   Abhimanyu, Coussens AK. The role of UV radiation and vitamin D in the seasonality and outcomes of infectious disease. *Photochemical & photobiological sciences :*

Official journal of the European Photochemistry Association and the European Society for Photobiology. 2017;16(3):314-338.

84.   Rondanelli M, Miccono A, Lamburghini S, et al. Self-Care for Common Colds: The Pivotal Role of Vitamin D, Vitamin C, Zinc, and Echinacea in Three Main Immune Interactive Clusters (Physical Barriers, Innate and Adaptive Immunity) Involved during an Episode of Common Colds-Practical Advice on Dosages and on the Time to Take These Nutrients/Botanicals in order to Prevent or Treat Common Colds. *Evidence-based complementary and alternative medicine : eCAM.* 2018;2018:5813095.

85.   Grant WB, Boucher BJ. Yes, vitamin D can be a magic bullet. *Clinical Nutrition.* 2020;39(5):1627.

86.   Hansdottir S, Monick MM, Hinde SL, Lovan N, Look DC, Hunninghake GW. Respiratory epithelial cells convert inactive vitamin D to its active form: potential effects on host defense. *Journal of immunology (Baltimore, Md : 1950).* 2008;181(10):7090-7099.

87.   Olliver M, Spelmink L, Hiew J, Meyer-Hoffert U, Henriques-Normark B, Bergman P. Immunomodulatory effects of vitamin D on innate and adaptive immune responses to Streptococcus pneumoniae. *The Journal of infectious diseases.* 2013;208(9):1474-1481.

88.   Grant WB, Lahore H, McDonnell SL, et al. Evidence that Vitamin D Supplementation Could Reduce Risk of Influenza and COVID-19 Infections and Deaths. *Nutrients.* 2020;12(4):988.

89.   Xu H, Soruri A, Gieseler RKH, Peters JH. 1, 25- Dihydroxyvitamin D3 exerts opposing effects to IL- 4 on MHC class- II antigen expression, accessory activity, and phagocytosis of human monocytes. *Scandinavian journal of immunology.* 1993;38(6):535-540.

90.   Hirsch D, Archer FE, Joshi-Kale M, Vetrano AM, Weinberger B. Decreased anti-inflammatory responses to vitamin D in neonatal neutrophils. *Mediators of inflammation.* 2011;2011.

91.   Risitano AM, Mastellos DC, Huber-Lang M, et al. Complement as a target in COVID-19? *Nature reviews Immunology.* 2020;20(6):343-344.

92.   Gralinski LE, Sheahan TP, Morrison TE, et al. Complement Activation Contributes to Severe Acute Respiratory Syndrome Coronavirus Pathogenesis. *mBio.* 2018;9(5).

93.   Karkeni E, Morin SO, Bou Tayeh B, et al. Vitamin D Controls Tumor Growth and CD8+ T Cell Infiltration in Breast Cancer. *Frontiers in immunology.* 2019;10:1307.

94.   Chadha MK, Fakih M, Muindi J, et al. Effect of 25-hydroxyvitamin D status on serological response to influenza vaccine in prostate cancer patients. *The Prostate.* 2011;71(4):368-372.

95.   Zheng M, Gao Y, Wang G, et al. Functional exhaustion of antiviral lymphocytes in COVID-19 patients. *Cellular & molecular immunology.* 2020;17(5):533-535.

96.   Moon C. Fighting COVID-19 exhausts T cells. *Nature reviews Immunology.* 2020;20(5):277.

97.   Minton K. DAMP-driven metabolic adaptation. *Nature reviews Immunology.* 2020;20(1):1.

98.   Xiao L, Sakagami H, Miwa N. ACE2: The key Molecule for Understanding the Pathophysiology of Severe and Critical Conditions of COVID-19: Demon or Angel? *Viruses.* 2020;12(5):491.

99.   van de Veerdonk F, Netea MG, van Deuren M, et al. Kinins and cytokines in COVID-19: a comprehensive pathophysiological approach. 2020.

100.   Imai Y, Kuba K, Rao S, et al. Angiotensin-converting enzyme 2 protects from severe acute lung failure. *Nature.* 2005;436(7047):112-116.

101.   Mitchell F. Vitamin-D and COVID-19: do deficient risk a poorer outcome? *The Lancet Diabetes & Endocrinology.* 2020.

102.   Machado CDS, Ferro Aissa A, Ribeiro DL, Antunes LMG. Vitamin D supplementation alters the expression of genes associated with hypertension and did not induce DNA damage in rats. *Journal of toxicology and environmental health Part A.* 2019;82(4):299-313.

103.   Gracia-Ramos AE. Is the ACE2 Overexpression a Risk Factor for COVID-19 Infection? *Archives of medical research.* 2020;51(4):345-346.