Nicholas Tentolouris (Orcid ID: 0000-0003-0615-2534)

Georgia Samakidou (Orcid ID: 0000-0001-5567-8044)

The effect of vitamin D supplementation on mortality and Intensive Care Unit admission of COVID-19 patients. A systematic review, meta-analysis and meta-regression

Short running title: Vitamin D supplementation and COVID-19

Nikolaos Tentolouris[1], Georgia Samakidou[1], Ioanna Eleftheriadou[1], Anastasios Tentolouris[1] and Edward B Jude[2]

[1]Diabetes Centre, First Department of Propaedeutic Internal Medicine, Medical School, National and Kapodistrian University of Athens, Laiko General Hospital, 17 Agiou Thoma Street, 11527, Athens, Greece

[2]Tameside and Glossop Integrated Care NHS Foundation Trust, Fountain Street, Ashton-under-Lyne OL6 9RW, UK

Corresponding author: Prof Nikolaos Tentolouris, Diabetes Centre, First Department of Propaedeutic Internal Medicine, Medical School, National and Kapodistrian University of Athens, Laiko General Hospital, 17 Agiou Thoma Street, 11527, Athens, Greece, Telephone: +30 213 2061 061, Fax: +30 213 2061 794, e-mail: ntentol@med.uoa.gr

E-mail addresses of all authors:

Nikolaos Tentolouris (ntentol@med.uoa.gr)

This article has been accepted for publication and undergone full peer review but has not been through the copyediting, typesetting, pagination and proofreading process, which may lead to differences between this version and the Version of Record. Please cite this article as doi: 10.1002/dmrr.3517.

Georgia Samakidou (geo.samak@gmail.com)

Ioanna Eleftheriadou (joeleftheriadou@yahoo.com)

Anastasios Tentolouris (antentolouris@hotmail.com)

Edward B Jude (Edward.Jude@tgh.nhs.uk)

Word counts for abstract: 240

Word counts for main text: 3655

Number of references: 48

Number of tables: 3

Number of figures: 2

Abstract

Aims: The aim of this systematic review and meta-analysis was to investigate the effect of vitamin D supplementation on mortality and admission to intensive care unit (ICU) of COVID-19 patients.

Methods: A systematic search of PubMed, Google Scholar, Embase, Web of Science and medRxiv with terms relative to vitamin D supplementation and COVID-19 was conducted on March, 26th, 2021. Comprehensive Meta-Analysis software was used for the quantitative assessment of data and random-effects model was applied. To investigate the association between the dose of vitamin D and the outcomes of interest, meta-regression analysis was performed.

Results: 2,078 patients from 9 studies with data on mortality were included (583 received vitamin D supplementation, while 1,495 did not). 61 (10.46%) individuals in the treated group died, compared to 386 (25.81%) in the non-treated group [odds ratio (OR): 0.597; 95% CI: 0.318-1.121; p=0.109]. 860 patients from 6 studies with data on ICU admission were included (369 received vitamin D supplementation, while 491 did not). 45 (12.19%) individuals in the treated group were admitted to ICU, compared to 129 (26.27%) in the non-treated group (OR:

0.326; 95%CI: 0.149-0.712; p=0.005). No significant linear relationship between vitamin D dose and log OR of mortality or log OR of ICU admission was observed.

Conclusion: This meta-analysis indicates a beneficial role of vitamin D supplementation on ICU admission, but not on mortality, of COVID-19 patients. Further research is urgently needed to understand the benefit of vitamin D in Covid-19.

Keywords

COVID-19, Vitamin D, cholecalciferol, calcifediol, mortality, intensive care unit

Introduction

In late December 2019, the first cases of coronavirus disease 2019 (COVID-19), a disease caused by a novel beta-coronavirus named "severe acute respiratory syndrome coronavirus 2" (SARS-CoV-2), were reported in Wuhan, China. By March 2020, the disease had already spread globally, leading to the declaration of a pandemic by the World Health Organization (WHO).[1] Since then, the global impact of COVID-19 has undoubtedly been tremendous, and until 29 May 2021 there have been approximately 173 million cases and 3.72 million deaths from COVID-19.[2]

The clinical manifestations of COVID-19 range from asymptomatic or mild cases with fever, dry cough and fatigue, to severe and even critical cases with dyspnoea, need for Intensive Care Unit (ICU) admission, acute respiratory distress syndrome (ARDS) and multi-organ failure and death.[1] Some of the risk factors that have been associated with COVID-19 severity are older age, black ethnicity, institutionalization, immunodeficiency, chronic kidney disease, chronic metabolic diseases (including diabetes) and obesity.[3,4] Interestingly, several of these factors have also been associated with increased risk of vitamin D deficiency.[5,6]

The link between vitamin D deficiency and COVID-19 positivity rates and severity has been investigated in several observational studies,[7-17] as well as in systematic reviews and meta-analyses.[18-22] Despite the inconsistency of the results and the need for their critical appraisal due to several reasons (including different designs of the studies and distinct population characteristics, presence of confounding factors and inability to establish causation), the growing amount of evidence points towards a link between serum 25-hydroxyvitamin D [25(OH)D] levels and the risk of infection and disease severity from SARS-CoV-2.[4]

All these observations led to the research question of whether vitamin D supplementation could improve the clinical outcomes of COVID-19 patients and reduce the risk of severe disease and mortality. Some studies have been published to address this question; however, the results are inconsistent. The aim of this systematic review and meta-analysis was to accumulate the existing evidence and investigate the effect of vitamin D supplementation on mortality and need for ICU admission of COVID-19 patients. In addition, using meta-regression analysis, we examined whether the dose of vitamin D after diagnosis of COVID-19 was associated with either mortality or need for ICU admission.

Methods

This systematic review and meta-analysis was conducted according to the recommendations of the Preferred Reporting Items for Systematic Reviews and Meta-Analysis (PRISMA)[23]. The PICOS approach was used for the development of the research questions (Supplementary Table 1).

Eligibility

The population of interest was adult patients with COVID-19 receiving any form of vitamin D supplementation.

Information sources and search strategy

A comprehensive search of PubMed, Google Scholar, Embase, Web of Science and medRxiv with terms relative to vitamin D supplementation and COVID-19 patients was conducted on March26th, 2021, without limitations in publication dates. The search strategy for PubMed was: ("Vitamin D" [Mesh] OR "Vitamin D" OR "25(OH)D" OR "25-hydroxyvitamin D" OR "cholecalciferol*" OR "ergocalciferol*"OR “calcifediol*”) AND ("COVID-19" [Mesh] OR "COVID-19" OR "SARS-CoV-2" OR "Coronavirus disease").

Outcomes

The outcomes of interest were mortality and ICU admissions.

Study selection and data extraction process

Randomized trials and certain observational studies (case-control, cross-sectional and observational cohort) involving vitamin D supplementation and reporting on the selected outcomes were included in this systematic review. Articles with distinct features (e.g., clinical case series, case reports, animal or laboratory studies, reviews, non-English articles) and studies not involving vitamin D supplementation were excluded. Two independent researchers (GS and IE) screened the results by titles and abstracts and assessed the selected full-text articles for eligibility. Any disagreements were resolved with re-evaluation and consensus. After the final assessment, 10 records were selected for qualitative and quantitative synthesis, and data from

the selected studies were extracted. Of the 10 studies selected, 2 of them were randomized studies [24,25] and 8 were non-randomized studies.[26-33] In case of missing information from certain studies, corresponding authors were contacted. The detailed PRISMA chart is available in Supplementary Figure 1 and the characteristics of the included studies in Table 1 and Supplementary table 2.

In addition, we calculated the dose of Vitamin D, cholecalciferol or calcifediol supplementation post-diagnosis of COVID-19, and the dose was averaged and expressed as dose per month.

Risk of bias and quality of the evidence assessment

The risk of bias assessment was performed by two independent reviewers (GS and IE) and any discrepancy was settled through re-evaluation and consensus. The version 2 of the Cochrane risk-of-bias tool for randomized trials (RoB 2 tool) was used for the assessment of risk of bias of randomized trials.[34] Each outcome of the included randomized trials was evaluated through the process of signaling questions for the presence of bias in the randomization process, due to deviations from intended intervention, due to missing outcome data, in measurement of the outcome and in selection of the reported results. Based on the response to each aforementioned domain, an overall judgement regarding the risk of bias of the outcome of interest was made (high, some concerns or low). The robvis tool was used for the production of the final images.[35] The ROBINS-I tool was used for the assessment of risk of bias of non-randomized trials.[36] According to ROBINS-I methodology, for each non-randomized trial a target (idealized) randomized trial was assumed and assessed for bias in 7 domains (confounding, selection of participants, classification of interventions, deviation from intended interventions,

missing data, measurement of outcomes and selection of the reported results). Depending on the score of each domain, an overall risk of bias judgment was established. For the overall rating of the quality of the evidence the GRADE approach[37] was followed and the GRADEpro Guideline Development Tool (GDT)[38] was used for the creation of the Summary of Findings (SoF) table.

Statistical analysis

Statistical analysis was performed using Comprehensive Meta-Analysis software (CMA, Version 2.0, Biostat, Inc, NJ, USA). The software was used for pooling the data and deriving cumulative effect of the intervention on outcome of interest. The results were specifically assessed for presence of heterogeneity using Q statistics (significant at $P<0.10$). I^2—a quantitative measure of heterogeneity—was used to categorize studies into various levels of heterogeneity (high: 75–100%, medium: 50–70% and low: 0-50%). Cumulative results showing mortality and ICU rates with vitamin D supplementation are presented using forest plots. Publication bias was assessed using both quantitative and qualitative methods. The presence or absence of significant bias was concluded from the quantitative results of Egger's and Begg's and Mazumdar rank correlation test, whereas visual inspection of bias was undertaken using Funnel plot. Forest plot was used to display the relative treatment effect [odds ratio (OR)] and its 95% confidence intervals (CI) for each study. To examine if the dose of vitamin D affects the outcomes (mortality, ICU admission), random effect meta-regression analysis was applied. For this, log odds ratio was used as dependent variable and the dose of vitamin D as moderator variable. Additionally, we performed a separate analysis where we made a distinction between studies with "high and low" vitamin D administered doses.

The primary outcomes of interest were the impact of vitamin D supplementation on mortality and ICU admission in hospitalized patients with COVID-19. We pre-specified a priori that results for dichotomous outcomes were to be quantitatively synthesized by individual studies with use of a random-effects model with inverse variance weighting to obtain summary effect estimates represented as OR with associated 95% CI. We consider that the random-effects model approach was more appropriate for this meta-analysis because the studies included did not have the same design, intervention, patient population, dose of vitamin D supplementation, and management strategies for COVID-19.

Results

A total of 2078 patients from 9 studies hospitalized for COVID-19 were included in this meta-analysis with available data for mortality as outcome; of them, 583 received vitamin D supplementation and 61 (10.46%) died.[24-31,33]A total of 1495 patients did not receive vitamin D supplementation and 386 (25,81%) died. The summary estimates indicated that vitamin D supplementation did not reduce mortality in hospitalized patients with COVID-19 (test for overall effect size using the random effects model OR: 0.597; 95% CI: 0.318-1.121; p=0.109) (Figure 1).

Though 3 studies favored the intervention arm, the degree of impact varied among the studies. In addition, significant heterogeneity was found in terms of between-study variance (Q statistic=21.27, p=0.006, I^2=62.40%) and that resulted in deviation from funnel shape (Supplementary Figure 2). In terms of publication bias, the Egger's and Begg's tests showed the absence of any significant publication bias (p>0.05) (Table 2). The quality of the evidence

regarding the effect of vitamin D supplementation on mortality of COVID-19 patients assessed with the GRADE approach was judged as “very low” (table 3).

Random effect meta-regression analysis was applied to estimate functional relationship of log OR of mortality and vitamin D dose; it was found that the regression coefficient of the slope was 0.0000 (p=0.72294), suggesting that there is no significant linear relationship between vitamin D dose and log OR of mortality (Supplementary Table 3, Supplementary Fig 3). In addition, in the analysis of variance of random effect meta-regression analysis of log OR of mortality on dose of vitamin D, Q values of the model (0.12569), the residual (7.31725), and the total (7.44295) were not significant, implying that the relationship between vitamin D dose and mortality were not significant, deviations among log OR values of mortality and regression line were also not significant, and that the amount of total variance is lower than we would expect based on within-study error, respectively (Supplementary Table 3).

A total of 860 patients from 6 studies hospitalized for COVID-19 were also included in this meta-analysis with available data for the need of ICU as outcome; of them, 369 received vitamin D supplementation and 45 (12.19%) were admitted to ICU. [24,25,28,29,32,33] A total of 491 patients did not receive vitamin D supplementation and 129 (26.27%) were admitted to ICU. Overall, vitamin D supplementation significantly reduced the need for admission to ICU in hospitalized patients with COVID-19 (test for overall effect size using the random effects model OR: 0.326; 95% CI: 0.149-0.712; p=0.005) (Figure 2).

Though two studies favored the intervention arm, the degree of impact varied among the studies. In addition, significant heterogeneity was found in terms of between-study variance (Q

statistic=12.53, p=0.028, I^2=60.09%) and that resulted in deviation from funnel shape (Supplementary Figure 4). In terms of publication bias, the Egger's and Begg's tests showed that there was significant publication bias (p<0.05) (Table 2). The quality of the evidence regarding the effect of vitamin D supplementation on ICU admission of COVID-19 patients assessed with the GRADE approach was judged as "very low" (table 3).

Random effect meta-regression analysis was applied to estimate functional relationship of log OR of ICU admission and vitamin D dose; it was found that the regression coefficient of the slope was 0.0000 (p=0.96331), suggesting that there is no significant linear relationship between vitamin D dose and log OR of ICU admission (Supplementary Table 4, Supplementary Figure 5). In addition, in the analysis of variance of random effect meta-regression analysis of log OR of ICU admission on dose of vitamin D, Q values of the model (0.00212), the residual (5.67562), and the total (5.67773) were not significant, implying that the relationship between vitamin D dose and ICU admission were not significant, deviations among log OR values of ICU admission and regression line were also not significant, and that the amount of total variance is lower than we would expect based on within-study error, respectively (Supplementary Table 4).

To further investigate the impact of the administered dose of vitamin D on the outcomes of interest we performed a separate analysis, where we categorized the included studies as studies with "high or low doses" of vitamin D supplementation. Significant heterogeneity between studies, as well as within participants in each study, regarding the dose and duration of vitamin D supplementation was observed, so the use of an arbitrary value of administered vitamin D as a threshold for the distinction between "high and low doses" seemed inappropriate. Therefore, we decided to analyze separately the 2 studies that administered very

high bolus doses of vitamin D, the RCT by Murai et al[24]and the study by Giannini et al[33], where 200.000 and 400.000 IU of vitamin D were administered respectively, ("high doses") while the remaining studies were categorized as "low doses". "High doses" of vitamin D supplementation did not significantly reduce mortality (OR: 1.444; 95% CI: 0.705- 2.959, p=0.316) nor ICU admission (OR: 0.603; 95% CI: 0.348- 1.045, p= 0.072) in patients with COVID-19 (supplementary figures 6 and 7). However, "low doses" of vitamin D supplementation significantly reduced both mortality (OR: 0.437; 95% CI: 0.220- 0.867, p= 0.018) and ICU admission (OR: 0.157; 95% CI: 0.033- 0.743, p= 0.02) in COVID-19 patients (supplementary figures 8 and 9).

Discussion

In this systematic review and meta-analysis we examined the effect of vitamin D supplementation on mortality and ICU admission rates of patients with COVID-19. We found that vitamin D supplementation was associated with a significant reduction of the risk for ICU admission, while as far as mortality is concerned, no significant benefit was observed. Moreover, no significant relationship was found between the administered dose of vitamin D and either mortality or ICU admission. The quality of the evidence based on the GRADE approach is characterized as "very low" for both outcomes of interest (table 3).

The potential protective actions of vitamin D against COVID-19 can be explained by the biological functions of its biologically active form, 1,25-dihydroxyvitamin-D [1,25$(OH)_2$D], also known as calcitriol. Firstly, calcitriol regulates the innate immune response through the induction of autophagy and the production of cathelicidin, also known as LL-37, by

macrophages and epithelial cells of the respiratory system. LL-37 exerts antiviral activities through the disruption of the viral envelope and through binding to SARS-CoV-2 S (spike) protein, interfering with the mechanism of viral entry into the host cells.[4,39,40] A second mechanism is the regulation of the adaptive immunity and specifically the shift of the immune response from Th1 and Th17 to Th2 and Treg profile, thereby reducing the production of pro-inflammatory cytokines and the risk of cytokine storm.[4] Additionally, calcitriol interacts with the renin-angiotensin-aldosterone system (RAAS), mainly through the suppression of renin and angiotensin converting enzyme (ACE) and the induction of angiotensin converting enzyme 2 (ACE2), which leads to a reduction in the levels of angiotensin II and an increase of angiotensin 1-7. These actions of calcitriol counteract the imbalance of ACE:ACE2 that is caused by the downregulation of ACE2 in lung cells, due to the binding of SARS-CoV-2, and, subsequently, reduce the risk for vasoconstriction, ARDS and cardiac injury.[4,41] Finally, calcitriol has been found to protect against endothelial dysfunction and to exert antithrombotic actions.[4]

The administered dose of vitamin D may also influence the impact of supplementation on the outcomes of COVID-19. The included studies in this meta-analysis present variability as far as the administered dose of vitamin D is concerned, ranging from low daily doses like 1000 IU of cholecalciferol[32] to high-dose boluses like 400,000 IU of cholecalciferol.[24,33] Recently, it has been advocated that the daily doses of vitamin D rather than the intermittent high-dose boluses are effective for the prevention or treatment of certain diseases, like acute respiratory infections, rickets and tuberculosis.[42] A plausible explanation for this is that high-dose boluses of vitamin D increase the activity of the inactivating enzyme 24-hydroxylase CYP24A1, as well as the levels of fibroblast growth factor 23 (FGF23). 24-hydroxylase CYP24A1 is an important

regulator of vitamin D metabolism, as it converts 25(OH)D and 1,25$(OH)_2$D to the largely inactive forms of 24,25$(OH)_2$D and 1,24,25$(OH)_3$D. This is a mechanism through which vitamin D regulates its own metabolism. FGF23 negatively regulates vitamin D metabolism, via the increased expression of 24-hydoxylase CYP24A1 and, at the same time, via the reduction of the mRNA levels of 1-a hydroxylase, the enzyme responsible for 1-a hydroxylation of 25(OH)D.[43] Consequently, the activation of these mechanisms after the administration of a high intermittent dose of vitamin D has a longstanding effect and may result to intracellular vitamin D deficiency, despite the apparently efficient circulating levels. In an attempt to investigate the effect of dose supplementation of the studies included in this meta-analysis on the outcomes of interest, we performed a meta-regression analysis; however, no significant relationship was found. Nevertheless, the sub-analysis of "high" and "low" doses showed a significant impact of "low" administered doses on both outcomes, while "high" doses were not associated with any significant result. The aforementioned auto-regulatory pathways of vitamin D metabolism could be reflected here and merely justify this lack of association. Additionally, the "high" doses of vitamin D were administered after the diagnosis of COVID-19 (mean of 10.3 days from symptom onset in the study of Murai et al and the second and third day of the in-hospital stay in the study of Giannini et al), a fact that could reduce the effectiveness of the intervention. However, as previously mentioned, the included studies differ substantially as far as their design, study populations, dose and duration of vitamin D supplementation, rendering the interpretation of these results challenging.

Another issue that has risen is whether the impact of vitamin D supplementation should be considered in the setting of pre-existing deficiency or insufficiency, as supplementation

irrespectively of baseline levels is not expected to be beneficial.[39] Indeed, levels of 25(OH)D are not measured in all studies included in this meta-analysis. However, even in the cases of measured levels, there is controversy regarding the impact of time of measurement. 25(0H)D is largely bound to vitamin D binding protein and albumin, whose concentrations tend to decrease during acute illness, as a negative acute phase response.[44] Consequently, the interpretation of these levels in patients with severe COVID-19 remains questionable, as they may reflect reverse causality.

Vitamin D deficiency "pandemic" has constituted a long-standing issue of debate between experts and medical organizations, concerning the definition of the desirable levels of serum 25(OH)D and the recommended doses of supplementation.[5] In the setting of the overwhelming impact of COVID-19 pandemic and the urgent need for effective treatments against SARS-CoV-2, a link between these 2 pandemics has been proposed and vitamin D supplementation has been advocated as a possible adjunctive intervention for the management of COVID-19 patients. Indeed, this possibility seems intriguing, as vitamin D supplementation is a low-cost and safe intervention. Our findings from this meta-analysis, suggest a beneficial role of vitamin D supplementation in the rates of ICU admissions of COVID-19, irrespectively of the administered dose; however, significant reduction of mortality was not observed.

Four other systematic-reviews and meta-analyses on the effect of vitamin D supplementation on mortality and ICU admissions of COVID-19 patients were retrieved from database searching.[45,46] The first one,[45] that included 532 COVID-19 patients from 3 studies, concluded that vitamin D supplementation was associated with significant lower rates of ICU admission ($p < 0.0001$), while no significant benefit for mortality was observed; these findings are similar to

the results of our study. However, compared to that study, our meta-analysis includes a larger number of patients because at the time we performed our search more studies had been published. Moreover, as previously explained, we decided a priori to use the random-effects model, which we believe to be more appropriate for this meta-analysis due to different designs of the included studies, while in the aforementioned study the significant result was obtained with the application of fixed effect model. The second meta-analysis, which was obtained from a preprint server,[46] included only clinical trials, quasi experimental and pilot studies. Only one of the included studies reported on ICU admissions, while 3 studies that included a total of 190 patients reported on mortality. The authors conducted a meta-analysis of these studies and concluded that vitamin D supplementation is associated with a significant reduction in the odds of mortality (p=0.008). However, they did not describe the model applied for their analysis, and also, no publication bias was reported. An interesting, recently published analysis of 2933 COVID-19 patients from 13 studies (3 RCTs and 10 observational) concluded that vitamin D supplementation significantly reduced the incidence of the composite outcome of ICU admission/mortality; the association remained significant when adjusted risk estimates were analyzed.[47] On the contrary, another recent analysis of 467 patients with COVID-19 that aimed to investigate the effect of vitamin D supplementation on clinical outcomes, including ICU admission and mortality, did not find any significant association.[48] However, this meta-analysis included only randomized and quasi-experimental trials; consequently, the number of the included patients was small. Another distinction of this meta-analysis from ours and the previously mentioned is that vitamin D was administration was prospective after the diagnosis of COVID-19; as previously mentioned the possible influence of the time of vitamin D

administration remains to de elucidated. Additionally, an important asset of our study is the meta-regression analysis regarding the relationship between the administered dose of vitamin D and the outcomes of interest, an approach that had not been applied in the abovementioned studies.

This meta-analysis has several limitations. Firstly, due to the scarcity of RCTs at the moment of data collection, non-randomized studies have been included. The included studies differ as far as their design and sample size is concerned, and most of them present a high risk of bias (supplementary figures 10 and 11, supplementary table 2). Finally, there is heterogeneity between the studies in terms of the form and dose of vitamin D supplementation, the timing of administration in respect of the diagnosis of COVID-19 infection, the baseline levels of 25(OH)D, as well as the characteristics of the studied populations and the presence of comorbidities. Despite these limitations, we believe that our study provides insight of the possible contribution of vitamin D supplementation in the management of COVID-19 patients. Nevertheless, before suggesting the use of vitamin D as a possible adjunct treatment in COVID-19 pandemic, robust evidence from high-quality RCTs is needed.

Conclusion

The findings of the present meta-analysis support a beneficial role of vitamin D supplementation in the rates of ICU admission in COVID-19 patients. However, validation of these findings from high-quality RCTs is necessary.

Role of the funding source

There was no funding source for this study. All authors had full access to all the data in the study and the corresponding author had final responsibility for the decision to submit for publication.

Conflicts of interest

The authors declare they have no conflict of interest.

Author contributions

GS and IE determined the search strategy, screened the selected studies and extracted the data. NT and AT performed the statistical analysis. All authors participated in the writing and revision of this paper. All authors have read and approved this final manuscript.

Data availability statement

The data that supports the findings of this study are available in the supplementary material of this article.

References

1. Hu B, Guo H, Zhou P, Shi ZL. Characteristics of SARS-CoV-2 and COVID-19. *Nat Rev Microbiol.* Mar 2021;19(3):141-154.
2. Dong E, Du H, Gardner L. An interactive web-based dashboard to track COVID-19 in real time. *Lancet Infect Dis.* May 2020;20(5):533-534.
3. Gao Y-d, Ding M, Dong X, et al. Risk factors for severe and critically ill COVID-19 patients: A review. *Allergy.* 2021;76(2):428-455.
4. Charoenngam N, Shirvani A, Holick MF. Vitamin D and Its Potential Benefit for the COVID-19 Pandemic. *Endocr Pract.* Mar 17 2021.
5. Holick MF. The vitamin D deficiency pandemic: Approaches for diagnosis, treatment and prevention. *Rev Endocr Metab Disord.* Jun 2017;18(2):153-165.
6. Boucher BJ. Vitamin D status as a predictor of Covid-19 risk in Black, Asian and other ethnic minority groups in the UK. *Diabetes Metab Res Rev.* Nov 2020;36(8):e3375.
7. Hastie CE, Mackay DF, Ho F, et al. Vitamin D concentrations and COVID-19 infection in UK Biobank. *Diabetes Metab Syndr.* Jul-Aug 2020;14(4):561-565.
8. 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.
9. 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.
10. 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 Netw Open.* Sep 1 2020;3(9):e2019722.
11. 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. *Clin Endocrinol (Oxf).* Oct 2020;93(4):508-511.
12. Pizzini A, Aichner M, Sahanic S, et al. Impact of Vitamin D Deficiency on COVID-19-A Prospective Analysis from the CovILD Registry. *Nutrients.* Sep 11 2020;12(9).
13. Szeto B, Zucker JE, LaSota ED, et al. Vitamin D Status and COVID-19 Clinical Outcomes in Hospitalized Patients. *Endocr Res.* Dec 30 2020:1-8.
14. Vassiliou AG, Jahaj E, Pratikaki M, Orfanos SE, Dimopoulou I, Kotanidou A. Low 25-Hydroxyvitamin D Levels on Admission to the Intensive Care Unit May Predispose COVID-19 Pneumonia Patients to a Higher 28-Day Mortality Risk: A Pilot Study on a Greek ICU Cohort. *Nutrients.* Dec 9 2020;12(12).
15. Angelidi AM, Belanger MJ, Lorinsky MK, et al. Vitamin D Status Is Associated With In-Hospital Mortality and Mechanical Ventilation: A Cohort of COVID-19 Hospitalized Patients. *Mayo Clin Proc.* Jan 9 2021.
16. Charoenngam N, Shirvani A, Reddy N, Vodopivec DM, Apovian CM, Holick MF. Association of vitamin D status with hospital morbidity and mortality in adult hospitalized COVID-19 patients. *Endocr Pract.* Mar 8 2021.
17. Hastie CE, Pell JP, Sattar N. Vitamin D and COVID-19 infection and mortality in UK Biobank. *Eur J Nutr.* Feb 2021;60(1):545-548.
18. Petrelli F, Luciani A, Perego G, Dognini G, Colombelli PL, Ghidini A. Therapeutic and prognostic role of vitamin D for COVID-19 infection: A systematic review and meta-analysis of 43 observational studies. *J Steroid Biochem Mol Biol.* Mar 26 2021;211:105883.

19. Liu N, Sun J, Wang X, Zhang T, Zhao M, Li H. Low vitamin D status is associated with coronavirus disease 2019 outcomes: a systematic review and meta-analysis. *Int J Infect Dis.* Mar 2021;104:58-64.
20. Kazemi A, Mohammadi V, Aghababaee SK, Golzarand M, Clark CCT, Babajafari S. Association of Vitamin D Status with SARS-CoV-2 Infection or COVID-19 Severity: A Systematic Review and Meta-analysis. *Adv Nutr.* Mar 5 2021.
21. Bassatne A, Basbous M, Chakhtoura M, Zein OE, Rahme M, Fuleihan GE. The link between COVID-19 and VItamin D (VIVID): a systematic review and meta-analysis. *Metabolism.* Mar 24 2021:154753.
22. Akbar MR, Wibowo A, Pranata R, Setiabudiawan B. Low Serum 25-hydroxyvitamin D (Vitamin D) Level Is Associated With Susceptibility to COVID-19, Severity, and Mortality: A Systematic Review and Meta-Analysis. *Front Nutr.* 2021;8:660420.
23. Liberati A, Altman DG, Tetzlaff J, et al. The PRISMA statement for reporting systematic reviews and meta-analyses of studies that evaluate healthcare interventions: explanation and elaboration. *BMJ.* 2009;339:b2700.
24. Murai IH, Fernandes AL, Sales LP, et al. Effect of a Single High Dose of Vitamin D3 on Hospital Length of Stay in Patients With Moderate to Severe COVID-19: A Randomized Clinical Trial. *Jama.* Feb 17 2021.
25. 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". *The Journal of steroid biochemistry and molecular biology.* 2020;203:105751-105751.
26. Annweiler G, Corvaisier M, Gautier J, et al. Vitamin D Supplementation Associated to Better Survival in Hospitalized Frail Elderly COVID-19 Patients: The GERIA-COVID Quasi-Experimental Study. *Nutrients.* Nov 2 2020;12(11).
27. Cereda E, Bogliolo L, Lobascio F, et al. Vitamin D supplementation and outcomes in coronavirus disease 2019 (COVID-19) patients from the outbreak area of Lombardy, Italy. *Nutrition (Burbank, Los Angeles County, Calif.).* 2021;82:111055-111055.
28. Hernández JL, Nan D, Fernandez-Ayala M, et al. Vitamin D Status in Hospitalized Patients with SARS-CoV-2 Infection. *J Clin Endocrinol Metab.* Mar 8 2021;106(3):e1343-e1353.
29. Jevalikar G, Mithal A, Singh A, 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.* Mar 18 2021;11(1):6258.
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.* Dec 11 2020;12(12).
31. Cangiano B, Fatti LM, Danesi L, et al. Mortality in an Italian nursing home during COVID-19 pandemic: correlation with gender, age, ADL, vitamin D supplementation, and limitations of the diagnostic tests. *Aging (Albany NY).* Dec 22 2020;12(24):24522-24534.
32. Tan CW, Ho LP, Kalimuddin S, et al. Cohort study to evaluate the effect of vitamin D, magnesium, and vitamin B(12) in combination on progression to severe outcomes in older patients with coronavirus (COVID-19). *Nutrition.* Nov-Dec 2020;79-80:111017.
33. Giannini S, Passeri G, Tripepi G, et al. Effectiveness of In-Hospital Cholecalciferol Use on Clinical Outcomes in Comorbid COVID-19 Patients: A Hypothesis-Generating Study. *Nutrients.* Jan 14 2021;13(1).
34. Sterne JAC, Savović J, Page MJ, et al. RoB 2: a revised tool for assessing risk of bias in randomised trials. *Bmj.* Aug 28 2019;366:l4898.

35. McGuinness LA, Higgins JPT. Risk-of-bias VISualization (robvis): An R package and Shiny web app for visualizing risk-of-bias assessments. *Research Synthesis Methods.* 2020/04/26 2020;n/a(n/a).
36. Sterne JA, Hernán MA, Reeves BC, et al. ROBINS-I: a tool for assessing risk of bias in non-randomised studies of interventions. *Bmj.* Oct 12 2016;355:i4919.
37. Higgins JPT, Thomas J, Chandler J, et al. Cochrane Handbook for Systematic Reviews of Interventions version 6.2 (updated February 2021). Cochrane. 2021; www.training.cochrane.org/handbook. Accessed 10/25/2021.
38. GRADEpro GDT: GRADEpro Guideline Development Tool [Software]. McMaster University and Evidence Prime, 2021. https://gradepro.org/. Accessed 10/25/2021.
39. Griffin G, Hewison M, Hopkin J, et al. Vitamin D and COVID-19: evidence and recommendations for supplementation. *R Soc Open Sci.* Dec 2020;7(12):201912.
40. Bilezikian JP, Bikle D, Hewison M, et al. MECHANISMS IN ENDOCRINOLOGY: Vitamin D and COVID-19. *Eur J Endocrinol.* Nov 2020;183(5):R133-r147.
41. Arnold RH. COVID-19 - Does This Disease Kill Due to Imbalance of the Renin Angiotensin System (RAS) Caused by Genetic and Gender Differences in the Response to Viral ACE 2 Attack? *Heart Lung Circ.* Jul 2020;29(7):964-972.
42. Griffin G, Hewison M, Hopkin J, et al. Perspective: Vitamin D supplementation prevents rickets and acute respiratory infections when given as daily maintenance but not as intermittent bolus: implications for COVID-19. *Clin Med (Lond).* Mar 2021;21(2):e144-e149.
43. Bringhurst FR, Demay MB, Kronenberg HM. Mineral Metabolism. *Williams Textbook of Endocrinology.* Vol 1. 14th ed: Elsevier - Health Sciences Division; 2020:1211-1217.
44. Rhodes JM, Subramanian S, Laird E, Griffin G, Kenny RA. Perspective: Vitamin D deficiency and COVID-19 severity - plausibly linked by latitude, ethnicity, impacts on cytokines, ACE2 and thrombosis. *J Intern Med.* Jan 2021;289(1):97-115.
45. Shah K, Saxena D, Mavalankar D. Vitamin D supplementation, COVID-19 and disease severity: a meta-analysis. *Qjm.* May 19 2021;114(3):175-181.
46. Nikniaz L, Akbarzadeh MA, Hosseinifard H, Hosseini M-S. The impact of vitamin D supplementation on mortality rate and clinical outcomes of COVID-19 patients: A systematic review and meta-analysis. *medRxiv.* 2021:2021.2001.2004.21249219.
47. Pal R, Banerjee M, Bhadada SK, Shetty AJ, Singh B, Vyas A. Vitamin D supplementation and clinical outcomes in COVID-19: a systematic review and meta-analysis. *J Endocrinol Invest.* Jun 24 2021:1-16.
48. Rawat D, Roy A, Maitra S, Shankar V, Khanna P, Baidya DK. "Vitamin D supplementation and COVID-19 treatment: A systematic review and meta-analysis". *Diabetes Metab Syndr.* Jul-Aug 2021;15(4):102189.

Legends to figures

Figure 1: Forest plot for vitamin D supplementation and mortality

Figure 2: Forest plot for vitamin D supplementation and ICU admission

Table 1: Baseline characteristics of the included studies

Authors (year)	Methods	Population	Serum 25 (OH)D (ng/ml)		Vitamin D administration	Outcomes
			Baseline	End of study		
Murai et al (2021)	Multicentre double-blinded, placebo-controlled trial (Brazil)	237 hospitalised patients from 2 hospitals in Brazil with moderate to severe COVID-19. Intervention group N:119 Mean (SD) age: 56.5(13.8) Control group: N: 118 Mean (SD) age: 56 (15)	Intervention: Mean (SD): 21.2 (10.1) Control: Mean (SD): 20.6 (8.1)	Intervention: Mean (SD): 44.4 (15) Control: Mean (SD): 19.8 (10.5)	Intervention: Single oral dose of 200.000 IU of D3 after the diagnosis of COVID in hospitalised patients with moderate or severe disease Control: Placebo	Primary: length of hospital stay Secondary: 1.Mortality during hospitalisation 2.N requiring ICU admission 3.N requiring and duration of mechanical ventilation 4.Serum levels of 25(OH)D, total calcium, creatinine, CRP
Entrenas Castillo et al (2020)	Pilot randomized open label, double masked clinical study (Spain)	76 Hospitalised COVID 19 patients Intervention group: N:50 Mean (SD) age: 53.1 (10.8) Control group: Mean (SD) age: 52.8 (9.4)	NR	NR	Intervention: Oral calcifediol 0,532 mg on day of admission, 0.266 on day 3 and 7 and then weekly until discharge or ICU admission	1.ICU admission 2.Mortality
Annweiler et al (2020)	Quasi- experimental study with retrospective collection of data from patients records (France)	77 patients hospitalised in a geriatric acute care unit mean (SD) age: 88 (5) years, range 78–100 years 45 patients receiving vit D sup (Group 1: 29, Group 2: 16) 32 patients with no vit D sup	NR	NR	Group 1: oral boluses of vit D sup over the preceding year (50,000 IU D3 per month, or 80,000 IU or 100,000 IU vitamin D3 every 2–3 month) Group 2: oral 80,000 IU D3 within hours of the diagnosis of COVID-19.	Primary: 14-day mortality Secondary: highest (worst) score on the ordinal scale for clinical improvement (OSCI) measured during COVID-19 acute phase
Cereda et al (2020)	Prospective observational study (Italy)	170 participants with data about in-hospital and vit D sup from 3 groups (group 1: patients with PD,	Mean (SD) levels of 25(OH)D of hospital inpatients (group 3): 13.2 (11.1)		oral intake of at least 25 000 IU/month (~800	1.In hospital mortality 2.Hospitalisation

		group 2: caregivers of patients with PD, group 3: COVID -19 patients admitted to a referral hospital). 18 received vit D sup in the previous 3 months, 152 did not receive vit D sup.	Values not available for 25(OH)D supplemented VS non-supplemented		IU/d) in the previous 3 months	
Hernandez et al (2020)	Retrospective case-control study (Spain)	216 patients with COVID-19 admitted to a university hospital. 19 patients were on oral vit D sup at admission, 197 patients were not on oral vit D sup	Group on vit D sup: 21.1 ± 5.9 Group not receiving vit D sup: 13.8 ± 7.2	NA	11 patients were taking cholecalciferol, 25 000 IU/monthly in 10 cases, and 5600 IU/weekly in 1, and 8 patients were on calcifediol, 0.266 mg/monthly	1.ICU admission 2.Mechanical ventilation 3.Radiological worsening 4.Secondary infection 5.Thrombotic events 6.Death 7.Composite severity endpoint 8.Length of stay
Jevalikar et al (2021)	prospective, single-centre, cross-sectional, observational study (India)	A total of 410 patients hospitalised for COVID-19. (127 females, 9 pediatric, 17 asymptomatic) with a median age of 54 years (range 6–92 years) were included 197 had VDD defined as 25(0H)D < 20 ng/ml and 128 of them were treated with cholecalciferol . (the outcomes of those treated with cholecalciferol were compared with the ones with no-treated VDD)	NR	NR	For most patients the treatment was administered as cholecalciferol granules (60,000 units per gram), depending on the decision of the treating physician, median administered dose 60.000 IU	Primary outcome: Proportion of severe cases in VDD group vs non-VDD Other outcomes: 1.admission to ICU, 2.administration of oxygen 3.inotropic support 4.renal replacement therapy 5.Deaths 6.Difference in the mean levels of inflammatory markers
Ling et al (2020)	retrospective multi-centre cross-sectional observational study (United Kingdom)	968 patients hospitalised with COVID-19 from 3 hospital trusts were included [a primary cohort of 444 and a validation cohort of 541 patients from 2 hospitals (RPH and UHL)], of whom 151 received cholecalciferol booster therapy	Primary cohort: median 25(OH)D level: 12.5 (IQR 7.6, 22), in 230 participants with available values Validation cohort:	NA	high-dose cholecalciferol booster therapy (approximately ≥ 280,000 IU in a time period of up to 7 weeks) in various regimens	COVID-19 mortality

			Median 25 (OH)D: 12.5 (7.6,22) at RPH and 17.2 (10.8,24) at UHL			
Cangiano et al (2020)	observational cohort study (Italy)	157 residents of a nursing home [mean age: 89.8 (6.53)], 98 of them were COVID-19 positive (20 were on vit D sup, while 78 were not)	NR	NA	Cholecalciferol 25.000 IU 2 times a month	1.Mortality 2.Positivity for SARS-CoV-2.
Tan et al (2020)	cohort observational study (Singapore)	43 patients with COVID-19 hospitalised in a tertiary hospital 17 of them received vit D sup [mean age (SD): 58.4 (7)] 26 of them did not receive vit D sup and were used as control group [mean age(SD): 64.1(7.9)]	NR	NR	a single daily oral 1000IU dose of vit D3, 150 mg of magnesium oxide, and 500 mg vitamin B12 (methylcobalamine) for ≤14 days	1.Requirement of oxygen therapy 2.ICU admission 3.Mortality
Giannini et al (2021)	Retrospective study (Italy)	91 patients with COVID-19 admitted in a general medicine ward of a university hospital. 36 received vit D sup [mean age (SD): 73(13)] while 55 did not receive vit D sup [mean age (SD): 74 (13)]	Group with vit D sup: Median 25(OH)D (IQR): 9.9(4.8,16.9) Group with no vitD sup: Median 25(OH)D (IQR): 14.4(7.6,30.8)	NA	400,000 IU vit D sup as bolus oral cholecalciferol 200.000 daily for two consecutive days (the second and third day of the in-hospital stay)	Composite outcome of transfer to ICU and/or death from any cause

N: number of participants, SD: standard deviation, 25(OH)D: 25-hydroxyvitamin D, ICU: intensive care unit, vit D sup: vitamin D supplementation, D3: cholecalciferol, PD: Parkinson's disease, VDD: vitamin D deficiency, IQR: interquartile range, NR: not reported, NA: non-applicable

Table 2: Publication bias

Mortality and vitamin D supplementation	
Egger's test	
Intercept	0.09719
95% CI	-2.74118 to 2.93557
Significance level	p=0.67666
Begg's test	
Kendall's Tau	-0.11111
Significance level	p=0.41712
ICU admission and vitamin D supplementation	
Egger's test	
Intercept	-3.00006
95% CI	-4.96067 to -1.03945
Significance level	p=0.011317
Begg's test	
Kendall's Tau	-0.86667
Significance level	-0.86667

Table 3: Summary of findings table

Summary of findings:

Vitamin D supplementation compared to no vitamin D supplementation for reducing mortality and ICU admission of COVID-19 patients

Patient or population: reducing mortality and ICU admission of COVID-19 patients

Setting: hospitalized patients (hospitals, geriatric acute care units, nursing homes)

Intervention: vitamin D supplementation

Comparison: no vitamin D supplementation

Outcomes	Anticipated absolute effects* (95% CI)		Relative effect (95% CI)	№ of participants (studies)	Certainty of the evidence (GRADE)	Comments
	Risk with no vitamin D supplementation	Risk with vitamin D supplementation				
Mortality	258 per 1,000	**172 per 1,000** (100 to 281)	**OR 0.597** (0.318 to 1.121)	2078 (9 studies)	⨁◯◯◯ Very low[a,b,c,d]	Vitamin D supplementation does not reduce mortality in hospitalized patients for COVID-19. The quality of evidence is very low and we have very little confidence in this result
ICU admission	263 per 1,000	**104 per 1,000** (50 to 202)	**OR 0.326** (0.149 to 0.712)	860 (6 studies)	⨁◯◯◯ Very low[a,b,c,d]	Vitamin D supplementation reduced the need for ICU admission for hospitalized patients for COVID-19. The quality of evidence is very low and we have very little confidence in this result,

***The risk in the intervention group** (and its 95% confidence interval) is based on the assumed risk in the comparison group and the **relative effect** of the intervention (and its 95% CI).

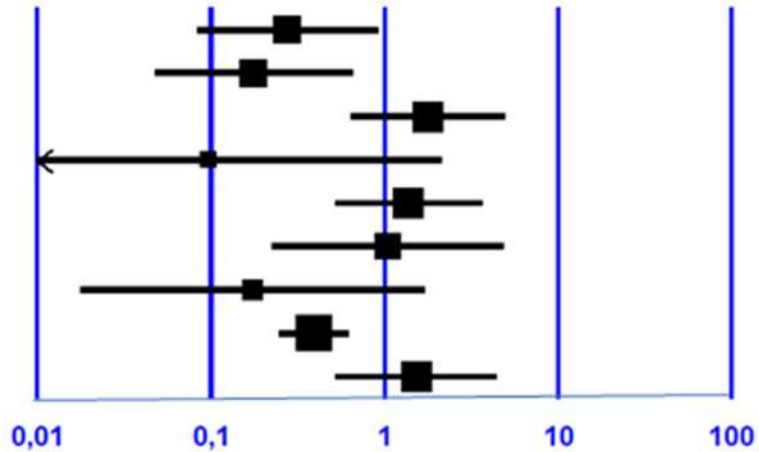
CI: confidence interval; **OR:** odds ratio

GRADE Working Group grades of evidence
High certainty: we are very confident that the true effect lies close to that of the estimate of the effect.
Moderate certainty: we are moderately confident in the effect estimate: the true effect is likely to be close to the estimate of the effect, but there is a possibility that it is substantially different.
Low certainty: our confidence in the effect estimate is limited: the true effect may be substantially different from the estimate of the effect.
Very low certainty: we have very little confidence in the effect estimate: the true effect is likely to be substantially different from the estimate of effect.

Explanations

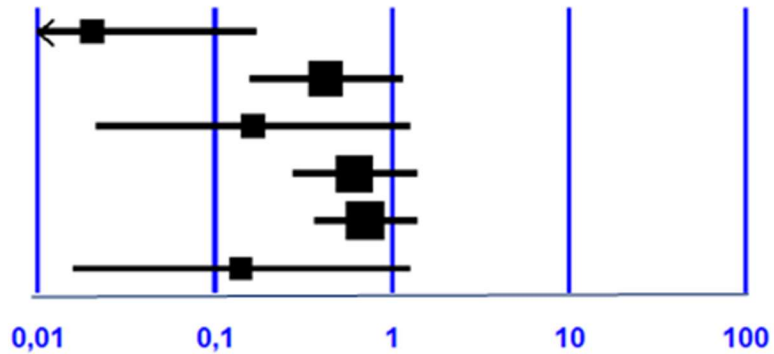
a. serious overall risk of bias of the included studies. The majority of the included studies are non-randomized (ROBINS-2 tool was used for the assessment of non-randomized studies and RoB2 tool for randomized studies), b. presence of significant inconsistency (minimal overlap of confidence intervals, large differences in estimation effects, statistical significance for heterogeneity $p < 0.05$ and $I^2=62.40$) with no robust explanation available, c. serious indirectness (differences in study populations, doses and forms of vitamin D and duration of therapy that affect generalizability), d. publication bias strongly suspected (funnel plot asymmetry)

Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value	Dead / Total: Treated	Dead / Total: Not treated	Relative weight
Annweiler, 2020	0,275	0,083	0,907	-2,121	0,034	5 / 45	10 / 32	7,31
Cangiano, 2020	0,176	0,048	0,651	-2,605	0,009	3 / 20	39 / 78	6,11
Cereda, 2021	1,782	0,646	4,912	1,116	0,264	7 / 18	40 / 152	10,12
Entrenas Castillo, 2020	0,097	0,004	2,100	-1,487	0,137	0 / 50	2 / 26	1,10
Giannini, 2021	1,378	0,522	3,636	0,648	0,517	10 / 36	12 / 55	11,06
Hernadez, 2020	1,041	0,224	4,839	0,051	0,959	2 / 19	20 / 197	4,41
Jevalikar, 2021	0,173	0,018	1,698	-1,505	0,132	1 / 128	3 / 69	2,00
Ling, 2020	0,392	0,247	0,622	-3,975	0,000	24 / 148	254 / 768	48,72
Murai, 2021	1,527	0,526	4,435	0,779	0,436	9 / 119	6 / 118	9,16
Total (95% CI):	0.597	0.318,	1.121			447/ 2078		100%



Heterogeneity: Q-value=21.27, df=8, p=0.006, I^2=62,4%.
Test for overall effect (random effects): Z= -1,604 (p=0,109)

Study name	Odds ratio	Lower limit	Upper limit	Z-Value	p-Value	ICU / Total: Treated	ICU / Total: Not-treated	Relative weight
Entrenas Castillo, 2020	0,020	0,002	0,171	-3,591	0,000	1 / 50	13 / 26	3,99
Giannini, 2021	0,422	0,157	1,139	-1,704	0,088	7 / 36	20 / 55	18,33
Hernadez, 2020	0,163	0,021	1,255	-1,742	0,082	1 / 19	50 / 197	4,33
Jevalikar, 2021	0,615	0,277	1,368	-1,191	0,234	16 / 128	13 / 69	28,22
Murai, 2021	0,707	0,365	1,367	-1,031	0,303	19 / 119	25 / 118	41,36
Tan, 2020	0,141	0,016	1,251	-1,759	0,079	1 / 17	8 / 26	3,77
Total (95% CI)	0.326	0.149,	0.712			174/ 860		100%



Heterogeneity: Q-value=12,53, df=65 p=0.028, I^2=60.09%.
Test for overall effect (random effects): Z= -2,813 (p=0,005)

Legends to supplementary figures:

Supplementary figure 1: PRISMA flow diagram

Supplementary figure 2: Funnel plot for vitamin D supplementation and mortality

Supplementary figure 3: Random effect meta-regression analysis on the functional relationship of log odds ratio of mortality and vitamin D dose; the regression coefficient of the slope is 0.0000 (p:0.72294). The dose of vitamin D (x-axis) given to the patients is in thousand units per month

Supplementary figure 4: Funnel plot for vitamin D supplementation and ICU admission

Supplementary figure 5: Random effect meta-regression analysis on the functional relationships of log odds ratio of ICU admission and vitamin D dose; the regression coefficient of the slope is 0.0000 (p=0.96331). The dose of vitamin D (x-axis) given to the patients is in thousand units per month

Supplementary figure 6: Forest plot for “high doses” of vitamin D supplementation and mortality

Supplementary figure 7: Forest plot for “high doses” of vitamin D supplementation and ICU admission

Supplementary figure 8: Forest plot for “low doses” of vitamin D supplementation and mortality

Supplementary figure 9: Forest plot for “low doses” of vitamin D supplementation and ICU admission

Supplementary figure 10: Risk of bias assessment of the outcome “mortality” of the randomized studies with the use of Rob2 tool

Supplementary figure 11: Risk of bias assessment of the outcome “ICU admission” of the randomized studies with the use of Rob2 tool