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Profesional paper

VITAMIN D STATUS IN PATIENTS WITH COVID-19 – SEX DIFFERENCES ASSOCIATED WITH SEVERITY OF THE DISEASE.

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ABSTRACT

Background: Patients with COVID-19 can develop abnormal inflammatory response, which can progress to multiorgan failure and death, with a higher prevalence observed in men. Besides, recent research suggests a complex interaction between viral infections and vitamin D.

Aims: This study evaluates sex differences of vitamin D status, circulating enzymes and biomarkers of inflammatory response at the time of diagnosis of COVID-19, depending on severity of disease the patients develop.

Methods: This retrospective observational study was conducted from September to December 2020. Patients (n = 115) were divided in two groups: Group A- asymptomatic or mild cases (36 men and 19 women; mean age 53.3 ± 1.4 y.) and Group B- patients who develop moderate or severe COVID-19 requiring hospitalization (38 men and 22 women; mean age 56.1 ± 1.3 y.).

Results: Patients of Group B had almost two-fold lower circulating 25(OH)D concentrations than patients of Group A (Group A: 61.6 ± 1.9 nmol/L; Group B: 32.2 ± 1.4 nmol/L; $p < 0.001$). Within Group A, the only parameter with a statistically significant sex difference was ferritin ($p < 0.01$). Within Group B, in addition to ferritin ($p < 0.05$), we observed a pronounced and statistically highly significant sex difference in circulating 25(OH)D concentrations (men: 35.8 ± 1.7 nmol/L; women: 26.0 ± 1.7; $p < 0.001$).

Conclusions: Our results demonstrate that inadequate vitamin D status is associated with the development of moderate or severe COVID-19 and indicate that circulating 25(OH)D might be associated to sex differences in disease outcome.

Key words: 25(OH)D, SARS-CoV-2, ferritin, women, gender

INTRODUCTION

COVID-19 is a disease that is characterized by high contagiousness and remarkable variability of the clinical presentation. Studies have shown that SARS-CoV-2 initially acts through mechanisms of immune evasion, which in some patients is followed by immune hyper-reaction and cytokine storm, as a common pathogenic mechanism with acute respiratory distress syndrome and systemic inflammatory distress syndrome, regardless the etiological factor [1]. Studies also indicate a complex interaction between viral infections and vitamin D, including functional immunoregulatory processes, interaction with cellular and viral factors, induction of autophagy and apoptosis, as well as genetic and epigenetic alterations [2]. Accordingly, previous systematic reviews and meta-analyses suggested that vitamin D supplementation significantly reduces the

risk of common upper respiratory tract infections [3,4]. Also, protective role of vitamin D has been shown in many conditions associated with pneumonia, cytokine overproduction and ARDS [1]. Research focused on the role of vitamin D in patients with COVID-19 highlights the importance of optimization of vitamin D status in COVID-19 management [5]. Inadequate vitamin D status in patients with COVID-19 is directly associated with increased concentrations of circulating CRP and IL-6 and increased risk of pneumonia and acute respiratory distress syndrome [6].

In parallel with these observations, another important observation emerged during the COVID-19 pandemic, that is significant sex difference in COVID-19 morbidity and mortality, with male patients having almost three times odds of requiring intensive treatment unit admission and higher odds of death [7].

Based on these observations, we hypothesized that circulating concentrations of 25(OH)D, which is a well-established biomarker of vitamin D status, may be among the key factors associated with marked variability of clinical presentation in patients with COVID-19. Therefore, the aim of this study was to evaluate, at the time of diagnosis of COVID-19, the circulating 25(OH)D concentrations, as well as aspartate aminotransferase (AST), alanine aminotransferase (ALT), <glutamyl transferase (GGT), lactate dehydrogenase (LDH), creatine kinase (CK) and its isoenzyme CK-MB, C-reactive protein (CRP), interleukin 6 (IL-6), white blood cell (WBC) count, erythrocyte sedimentation rate (Se), ferritin (FER), and serum iron (IRN), depending on severity of disease the patients develop, for men and women separately.

METHODS & MATERIAL

Design of this study has been reported in detail previously [8]. In brief, this was a retrospective observational study that was conducted in the General City Hospital "8th September", Skopje, North Macedonia, which, during the most difficult months of the pandemic, was reorganized to operate as a COVID center covering the whole country. The study was conducted from September to December 2020, and was approved by the Ethics Committee of the Hospital. Eligibility criteria for enrolment in this study were the following: adults, at the time of diagnosis of COVID-19 that was confirmed with positive PCR test for SARS-CoV-2, without comorbidities such as diabetes, history of cancer, end-stage kidney disease on hemodialysis, or chronic obstructive lung disease. Pregnant women were not included in this study. In total, n = 115 patients were enrolled and divided in two groups: Group A- asymptomatic or mild cases, n = 55 (36 men and 19 women) and Group B- patients who develop moderate or severe COVID-19 requiring hospitalization, n = 60 (38 men and 22 women).

Serum concentrations of 25(OH)D were measured using automated immunochemistry analyzer Advia Centaur XP, serum concentrations of CRP and FER were measured using the automated nephelometer BN ProSpec System, serum concentrations of IL-6 were measured using the automated immunochemistry analyzer Immulite 2000 Xpi, and serum concentrations of AST, ALT, GGT, LDH, CK, CK-MB, and IRN were measured using the automated biochemistry analyzer Dimension RxL. WBC count was determined using the automated hematology analyzer Advia 2120i. For all these automated analyzers, original reagents were used. Se was measured using the automated analyzer Alifax.

Statistical data analysis was conducted using the SPSS program.

RESULTS

First, we analyzed the age of patients and found no statistically significant difference between two groups (Group A: 53.3 ± 1.4 y.; Group B: 56.1 ± 1.3 y.; $p > 0.05$), which ruled out potential influence of age on biochemical parameters and severity of disease [9,8]. Next, we showed that serum 25(OH)D concentrations are significantly lower in patients who develop moderate or severe COVID-19 requiring hospitalization, i.e., almost two-fold lower, $p < 0.001$ (Table 1). Comparative analyses of circulating enzymes and biomarkers of inflammatory response in these two groups have been reported previously [9,8].

Table 1. Vitamin D status in patients with COVID-19.

	Group A		Group B	
	Mean ± standard error	95% Confidence interval for mean	Mean ± standard error	95% Confidence interval for mean
25(OH)D (nmol/L)	61.5 ± 1.9	57.7 – 65.2	32.2 ± 1.4***	29.5 – 34.9

Group A- asymptomatic or mild cases of COVID-19.

Group B- patients who develop moderate or severe COVID-19 requiring hospitalization.

***$p < 0.001$

Next, we asked whether there were significant sex differences in the values of analyzed clinical chemistry parameters. To this end, using Mann-Whitney U-test, we analyzed the statistical significance for all parameters depending on gender, i.e., men vs. women, for Group A and Group B separately.

These results showed that in patients of Group A there is a statistically significant sex difference only for FER ($p < 0.01$), while there is almost no difference in serum concentrations of 25(OH)D between men and women (Table 2). Within Group B, a statistically significant difference for FER was also observed ($p < 0.05$). In addition, in patients of Group B a striking sex difference in serum concentrations of 25(OH)D was observed ($p < 0.001$), with women having nearly 10 units lower serum concentrations (Table 3).

Table 2. Vitamin D status, circulating enzymes and biomarkers of inflammatory response in patients with COVID-19; men and women of Group A- asymptomatic or mild cases.

	Men		Women	
	Mean ± standard error	95% Confidence interval for mean	Mean ± standard error	95% Confidence interval for mean
Age (y.)	52.5 ± 1.8	49.0 – 56.1	54.8 ± 2.3 ns	50.0 – 59.6
AST (U/L)	45.1 ± 5.0	35.0 – 55.2	32.3 ± 2.7 ns	26.6 – 37.9
ALT (U/L)	71.6 ± 10.7	49.8 – 93.4	44.5 ± 5.3 ns	33.4 – 55.7
LDH (U/L)	334.7 ± 29.2	275.4 – 394.0	327.0 ± 38.9 ns	245.3 – 408.6
GGT (U/L)	53.6 ± 6.3	40.8 – 66.4	41.5 ± 5.4 ns	30.2 – 52.8
CK (U/L)	226.6 ± 83.8	56.4 – 396.8	90.3 ± 13.3 ns	62.3 – 118.3
CK-MB (U/L)	19.7 ± 1.2	17.2 – 22.2	19.6 ± 1.8 ns	15.8 – 23.4
Se (mm/h)	25.1 ± 2.0	21.0 – 29.3	30.6 ± 3.7 ns	22.8 – 38.5
WBC (x109)	6.7 ± 0.4	6.0 – 7.5	7.1 ± 0.6 ns	5.8 – 8.4
CRP (mg/L)	39.1 ± 5.9	27.1 – 51.1	35.3 ± 8.4 ns	17.7 – 52.9
IL-6 (pg/mL)	17.8 ± 3.3	11.0 – 24.5	13.5 ± 3.1 ns	7.1 – 19.9
FER (µg/L)	518.1 ± 54.5	407.5 – 628.7	248.0 ± 53.5**	135.5 – 360.4
IRN (µmol/L)	7.7 ± 0.9	5.9 – 9.5	5.8 ± 0.9 ns	3.9 – 7.8
25(OH)D (nmol/L)	61.6 ± 2.2	57.2 – 66.0	61.3 ± 3.7 ns	53.4 – 69.1

**p < 0.01; ns – nonsignificant; compared to men

Table 3. Vitamin D status and circulating enzymes and biomarkers of inflammatory response in patients with COVID-19; men and women of Group B- patients who develop moderate or severe COVID-19 requiring hospitalization.

	Men		Women	
	Mean ± standard error	95% Confidence interval for mean	Mean ± standard error	95% Confidence interval for mean
Age (y.)	57.8 ± 1.4	55.1 – 60.6	53.1 ± 2.5 ns	47.8 – 58.3
AST (U/L)	54.9 ± 4.8	45.1 – 64.7	54.0 ± 6.3 ns	40.9 – 67.0
ALT (U/L)	68.5 ± 10.0	48.3 – 88.6	56.1 ± 10.9 ns	33.5 – 78.7
LDH (U/L)	449.7 ± 34.9	378.9 – 520.5	420.6 ± 22.6 ns	373.7 – 467.5
GGT (U/L)	101.6 ± 15.5	70.1 – 133.0	90.8 ± 23.5 ns	41.9 – 139.7
CK (U/L)	220.4 ± 42.1	135.2 – 305.6	203.1 ± 58.2 ns	82.0 – 324.1
CK-MB (U/L)	22.4 ± 1.8	18.8 – 26.0	22.8 ± 2.2 ns	18.2 – 27.4
Se (mm/h)	37.8 ± 3.3	31.2 – 44.5	34.2 ± 4.3 ns	25.3 – 43.1
WBC (x109)	8.3 ± 0.6	7.1 – 9.5	7.9 ± 0.7 ns	6.4 – 9.4
CRP (mg/L)	93.4 ± 7.9	77.5 – 109.3	112.3 ± 14.9 ns	81.3 – 143.3
IL-6 (pg/mL)	65.8 ± 11.8	41.9 – 89.6	82.3 ± 24.2 ns	32.1 – 132.6
FER (µg/L)	969.2 ± 132.8	700.0 – 1238.3	794.0 ± 271.0*	230.3 – 1357.6
IRN (µmol/L)	5.3 ± 0.5	4.3 – 6.4	4.8 ± 0.7 ns	3.3 – 6.3
25(OH)D (nmol/L)	35.8 ± 1.7	32.4 – 39.2	26.0 ± 1.7 ***	22.5 – 29.4

***p < 0.001; *p < 0.05; ns – nonsignificant; compared to men

DISCUSSION

SARS-CoV-2, the causative pathogen of COVID-19 disease, first emerged in Wuhan, China, in December 2019, and spread with a skyrocket speed all over the world. COVID-19 pandemic, declared by the WHO shortly after, i.e., in March 2020 [10, has resulted in millions of infected people and a significant number of deaths. This situation has led to an overburden of healthcare systems in most countries [11,12], but especially in low- and middle-income countries [13]. About 80% of people who had COVID-19 were asymptomatic or developed only mild symptoms of the disease. The rest of the infected developed a serious

form of the disease [14]. Faced with this serious public health emergency, with no specific drugs or vaccines, drug repurposing was among the first responses suggested by scientists responding to this global crisis [15]. In addition, since the beginning of the pandemic, vitamin D has been highlighted as one of the key factors that determine the course of disease [14]. Suggested putative molecular mechanisms underlying the protective effects of vitamin D in COVID-19 include involvement in maintaining tight junctions, gap junctions, and adherens junctions to decrease viral infection, enhancing cellular innate immunity partly through induction of antimicrobial peptides, reducing the production of pro-inflammatory cytokines, as well as increasing the expression of anti-inflammatory cytokines [16], which led to the recommendation to start using vitamin D to combat COVID-19 that was publicly released as early as on 13 March 2020, Website https://www.foxnews.com/opinion/former-cdc-chief-tom-frieden-coronavirus-risk-may-be-reduced-with-vitamin-d (accessed on 28.02.2023).

In addition, since the beginning of the pandemic, a significant sex difference in morbidity and mortality from COVID-19 has been observed [7], with men having a significantly higher mortality than women, approximately 3:1, which indicates the presence of sex-dependent biological factors underlying the observed differences in disease outcome [17]. Suggested putative biological factors include sex-dependent dimorphic immune responses to viral infections [18], sex-dependent modulation of the expression of ACE2 and TMPRSS2, the key proteins for SARS-CoV-2 entrance in human host cells [19], and the protective role of estrogen on the cardiovascular system [20].

Despite significantly lower prevalence, some women still develop a severe form of COVID-19. In this context, the results of our study clearly showed that in patients who develop moderate or severe COVID-19 requiring hospitalization, there was a significant sex difference in circulating 25(OH)D concentrations, with women having particularly pronounced vitamin D deficiency. These results indicate that extreme vitamin D deficiency in women may overcome protective genetic and hormonal factors, leading to a severe form of the disease. This observation derives from the specific, original design of our study and the narrow and well-defined time frame of data collection, which is the period September – December 2020.

The results of our study are in accordance with the data from the literature [21]. Understandably, at the beginning of the pandemic, reports on the role of vitamin D in COVID-19 were scarce [22,1]. At that time, this role of vitamin D could only be assumed from the existing scientific literature on its immunomodulatory effects [1] as well as from the clinical practice in management of patients with COVID-19 [22]. However, subsequent studies highlighted the role of vitamin D in COVID-19. For example, it has been shown that low 25(OH)D levels assessed at the time of hospitalization are associated with greater COVID-19 severity, requirement for intensive care unit admission, and COVID-19 related mortality [23]. Besides, it has been shown that among hospitalized COVID-19 patients, pre-infection deficiency of vitamin D was associated with increased disease severity and mortality [24]. Also, a recent meta-analysis of thirty-two observational studies has shown that vitamin D deficiency in COVID-19 patients is associated with significantly higher levels of inflammatory biomarkers, such as IL-6, CRP, and FER [25]. In addition, another meta-analysis of twenty-four observational studies showed a potentially increased risk of developing severe COVID-19 infection among patients with low vitamin D levels [26].

In the context of striking sex differences in morbidity and mortality of COVID-19, putative sex differences in protective effects of vitamin D in COVID-19 have also been addressed [27,17]. Previous studies have provided evidence about sex differences in immunomodulatory and anti-inflammatory properties of vitamin D in autoimmune diseases [28], estrogen-dependent vitamin D control of T-cell differentiation [29], or tissue-specific effects of vitamin D on peripheral estrogen and androgen metabolism [30]. However, studies specifically addressing sex-dependent differences in vitamin D status in COVID-19 are rare [31]. To the best the best of our knowledge, sex difference in circulating 25(OH)D concentrations at the time of diagnosis of COVID-19 in patients who develop moderate or severe COVID-19 requiring hospitalization has not been reported so far. As such, the results of our study represent a foundation for future studies designed to address molecular mechanisms underlying our clinical observation.

In addition to 25(OH)D, we have also observed a significant sex difference for serum FER. Unlike 25(OH)D, a statistically significant sex difference for FER was observed in both Group A and Group B. FER is the major iron storage protein, and as such is essential to iron homeostasis. Low serum FER in otherwise healthy

individuals is highly specific for iron deficiency anemia [32]. On the other side, elevated serum FER occurs not only because of iron overload, but also due to inflammation, liver or kidney disease, cancer, or metabolic syndrome [33]. Accordingly, during the COVID-19 pandemic, serum FER emerged as one of the key inflammatory biomarkers and an important predictor of disease outcome [34]. The results on circulating FER we report here are consistent with the data from the literature, where a sex difference has also been observed, with men having higher mean values than women [35]. This observation can be explained by the sex differences in the reference ranges for this parameter, which are lower for women, but it is also tempting to speculate that the higher circulating concentrations of FER could contribute to the poorer prognosis and worse outcome in men [35].

In conclusion, the outcome of COVID-19 appears to depend on the interaction between genetic and hormonal factors, age and previous comorbidities, and environmental factors that also include, among others, vitamin D status. Our study confirms that inadequate vitamin D status is associated with moderate or severe COVID-19 requiring hospitalization. However, to identify the causal relationships and mechanisms underlying our observation of a significant sex difference in circulating 25(OH)D concentrations in patients who develop moderate or severe COVID-19 requiring hospitalization, additional clinical and basic studies are warranted.

Conflict of interest

The authors declare that they have no conflict of interest.

REFERENCES

1. Jakovac H (2020) COVID-19 and vitamin D-Is there a link and an opportunity for intervention? Am J Physiol Endocrinol Metab 318:E589. https://doi.org/10.1152/ajpendo.00138.2020
2. Teymoori-Rad M, Shokri F, Salimi V, Marashi SM (2019) The interplay between vitamin D and viral infections. Rev Med Virol 29:e2032. https://doi.org/10.1002/rmv.2032
3. Autier P, Mullie P, Macacu A, Dragomir M, Boniol M, Coppens K, Pizot C, Boniol M (2017) Effect of vitamin D supplementation on non-skeletal disorders: a systematic review of meta-analyses and randomised trials. Lancet Diabetes Endocrinol 5:986-1004. https://doi.org/10.1016/s2213-8587(17)30357-1
4. Martineau AR, Jolliffe DA, Hooper RL, Greenberg L, Aloia JF, Bergman P, Dubnov-Raz G, Esposito S, Ganmaa D, Ginde AA, Goodall EC, Grant CC, Griffiths CJ, Janssens W, Laaksi I, Manaseki-Holland S, Mauger D, Murdoch DR, Neale R, Rees JR, Simpson S, Jr., Stelmach I, Kumar GT, Urashima M, Camargo CA, Jr. (2017) Vitamin D supplementation to prevent acute respiratory tract infections: systematic review and meta-analysis of individual participant data. Bmj 356:i6583. https://doi.org/10.1136/bmj.i6583
5. Laird E, Rhodes J, Kenny RA (2020) Vitamin D and Inflammation: Potential Implications for Severity of Covid-19. Ir Med J 113:81
6. Saponaro F, Franzini M, Okoye C, Antognoli R, Campi B, Scalese M, Neri T, Carrozzi L, Monzani F, Zucchi R, Celi A, Paolicchi A, Saba A (2021) Is There a Crucial Link Between Vitamin D Status and Inflammatory Response in Patients With COVID-19? Front Immunol 12:745713. https://doi.org/10.3389/fimmu.2021.745713
7. Peckham H, de Gruijter NM, Raine C, Radziszewska A, Ciurtin C, Wedderburn LR, Rosser EC, Webb K, Deakin CT (2020) Male sex identified by global COVID-19 meta-analysis as a risk factor for death and ITU admission. Nat Commun 11:6317. https://doi.org/10.1038/s41467-020-19741-6
8. Pop-Kostova A, Ruskovska T (2021) Clinical significance of circulating enzymes in patients with COVID-19. Knowledge - International Journal 47:611-616
9. Pop-Kostova A, Ruskovska T (2021) Inflammatory biomarkers in patients with COVID-19. Knowledge - International Journal 49:595-600
10. Cucinotta D, Vanelli M (2020) WHO Declares COVID-19 a Pandemic. Acta Biomed 91:157-160. https://doi.org/10.23750/abm.v91i1.9397
11. Arsenault C, Gage A, Kim MK, Kapoor NR, Akweongo P, Amponsah F, Aryal A, Asai D, Awoonor-Williams JK, Ayele W, Bedregal P, Doubova SV, Dulal M, Gadeka DD, Gordon-Strachan G, Mariam DH, Hensman D, Joseph JP, Kaewkamjornchai P, Eshetu MK, Gelaw SK, Kubota S, Leerapan B, Margozzini P, Mebratie AD, Mehata S, Moshabela M, Mthethwa L, Nega A, Oh J, Park S, Passi-Solar Á, Pérez-Cuevas R, Phengsavanh A, Reddy T, Rittiphairoj T, Sapag JC, Thermidor R, Tlou B, Valenzuela Guiñez F, Bauhoff S, Kruk ME (2022) COVID-19 and resilience of healthcare systems in ten countries. Nat Med 28:1314-1324. https://doi.org/10.1038/s41591-022-01750-1

12. Mogharab V, Ostovar M, Ruszkowski J, Hussain SZM, Shrestha R, Yaqoob U, Aryanpoor P, Nikkhoo AM, Heidari P, Jahromi AR, Rayatdoost E, Ali A, Javdani F, Farzaneh R, Ghanaatpisheh A, Habibzadeh SR, Foroughian M, Ahmadi SR, Akhavan R, Abbasi B, Shahi B, Hakemi A, Bolvardi E, Bagherian F, Motamed M, Boroujeni ST, Jamalnia S, Mangouri A, Paydar M, Mehrasa N, Shirali D, Sanmarchi F, Saeed A, Jafari NA, Babou A, Kalani N, Hatami N (2022) Global burden of the COVID-19 associated patient-related delay in emergency healthcare: a panel of systematic review and meta-analyses. Global Health 18:58. https://doi.org/10.1186/s12992-022-00836-2

13. Hamid H, Abid Z, Amir A, Rehman TU, Akram W, Mehboob T (2020) Current burden on healthcare systems in low- and middle-income countries: recommendations for emergency care of COVID-19. Drugs Ther Perspect 36:466-468. https://doi.org/10.1007/s40267-020-00766-2

14. Aslan MT, Aslan İ, Özdemir Ö (2020) Letter to the Editor: Is Vitamin D One of the Key Elements in COVID-19 Days? J Nutr Health Aging 24:1038-1039. https://doi.org/10.1007/s12603-020-1413-5

15. Kato S, Kurzrock R (2020) Repurposing Interleukin-6 Inhibitors to Combat COVID-19. J Immunother Precis Oncol 3:52-55. https://doi.org/10.36401/jipo-20-11

16. Grant WB, Lahore H, McDonnell SL, Baggerly CA, French CB, Aliano JL, Bhattoa HP (2020) Evidence that Vitamin D Supplementation Could Reduce Risk of Influenza and COVID-19 Infections and Deaths. Nutrients 12. https://doi.org/10.3390/nu12040988

17. Pagano MT, Peruzzu D, Ruggieri A, Ortona E, Gagliardi MC (2020) Vitamin D and Sex Differences in COVID-19. Front Endocrinol (Lausanne) 11:567824. https://doi.org/10.3389/fendo.2020.567824

18. Ghosh S, Klein RS (2017) Sex Drives Dimorphic Immune Responses to Viral Infections. J Immunol 198:1782-1790. https://doi.org/10.4049/jimmunol.1601166

19. Mjaess G, Karam A, Aoun F, Albisinni S, Roumeguère T (2020) COVID-19 and the male susceptibility: the role of ACE2, TMPRSS2 and the androgen receptor. Prog Urol 30:484-487. https://doi.org/10.1016/j.purol.2020.05.007

20. Mendelsohn ME (2002) Protective effects of estrogen on the cardiovascular system. Am J Cardiol 89:12E-17E; discussion 17E-18E. https://doi.org/10.1016/s0002-9149(02)02405-0

21. Takase T, Tsugawa N, Sugiyama T, Ikesue H, Eto M, Hashida T, Tomii K, Muroi N (2022) Association between 25-hydroxyvitamin D levels and COVID-19 severity. Clin Nutr ESPEN 49:256-263. https://doi.org/10.1016/j.clnesp.2022.04.003

22. Jain A, Chaurasia R, Sengar NS, Singh M, Mahor S, Narain S (2020) Analysis of vitamin D level among asymptomatic and critically ill COVID-19 patients and its correlation with inflammatory markers. Sci Rep 10:20191. https://doi.org/10.1038/s41598-020-77093-z

23. Chiodini I, Gatti D, Soranna D, Merlotti D, Mingiano C, Fassio A, Adami G, Falchetti A, Eller-Vainicher C, Rossini M, Persani L, Zambon A, Gennari L (2021) Vitamin D Status and SARS-CoV-2 Infection and COVID-19 Clinical Outcomes. Front Public Health 9:736665. https://doi.org/10.3389/fpubh.2021.736665

24. Dror AA, Morozov N, Daoud A, Namir Y, Yakir O, Shachar Y, Lifshitz M, Segal E, Fisher L, Mizrachi M, Eisenbach N, Rayan D, Gruber M, Bashkin A, Kaykov E, Barhoum M, Edelstein M, Sela E (2022) Pre-infection 25-hydroxyvitamin D3 levels and association with severity of COVID-19 illness. PLoS One 17:e0263069. https://doi.org/10.1371/journal.pone.0263069

25. Hopefl R, Ben-Eltriki M, Deb S (2022) Association Between Vitamin D Levels and Inflammatory Markers in COVID-19 Patients: A Meta-Analysis of Observational Studies. J Pharm Pharm Sci 25:124-136. https://doi.org/10.18433/jpps32518

26. Ben-Eltriki M, Hopefl R, Wright JM, Deb S (2022) Association between Vitamin D Status and Risk of Developing Severe COVID-19 Infection: A Meta-Analysis of Observational Studies. J Am Nutr Assoc 41:679-689. https://doi.org/10.1080/07315724.2021.1951891

27. Getachew B, Tizabi Y (2021) Vitamin D and COVID-19: Role of ACE2, age, gender, and ethnicity. J Med Virol 93:5285-5294. https://doi.org/10.1002/jmv.27075

28. Correale J, Ysrraelit MC, Gaitán MI (2010) Gender differences in 1,25 dihydroxyvitamin D3 immunomodulatory effects in multiple sclerosis patients and healthy subjects. J Immunol 185:4948-4958. https://doi.org/10.4049/jimmunol.1000588

29. Spanier JA, Nashold FE, Mayne CG, Nelson CD, Hayes CE (2015) Vitamin D and estrogen synergy in Vdr-expressing CD4(+) T cells is essential to induce Helios(+)FoxP3(+) T cells and prevent autoimmune demyelinating disease. J Neuroimmunol 286:48-58. https://doi.org/10.1016/j.jneuroim.2015.06.015

30. Lundqvist J, Norlin M, Wikvall K (2011) 1α,25-Dihydroxyvitamin D3 exerts tissue-specific effects

on estrogen and androgen metabolism. Biochim Biophys Acta 1811:263-270. https://doi.org/10.1016/j.bbalip.2011.01.004

31. Hars M, Mendes A, Serratrice C, Herrmann FR, Gold G, Graf C, Zekry D, Trombetti A (2020) Sex-specific association between vitamin D deficiency and COVID-19 mortality in older patients. Osteoporos Int 31:2495-2496. https://doi.org/10.1007/s00198-020-05677-6
32. Knovich MA, Storey JA, Coffman LG, Torti SV, Torti FM (2009) Ferritin for the clinician. Blood Rev 23:95-104. https://doi.org/10.1016/j.blre.2008.08.001
33. Cullis JO, Fitzsimons EJ, Griffiths WJ, Tsochatzis E, Thomas DW (2018) Investigation and management of a raised serum ferritin. Br J Haematol 181:331-340. https://doi.org/10.1111/bjh.15166
34. Kaushal K, Kaur H, Sarma P, Bhattacharyya A, Sharma DJ, Prajapat M, Pathak M, Kothari A, Kumar S, Rana S, Kaur M, Prakash A, Mirza AA, Panda PK, Vivekanandan S, Omar BJ, Medhi B, Naithani M (2022) Serum ferritin as a predictive biomarker in COVID-19. A systematic review, meta-analysis and meta-regression analysis. J Crit Care 67:172-181. https://doi.org/10.1016/j.jcrc.2021.09.023
35. Gandini O, Criniti A, Gagliardi MC, Ballesio L, Giglio S, Balena A, Caputi A, Angeloni A, Lubrano C (2021) Sex-disaggregated data confirm serum ferritin as an independent predictor of disease severity both in male and female COVID-19 patients. J Infect 82:414-451. https://doi.org/10.1016/j.jinf.2020.10.012