Peer Review Report

Review Report on Seasonal Variation of Total and Bioavailable 25-Hydroxyvitamin D (25(OH)D) in Healthy Adult Slovenian Population

Original Research, Acta Biochim. Pol.

Reviewer: Marta Karaźniewicz-Łada

Submitted on: 12 May 2024

Article DOI: 10.3389/abp.2024.13108

EVALUATION

Q 1 Please summarize the main findings of the study.

The article presents results of the seasonal changes of free and bioavailable vitamin D concentrations in healthy volunteers. The Authors reported significantly lower 25(OH)D levels in winter compared to summer. Moreover, they observed a positive correlation between the total and bioavailable 25(OH)D but the strength of the correlation depended on the season. They presented possible explanations for this observations.

Q 2 Please highlight the limitations and strengths.

Limitations of the study:

- 1) it is unknown how the Authors solved the problem with a variable daily exposure to the sun. Due to the wide range of participants' age (19-70 years), the subjects may vary in daily exposure to the sun. Younger people spend more time outdoors compared to older people who spend most of the day at home. This may influence the skin synthesis of vitamin D.
- 2) The novelty of the study is limited. The Authors did not provide any plausible explanations on clinical significance of the obtained results

Q 3 Please comment on the methods, results and data interpretation. If there are any objective errors, or if the conclusions are not supported, you should detail your concerns.

The methods for determination of the analyses compounds should be better described. For example it is unknown how the albumin levels were determined. Moreover, the formulas used for determination of free and bioavailable vitamin D fraction should be added and the calculation process should be described. Conclusions provided by the Authors are rather general statement regarding vitamin D supplementation and fortification of food. Authors should provide conclusions referring to the obtained results.

Check List

Q 4 Please provide your detailed review report to the editor and authors (including any comments on the Q4 Check List)

The article presents an interesting study on the seasonal variation of total and bioavailable 25(OH)D in healthy Slovenian subjects. Study design and all statistical analyses seem to be performed properly. However, several issues need to be addressed before the manuscript acceptance.

- 1) Problem with abbreviations: abbreviations should be defined when used for the first time. The Authors explained the same abbreviation several times (for example VDBP). Moreover, it is not clear if VDBP and DBP mean the same parameter as they are used interchangeably. In the abbreviation list, there is a mistake in the formula of 1,25-dihydroxycholecalciferol
- 2) lines 58-69: Please review the list of physiological functions as the text contains several unnecessary repetitions (for example point 3)
- 3) lines 73-76: provide the vitamin D ranges defining normal, insufficient and deficient vitamin D body status

- 4) lines 78-184: the section should be shortened as it contained repetitions of the same information, for example the information that 25(OH)D is used as a measure of vitamin D status is repeated in lines 102, 119, 166 and 181.
- 5) line 176: do the Authors mean 25(OH)D?
- 6) lines 207–227: what was the mean daily exposure to the sun? Due to the wide range of participants' age (19–70 years), the subjects may vary in daily exposure to the sun. Younger people spend more time outdoors compared to older people who spend most of the day at home. This may influence the skin synthesis of vitamin D.
- 7) How did the Authors estimate the sample size of the study? Is the current sample size power-based?
- 8) line 236: please explain the abbreviations: "S-25(OH)D", "S-albumin", "S-DBP"
- 9) The methods for determination of the analyses compounds should be better described. For example it is unknown how the albumin levels were determined. Moreover, the formulas used for determination of free and bioavailable vitamin D fraction should be added and the calculation process should be described.
- 10) Discussion: lines 315–317 and line 321–324 present the same information on the study results. Moreover, it is unclear what do "r(85)" mean. The Authors should better explain what they mean by "stored vitamin D" (bound to DBP?). Units for the determined parameters should be provided (lines 365, 372).
- 11) Lines 390–413: it is not clear how the presented description on supplementation refer to the obtained results. It should be explained or at least referenced to the literature data.
- 12) Conclusions provided by the Authors are rather general statement regarding vitamin D supplementation and fortification of food. Authors should provide conclusions referring to the obtained results.
- 13) Figure 1: the figure legend is too long. The description should be shortened or moved to the Introduction part
- 14) Table 2: The plausible explanation for the presented results on correlations should be described in the discussion. For example, it is not clear why the Authors analysed the correlation between total 25(OH)D in winter and bioavailable 25(OH)D in summer.

Q 5	Is the English language of sufficient quality?
Yes.	
Q 6	Is the quality of the figures and tables satisfactory?
Yes.	
0.7	Describe reference list seconds relevant literature de motele and in an orbital manage.
Q /	Does the reference list cover the relevant literature adequately and in an unbiased manner?
Yes.	
Q 8	Are the statistical methods valid and correctly applied? (e.g. sample size, choice of test)
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No.	

Q 10 Are the data underlying the study available in either the article, supplement, or deposited in a repository? (Sequence/expression data, protein/molecule characterizations, annotations, and taxonomy data are required to be deposited in public repositories prior to publication)							
Yes.							
Q 11 procedu	Does the study adhere to ethical standards re?	including ethics	committee	approv	al and consen		
Yes.							
Q 12	Have standard biosecurity and institutional	safety procedu	res been ac	lhered to	9?		
Yes.							
QUALITY A	ASSESSMENT						
Q 13	Originality						
Q 14	Rigor						
Q 15	Significance to the field						
Q 16	Interest to general audience						
Q 17	Quality of the writing						
Q 18	Overall quality of the study						