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Rising trend in 25OHD during COVID-19 pandemic in Ireland

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ABSTRACT

Objectives: Vitamin D status has improved substantially in Ireland over the past 40 years. Since the onset of the Covid-19 pandemic in 2020, there have been plausible suggestions about the need to augment vitamin D intake by supplementation on a population basis in order to prevent SARS-Co-V2 infection and reduce mortality. Some groups have advocated supplementations for all adults, but governmental agencies have advocated targeted supplementation. We sought to explore the effect of COVID-19 pandemic on vitamin D status from April 2020 to March 2021 by comparison with prior trend analysis of vitamin D status over the past 28 years.

Setting: University hospital, Dublin, Ireland.

Participants: Laboratory-based samples of circulating 25-hydroxyvitamin D (25OHD).

Primary and secondary outcome measures: Primary outcome: comparing yearly average 25OHD in the 12 months prior to the pandemic (April 2019 to March 2020) with the first 12 months (April 2020 to March 2021) of the pandemic. Secondary outcome: comparing prevalence of vitamin D deficiency and vitamin D excess during the two time periods.

Results: Regarding the primary outcome in a large sample (n = 100,505), we noted the average yearly 25OHD increased by 2.8 nmol/L (61.4, 95%CI 61.5 – 61.7 vs 58.6, 95% CI 58.4-58.9, p <.001). This yearly increase is almost 3-fold higher than the yearly increase in average 25OHD based on two similar trend analyses that we conducted between 1993 and 2016. Regarding secondary outcome, we showed a lower prevalence of low 25OHD indicating benefit, but we also showed a higher prevalence of high 25OHD.

Conclusions: The pandemic has emphasised the need to correct vitamin D deficiency. Rather than a blanket recommendation about vitamin D supplementation for all adults during the

pandemic, we recommend a targeted approach of supplementation within current governmental guidelines that augments vitamin D intake in at-risk groups.

Strengths and limitations

- This is a laboratory-based 25OHD trend analysis that includes a large sample size.
- This trend analysis is compared to with two prior trend analyses dating back to 1993.
- This is not a population-based representative sample and is subject to selection bias.
- There is no clinical information about reason for 25OHD testing or about vitamin D supplementation.

INTRODUCTION

Vitamin D is an essential micronutrient in all age groups for bone and muscle health[1-4]. Vitamin D may have a role in extraskeletal health such as the immune response to acute respiratory illnesses[5], which is pertinent during the COVID-19 pandemic[6]. Following the onset of the pandemic, some reports advocated blanket oral supplementation to entire populations with doses ranging from 20 µg to 50 µg daily[7-10], which are in excess of vitamin D intake requirement as specified by governmental agencies in Europe and North America[1-4]. By contrast, other groups have countered this blanket recommendation, favouring a targeted approach based on modelling of total vitamin D intakes[11 12].

These governmental reports, which have been issued over the past decade prior to the pandemic, are based on similar health outcomes such as musculoskeletal health, falls and total risk of mortality, but not on immune response to infection. Subsequently, governmental agencies from England and Ireland issued advice about vitamin D supplementation during the pandemic, targeting at-risk populations, including measures to facilitate supplementation[13 14](Table 1). Given the ongoing concerns about vitamin D inadequacy across Europe[15 16], we suggest that these governmental measures at augmenting vitamin D status during the pandemic can be beneficial to at-risk groups but that blanket recommendations may predispose to unnecessary self-supplementation of vitamin D doses more than requirement in healthy persons.

Table 1 Vitamin D intake recommendations

Region	Year	Vitamin D Intake	Population Reference Intake µg/day (age group)
USA & Canada[1]	2011	Total intake	15 (<70 y) 20 (≥70 y)
Nordic Countries[2]	2014	Total intake	10 (<75 y) 20 (≥75 y)
European Union[3]	2016	Total intake	15 (≥18)
United Kingdom[4]	2016	Total intake	10 (all adults)
COVID-19 vitamin D advocates[7-10]	2020	Supplemental intake	20-50 (all adults, all year)
Post COVID-19 United Kingdom[13]	2020	Supplemental intake	10 (October to March for healthy adults) 10 (all year for those with limited sunlight exposure)
Post COVID-19 Ireland[14]	2020	Supplemental intake	10 (October to March >65 y) 15 (all year for housebound >65 y)

Total intake refers to vitamin D intake from all sources: skin production and oral intake (natural foods, fortified foods, supplements). Population reference intakes for governmental agencies refers to intake that meets the needs of 97.5% on the population; these intakes are based on health outcomes such as musculoskeletal health, falls and total risk of mortality,

Ensuring adequate vitamin D intake across all age groups is a challenging population health task. Vitamin D intake requirements must be modelled to cover total intake because vitamin D supply has various sources: skin production on exposure to ultra-violet light, natural foods, fortified foods, and supplements. There is almost complete unanimity about total vitamin D intake requirements across governmental agencies for North America and for Europe[1-4]. In circumstances of minimal or no sunlight exposure, the total oral intake requirement approximates to between 10 µg and 20 µg daily (400 IU to 800 IU daily)[1-4]. The pre-eminent measure of vitamin D status is the measurement of the circulating vitamin D metabolite, 25-hydroxyvitamin D (25OHD).

We have been engaged in clinical research on 25OHD since the 1970s[17]. We have noted substantial improvement in vitamin D status over the past 45 years. In our early studies in Ireland, we noted the primacy of oral intake over sunlight exposure for both the prevention and correction of vitamin D deficiency[18-21]. Following the advent of voluntary milk fortification in Ireland in the 1980s at 1 μg/100 ml (but more recently some milk products fortify at 2 μg/100 ml) and the ready availability of low-dose vitamin D supplements, vitamin D status improved substantially[22]. In more recent years, we reported instances of hypervitaminosis D raising concerns about excessive oral intake of vitamin D[23].

We have published two trend analyses of laboratory-based 25OHD results: the first trend study reported 25OHD from 1993 to 2013 that incorporated a time series analysis to predict 25OHD trend from 2014 to 2016[24]; the second trend study reported 25OHD from 2014 to 2016 that confirmed the forecast analysis from the first study[25]. Our analyses over the past 24 years in Ireland show yearly average 25OHD concentrations increased by about 1 nmol/L/year. As early as 2014, we recognized a dual concern about vitamin D status in Ireland: hypovitaminosis D in at risk groups; and hypervitaminosis D due to high supplemental intake, especially from over-the-counter preparations in individuals who already have adequate vitamin D status[26]. One of the consequences of raising public awareness, whether it be from governmental agencies or from professional bodies, is the increased supply of vitamin D supplements, which are available for over-the-counter purchase.

We sought to explore the effect of COVID-19 pandemic on vitamin D status from April 2020 to March 2021 given the conflicting advice: governmental agencies promoting vitamin D supplementation in at-risk groups, and groups advocating blanket recommendations for vitamin D supplementation to all adults.

METHODS

Data mining

Our laboratory data system was mined to identify all 25OHD samples over a 24-month period from April 2019 to March 2021: pre-COVID-19 era being represented by 12 months from April 2019 to March 2020, and the COVID-19 era being represented by April 2020 to March 2021. In order to identify duplicate samples, the following identifiers were extracted: date of birth and medical record number. Additional data included age, sex, referral source (either hospital consultant or general practitioner), and date of 25OHD test.

The sample size for the 24 months from April 2019 to March 2021 was 137,963; after excluding those with more than one sample during the 2 years (n = 37,458) the final sample was trimmed to 100,505. Regarding those cases with more than one 25OHD sample, the analysis was based on the 25OHD being first sample, which hereafter is termed the first in sequence. A secondary analysis was performed, whereby the selected 25OHD of those cases with more than one sample was the final sample, which hereafter is termed the last in sequence .

Research Ethics Approval

The Clinical Audit Committee, St Vincent's Healthcare Group, approved the extraction and audit of the data from our hospital laboratory system (Reference number: 3174).

Public and Patient Involvement

It was not appropriate or possible to involve patients or the public in the design, or conduct, or reporting, or dissemination plans of our research

Study design

The primary analysis related to comparison of 25OHD in the pre-Covid-19 era, hereafter referred to as group 1, with 25OHD in the Covid-19 era, hereafter referred to as group 2. The prevalence estimates for categories of 25OHD in the two groups was calculated according to the following thresholds: <30 nmol/L; 30-50 nmol/L; 50.1-125 nmol/L; 125.1 to 175 nmol/L; >175 nmol/L. In addition, a composite analysis of the entire group over the 2 years was performed in order to assess vitamin D status according to age, sex, and different age groups.

Analytical Methods

Serum 25OHD concentrations were quantified using the Elecsys Vitamin D Total (Roche Diagnostics GmbH, Mannheim, Germany) automated competitive binding protein assay, which measures total vitamin D, including isomers in the form of the C3 epimer as well as 24,25OHD metabolites. This is not specifically corrected for; rather, an assumption is made that there is a non-statistically significant difference in the percent concentration of vitamin D metabolites relative to the measured concentration in patient samples tested over the three-year period. The average inter-assay coefficients of variation (CV) for the 25OHD assay determined over the period studied were as follows: 14.6% at a mean concentration of 37.7 nmol/L, 8.7% at a mean concentration of 74.6 nmol/L, and 7.6% at a mean concentration of 112.1 nmol/L. Functional sensitivity was verified at 15 nmol/L (%CV <20%). To ensure a high standard of analysis for serum 25OHD concentrations, the laboratory participates in an external quality assurance scheme: the Vitamin D External Quality Assessment Scheme (DEQAS)[27]. During the 2-year period 2019 to 2021, our assay displayed a mean bias of 1.12 % from target values provided by the Centers for Disease Control and Prevention (Atlanta,

Georgia, USA) using their isotope dilution LC-MS/MS Reference Measurement Procedure[28]. Assay performance met DEQAS defined performance criteria set at ±25%. For samples with undetectable 25OHD (<15 nmol/L), the result was censored at 14.9 pmol/L. For samples with high 25OHD (>175 mol/L), the result was censored at 175.1 nmol/L.

Statistical analysis

Descriptive statistics are presented as number and percent for categorical variables and as mean (95% confidence intervals) for continuous variables. Differences in independent categorical variables were tested by chi-square. The distribution for 25OHD exhibited positive skewness and thus was log-transformed prior to parametric statistical tests. Differences between two means were tested by independent-samples *t* test using Levene's test for equality of variances. Statistics were considered significant if *p* value <.05. Analyses were performed using IBM SPSS Statistics version 25 (Armonk, NY, USA).

RESULTS

Group 1 and group 2 were similar with respect to age, sex, and source of referral (Table 1). Mean 25OHD was 2.8 nmol/L higher in group 2 compared to group 1 (61.4, 95%CI 61.5, 61.7 nmol/L vs 58.6, 95% CI 58.4, 58.9 nmol/L, *p* <.001) (Table 1). If the 25OHD duplicate result was selected as last in sequence, then mean 25OHD was 5.1 nmol/L higher in group 2 compared to group 1 (63.3, 95%CI 63.2, 63.6 nmol/L vs 58.2, 95% CI 58.0, 58.5 nmol/L, *p* <.001). In group 2 compared to group 1, there was a lower percent (12.0% vs 13.4%) of low vitamin D status (25OHD <30 nmol/L) but a higher percent (2.1% vs 1.7%) of high vitamin D status (25OHD >125 nmol/L) (*p* <.001) (Table 1). The monthly 25OHD trimmed values for both groups are plotted showing the seasonal variation (Figure 1). The average seasonal change in

25OHD from nadir to peak was almost identical for both at 20.2 nmol/L in group 1 and 20.1 nmol/L in group 2.

Table 2: Descriptive statistics

Variable	Group 1 (*n* = 58,642)	Group 2 (*n* = 41,863)
Age, years	52.5 (52.3, 52.7)	52.3 (52.1, 52.5)
Women : Men, %	66.4 : 33.6	64.3 : 35.7
Hospital : Primary Care, %	30.0 : 70.0	25.5 : 74.5
25OHD status, %		
<30 nmol/L	13.4	12.0
30-50 nmol/L	28.4	25.1
51-125 nmol/L	56.6	60.7
125.1-175 nmol/L	1.4	1.8
>175 nmol/L	0.3	0.3
25OHD, nmol/L	58.6 (58.4-58.9)	61.4 (61.5 – 61.7)

Results are presented as % for categorical variables and as mean (95%CI) for continuous variables

For the composite analysis, mean 25OHD was higher in women compared to men (61.3, 95%CI 61.1 – 61.5 nmol/L vs 56.9, 95% CI 56.7-57.3 nmol/L, p <.001), and in those sourced from primary care compared to hospital care (60.7, 95%CI 60.5 – 60.9 nmol/L vs 57.4, 95% CI 57.0-57.7 nmol/L, p <.001). Vitamin D status according to age categories showed that infants and toddlers had the lowest prevalence of 25OHD <30 nmol/L and the highest prevalence for 25OHD >125 nmol/L (Table 3). Regarding vitamin D status according to age categories and sex, adult females had better vitamin D status than males, but in infants a greater percent of females compared to males had both the lowest and the highest prevalence of vitamin D status, but the numbers were small (Table 3).

Table 2. Crosstabulation of vitamin D status according to age categories and sex

Age Categories		Vitamin D Status, nmol/L				
		<30	30 - 50	50.1 - 125	125.1 - 175	>175
Infants	Female (*n*=87)	8.0	6.9	67.8	10.3	6.9
	Male (*n*=128)	4.7	10.2	76.6	7.8	0.8
	Total (*n*=215)	6.0	8.8	73.0	8.8	3.3
Toddlers	Female (*n*=250)	3.6	17.6	74.4	3.2	1.2
	Male (*n*=288)	6.3	15.6	73.3	4.5	0.3
	Total (*n*=538)	5.0	16.5	73.8	3.9	0.7
Children & adolescents	Female (*n*=3,253)	16.5	33.4	49.5	0.6	0.1
	Male (*n*=2,271)	13.7	30.8	54.3	1.0	0.2
	Total (*n*=5,524)	15.4	32.3	51.4	0.7	0.1
Young adults	Female (*n*=42,757)	12.0	28.4	57.8	1.6	0.3
	Male (*n*=20,533)	15.9	31.0	51.7	1.1	0.3
	Total (*n*=63,290)	13.3	29.2	55.8	1.4	0.3
Older adults	Female (*n*=19,493)	10.0	19.1	68.3	2.3	0.4
	Male (*n*=11,415)	14.1	26.5	57.7	1.4	0.2
	Total (*n*=30,908)	11.6	21.8	64.3	2.0	0.3
Total	Female (*n*=65,840)	11.6	25.8	60.5	1.8	0.3
	Male (*n*=34,635)	15.1	29.3	54.1	1.3	0.2
	Total (*n*=100,475)	12.8	27.0	58.3	1.6	0.3

Results are presented as %

DISCUSSION

In a trend analysis of laboratory-based 25OHD samples comparing yearly average 25OHD in the 12 months before onset of the Covid-19 pandemic (April 2019 to March 2020) with the first 12 months of the Covid-19 pandemic in Ireland (April 2020 to March 2021), we showed the average yearly 25OHD increased by 2.8 nmol/L/year. This yearly trend was nearly 3-fold higher than the average yearly increase in 25OHD of 1 nmol/L/year that we recorded in two prior trend analyses of laboratory-based samples from 1993 to 2016[24 25]. If the 25OHD duplicate was selected as last in sequence for the trend analysis, then average 25OHD increase during the pandemic was even higher at 5.1 nmol/L/year. We did observe benefit

with respect to their being lower prevalence of 25OHD <30 nmol/L, but to a lesser extent there was a higher prevalence of 25OHD >125 nmol/L.

The 25OHD threshold for diagnosis of vitamin D deficiency depends on the approach: whether is it viewed as being population-based[1 4] or as being case-based[29 30]. There are important differences. For instance, the Institute of Medicine (IOM) for the USA and Canada in 2011 used a statistical probability method to determine the prevalence of nutrient inadequacy[31]. The IOM set a 25OHD threshold of 30 nmol/L. IOM referred to 25OHD as a "biomarker of exposure" but not as a "biomarker of effect", which means that 25OHD is the preeminent measure of total vitamin D intake, but it only estimates risk of disease. Thus, 25OHD below 30 nmol/L was defined by IOM as "risk of deficiency". Choosing a higher 25OHD threshold for defining vitamin D deficiency inflates the prevalence[31]. Similarly, the Scientific Advisory Committee on Nutrition (SACN) for the UK set a 25OHD threshold of 25 nmol/L[4]. Another key difference is that governmental agencies set their specifications for vitamin D intake based on total vitamin D intake. Modelling intake from all sources estimates the shortfall in vitamin D intake that can be bridged by supplemental intake[32].

Whereas, in a case-based approach, the 25OHD threshold is agreed by expert clinical opinion on optimal vitamin D status in individuals, such as the European Calcified Tissue Society that set a 25OHD threshold at 50 nmol/L[29]. The Endocrine Society, which set an even higher 25OHD threshold at 75 nmol/L, specified vitamin D intakes as supplemental intake not total vitamin D intake up to 37µg to 50µg that were up to 5-fold higher than those specified by IOM[30]. The higher 25OHD threshold and higher vitamin D intakes were critiqued as lacking evidence and as overestimating the 25OHD response to vitamin D supplementation[33 34]. That critique has been validated by the findings of the subsequent VITAL trial using 50 µg vitamin

D daily for 5 years with 25,871 participants that reported no benefit with respect to lowering incidence of invasive cancer or cardiovascular events[35].

Governmental agencies have adopted a precautionary approach to vitamin D harm. The tolerable upper intake level (UL) for vitamin D is 100 µg (4000 IU) daily, as set by governmental agencies [1 36]. The UL is 10-fold higher than the average total vitamin D intake requirement for healthy adults[1]. The UL is not intended as a target intake; rather, the risk for harm begins to increase once vitamin D intake surpasses this level[37]. In Ireland, over-the-counter vitamin D products are regulated by the Food Safety Authority of Ireland. The UL determines the maximum dose of a vitamin D product that can be marketed over-the-counter. Labelling requires that the dose not exceed the UL. There is no requirement to model total vitamin D intake, such that a healthy adult with vitamin D intake from other sources can self-medicate with a vitamin D supplement at the threshold of the UL.

Risk of harm from vitamin D supplementation is vaguely defined. Defining harm just in terms of hypercalcemia sets the vitamin D dose and the 25OHD threshold at high levels[30]. Some vitamin D studies, where the primary endpoint was prevention of fracture or prevention of falls or increase in bone mineral density, have suggested increased risk of the primary outcome: (1) two studies showed more fractures using vitamin D 12,500 µg yearly[38] or using 7,500 µg yearly[39]; (2) five studies showed more falls using vitamin D 12,500 µg yearly[38], or using daily dose of 100 µg vitamin D daily [40], or using 1500 µg vitamin D monthly[41], or using 2500 µg vitamin D monthly, or using a range of daily vitamin D doses from 50 µg to 100 µg[42]; (3) and one study showed lower bone mineral density using 100 µg or 250 µg vitamin D daily[43]. A recent meta-analysis of RCTs showed that vitamin D did not have a beneficial effect on muscle but may have adverse effects on muscle health[44]. An RCT investigating distal radius

fracture healing with vitamin D bolus doses (placebo, 750 µg, or 1875 µg twice 6 weeks apart) showed no benefit of the lower dose compared to control dose but adverse effects of the highest dose compared to controls[45]. Most participants in these trials did not have 25OHD below 30 nmol/L; also, intermittent boluses have a different pharmacokinetic profile to daily dosing, thus limiting dose comparisons[29].

During the Covid-19 pandemic, some groups have advocated the need for mass vitamin D supplementation in order to enhance immune response to SARS-Co-V2 infection [7 9 10 46 47]. EFSA permits stating that benefit from vitamin D supplementation covers the normal functioning of the immune system without specifying a vitamin D intake for this benefit[48]. The is some evidence of benefit from vitamin D randomised controlled trials (RCTs) that have been conducted during the pandemic but there is wide variation in vitamin D doses[49-54]. Further studies should provide clarity about benefit and optimal vitamin D schedules. These studies might provide cogent evidence for higher vitamin D intake requirements that could forming part of population-based or case-based recommendations. Meanwhile, the likely effect of advocating for mass supplementation is that individuals, who are best able to self-medicate, are the ones who are least likely to need supplementation. Frail older adults, lower socioeconomic groups and minority ethnic groups are more likely to have lower 25OHD and are less likely to afford the means for supplementation[55 56]. It is better to have a targeted approach to vitamin D supplementation such as the frail older adult[13 14]. Mandatory fortification of foodstuffs with vitamin D, which has been shown to be effective in Finland[57], poses many challenges[58]. Voluntary fortification, while less satisfactory than mandatory fortification, is effective at ameliorating seasonal decline in 25OHD as has been shown in Ireland[22]. Fortification with any nutrient (whether mandatory or voluntary) in addition to supplementation (whether mandatory or voluntary), can result in total nutrient intakes that

are higher than requirement and may even exceed the UL, especially if nutrient intake targets the RDA and not the average requirement[59].

Our trend analysis has many limitations. First and foremost, it is not a representative sample because samples are not collected as part of a population-based survey. We do not have information on the clinical indication for the test nor do we know whether patients were on vitamin D supplements or had an underlying condition that predisposed to vitamin D deficiency. We only had limited information on calcium status and parathyroid status (not shown). The 12-month trend analysis is too short to declare with any certainty that the pandemic has contributed to a shift upwards in the yearly average 25OHD increase.

In conclusion, we report in Ireland a laboratory-based trend analysis of 25OHD showing that the yearly average 25OHD has increased about 3-fold during the first year of Covid-19 pandemic compared to prior trend analysis. This trend reflects benefit for those with low 25OHD. Public health efforts should be redoubled at maximising the provision of specified daily vitamin D supplements in at-risk groups and clinically vulnerable patients. There should be a precautionary approach to population-based blanket recommendations for vitamin D supplementation to healthy adults.

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Author Contributions:

All authors read, edited, and approved the submitted manuscript. Conception and design: MMcK. Laboratory analysis: PT and MK. Statistical analysis and data interpretation: MMcK.

Drafting manuscript: MMcK. Critical review and revising manuscript: all authors. MMcK takes responsibility for the integrity of the data analysis.

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All authors declare that they have no competing interests.

Data sharing statement:

Data described in the manuscript, code book, and analytic code will be made available upon request pending application to and approval by the corresponding author.

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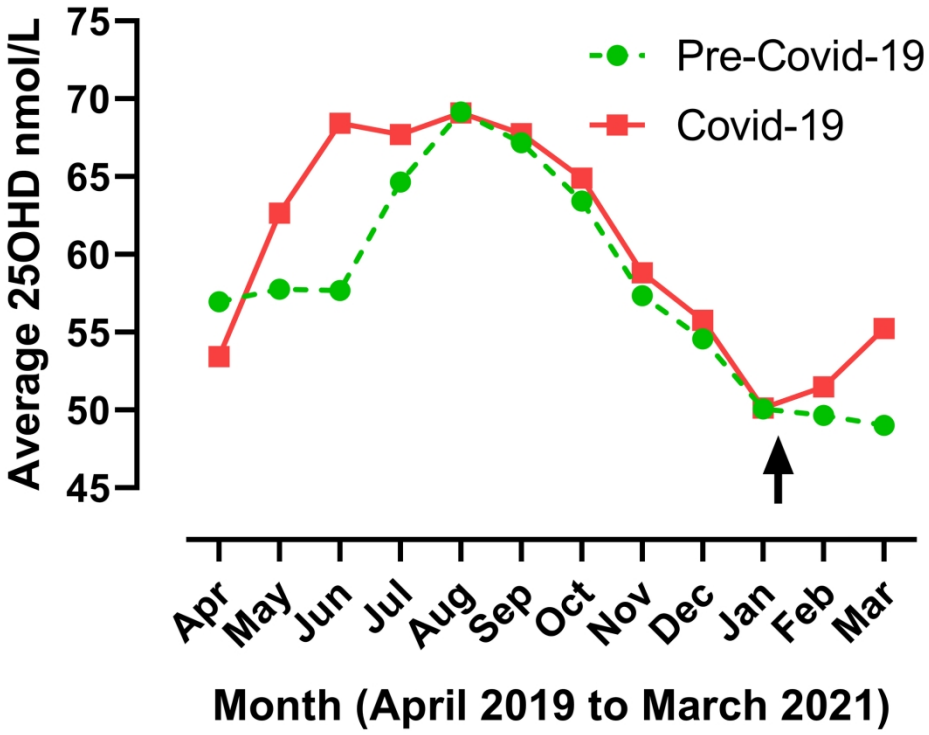

Seasonal variation in monthly average 25OHD over 1 year for both pre-Covid-19 group (green circles) and Covid-19 group (red squares). Yearly average 25OHD was higher in Covid-19 group compared to pre-Covid-19 group (p <.001). The time of highest infection rate and admission rate to intensive care was in early January 2021 (black arrow).

98x80mm (600 x 600 DPI)

STROBE Statement for **bmjopen-2021-059477**

	Item No	Recommendation
Title and abstract	1	(*a*) Indicate the study's design with a commonly used term in the title or the abstract
		(*b*) Provide in the abstract an informative and balanced summary of what was done and what was found. **Done**
Introduction		
Background/rationale	2	Explain the scientific background and rationale for the investigation being reported. **Done**
Objectives	3	State specific objectives, including any prespecified hypotheses. **Done**
Methods		
Study design	4	Present key elements of study design early in the paper **Done**
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection **Done**
Participants	6	(*a*) *Cohort study*—Give the eligibility criteria, and the sources and methods of selection of participants. Describe methods of follow-up **Consecutive laboratory-based samples formed the cohort, as described** *Case-control study*—Give the eligibility criteria, and the sources and methods of case ascertainment and control selection. Give the rationale for the choice of cases and controls *Cross-sectional study*—Give the eligibility criteria, and the sources and methods of selection of participants
		(*b*) *Cohort study*—For matched studies, give matching criteria and number of exposed and unexposed *Case-control study*—For matched studies, give matching criteria and the number of controls per case
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers. Give diagnostic criteria, if applicable **Done**
Data sources/ measurement	8*	For each variable of interest, give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group **Done**
Bias	9	Describe any efforts to address potential sources of bias. **Bias explained in Methods**
Study size	10	Explain how the study size was arrived at **Sample size was determined by number of laboratory samples during the 2-year time period.**
Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen and why. **Done**
Statistical methods	12	(*a*) Describe all statistical methods, including those used to control for confounding. **Done**
		(*b*) Describe any methods used to examine subgroups and interactions **Done**
		(*c*) Explain how missing data were addressed. **Not pertinent**
		(*d*) *Cohort study*—If applicable, explain how loss to follow-up was addressed **Not pertinent** *Case-control study*—If applicable, explain how matching of cases and controls was addressed

		Cross-sectional study—If applicable, describe analytical methods taking account of sampling strategy
		(*e*) Describe any sensitivity analyses **Not performed**
Results		
Participants	13*	(a) Report numbers of individuals at each stage of study—eg numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed. **Done**
		(b) Give reasons for non-participation at each stage **Not pertinent**
		(c) Consider use of a flow diagram **Not pertinent**
Descriptive data	14*	(a) Give characteristics of study participants (eg demographic, clinical, social) and information on exposures and potential confounders **Done**
		(b) Indicate number of participants with missing data for each variable of interest **Not pertinent**
		(c) *Cohort study*—Summarise follow-up time (eg, average and total amount) **Not pertinent**
Outcome data	15*	*Cohort study*—Report numbers of outcome events or summary measures over time **Done**
		Case-control study—Report numbers in each exposure category, or summary measures of exposure
		Cross-sectional study—Report numbers of outcome events or summary measures
Main results	16	(*a*) Give unadjusted estimates and, if applicable, confounder-adjusted estimates and their precision (eg, 95% confidence interval). Make clear which confounders were adjusted for and why they were included **Not pertinent**
		(*b*) Report category boundaries when continuous variables were categorized **Done**
		(*c*) If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period **Not pertinent**
Other analyses	17	Report other analyses done—eg analyses of subgroups and interactions, and sensitivity analyses **Done**
Discussion		
Key results	18	Summarise key results with reference to study objectives **Done**
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or imprecision. Discuss both direction and magnitude of any potential bias **Done**
Interpretation	20	Give a cautious overall interpretation of results considering objectives, limitations, multiplicity of analyses, results from similar studies, and other relevant evidence **Done**
Generalisability	21	Discuss the generalisability (external validity) of the study results **Done**
Other information		
Funding	22	Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based **No funding for study is stated**

BMJ Open

The COVID-19 pandemic and vitamin D: rising trends in status and in daily amounts of vitamin D provided by supplements

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The COVID-19 pandemic and vitamin D: rising trends in status and in daily amounts of vitamin D provided by supplements

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ABSTRACT

Objectives: Since the onset of the COVID-19 pandemic in 2020, there have been plausible suggestions about the need to augment vitamin D intake by supplementation in order to prevent SARS-Co-V2 infection and reduce mortality. Some groups have advocated supplementations for all adults, but governmental agencies have advocated targeted supplementation. We sought to explore the effect of COVID-19 pandemic on both vitamin D status and on the dose of new-to-market vitamin D supplements.

Setting: University hospital, Dublin, Ireland.

Participants: Laboratory-based samples of circulating 25-hydroxyvitamin D (25OHD) (*n* = 100,505)

Primary and secondary outcome measures: Primary outcomes: comparing yearly average 25OHD prior to the pandemic (April 2019 to March 2020) with during the pandemic (April 2020 to March 2021) and comparing the dose of new-to-market vitamin D supplements between 2017 and 2021. Secondary outcome: comparing prevalence of vitamin D deficiency and vitamin D excess during the two time periods.

Results: The average yearly serum 25OHD measurement increased by 2.8 nmol/L (61.4, 95%CI 61.5 – 61.7 vs 58.6, 95% CI 58.4-58.9, *p* <.001), which was almost 3-fold higher than two similar trend analyses that we conducted between 1993 and 2016. There was a lower prevalence of low 25OHD and a higher prevalence of high 25OHD. The dose of new-to-market vitamin D supplements was higher in the years 2020-2021 compared to the years 2017-2019 (*p* <.001).

Conclusions: We showed significant increases in serum 25OHD and in the dose of new-to-market vitamin D supplements. The frequency of low vitamin D status reduced indicating

benefit, but the frequency of vitamin D excess increased indicating risk of harm. Rather than a blanket recommendation about vitamin D supplementation for all adults, we recommend a targeted approach of supplementation within current governmental guidelines to at-risk groups and cautioning consumers about adverse effects of high dose supplements on the market.

Strengths and limitations

- This is a laboratory-based 25OHD trend analysis that includes a large sample size.
- This trend analysis is compared to with two prior trend analyses dating back to 1993.
- This is not a population-based representative sample and is subject to selection bias.
- There is no clinical information about reason for 25OHD testing or about vitamin D supplementation.

INTRODUCTION

Vitamin D is an essential micronutrient in all age groups for bone and muscle health[1-4]. Vitamin D may have a role in extraskeletal health such as the immune response to acute respiratory illnesses[5], which is pertinent during the COVID-19 pandemic[6]. Following the onset of the pandemic, some reports advocated blanket oral supplementation to entire populations with doses ranging from 20 µg to 50 µg daily[7-10], which are in excess of vitamin D intake requirement as specified by governmental agencies in Europe and North America[1-4]. By contrast, other groups have countered this blanket recommendation, favouring a targeted approach based on modelling of total vitamin D intakes[11, 12].

Governmental reports, which have been issued over the past decade prior to the pandemic, are based on similar health outcomes such as musculoskeletal health, falls and total risk of mortality, but not on immune response to infection[1, 3, 4]. Subsequently, governmental agencies from England and Ireland issued advice about vitamin D supplementation during the pandemic[13, 14]. This advice targeting at-risk populations included measures to facilitate supplementation[13, 14](Table 1) given the ongoing concerns about vitamin D inadequacy across Europe[15, 16].

Table 1 Vitamin D intake recommendations

Region	Year	Vitamin D Intake	Population Reference Intake µg/day (age group)
USA & Canada[1]	2011	Total intake	15 (<70 y) 20 (≥70 y)
Nordic Countries[2]	2014	Total intake	10 (<75 y) 20 (≥75 y)
European Union[3]	2016	Total intake	15 (≥18)
United Kingdom[4]	2016	Total intake	10 (all adults)
COVID-19 vitamin D advocates[7-10]	2020	Supplemental intake	20-50 (all adults, all year)
Post COVID-19 United Kingdom[13]	2020	Supplemental intake	10 (October to March for healthy adults) 10 (all year for those with limited sunlight exposure)
Post COVID-19 Ireland[14]	2020	Supplemental intake	10 (October to March >65 y) 15 (all year for housebound >65 y)

Total intake refers to vitamin D intake from all sources: skin production and oral intake (natural foods, fortified foods, supplements). Population reference intakes for governmental agencies refers to intake that meets the needs of 97.5% on the population; these intakes are based on health outcomes such as musculoskeletal health, falls and total risk of mortality,

Ensuring adequate vitamin D intake across all age groups is a challenging population health task. Vitamin D intake requirements must be modelled to cover total intake because vitamin D supply has various sources: skin production on exposure to ultra-violet light, natural foods, fortified foods, and supplements. There is almost complete unanimity about total vitamin D intake population requirements across governmental agencies for North America and for Europe[1-4]. In circumstances of minimal or no sunlight exposure, the total oral intake requirement varies between 10 µg and 20 µg daily (400 IU to 800 IU daily)[1-4]. The pre-eminent measure of vitamin D status is the measurement of the circulating vitamin D metabolite, 25-hydroxyvitamin D (25OHD).

We have been engaged in clinical research on 25OHD since the 1970s[17]. We have noted substantial improvement in vitamin D status over the past 5 decades. In our early studies in Ireland, we noted the primacy of oral intake over sunlight exposure for both the prevention and correction of vitamin D deficiency[18-21]. Following the advent of voluntary milk fortification in Ireland in the 1980s at 1 μg/100 ml (but more recently some milk products fortify at 2 μg/100 ml) and the ready availability of low-dose vitamin D supplements, vitamin D status has improved substantially[22]. In more recent years, we reported instances of hypervitaminosis D raising concerns about excessive oral intake of vitamin D[23].

We have published two trend analyses of laboratory-based 25OHD results: the first trend study reported 25OHD from 1993 to 2013 that incorporated a time series analysis to predict 25OHD trend from 2014 to 2016[24]; the second trend study reported 25OHD from 2014 to 2016 that confirmed the forecast analysis from the first study[25]. We reported that over 24 years in Ireland from 1993 to 2016 that the yearly average 25OHD concentration increased by about 1 nmol/L/year. As early as 2014, we recognized a dual concern about vitamin D status in Ireland: hypovitaminosis D in at-risk groups; and hypervitaminosis D due to high supplemental intake, especially from over-the-counter preparations in individuals who already have adequate vitamin D status[23]. One of the consequences of raising public awareness, whether it be from governmental agencies or from professional bodies, is the increased supply of vitamin D supplements, which are available for over-the-counter purchase.

We sought to explore the effect of COVID-19 pandemic on vitamin D status from April 2020 to March 2021 given the conflicting advice: governmental agencies promoting vitamin D supplementation in at-risk groups, and groups advocating blanket

recommendations for vitamin D supplementation to all adults. We examined vitamin D status before and during the first year of the pandemic when public debate and temporary emergency legislative measures (such as social distancing and mask wearing) were focused on how people could protect themselves against COVID-19. We also assessed the Food Safety Authority of Ireland (FSAI) notification database for new-to-market vitamin D supplements notified between 2017 and 2021 ; all food supplements placed on the Irish market must be notified to FSAI.[26]

METHODS

Data mining

Our laboratory data system was mined to identify all 25OHD samples over a 24-month period from April 2019 to March 2021: pre-COVID-19 era being represented by 12 months from April 2019 to March 2020, and the COVID-19 era being represented by April 2020 to March 2021. In order to identify duplicate samples, the following identifiers were extracted: date of birth and medical record number. Additional data included age, sex, referral source (either hospital consultant or general practitioner), and date of 25OHD test. The sample size for the 24 months from April 2019 to March 2021 was 137,963; after excluding those with more than one sample during the 2 years (*n* = 37,458) the final sample was trimmed to 100,505. Regarding those cases with more than one 25OHD sample, the analysis was based on the 25OHD being the first sample, which hereafter is termed the first in sequence. A secondary analysis was performed, whereby the selected 25OHD of those cases with more than one sample was the final sample, which hereafter is termed the last in sequence.

To enable closer monitoring of food supplements, Ireland took up the option within the EU Directive regulating food supplements of mandating food businesses placing food supplements on the Irish market to notify all details on the products to the FSAI. We mined

this FSAI notification database about new-to-market vitamin D supplements that were notified between January 2017 and December 2021 (*n* = 2688).

Research Ethics Approval

The Clinical Audit Committee, St Vincent's Healthcare Group, approved the extraction and audit of the data from our hospital laboratory system (reference number: 3174). Audits reviewed and approved in Ireland by an institutional clinical audit committee are neither subject to Research Ethics Committee approval nor require individual consent, as per Irish Health Research Regulations 2018.

Public and Patient Involvement

It was not appropriate or possible to involve patients or the public in the design, or conduct, or reporting, or dissemination plans of our research.

Study design

The primary analysis entailed a comparison of serum 25OHD concentration in the pre-COVID-19 era, hereafter referred to as group 1, with 25OHD in the COVID-19 era, hereafter referred to as group 2. The prevalence estimates for categories of 25OHD in the two groups were calculated according to the following thresholds: <30 nmol/L; 30-50 nmol/L; 50.1-125 nmol/L; and >125 nmol/L. In addition, a composite analysis of the entire group over the 2 years was performed in order to assess vitamin D status according to age, sex, and different age groups.

The list of vitamin D supplements that were notified to the FSAI between 2017 and 2020 was collated with respect to the total dose of vitamin D. Vitamin D supplements were categorised as high dose according to two different standards: firstly, if they exceeded the

tolerable upper intake level (UL) of 100 µg, which is defined as is the highest level of long-term daily intake of a nutrient, from all sources, judged to be unlikely to pose a risk of adverse health effects to humans[27]; and secondly, if they exceeded the maximum safe level (MSL) of 75 µg, which is defined as maximum amount of vitamin D that can safely be added to food supplements targeting teenagers and adults in Ireland. The MSL is calculated using a risk assessment approach: it is equal to the UL minus the estimated intake of vitamin D intake in the highest consumers (95th percentile of intake from both base diet and fortified foods)[28].

Analytical Methods

Serum 25OHD concentrations were quantified using the Elecsys Vitamin D Total (Roche Diagnostics GmbH, Mannheim, Germany) automated competitive binding protein assay, which measures total vitamin D, including isomers in the form of the C3 epimer as well as 24,25OHD metabolites. This is not specifically corrected for; rather, an assumption is made that there is a non-statistically significant difference in the percent concentration of vitamin D metabolites relative to the measured concentration in patient samples tested over the three-year period. The average inter-assay coefficients of variation (CV) for the 25OHD assay determined over the period studied were as follows: 14.6% at a mean concentration of 37.7 nmol/L, 8.7% at a mean concentration of 74.6 nmol/L, and 7.6% at a mean concentration of 112.1 nmol/L. Functional sensitivity was verified at 15 nmol/L (%CV <20%). To ensure a high standard of analysis for serum 25OHD concentrations, the laboratory participates in an external quality assurance scheme: the Vitamin D External Quality Assessment Scheme (DEQAS)[29]. During the 2-year period 2019 to 2021, our assay displayed a mean bias of 1.12 % from target values provided by the Centers for Disease Control and Prevention (Atlanta, Georgia, USA) using their isotope dilution LC-MS/MS Reference Measurement Procedure[30].

Assay performance also met DEQAS defined quality assurance performance criteria. For samples with undetectable 25OHD (<15 nmol/L), the result was censored at 14.9 pmol/L. For samples with high 25OHD (>175 mol/L), the result was censored at 175.1 nmol/L.

Statistical analysis

Descriptive statistics are presented as number and percent for categorical variables and as mean (95% confidence intervals) or median (interquartile range) for continuous variables. Differences in independent categorical variables were tested by chi-square. Differences between two means for 25OHD (both yearly and monthly) were tested by independent-samples *t* test using Levene's test for equality of variances. To account for multiple testing of monthly mean 25OHD, the Benjamini-Hochberg correction method was applied with a false discovery rate of 0.05; *p* values were converted to corresponding *q* values for the determination of true significance. A one-way analysis of variance (ANOVA) was conducted to explore the impact of year on dose of newly notified vitamin D supplements; post-hoc comparisons were made using Tukey HSD test. Statistics were considered significant if *p* value <.05. Analyses were performed using IBM SPSS Statistics version 25 (Armonk, NY, USA).

RESULTS

Group 1 and group 2 were similar with respect to age, sex, and source of referral (Table 1). Mean 25OHD was 2.8 nmol/L higher in group 2 compared to group 1 (61.4, 95%CI 61.5, 61.7 nmol/L vs 58.6, 95% CI 58.4, 58.9 nmol/L, *p* <.001) (Table 1). If the 25OHD duplicate result was selected as last in sequence, then mean 25OHD was 5.1 nmol/L higher in group 2 compared to group 1 (63.3, 95%CI 63.2, 63.6 nmol/L vs 58.2, 95% CI 58.0, 58.5 nmol/L, *p* <.001). In group 2 compared to group 1, there was a lower percent (12.0% vs 13.4%) of low

vitamin D status (25OHD <30 nmol/L) but a higher percent (2.1% vs 1.7%) of high vitamin D status (25OHD >125 nmol/L) (p <.001) (Table 2).

Table 2: Descriptive statistics

Variable	Group 1 (*n* = 58,642)	Group 2 (*n* = 41,863)
Age, years	52.5 (52.3, 52.7)	52.3 (52.1, 52.5)
Women : Men, %	66.4 : 33.6	64.3 : 35.7
Hospital : Primary Care, %	30.0 : 70.0	25.5 : 74.5
25OHD status, %		
<30 nmol/L	13.4	12.0
30-50 nmol/L	28.4	25.1
51-125 nmol/L	56.6	60.7
>125 nmol/L	1.7	2.1
25OHD, nmol/L	58.6 (58.4-58.9)	61.4 (61.5 – 61.7)

Results are presented as % for categorical variables and as mean (95%CI) for continuous variables

The monthly 25OHD trimmed values for both groups are plotted showing the seasonal variation (Figure 1). The average seasonal change in 25OHD from nadir to peak was almost identical for both at 20.2 nmol/L in group 1 and 20.1 nmol/L in group 2. Starting the monthly sequence in April 2020, the monthly mean 25OHD in group 2 compared to group 1 following Benjamini-Hochberg correction for multiple comparisons was significantly higher in the following months: May (q <.001), June (q <.001), July (q <.001), October (q =.012), November (q =.012), December (q =.012), February (q =.003), and March (q <.001) (Figure 1).

higher in May, June, July, October, November, December, February, and March (see results).

For the composite analysis, mean 25OHD was higher in women compared to men (61.3, 95%CI 61.1 – 61.5 nmol/L vs 56.9, 95% CI 56.7-57.3 nmol/L, *p* <.001), and in those sourced from primary care compared to hospital care (60.7, 95%CI 60.5 – 60.9 nmol/L vs 57.4, 95% CI 57.0-57.7 nmol/L, *p* <.001). Vitamin D status according to age categories showed that infants and toddlers had the lowest prevalence of 25OHD <30 nmol/L and the highest prevalence for 25OHD >125 nmol/L (Table 3). Regarding vitamin D status according to age categories and sex, adult females had better vitamin D status than males, but in infants a greater percent of females compared to males had both the lowest and the highest prevalence of vitamin D status, but the numbers were small (Table 3).

Table 3. Crosstabulation of vitamin D status according to age categories and sex

Age Categories		Vitamin D Status, nmol/L			
		<30	30 - 50	50.1 -125	>125
Infants	Female (*n*=87)	8.0	6.9	67.8	17.2
	Male (*n*=128)	4.7	10.2	76.6	8.6
	Total (*n*=215)	6.0	8.8	73.0	12.1
Toddlers	Female (*n*=250)	3.6	17.6	74.4	4.4
	Male (*n*=288)	6.3	15.6	73.3	4.8
	Total (*n*=538)	5.0	16.5	73.8	4.6
Children & adolescents	Female (*n*=3,253)	16.5	33.4	49.5	0.7
	Male (*n*=2,271)	13.7	30.8	54.3	1.2
	Total (*n*=5,524)	15.4	32.3	51.4	0.8
Young adults	Female (*n*=42,757)	12.0	28.4	57.8	1.9
	Male (*n*=20,533)	15.9	31.0	51.7	1.4
	Total (*n*=63,290)	13.3	29.2	55.8	1.7
Older adults	Female (*n*=19,493)	10.0	19.1	68.3	2.7
	Male (*n*=11,415)	14.1	26.5	57.7	1.6
	Total (*n*=30,908)	11.6	21.8	64.3	2.3
Total	Female (*n*=65,840)	11.6	25.8	60.5	2.1
	Male (*n*=34,635)	15.1	29.3	54.1	1.5
	Total (*n*=100,475)	12.8	27.0	58.3	1.9

Results are presented as %

Regarding the analysis of the FSAI notification database of new-to-market vitamin D supplements notified between 2017 and 2021, there was a significant difference in mean vitamin D doses over the 5 years ($p < .001$). Post-hoc comparisons showed no differences between 2017, 2018, and 2019, but higher in 2020 compared to the 2017, 2108, and 2019 (respectively, $p = .002$ $p = .021$ $p = .001$) and higher in 2021 compared to 2020 ($p < .002$) (Figure 2). Regarding the proportion of food supplement products notified that provide daily amounts of vitamin D exceeding the UL of 100 µg (1%, $n = 9$) and the MSL of 75 µg (3%, $n = 80$), the majority were notified during the COVID-19 pandemic (n =3 in 2017–2019 *vs*. n = 6 in 2020–2021 above the UL; $n = 18$ in 2017–2019 *vs*. $n = 62$ in 2020–2021 above the MSL) (Table 4).

Table 4

Year	Number of supplements notified	Mean amount of vitamin D (ug)	SD	IQR	Median	25th centile	75th centile	Min. amount of vitamin D (ug)	Max. amount of vitamin D (ug)	Number above UL (100 ug)	Number above MSL (75 ug)
2017	491	10.6	14.3	5.0	5.0	5.0	10.0	0.08	100	0	4 (0.8%)
2018	383	11.3	16.5	5.7	5.0	4.3	10.0	0.002	200	1 (0.3%)	3 (0.8%)
2019	442	11.1	18.1	6.8	5.0	3.2	10.0	0.63	125	2 (0.5%)	11 (2.5%)
2020	554	16.2	22.2	15.0	10.0	5.0	20.0	0.34	200	1 (0.2%)	17 (3.1%)
2021	819	21.3	34.5	20.0	10.0	5.0	25.0	0.13	500	5 (0.6%)	45 (5.5%)

UL = upper tolerable intake level: MSL = maximum safe level

DISCUSSION

In a trend analysis of laboratory-based 25OHD samples comparing yearly average 25OHD in the 12 months before onset of the COVID-19 pandemic (April 2019 to March 2020) with the first 12 months of the COVID-19 pandemic in Ireland (April 2020 to March 2021), we showed the average yearly 25OHD increased by 2.8 nmol/L/year. This yearly trend was nearly 3-fold higher than the average yearly increase in 25OHD of 1 nmol/L/year that we recorded in two prior trend analyses of laboratory-based samples from 1993 to 2016[24, 25]. If the 25OHD duplicate was selected as last in sequence for the trend analysis, then average 25OHD increase during the pandemic was even higher at 5.1 nmol/L/year. We observed benefit with respect to their being lower prevalence of 25OHD <30 nmol/L, but to a lesser extent there was a higher prevalence of 25OHD >125 nmol/L. Higher average monthly 25OHD was noted in nearly all months except at the end of summer, suggesting an increase in the prevalence of vitamin D supplementation. The dose of new-to-market vitamin D supplements increased significantly during the pandemic with an increase in the frequency of supplements exceeding the UL and MSL.

The 25OHD threshold for diagnosis of vitamin D deficiency depends on the approach: whether is it viewed as being population-based[1, 4] or as being case-based[31]. For a population-based approach, the Institute of Medicine (IOM) for the USA and Canada in 2011 used a statistical probability method to determine the prevalence of nutrient inadequacy[32]. The IOM set a 25OHD threshold of 30 nmol/L. IOM referred to 25OHD as a “biomarker of exposure” but not as a “biomarker of effect”, which means that 25OHD is the preeminent measure of total vitamin D intake, but it only estimates risk of disease. Thus, 25OHD below 30 nmol/L was defined by IOM as “risk of deficiency”. Similarly, the Scientific Advisory

Committee on Nutrition (SACN) for the UK set a 25OHD threshold of 25 nmol/L[4]. Governmental agencies set their specifications for vitamin D intake based on total vitamin D intake. Modelling intake from all sources estimates the shortfall in vitamin D intake that can be bridged by supplemental intake[33]. Whereas, a case-based approach, which is guiding clinicians about the need for supplemental vitamin D on an individual basis, sets higher 25OHD thresholds – for example the European Calcified Tissue Society set a 25OHD threshold at 50 nmol/L[31]. Case-based approaches tend to give guidance about 25OHD monitoring.

Governmental agencies have adopted a precautionary approach to vitamin D harm. The tolerable upper intake level (UL) from all oral sources of vitamin D (natural foods, fortified foods, and supplement) in those with minimal sunlight is 100 µg (4000 IU) daily [1, 34]. The UL is 10-fold higher than the average total vitamin D intake requirement for healthy adults[1]. The UL is not intended as a target intake; rather, the risk for harm begins to increase once vitamin D intake surpasses this level[35]. In Ireland, over-the-counter vitamin D products are regulated by the FSAI. The UL determines the maximum dose of a vitamin D product that can be marketed. Labelling requires that the dose not exceed the UL.

In addition to the UL, in Ireland the FSAI has also published guidance for food business operators regarding the MSL of vitamin D that can be added to food supplements in Ireland[28]. Since the UL encompasses daily oral vitamin D intake from all sources (base diet, fortified foods, and food supplements), then the maximum safe dose of a vitamin D supplement should be less than the UL. Following risk assessment approach, the FSAI deemed that the MSL for vitamin D in food supplements is 75 µg per day for teenagers and adults[28]. In our analysis of new-to-market vitamin D supplements, we noted that the frequency of supplements exceeding the MSL had increased from 0.8% to 6.1% between 2017 and 2021.

During the COVID-19 pandemic, some groups have advocated the need for mass vitamin D supplementation in order to enhance immune response to SARS-Co-V2 infection [7, 9, 10, 36, 37]. The European Food Safety Authority permits stating that benefit from vitamin D supplementation covers the normal functioning of the immune system without specifying a vitamin D intake for this benefit[38]. There is some evidence of benefit from vitamin D in randomised controlled trials (RCTs) that have been conducted during the pandemic but there is wide variation in vitamin D doses[39-44]. A Mendelian randomization study did not support an association between 25OHD and COVID-19 susceptibility, severity[45], or hospitalization; similar findings have been reported in a UK biobank study[46]. Further studies should provide clarity about benefit and optimal vitamin D schedules. These studies might provide cogent evidence for higher vitamin D intake requirements that could form part of population-based or case-based recommendations. Meanwhile, the likely effect of advocating for mass supplementation is that individuals, who are best able to self-medicate, are the ones who are least likely to need supplementation. Frail older adults, lower socioeconomic groups and minority ethnic groups are more likely to have lower 25OHD and are less likely to afford the means for supplementation[47, 48]. It is better to have a targeted approach to vitamin D supplementation such as the frail older adult[13, 14].

Mandatory fortification of foodstuffs with vitamin D, which has been shown to be effective in Finland[49], poses many challenges[50], but has the major advantage of reaching lower socio-economic groups excluded from the benefits of foods voluntarily fortified with vitamin D due to the significantly higher prices of such foods. Voluntary fortification, while less satisfactory than mandatory fortification, is effective at ameliorating seasonal decline in 25OHD, as has been shown in Ireland[22]. Fortification with any nutrient (whether mandatory or voluntary) in addition to supplementation (whether mandatory or voluntary), can result in

total nutrient intakes that are higher than requirement and may even exceed the UL, especially if nutrient intake targets the RDA and not the average requirement[51].

Our trend analysis has many limitations. First and foremost, it is not a representative sample because samples are not collected as part of a population-based survey. We do not have information on the clinical indication for the test nor do we know whether patients were on vitamin D supplements or had an underlying condition that predisposed to vitamin D deficiency. The plot of the seasonal variation of 25OHD is open to many interpretations, such as: more outdoor activity during the first lockdown accounting for the increased in 25OHD early in the pandemic from May 2020 to July 2020, coupled with higher supplemental intake during the winter months accounting for higher 25OHD from October 2020 to March 2021 (excepting January 2021). The 12-month trend analysis is too short to declare with any certainty that the COVID-19 pandemic has contributed to a shift upwards in the yearly average 25OHD increase or if it just a transient trend upwards due to the unique circumstances of living through legislatively enforced measures implemented globally to protect people from a pandemic while the search for solutions - such as the potential benefit of vitamin D - was the highest profile news story.

In conclusion, we report in Ireland a laboratory-based trend analysis of 25OHD showing that the yearly average 25OHD has increased about 3-fold during the first year of COVID-19 pandemic compared to prior trend analysis. This trend reflects benefit for those with low vitamin D status but risk for those with high vitamin D status, especially since there is a trend for greater availability of high dose supplements. Public health efforts should be redoubled at maximising the provision of specified daily vitamin D supplements in at-risk groups and clinically vulnerable patients and should advise about safe vitamin D supplement

use. There should be a precautionary approach to population-based blanket recommendations for vitamin D supplementation to healthy adults, as well as a caution to consumers about adverse effects of high dose vitamin D supplements on the market.

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Data described in the manuscript, code book, and analytic code will be made available upon request pending application to and approval by the corresponding author.

Research Ethics Approval Statement:

The Clinical Audit Committee, St Vincent's Healthcare Group, approved the extraction and audit of the data from our hospital laboratory system (reference number: 3174). Audits reviewed and approved in Ireland by an institutional clinical audit committee are neither subject to Research Ethics Committee approval nor require individual consent, as per Irish Health Research Regulations 2018.

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Legends to Figures:

Figure 1. Monthly mean 25OHD during COVID-19 pandemic was significantly higher than prior to the pandemic for the following months: May, June, July, October, November, December, February, and March (see results).

Figure 2. Yearly mean (95% confidence intervals) vitamin D supplement doses.

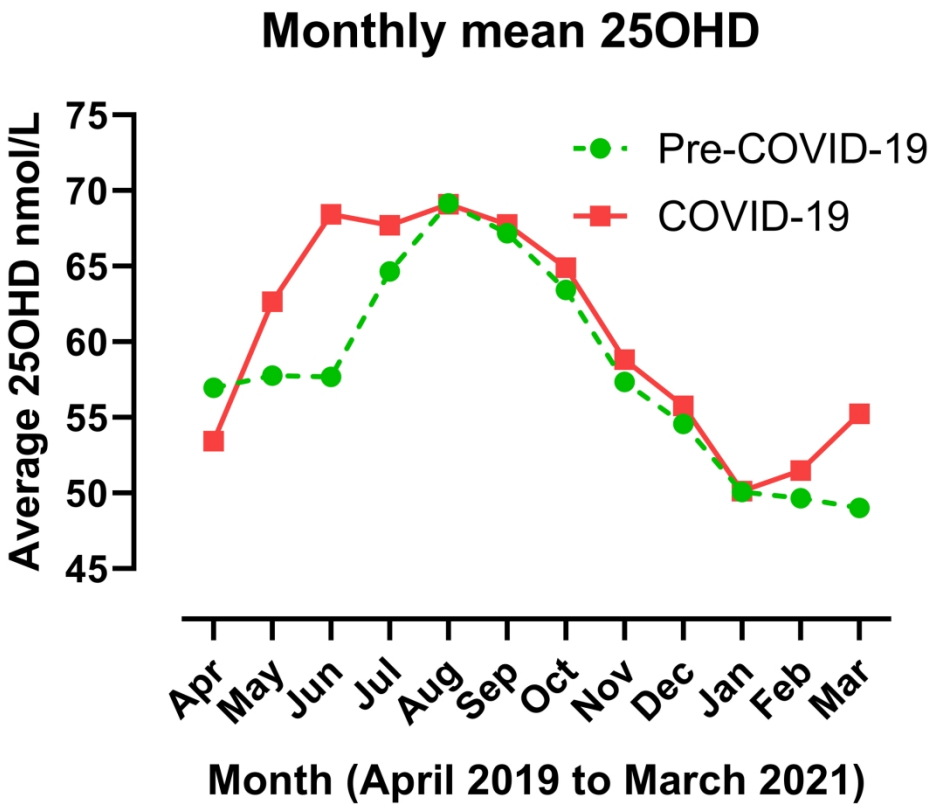

Figure 1. Monthly mean 25OHD during COVID-19 pandemic was significantly higher than prior to the pandemic for the following months: May, June, July, October, November, December, February, and March (see results).

100x87mm (600 x 600 DPI)

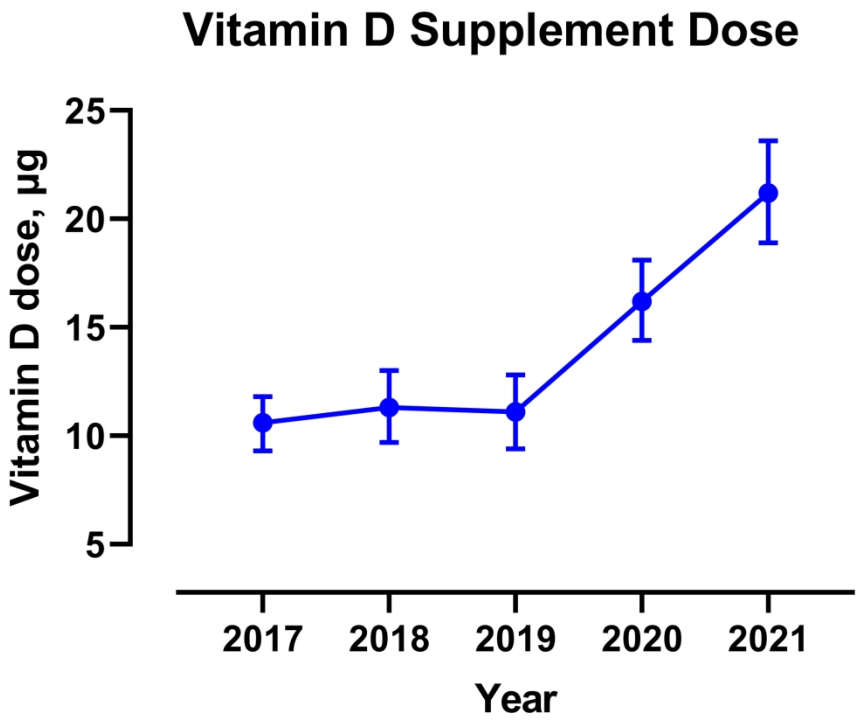

Figure 2. Yearly mean (95% confidence intervals) vitamin D supplement doses

105x82mm (600 x 600 DPI)

STROBE Statement for **bmjopen-2021-059477**

	Item No	Recommendation
Title and abstract	1	(*a*) Indicate the study's design with a commonly used term in the title or the abstract
		(*b*) Provide in the abstract an informative and balanced summary of what was done and what was found. **Done**
Introduction		
Background/rationale	2	Explain the scientific background and rationale for the investigation being reported. **Done**
Objectives	3	State specific objectives, including any prespecified hypotheses. **Done**
Methods		
Study design	4	Present key elements of study design early in the paper **Done**
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection **Done**
Participants	6	(*a*) *Cohort study*—Give the eligibility criteria, and the sources and methods of selection of participants. Describe methods of follow-up **Consecutive laboratory-based samples formed the cohort, as described** *Case-control study*—Give the eligibility criteria, and the sources and methods of case ascertainment and control selection. Give the rationale for the choice of cases and controls *Cross-sectional study*—Give the eligibility criteria, and the sources and methods of selection of participants
		(*b*) *Cohort study*—For matched studies, give matching criteria and number of exposed and unexposed *Case-control study*—For matched studies, give matching criteria and the number of controls per case
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers. Give diagnostic criteria, if applicable **Done**
Data sources/ measurement	8*	For each variable of interest, give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group **Done**
Bias	9	Describe any efforts to address potential sources of bias. **Bias explained in Methods**
Study size	10	Explain how the study size was arrived at **Sample size was determined by number of laboratory samples during the 2-year time period.**
Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen and why. **Done**
Statistical methods	12	(*a*) Describe all statistical methods, including those used to control for confounding. **Done**
		(*b*) Describe any methods used to examine subgroups and interactions **Done**
		(*c*) Explain how missing data were addressed. **Not pertinent**
		(*d*) *Cohort study*—If applicable, explain how loss to follow-up was addressed **Not pertinent** *Case-control study*—If applicable, explain how matching of cases and controls was addressed

		Cross-sectional study—If applicable, describe analytical methods taking account of sampling strategy
		(*e*) Describe any sensitivity analyses **Not performed**
Results		
Participants	13*	(a) Report numbers of individuals at each stage of study—eg numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed. **Done**
		(b) Give reasons for non-participation at each stage **Not pertinent**
		(c) Consider use of a flow diagram **Not pertinent**
Descriptive data	14*	(a) Give characteristics of study participants (eg demographic, clinical, social) and information on exposures and potential confounders **Done**
		(b) Indicate number of participants with missing data for each variable of interest **Not pertinent**
		(c) *Cohort study*—Summarise follow-up time (eg, average and total amount) **Not pertinent**
Outcome data	15*	*Cohort study*—Report numbers of outcome events or summary measures over time **Done**
		Case-control study—Report numbers in each exposure category, or summary measures of exposure
		Cross-sectional study—Report numbers of outcome events or summary measures
Main results	16	(*a*) Give unadjusted estimates and, if applicable, confounder-adjusted estimates and their precision (eg, 95% confidence interval). Make clear which confounders were adjusted for and why they were included **Not pertinent**
		(*b*) Report category boundaries when continuous variables were categorized **Done**
		(*c*) If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period **Not pertinent**
Other analyses	17	Report other analyses done—eg analyses of subgroups and interactions, and sensitivity analyses **Done**
Discussion		
Key results	18	Summarise key results with reference to study objectives **Done**
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or imprecision. Discuss both direction and magnitude of any potential bias **Done**
Interpretation	20	Give a cautious overall interpretation of results considering objectives, limitations, multiplicity of analyses, results from similar studies, and other relevant evidence **Done**
Generalisability	21	Discuss the generalisability (external validity) of the study results **Done**
Other information		
Funding	22	Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based **No funding for study is stated**