Efficacy of various prescribed vitamin D supplementation regimens on 25-hydroxyvitamin D serum levels in long-term care

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This is an Accepted Manuscript for Public Health Nutrition as part of the Cambridge Coronavirus Collection. This peer-reviewed article has been accepted for publication but not yet copyedited or typeset, and so may be subject to change during the production process. The article is considered published and may be cited using its DOI 10.1017/S1368980021001609

Public Health Nutrition is published by Cambridge University Press on behalf of The Nutrition Society
Short title: Efficacy of vitamin D supplementation regimens

Acknowledgements: The authors would like to thank Brian Haward with Significant Difference LLC for his statistical support and guidance.

Financial Support: This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

Conflict of Interest: Authors have no conflict of interest to report.

Authorship: All authors have made substantial contributions to all of the following: (1) the conception and design of the study, or acquisition of data, or analysis and interpretation of data, (2) drafting the article or revising it critically for important intellectual content, (3) final approval of the version to be submitted.

Ronna Robbins: conceptualization, methodology, data curation, formal analysis, supervision, writing-original draft
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Ethical Standards Disclosure: This study was conducted according to the guidelines laid down in the Declaration of Helsinki and all procedures involving research study participants were approved by the Institutional Review Board, University of Texas at Austin (2018-03-0030). Written and Verbal informed consent was obtained from all subjects/patients or legal proxies. Verbal consent was witnessed and formally recorded.
ABSTRACT

Objective: The aims of this study were to examine the efficacy among various vitamin D supplementation regimens on serum 25(OH)D concentrations and determine the minimal dose rate required to achieve sufficient serum concentrations (≥75 nmol/L) among older adults in long-term care (LTC).

Design: A one-year medical history was abstracted from medical records, and a one-time blood draw to measure serum 25(OH)D concentrations was obtained. Individuals were stratified into vitamin D supplemented and non-supplemented groups. The supplemented group was further categorized into four treatment forms: single ingredient vitamin D$_{2or3}$, multivitamin, calcium with vitamin D, or combination of the three, and by daily prescribed doses: 0-399, 400-799, 800-1999, 2000-3999, and >4000 IU/day.

Setting: Five LTC communities in Austin, Texas.

Participants: 173 older (≥65 years) adults.

Results: Of the participants, 62% received a vitamin D supplement and 55% had insufficient (≤75 nmol/L) 25(OH)D serum concentrations. Individuals receiving single ingredient vitamin D$_{2or3}$ supplementation received the highest daily vitamin D mean dose (2900 IU/d), while combination of forms was the most frequent treatment (44%) with the highest mean serum concentration (108 nmol/L). All supplementation doses were successful at reaching sufficient serum concentrations, except those <800 IU/d. Using a prediction model, it was observed that one IU/d of vitamin D supplementation resulted in a 0.008 nmol/L increase in serum 25(OH)D concentrations.

Conclusion: Based on the predictive equation, results suggest that supplementation of 1500 IU/d of vitamin D$_{2or3}$ or combination of vitamin D is most likely to achieve sufficient serum 25(OH)D concentrations in older adults in LTC.

KEY WORDS

Vitamin D, supplementation, 25-hydroxyvitamin D, long-term care, older adults, skilled-nursing
Introduction

The classical function of vitamin D in calcium and phosphate homeostasis and bone metabolism has long been recognized. Over the past decades, a more expansive role of vitamin D in non-skeletal physiological processes has emerged \(^{(1,2)}\). Research shows that insufficient serum vitamin D concentrations are associated with an increased rate of respiratory tract infection, influenza, and other infectious diseases, along with several chronic health conditions, including dementia, depression, cardiovascular disease, and cancer \(^{(3-7)}\). As the world deals with the health and economic burden of the COVID-19 pandemic, insufficient vitamin D concentrations have emerged as a possible risk factor for SARS-CoV-1 infection, the virus that causes COVID-19. As a result, the interest in vitamin D supplementation to reduce the rate of infection, lessen severe illness, and or accelerate recovery has surfaced \(^{(5-7)}\).

Older adults, especially those in long-term care (LTC) experience high rate of vitamin D insufficiency (40-100\%) due to inadequate sunlight exposure, medication interactions, and limited dietary sources \(^{(1,4,6,8,9)}\). LTC older adults are also among the most vulnerable populations for SARS-CoV-1 outbreaks and at the greatest risk for severe illness and morality from COVID-19 \(^{(10,11)}\). Correcting insufficient vitamin D concentrations in these individuals is critical secondary to vitamin D’s role in regulating innate and adaptative immunity, and potential to reduce the risk of viral infection, progression, and severity \(^{(5,6)}\). Since LTC residents are often exposed to limited sunlight exposure, oral nutrition becomes an essential route for intake of vitamin D; hence, supplementation is recommended to maintain optimal serum 25-hydroxyvitamin D (25(OH)D) concentrations \(^{(12,13)}\).
The National Academy of Medicine (NAM), formerly known as Institute of Medicine (IOM), set the dietary reference intake (DRI) for vitamin D in older adults (70+ years) at 800 IU/d, which is sufficient to reach serum 25(OH)D concentration of ≥50 nmol/L (20 ng/mL) and maintain bone health (12-15). However, a growing body of evidence suggests that 800 IU/d will not raise serum 25(OH)D concentrations above the Endocrine’s Society concentration recommended to achieve sufficiency (75 nmol/L (30 ng/mL)) (12). Thus, the DRI may not be protective of non-skeletal health conditions including COVID-19 (6,12,16). Even with general agreement among health organizations and experts for universal vitamin D supplementation in older adults (≥65 years), the recommended supplementation dose rate and 25(OH)D target serum concentrations remain controversial (12,16,17). Health organizations and experts suggest a variety of supplementation dose rates ranging from 1000 to 4000 IU/d to achieve sufficient serum 25(OH)D concentrations (18-21).

Despite the known health consequences, environmental and physiological risk factors, and recommendations from experts for universal blood screening and supplementation, vitamin D insufficiency is routinely not diagnosed and/or undertreated within the LTC population (1,8,9,16,17,22). Several factors contribute to poor testing and treatment, including limited coverage of 25(OH)D blood test by Medicaid/Medicare and lack of systematic supplementation by practitioners in LTC, resulting from a perception that supplementation and/or correcting for insufficiency is not considered a health priority (17,23-25).

The objectives of this study were to determine the prevalence of vitamin D supplementation and examine the efficacy among various vitamin D supplementation regimens on serum 25-hydroxyvitamin D concentration in older adults living in a LTC community. Additionally, we aimed to determine the minimal vitamin D supplementation dose rate required to achieve
sufficient serum 25(OH)D concentration (≥75 nmol/L) in these LTC patients.

Methods

Participants

For this cross-sectional study, older adults from five LTC communities in the metropolitan area of Austin, Texas, were recruited to participate. To be eligible, participants had to be ≥65 years, and reside within skilled nursing or assisted living units. There were no exclusion criteria for 25(OH)D serum concentrations or vitamin-mineral supplementation. Written consent was obtained from medical power of attorney for all participants, along with verbal assent from individuals without cognitive impairment (assessed by each patient’s nursing staff or Social Worker).

Data Collection

A one-year medical history was collected from on-site electronic medical records (EMR) using double-blinded protocols (26,27). Data abstracted from EMR included demographics (age, sex, race, level of care), lifestyle factors (alcohol and tobacco use), and medical history (weight, height, diagnoses, medications, number of infections, falls, hospitalizations). Race was categorized as Caucasian or non-Caucasian based on the U.S Census Bureau’s 2013-2017 American Community Survey (28). Race was included as a covariate to account for any potential difference in serum concentrations secondary to skin pigmentation and UV-B (sun) exposure.

Height and weight from the EMR were used to calculate body mass index (BMI) as kg/m² and then categorized into the Center for Disease Control Adult standard weight status categories: underweight: <18.5 kg/m²; healthy weight: 18.5 to 24.9 kg/m²; overweight: 25.0 to 29.9 kg/m²; and obese: ≥30.0 kg/m² (29). The dosage, start, and or discontinue date for all medications were
collected and then categorized according to the Food and Drug Administration’s U.S. Pharmacopeia Therapeutic Category and Pharmacologic Classification Guidelines (e.g., antidepressants, diuretics, bisphosphates)\(^{(30)}\). Medications that inhibit or induce cytochrome P450 25-hydroxylase (CYP3A4) enzyme activity, which is responsible for converting ergo- and cholecalciferol (dietary sources of vitamin D) to the circulating metabolite 25(OH)D, were further categorized as having a drug-vitamin D interaction and used as a covariate in statistical analyses \(^{(31)}\).

Vitamin-mineral supplementation along with dosage, start and or discontinue date were also collected from the medical record. Vitamin D supplementation was defined as a supplement containing \(\geq 200\) IU/d of vitamin D\(_2\) or D\(_3\), which included single-ingredient vitamin D supplement, calcium with vitamin D, and multivitamins. To determine the specific amount of vitamin D provided by multivitamins, each LTC community provided their house multivitamin formulary.

**Nutritional Analysis**

Each community provided their spring/summer 2018 cycle menus along with the corresponding nutritional analyses and serving sizes. The nutritional analyses did not include vitamin D, so a trained LTC-Registered Dietitian Nutritionist used the USDA Nutrient Composition Database to calculate the estimated daily average of vitamin D (IU) provided in meals \(^{(32)}\). Nutritional analyses were conducted using generic recipes and community specific serving sizes. To ensure the calculated analysis was credible, the macro and micro-nutrients of the calculated analyses were compared to the nutritional analysis provide by each LTC community. The amount of
vitamin D provided from oral nutritional supplementation (i.e., calorie/protein shakes) was documented and included in the daily vitamin D meal total for each participant.

**Measurements/Serum Analysis**

Despite the expected time spent outdoors and accompanying sun exposure to be minimal for LTC patients, fasting venous blood draws were obtained from all participants during the summer of 2018, when serum 25(OH)D concentrations were predicted to be the highest (average high temperature of 35.1°C (95.2°F) in Austin, Texas) (12,33,34). The Endocrine Society clinical practice guidelines were used to define serum 25(OH)D concentrations as sufficient (≥75 nmol/L) and insufficient (<75 nmol/L) (12). To ensure each LTC community maintained regulatory compliance with the Texas Department of Health and Human Services and the Center for Medicare and Medicaid Services (CMS), a mobile diagnostic laboratory and phlebotomy company that was both College of American Pathologists Accredited and CLIA-88 certified (Clinical Laboratory Improvement Amendments) was contracted to collect blood samples. All test procedures followed the Clinical Laboratory Standards Institute (CLSI) Evaluation Protocols. An Access 25(OH)D vitamin D Total chemiluminescent immunoassay was used to measure serum 25(OH)D concentrations using the Access2 Immunoassay System (Beckman Coulter, Brea, CA, USA). The Beckman Coulter Access 25(OH)D vitamin D Total assay is standardized and traceable to the gold standard 25(OH)D vitamin D Reference Measurement Procedure (RMP) from Ghent University (Ghent, Belgium) (35). Using Passing-Bablok regression and Spearman correlation, a measurement procedure comparison evaluated serum samples (n=110) with Access 25(OH)D vitamin D Total assay (ng/mL) on the Access2 System and an isotope-dilution-liquid chromatography tandem-mass spectrometry (ID-LC-MS/MS) 25(OH)D vitamin D (RMP by
Ghent University) and produced the following results: r-value 0.95 [intercept-2.87, CI, -5.44 to -0.88, slope 1.01 (0.94 to 1.10)]\(^{(35)}\). Beckman Coulter reports the Access 25(OH)D vitamin D assay to have a total coefficient of variation (CV) \(<10\%\), which meets the vitamin D Standardization Program criteria\(^{(35)}\). Supporting this claim is an independent 2017 study by Madenci et al., which evaluated the 25(OH)D vitamin D Total assay (ng/mL) on the Access2 System analytical performance and found a CV% of 8.1 and 7.7% for low and high concentrations, respectively\(^{(36)}\). Per manufactures specification and lab policy, the Access 25(OH) vitamin D Total assay undergoes quantitative assay calibration every 28 days using assay calibrators, which are traceable to the Joint Committee for Traceability in Laboratory Medicine (JCTLM)-approved ID-LC-MS/MS and RMP developed at Ghent University. The calibrator’s RMP is further traceable to the National Institute of Standards and Technology (NIST) standard reference material (SRM) 2972\(^{(33)}\). To validate the accuracy of analysis, all critical concentrations were automatically re-analyzed, a delta check was conducted on all specimens, and a significant deviation (p<0.01) prompted a re-test.

Individuals were stratified into groups based on whether they received vitamin D supplementation or were non-supplemented. The vitamin D supplemented group was further categorized based on the following treatment forms: single ingredient vitamin D (vitamin D\(_{2/3}\) ), calcium with vitamin D, multivitamin, and combination of the three treatments. The total study population was categorized by supplementation dose rate prescribed per day: 0-399, 400-799, 800-1999, 2000-3999, and >4000 IU/day. The 0-399 IU/d supplemented group included those not receiving any supplementation. The dose range of 400-799 IU/d was determined based on multiple studies that showed supplementation less than 800 IU/d does not raise serum 25(OH)D
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concentrations above 75 nmol/L \(^{(20-22,37)}\). The dose range of 800-1999 IU/d was determined by the DRI recommendation of 800 IU/d and several randomized control trials that did not detect serum differences in supplementation dose rates between 800 and 2000 IU/d \(^{(15,21,38-40)}\). The dose range of 2000-3999 IU/d was determined based on limited prior data available on serum concentration effects for these doses. Finally, the dose range >4000 IU/d was selected based on randomized control trials showing supplementation of 4000 IU/d or higher resulted in serum concentrations ≥75 nmol/L \(^{(12,21,41)}\).

Statistical Analysis

Statistical analyses were performed using Stata v16 (StataCorp, College Station, Texas). Descriptive analyses summarized population characteristics; 25(OH)D serum concentrations; supplementation treatment types and dose rates; and the prevalence of vitamin D sufficiency (≥75 nmol/L) and insufficiency (<75 nmol/L). Continuous variables are presented as mean ± standard error of the mean (SEM) and as percentages for categorical variables. Depending on variable t-test, \(X^2\), or Analysis of variance (ANOVA) determined mean differences in serum 25(OH)D concentrations. Factorial analysis of covariance (ANCOVA) compared serum 25(OH)D concentrations across different dietary supplementation treatment types and dose ranges. Covariates in ANCOVA included: vitamin D provided in meals (IU/d), years living in the LTC community, age, race, sex, BMI, diagnosis of renal and liver disease, and prescribed medications with a drug-vitamin D interaction. Bonferroni type adjustment corrected for multiple comparisons with an adjusted significant p-value of \(p ≤ 0.005\). Linear Regression determined the minimal dose of vitamin D supplementation required to achieve sufficient serum concentrations (≥75 nmol/L). Multiple logistic regression was used to determine significant predictors of
sufficient serum 25(OH)D concentrations among the various treatment regimens within supplementation dose rates and treatment forms while controlling for the above covariates. P-values of \(<0.05\) were set as the threshold for statistical significance for the ANCOVA models.

**RESULTS**

A total of 180 participants were recruited; however, four refused blood draws, and three died during the study period. Therefore, analyses were performed on the 173 participants with complete data. Participants were older (age: 83 ± 0.82 years) and the majority were Caucasian (89%), women (61%), and overweight (BMI: 26 ± 0.43 kg/m²). Thirty-eight% were not prescribed a vitamin D supplement, and 55% of participants had insufficient 25(OH)D (<75 nmol/L). Participants were prescribed an average of 11 (range 1-22) medications per day (including on average 2 vitamin-mineral supplements), with 52% receiving at least one medication with a drug- vitamin D interaction. The calculated estimated daily average intake of vitamin D provided in meals (which included fortified foods) was 200 IU/d. As seen in Table 1, the mean serum dose rate of vitamin D supplementation and 25(OH)D concentrations did not differ between demographic characteristic groups (sex, race, and level of care).

*Mean Comparisons between different supplementation treatment forms*

Only four individuals received vitamin D₂ alone, so vitamin D₂ and D₃ were combined into one treatment category (vitamin D₂or3). As seen in Table 1, the most commonly provided vitamin D supplement was a combination (44%), with the least being coming from a calcium + vitamin D supplement (16%). The mean dose rate of those supplemented was 2030 ± 227 IU/d (range 200-15000 IU/d), with the highest dose provided by the vitamin D₂or3, followed by the combination
treatment forms. Mean serum 25(OH)D concentration was 94 ± 4 nmol/L. As expected, when compared to non-supplemented individuals, those supplemented had a higher vitamin D intake (2030 vs. 8 IU/d, p<0.001) and serum 25(OH)D concentrations (94 ± 4 vs. 60 ± 3 nmol/L, p<0.001), on average. Insufficient serum concentrations were observed in 42% of those supplemented compared to 81% non-supplemented (p<0.001).

Table 1 also demonstrates that mean supplementation dose rates, serum 25(OH)D concentrations, and prevalence of insufficient serum concentrations differed significantly across all four forms of vitamin D supplementation treatment types (all p<0.001), with those receiving vitamin D from a D2or3 or combination sources achieving the highest serum concentrations. Serum 25(OH)D also significantly increased as supplementation dose increased (all p’s<0.001). As expected, a higher prevalence of serum vitamin D insufficiency was observed in those in the 0-399 IU/d group compared to the >4000 IU/d group (79 vs. 19%; p<0.001).

Table 2 shows pairwise comparisons of serum 25(OH)D concentrations across the different supplementation treatment forms. It was observed that compared to the non-supplemented group, those in the vitamin D2or3 and combination groups had approximately 30-40 nmol/L higher serum 25(OH)D concentrations (p’s<0.001). Further, consuming a combination of supplements also results in approximately 30 nmol/L higher serum 25(OH)D concentrations than that observed in the multivitamin group (p=0.003). Table 2 also shows pairwise comparisons of mean serum 25(OH)D concentrations across the different supplementation dose ranges. In general, it was observed that serum concentration significantly increased as supplementation dose increased.

Linear Regression was performed to determine the predicted vitamin D supplementation dose
rate required to achieved sufficient serum 25(OH)D concentration. The model (R²=0.31 (CI: 0.03 to 0.005), p<0.001) explained 36% of the variability between serum concentrations and supplementation dose rate. The equation predicting minimal supplementation dose rate required to achieved sufficient serum 25(OH)D concentrations of ≥75 nmol/L is:

$$25(\text{OH})\text{D Serum Concentrations} = 26.11 + 0.008 \text{ (IU/d vitamin D)}$$

Thus, one IU/d of vitamin D supplementation resulted in a 0.008 nmol/L increase in serum 25(OH)D concentrations. Using this predictive equation, a supplementation dose rate of 1500 IU/d would be required to achieve sufficient serum 25(OH)D concentration of 75 nmol/L.

Of the treatment forms only vitamin D₂or₃ (odds ratio (OR): 9.0 (CI 3.1 to 25.7), p<0.001), and combination of supplementation treatment forms (OR 6.8 (CI: 2.7 to 17), p<0.001) significantly increased the odds of having sufficient serum 25(OH)D concentration, whereas calcium with vitamin D and multivitamins were not significant determinates when compared to the non-supplemented group. Further, compared to the 0-399 IU/d group, all supplementation dose range categories significantly increased the odds of having sufficient serum concentrations 800-1999 IU/d: (OR 4.2 (CI 1.6 to 10.6), p<0.03); 2000-3999 IU/d: (OR 10.8 (CI 4.0, 30.6), p<0.001); >4000 IU/d: (OR 14.7 (CI 3.2 to 68.5), p<0.001), except for the 400-799 IU/d group (OR 2.4 (CI 0.9 to 6.3), p=0.11).

**Discussion**

With vitamin D’s role in the regulation of immunity, along with its potential to reduce the risk of viral infections, correcting insufficient serum 25(OH)D concentrations in older adults, especially those living in LTC communities, has never been more important than during the COVID-19
Our study adds to the LTC literature because it suggests that supplementation with vitamin D₂ or D₃ alone or a combination of therapies (including vitamin D, calcium with vitamin D, and/or a multivitamin) may lead to a better ability to raise serum 25(OH)D to sufficient concentrations compared to a multivitamin or calcium with vitamin D supplement alone. Further, it also suggests that at least 1500 IU/d of vitamin D is needed to maintain or treat insufficient serum 25(OH)D concentrations in older adults living in LTC.

The majority of national and international health organizations agree that individuals living in LTC communities should receive a vitamin D supplement; however, recommendations on the dose rate required and target serum 25(OH)D concentration remains controversial (12,13,42). The wide variability in 25(OH)D assays has been cited as a limiting factor preventing consensus of serum cut-off points (43). This study adds to the literature because it used 25(OH)D assays that were standardized and traceable to the gold standard 25(OH) vitamin D Reference Measurement Procedure (RMP) from Ghent University (Ghent, Belgium) (35). Further, we analyzed these results in light of current Endocrine Society cutoff values for sufficiency/insufficiency; however, serum 25(OH)D concentration cut-off points used to define deficient, insufficient, and sufficient remains controversial and highly debated among leading health organizations, specifically between The Endocrine Society’s 2012 and NAM’s 2011 guidelines (43). It should be noted that the target audience on which the guidelines are based is a contributing factor to the controversy. The Endocrine Society’s guidelines are designed for clinical practice, while the NAM’s are designed for overall public health targeted at healthy non-diseased populations (43).

Our results suggest that less than half of LTC residents had sufficient serum 25(OH)D
concentration of ≥75 nmol/L. These results are similar to a study conducted in Pittsburgh, Pennsylvania showing that 52% of LTC residents have sufficient serum concentrations (16). The number of LTC residents who received at least 200 IU/d of vitamin D supplementation in the current study (62%) was higher than expected based upon prior evidence suggesting ranges of 10-50%; however, 42% of those supplemented failed to reach sufficient serum 25(OH)D concentrations (17,24,44). These data indicate that despite the higher-than-expected prevalence of supplementation, the prescribed supplementation dose rates were possibly too low, not treated for a long enough duration, or serum 25(OH)D concentrations were very low at the start of supplementation. These results are supported by multiple studies, including a 2015 randomized control trial of vitamin D deficient LTC residents that showed supplementation of 800 IU/d took at least 12 weeks of supplementation for serum 25(OH)D concentrations to reach a steady-state, and 60% of those never achieve the target serum 25(OH)D concentration of ≥75 nmol/L (16,22,37). In the current study all supplementation dose ranges were successful at reaching the target serum 25(OH)D concentrations of 75 nmol/L, expect dose ranges of <800 IU/d in at least some residents.

As expected, this study showed that as supplementation dose rate increases, serum 25(OH)D concentration also increase. Based on our predictive regression equation, we find that serum 25(OH)D concentrations increases approximately 8.0 nmol/L (3.0 ng/mL) for every 1000 IU of vitamin D. A meta-analysis of 16 randomized control trials of free-living adults by Black et al. also developed a predictive equation and determined serum 25(OH)D concentrations increased 1.2 nmol/L for every 1 μg (40 IU) of ingested fortified foods (45). Black et al. equation predicts a greater increase in serum concentration per IU supplemented than our study; however, the author
acknowledges that the results should be interpreted with caution due to a high level of heterogeneity across the studies (i.e., environmental and methodology variability, population and age differences, range of assays used to measure 25(OH)D, daily doses of vitamin D, and food sources). On the other hand, a study with a similar design as ours by Singh et al. concluded that a 5.0 nmol/L (2.0 ng/mL) increase in serum 25(OH)D concentrations occurred for every 1000 IU of vitamin D supplemented in older adults in LTC communities (19). Nevertheless, our study’s predicted rise in serum concentration of 8 nmol/L for every 1000 IU of vitamin D shows that more vitamin D supplementation is required in the LTC population and is a concern even if targeted serum concentrations were <75 nmol/L.

Using the regression equation developed in our study, the findings suggest 1500 IU/d of vitamin D is needed to achieve sufficient serum 25(OH)D concentrations. Several studies have also found similar results, including a study by Bischoff-Ferrari et al. that determined a supplementation dose rate >1000 IU/d is required for older adults in LTC to achieve serum 25(OH)D concentration of ≥75 nmol/L (20). Further, Kotlarczy et al. concluded that supplementation of 800 IU/d in LTC residents brought serum 25(OH)D concentrations to the NAM serum recommendation of ≥50 nmol/L, but these rates were not able to reach ≥75 nmol/L (16). Thus, our results support the current recommendations set by the U.S. Endocrine Society Clinical Practice Guidelines that 1500-2000 IU/d of vitamin D is needed to reach and maintain 25(OH)D ≥75 nmol/L (12).

Our results should be interpreted in light of several study strengths and limitations. With regard to strengths, we were able to identify and control for medications metabolized by the cytochrome
P450 25-hydroxylase (CYP3A4) enzyme, which has the potential to affect vitamin D status or alter supplementation effectiveness. In this study 52% of participants received at least one medication with a drug-vitamin D interaction. Further, we were able to estimate daily vitamin D content provided in meals. Suominen et al. similarly found the nutrient content of vitamin D served to residents in LTC to be 200 IU/d with an estimated consumption of <100 IU/d \(^\text{46}\). Several study limitations also exist. Due to limitations of the retrospective study design and because of our desire to understand current LTC practices, we were unable to standardize food intake and physical activity (particularly outdoor activity reflective of direct sun exposure). However, it should be noted that it is a common practice by dietary supplement manufacturers to add ingredients greater than the label-claim (overage amount) to account for shelf life and losses during processing. These overages were not estimated in this study. With vitamin D overage amounts estimated to be up to ~40\% \(^\text{47}\), this could have resulted in an underestimate of vitamin D intake from supplementation and its effect on serum concentrations. Further, due to the available information in the medical record, we also were unable to determine total supplement duration and utilization of loading doses. Loading doses are often prescribed to quicken return to a steady state, which often is not reached until three to four months of supplementation \(^\text{21,48,49}\). Compliance with supplementation was also not available; however, individuals living in LTC are reported to be 96\% compliant when taking medications \(^\text{50}\). Nevertheless, because this study examines commonly used supplementation patterns in LTC residents, it does have high generalizability.

**Conclusion**

Our results add to the growing need for health organizations to agree on serum cut-off
concentrations used to define vitamin D status and provided clear clinical practice guidelines on dose rates and target serum 25(OH)D concentration for practitioners. We find that the current DRI of 800 IU/d may be too low to reach and maintain sufficient serum concentrations in older LTC residents. Our results support the U.S. Endocrine Society Clinical Practice Guidelines, which recommends 1500-2000 IU/d to maintain serum 25 (OH)D concentrations above 75 nmol/L, but adds to this by suggesting that supplementation with either vitamin D$_{2}$ or D$_{3}$ alone or a combination vitamin D supplementation may be needed to achieve sufficient serum 25(OH)D concentration in patients living in LTC communities. Until a consensus is reached, individuals in LTC may continue to receive supplementation dosages that are ineffective at reaching sufficient serum 25(OH)D concentrations (>75 nmol/L), thus are potentially increasing the risk for adverse outcomes associated with non-skeletal health conditions. These results have immediate implication and can be used by practitioners (physicians, dietitians, and nurses) to maintain or achieve sufficient serum 25(OH)D concentrations in LTC older adults and potentially reduce COVID-19 infection rates, progression, and disease severity.
Reference:

1. Holick MF. Vitamin D deficiency. *N Engl J Med.* 2007;357(3):266-281.
2. Grant WB. Epidemiology of disease risks in relation to vitamin D insufficiency. *Prog Biophys Mol Biol.* 2006;92(1):65-79.
3. Wimalawansa SJ. Non-musculoskeletal benefits of vitamin D. *J Steroid Biochem Mol Biol.* 2018;175:60-81.
4. Nair R, Maseeh A. Vitamin D: The "sunshine" vitamin. *J Pharmacol Pharmacother.* 2012;3(2):118-126.
5. Ali N. Role of vitamin D in preventing of COVID-19 infection, progression and severity. *J Infect Public Health.* 2020;13(10):1373-1380.
6. Grant WB, Lahore H, McDonnell SL, et al. Evidence that Vitamin D Supplementation Could Reduce Risk of Influenza and COVID-19 Infections and Deaths. *Nutrients.* 2020;12(4).
7. Bilezikian JP, Bikle D, Hewison M, et al. Mechanisms in Endocrinology: Vitamin D and COVID-19. *Eur J Endocrinol.* 2020;183(5):R133-r147.
8. van Schoor NM, Lips P. Worldwide vitamin D status. *Best Pract Res Clin Endocrinol Metab.* 2011;25(4):671-680.
9. Diekmann R, Winning K, Bauer JM, et al. Vitamin D status and physical function in nursing home residents: a 1-year observational study. *Z Gerontol Geriatr.* 2013;46(5):403-409.
10. COVID-19; Older Adults. Center for Disease Control and Prevention website. Updated December 13, 2020. Accessed January 20, 2021. https://www.cdc.gov/coronavirus/2019-ncov/need-extra-precautions/older-adults.html.
11. Morbidity and Mortality Weekly Report: Rates of COVID-19 among residents and staff in nursing homes. Center for Disease Control and Prevention website. Updated January 14, 2021. Accessed January 21, 2021. https://www.cdc.gov/mmwr/volumes/70/wr/mm7002e2.htm.
12. Holick MF, Binkley NC, Bischoff-Ferrari HA, et al. Evaluation, treatment, and prevention of vitamin D deficiency: an Endocrine Society clinical practice guideline. *J Clin Endocrinol Metab.* 2011;96(7):1911-1930.
13. Pilz S, Zittermann A, Trummer C, et al. Vitamin D testing and treatment: a narrative review of current evidence. *Endocr Connect.* 2019;8(2):R27-r43.
14. Rosen CJ, Abrams SA, Aloia JF, et al. IOM committee members respond to Endocrine Society vitamin D guideline. *J Clin Endocrinol Metab.* 2012;97(4):1146-1152.
15. Ross AC TC, Yaktine AL, del Valle, HB. Dietary Reference Intakes for Calcium and Vitamin D; Committee to Review Dietary Reference Intake for Vitamin D and Calcium. In: Medicine Io, ed. Washington DC, USA: National Acaemic PRess; 2010.
16. Kotlarczyk MP, Perera S, Ferchak MA, Nace DA, Resnick NM, Greenspan SL. Vitamin D deficiency is associated with functional decline and falls in frail elderly women despite supplementation. Osteoporos Int. 2017;28(4):1347-1353.

17. Rolland Y, de Souto Barreto P, Abellan Van Kan G, et al. Vitamin D supplementation in older adults: searching for specific guidelines in nursing homes. J Nutr Health Aging. 2013;17(4):402-412.

18. Holick MF. Vitamin D deficiency in 2010: health benefits of vitamin D and sunlight: a D-bate. Nat Rev Endocrinol. 2011;7(2):73-75.

19. Singh G, Bonham AJ. A predictive equation to guide vitamin D replacement dose in patients. J Am Board Fam Med. 2014;27(4):495-509.

20. Bischoff-Ferrari HA, Giovannucci E, Willett WC, Dietrich T, Dawson-Hughes B. Estimation of optimal serum concentrations of 25-hydroxyvitamin D for multiple health outcomes. Am J Clin Nutr. 2006;84(1):18-28.

21. Schwartz JB, Kane L, Bikle D. Response of Vitamin D Concentration to Vitamin D3 Administration in Older Adults without Sun Exposure: A Randomized Double-Blind Trial. J Am Geriatr Soc. 2016;64(1):65-72.

22. Chel V, Wijnhoven HA, Smit JH, Ooms M, Lips P. Efficacy of different doses and time intervals of oral vitamin D supplementation with or without calcium in elderly nursing home residents. Osteoporos Int. 2008;19(5):663-671.

23. Rolland Y, Abellan van Kan G, Hermabessiere S, Gerard S, Guyonnet Gillette S, Vellas B. Descriptive study of nursing home residents from the REHPA network. J Nutr Health Aging. 2009;13(8):679-683.

24. Buckinx F, Reginster JY, Cavalier E, et al. Determinants of vitamin D supplementation prescription in nursing homes: a survey among general practitioners. Osteoporos Int. 2016;27(3):881-886.

25. Medical Necessity for Vitamin Assay D Testing. Center for Medicare and Medicaid Services website. Published 2019. Accessed April 5, 2019. https://www.cms.gov/Research-Statistics-Data-and-Systems/Monitoring-Programs/Medicare-FFS-Compliance-Programs/Recovery-Audit-Program/Approved-RAC-Topics-Items/0143-Medical-Necessity-for-Vitamin-D-Assay-Testing-CPT-82306-CPT-82652.

26. Karanicolas PJ, Farrokhyar F, Bhandari M. Practical tips for surgical research: blinding: who, what, when, why, how? Can J Surg. 2010;53(5):345-348.

27. Li T, Vedula SS, Hadar N, Parkin C, Lau J, Dickersin K. Innovations in data collection, management, and archiving for systematic reviews. Ann Intern Med. 2015;162(4):287-294.
28. 2013-2017 ACS 5-year Estimates. U.S. Census Bureau website. Updated November 28, 2018. Accessed April 3, 2019. https://www.census.gov/programs-surveys/acs/technical-documentation/table-and-geography-changes/2017/5-year.html.

29. About Adult BMI. Center for Disease Control and Prevention website. Updated September 17, 2020. Accessed January 23, 2018. https://www.cdc.gov/healthyweight/assessing/bmi/adult_bmi/index.html.

30. USP Therapeutic Categories Model Guidelines. Food and Drug Administration website. Updated 3/28/2018. Accessed April, 2018. https://www.fda.gov/regulatory-information/fdaaa-implementation-chart/usp-therapeutic-categories-model-guidelines.

31. Robien K, Oppeneer SJ, Kelly JA, Hamilton-Reeve JM. Drug-vitamin D interactions: a systematic review of the literature. *Nutr Clin Pract.* 2013;28(2):194-208.

32. USDA Food Composition Databases. U.S. Department of Agriculture website. Published 2018. Updated October 2019. Accessed June, 2018. https://fdc.nal.usda.gov.

33. Lips P, van Schoor NM, de Jongh RT. Diet, sun, and lifestyle as determinants of vitamin D status. *Ann N Y Acad Sci.* 2014;1317:92-98.

34. Sliney DH, Wengraitis S. Is a differentiated advice by season and region necessary? *Prog Biophys Mol Biol.* 2006;92(1):150-160.

35. Instructions for Use: Access 25(OH)D Vitamin D Total 25(OH)D vitamin D. In. B29609 D: kman Coulter , Inc.; 2017:1-16.

36. Madenci Ö, Orçun A, Yildiz Z, et al. Evaluation of new Beckman Coulter 25(OH) Vitamin D assay and potential improvement of clinical interpretation. *Biochem Med (Zagreb).* 2017;27(2):332-341.

37. Wijnen H, Salemink D, Roovers L, Taekema D, de Boer H. Vitamin D supplementation in nursing home patients: randomized controlled trial of standard daily dose versus individualized loading dose regimen. *Drugs Aging.* 2015;32(5):371-378.

38. Medicine) Ilo. 2011 Dietary reference intake for calcium and vitamin D. *Washington DC: The National Academies Press.*

39. Gallagher JC, Sai A, Templin T, 2nd, Smith L. Dose response to vitamin D supplementation in postmenopausal women: a randomized trial. *Ann Intern Med.* 2012;156(6):425-437.

40. Gallagher JC, Peacock M, Yalamanchili V, Smith LM. Effects of vitamin D supplementation in older African American women. *J Clin Endocrinol Metab.* 2013;98(3):1137-1146.

41. Recommendations abstracted from the American Geriatrics Society Consensus Statement on vitamin D for Prevention of Falls and Their Consequences. *J Am Geriatr Soc.* 2014;62(1):147-152.
42. Fuleihan Gel H, Bouillon R, Clarke B, et al. Serum 25-Hydroxyvitamin D Levels: Variability, Knowledge Gaps, and the Concept of a Desirable Range. *J Bone Miner Res.* 2015;30(7):1119-1133.

43. Sempos CT, Binkley N. 25-Hydroxyvitamin D assay standardisation and vitamin D guidelines paralysis. *Public Health Nutr.* 2020;23(7):1153-1164.

44. Bruyere O, Cavalier E, Souberbielle JC, et al. Effects of vitamin D in the elderly population: current status and perspectives. *Arch Public Health.* 2014;72(1):32.

45. Black LJ, Seamans KM, Cashman KD, Kiely M. An updated systematic review and meta-analysis of the efficacy of vitamin D food fortification. *J Nutr.* 2012;142(6):1102-1108.

46. Suominen MH, Hosia-Randell HM, Muurinen S, et al. Vitamin D and calcium supplementation among aged residents in nursing homes. *J Nutr Health Aging.* 2007;11(5):433-437.

47. Andrews KW, Gusev PA, McNeal M, et al. Dietary Supplement Ingredient Database (DSID) and the Application of Analytically Based Estimates of Ingredient Amount to Intake Calculations. *J Nutr.* 2018;148(suppl_2):1413s-1421s.

48. Heaney RP, Davies KM, Chen TC, Holick MF, Barger-Lux MJ. Human serum 25-hydroxycholecalciferol response to extended oral dosing with cholecalciferol. *Am J Clin Nutr.* 2003;77(1):204-210.

49. Jones KS, Assar S, Harnpanich D, et al. 25(OH)D2 half-life is shorter than 25(OH)D3 half-life and is influenced by DBP concentration and genotype. *J Clin Endocrinol Metab.* 2014;99(9):3373-3381.

50. Veleva BI, Chel VG, Achterberg WP. Efficacy of daily 800 IU vitamin D supplementation in reaching vitamin D sufficiency in nursing home residents: cross-sectional patient file study. *BMC Geriatr.* 2014;14:103.
### Table 1. Population Characteristics: Mean Supplementation Dose Rates and Serum 25(OH)D Concentrations

|                          | n, %   | Dietary supplement intake dose rate (IU/d) (mean, SE)† | p-value‡  | Serum 25(OH)D (nmol/L) (mean, SE)† | p-value‡ | Insufficient (<75 nmol/L) (%) | p-value‡ |
|--------------------------|--------|------------------------------------------------------|-----------|-----------------------------------|-----------|-------------------------------|-----------|
| **Sex**                  |        |                                                      |           |                                   |           |                               |           |
| Female                   | 107, 61| 1200, 221                                            | 0.957     | 83, 4                             | 0.731     | 53                            | 0.524     |
| Male                     | 66, 39 | 1184, 182                                            |           | 80, 3                             |           | 58                            |           |
| **Race**                 |        |                                                      |           |                                   |           |                               |           |
| Caucasian                | 159, 89| 1235, 254                                            | 0.460     | 83, 3                             | 0.216     | 53                            | 0.813     |
| Non-Caucasian            | 19, 11 | 840, 164                                             |           | 70, 7                             |           | 53                            |           |
| **Level of Care**        |        |                                                      |           |                                   |           |                               |           |
| AL                        | 62, 35 | 1300, 337                                            |           | 89, 3                             |           | 48                            |           |
| SNF                      | 111, 65| 1100, 145                                            | 0.501     | 77, 3                             | 0.063     | 59                            | 0.181     |
| **Medical Diagnosis**    |        |                                                      |           |                                   |           |                               |           |
| Liver Disease*           | 8, 5   | 850, 235                                             | 0.629     | 55, 15                            | 0.061     | 87                            | 0.060     |
| Renal Disease*           | 55, 32 | 1500, 250                                            | 0.213     | 83, 4                             | 0.942     | 44                            | 0.024     |
| Medication Interaction‡‡ | 90, 52 | 1280, 261                                            | 0.555     | 75, 6                             | 0.155     | 45                            | 0.563     |
| **Total Population**     | 173, 100| 1190, 152                                           |           | 80, 3                             |           | 55                            |           |
| Vit D Supplemented        | 108, 62| 2030, 227                                            |           | 94, 4                             |           | 42                            |           |
| Non-supplemented          | 65, 38 | 8, 5                                                 | <0.001    | 60, 3                             | <0.001    | 81                            | <0.001    |
| **Supplementation Treatment Forms** ||| | | | | |
| Vit D<sub>2or3</sub>     | 29, 27 | 2900, 539                                            |           | 92, 6                             |           | 35                            |           |
| Ca+D                     | 14, 13 | 500, 65                                              |           | 73, 6                             |           | 50                            |           |
| MVI                       | 17, 16 | 466, 46                                              |           | 71, 5                             |           | 77                            |           |
| Combo                    | 48, 44 | 2300, 317                                            | <0.001    | 108, 8                            | <0.001    | 30                            | <0.001    |
| **Supplement Dose Range (IU/d)** ||| | | | | |
| 0-399                     | 70, 41 | 8, 5                                                 |           | 63, 3                             |           | 79                            |           |
| 400-799                   | 27, 45 | 418, 15                                              |           | 73, 5                             |           | 59                            |           |
| 800-1999                  | 30, 17 | 1090, 66                                             |           | 88, 5                             |           | 43                            |           |
| 2000-3999                 | 30, 17 | 2300, 51                                             |           | 104, 6                            |           | 27                            |           |
| >4000                     | 16, 9  | 6100, 902                                            | <0.001    | 133, 19.5                         | <0.001    | 19                            | <0.001    |

*AL, Assisted Living; SNF, Skilled Nursing Facility; Vitamin D<sub>2or3</sub>, supplement containing either vitamin D<sub>2</sub> or D<sub>3</sub>; MVI, multivitamin; Ca+D, calcium+ vitamin D; Combo, combination of vitamin
†Supplementation continuous variable reported as means ± standard error.
‡p-value determined by t-test, Chi-square, or ANCOVA depending on variable.
*Compared to not having diagnosis of liver or renal
‡‡Compared to medication without drug-vitamin D interaction
**Percentages within each category
ANCOVA Covariates: age, BMI, sex, years living in community, diagnosis of liver or renal disease, vitamin D provided in meals and oral supplements, and medication with a drug-vitamin D interaction.
Table 2. Pairwise Comparison of Serum 25(OH)D Concentrations Across Supplementation Treatment Forms and Dose Ranges

| Supplement Treatment Type | Mean Difference$^\dagger$ (nmol/L) | Bonferroni 95% (CI) | p-value$^\ddagger$ |
|---------------------------|-------------------------------------|---------------------|-------------------|
| Vit D$_{2or3}$ vs No D supplement | 30.3 | 7.5, 53.3 | 0.001 |
| Ca+Vit D vs No D supplement | 22.0 | -12.0, 48.0 | 0.070 |
| MVI vs No D supplement | 8.0 | -20.0, 35.6 | 1.000 |
| Combo vs No D supplement | 39.5 | 27.8, 66.5 | 0.001 |
| Ca+Vit D vs Vit D$_{2or3}$ | -8.3 | -45.6, 20.5 | 1.000 |
| MVI vs Vit D$_{2or3}$ | -22.3 | -54.0, 8.0 | 0.130 |
| Combo vs Vit D$_{2or3}$ | 9.3 | -7.5, 40.5 | 1.000 |
| MVI vs Ca+Vit D | -14.0 | -47.0, 26.5 | 1.000 |
| Combo vs Ca+Vit D | 17.5 | -1.8, 60.0 | 0.410 |
| Combo vs MVI | 31.5 | 10.8, 68.3 | 0.003 |

| Supplement Dose Range (IU/d) | Mean Difference$^\dagger$ (nmol/L) | Bonferroni 95% (CI) | p-value$^\ddagger$ |
|-----------------------------|-------------------------------------|---------------------|-------------------|
| 400-799 vs 0-399 | 11.0 | -11.0, 33.0 | 1.000 |
| 800-1999 vs 0-399 | 24.5 | 3.0, 46.0 | 0.001 |
| 2000-3999 vs 0-399 | 42.5 | 21.3, 64.0 | 0.001 |
| >4000 vs 0-399 | 69.0 | 42.3, 69.0 | 0.001 |
| 800-1999 vs 400-799 | 13.3 | -12.8, 39.0 | 1.000 |
| 2000-3999 vs 400-799 | 31.5 | 5.5, 57.0 | 0.005 |
| >4000 vs 400-799 | 58.3 | 27.5, 89.0 | 0.001 |
| 2000-3999 vs 800-1999 | 18.3 | -7.25, 43.5 | 0.430 |
| >4000 vs 800-1999 | 44.8 | 14.5, 75 | 0.001 |
| >4000 vs 2000-3999 | 26.5 | -3.5, 56.8 | 0.130 |

Vitamin D$_{2or3}$, supplement containing either vitamin D$_{2}$ or D$_{3}$; MVI, multivitamin; Ca+Vit D, calcium + vitamin D; Combo, combination of vitamin D supplementation types.
CI, Confidence Intervals
$^\dagger$Difference of means between pairs (nmol/L)
$^\ddagger$p-value determined by ANCOVA with Bonferroni adjustments.
Adjusted p-value significant at p<0.005
Covariates: age, BMI, sex, years living in community, diagnosis of liver or renal disease, vitamin D provided in meals, and medication that interferes with vitamin D metabolism