Published in final edited form as:

Mayo Clin Proc. 2018 June ; 93(6): 721–730. doi:10.1016/j.mayocp.2018.03.006.

Serum 25-Hydroxyvitamin D Values and Risk of All-Cause and Cause-Specific Mortality: A Population-Based Cohort Study

Daniel V. Dudenkov, MD,

Division of General Internal Medicine, Mayo Clinic

Kristin C. Mara, MS,

Division of Biomedical Statistics and Informatics, Mayo Clinic

Tanya M. Petterson, MS,

Division of Biomedical Statistics and Informatics, Mayo Clinic

Julie A. Maxson, CCRP, and

Department of Family Medicine, Mayo Clinic

Tom D. Thacher, MD

Department of Family Medicine, Mayo Clinic

Abstract

Objective—To determine the relationship between 25(OH)D values and all-cause and cause-specific mortality.

Patients and Methods—We identified all serum 25(OH)D measurements in adults residing in Olmsted County, MN between 01/01/2005 and 12/31/2011, through the Rochester Epidemiology Project. All-cause mortality was the primary outcome. Subjects were followed until their last clinical visit as an Olmsted County resident, 12/31/2014, or death. Multivariate analyses were adjusted for age, sex, race, month of measurement, and Charlson comorbidity index.

Results—A total of 11,022 unique persons had a 25(OH)D measurement between 01/01/2005 and 12/31/2011, with a mean ($\pm$ SD) value of 30.0 ± 12.9 ng/mL. Mean age was 54.3 ± 17.2 years, and the majority were female (77.1%) and white (87.6%). There were 723 deaths after a median follow-up of 4.8 years (IQR 3.4–6.2). Unadjusted all-cause mortality hazard ratios (HRs) (95% confidence interval) for 25(OH)D values <12, 12–19, and >50 ng/mL were 2.6 (2.0–3.2), 1.3 (1.0–1.6) and 1.0 (0.72–1.5), respectively, compared with the reference 20–50 ng/mL. In a multivariate model, the interaction between the effect of 25(OH)D and race on mortality was significant ($P < .001$). In whites, adjusted HRs for 25(OH)D values <12, 12–19, 20–50, and >50 ng/mL were 2.5

Reprints and correspondence: Daniel Dudenkov, MD 200 First Street S.W. Rochester, MN 55905 dudenkov.daniel@mayo.edu.

Disclosure

Dr. Tom Thacher is a consultant for Biomedical Systems and Ultragenyx Pharmaceutical. The other authors have nothing to disclose.

Publisher's Disclaimer: This is a PDF file of an unedited manuscript that has been accepted for publication. As a service to our customers we are providing this early version of the manuscript. The manuscript will undergo copyediting, typesetting, and review of the resulting proof before it is published in its final citable form. Please note that during the production process errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.

(2.2–2.9), 1.4 (1.2–1.6), 1.0 (referent), and 1.0 (0.81–1.3), respectively. In non-whites, adjusted HRs were 1.9 (1.5–2.3), 1.7 (1.1–2.6), 1.5 (1.0–2.0), and 2.1 (0.77–5.5).

Conclusion—White patients with 25(OH)D values <20 ng/mL had greater all-cause mortality than those with values 20–50 ng/mL, and white patients had greater mortality associated with low 25(OH)D values than non-white patients. Values of 25(OH)D >50 ng/mL were not associated with all-cause mortality.

Keywords

vitamin D; epidemiology; mortality; race; public health

Introduction

Vitamin D deficiency is a globally prevalent condition that is associated with multiple adverse health outcomes including mortality.^{1–3} An inverse relationship between serum 25-hydroxyvitamin D [25(OH)D], the accepted measure of vitamin D status, and all-cause mortality has been reported for 25(OH)D values up to 20–36 ng/mL.^{4–9} Higher cardiovascular, cancer and respiratory mortality have been associated with low vitamin D status.^{7,10}

More recent debate has surfaced regarding the effect of high 25(OH)D levels on mortality. A handful of studies have demonstrated a reverse J-shaped association between serum 25(OH)D and mortality with increased risk of all-cause mortality^{4,11,12} and cardiovascular disease mortality⁴ at 25(OH)D values >50–60 ng/mL. We previously described an increasing incidence of 25(OH)D values >50 ng/mL, which raises potential concern for the long-term impact of this change.¹³ Taking a cautious approach to the literature, the Institute of Medicine committee formulating Dietary Reference Intakes for vitamin D recommended a tolerable upper intake level of 4,000 IU, corresponding to a serum 25(OH)D concentration of approximately 50 ng/mL.¹⁴ The committee noted that individual vitamin D requirements are variable, but they identified serum 25(OH)D values below 20 ng/mL as increasing the risk of vitamin D deficiency.

Most previous studies of vitamin D and mortality have been limited by low numbers of subjects with high serum 25(OH)D values, lack of racial diversity, lack of information on cause-specific mortality, or were not population-based. Our objective was to conduct a population-based study in the United States of the relationship between vitamin D status and all-cause and cause-specific mortality. We hypothesized that we would observe a reverse J-shaped association between 25(OH)D concentrations and mortality.

Methods

Olmsted County and the Rochester Epidemiology Project

Data from the Rochester Epidemiology Project (REP) was used to retrieve the cohort and outcomes data. The REP database is a rare example of a population-based medical record linkage system that includes over fifty years of health care utilization, diagnostic and laboratory data from virtually all medical care providers within Olmsted County, Minnesota,

covering 98% of all health care services provided for Olmsted County residents.^{15,16} The county is served by two large integrated health systems, the Mayo Clinic and the Olmsted Medical Center, including primary through tertiary services, outpatient and hospital care.¹⁷ Over 95% of the Olmsted County population has granted the medical records research authorization required by Minnesota law, thus allowing their records to be used for research.^{18,19}

Olmsted County, MN is located in the Upper-Midwestern United States (44° north latitude) and has limited sun exposure for residents in winter months. The population of Olmsted County increased from 135,897 to 148,700 between 2002 and 2011. In the 2000 and 2010 censuses, the proportions of Olmsted County residents classified as white, black, Asian and Hispanic were 90% and 86%, 2.7% and 4.8%, 4.3% and 5.5%, and 2.4% and 4.2%, respectively. The proportions of males were 49% and 48%, and of individuals aged 65 years and older 11% and 13%, respectively. Compared with the entire U.S. 2010 population, the county is less ethnically diverse (72% versus 86% white), more educated (85% versus 94% high school graduates) and wealthier (\$51,914 versus \$64,090 median household income). However, characteristics of the population are very similar to the overall population of the upper Midwest.¹⁹

Patient Selection and Outcomes

We identified all initial serum 25(OH)D measurements (index 25(OH)D measurement) in persons age 18 years and older residing in Olmsted County, MN between 01/01/2005 and 12/31/2011, using the resources of the Rochester Epidemiology Project. Patients were followed until their last clinical visit in Olmsted County as an Olmsted County resident, 12/31/2014, or death, whichever came first. We required that survival be conditional on surviving the first 30 days following the first ever 25(OH)D measurement due to concerns about sampling bias; our fear was some patients may have had 25(OH)D measured because they were already so ill that their immediate life expectancy was low.

We collected data on age at medical visit, sex, race, and Charlson comorbidity index. Cause-specific mortality was assessed according to four ICD-10 subsets: circulatory system diseases (includes cardiovascular, cerebrovascular and other vascular causes), I00-I99; respiratory diseases (includes respiratory infections and chronic respiratory diseases), J00-J98; all malignant cancers, C00-C97; and all other causes.

Laboratory Methods

All 25(OH)D tests ordered in Olmsted County were measured at the Mayo Clinic Laboratory during the study interval, with results and date of blood draw recorded. The method of 25(OH)D measurement was isotope-dilution liquid chromatography tandem mass spectrometry (LC-MS/MS) with an inter-assay coefficient of variation of 3.7–11% (Dr. Ravinder Singh, personal communication).

Statistical Analysis

Serum 25(OH)D was examined as a continuous variable and as a categorical variable using predetermined ranges of interest: 25(OH)D values <12, 12–19, 20–50 (reference category),

and >50 ng/mL. The Kaplan-Meier estimator (product limit method) was used to calculate the probability of survival for each of the four categories of 25(OH)D. Cox proportional hazards regression was used to assess the relationship of 25(OH)D and all-cause mortality, univariately and in a multivariable model. Multivariable analysis was adjusted for age, sex, race, month of 25(OH)D measurement, and Charlson comorbidity index at the time of 25(OH)D measurement. All 2-way interactions between main effects were examined. Ties in event and censoring times were handled using the Efron method. The functional form of continuous variables was assessed using Martingale residuals and the proportional hazards assumption was assessed via plots of the Schoenfeld residuals.²⁰ Additionally, after examining a penalized spline fit to the Martingale residuals, an outcome-based cut point analysis²¹ with a log-rank based test²² was used to look for the best initial 25(OH)D value separating 25(OH)D values into the most homogeneous groups with respect to survival. After looking at all-cause mortality, we performed similar analyses looking at the cause-specific deaths: cancer-related deaths, circulatory-related deaths, respiratory-related deaths, and other deaths (determined based on the diagnosis listed first on the death certificate; circulatory ICD10: I00-I99; respiratory ICD10: J00-J98; cancer ICD10: C00-C97; other - all remaining deaths).

Results

A total of 13,599 unique persons had a 25(OH)D measurement between 01/01/2005 and 12/31/2011. After excluding those with a death or censored within 30 days following first ever 25(OH)D measurement, we were left with a sample size of 11,022 patients (Table 1). The mean ($\pm$ SD) age was 54.3 $\pm$ 17.2 years with a predominance of women (77.1%) and whites (87.6%). The mean 25(OH)D value was 30.0 $\pm$ 12.9 ng/mL. Of the total cohort, the proportions of patients with 25(OH)D values <12, 12–19, 20–50 and >50 ng/mL were 5.8%, 14.6%, 74.5% and 5.1%, respectively. Among non-whites, a greater proportion were in the lower 25(OH)D ranges than whites.

There were 723 deaths (123 cancer-related, 125 circulatory-related, 159 respiratory-related, 316 other) after a median observed follow-up of 4.8 y (IQR 3.4–6.2). Supplemental Table 1 includes the numbers of deaths according to 25(OH)D category, sex and race. The overall survival was 93.6% (95% CI: 93.1–94.2%) at 5 y after index 25(OH)D. On univariate analysis, the risk of death was greater in males (hazard ratio [HR] 1.88, 95% CI 1.61–2.19), those who were older (HR per 10 year increase 2.45, 95% CI 2.31–2.60), and those with a higher Charlson comorbidity index (HR per unit 1.29, 95% CI 1.27–1.31) (Table 2). All-cause mortality unadjusted HRs for 25(OH)D values <12, 12–20, and >50 ng/mL were 2.56 (95% CI 2.04–3.21), 1.27 (95% CI 1.04–1.55) and 1.03 (95% CI 0.72–1.48), respectively, compared with 25(OH)D values 20–50 ng/mL. There was a significant interaction between 25(OH)D values and race on mortality ($P=.003$). For whites, 25(OH)D values <12, 12–20, and >50 ng/mL had HRs of 3.57 (95% CI 2.81–4.54), 1.46 (95% CI 1.18–1.81) and 1.06 (95% CI 0.73–1.54), respectively, compared with the reference category of whites with 25(OH)D values 20–50 ng/mL. For non-whites, the respective HRs were 0.79 (95% CI 0.51–1.22), 0.80 (95% CI 0.57–1.12) and 1.23 (95% CI 0.46–3.29), compared with the same reference category (Table 2). There were no significant interactions of sex and age with 25(OH)D values and mortality.

The final model for all-cause mortality included age (quadratic), sex, Charlson comorbidity index, index month, race, vitamin D categories, and the interaction between race and serum 25(OH)D (Table 3). As in the univariate analysis, we observed a greater risk of death in males, those who were older, and those with a higher Charlson index. For low 25(OH)D levels, whites were at a much higher risk of death than non-whites (Figure 1 and Supplemental Figure). In whites, 25(OH)D values <20 ng/mL were associated with increased all-cause mortality, but this association did not exist for non-whites. For the entire cohort, 25(OH)D values >50 ng/mL were not associated with increased mortality.

The cut-point analysis of the relationship of overall survival with 25(OH)D values identified nodes at approximately 12 and 20 ng/mL, which were consistent with our predetermined 25(OH)D categories. We estimated a 25(OH)D value of 20–25 ng/mL as the lower bound of the range without a significant association between 25(OH)D and mortality. Cut-point analysis did not reveal higher values of 25(OH)D that would suggest an association with high values of 25(OH)D and mortality (i.e., the risk of death was similar across 25(OH)D values for those with 25(OH)D values greater than 20–25 ng/mL (Figure 1).

The final models for cause-specific mortality included age, sex, Charlson comorbidity index, index month, race, and 25(OH)D categories (Supplemental Table 2). The risk of cancer-related death was greater in those who were older and in those with a higher Charlson index. Low 25(OH)D values were associated with a significantly greater risk of cancer-related death compared with the reference category 20–50 ng/mL (HR for <12 ng/mL 2.56, 95% CI 1.45–4.53; HR for 12–19 ng/mL 1.69, 95% CI 1.06–2.70). The risk of circulatory-related death was greater in those who were older, males, non-whites, and in those with a higher Charlson index. Very low 25(OH)D values <12 ng/mL were associated with a significantly greater risk of circulatory-related death compared with the reference category 20–50 ng/mL (HR 2.48, 95% CI 1.53–4.02). The risk of respiratory-related death was greater in those who were older, males, and in those with a higher Charlson index. Those with 25(OH)D values >50 ng/mL were at marginally greater risk of respiratory-related death compared with the reference category (HR 1.95, 95% CI 1.004–3.78). The risk of other causes of death was greater in those who were older, males, and in those with a higher Charlson index. Very low 25(OH)D values <12 ng/mL were associated with a significantly greater risk of other causes of death compared with the reference category 20–50 ng/mL (HR 2.72, 95% CI 1.91–3.89).

Discussion

We found a strong association between 25(OH)D concentrations <20 ng/mL and all-cause mortality. There was a significant interaction by race, with no association for non-whites even at very low 25(OH)D values (<12 ng/mL). Values of 25(OH)D >50 ng/mL were not associated with greater all-cause mortality.

We confirmed the association between low 25(OH)D levels and higher all-cause mortality that has been demonstrated in previous studies, including several meta-analyses.^{5–7,10,23} Previous studies have reported inverse relationships between serum 25(OH)D and mortality up to 25(OH)D concentrations of 20–36 ng/mL.^{4,5,23} We also found that the association

between all-cause mortality and 25(OH)D values of at least 20–25 ng/mL was no longer statistically significant, which was in agreement with our a priori cut-off of 20 ng/mL.

Lower 25(OH)D values may not be causal in increasing mortality, but may be associated with illness and a prognostic marker for mortality.²⁴ However, the effects of vitamin D and its metabolites on inflammation, cellular proliferation, genetic regulation, calcium homeostasis, and immune modulation could have a salutary effect in a variety of diseases that contribute to mortality.^{10,25} Low 25(OH)D values have an adverse effect on mortality independent of PTH levels,²⁶ and high PTH concentrations are associated with increased mortality independent of 25(OH)D values.²⁷ It is not clear that changing the 25(OH)D concentration would alter the mortality risk. Randomized trials have demonstrated a modest mortality benefit with vitamin D supplementation, although most of the included studies were not designed to evaluate mortality as a primary outcome, and most participants were white.²⁸

Values of 25(OH)D >50 ng/mL were not associated with higher all-cause mortality. Although most previous studies found no increase in all-cause mortality associated with 25(OH)D values >50 ng/mL,^{6,23,25} a few found an increased risk with 25(OH)D concentrations >50–60 ng/mL.^{4,11,12} However, vitamin D assay standardization appears to abolish the apparent increased mortality risk associated with high 25(OH)D concentrations, because the radioimmunoassay used in the NHANES III tends to inflate values of 25(OH)D greater than 40 ng/mL.²⁹ Most individuals with 25(OH)D values >50 ng/mL are taking vitamin D to achieve these levels. Some of these patients are possibly being treated with vitamin D because they have had or are at risk for vitamin D deficiency.³⁰ Individuals with a disease associated with vitamin D deficiency (i.e. osteoporosis) may take vitamin D even in the absence of documented vitamin D deficiency. Thus, the reported increase in all-cause mortality is likely confounded by the indication for vitamin D intake. Given the rapid rise in the incidence of 25(OH)D values >50 ng/mL in the population,¹³ our finding is reassuring that all-cause mortality was not increased in this group.

A novel finding of our study was the significant interaction of race and serum 25(OH)D in the relationship with all-cause mortality. Despite a strong association between 25(OH)D concentrations <20 ng/mL and all-cause mortality in whites, this relationship was absent in non-whites, even at very low 25(OH)D values. Most previous studies involved predominantly white populations of European descent with minimal inclusion of non-white races. Of studies that adjusted for race,^{8,11,26,31–34} an interaction between the effect of race and 25(OH)D on all-cause mortality was not described. All-cause mortality data from the Third National Health and Nutrition Examination Survey (NHANES-III) included an oversampling of non-white participants, and in a subgroup analysis, investigators reported a nonsignificant interaction by race.⁹ In a 15-year follow-up of the same study, non-Hispanic whites had an increased all-cause mortality in all 25(OH)D categories <30 ng/mL, whereas non-Hispanic blacks only had increased mortality with 25(OH)D values <8 ng/mL, when compared with a 25(OH)D reference category of 30–39 ng/mL.¹¹

The biological and clinical implications of the race interaction in our study are unclear. Non-white individuals generally have lower 25(OH)D concentrations than whites,^{35,36} an effect

that is accentuated by greater latitude from the equator.³⁷ However, the association of low 25(OH)D values with adverse outcomes in non-white individuals is only evident at very low 25(OH)D concentrations.^{11,38} Despite their lower values of 25(OH)D, African Americans have a lower risk of osteoporosis, a situation called “the vitamin D paradox.”³⁹ Unlike whites, low 25(OH)D values were not associated with fracture risk in blacks.⁴⁰ Non-whites may have a different association between low 25(OH)D concentrations and other comorbidities.^{41,42} Investigators using NHANES-III data found that 25(OH)D concentrations <15 ng/mL were associated with higher fatal stroke compared to concentrations ≥15 ng/mL in whites but not in blacks, after adjusting for multiple covariates.⁴³ Others using the NHANES-III data found that the excess cardiovascular mortality seen in blacks was completely eliminated after adjusting for 25(OH)D levels and income.⁴⁴

The strengths of our study include a large community-based population and access to comprehensive data from primary through tertiary care for nearly the entire population. All measurements of 25(OH)D were performed in the same laboratory with LC-MS/MS, which provides optimal accuracy of 25(OH)D values, and no standardization adjustment was necessary. However, our study had limitations that are worth noting. We do not have a true population sample, and women are overrepresented. This may be related to more frequent measurement of 25(OH)D in women, who are at greater risk of osteoporosis than men. Subjects were those who presented for care and had 25(OH)D measured, so some selection bias is likely. Our population is less ethnically diverse than the US population, which may limit generalizability; in particular, among our non-white population, only 75 individuals died (Supplemental Table 1). Compared to similar previous studies, most of which are from predominantly white populations of European descent, our cohort is actually more ethnically diverse. A larger number of non-white subjects would have provided greater power to detect an increase in mortality among non-whites associated with 25(OH)D values below 12 ng/mL. This is visually represented in Figure 1 by the line for the 95% CI rising above 1 for non-whites. However, our finding of a difference in mortality between whites and non-whites associated with low 25(OH)D values remains valid, based on the statistical significance of the interaction. We could not establish causality of the association of low 25(OH)D with mortality, due to the retrospective nature of the study. Furthermore, a single measurement of serum 25(OH)D may not represent long-term vitamin D status, but 25(OH)D values do appear to remain relatively stable over time.⁴⁵

Conclusion

White patients with 25(OH)D values <20 ng/mL had a greater all-cause mortality than those with 25(OH)D values 20–50 ng/mL. White patients had greater mortality associated with low 25(OH)D values than non-white patients. Values of 25(OH)D >50 ng/mL were not associated with an increased risk of all-cause mortality.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

Financial support

This study was made possible using the resources of the Rochester Epidemiology Project, which is supported by the National Institute of Aging of the National Institutes of Health (NIH) under Award Number R01AG034676. The study was also supported by the Mayo Clinic CTSA through grant number UL1 TR000135 from the National Center for Advancing Translational Sciences (NCATS), a component of the NIH. The content is solely the responsibility of the authors and does not necessarily represent the official views of the NIH.

List of Abbreviations

25(OH)D	25-hydroxyvitamin D
REP	Rochester Epidemiology Project
HR	hazard ratio
CI	confidence interval
ng/mL	nanograms per milliliter

References

1. Holick MF. Vitamin D deficiency. *N Engl J Med*. 2007; 357(3):266–281. [PubMed: 17634462]
2. Thacher TD, Clarke BL. Vitamin D insufficiency. *Mayo Clin Proc*. 2011; 86(1):50–60. [PubMed: 21193656]
3. Ross AC, Manson JE, Abrams SA, et al. The 2011 report on dietary reference intakes for calcium and vitamin D from the Institute of Medicine: what clinicians need to know. *J Clin Endocrinol Metab*. 2011; 96(1):53–58. [PubMed: 21118827]
4. Durup D, Jorgensen HL, Christensen J, Schwarz P, Heegaard AM, Lind B. A reverse J-shaped association of all-cause mortality with serum 25-hydroxyvitamin D in general practice: the CopD study. *J Clin Endocrinol Metab*. 2012; 97(8):2644–2652. [PubMed: 22573406]
5. Zittermann A, Iodice S, Pilz S, Grant WB, Bagnardi V, Gandini S. Vitamin D deficiency and mortality risk in the general population: a meta-analysis of prospective cohort studies. *Am J Clin Nutr*. 2012; 95(1):91–100. [PubMed: 22170374]
6. Gaksch M, Jorde R, Grimnes G, et al. Vitamin D and mortality: Individual participant data meta-analysis of standardized 25-hydroxyvitamin D in 26916 individuals from a European consortium. *PloS One*. 2017; 12(2):e0170791. [PubMed: 28207791]
7. Schottker B, Haug U, Schomburg L, et al. Strong associations of 25-hydroxyvitamin D concentrations with all-cause, cardiovascular, cancer, and respiratory disease mortality in a large cohort study. *Am J Clin Nutr*. 2013; 97(4):782–793. [PubMed: 23446902]
8. Ginde AA, Scragg R, Schwartz RS, Camargo CA Jr. Prospective study of serum 25-hydroxyvitamin D level, cardiovascular disease mortality, and all-cause mortality in older U.S. adults. *J Am Geriatr Soc*. 2009; 57(9):1595–1603. [PubMed: 19549021]
9. Melamed ML, Michos ED, Post W, Astor B. 25-hydroxyvitamin D levels and the risk of mortality in the general population. *Arch Intern Med*. 2008; 168(15):1629–1637. [PubMed: 18695076]
10. Chowdhury R, Kunutsor S, Vitezova A, et al. Vitamin D and risk of cause specific death: systematic review and meta-analysis of observational cohort and randomised intervention studies. *BMJ*. 2014; 348:g1903. [PubMed: 24690623]
11. Sempos CT, Durazo-Arvizu RA, Dawson-Hughes B, et al. Is there a reverse J-shaped association between 25-hydroxyvitamin D and all-cause mortality? Results from the U.S. nationally representative NHANES. *J Clin Endocrinol Metab*. 2013; 98(7):3001–3009. [PubMed: 23666975]

12. Amrein K, Quraishi SA, Litonjua AA, et al. Evidence for a U-shaped relationship between prehospital vitamin D status and mortality: a cohort study. *J Clin Endocrinol Metab.* 2014; 99(4): 1461–1469. [PubMed: 24423347]
13. Dudenkov DV, Yawn BP, Oberhelman SS, et al. Changing Incidence of serum 25-hydroxyvitamin D values above 50 ng/mL: A 10-year population-based study. *Mayo Clin Proc.* 2015; 90(5):577–586. [PubMed: 25939935]
14. Institute of Medicine. Dietary reference intakes for calcium and vitamin D. Washington, DC: The National Academies Press; 2011.
15. Melton LJ 3rd. History of the Rochester Epidemiology Project. *Mayo Clin Proc.* 1996; 71(3):266–274. [PubMed: 8594285]
16. Rocca WA, Yawn BP, St Sauver JL, Grossardt BR, Melton LJ 3rd. History of the Rochester Epidemiology Project: half a century of medical records linkage in a US population. *Mayo Clin Proc.* 2012; 87(12):1202–1213. [PubMed: 23199802]
17. St Sauver JL, Grossardt BR, Yawn BP, Melton LJ 3rd, Rocca WA. Use of a medical records linkage system to enumerate a dynamic population over time: the Rochester epidemiology project. *Am J Epidemiol.* 2011; 173(9):1059–1068. [PubMed: 21430193]
18. Jacobsen SJ, Xia Z, Champion ME, et al. Potential effect of authorization bias on medical record research. *Mayo Clin Proc.* 1999; 74(4):330–338. [PubMed: 10221460]
19. St Sauver JL, Grossardt BR, Leibson CL, Yawn BP, Melton LJ 3rd, Rocca WA. Generalizability of epidemiological findings and public health decisions: an illustration from the Rochester Epidemiology Project. *Mayo Clin Proc.* 2012; 87(2):151–160. [PubMed: 22305027]
20. Grambsch PM, Therneau TM. Proportional hazards tests and diagnostics based on weighted residuals. *Biometrika.* 1994; 81(3):515–526.
21. William, BA., Mandrekar, JN., Mandrekar, SJ., Cha, SS., Furth, AF. Finding optimal cutpoints for continuous covariates with binary and time-to-event outcomes. Department of Health Sciences Research, Mayo Clinic; 2006.
22. Contal C, O'Quigley J. An application of changepoint methods in studying the effect of age on survival in breast cancer. *Computational Statistics and Data Analysis.* 1999; 30:253–270.
23. Garland CF, Kim JJ, Mohr SB, et al. Meta-analysis of all-cause mortality according to serum 25-hydroxyvitamin D. *Am J Public Health.* 2014; 104(8):e43–50.
24. Schottker B, Saum KU, Perna L, Ordonez-Mena JM, Holleczeck B, Brenner H. Is vitamin D deficiency a cause of increased morbidity and mortality at older age or simply an indicator of poor health? *Eur J Epidemiol.* 2014; 29(3):199–210. [PubMed: 24682834]
25. Schottker B, Ball D, Gellert C, Brenner H. Serum 25-hydroxyvitamin D levels and overall mortality. A systematic review and meta-analysis of prospective cohort studies. *Ageing Res Rev.* 2013; 12(2):708–718. [PubMed: 22343489]
26. Kestenbaum B, Katz R, de Boer I, et al. Vitamin D, parathyroid hormone, and cardiovascular events among older adults. *J Am Coll Cardiol.* 2011; 58(14):1433–1441. [PubMed: 21939825]
27. Kritchevsky SB, Tooze JA, Neiberg RH, et al. 25-Hydroxyvitamin D, parathyroid hormone, and mortality in black and white older adults: the health ABC study. *J Clin Endocrinol Metab.* 2012; 97(11):4156–4165. [PubMed: 22942386]
28. Bjelakovic G, Gluud LL, Nikolova D, et al. Vitamin D supplementation for prevention of mortality in adults. *Cochrane Database Syst Rev.* 2014; (1):CD007470. [PubMed: 24414552]
29. Durazo-Arvizu RA, Dawson-Hughes B, Kramer H, et al. The Reverse J-Shaped Association Between Serum Total 25-Hydroxyvitamin D Concentration and All-Cause Mortality: The Impact of Assay Standardization. *Am J Epidemiol.* 2017; 185(8):720–726. [PubMed: 28338905]
30. Kroll MH, Bi C, Garber CC, et al. Temporal relationship between vitamin D status and parathyroid hormone in the United States. *PloS One.* 2015; 10(3):e0118108. [PubMed: 25738588]
31. Saliba W, Barnett O, Rennert HS, Rennert G. The risk of all-cause mortality is inversely related to serum 25(OH)D levels. *J Clin Endocrinol Metab.* 2012; 97(8):2792–2798. [PubMed: 22648653]
32. Kramer H, Sempos C, Cao G, et al. Mortality rates across 25-hydroxyvitamin D (25[OH]D) levels among adults with and without estimated glomerular filtration rate <60 ml/min/1.73 m²: the third national health and nutrition examination survey. *PloS One.* 2012; 7(10):e47458. [PubMed: 23112816]

33. Eaton CB, Young A, Allison MA, et al. Prospective association of vitamin D concentrations with mortality in postmenopausal women: results from the Women's Health Initiative (WHI). *Am J Clin Nutr*. 2011; 94(6):1471–1478. [PubMed: 22030222]
34. Daraghme AH, Bertoia ML, Al-Qadi MO, Abdalbaki AM, Roberts MB, Eaton CB. Evidence for the vitamin D hypothesis: The NHANES III extended mortality follow-up. *Atherosclerosis*. 2016; 255:96–101. [PubMed: 27855294]
35. Powe CE, Evans MK, Wenger J, et al. Vitamin D-binding protein and vitamin D status of black Americans and white Americans. *N Engl J Med*. 2013; 369(21):1991–2000. [PubMed: 24256378]
36. Lips P. Worldwide status of vitamin D nutrition. *J Steroid Biochem Mol Biol*. 2010; 121(1–2):297–300. [PubMed: 20197091]
37. Durazo-Arvizu RA, Camacho P, Bovet P, et al. 25-Hydroxyvitamin D in African-origin populations at varying latitudes challenges the construct of a physiologic norm. *Am J Clin Nutr*. 2014; 100(3):908–914. [PubMed: 25008852]
38. Signorello LB, Han X, Cai Q, et al. A prospective study of serum 25-hydroxyvitamin D levels and mortality among African Americans and non-African Americans. *Am J Epidemiol*. 2013; 177(2):171–179. [PubMed: 23125439]
39. Aloia JF. African Americans, 25-hydroxyvitamin D, and osteoporosis: a paradox. *Am J Clin Nutr*. 2008; 88(2):545S–550S. [PubMed: 18689399]
40. Cauley JA, Danielson ME, Boudreau R, et al. Serum 25-hydroxyvitamin D and clinical fracture risk in a multiethnic cohort of women: the Women's Health Initiative (WHI). *J Bone Miner Res*. 2011; 26(10):2378–2388. [PubMed: 21710614]
41. Gutierrez OM, Farwell WR, Kermah D, Taylor EN. Racial differences in the relationship between vitamin D, bone mineral density, and parathyroid hormone in the National Health and Nutrition Examination Survey. *Osteoporos Int*. 2011; 22(6):1745–1753. [PubMed: 20848081]
42. Harris SS. Does vitamin D deficiency contribute to increased rates of cardiovascular disease and type 2 diabetes in African Americans? *Am J Clin Nutr*. 2011; 93(5):1175S–1178S. [PubMed: 21367947]
43. Michos ED, Reis JP, Post WS, et al. 25-Hydroxyvitamin D deficiency is associated with fatal stroke among whites but not blacks: The NHANES-III linked mortality files. *Nutrition*. 2012; 28(4):367–371. [PubMed: 22261577]
44. Fiscella K, Franks P. Vitamin D, race, and cardiovascular mortality: findings from a national US sample. *Ann Fam Med*. 2010; 8(1):11–18. [PubMed: 20065273]
45. McKibben RA, Zhao D, Lutsey PL, et al. Factors Associated With Change in 25-Hydroxyvitamin D Levels Over Longitudinal Follow-Up in the ARIC Study. *J Clin Endocrinol Metab*. 2016; 101(1):33–43. [PubMed: 26509869]

Figure 1.

Spline plots displaying unadjusted hazard ratios (HR) for all-cause mortality with serum 25(OH)D values as a continuous variable. Dotted lines indicate 95% confidence intervals. A) All patients. HRs with 95% CI are shown for categorical 25(OH)D ranges based on cut-point analysis (<12 ng/mL, 12–19 ng/mL, and >19 ng/mL. B) Separated by race.

Table 1

Baseline characteristics of Olmsted County residents with a serum 25(OH)D measurement between 01/01/2005 and 12/31/2011.

Characteristics	Serum 25(OH)D Categories (ng/mL)				P-value
	Total	<12	12–19	20–50	>50
n	11,022	643	1605	8210	564
25(OH)D, mean (SD), ng/mL	30.0 (12.9)	8.5	15.9	32.4	60.9
Age, mean (SD), y	54.3 (17.2)	51.4 (18.0)	51.8 (17.0)	54.9 (17.1)	55.8 (18.0)
					.10 ^a
Age groups, n (%)					
18–49 y	4244 (38.5)	311 (48.4)	725 (45.2)	3014 (36.7)	194 (34.4)
50–64 y	3784 (34.3)	195 (30.3)	546 (34.0)	2851 (34.7)	192 (34.0)
65 y	2994 (27.2)	137 (21.3)	334 (20.8)	2345 (28.6)	178 (31.6)
Sex, n (%)					<.001
Women	8496 (77.1)	463 (72.0)	1176 (73.3)	6357 (77.4)	500 (88.7)
Men	2526 (22.9)	180 (28.0)	429 (26.7)	1853 (22.6)	64 (11.3)
Race, n (%)					<.001
White	9653 (87.6)	420 (65.3)	1209 (75.3)	7482 (91.1)	542 (96.1)
Non-white	1369 (12.4)	223 (34.7)	396 (24.7)	728 (8.9)	22 (3.9)
Season, n (%)					<.001
Winter, Dec–Feb	2751 (25.0)	225 (35.0)	483 (30.1)	1934 (23.6)	109 (19.3)
Spring, Mar–May	2863 (26.0)	209 (32.5)	510 (31.8)	1992 (24.3)	152 (27.0)
Summer, Jun–Aug	2525 (22.9)	92 (14.3)	247 (15.4)	2032 (24.8)	154 (27.3)
Fall, Sept–Nov	2883 (26.1)	117 (18.2)	365 (22.7)	2252 (27.4)	149 (26.4)
Charlson index, mean (SD)	3.37 (3.4)	3.75 (4.1)	3.23 (3.5)	3.36 (3.4)	3.54 (3.2)
					.06 ^b

^a Pearson Correlation,

^b Spearman Correlation

Table 2

Associations between serum 25(OH)D values and all-cause mortality. Univariate hazard ratios for serum 25(OH)D, as categorized, from Cox proportional hazards modeling of time to death.^a

Univariate Model			Univariate Model Adjusted for One Additional Risk Factor Serum 25(OH)D Categories (ng/mL)							
Adjusting Risk Factor	HR (95% CI)	P-value	20–50 HR ^b (95% CI)	P-value	<12 HR ^b (95% CI)	P-value	12–19 HR ^b (95% CI)	P-value	>50 HR ^b (95% CI)	P-value
25(OH)D, ng/mL										
20–50	1.0									
<12	2.56 (2.04–3.21)	<.001								
12–19	1.27 (1.04–1.55)	.02								
>50	1.03 (0.72, 1.48)	.87								
Sex										
Female	1.0		ref.		2.51 (2.00, 3.15)	P<.001	1.24 (1.02, 1.52)	P=.03	1.12 (0.78, 1.61)	P=.55
Male	1.88 (1.61, 2.19)	<.001								
Age (per 10 y)										
	2.45 (2.31, 2.60)	<.001	ref.		3.08 (2.46, 3.87)	P<.001	1.52 (1.24, 1.86)	P<.001	0.93 (0.65, 1.34)	P=.70
Race										
White	1.0		ref.		3.57 (2.81, 4.54)		1.46 (1.18, 1.81)		1.06 (0.73, 1.54)	
Non-White	0.97 (0.77, 1.24)	.82	1.41 (1.05, 1.90)		0.79 (0.51, 1.22)		0.80 (0.57, 1.12)		1.23 (0.46, 3.29)	
CCI ^d (per unit)										
	1.29 (1.27, 1.31)	<.001								
Season										
Winter, Dec–Feb	1.0	.45 ^e	ref.		2.59 (2.25, 2.98)	P<.001	1.38 (1.22, 1.56)	P=.01	0.93 (0.73, 1.18)	P=.91
Spring, Mar–May	1.16 (0.94, 1.42)	.16								
Summer, Jun–Aug	1.17 (0.94, 1.45)	.15								
Fall, Sept–Nov	1.12 (0.91, 1.38)	.28								

^aTime 0 is 30 days after index 25(OH)D measurement

Author Manuscript

Author Manuscript

Author Manuscript

Author Manuscript

q From model containing 25(OH)D level and one adjusting risk factor (except for the model with race, which also includes the interaction between 25(OH)D level and race in the model)
 c Interaction P -value. HRs presented separately by race due to race-25(OH)D interaction
 d CCI: Charlson comorbidity index
 e Joint test

Table 3Hazard ratios from multivariable Cox proportional hazards modeling of time to death.^a

	Adjusted HR (95% CI)		P-value
Age, y^b			<.001
50	1.0		
60	1.48 (1.39, 1.57)		
70	2.47 (2.24, 2.73)		
Sex			<.001
Female	1.0		
Male	1.52 (1.38, 1.68)		
Charlson Comorbidity Index (per unit)	1.16 (1.15, 1.17)		<.001
Season			.24 ^c
Winter, Dec–Feb	1.0		
Spring, Mar–May	1.01 (0.88, 1.15)		.94
Summer, Jun–Aug	1.12 (0.98, 1.28)		.11
Fall, Sept–Nov	1.10 (0.96, 1.25)		.18
Vitamin D*Race	White	Non-White	<.001 ^d
25(OH)D 20–50 ng/mL	1.0	1.86 (1.50, 2.31)	
25(OH)D <12 ng/mL	2.52 (2.17, 2.91)	1.69 (1.10, 2.62)	
25(OH)D 12–19 ng/mL	1.43 (1.25, 1.63)	1.46 (1.04, 2.04)	
25(OH)D >50 ng/mL	1.04 (0.81, 1.33)	2.06 (0.77, 5.51)	

^aTime 0 is 30 days after index 25(OH)D measurement^bAge is modeled as a quadratic function; the hazard ratios are compared to a 50 year old^cJoint test^dInteraction *P*-value