

# Serum Total 25-OH Vitamin D Adds Little Prognostic Value in Patients Undergoing Coronary Catheterization

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**Background**—Vitamin D deficiency is associated with an increased risk of cardiovascular disease; however, it is unclear whether vitamin D status should be considered in clinical risk assessments of patients with cardiovascular disease.

**Methods and Results**—This study included 2975 patients who had their first serum total 25-hydroxy vitamin D (25-OH vitamin D) measurement before their first coronary catheterization in Alberta, Canada. Cox regression was used to examine associations between 25-OH vitamin D and mortality risk after adjusting for demographic and clinical risk factors. Interactions were tested using multiplicative terms, and prognostic value was assessed using measures of model discrimination, fit, calibration and net reclassification improvement. There were 401 deaths over a median of 5.8 years of follow-up. Serum total 25-OH vitamin D was inversely associated with mortality after adjusting for demographic and clinical risk factors, which was largely driven by excess risk in the bottom quintile (hazard ratio 1.84 for bottom versus top quintile, 95% CI 1.36–2.50, *P* for trend <0.001). Associations were weaker in the presence of several competing risk factors (e.g., advanced age; *P* for interactions <0.05). Adding 25-OH vitamin D to a model containing demographic and clinical risk factors yielded similar discrimination, model fit, and calibration and only modest improvements in risk reclassification (net reclassification improvement 1.9% for deaths, 2.3% for survivors).

**Conclusions**—Pre-catheterization, serum total 25-OH vitamin D was inversely associated with mortality risk after adjusting for established demographic and clinical risk factors. This association was attenuated by several competing risk factors. Overall, 25-OH vitamin D added little prognostic value over established risk factors; therefore, its measurement is not warranted in patients undergoing coronary catheterization. (*J Am Heart Assoc.* 2016;5:e004289 doi: 10.1161/JAHA.116.004289)

**Key Words:** coronary artery disease • mortality • prognostication • risk reclassification • vitamin D

An abundance of epidemiological evidence links vitamin D deficiency to cardiovascular disease (CVD). In 2 large meta-analyses of prospective cohort and intervention studies, lower serum total 25-hydroxyvitamin D (25-OH vitamin D; the major circulating form of vitamin D) was associated with a

greater risk of fatal and nonfatal CVD events in both healthy persons and those with preexisting CVD.<sup>1,2</sup> Combined with biochemical evidence of 25-OH vitamin D's role in the development of cardiovascular risk factors such as insulin resistance,<sup>3</sup> many have suggested that vitamin D deficiency is causally related to CVD. Given the high prevalence of vitamin D deficiency in the Northern Hemisphere,<sup>4</sup> findings such as these have helped increase demand for 25-OH vitamin D testing and supplementation rates.<sup>5</sup> Nevertheless, because of a lack of high-quality trials of vitamin D supplementation,<sup>6</sup> both the US Agency for Healthcare Research and Quality<sup>7</sup> and the US Institute of Medicine<sup>8</sup> have stated that evidence is insufficient to support a causal role of vitamin D in CVD. Others have noted that vitamin D deficiency may simply indicate ill health.<sup>9</sup> As such, 25-OH vitamin D could still be valuable as a clinical prognostic marker.<sup>8,10–12</sup>

While several clinical studies have evaluated the association between 25-OH vitamin D and risk of CVD outcomes, complications, and mortality following surgery or hospitalization,<sup>13–20</sup> few have fully evaluated the prognostic value of 25-OH vitamin D by contrasting model discrimination, fit, and

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An accompanying Table S1 is available at <http://jaha.ahajournals.org/content/5/10/e004289/DC1/embed/inline-supplementary-material-1.pdf>

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calibration to those from models containing established risk factors. Furthermore, none have examined the ability of 25-OH vitamin D to appropriately reclassify patient risk when added to models containing established risk factors.

The objectives of this study were therefore to (1) examine the association between serum total 25-OH vitamin D concentration and total mortality in a population of patients undergoing coronary catheterization; (2) examine variation in this association according to timing between 25-OH vitamin D testing and catheterization, demographic risk factors, clinical risk factors, and season of testing; and (3) determine whether 25-OH vitamin D provides additional prognostic value (superior model discrimination, fit, calibration and risk reclassification) above and beyond established risk factors.

## Methods

### Study Population

Our study included patients from the Alberta Provincial Project for Outcome Assessment in Coronary Heart Disease (APPROACH). Established in 1995, APPROACH is a cardiac admission, catheterization, and revascularization registry that collects demographic, clinical, and outcome data on all patients undergoing invasive cardiac procedures in the province of Alberta, Canada.<sup>21</sup> At the time of analysis, follow-up was available until August 20, 2015. All patients provided informed consent at the time of enrollment. This study complies with the Declaration of Helsinki and was approved by the University of Calgary Conjoint Health Research Ethics Board (ethics identifier E25065).

### Exposure

Results for serum total 25-OH vitamin D, date of testing, and personal health number were extracted from the laboratory information systems of Calgary (Calgary Laboratory Services, August 9, 2007, to June 26, 2013; n=32 610) and Edmonton (Alberta Health Services, August 26, 2008, to September 26, 2013; n=13 784) Alberta. During the time period of this study, 25-OH vitamin D testing was available to all Alberta physicians for any purpose. Because 25-OH vitamin D was measured by liquid chromatography–tandem mass spectrometry in Edmonton and by immunoassay (Liaison; DiaSorin) in Calgary, 240 identical samples were run by each method ( $R^2=0.8$ , slope=1.0, intercept=-8), and Edmonton results were corrected to match those from Calgary by subtracting 8 nmol/L. Results below each assay's limit of quantification were replaced with the respective limit of quantification and results above each assay's upper reporting limit were replaced with the respective upper reporting limit.

## Outcome

The outcome used was total (all-cause) mortality, provided by linkage of the APPROACH registry to Alberta Vital Statistics.

## Covariates

Established prognostic factors were assessed at catheterization in the APPROACH registry. These included both demographic (age, sex) and clinical risk factors (body mass index [BMI; kg/m<sup>2</sup>], renal disease, hypertension, hyperlipidemia, type 2 diabetes mellitus, congestive heart failure, smoking status [current and prior], prior history of myocardial infarction, family history of heart disease, ejection fraction), including a modified 5-point Duke severity score (Duke5) based on the number of coronary vessels affected and the extent of occlusion.

## Statistical Analysis

Laboratory data was merged to the APPROACH registry by personal health number. To eliminate the effect of a change in lifestyle, supplementation, or prior procedure on 25-OH vitamin D concentration only the first 25-OH vitamin D result prior to the first recorded catheterization was used. Patient characteristics were tabulated according to quintiles of 25-OH vitamin D. Trends were evaluated using linear regression (continuous variables) and logistic regression (dichotomous variables). Demographic and clinical risk factors of patients who were tested were compared to those who were not using the Student *t* test (continuous variables) and the chi-squared test (categorical variables).

Cox proportional hazards models were used to evaluate the shape and association between pre-catheterization 25-OH vitamin D quintiles (top quintile as reference) and post-catheterization mortality. Follow-up time was calculated from catheterization until death or censoring. An ordinal variable for vitamin D quintile was used to test for linear trends across quintiles. A second model was adjusted for demographic risk factors (age, sex). A third model was further adjusted for clinical risk factors (BMI, smoking status, renal disease, hypertension, hyperlipidemia, type 2 diabetes mellitus, family history of heart disease, prior myocardial infarction, congestive heart failure, whether ejection fraction was measured, ejection fraction [ $<20\%$ ,  $20\text{--}34\%$ ,  $35\text{--}50\%$ ,  $>50\%$  (reference)], and Duke5 score (0–5)).

Interaction terms (eg, 25-OH vitamin D quintile multiplied by age) were used to test whether associations differed according to the length of time between 25-OH vitamin D testing and catheterization, demographic risk factors, clinical risk factors, and the season of testing (based on dates of solstices and equinoxes in the Northern Hemisphere).

**Table 1.** Patient Characteristics by Quintile of Serum Total 25-OH Vitamin D

Characteristic	Quintile					P for Trend
	1	2	3	4	5	
Patients, n	595	597	593	597	593	
25-OH vitamin D, nmol/L, median (range)	30.4 ( $\leq$ 40.2)	48.4 (40.3–56.3)	64.0 (56.4–71.9)	80.5 (72.0–91.6)	109.0 ( $\geq$ 91.8)	
Age, y (SD)	59.9 (12.1)	62.7 (12.1)	63.3 (11.5)	65.8 (11.5)	66.5 (11.6)	<0.001
Male, % (n)	65.5 (390)	64.5 (385)	61.7 (366)	58.1 (347)	51.8 (307)	<0.001
BMI, kg/m <sup>2</sup> (SD)	28.8 (5.9)	29.0 (6.1)	28.8 (5.7)	28.0 (5.6)	27.3 (5.7)	<0.001
Current smoker, % (n)	22.5 (134)	18.6 (111)	17.2 (102)	13.9 (83)	12.1 (72)	<0.001
Prior smoker, % (n)	27.1 (161)	30.5 (182)	34.6 (205)	32.7 (195)	34.6 (205)	0.005
Renal disease, % (n)	6.7 (40)	4.9 (29)	3.9 (23)	4.5 (27)	4.2 (25)	0.055
Hypertension, % (n)	66.6 (396)	70.5 (421)	69.0 (409)	67.3 (402)	68.6 (407)	0.873
Hyperlipidemia, % (n)	68.1 (405)	69.2 (413)	65.4 (388)	64.8 (387)	65.1 (386)	0.092
Type 2 diabetes mellitus, % (n)	37.3 (222)	30.2 (180)	23.8 (141)	23.8 (142)	21.4 (127)	<0.001
Family history of heart disease, % (n)	30.0 (178)	31.2 (186)	34.1 (202)	31.7 (189)	32.4 (192)	0.369
Prior myocardial infarction, % (n)	8.1 (48)	7.2 (43)	7.8 (46)	9.1 (54)	7.4 (44)	0.873
Congestive heart failure, % (n)	14.0 (83)	12.6 (75)	9.8 (58)	13.4 (80)	13.5 (80)	0.985
Ejection fraction missing, % (n)	23.5 (140)	23.5 (140)	19.6 (116)	27.3 (163)	28.5 (169)	0.014
Ejection fraction <50, % (n)*	21.3 (97)	22.1 (101)	17.6 (84)	21.7 (94)	19.1 (81)	0.416
Duke5 score, score (SD) <sup>†</sup>	1.9 (1.2)	1.9 (1.1)	1.8 (1.1)	1.8 (1.1)	1.8 (1.2)	0.048
Season of 25-OH vitamin D testing						
Spring, % (n)	22.5 (134)	27.1 (162)	24.1 (143)	26.1 (156)	24.6 (146)	0.569
Summer, % (n)	19.5 (116)	25.3 (151)	24.8 (147)	24.0 (143)	25.5 (151)	0.055
Fall, % (n)	25.9 (154)	26.5 (158)	25.3 (150)	28.1 (168)	28.2 (167)	0.278
Winter, % (n)	31.8 (189)	21.1 (126)	25.8 (153)	21.8 (130)	21.8 (129)	<0.001

25-OH vitamin D indicates 25-hydroxyvitamin D; BMI, body mass index; Duke5 scale, modified 5-point Duke severity scale; SD, Standard deviation.

\*Ejection fraction was coded as a 4-point scale.

<sup>†</sup>Duke5 score is a 5-point scale.

Continuous covariates were dichotomized according to the median prior to testing, and strata-specific associations were shown for any significant interactions.

The ability of models to discriminate between patients who died versus those who did not was evaluated using the concordance probability estimate (C-metric).<sup>22</sup> Model fit was assessed using the Akaike Information Criterion (AIC), a measure that assigns a penalty for additional variables in a model. Calibration was evaluated using the Greenwood-Nam-D'Agostino goodness-of-fit test.<sup>23</sup> Risk reclassification was assessed using net reclassification improvement (NRI)<sup>24</sup> from a model containing demographic and clinical covariates versus a model also containing 25-OH vitamin D. The NRI describes (1) the proportion of cases (deaths) correctly reclassified to higher risk categories (case NRI=[number of cases reclassified higher–number of cases reclassified lower]/number of cases) and (2) the proportion of controls (survivors) correctly reclassified to lower risk categories (control NRI=[number of controls

reclassified lower–number of controls reclassified higher]/number of controls). An “overall” NRI can be calculated as the sum of both, however this cannot be expressed as a percentage because it is the sum of 2 fractions with different denominators.<sup>24</sup> Risk categories of 0% to <5%, 5% to <10%, 10% to <20%, and  $\geq$ 20% were selected for use in NRI calculations after physician consultation and literature review.<sup>25</sup> For calibration and net reclassification, we used predicted probabilities calculated at 5 years of follow-up.

All statistical analyses were performed using SAS version 9.4 (SAS Institute). Analyses were considered statistically significant at  $\alpha < 0.05$ .

## Results

The final data set contained 2975 patients (mean age 64 years, 60% male) catheterized between November 2007

and March 2012. Patients were followed for a median of 5.8 years (16 413 person-years) from 25-OH vitamin D measurement until death or censoring. There were 401 deaths, of which 44(11%) occurred  $\leq 30$  days after catheterization. The median time between 25-OH vitamin D testing and catheterization was 0.95 years, whereas the median time from catheterization until death or censoring was 4.4 years (12 945 person-years).

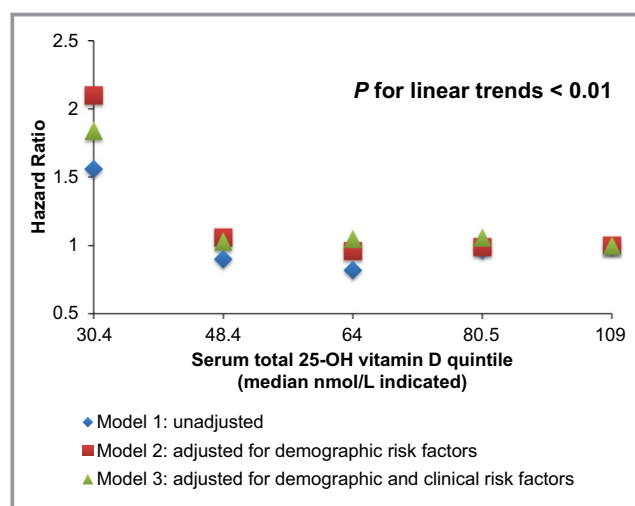
Increasing 25-OH vitamin D quintile was significantly associated with greater patient age; higher proportions of past smokers and patients without a measure of ejection fraction; lower BMI; lower proportions of men, current smokers, those with type 2 diabetes mellitus and those who were tested during the winter season (Table 1).

Compared with patients who were tested, the remaining 26 704 who were not tested were significantly younger, were more likely to be male and currently smoke (Table S1). They also had higher BMI; a greater prevalence of a family history of coronary heart disease, prior myocardial infarction, congestive heart failure, missing ejection fraction, and low ejection fraction; and lower prevalence of renal disease and type 2 diabetes mellitus. These patients also had higher Duke5 scores and longer follow-up after catheterization.

Serum total 25-OH vitamin D quintile was inversely associated with risk of mortality following coronary catheterization (hazard ratio 1.56 for bottom versus top quintile, 95% CI 1.17–2.08,  $P$  for trend  $<0.001$ ), which was driven by excess risk in the first quintile (Figure). The association was strengthened (hazard ratio 2.07, 95% CI 1.54–2.78,  $P$  for trend  $<0.001$ ) after adjusting for demographic risk factors (age, sex) but was weakened (hazard ratio 1.84, 95% CI 1.36–2.50,  $P$  for trend  $<0.001$ ) after further adjusting for clinical risk factors. In a sensitivity analysis, adjustment for medication use, including lipid-lowering agents, antiarrhythmia medications, blood pressure-lowering agents, anticoagulants, thrombolytic agents, antiplatelet agents, nitrates, insulin, and other blood sugar-lowering medications did not substantially change the association (data not shown). In addition, using the last 25-OH vitamin D test result prior to catheterization weakened the association slightly (data not shown).

The overall association between 25-OH vitamin D and mortality was attenuated among patients who were older, who had an ejection fraction  $<50\%$ , or who had a higher Duke5 score ( $P$  for interactions  $<0.05$ ) (Table 2). Interestingly, the association was not significantly affected by the time between testing and catheterization or season of testing (data not shown).

Despite being inversely associated with mortality, 25-OH vitamin D quintile had poor discriminative ability and model fit (c-metric 0.55, AIC 6212) compared with demographic and clinical risk factors together (c-metric 0.70, AIC 5924) (Table 3). Adding 25-OH vitamin D quintile to the model



**Figure.** Associations between serum total 25-OH vitamin D quintile and mortality. Median 25-OH vitamin D is shown for each quintile. Demographic risk factors were age and sex. Clinical risk factors were body mass index, smoking status (current, prior vs never), renal disease, hypertension, hyperlipidemia, type 2 diabetes mellitus, family history of heart disease, prior myocardial infarction, congestive heart failure, whether ejection fraction was measured, ejection fraction ( $<20\%$ ,  $20\text{--}34\%$ ,  $35\text{--}50\%$ ,  $>50\%$ ), and Duke5 score (0–5). The number of deaths in each quintile were 115 in quintile 1, 71 in quintile 2, 64 in quintile 3, 75 in quintile 4, and 76 in quintile 5. 25-OH vitamin D indicates 25-hydroxyvitamin D.

containing demographic and clinical risk factors only slightly improved overall discrimination and fit (c-metric 0.71, AIC 5908). All models were acceptably calibrated by the Greenwood-Nam-D'Agostino test ( $P \geq 0.05$ ); however, the model containing only 25-OH vitamin D quintile was the most well calibrated.

When 25-OH vitamin D quintile was added to the model containing demographic and clinical covariates, 20% of all patients were reclassified to a new 5-year risk category (Table 4). Among deaths, 9.0% were correctly reclassified to a higher risk category and 7.1% were incorrectly reclassified to a lower risk category, resulting in an NRI for deaths of 1.9% (Table 4). Among survivors, 11.3% were correctly reclassified to a lower risk category and 9.0% were incorrectly reclassified to a higher risk category, resulting in an NRI of 2.3% for survivors (Table 5). The overall NRI was 0.041 (Table 5).

## Discussion

In a population of patients undergoing their first coronary catheterization, higher pre-catheterization serum total 25-OH vitamin D was significantly associated with a lower risk of mortality, largely due to the excess risk in the bottom quintile. This association was attenuated by several competing risk factors. Considering 25-OH vitamin D in addition to



**Table 2.** Significant Interactions Among Serum Total 25-Hydroxyvitamin D Quintiles and Demographic and Clinical Risk Factors

Variable	Quintile, Hazard Ratio (95% CI)			P for Trend	P for Interaction
	1	3	5		
<b>Age*</b>					
≥63 y	1.28 (0.88–1.86)	0.92 (0.63–1.36)	1.0	0.604	<0.002
<63 y	3.17 (1.68–5.97)	1.48 (0.72–3.07)	1.0	<0.001	
<b>Ejection fraction</b>					
≥50%	2.03 (1.24–3.30)	0.85 (0.49–1.47)	1.0	0.010	0.026
<50%	0.89 (0.42–1.90)	1.41 (0.68–2.91)	1.0	0.446	
<b>Duke5 score*</b>					
≥3	1.48 (1.03–2.13)	0.98 (0.65–1.48)	1.0	0.159	0.001
<3	2.98 (1.70–5.23)	1.15 (0.63–2.11)	1.0	<0.001	

Models are adjusted for demographic and clinical covariates except for the stratification variable. \*Continuous variables were dichotomized according to the median.

demographic and clinical risk factors only marginally improved prognostic value. Although we found similar associations observed in other studies of vitamin D deficiency, CVD and mortality risk, to our knowledge ours is the first to use risk-reclassification measures to evaluate the clinical utility of vitamin D status.

Vitamin D deficiency, defined by the Endocrine Society clinical practice guidelines as a serum total 25-OH vitamin D concentration <50 nmol/L (<20 ng/mL),<sup>26</sup> is relatively common. In a systematic review of vitamin D status in 195 studies from 44 countries, 37% reported mean 25-OH vitamin D concentration <50 nmol/L.<sup>27</sup> Although vitamin D deficiency can lead to rickets in children and osteomalacia in adults,<sup>28</sup> a growing body of epidemiological evidence also implicates vitamin D deficiency as a risk factor for cancer, autoimmune

disorders, infectious diseases, and CVD as well as its risk factors such as type 2 diabetes mellitus.<sup>1,2,10,28</sup> Cardiovascular-related associations seem plausible given that the vitamin D receptor is present on numerous cardiovascular cell types and is involved in biochemical pathways responsible for blood pressure regulation, inflammation, and thrombosis<sup>29</sup>; however, these associations could be due to confounding. Vitamin D deficiency is associated with old age, poor-quality diet, sedentary lifestyle, obesity, and smoking, all of which are causally related to the development of cardiovascular risk factors and CVD itself.<sup>8,11,12</sup>

Even if 25-OH vitamin D is simply a risk marker correlated with causal risk factors, there remains strong interest in whether it can better identify high- and low-risk patients compared to established risk factors. In several prospective cohort studies of postsurgical or intensive care unit patients, 25-OH vitamin D was inversely associated with late discharge,<sup>16,17</sup> in-hospital death,<sup>18–20</sup> serious infections,<sup>18</sup> disease severity, and secondary CVD events.<sup>18</sup> Furthermore, in a study of 3371 patients undergoing coronary artery bypass grafting or valve surgery, 25-OH vitamin D was inversely associated with postoperative cardiac complications including in-hospital death, myocardial infarction, low output syndrome, and stroke.<sup>13</sup> In contrast, other studies have not found associations with postoperative mortality or cardiac comorbidities (eg, arrhythmias, low cardiac output) in cardiac surgery patients,<sup>14,15</sup> perhaps because of small sample sizes and presence of competing risk factors. Interestingly, serum 25-OH and 1,25 (OH)<sub>2</sub> vitamin D fluctuate during hospital stays<sup>16</sup> and following cardiac surgery,<sup>15,30</sup> likely due to fluid administration. This finding clearly supports measuring 25-OH vitamin D in the pre-procedural rather than peri- or post-procedural states, and may explain heterogeneity between studies.

**Table 3.** Measures of Model Performance

Model	Discrimination: C-Metric (SE)	Model Fit, AIC	Calibration,* Greenwood-Nam-D'Agostino Test, P Value (χ <sup>2</sup> )
25-OH vitamin D quintile only	0.55 (0.015)	6212	0.989 (0.30)
Demographic and clinical covariates only	0.70 (0.01)	5924	0.971 (2.95)
25-OH vitamin D plus demographic and clinical covariates	0.71 (0.01)	5908	0.058 (16.4)

Higher values of the c-metric indicate better discrimination between deaths and survivors. A lower AIC indicates better model fit. A nonsignificant (P≥0.05) value for the Greenwood-Nam-D'Agostino test indicates that observed and predicted outcomes do not differ significantly. 25-OH vitamin D indicates 25-hydroxyvitamin D; AIC, Akaike Information Criterion

\*Calibration was determined based on 5-year predicted probabilities.

**Table 4.** Reclassification of 5-Year Predicted Mortality Risk Among Deaths and Survivors When Adding Serum Total 25-OH Vitamin D to a Model Containing Clinical and Demographic Covariates

Model* Without 25-OH Vitamin D	Group	Model* With 25-OH Vitamin D				Total, % (n)
		0% to <5%	5% to <10%	10% to <20%	≥20%	
0% to <5%	Deaths, % (n)	68.8 (11)	31.3 (5)	0 (0)	0 (0)	4.2 (16)
	Survivors, % (n)	87.2 (510)	12.8 (75)	0 (0)	0 (0)	22.5 (585)
5% to <10%	Deaths, % (n)	10.0 (6)	68.3 (41)	21.7 (13)	0 (0)	15.9 (60)
	Survivors, % (n)	14.7 (122)	73.9 (614)	11.4 (95)	0 (0)	32.0 (831)
10% to <20%	Deaths, % (n)	0 (0)	7.8 (9)	78.5 (91)	13.8 (16)	30.7 (116)
	Survivors, % (n)	0 (0)	14.2 (108)	77.5 (588)	8.3 (63)	29.2 (759)
≥20%	Deaths, % (n)	0 (0)	0 (0)	6.5 (12)	93.5 (174)	49.2 (186)
	Survivors, % (n)	0 (0)	0 (0)	14.9 (63)	85.1 (359)	16.3 (422)
Total, % (n)	Deaths, % (n)	4.5 (17)	14.6 (55)	30.7 (116)	50.3 (190)	100 (378)
	Survivors, % (n)	24.3 (632)	30.7 (797)	28.7 (746)	16.3 (422)	100 (2597)

25-OH vitamin D indicates 25-hydroxyvitamin D.

\*Both models contained demographic and clinical covariates. A total of 20% of patients were reclassified to a new risk category when 25-OH vitamin D was added to the model. The total number of deaths considered in this analysis was 378 because 23 patients died after 5 years.

In our study, serum total 25-OH vitamin D measured prior to catheterization was inversely associated with mortality. Patients with a 25-OH vitamin D concentration in the bottom quintile had nearly twice the risk of death compared to those in the top quintile, even after adjusting for BMI. Adipose tissue, which is strongly associated with increased BMI, is a sink for fat-soluble 25-OH vitamin D<sup>8</sup> and is causally related to development of dyslipidemias, type 2 diabetes mellitus, high blood pressure, and inflammation.<sup>31</sup> Interestingly, this association remained significant after adjusting for other risk factors and possible mediators (eg, type 2 diabetes mellitus, hypertension, coronary artery disease severity) which could indicate that 25-OH vitamin D affects mortality risk through an independent pathway; however, because of a lack of information on diet, physical activity, and other correlated risk factors, this association is more likely due to residual and unmeasured confounding.

We found that this association was significantly attenuated in older patients, among those with a low ejection fraction or a high Duke5 score. This may be expected if other risk factors are more important than vitamin D status and tend to “wash out” associations attributed to 25-OH vitamin D. Another

explanation is simply that these patients are more likely to take vitamin D supplements which would result in exposure misclassification and attenuation of the association. Interestingly, elapsed time between 25-OH vitamin D testing and catheterization did not alter this association, suggesting that 25-OH vitamin D concentration is relatively stable over long periods of time; however, using the last 25-OH vitamin D measure before catheterization attenuated the overall association, which may be due to lifestyle modification, supplementation, and other changes initiated by patients after receiving results from the first test.

According to the American Heart Association, the prognostic value of novel risk markers should be evaluated in terms of model discrimination, fit, calibration, and particularly the ability to correctly reclassify cases and controls compared to established risk factors.<sup>32</sup> Nevertheless, few epidemiological studies of 25-OH vitamin D followed these recommendations. In our study, serum total 25-OH vitamin D was a poor discriminator of patients who died and those who survived. As well, adding 25-OH vitamin D to a model containing established demographic and clinical risk factors led to only minor improvements in model discrimination and fit. However, as the c-metric is insensitive to changes in absolute probability,<sup>33</sup> we calculated the NRI.<sup>24</sup> As NRIs for both deaths and survivors were <5%, this suggested that serum total 25-OH vitamin D provides little prognostic value above and beyond established cardiovascular risk factors.

Our study has several strengths. First, we examined only the first 25-OH vitamin D measurement occurring before the first catheterization to reduce the possibility that patients changed their diet, physical activity, or supplement use in

**Table 5.** Net Reclassification Improvement

Net Reclassification Improvement	Result
Deaths	1.9%
Survivors	2.3%
Overall	0.042

response to a diagnosis of vitamin D deficiency or CVD, which could bias associations. Second, we were able to evaluate the impact of and adjust for multiple risk factors and coronary artery disease severity in our models. Third, to our knowledge, our study is the first to use reclassification measures to evaluate the prognostic value of serum total 25-OH vitamin D compared with established risk factors. Fourth, we added depth to a well-established and long-running cardiac catheterization registry by linking to provincial repositories of laboratory data containing abundant 25-OH vitamin D test data.

Our study also has some limitations. First, not all APPROACH patients were tested for 25-OH vitamin D; therefore, our findings may not be generalizable to all patients receiving coronary catheterization. Associations were slightly stronger in younger and healthier participants who made up a larger proportion of untested patients; however, these small differences are unlikely to change our overall findings. Second, we had no control over when patients were tested for 25-OH vitamin D relative to catheterization; however, the association of 25-OH vitamin D and mortality did not significantly change according to time between testing and catheterization. Third, although cause of death was not available in our data, a large proportion of patients would have died from cardiovascular causes. Fourth, although there is likely residual and unmeasured confounding in our study because of individual differences in sun exposure, physical activity, diet, or supplement use, we attempted to limit the effect of lifestyle changes that might occur after a diagnosis of vitamin D deficiency or catheterization by including only the first 25-OH vitamin D test result prior to the first catheterization.

## Conclusions

Pre-catheterization serum total 25-OH vitamin D was inversely associated with mortality risk after adjusting for established demographic and clinical risk factors. This association was attenuated by several competing risk factors. Overall, 25-OH vitamin D added little prognostic value over established risk factors; therefore, its measurement is not warranted in patients undergoing coronary catheterization.

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Gerling acquired, analyzed, and interpreted the data, drafted the manuscript and performed statistical analysis. James, Wilton, Naugler contributed to design of the study, interpreted data, revised the manuscript and provided supervisory support to Gerling. Southern contributed to the acquisition of data, revisions of the manuscript and statistical analysis. Galbraith contributed to design of the study, revisions of the manuscript and administrative support. Knudtson

contributed to the interpretation of data, administrative support and obtained funding. de Koning conceived of the idea for this study, interpreted the data, revised the manuscript, provided statistical support and supervised Gerling. de Koning had full access to all the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis. We thank Mei Zhang and Zhi Tan for additional statistical support, and Dr Alex Chin for sharing 25-OH vitamin D method comparison data. All authors declare no conflicts of interest. Written permission has been obtained from all persons named in the acknowledgments.

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## Disclosures

None.

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## Supplemental Material

**Table S1. Patient characteristics according to 25-OH vitamin D testing status**

Predictors			p for
	Yes (n=2975)	No (n=26704)	difference
Age, y (sd)	63.6 (12.0)	62.8 (12.5)	<0.001
Male, % (n)	60.3 (1795)	72.1 (19246)	<0.001
BMI, kg/m <sup>2</sup> (sd)	28.4 (5.8)	29.1 (5.7)	<0.001
Current smoker, % (n)	16.9 (502)	29.5 (7886)	<0.001
Prior smoker, % (n)	31.9 (948)	31.3 (8352)	0.511
Renal Disease, % (n)	4.9 (144)	3.9 (1032)	0.010
Hypertension, % (n)	68.5 (2035)	68.3 (18234)	0.893
Hyperlipidemia, % (n)	66.6 (1979)	66.7 (17834)	0.773
Type 2 Diabetes, % (n)	27.3 (812)	23.0 (6127)	<0.001
Family history of heart disease, % (n)	31.8 (947)	34.3 (9173)	0.006
Prior Myocardial Infarction, % (n)	7.9 (235)	9.9 (2631)	<0.001
Congestive Heart Failure, % (n)	12.7 (376)	14.1 (3772)	0.027
Ejection fraction missing, % (n)	24.5 (728)	30.1 (7986)	<0.001
Ejection fraction <50, % (n)	20.4 (457)	28.5 (5343)	<0.001
Duke5 score, score (sd)	1.8 (1.3)	2.0 (1.1)	<0.001
<b>Outcomes</b>			
Crude mortality rate, % (n)	13.5 (401)	13.6 (3647)	0.872
Median follow-up time from catheterization, y (sd)	4.4 (1.4)	5.0 (1.8)	<0.001

Continuous variables are presented as mean (standard deviation [SD]) unless otherwise indicated.