Vitamin D status and metabolic syndrome in the elderly: the Rotterdam Study

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Abstract

Objective: The effects of vitamin D in the elderly are inconsistent. The aim of this study was to evaluate the association between vitamin D status and the metabolic syndrome (MetS) in the elderly, as well as between vitamin D status and the components of MetS (i.e. serum glucose, triglycerides (TG), HDL cholesterol (HDL-C), waist circumference (WC), and blood pressure (BP)).

Methods: The study was embedded in the Rotterdam Study, a population-based cohort of middle-aged and elderly adults. We analyzed data from 3240 people (median age 71.2 years) who did not have type 2 diabetes mellitus at baseline.

Results: We found higher 25-hydroxyvitamin D (25(OH)D) concentrations associated with lower prevalence of MetS (odds ratio (OR); 95% CI: 0.61; 0.49, 0.77 for adequate levels ($\geq$75 nmol/l) vs deficiency ($<$50 nmol/l). In addition, in analysis of the individual components, the ORs for adequate vs deficient vitamin D levels were: 0.66 (95% CI 0.53, 0.83) for elevated WC, 0.67 (95% CI 0.52, 0.86) for reduced HDL-C, 0.69 (95% CI 0.54, 0.88) for elevated TG, and 0.80 (95% CI 0.65, 0.99) for elevated fasting glucose. Vitamin D was not associated with elevated blood pressure, and ORs for adequacy vs deficiency were 0.82 (95% CI 0.65, 1.03).

Conclusion: Higher 25(OH)D concentrations in the elderly are associated with lower prevalence of MetS and, in particular, with more beneficial HDL-C, TG, WC, and serum glucose. Since the prevalence of vitamin D deficiency is common worldwide and its risk increases with age, if causality is proven, benefits of improving vitamin D status among the elderly may be great.

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Introduction

Vitamin D was discovered at the beginning of the 20th century, when it presented a long-awaited cure for rickets, a childhood disease consisting of weak bones (1). Since that time, vitamin D has been established as a major factor influencing metabolism of bones and calcium metabolism (1, 2).

Active vitamin D binds to a receptor from the nuclear family called vitamin D receptor (VDR), which, subsequently, increases transcription of its target genes. Recently, scientists have discovered that the enzyme needed for the activation of vitamin D, 1α hydroxylase, is present in most cells and tissues in the human body, as are VDRs. This discovery led to additional research into other vitamin D effects beside calcium metabolism and bone health. Currently, we know there are numerous binding sites for vitamin D in the human genome, suggesting a wide range of vitamin D effects (3).

However, the cutoff points for adequate vitamin D status are still being debated. Some experts consider serum 25(OH)D concentrations higher than 75 nmol/l as adequate not just for bone health, but for the non-skeletal effects as well (3, 4). Others suggest that serum 25(OH)D concentrations of 50 nmol/l are sufficient (5).

Metabolic syndrome (MetS) is defined as a cluster of risk factors for cardiovascular disease and type 2 diabetes mellitus (DM) (6). Several commonly used

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definitions of MetS have been proposed and (6), although these definitions differ, they generally agree on the following criteria for the diagnosis of MetS: central obesity, dyslipidemia (elevated triglycerides (TG) and reduced HDL cholesterol (HDL-C)), hypertension, and hyperglycemia (6).

The association between vitamin D status and MetS has previously been studied in different populations (7, 8). Animal models and *in vitro* studies provide an insight into mechanisms (9, 10) of vitamin D metabolic actions, while meta-analyses of epidemiological studies confirm inverse associations between serum vitamin D status and MetS (7, 8). However, the most of the studies published on this topic were conducted in younger populations (11), and subgroup analyses of data from the elderly (aged 65 years and older) are not conclusive and two of the studies focusing on this age group yielded contradictory results (12, 13). In addition, there has not been sufficient attention paid for finding the specific groups among the elderly who may benefit the most from improved vitamin D status (e.g. gender or those with obesity or impaired kidney function).

Thus, our study aimed to evaluate whether vitamin D status is associated with risk of MetS in the elderly. Furthermore, we aimed to evaluate the association of vitamin D status with individual components of the MetS and to assess whether the association differed by age, gender, BMI, and kidney function. Finally, we assessed which, if any, of the associations between vitamin D status and individual components of MetS were the strongest.

Subjects and methods

Study design

This study was embedded in the Rotterdam Study, a large prospective population-based cohort study conducted among residents in Ommoord, a district of Rotterdam, The Netherlands (14). These participants were aged 55 years or older at baseline. Of 10 275 eligible subjects, 7983 (78%) participated in the baseline examinations between 1989 and 1993. All participants were interviewed at home and visited the research center for further examinations. Additional follow-up visits were conducted every 3–4 years.

The study was approved by the medical ethics committee at Erasmus University Rotterdam, The Netherlands and written informed consent was obtained from all participants.

25-hydroxyvitamin D

Between 1997 and 1999, in the third survey of the cohort described previously, serum 25 hydroxivitamin D concentrations were assessed using electrochemiluminescence immunoassay (COBAS, Roche Diagnostics GmbH). The sensitivity of the test was 10 nmol/l and serum 25(OH)D concentrations detected were within range of 7.5 and 175 nmol/l. Within-run precision was $<7.8\%$ and the inter mediate precision was $<13.1\%$. Concentrations of serum 25(OH)D under 50 nmol/l were categorized as 'deficient', and concentrations of serum 25(OH)D from 50 to 75 nmol/l as 'insufficient,' according to cutoffs described by Holick (3).

MetS and its components

To define MetS, we used the joint interim statement of the International Diabetes Federation Task Force on Epidemiology and Prevention; the National Heart, Lung, and Blood Institute; the American Heart Association; the World Heart Federation; the International Atherosclerosis Society; and the International Association for the Study of Obesity (6). According to this definition, an individual had MetS if three out of five of the following conditions are met: i) waist circumference ≥ 102 cm for men and ≥ 88 cm for women; ii) elevated TG (≥ 1.7 mmol/l) or alternatively drug treatment for elevated TG; iii) reduced concentration of HDL-C (≤ 1.0 mmol/l in males and ≤ 1.3 mmol/l in females) or, alternatively, active drug treatment for reduced HDL-C; iv) elevated blood pressure (systolic ≥ 130 mmHg and/or diastolic ≥ 85 mmHg) or, alternatively, active antihypertensive drug treatment in a patient with a history of hypertension; v) elevated fasting glucose (≥ 100 mg/dl) or active drug treatment of hyperglycemia (6). All components of MetS were assessed during the third survey of the Rotterdam Study cohort (1997–1999).

Waist circumference was measured half way between the lower rib margin and the iliac crest and presented in centimeters. During measurement, participants were standing, wearing only light clothes, breathing gently.

Serum glucose was measured by hexokinase enzymatic method after overnight fasting. Fasting serum glucose was not assessed in subjects with diagnosis of type 2 diabetes mellitus. Hence, these subjects were excluded from the analysis ($n=588$).

TG were measured in fasting serum using enzymatic method. HDL-C was measured by automatic enzymatic method from fasting serum after precipitation of non-HDL fraction. Blood pressure was measured at the right brachial

artery using random-zero sphygmomanometer after 5 min of rest with the participants in sitting position. Mean of the two consecutive measurements was used.

Confounders and effect modifiers

At the baseline cohort visit (1989–1993), information on multiple factors was collected. Trained interviewers conducted home interviews, after which participants were invited to the research center for the clinical examination and laboratory tests. In our analyses, we used the following information collected at that time point: diet, family history of diseases, educational level, and household income.

Family history of diseases included following diseases in siblings, children, or parents: diabetes mellitus, myocardial infarction, and cerebrovascular accident. Level of education was split into low (primary education or less) and high (more than primary education). The cutoff value of 2699 euro per month (35 999 euro per year) was used to assess household income. Income below cutoff was considered as low household income, and everything above the cutoff as high household income. In order to adjust for overall dietary quality, the Dutch Healthy Diet (DHD) Index was used. DHD Index is a representative continuous score of dietary compliance to the Dutch Guidelines for a Healthy Diet, which has been assessed from the FFQ assessed at baseline and has been described in detail elsewhere (15).

Information on following factors we used in our analyses was collected during the third visit of the cohort participants (1997–1999) to the research center: weight, height, prevalent cardio-metabolic diseases, serum creatinine, smoking, and physical activity.

BMI was calculated by dividing body weight (kg) by height squared (m^2). Serum creatinine was measured using an enzymatic assay method and used to calculate estimated glomerular filtration rate (eGFR) with simplified modification of diet in renal disease (MDRD) formula. Chronic kidney disease was defined as eGFR <60 ml/min per 1.73 m^2. Cardio-metabolic baseline diseases (values: 0=no and 1=yes, subjects with diabetes mellitus were excluded) were identified on the basis of the presence of at least one of the following: heart failure, coronary heart disease, atrial fibrillation, cerebrovascular accident, and chronic kidney disease. Smoking status was assessed on the basis of three categories: non-smoker, former smoker, and current smoker. Physical activity levels (minutes/week) were assessed using a validated questionnaire of the Zutphen Study (16). Originally, the questionnaire provided information about walking, cycling, gardening, diverse sports and hobbies hence information about outdoors activities. Later, questions about housekeeping activities were also included.

Population for analysis

The initial study population consisted of 7983 participants. We used the third visit of the cohort (1997–1999) in our analyses. A subsample of the initial population had serum 25(OH)D measured at the third visit ($n=3828$). We excluded participants with diabetes mellitus that had been ascertained at the first visit or had been diagnosed between the first (1989–1993) and the third visit of the cohort (1997–1999). These participants had no fasting blood samples drawn at the third visit due to risk of hypoglycemia. After exclusion of participants with previously identified type 2 diabetes mellitus diagnosis, 3240 participants with serum 25(OH)D measurements were available for analysis, including those with pre-diabetes and insulin resistance syndrome.

Statistical analyses

To determine the association between vitamin D status and prevalence of MetS (and each of its components) we used logistic regression models.

Firstly, a crude model was built adjusted for age and sex. Secondly, a multivariate model was built, further adjusted for potential confounders. The confounders selected based on previously published literature, univariate inspection based on 10% change in the effect estimate, and association with the outcome and/or the exposure were: dietary quality score, baseline diseases, family history of diseases, physical activity, season when blood was drawn, level of education, and household income (17, 18). Serum 25(OH)D concentration was used as continuous variable and also as categorical variable (cutoff points described by Holick (3)).

We further tested whether the associations differed by age, gender, eGFR, and BMI. In case of significant effect modification, stratified analyses were performed ($P_{\text{interaction}} <0.05$).

In addition, we tested which of the associations between vitamin D status and individual components of MetS were independent from the rest of the MetS components. For this purpose, we used multivariate model further adjusted for the rest of the components of MetS, tested individually.

Finally, to assess the effect of BMI on the association between vitamin D status and MetS, and the effect of BMI on the association between vitamin D status and individual components of MetS, we further adjusted the multivariate model to include BMI.

To reduce the potential for any biases associated with missing data, a multiple imputation procedure was performed, $n=10$ imputations (Supplementary Table 1, see section on supplementary data given at the end of this article). The multiple imputation procedure is based on prediction of missing data based on correlation with other observed data (Supplementary Table 1, see section on supplementary data given at the end of this article). The missing data are imputed with randomly selected values from the predicted distribution. In the next step, ten different copies of the original data set with missing data imputed by this random selection of predicted values are created. The analyses are performed separately in all of the ten datasets and pooled results from these ten imputed datasets are reported. Final results are presented after the multiple imputation procedure.

Main results are presented as odds ratios (ORs) and 95% CIs for MetS and for components of MetS. A P value <0.05 was considered to be statistically significant. All analyses were performed with IBM SPSS Statistics version 20 (SPSS, Inc.).

Sensitivity analyses were performed using different cutoff points for serum 25(OH)D concentrations as suggested by the Institute of Medicine (19). According to these cutoffs, 25(OH)D concentrations <40 nmol/l were defined as covering the needs of half of the population, while concentrations of 50 nmol/l were defined as covering the needs of 97.5% of the population (19). Additional sensitivity analysis included use of alternative cutoff points for waist circumference being 94 cm for men and 80 cm for women (6).

Results

Table 1 presents the characteristics of our study population by vitamin D status. Only 16% of our study participants had adequate vitamin D status (serum 25(OH)D concentrations of 75 nmol/ and higher), 27% had a vitamin D insufficiency (serum 25(OH)D concentrations between 50 and 75 nmol/l), and 57% had a vitamin D deficiency (serum 25(OH)D concentrations

Table 1 Baseline (RS-I-3) characteristics of the study population according to serum 25(OH)D. Prevalent diabetes cases were excluded in the analyses. Data presented after multiple imputation procedure.

	Deficiency (<50 nmol/l)	**Insufficiency** (50–75 nmol/l)	**Adequate** (≥75 nmol/l)	
	$n=1833$ (56.6%)	$n=874$ (27.0%)	$n=533$ (16.4%)	***P* value**
Serum 25(OH)D[a] (nmol/l)	31.3 (10.8)	61.2 (7.0)	93.3 (15.4)	<0.001
Metabolic syndrome (yes) *n* (%)	766 (41.8)	286 (32.7)	158 (29.6)	<0.001
Age[a] (years)	74.1 (7.6)	70.3 (5.8)	69.5 (5.9)	<0.001
Gender (female) *n* (%)	1244 (67.9)	459 (52.5)	226 (42.4)	<0.001
Glucose in serum[a] (mmol/l)	5.54 (0.59)	5.52 (0.56)	5.50 (0.54)	0.273
HDL cholesterol[a] (mmol/l)	1.42 (0.41)	1.43 (0.39)	1.39 (0.38)	0.082
Total cholesterol[a] (mmol/l)	5.85 (1.02)	5.87 (0.93)	5.83 (0.99)	0.699
Triglycerides[a] (mmol/l)	1.49 (0.70)	1.41 (0.62)	1.42 (0.67)	0.005
Lipid lowering medication (yes) *n* (%)	254 (13.9)	110 (12.6)	68 (12.8)	0.902
Systolic blood pressure[a] (mmHg)	144 (21)	141 (20)	141 (21)	0.001
Diastolic blood pressure[a] (mmHg)	75 (12)	75 (11)	76 (10)	0.107
Blood pressure lowering medication (yes) *n* (%)	428 (23.3)	184 (21.0)	89 (16.7)	0.001
Cardiovascular diseases (yes) *n* (%)	378 (20.6)	138 (15.8)	78 (14.6)	0.001
Waist circumference[a] (cm)	93.1 (11.8)	92.3 (10.7)	92.4 (10.6)	0.204
BMI[a] (kg/m^2)	26.9 (4.1)	26.3 (3.4)	26.1 (3.4)	<0.001
Physical activity[a] (min/week)	2558 (1150)	2699 (1094)	2753 (1084)	<0.001
DHDI-score[a]/[b]	49.0 (10.1)	48.8 (10.2)	47.9 (10.0)	0.094
Smoking (current) *n* (%)	403 (22.0)	187 (21.4)	111 (20.8)	<0.001
Family history of diseases (yes)[b] *n* (%)	1212 (66.1)	565 (64.6)	343 (64.4)	0.831
Education (low)[b] *n* (%)	618 (33.7)	229 (26.2)	124 (23.3)	<0.001
Income (below the average)[b] *n* (%)	1035 (56.5)	398 (45.5)	233 (43.7)	<0.001

[a]Mean (S.D.).
[b]Also assessed at the previous visit to the research center (1989–1993).

Table 2 Vitamin D status and prevalence of metabolic syndrome. Crude model adjusted for age and sex only. Multivariate model is adjusted for age, sex, physical activity, diet quality score, family history of cardio-metabolic diseases, baseline cardio-metabolic diseases, smoking, education, income, season of blood draw, and year or blood draw. Prevalent diabetes cases were excluded in the analyses. Data presented after multiple imputation procedure.

	n	**Crude model OR** (95% CI)	**Multivariate model OR** (95% CI)
Serum 25(OH)D continuous	3240	0.91 (0.88, 0.94)*	0.90 (0.87, 0.93)*
Serum 25(OH)D cut-offs (nmol/l)			
<50	1833	Reference	Reference
50–75	874	0.72 (0.60, 0.86)*	0.70 (0.58, 0.84)*
≥75	533	0.65 (0.52, 0.81)*	0.61 (0.49, 0.77)*

*P value <0.001. OR, odds ratio (for continuous serum 25(OH)D per ten unit serum 25(OH)D increase).

lower than 50 nmol/l). Participants with adequate vitamin D status were younger and more likely to be men (Table 1). Also, among participants who were vitamin D deficient, there was a greater prevalence of MetS and these participants were also likely to have high blood pressure. However, there were no differences in mean waist circumference, serum glucose, HDL-C and total cholesterol across serum 25(OH)D strata (Table 1).

Vitamin D status and prevalence of MetS and its components

Every 10 nmol/l increase in serum 25(OH)D was significantly associated with a lower prevalence of MetS (OR=0.91, 95% CI 0.88, 0.94 and OR=0.90, 95% CI 0.87, 0.93 for crude model and multivariate model respectively) among our study participants. Both insufficient and adequate categories of vitamin D status were associated with lower prevalence of MetS when compared with those with a vitamin D deficiency (Table 2).

Regarding the individual components of the MetS, vitamin D status was significantly associated with lower prevalence of elevated WC (multivariate adjusted OR=0.92, 95% CI 0.89, 0.95), lower prevalence of elevated TG (multivariate adjusted OR=0.93, 95% CI 0.89, 0.96), lower prevalence of reduced HDL-C (multivariate adjusted OR=0.93, 95% CI 0.90, 0.97), and lower prevalence of elevated fasting glucose (multivariate adjusted OR=0.96, 95% CI 0.93, 0.99) (Table 3). We also found an association between vitamin D status and lower prevalence of elevated blood pressure (multivariate adjusted OR=0.96, 95% CI 0.93, 1.00); however, this finding was no longer significant when assessing the cutoffs (Table 3).

Furthermore, we found WC to be significantly associated with serum 25(OH)D independently of the other MetS components, whereas no independent association was found for the other MetS components after mutual adjustment (Table 4).

After additional adjustment for BMI, higher serum 25(OH)D concentrations were still significantly associated with lower odds of MetS. Furthermore, WC was significantly associated with serum 25(OH)D only when serum 25(OH)D was assessed continuously. The effect estimates for TG and HDL-C were slightly attenuated; however, direction of the effects remained the same (Supplementary Table 2, see section on supplementary data given at the end of this article).

Table 3 Vitamin D status and individual components of metabolic syndrome. Multivariate model is adjusted for age, sex, physical activity, diet quality score, family history of cardio-metabolic diseases, baseline cardio-metabolic diseases, smoking, education, income, season of blood draw, and year of blood draw. Prevalent diabetes cases were excluded in the analyses. Data presented after multiple imputation procedure.

		Multivariate model OR (95% CI)				
	n	Elevated waist circumference	Elevated triglycerides	Reduced HDL cholesterol	Elevated glucose	Elevated blood pressure
Serum 25(OH)D continuous	3240	0.92 (0.89, 0.95)*	0.93 (0.89, 0.96)*	0.93 (0.90, 0.97)*	0.96 (0.93, 0.99)	0.96 (0.93, 1.00)
Serum 25(OH)D cut-offs (nmol/l)						
<50	1833	Reference	Reference	Reference	Reference	Reference
50–75	574	0.76 (0.63, 0.91)	0.74 (0.61, 0.90)	0.76 (0.62, 0.93)	0.94 (0.79, 1.12)	0.98 (0.80, 1.20)
≥75	533	0.66 (0.53, 0.83)*	0.69 (0.54, 0.88)	0.67 (0.52, 0.86)	0.80 (0.65, 0.99)	0.82 (0.65, 1.03)

*P value <0.001. OR, odds ratio (for continuous serum 25(OH)D per ten unit serum 25(OH)D increase).

Table 4 Vitamin D status and individual components of metabolic syndrome after mutual adjustment of the other MetS components. Multivariate model is adjusted for age, sex, physical activity, diet quality score, family history of cardio-metabolic diseases, baseline cardio-metabolic diseases, smoking, education, income, season of blood draw, year of blood draw plus additionally adjusted for the rest of the individual components of the MetS; for example: for WC as an outcome model was adjusted for TG, HDL, glucose and BP. Prevalent diabetes cases were excluded in the analyses. Data presented after multiple imputation procedure.

		Multivariate model OR (95% CI)				
	n	Elevated waist circumference	Elevated triglycerides	Reduced HDL cholesterol	Elevated glucose	Elevated blood pressure
Serum 25(OH)D continuous	3240	0.93 (0.90, 0.97)*	0.96 (0.92, 1.00)	0.97 (0.93, 1.01)	0.99 (0.96, 1.02)	0.98 (0.94, 1.01)
Serum 25(OH)D cut-offs (nmol/l)						
<50	1833	Reference	Reference	Reference	Reference	Reference
50–75	874	0.82 (0.68, 0.99)	0.90 (0.72, 1.12)	0.89 (0.71, 1.11)	1.05 (0.88, 1.26)	1.04 (0.85, 1.27)
≥75	533	0.71 (0.57, 0.90)	0.82 (0.63, 1.07)	0.78 (0.59, 1.04)	0.91 (0.73, 1.13)	0.89 (0.70, 1.12)

*P value <0.001. OR, odds ratio (for continuous serum 25(OH)D per ten unit serum 25(OH)D increase); WC, waist circumference; TG, triglycerides; HDL-C, HDL cholesterol; BP, blood pressure.

Subgroup analysis

We found significant effect modification by gender ($P_{\text{interaction}}$ <0.05).

In women, higher serum 25(OH)D concentrations were significantly associated with lower prevalence of elevated TG (data not shown), while lower prevalence of MetS was significantly associated with higher serum 25(OH)D in both men and women, though results were stronger among women (data not shown).

Sensitivity analysis

We conducted sensitivity analyses using alternative cutoff points for WC as suggested by Alberti *et al.* (6). For this analysis, we considered a WC value of ≥94 cm to be elevated for men, and a WC value of ≥80 cm to be elevated for women. The effect estimates of this analysis were similar for both groups (data not shown). We also evaluated whether the use of different cutoff points for serum 25(OH)D, as suggested by the Institute of Medicine, would have any impact on our findings (20, 21), but the use of the IoM cutoff points yielded similar effect estimates (Supplementary Tables 3 and 4, see section on supplementary data given at the end of this article).

Discussion

Main results

We found that vitamin D status, measured by serum 25(OH)D concentrations, was inversely associated with the prevalence of MetS in the elderly. This association was mainly driven by elevated waist circumference. In particular, we found that vitamin D status was inversely associated with the prevalence of elevated serum TG, elevated WC, reduced HDL-C levels, and elevated glucose levels. No significant association was found between serum 25(OH)D and blood pressure.

Comparison with other studies

Other previous studies have found an inverse association between vitamin D status and MetS (4, 7, 8, 11) and our estimates fall within the range reported. However, the majority of the associations reported elsewhere were observed in younger populations (7, 8), such as the findings from a longitudinal study conducted among Australian adults by Gagnon *et al.* (11). They found that lower concentrations of serum vitamin D status were associated with increased risk of MetS (11). Within elderly populations, we identified just two cross-sectional studies which reported the association between vitamin D status and MetS: the Rancho Bernardo Study (12) and the Longitudinal Ageing Study Amsterdam (LASA) (13). The LASA study found a significant association between higher serum 25(OH)D (>50 nmol/l) and lower prevalence of MetS. Similar to our results, the LASA study also found a significant association with HDL-C and WC. The Rancho Bernardo Study (mean age >75 years) found no significant association between serum 25(OH)D and MetS, but they did find a significant association between serum 25(OH)D and glucose levels, though that finding was in men only. The discrepancy between our findings and those of The Rancho Bernardo study may be explained by the

relatively high serum 25(OH)D concentrations in the population of The Rancho Bernardo Study. In addition, this study took place in California where high exposure to UVB radiation caused serum 25(OH)D concentrations to be much higher than the mean concentration in the US elderly population in general (22) (in The Rancho Bernardo Study the mean serum 25(OH)D concentration was above 100 nmol/l with the bottom quintile of 87.5 nmol/l). It was hypothesized that there are different thresholds up to which vitamin D can have beneficial effects while raising serum 25(OH)D concentrations above the threshold produces small or no additional benefits (11, 13, 23, 24, 25).

The results we found are more closely aligned with those from LASA, where an association between vitamin D status and prevalence of MetS was found, odds ratio of 1.54 for serum 25(OH)D concentrations below 50 nmol/l vs above 50 nmol/l. These results were comparable with those we report here: multivariate adjusted odds ratio of 0.61 for serum 25(OH)D concentrations $\geq$75 nmol/l vs $<$50 nmol/l.

Based on results from these two studies and our results, we speculate that there might be a certain threshold effect of vitamin D status on MetS. Raising serum 25(OH)D concentrations above a certain threshold (e.g., $>$75 nmol/l) adds little effect.

Potential mechanisms

The association between vitamin D status and MetS and its components can be explained in several ways. Firstly, vitamin D influences formation of HDL particles (26, 27, 28, 29, 30). Secondly, serum 25(OH)D inhibits adipocyte differentiation and may thereby influence the development of adiposity (9). Also, vitamin D deficiency causes an increase in parathyroid hormone (PTH), which is known to favor the process of lipid storage (31, 32). Thirdly, serum 25(OH)D regulates an enzyme directly involved in lipoprotein mechanism – lipoprotein lipase (LPL) (33). Fourthly, the active form, serum 1,25-dihydroxyvitamin D, acts as a suppressor of the renin–angiotensin system (RAAS) (34, 35, 36). Finally, angiotensin II has been shown to cause insulin resistance and, since vitamin D inhibits RAAS, it might indirectly improve insulin sensitivity (37, 38). Also, vitamin D helps insulin secretion from pancreatic beta cells, it enhances insulin sensitivity by stimulating the expression of insulin receptors, and it is involved in the regulation of intracellular Ca^{2+} (39).

Subgroup analysis

We found an effect modification by gender. We found vitamin D status significantly associated with a lower prevalence of elevated TG in women. However, vitamin D status was inversely associated with MetS in men and in women. The effect magnitude was slightly greater in women than in men. These results may be explained by the increased risk of both MetS and vitamin D deficiency in women (3, 20).

Overall, the subgroup analysis suggests that the effect of serum 25(OH)D may differ in magnitude among risk groups for both MetS and vitamin D deficiency. Also, it seems that women might benefit more from adequate vitamin D status. These findings warrant further study in the future.

Strengths and limitations

The main strengths of our study were its prospective design and the extensive records of population characteristics. However, some limitations need to be taken into account as well. Firstly, we did not assess PTH levels, which is an important factor related to serum 25(OH)D and calcium metabolism. Secondly, we were not able to exclude patients with MetS before serum 25(OH)D assessment because we had not measured triglyceride levels and we had no fasting blood samples at that time. Although we excluded subjects who had type 2 diabetes mellitus before serum 25(OH)D assessment, reverse causality cannot be fully ruled out (reverse causality occurs when the outcome is related to the exposure being studied. Specifically, the participant's ill health could cause low serum 25(OH)D concentrations rather than the other way around, which we investigated). Thirdly, some of the population characteristics we used in our analysis were recorded at the examination round of the cohort before serum 25(OH)D assessment, so some changes in serum concentration may have occurred within that period. However, only diet, family medical history, income, and education were assessed at that time, so the time delay may have influenced our results only in very specific circumstances. Specifically, this influence may have occurred if the particular change in the variables differently confounded the relationship between vitamin D status and MetS at the moment of serum 25(OH)D assessment than before the serum 25(OH)D assessment, which is unlikely in the elderly. Fourthly, although we adjusted for many potential confounders, this study is of observational design, so residual confounding may remain. For example, confounding may have occurred

due to lack of data on time spent outdoors. Finally, we could not explore the relationship longitudinally, because not all components of MetS were measured in the subsequent rounds of the Rotterdam Study.

Main conclusion and future directions

Higher serum 25(OH)D concentrations were associated with lower prevalence of MetS in the elderly. Moreover, vitamin D status was associated with lower prevalence of dyslipidemia, abdominal obesity, and hyperglycemia. In addition, vitamin D status was associated with abdominal obesity independently of other MetS components. The beneficial effects of vitamin D might differ in magnitude in different risk groups for the MetS. Some effects of vitamin D were stronger in females. We conclude that the elderly might benefit from higher serum 25(OH)D, especially women. However, the causality between vitamin D status and MetS still needs to be investigated and, therefore, well designed supplementation trials are needed.

Supplementary data

This is linked to the online version of the paper at http://dx.doi.org/10.1530/EJE-14-0580.

Declaration of interest

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