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# Higher serum 25-hydroxyvitamin D concentrations are related to a reduced risk of depression

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## Abstract

Vitamin D has been suggested to protect against depression, but epidemiological evidence is scarce. The present study investigated the relationship of serum 25-hydroxyvitamin D (25(OH)D) with the prevalence of depressive and anxiety disorders. The study population consisted of a representative sample of Finnish men and women aged 30–79 years from the Health 2000 Survey. The sample included 5371 individuals, of which 354 were diagnosed with depressive disorder and 222 with anxiety disorder. Serum 25(OH)D concentration was determined from frozen samples. In a cross-sectional study, a total of four indicators of depression and one indicator of anxiety were used as dependent variables. Serum 25(OH)D was the risk factor of interest, and logistic models used further included sociodemographic and lifestyle variables as well as indicators of metabolic health as confounding and/or effect-modifying factors. The population attributable fraction (PAF) was estimated. Individuals with higher serum 25(OH)D concentrations showed a reduced risk of depression. The relative odds between the highest and lowest quartiles was 0·65 (95 % CI 0·46, 0·93; *P* for trend=0·006) after adjustment for sociodemographic, lifestyle and metabolic factors. Higher serum 25(OH)D concentrations were associated with a lower prevalence of depressive disorder especially among men, younger, divorced and those who had an unhealthy lifestyle or suffered from the metabolic syndrome. The PAF was estimated to be 19 % for depression when serum 25(OH)D concentration was at least 50 nmol/l. These results support the hypothesis that higher serum 25(OH)D concentrations protect against depression even after adjustment for a large number of sociodemographic, lifestyle and metabolic factors. Large-scale prospective studies are needed to confirm this finding.

**Key words: Vitamin D: Depression: Cross-sectional studies**

Depression is a major cause of illness burden to societies worldwide, with increasing effects on personal suffering, work disability, and use of healthcare resources[1]. The 1-year prevalence of depressive disorders in Finland has been estimated to be about 8 % in women and 5 % in men[2].

The pathogenesis of depression is thought to involve multiple interacting risk factors, covering biological[3], psychological and psychosocial factors[4–6], for example maladaptive consequences on personality and behaviour of being exposed to adverse childhood experiences, losses, and other stressful life events in biologically vulnerable persons. The possible pathophysiological mechanisms of depression have been linked to functional and structural brain abnormalities[7]. They are mostly related to genetic vulnerability, the malfunction of noradrenergic and serotonergic neurotransmitter systems that modulate brain areas concerning mood, thinking, sleep, appetite and behaviour, as well as to the role of dysregulation of the stress hormone cortisol[3] and the immune response system[8]. Depression is a disorder with a high prevalence[9] and a frequently recurrent and chronic course[10]. For this reason, research is needed to identify potential biological, psychological and environmental risk factors in order to prevent its prevalence.

Vitamin D has many physiological roles[11]. In addition to its classic role in bone metabolism, vitamin D may also have many potential non-skeletal functions. This is supported by the finding that vitamin D receptors are expressed not only in cells related to Ca and P metabolism, but also in other cells and tissues in the human body[11]. Vitamin D appears to play an important role in brain development and function, and may be directly involved in the autocrine or paracrine regulation of the brain[12,13].

Some recently published larger cross-sectional studies have demonstrated an inverse association between serum 25-hydroxyvitamin D (25(OH)D) concentration and depression. For example, a Norwegian population-based study on 10 086

**Abbreviations:** 25(OH)D, 25-hydroxyvitamin D; BDI, Beck Depression Inventory; CES-D, Center for Epidemiologic Studies-Depression; mAHEI, modified Alternate Healthy Eating Index; PAF, population attributable fraction.

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individuals has indicated that low serum 25(OH)D concentrations are associated with depressive symptoms measured by the Hopkins Symptoms Check List 10[14]. In addition, one study conducted in a sample of 12 594 US men and women showed that low serum 25(OH)D concentration is associated with depressive symptoms, measured by the Center for Epidemiologic Studies-Depression (CES-D) scale, particularly in subjects with a history of depression[15]. Similar results have also been found in a large, nationally representative sample of 7970 young adults in the USA assessed by the Diagnostic Interview Schedule[16]. Furthermore, an inverse association has been demonstrated in three studies on older adults where depression or depressive symptoms were measured using the CES-D scale and the Diagnostic Interview Schedule[17], the Geriatric Depression Scale (GDS10)[18] and the Beck Depression Inventory (BDI)[19].

It has been shown that serum 25(OH)D is associated with a large number of sociodemographic, lifestyle and metabolic health-related variables[20]. This, and the finding of a lack of association with depression after controlling for potential confounding factors in two studies[21,22], underlines the need for large-scale studies with a comprehensive set of serum 25(OH)D determinants. The objective of the present study was to assess the cross-sectional associations between serum 25(OH)D concentration and the prevalence of depression after comprehensively controlling for potential confounding factors in a representative sample of the Finnish adult population. To ensure that there is a specific association between serum 25(OH)D and depression, we also evaluated the association between serum 25(OH)D and anxiety disorder.

## Subjects and methods

### Study population

The present cross-sectional study was based on the Health 2000 Survey, which was carried out in Finland from 2000 to 2001[23]. The Health 2000 Survey included interviews, self-administered questionnaires and a comprehensive health examination. The study population consisted a two-stage stratified cluster sample representing the adult population aged 30 years and over living in mainland Finland. The sample frame was regionally stratified according to five university hospital regions. In the first stage of sampling, eighty healthcare districts were sampled as a cluster. In the second stage, a random sample of individuals from each of the eighty healthcare districts was drawn from a national population register. The study population of the Health 2000 Survey consisted of 8028 individuals, of whom 85 % participated in a health examination. The present study included 2524 men and 2847 non-pregnant women aged 30–79 years whose serum 25(OH)D concentrations were determined and whose depressive symptoms and diagnoses of depressive and anxiety disorders were assessed.

Written informed consent was obtained from all the participants of the survey. The present study was approved by the Ethical Committee for Research in Epidemiology and Public Health at the Hospital District of Helsinki and Uusimaa in Finland.

### Methods

*Current depressive symptoms and depressive and anxiety disorders*. Current depressive symptoms were measured using the BDI[24], and the cut-off point ≥10 points[25] indicated the presence of symptoms (1404 cases). Diagnoses of depressive disorder (354 cases), major depressive disorder (271 cases), dysthymic disorder (131 cases) and anxiety disorder (222 cases) were assessed using a computerised version of the Munich-Composite International Diagnostic Interview, allowing the estimation of diagnoses for mental disorders during the past 12 months, according to the Diagnostic and Statistical Manual of Mental Disorders (DSM-IV)[26,27].

*Serum 25-hydroxyvitamin D assay*. Serum samples were collected from the subjects between September 2000 and March 2011. Serum 25(OH)D concentrations were determined based on serum samples stored at −70°C using RIA (DiaSorin) between January 2001 and November 2002. The inter-assay CV for 25(OH)D concentration measurements was 7·80 % at a mean concentration of 47·3 nmol/l. The laboratory where the analyses were carried out participated in an external quality control programme run by Labquality Limited. Serum 25(OH)D concentration was categorised according to quartiles (7–33, 34–43, 44–55 and 56–134 nmol/l). Serum 25(OH)D concentration less than 50 nmol/l was considered as a cut-off point of vitamin D deficiency[28,29].

*Sociodemographic and lifestyle factors and chronic diseases*. Interviews and self-administered questionnaires provided information on marital status (unmarried, married or cohabiting, divorced and widow/widower), education (<7, 7–12 and >12 years), leisure-time physical activity (no, moderate ≥4 h/week and vigorous ≥3 h/week), smoking status (never, former smoker and current smoker) and alcohol consumption (male 0, 1–199 and ≥200 g/week; female 0, 1–99 and ≥100 g/week)[23]. Currently perceived economic problems were also determined using a questionnaire on a 5-point ordinal scale that asked whether the household had to make considerable or major compromises in consumption. Likewise, currently perceived lack of social support was determined by asking the respondent whether there was no one close by to get help or support from in time of exhaustion, when in need of practical help or emotional support. Perceived health was determined in the interview based on a 5-point scale that ranged from good to poor. Also, chronic diseases were self-reported in the interview (stroke, coronary artery disease, diabetes and cancer) or diagnosed in the health examination (musculoskeletal disorder).

*Diet and vitamin D supplementation*. Data on diet were collected using a self-administered FFQ that was designed to estimate average food intakes over the preceding year[30,31]. Participants recorded their average consumption of food items and prepared dishes in nine frequency categories ranking from 'never or seldom' to 'at least six times per d'. Average food consumption and intakes of nutrients per d were calculated using software developed at the National Institute for Health and Welfare (Finessi) and the National Finnish Food Composition Database (Fineli®)[32]. The FFQ also included questions about the regular use of vitamin D

supplements, which included single-ingredient vitamin D supplements and multivitamin supplements including vitamin D.

*Dietary index*. A dietary index (modified Alternate Healthy Eating Index, mAHEI) was created based on the definition of the nine-item Alternate Healthy Eating Index[33,34]. The mAHEI was adapted to the Finnish diet and included seven components (vegetables, fruits and berries, legumes, nuts, seeds and soyabeans, white:red meat ratio, rye, polyunsaturated: saturated fat ratio, and *trans*-fat intake). The components were divided into quintiles and, with the exception of *trans*-fat intake, received scores in ascending order, such that the lowest quintile received 1 point and the highest quintile 5 points. Alcohol consumption and multivitamin use were excluded because alcohol consumption was considered an independent lifestyle factor in the present study and habitual multivitamin use is not recommended in Finland. Scores of all components were summed to the total score that ranged from 7 (lowest) to 35 (highest). A higher score indicated a healthier diet.

*Lifestyle index*. A lifestyle index was created based on the following five factors: BMI; physical activity; smoking status; alcohol consumption; diet[35]. A healthy lifestyle was defined as a BMI $<25{\cdot}0\,kg/m^2$, regular leisure-time physical activity at least approximately 30 min/d (moderate $\geq 4$ h/week or vigorous $\geq 3$ h/week), no current smoking, alcohol consumption ranging from 1 to 99 g/week in women or 1 to 199 g/week in men, and an above-median total score on the mAHEI index (21 points)[36]. For each factor, participants who met the criteria for a healthy lifestyle received 1 point, while those who did not meet the criteria were scored 0. Thus, the total score ranged from 0 to 5, with higher scores suggesting a healthier lifestyle than lower scores.

*Metabolic health-related factors*. Weight, height and waist circumference were measured with subjects wearing light clothing and without shoes, and BMI ($kg/m^2$) was calculated[23]. Blood pressure was measured twice by the auscultation method, and the mean value of the two measurements was used. Fasting blood samples were taken and stored at $-70°C$. Serum fasting glucose and TAG concentrations were analysed by enzymatic methods (Triglycerides GPO PAP, Glucose and Hexokinase; Olympus System Reagent) and serum HDL-cholesterol by a direct method (HDL-C Plus; Roche Diagnostics GmbH). The positive metabolic syndrome was defined according to the harmonisation definition[37] as the presence of any three of the following five risk factors: elevated waist circumference ($\geq 94$ cm in men and $\geq 80$ cm in women); elevated serum TAG ($\geq 1{\cdot}7$ mmol/l); reduced HDL-cholesterol ($<1{\cdot}0$ mmol/l in men and $<1{\cdot}3$ mmol/l in women); elevated mean blood pressure (systolic blood pressure $\geq 130$ mmHg or diastolic blood pressure $\geq 85$ mmHg or antihypertensive drug treatment); elevated serum fasting glucose level ($\geq 5{\cdot}6$ mmol/l).

### Statistical analyses

The associations between serum 25(OH)D concentration and potential confounding and effect-modifying factors were estimated using linear models. Multivariate logistic regression was used to evaluate the cross-sectional association between serum 25(OH)D concentration and the prevalence of depressive disorder. Relative odds (OR) and 95 % CI between quartiles of serum 25(OH)D were calculated. Test for trend was performed using the likelihood ratio test, with serum 25(OH)D included as a continuous variable in the model. The population attributable fraction (PAF) was assessed based on the estimated OR. The PAF estimated the proportion of cases (e.g. depressive disorder) in a given population that would theoretically not have occurred if all of the individuals had low-risk target values of the risk factors (i.e. serum 25(OH)D concentration at least 50 nmol/l) of interest instead of their true values[38]. This was done by combining information about the prevalence of the risk factor in the population with estimates of the strength of the association between the risk factor and the outcome. The rest of the factors in the model retained their values. In the calculation of PAF, a causal relationship between the risk factors and the outcome was assumed. Two-sided 95 % CI of the PAF were estimated by the delta method.

We defined three main-effects models. Model 1 (OR only) included age, sex and month of serum sampling for serum 25(OH)D determinations. Model 2 (OR only) further included education and four lifestyle factors (BMI, leisure-time physical activity, smoking status and alcohol consumption). Model 3 (OR and PAF) was additionally adjusted for the components of the metabolic syndrome excluding waist circumference (i.e. blood pressure, serum HDL-cholesterol, serum TAG and fasting glucose). Waist circumference was excluded because BMI was already included as a lifestyle factor, both of which are measures of obesity.

Possible modification by sex, age, marital status, healthy lifestyle index, healthy dietary index (mAHEI), the metabolic syndrome, perceived economic problems, perceived social support, anxiety disorder, chronic diseases and perceived health on the prevalence of depressive disorder was studied by including an interaction term between serum 25(OH)D and the potential effect-modifying factor in model 3 one at a time.

Statistical analyses were carried out using SAS 9.2 (SAS Institute, Inc.)[39].

## Results

Participants with higher serum 25(OH)D concentrations (highest quartile range 56–134 nmol/l) were somewhat older, generally married or cohabiting, more educated and with no economic problems compared with others (Table 1). Participants with higher serum 25(OH)D concentrations also had a healthier lifestyle by being leaner, performing more leisure-time physical activity, smoking less often and consuming less alcohol. Their vitamin D intake from diet and supplements was higher and eating habits were healthier. In addition, their metabolic health was better, showing lower blood pressure and lower serum TAG and serum fasting glucose concentrations, and higher serum HDL-cholesterol concentrations. They also perceived their health to be better. Lower serum 25(OH)D values were more often measured during the winter season.

An inverse association was found between serum 25(OH)D concentration and depressive disorder during the past

**Table 1.** Descriptive variables of the total study population by quartiles of serum 25-hydroxyvitamin D (25(OH)D) concentrations
(Mean values and standard deviations or percentages, adjusted for age (continuous) and sex)

| | | Serum 25(OH)D quartiles* | | | | |
|---|---|---|---|---|---|---|
| | Total (*n* 5371) | 1 (*n* 1276) | 2 (*n* 1414) | 3 (*n* 1371) | 4 (*n* 1310) | *P* for heterogeneity |
| Sociodemographic factors | | | | | | |
| Age (years) | | | | | | <0·001 |
| Mean | 50·4 | 48·3 | 49·5 | 51·0 | 52·7 | |
| SD | 12·7 | 13·2 | 12·4 | 12·3 | 12·5 | |
| Sex, women (%) | 53·0 | 52·2 | 55·4 | 53·8 | 50·4 | 0·058 |
| Married or cohabiting (%)† | 73·1 | 66·5 | 72·9 | 75·1 | 77·6 | <0·001 |
| Education >12 years (%)† | 37·2 | 33·9 | 34·8 | 39·7 | 40·2 | 0·003 |
| Economic problems (%)† | 14·5 | 17·9 | 16·3 | 13·4 | 10·6 | <0·001 |
| Lack of social support (%)† | 8·81 | 10·5 | 8·60 | 8·52 | 7·70 | 0·084 |
| Lifestyle | | | | | | |
| BMI (kg/m$^2$)† | | | | | | <0·001 |
| Mean | 26·9 | 27·6 | 27·2 | 26·9 | 26·0 | |
| SD | 4·64 | 5·20 | 4·67 | 4·50 | 4·08 | |
| Moderate or vigorous leisure-time physical activity (%)† | 75·1 | 64·2 | 75·0 | 77·6 | 83·1 | <0·001 |
| Non-smoker (%)† | 74·5 | 67·0 | 72·4 | 76·6 | 81·8 | <0·001 |
| Alcohol consumption (g/week)† | | | | | | 0·0001 |
| Mean | 81·3 | 96·8 | 74·6 | 78·4 | 76·4 | |
| SD | 149 | 196 | 129 | 139 | 124 | |
| Vitamin D and dietary factors | | | | | | |
| Serum 25(OH)D (nmol/l) | | | | | | <0·001 |
| Mean | 45·6 | 26·7 | 38·5 | 49·1 | 68·1 | |
| SD | 16·6 | 5·25 | 2·87 | 3·39 | 12·7 | |
| Vitamin D intake from diet (μg/d)‡ | | | | | | <0·001 |
| Mean | 6·84 | 6·04 | 6·80 | 7·05 | 7·41 | |
| SD | 4·06 | 3·65 | 4·09 | 4·01 | 4·26 | |
| Vitamin D supplement use (%)‡ | 11·1 | 3·41 | 7·39 | 14·0 | 19·2 | <0·001 |
| mAHEI (total score)‡ | | | | | | <0·001 |
| Mean | 21·1 | 20·0 | 20·8 | 21·3 | 22·1 | |
| SD | 4·92 | 4·89 | 4·90 | 4·84 | 4·80 | |
| Metabolic health and chronic diseases | | | | | | |
| Normal blood pressure§ (%)† | 38·0 | 36·2 | 35·9 | 39·1 | 40·7 | 0·012 |
| Serum HDL-cholesterol (mmol/l)† | | | | | | <0·001 |
| Mean | 1·33 | 1·25 | 1·30 | 1·35 | 1·43 | |
| SD | 0·38 | 0·36 | 0·36 | 0·38 | 0·38 | |
| Serum TAG (mmol/l)† | | | | | | <0·001 |
| Mean | 1·58 | 1·77 | 1·63 | 1·54 | 1·39 | |
| SD | 1·05 | 1·33 | 1·07 | 0·92 | 0·78 | |
| Serum fasting glucose (mmol/l)† | | | | | | <0·001 |
| Mean | 5·52 | 5·65 | 5·52 | 5·50 | 5·42 | |
| SD | 1·17 | 1·54 | 1·14 | 0·98 | 0·95 | |
| Metabolic syndrome (%)† | 42·5 | 49·0 | 45·9 | 41·6 | 33·3 | <0·001 |
| Chronic disease‖ (%)† | 44·7 | 46·2 | 46·2 | 43·1 | 43·4 | 0·144 |
| Good perceived health (%)† | 65·3 | 60·0 | 65·5 | 65·6 | 69·8 | <0·001 |
| Diagnoses of depression and anxiety | | | | | | |
| Depressive disorder (%) | 6·59 | 8·38 | 7·17 | 5·65 | 5·21 | 0·004 |
| Major depressive disorder (%) | 5·05 | 6·30 | 5·34 | 4·00 | 4·61 | 0·042 |
| Dysthymic disorder (%) | 2·44 | 3·57 | 2·81 | 1·80 | 1·59 | 0·003 |
| Anxiety disorder (%) | 4·13 | 4·50 | 3·80 | 3·82 | 4·46 | 0·678 |
| Depressive symptoms | | | | | | |
| BDI ≥10 (%) | 26·1 | 30·0 | 26·3 | 25·3 | 23·4 | 0·003 |
| Season | | | | | | |
| Winter season (November to February) (%) | 64·9 | 78·3 | 68·4 | 61·3 | 51·6 | <0·001 |

mAHEI, modified Alternate Healthy Eating Index; BDI, Beck Depression Inventory.
* 7–33, 34–43, 44–55 and 56–134 nmol/l.
† One to thirty-seven participants were excluded because of missing data.
‡ 367 participants were excluded because of missing data on diet.
§ Normal blood pressure: systolic blood pressure <130 mmHg, diastolic blood pressure <85 mmHg and no antihypertensive medication.
‖ At least one of the following: stroke (self-reported); coronary artery disease (self-reported); diabetes (self-reported); cancer (self-reported); musculoskeletal disorder (diagnosed).

12 months after adjustment for age, sex and month for serum 25(OH)D measurements. The relative odds (OR) of the disease comparing the highest with the lowest quartile of serum vitamin D concentration was 0·56 (95% CI 0·40, 0·78; *P* for trend <0·001; Table 2). After further adjustment for potential confounding sociodemographic and lifestyle factors, the OR was slightly changed (OR 0·66, 95% CI 0·47, 0·93; *P* for trend=0·007). Further adjustment for the indicators of

**Table 2.** Depression, depressive symptoms and anxiety disorder by quartiles of serum 25-hydroxyvitamin D (25(OH)D) concentration (Odds ratios and 95 % confidence intervals)

| | Serum 25(OH)D quartiles* | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | 1 | 2 | | 3 | | 4 | | |
| | OR | OR | 95 % CI | OR | 95 % CI | OR | 95 % CI | *P* for trend |
| Diagnoses of depression | | | | | | | | |
| Depressive disorder | | | | | | | | |
| Model 1† | 1·00 | 0·83 | 0·62, 1·10 | 0·63 | 0·47, 0·86 | 0·56 | 0·40, 0·78 | <0·001 |
| Model 2‡ | 1·00 | 0·90 | 0·67, 1·20 | 0·72 | 0·52, 0·98 | 0·66 | 0·47, 0·93 | 0·007 |
| Model 3§ | 1·00 | 0·90 | 0·67, 1·20 | 0·71 | 0·52, 0·98 | 0·65 | 0·46, 0·93 | 0·006 |
| Major depressive disorder | | | | | | | | |
| Model 1† | 1·00 | 0·83 | 0·60, 1·14 | 0·60 | 0·42, 0·87 | 0·68 | 0·48, 0·98 | 0·01 |
| Model 2‡ | 1·00 | 0·89 | 0·64, 1·24 | 0·64 | 0·44, 0·93 | 0·74 | 0·51, 1·09 | 0·04 |
| Model 3§ | 1·00 | 0·90 | 0·65, 1·26 | 0·64 | 0·44, 0·93 | 0·74 | 0·50, 1·09 | 0·04 |
| Dysthymic disorder | | | | | | | | |
| Model 1† | 1·00 | 0·75 | 0·49, 1·16 | 0·47 | 0·28, 0·77 | 0·40 | 0·23, 0·68 | <0·001 |
| Model 2‡ | 1·00 | 0·88 | 0·56, 1·39 | 0·64 | 0·38,1·08 | 0·62 | 0·35, 1·08 | 0·04 |
| Model 3§ | 1·00 | 0·88 | 0·56, 1·40 | 0·66 | 0·39, 1·10 | 0·63 | 0·36, 1·11 | 0·05 |
| Depressive symptoms | | | | | | | | |
| BDI[3] ≥10 | | | | | | | | |
| Model 1† | 1·00 | 0·83 | 0·69, 0·99 | 0·77 | 0·65, 0·92 | 0·69 | 0·58, 0·83 | <0·001 |
| Model 2‡ | 1·00 | 0·90 | 0·75, 1·07 | 0·89 | 0·74, 1·07 | 0·83 | 0·69, 1·01 | 0·08 |
| Model 3§ | 1·00 | 0·90 | 0·75, 1·08 | 0·88 | 0·73, 1·06 | 0·82 | 0·68, 1·00 | 0·06 |
| Diagnosis of anxiety | | | | | | | | |
| Anxiety disorder | | | | | | | | |
| Model 1† | 1·00 | 0·82 | 0·56, 1·20 | 0·81 | 0·55, 1·19 | 0·92 | 0·62, 1·36 | 0·68 |
| Model 2‡ | 1·00 | 0·91 | 0·62, 1·34 | 0·96 | 0·64, 1·43 | 1·19 | 0·79, 1·80 | 0·39 |
| Model 3§ | 1·00 | 0·91 | 0·62, 1·35 | 0·97 | 0·65, 1·45 | 1·22 | 0·80, 1·84 | 0·35 |

BDI, Beck Depression Inventory.
* 7–33, 34–43, 44–55 and 56–134 nmol/l.
† Age (continuous), sex and month of blood sampling (September to March).
‡ Further included education (<7, 7–12 and >12 years), BMI (kg/m$^2$, continuous), leisure-time physical activity (no, moderate ≥4 h/week and vigorous ≥3 h/week), smoking (never, former smoker and current smoker) and alcohol consumption (male 0, 1–199 and ≥200 g/week; female 0, 1–99 and ≥100 g/week).
§ Further included blood pressure (optimal, normal, high normal and hypertension), serum HDL-cholesterol (mmol/l, continuous), serum TAG (mmol/l, continuous) and serum fasting glucose (mmol/l, continuous).

metabolic health did not notably alter the results (OR 0·65, 95 % CI 0·46, 0·93; *P* for trend=0·006). The results for major depressive disorder (OR 0·68–0·74), dysthymic disorder (OR 0·40–0·63) and current depressive symptoms according to the BDI (OR 0·69–0·82) were quite similar; however, in models 2 and 3, the association between serum 25(OH)D concentration and current depressive symptoms attenuated to a non-significant suggestive association. However, anxiety disorders showed a different picture with no association in the different models, the OR varying from 0·92 to 1·22 between the three models.

The inclusion of an interaction term between serum 25(OH)D and potential effect-modifying factors in model 3 one at a time (sex, age, marital status, healthy lifestyle index, healthy dietary index (mAHEI), the metabolic syndrome, perceived economic status, perceived social support, diagnosed anxiety disorder, chronic disease and perceived health) showed that the strength of the association between serum 25(OH)D concentration and the prevalence of depression varied between several subgroups (Table 3). Higher serum 25(OH)D concentrations were associated with a lower prevalence of depressive disorder especially among men (OR 0·40, 95 % CI 0·22, 0·73), and among young and divorced individuals. Similarly, participants who had an unhealthy lifestyle or poor dietary habits, or suffered from the metabolic syndrome, showed a stronger association. By contrast, for participants who received social support or were free from anxiety disorder or chronic disease, higher serum 25(OH)D concentration was related to a lower risk of depressive disorder.

Serum 25(OH)D appeared to be strongly associated with depressive disorder in terms of the PAF (Table 4). If a causal relationship between serum 25(OH)D and depressive disorder is assumed, 19 (95 % CI 4, 31) % of all cases could have been avoided if serum 25(OH)D concentration of each individual had been at least 50 nmol/l. The association was non-significant for major depressive disorder (PAF 15 %, 95 % CI −3, 29 %) and dysthymic disorder (PAF 23 %, 95 % CI −5, 43 %) and weaker for depressive symptoms (PAF 10 %, 95 % CI 1, 18 %). Anxiety disorders did not show any reduction at higher concentrations of serum 25(OH)D (PAF −3 %, 95 % CI −22, 12 %).

## Discussion

### Findings

The present cross-sectional study suggests that a higher serum 25(OH)D concentration is associated with a reduced risk of depressive disorder and depressive symptoms in a representative sample of the Finnish adult population. This association remained significant after adjustment for potential

**Table 3.** Depressive disorder between the highest and lowest quartiles of serum 25-hydroxyvitamin D (25(OH)D) concentration in the categories of potential effect-modifying factors

(Odds ratios and 95 % confidence intervals*)

| Effect-modifying factors | Total population (*n*) | *n*† | Serum 25(OH)D (highest *v.* lowest quartile) OR‡ | 95 % CI‡ |
|---|---|---|---|---|
| Sex | | | | |
| Men | 2473 | 111 | 0·40 | 0·22, 0·73 |
| Women | 2787 | 236 | 0·85 | 0·56, 1·29 |
| Age (years) | | | | |
| 30–59 | 3947 | 291 | 0·58 | 0·40, 0·85 |
| 60–79 | 1313 | 56 | 1·13 | 0·49, 2·60 |
| Marital status | | | | |
| Divorced | 524 | 68 | 0·43 | 0·19, 0·97 |
| Other | 4736 | 279 | 0·75 | 0·51, 1·10 |
| Lifestyle index score§ | | | | |
| 0–2 points, unhealthy lifestyle | 1601 | 128 | 0·52 | 0·28, 0·97 |
| 3–5 points, healthy lifestyle | 3304 | 189 | 0·70 | 0·45, 1·09 |
| mAHEI index score | | | | |
| <21 points, unhealthy diet | 2249 | 153 | 0·54 | 0·31, 0·92 |
| ≥21 points, healthy diet | 2656 | 164 | 0·80 | 0·49, 1·28 |
| Metabolic syndrome‖ | | | | |
| Yes | 2225 | 127 | 0·47 | 0·26, 0·85 |
| No | 3041 | 220 | 0·77 | 0·50, 1·16 |
| Perceived economic status | | | | |
| Economic problems | 758 | 103 | 0·62 | 0·32, 1·21 |
| Other | 4494 | 244 | 0·72 | 0·48, 1·08 |
| Perceived social support | | | | |
| Lack of social support | 460 | 71 | 1·07 | 0·50, 2·29 |
| Other | 4791 | 275 | 0·59 | 0·40, 0·87 |
| Anxiety disorder | | | | |
| Yes | 217 | 83 | 0·70 | 0·31, 1·58 |
| No | 5043 | 264 | 0·59 | 0·39, 0·87 |
| Chronic disease¶ | | | | |
| Yes | 2339 | 160 | 0·82 | 0·50, 1·35 |
| No | 2916 | 186 | 0·55 | 0·34, 0·88 |
| Perceived poor health | | | | |
| Yes | 1806 | 180 | 0·70 | 0·43, 1·13 |
| No | 3443 | 166 | 0·65 | 0·40, 1·05 |

mAHEI, modified Alternate Healthy Eating Index.

* Model: age (continuous); sex; month of blood sampling (September to March); education (<7, 7–12 and >12 years); BMI (kg/m$^2$, continuous); leisure-time physical activity (no, moderate ≥4 h/week and vigorous ≥3 h/week); smoking status (never, former smoker and current smoker); alcohol consumption (male 0, 1–199 and ≥200 g/week; female 0, 1–99 and ≥100 g/week); blood pressure (optimal, normal, high normal and hypertension); serum HDL-cholesterol (mmol/l, continuous); serum TAG (mmol/l, continuous); serum fasting glucose (mmol/l, continuous).

† Depressive disorder cases in the total population.

‡ OR and 95 % CI for the fourth quartile of serum 25(OH)D with the first quartile as a referent group.

§ BMI, leisure-time physical activity, smoking and alcohol consumption were excluded from the model.

‖ BMI, blood pressure, serum HDL-cholesterol, TAG and fasting glucose were excluded from the model.

¶ At least one of the following: stroke (self-reported); coronary artery disease (self-reported); diabetes (self-reported); cancer (self-reported); musculoskeletal disorder (diagnosed).

confounding factors related to sociodemographic status, lifestyle and metabolic health. Conversely, there was no association between serum 25(OH)D concentration and anxiety disorder.

The present results confirm the findings of previous cross-sectional studies indicating that serum 25(OH)D is inversely associated with the diagnosis of depression[16] or depressive symptoms[14,15]. In contrast, two cross-sectional studies among US adults[22] and elderly Chinese[21] have reported a lack of association between serum 25(OH)D and depression after controlling for potential confounders (e.g. sociodemographic factors, lifestyle and chronic diseases). However, in the present study, adjustment for a large number of potential confounding factors only slightly attenuated the association, thereby reducing the possibility that the association is caused by confounding factors. In addition, few randomised trials and cohort studies have reported contradictory results. In one small randomised double-blind trial among overweight and obese Norwegians, it has been indicated that 1 year of supplementation with vitamin D at high doses reduced depressive symptoms, as measured by the BDI[40]. However, in a randomised double-blind trial on postmenopausal US women, 2 years of supplementation with vitamin D did not reduce depressive symptoms, as measured by the Burnam scale[41]. Also, a few cohort studies, most of them based on some subpopulation of age, sex or disease, investigating the prediction of vitamin D status on the incidence of depression have given contradictory results, showing either an inverse association[42,43] or no association[44]. Recently, a study based on depressed individuals has shown an inverse association of serum 25(OH)D concentrations with the severity of depressive

**Table 4.** Population attributable fractions (PAF)* for diagnoses of depression, anxiety and depressive symptoms at a serum 25-hydroxyvitamin D (25(OH)D) concentration of at least 50 nmol/l
(Odds ratios and 95 % confidence intervals)

| | Serum 25(OH)D | | | | |
|---|---|---|---|---|---|
| | ≥ 50 nmol/l | < 50 nmol/l | | | |
| | OR | OR | 95 % CI | PAF | 95 % CI |
| Diagnosis of depression | | | | | |
| Depressive disorder | 1 | 1·40 | 1·08, 1·81 | 18·6 | 3·61, 31·2 |
| Major depressive disorder | 1 | 1·29 | 0·97, 1·73 | 14·8 | −2·70, 29·3 |
| Dysthymic disorder | 1 | 1·46 | 0·95, 2·24 | 22·6 | −5·38, 43·2 |
| Depressive symptoms | | | | | |
| BDI ≥ 10 | 1 | 1·18 | 1·02, 1·36 | 10·2 | 1·17, 18·4 |
| Diagnosis of anxiety | | | | | |
| Anxiety disorder | 1 | 0·94 | 0·69, 1·27 | −3·43 | −22·0, 12·3 |

BDI, Beck Depression Inventory.
* Model: age (continuous); sex; month of blood sampling (continuous); education (< 7, 7–12 and > 12 years); BMI (kg/m$^2$, continuous); leisure-time physical activity (no, moderate ≥ 4 h/week and vigorous ≥ 3 h/week); smoking (never, former smoker and current smoker); alcohol consumption (male 0, 1–199 and ≥ 200 g/week; female 0, 1–99 and ≥ 100 g/week); blood pressure (optimal, normal, high normal and hypertension); serum HDL-cholesterol (mmol/l, continuous); serum TAG (mmol/l, continuous); serum fasting glucose (mmol/l, continuous).

symptoms and the risk of developing depressive disorder during a 2-year follow-up[45].

A potential role of vitamin D in the pathophysiology of depression and the exact biological mechanisms behind this association remain mainly unknown, but some plausible hypotheses have been presented. Because receptors of vitamin D and vitamin D-activating enzyme 1α-hydroxylase are present in human brain regions (e.g. prefrontal cortex and hippocampus) known to be associated with the pathophysiology of depression[12,13] it is possible that vitamin D may play a role in this process. It has been suggested that vitamin D deficiency may contribute to disruptions in the neuroendocrine and central nervous systems[46]. These disruptions may lead to further dysregulation of neurotransmission and neurotransmitter metabolism and signalling, neuroprotection, neuroimmunomodulation, and the modulation of glucocorticoid-related actions in hippocampal cells, potentially leading to depression[46,47]

The present study investigating the interactions between serum 25(OH)D and selected potential effect-modifying factors showed, in contrast to two previous studies[14,42], that the association between serum 25(OH)D concentration and the prevalence of depression was stronger among men than among women. We also found that vitamin D protects against depression in the younger age group (30–59 years), but not among older adults. One explanation for the weaker association in the older age group might be that in the present data, low vitamin D concentrations are more common among younger individuals[20]. Another explanation might be the small number of depressive disorder cases (*n* 56) among older adults. We also made an interesting finding that the higher the serum 25(OH)D concentration, the lower the risk of depression, particularly among those who were divorced, had an unhealthier diet or lifestyle or had the metabolic syndrome. Assuming that there really is a causal relationship between vitamin D and depression, this may suggest that vitamin D provides protection against depression, with currently unknown mechanisms, especially for individuals with poor socio-economic status, lifestyle choices and metabolic health. Conversely, among individuals free from chronic diseases or anxiety disorder, the association between serum 25(OH)D concentration and the risk of depression was stronger than among those suffering from these diseases.

## Methodological issues

The study has several strengths. First, the results are based on a large representative sample of the Finnish adult population, allowing the estimation of the PAF. Second, depression and anxiety were assessed with widely used and validated instruments. Third, we used serum 25(OH)D concentration as the indicator of vitamin D status. Serum 25(OH)D concentration reflects vitamin D obtained from both diet and supplements and through subcutaneous synthesis stimulated by exposure to UVB radiation from sunlight. Fourth, we took into account a large variety of potential confounding and effect-modifying factors, which were determined by reliable methods[23]. Finally, we compared the results according to depressive and anxiety disorders. Serum 25(OH)D concentration was not associated with anxiety disorders, which reduces the possibility that vitamin D is generally associated with mental health. It may support the hypothesis that there is a biological mechanism for the association between serum 25(OH)D concentration and the prevalence of depression.

There are also some limitations. First, due to the cross-sectional nature of the study, any conclusions about causality cannot be drawn. It is possible that low serum 25(OH)D concentration may be a result of depression. Individuals suffering from depression may spend less time outdoors or have a poor diet low in natural (e.g. fish) or fortified (milk products and margarine) vitamin D. Second, despite the large number of potential confounding and effect-modifying factors considered in the present study, it is possible that residual confounding still remains. Finally, there are also some limitations relating to serum

25(OH)D measurements. Serum 25(OH)D concentration was measured only once, thus reflecting only recent exposure and providing no information on intra-individual seasonal variation. Furthermore, the possibility that the concentrations may have declined during storage, although only a small decline has been reported in the analyses based on frozen serum samples[48,49], cannot be ruled out. It should also be noted that mean serum 25(OH)D concentration reported in the present study is an underestimate of the full-year average, because the present data do not include any values for the summer months. In addition, the RIA used may have underestimated serum 25(OH)D concentrations[50].

## Conclusions

In conclusion, the present study suggests a protective effect of vitamin D against depressive disorder, but not against anxiety disorder, even after adjustment for a large number of potential confounding factors. If this association is causal, our finding may be of importance for public health policy, especially in populations with low concentrations of serum vitamin D. For example, in Finland, the annual costs due to depression are about 1 billion euros[51]. In the case of vitamin D providing protection against depression, elevation of serum 25(OH)D concentration to a level above 50 nmol/l in the total population would result in the avoidance of every fifth prevalent case with depressive disorder and in subsequent savings. Because the hypothesis of the inverse association between serum 25(OH)D concentration and the prevalence of depression is still mainly based on observational studies with a cross-sectional design, and because the determinants of vitamin D are not yet fully understood, large-scale cohort studies and experimental evidence are needed before any firm conclusions can be drawn.

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## References

1. Wittchen HU, Jacobi F, Rehm J, *et al.* (2011) The size and burden of mental disorders and other disorders of the brain in Europe 2010. *Eur Neuropsychopharmacol* **21**, 655–679.
2. Pirkola SP, Isometsä E, Suvisaari J, *et al.* (2005) DSM-IV mood-, anxiety- and alcohol use disorders and their comorbidity in the Finnish general population – results from the Health 2000 Study. *Soc Psychiatry Psychiatr Epidemiol* **40**, 1–10.
3. Belmaker RH & Agam G (2008) Major depressive disorder. *N Engl J Med* **358**, 55–68.
4. Kendler KS, Gardner CO & Prescott CA (2006) Toward a comprehensive developmental model for major depression in men. *Am J Psychiatry* **163**, 115–124.
5. Kendler KS, Gardner CO & Prescott CA (2002) Toward a comprehensive developmental model for major depression in women. *Am J Psychiatry* **159**, 1133–1145.
6. Kendler KS, Gatz M, Gardner CO, *et al.* (2006) Personality and major depression: a Swedish longitudinal, population-based twin study. *Arch Gen Psychiatry* **63**, 1113–1120.
7. Palazidou E (2012) The neurobiology of depression. *Br Med Bull* **101**, 127–145.
8. Bufalino C, Hepgul N, Aguglia E, *et al.* (2013) The role of immune genes in the association between depression and inflammation: a review of recent clinical studies. *Brain Behav Immun* **31**, 31–47.
9. Baumeister H & Harter M (2007) Prevalence of mental disorders based on general population surveys. *Soc Psychiatry Psychiatr Epidemiol* **42**, 537–546.
10. Kessler RC, Berglund P, Demler O, *et al.* (2003) The epidemiology of major depressive disorder: results from the National Comorbidity Survey Replication (NCS-R). *JAMA* **289**, 3095–3105.
11. DeLuca HF (2004) Overview of general physiologic features and functions of vitamin D. *Am J Clin Nutr* **80**, 1689S–1696S.
12. Eyles DW, Smith S, Kinobe R, *et al.* (2005) Distribution of the vitamin D receptor and 1 alpha-hydroxylase in human brain. *J Chem Neuroanat* **29**, 21–30.
13. Eyles DW, Burne TH & McGrath JJ (2013) Vitamin D, effects on brain development, adult brain function and the links between low levels of vitamin D and neuropsychiatric disease. *Front Neuroendocrinol* **34**, 47–64.
14. Kjaergaard M, Joakimsen R & Jorde R (2011) Low serum 25-hydroxyvitamin D levels are associated with depression in an adult Norwegian population. *Psychiatry Res* **190**, 221–225.
15. Hoang MT, Defina LF, Willis BL, *et al.* (2011) Association between low serum 25-hydroxyvitamin D and depression in a large sample of healthy adults: the Cooper Center longitudinal study. *Mayo Clin Proc* **86**, 1050–1055.
16. Ganji V, Milone C, Cody MM, *et al.* (2010) Serum vitamin D concentrations are related to depression in young adult US population: the Third National Health and Nutrition Examination Survey. *Int Arch Med* **3**, 29.
17. Hoogendijk WJ, Lips P, Dik MG, *et al.* (2008) Depression is associated with decreased 25-hydroxyvitamin D and increased parathyroid hormone levels in older adults. *Arch Gen Psychiatry* **65**, 508–512.
18. Stewart R & Hirani V (2010) Relationship between vitamin D levels and depressive symptoms in older residents from a national survey population. *Psychosom Med* **72**, 608–612.
19. Lee DM, Tajar A, O'Neill TW, *et al.* (2011) Lower vitamin D levels are associated with depression among community-dwelling European men. *J Psychopharmacol* **25**, 1320–1328.
20. Jääskeläinen T, Knekt P, Marniemi J, *et al.* (2013) Vitamin D status is associated with sociodemographic factors, lifestyle and metabolic health. *Eur J Nutr* **52**, 513–525.
21. Pan A, Lu L, Franco OH, *et al.* (2009) Association between depressive symptoms and 25-hydroxyvitamin D in middle-aged and elderly Chinese. *J Affect Disord* **118**, 240–243.
22. Zhao G, Ford ES, Li C, *et al.* (2010) No associations between serum concentrations of 25-hydroxyvitamin D and

parathyroid hormone and depression among US adults. *Br J Nutr* **104**, 1696–1702.
23. Heistaro S (editor) (2008) *Methodology Report. Health 2000 Survey*. Helsinki: National Public Health Institute.
24. Beck AT, Ward CH, Mendelson M, *et al.* (1961) An inventory for measuring depression. *Arch Gen Psychiatry* **4**, 561–571.
25. Beck AT, Steer RA & Garbin MG (1988) Psychometric properties of the Beck Depression Inventory: twenty-five years of evaluation. *Clin Psych Rev* **8**, 77–100.
26. American Psychiatric Association (1994) *Diagnostic and Statistical Manual of Mental Disorders*. Washington, DC: American Psychiatric Association.
27. Wittchen HU, Lachner G, Wunderlich U, *et al.* (1998) Test–retest reliability of the computerized DSM-IV version of the Munich-Composite International Diagnostic Interview (M-CIDI). *Soc Psychiatry Psychiatr Epidemiol* **33**, 568–578.
28. Holick MF (2007) Vitamin D deficiency. *N Engl J Med* **357**, 266–281.
29. IOM (Institute of Medicine Committee) (2011) Dietary reference intakes for adequacy: calcium and vitamin D. In *DRI Dietary Reference Intakes for Calcium and Vitamin D*, pp. 345–402 [AC Ross, CL Taylor, AL Yaktine and HB Del Valle, editors]. Washington, DC: The National Academies Press.
30. Männistö S, Virtanen M, Mikkonen T, *et al.* (1996) Reproducibility and validity of a food frequency questionnaire in a case–control study on breast cancer. *J Clin Epidemiol* **49**, 401–409.
31. Paalanen L, Männistö S, Virtanen MJ, *et al.* (2006) Validity of a food frequency questionnaire varied by age and body mass index. *J Clin Epidemiol* **59**, 994–1001.
32. Reinivuo H, Hirvonen T, Ovaskainen ML, *et al.* (2010) Dietary survey methodology of FINDIET 2007 with a risk assessment perspective. *Public Health Nutr* **13**, 915–919.
33. McCullough ML, Feskanich D, Stampfer MJ, *et al.* (2002) Diet quality and major chronic disease risk in men and women: moving toward improved dietary guidance. *Am J Clin Nutr* **76**, 1261–1271.
34. Sääksjärvi K, Knekt P, Lundqvist A, *et al.* (2013) A cohort study on diet and the risk of Parkinson's disease: the role of food groups and diet quality. *Br J Nutr* **109**, 329–337.
35. Hu FB, Manson JE, Stampfer MJ, *et al.* (2001) Diet, lifestyle, and the risk of type 2 diabetes mellitus in women. *N Engl J Med* **345**, 790–797.
36. Laaksonen MA, Knekt P, Rissanen H, *et al.* (2010) The relative importance of modifiable potential risk factors of type 2 diabetes: a meta-analysis of two cohorts. *Eur J Epidemiol* **25**, 115–124.
37. Alberti KG, Eckel RH, Grundy SM, *et al.* (2009) Harmonizing the metabolic syndrome: a joint interim statement of the International Diabetes Federation Task Force on Epidemiology and Prevention; National Heart, Lung, and Blood Institute; American Heart Association; World Heart Federation; International Atherosclerosis Society; and International Association for the Study of Obesity. *Circulation* **120**, 1640–1645.
38. Laaksonen MA, Härkänen T, Knekt P, *et al.* (2010) Estimation of population attributable fraction (PAF) for disease occurrence in a cohort study design. *Stat Med* **29**, 860–874.
39. SAS Institute, Inc. (2008) *SAS/STAT 9.2 User's Guide*. Cary, NC: SAS Institute, Inc.
40. Jorde R, Sneve M, Figenschau Y, *et al.* (2008) Effects of vitamin D supplementation on symptoms of depression in overweight and obese subjects: randomized double blind trial. *J Intern Med* **264**, 599–609.
41. Bertone-Johnson ER, Powers SI, Spangler L, *et al.* (2012) Vitamin D supplementation and depression in the women's health initiative calcium and vitamin D trial. *Am J Epidemiol* **176**, 1–13.
42. Milaneschi Y, Shardell M, Corsi AM, *et al.* (2010) Serum 25-hydroxyvitamin D and depressive symptoms in older women and men. *J Clin Endocrinol Metab* **95**, 3225–3233.
43. May HT, Bair TL, Lappe DL, *et al.* (2010) Association of vitamin D levels with incident depression among a general cardiovascular population. *Am Heart J* **159**, 1037–1043.
44. Chan R, Chan D, Woo J, *et al.* (2011) Association between serum 25-hydroxyvitamin D and psychological health in older Chinese men in a cohort study. *J Affect Disord* **130**, 251–259.
45. Milaneschi Y, Hoogendijk W, Lips P, *et al.* (2014) The association between low vitamin D and depressive disorders. *Mol Psychiatry* **19**, 444–451.
46. Garcion E, Wion-Barbot N, Montero-Menei CN, *et al.* (2002) New clues about vitamin D functions in the nervous system. *Trends Endocrinol Metab* **13**, 100–105.
47. Obradovic D, Gronemeyer H, Lutz B, *et al.* (2006) Cross-talk of vitamin D and glucocorticoids in hippocampal cells. *J Neurochem* **96**, 500–509.
48. Antoniucci DM, Black DM & Sellmeyer DE (2005) Serum 25-hydroxyvitamin D is unaffected by multiple freeze-thaw cycles. *Clin Chem* **51**, 258–261.
49. Ocke MC, Schrijver J, Obermann-de Boer GL, *et al.* (1995) Stability of blood (pro)vitamins during four years of storage at − 20 degrees C: consequences for epidemiologic research. *J Clin Epidemiol* **48**, 1077–1085.
50. Perna L, Haug U, Schöttker B, *et al.* (2012) Public health implications of standardized 25-hydroxyvitamin D levels: a decrease in the prevalence of vitamin D deficiency among older women in Germany. *Prev Med* **55**, 228–232.
51. Sillanpää M, Andlin-Sobocki P & Lönnqvist J (2008) Costs of brain disorders in Finland. *Acta Neurol Scand* **117**, 167–172.