Vitamin status in morbidly obese patients: a cross-sectional study[1–3]

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ABSTRACT

Background: Morbid obesity is associated with low circulating concentrations of 25-hydroxyvitamin D. Few data on the concentrations of other vitamins in morbidly obese patients are available.

Objective: The objective was to compare serum and blood vitamin concentrations in morbidly obese patients with those in healthy subjects.

Design: In 2 public hospital departments (southeast Norway), we prospectively examined 110 consecutive patients (76 women) and 58 healthy controls (30 women) not taking multivitamin supplements. Patients and controls did not differ significantly in age or ethnicity. The mean (±SD) body mass index (in kg/m^2) was 45 ± 7 in the patients and was 24 ± 3 in the controls. Patients with vitamin concentrations lower than 2 SD below the sex-specific mean in controls were considered to have inadequate vitamin status.

Results: The morbidly obese women and men had significantly lower concentrations of vitamin B-6, vitamin C, 25-hydroxyvitamin D, and lipid-standardized vitamin E than did the healthy controls ($P < 0.01$ for each). The status of these vitamins was inadequate in a substantial proportion of the patients (11–38%). The status of vitamins A, B-1, B-2, and B-12 and of folic acid was adequate in most of the patients (95–100%). A moderately elevated C-reactive protein concentration was associated with lower vitamin A, B-6, and C concentrations. In a multiple regression analysis, concentrations of alkaline phosphatase (inverse relation) and vitamin C were the strongest determinants of serum vitamin B-6 concentrations.

Conclusions: Low concentrations of vitamin B-6, vitamin C, 25-hydroxyvitamin D, and vitamin E adjusted for lipids are prevalent in morbidly obese Norwegian patients seeking weight-loss treatment. *Am J Clin Nutr* 2008;87:362–9.

KEY WORDS Morbid obesity, body mass index, vitamin deficiency, vitamin B-1, riboflavin, pyridoxal phosphate, ascorbic acid, retinol, α-tocopherol, 25-hydroxyvitamin D

INTRODUCTION

Morbid obesity, which is defined as a body mass index (BMI; in kg/m^2) >40 or >35 with a weight related comorbidity, impairs quality of life (1), increases the risk of coronary heart disease (2), and shortens life expectancy (3). Currently, an estimated 7% of adult women in the United States have a BMI >40 (4). Although morbidly obese persons have greater intakes than do nonobese persons, morbidly obese persons may have nutritional deficiencies. The most common vitamin deficiency associated with obesity seems to be low concentrations of 25-hydroxyvitamin D (5, 6), which is associated with an increased risk of diabetes and other cardiovascular disease risk factors (7, 8) and depression (9). Few data about other possible vitamin deficiencies in morbidly obese patients are available (10), although their dietary habits obviously deviate from those of nonobese individuals (11).

Bariatric surgery is used with increasing frequency and is the therapeutic option that offers patients with morbid obesity the most pronounced and lasting weight reduction (12). However, bariatric surgical procedures may induce malabsorption and frequently result in nutritional deficiencies (13). The combination of a low preoperative vitamin concentration and the malabsorption that often follows bariatric surgery may render these patients prone to severe vitamin deficiencies.

Observational studies in the general population have shown associations between vitamin status and morbidity, beyond the traditional vitamin deficiency disorders. Low vitamin B-6 concentrations have been linked with symptoms of depression (14) and increased risk of stroke (15) and colorectal neoplasia (16). A low vitamin C concentration in plasma may be associated with increased all-cause mortality (17), risk of myocardial infarction (18), and gallbladder disease (19). The aim of the present study was to assess concentrations of vitamins A, B-1, B-2, B-6, C, D, and E in morbidly obese patients seeking weight reduction and to compare the concentrations with those observed in a healthy control group.

SUBJECTS AND METHODS

The study took place in 2 public hospitals in southeast Norway with departments specialized in the treatment of morbid obesity by intensive lifestyle treatment, bariatric surgery, or both. The Regional Ethics Committees for Medical Research approved the study protocol, and all participants gave informed written consent before enrollment.

Patients

Patients were referred from primary or secondary care and were offered an appointment if they had a BMI (in kg/m^2) >40

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or >35 in patients with obesity-related conditions associated with a high cardiovascular disease risk (such as sleep apnea or diabetes mellitus) or induce physical problems that interfere with lifestyle (joint disorders or body size problems interfering with employment, family function, or ambulation), in accordance with criteria commonly used when considering weight-loss surgery (20). Between December 2005 and April 2006, 132 patients seeking weight reduction were prospectively examined at their first visit. Patients were excluded if they used multivitamin supplements ($n = 10$), refused to comply with study procedures ($n = 2$), were >60 y of age ($n = 5$), were currently being treated for a severe psychiatric disorder ($n = 2$), had an alcohol intake >50 g/d ($n = 2$), or had thyroid abnormalities ($n = 1$, excessive thyroxine substitution). Ten patients undergoing thyroxine substitution who had normal serum concentrations of thyroid-stimulating hormone (TSH) and thyroxine were included. The final study sample consisted of 110 patients.

The medical history of the enrolled patients included depression ($n = 52$), diabetes ($n = 29$), previous cholecystectomy ($n = 7$), and a reversed gastric banding procedure ($n = 3$). Drugs currently being used by the subjects included blood pressure–lowering agents ($n = 36$), analgesics ($n = 18$), antidepressants ($n = 16$), metformin ($n = 15$), insulin ($n = 3$), aspirin ($n = 9$), antacids ($n = 8$), oral contraceptives ($n = 6$), sibutramin ($n = 1$), and orlistat ($n = 1$). Four patients not taking thyroxine had elevated concentrations of TSH (concentrations between 5.9 and 15.0 mU/L) and thyroxine concentrations in the normal range.

Controls

Healthy controls were recruited via local advertisements at Aker University Hospital. Between January and May 2007, 58 nonobese subjects (30 women) aged 19–59 y were examined. Exclusion criteria included chronic disease and regular medication or multivitamin supplement use. Subjects using contraceptive medication ($n = 10$) or thyroxine substitution ($n = 4$) were allowed to participate.

Clinical characteristics

Demographic data, medical history, and the use of tobacco, alcohol, and vitamin supplements were recorded on standardized forms. Height and weight were measured while the participants were wearing light clothing without shoes. Waist and hip circumferences were assessed with a tape measure at the midpoint between the lowest rib margin and the iliac crest and at the level of the major trochanter, respectively. Blood pressure was measured while the subjects were sitting after they had rested for 5 min. Patients with a history of diabetes ($n = 20$) or with a fasting plasma glucose concentration ≥7.0 mmol/L ($n = 9$) were classified as having diabetes. The homeostasis model assessment of insulin resistance was calculated in participants without diabetes from fasting concentrations of glucose and insulin by using the calculator from the Diabetes Trials Unit (Internet: http://www.dtu.ox.ac.uk; accessed 23 November 2006).

Preparation of blood samples

Blood was collected by venipuncture after an overnight fast. Samples clotted 30 min at room temperature, and serum was separated by centrifugation at 1700 × *g* for 10 min. Aliquots were immediately stored at −20 °C (−80 °C for assays of vitamin B-2, vitamin C, and 25-hydroxyvitamin D). Samples prepared at Vestfold Hospital were kept on dry ice (−57 °C) for up to 16 h during transportation to Aker University Hospital. Laboratory assays were performed within 10 d of blood sampling, except for vitamin B-2 (within 90 d) and 25-hydroxyvitamin D (within 1 y). We previously performed extensive assessment of protocols for preparing specimens for vitamin C analysis with respect to deterioration of ascorbic acid concentrations at different storage conditions. The results are highly reliable when serum aliquots are frozen at −80 °C within 2 h of blood sampling and analyzed within 2 wk.

Laboratory analysis

Routine laboratory analyses were performed in blood, serum, or plasma (eg, homocysteine) with a Hitachi 717 Modular multianalyzer (Boehringer Mannheim, Mannheim, Germany) in the Department of Clinical Chemistry at Aker University Hospital and with a Vitros 950 Chemistry System (Ortho-Clinical Diagnostics, Rochester, NY) at Vestfold Hospital. Data on folic acid, vitamin B-12, and homocysteine were only available for participants at Aker University Hospital. The assays for C-reactive protein (CRP) have detection limits of <1 and <7 mg/L at Aker and Vestfold, respectively; both assays were calibrated by using European Community Bureau of Reference Certified Reference Material 470 (CRM470). A reference population for the assessment of 25-hydroxyvitamin D status in our laboratory was described previously (21).

HPLC was used to assay vitamin B-1 (thiamine pyrophosphate in heparinized blood) (22), vitamin B-2 (flavin mononucleotide in EDTA-blood; Chromsystems, Munich, Germany), vitamin B-6 (pyridoxal-5'-phosphate in serum; Chromsystems), vitamin A (retinol in serum; Bio-Rad Laboratories, Munich, Germany), and vitamin E (α-tocopherol in serum; Bio-Rad Laboratories). Serum samples were analyzed for vitamin C (ascorbic acid) in an environment acidified with *ortho*-phosphoric acid according to the method of Zannoni (23). Serum was also analyzed for 25-hydroxyvitamin D (the sum of 25-hydroxyvitamin D_2 and 25-hydroxyvitaminD_3) and 1,25-hydroxyvitamin D by radioimmunoassay (DiaSorin, Stillwater, MN), intact parathyroid hormone by chemiluminoimmunometric assay (Diagnostic Products Corporation, Los Angeles, CA), and ionized calcium with a Rapidlab 348 analyzer (Instru-Med Inc, Atlanta, GA). The interassay CVs with these methods in our laboratories ranged from 3% to 9%, except for 25-hydroxyvitamin D (14%), based on an analysis of ≥12 replicate samples on 2 different days. Ascorbic acid and 25-hydroxyvitamin D were analyzed in duplicate, and the analysis of 25-hydroxyvitamin D was performed on the same day for patients and controls. All vitamin assays included quality controls with high and low concentrations supplied by the manufacturers plus internal controls. For vitamins B-1 and C, external controls were unavailable and standards were prepared from dry substances (Sigma-Aldrich, St Louis, MO). We observed no significant laboratory drift within the study period when reviewing the results of the quality controls.

Sample size and statistical analysis

The primary objective was to estimate the prevalence of inadequate vitamin concentrations in morbidly obese patients

seeking weight-loss treatment. Reference intervals for each vitamin were calculated separately for women and men based on the mean ± 2 SD in the control group. (Reference intervals for vitamins with a log normal distribution were obtained by calculating the mean ± 2 SD of log-transformed values and back-transforming the result). Vitamin inadequacy was defined as a vitamin concentration below the reference interval. Assuming vitamin inadequacy rates of 2.5% in the control group and 10% in the obese patients, we used an exact binomial test with a 0.05 significance level to estimate that a sample size of 110 patients would give 93% power to detect a difference between the groups.

Data are presented as means ± SDs unless otherwise noted. Skewed data were log transformed when appropriate for further statistical analysis (ie, for the comparison of means or calculation of reference intervals). A Fisher's exact test or chi-square test was used to compare categorical data between groups, a Student's *t* test or Mann-Whitney *U* test was used to compare continuous data between groups, and Spearman's rank correlation was calculated to explore relations between continuous variables. The degree of association between vitamin B-6 (log μmol/L) and various clinical variables was determined by linear regression models that included dichotomous and continuous predictor variables. Covariates were variables known to influence vitamin B-6 concentrations and significant predictors in univariate analyses. The final model was obtained using a backward stepwise method. No significant interactions between the remaining variables were identified. The level of significance was $P < 0.05$, and all *P* values are 2-tailed. No adjustment was made for multiple testing. Statistical analyses were done in SPSS 14.0 (SPSS Inc, Chicago, IL) for WINDOWS.

RESULTS

All participants were of Europoid origin, except for 4 patients and 1 control subject of Indian subcontinent origin. Mean age was not significantly different in the patient and control groups, who had mean BMIs of 45 ± 7 and 24 ± 3, respectively. The patients had a higher mean systolic blood pressure level and a less favorable lipid profile than did the controls (**Table 1**).

Vitamin status

Overall, vitamin status in the morbidly obese patients was strikingly different from that in the healthy control group, and significant differences were observed in 6 of 9 vitamins assayed. Compared with controls, the obese patients had significantly lower mean serum concentrations of vitamins A, B-6, C, 25-hydroxyvitamin D, and lipid-standardized vitamin E. Conversely, the patients had higher mean blood concentrations of vitamin B-1 than did the controls. These observations were consistent in both women and men (**Table 2**). Moreover, a substantial proportion of patients (11–38%) were considered to have an inadequate vitamin B-6, vitamin C, 25-hydroxyvitamin D, and lipid-standardized vitamin E status (**Figure 1**; *see* Supplemental Table 1 under "Supplemental data" in the issue online at www.ajcn.org). Female patients had a significantly higher risk of inadequate vitamin C concentrations and a lower risk of inadequate 25-hydroxyvitamin D concentrations than did male patients ($P < 0.001$ for both), whereas inadequacy rates for other vitamins were not significantly different between men and women (*P* values not shown). Most patients (95–100%) had adequate concentrations of vitamins A, B-1, B-2, and B-12 and

TABLE 1
Clinical status of morbidly obese patients and healthy controls

	Women			Men		
	Controls ($n = 30$)	Patients ($n = 76$)	P^1	Controls ($n = 28$)	Patients ($n = 34$)	P^1
Age (y)	39 ± 11[2]	41 ± 11	0.359	39 ± 11	41 ± 10	0.521
BMI (kg/m^2)	23 ± 3	45 ± 7	<0.001	25 ± 3	45 ± 7	<0.001
Waist circumference (cm)	72 ± 7	133 ± 13	<0.001	85 ± 9	145 ± 14	<0.001
Waist-hip ratio	0.74 ± 0.1	1.0 ± 0.1	<0.001	0.87 ± 0.1	1.1 ± 0.1	<0.001
Alcohol intake (g/d)	3.0 ± 3.3	1.6 ± 2.5	0.002	6.9 ± 6.8	1.7 ± 3.1	<0.001
Current smoker [*n* (%)]	1 (3.3)	24 (31.6)	0.002	4 (14.3)	10 (29.4)	0.156
Systolic blood pressure (mm Hg)	117 ± 9	136 ± 18	<0.001	122 ± 13	142 ± 21	<0.001
Diastolic blood pressure (mm Hg)	80 ± 7	84 ± 11	0.026	81 ± 10	87 ± 13	0.052
Hemoglobin (g/dL)	12.9 ± 0.9	13.8 ± 1.0	<0.001	14.6 ± 0.6	15.2 ± 1.2	0.006
Cholesterol						
Total (mmol/L)	4.6 ± 0.9	4.8 ± 1.1	0.429	4.9 ± 1.0	4.9 ± 1.0	0.939
HDL (mmol/L)	1.8 ± 0.3	1.2 ± 0.3	<0.001	1.4 ± 0.3	1.1 ± 0.2	<0.001
Triacylglycerols (mmol/L)	0.8 ± 0.3	1.8 ± 1.1	<0.001	1.0 ± 0.6	1.9 ± 0.8	<0.001
Alanine aminotransferase (U/L)	18 ± 8	34 ± 22	<0.001	23 ± 9	46 ± 35	<0.001
Albumin (g/L)	44.3 ± 2.2	41.5 ± 2.6	<0.001	44.9 ± 2.2	43.0 ± 2.5	0.003
Alkaline phosphatase (U/L)	54 ± 16	82 ± 23	<0.001	63 ± 16	82 ± 22	<0.001
Creatinine (μmol/L)	65 ± 7	59 ± 10	0.003	76 ± 10	76 ± 20	0.755
C-reactive protein (mg/L)	1.2 ± 1.5	14.6 ± 11.8	<0.001	1.3 ± 0.8	8.0 ± 4.5	<0.001
HOMA-IR[3]	1.2 ± 0.4	3.3 ± 1.8	<0.001	1.5 ± 1.1	3.8 ± 1.9	<0.001

[1] Means were compared by *t* test (skewed data were log transformed where appropriate), proportions by Fisher's exact test, and alcohol intake and C-reactive protein by Mann-Whitney *U* test.

[2] $\bar{x}$ ± SD (all such values).

[3] Homeostasis model assessment of insulin resistance in participants without diabetes.

TABLE 2
Vitamin concentrations in morbidly obese patients and healthy controls

			Women			Men		
Vitamin	Analyte	Unit	Controls ($n = 30$)	Patients ($n = 76$)	P^1	Controls ($n = 28$)	Patients ($n = 34$)	P^1
A	Serum retinol	μmol/L	1.9 ± 0.5[2]	1.7 ± 0.4	0.006	2.3 ± 0.6	1.9 ± 0.4	0.013
B-1	Blood thiamine pyrophosphate	nmol/L	99 ± 19	125 ± 34	<0.001	106 ± 17	151 ± 37	<0.001
B-1/Hb	B-1/hemoglobin	pmol/g Hb	766 ± 129	907 ± 247	0.004	732 ± 123	990 ± 227	<0.001
B-2	Blood flavin mononucleotide	nmol/L	20 ± 7	20 ± 9	0.886	25 ± 14	21 ± 7	0.247
B-6	Serum pyridoxal-5'-phosphate	nmol/L	46 ± 24	29 ± 30	<0.001	58 ± 31	39 ± 33	0.002
Folic acid	Serum folic acid	nmol/L	16 ± 4	16 ± 8[3]	0.383	16 ± 5	13 ± 3[3]	0.023
B-12	Serum cobalamin	pmol/L	307 ± 98	303 ± 69[3]	0.866	368 ± 116	331 ± 97[3]	0.526
	Plasma homocysteine	μmol/L	11 ± 2	11 ± 4	0.975	12 ± 2	13 ± 7	0.953
C	Serum ascorbic acid	mmol/L	74 ± 14	48 ± 18	<0.001	63 ± 15	48 ± 14	<0.001
D	Serum 25-hydroxyvitamin D	nmol/L	54 ± 22	40 ± 16	0.007	59 ± 20	34 ± 15	<0.001
	Serum parathyroid hormone	pmol/L	4.5 ± 1.9	6.7 ± 2.6	<0.001	4.0 ± 2.1	6.8 ± 3.2	<0.001
E	Serum α-tocopherol	μmol/L	26.6 ± 4.9	28.0 ± 6.9	0.325	29.4 ± 9.5	26.8 ± 6.1	0.202
E/lipids	E/(cholesterol + triacylglycerols)	μmol/mmol	5.0 ± 0.7	4.3 ± 0.8	<0.001	5.0 ± 0.8	4.0 ± 0.7	<0.001

[1] Means were compared by *t* test (vitamins A, B-2, B-6, D, and E; folic acid; and homocysteine were compared log transformed).
[2] $\bar{x}$ ± SD (all such values).
[3] $n = 35$ women and $n = 16$ men.

folic acid (*see* Supplemental Table 1 under "Supplemental data" in the issue online at www.ajcn.org).

Vitamin concentrations in patients related to BMI and vitamin B-6 status

Patients with a BMI above the median of 45 had significantly lower concentrations of vitamins B-6 and 25-hydroxyvitamin D than did patients with a BMI <45 (**Table 3**). Inadequate vitamin B-6 status was associated with lower concentrations of several other vitamins (Table 3) and higher alkaline phosphatase concentrations: 98 ± 25 compared with 76 ± 19 U/L ($P < 0.001$). Antidepressant medication use was more frequent among female patients ($n = 15$, 20%) than among male patients ($n = 1$; 3%; $P = 0.020$). Women using antidepressants had a lower mean vitamin B-6 concentration than did the women not taking such drugs: 17 ± 7 compared with 32 ± 32 nmol/L ($P = 0.018$).

Vitamin concentrations related to inflammation

Whereas all control subjects had CRP concentrations <8 mg/L, 65 patients (59%) had moderately elevated CRP concentrations, with concentrations ranging from 8 to 69 mg/L. This finding was more frequent in women ($n = 52$; 68%) than in men ($n = 13$; 38%) ($P = 0.003$). Patients with moderately elevated CRP concentrations had significantly lower mean concentrations of vitamins A, B-6, and C than did patients with lower CRP concentrations (Table 3).

Vitamin concentrations related to other patient characteristics

Homocysteine concentrations (Table 2) were negatively correlated with folic acid concentrations (rank correlation: −0.38, $P = 0.006$) and not significantly correlated with vitamin B-2, B-6, or B-12 concentrations (data not shown). Parathyroid hormone concentrations were negatively correlated with 25-hydroxyvitamin D concentrations (rank correlation: −0.32, $P = 0.001$). All patients had normal concentrations of ionized calcium ($n = 51$) and 1,25-hydroxyvitamin D (data not shown).

Vitamin concentrations were not significantly associated with age and were not different between groups according to smoking habit or diabetes status. However, in patients without diabetes, vitamin B-6, vitamin C, and 25-hydroxyvitamin D were all negatively correlated with insulin resistance estimated by the homeostasis model assessment (rank correlations: −0.28, −0.34, and −0.28, respectively; $P \leq 0.01$ for each). Patients consuming alcohol (range: 1–14 g/d) had significantly higher concentrations of vitamins B-6 and C than did nonconsumers (data not shown).

Determinants of vitamin B-6 concentrations

Determinants of serum vitamin B-6 concentrations in the obese patients were evaluated in multiple linear regression models. The initial model included variables previously reported to be associated with vitamin B-6 concentrations (sex, age, smoking habit, alcohol intake, and concentrations of alkaline phosphatase, phosphate, albumin, creatinine, CRP, and vitamin B-2) and variables that were significant predictors of inadequate vitamin B-6 concentrations in a univariate analysis (BMI, antidepressant medication use, and concentrations of vitamin C and hemoglobin-adjusted vitamin B-1). The variable with the highest *P* value was removed stepwise until all remaining variables had *P* values <0.05. Of the remaining variables, the strongest determinants of vitamin B-6 concentrations were alkaline phosphatase (inverse relation) and vitamin C concentrations (**Table 4**). The procedure was then repeated with sex forced to be kept in the model. The results were essentially the same as those in Table 4, except that, in the new model, antidepressant use was nearly significant ($P = 0.066$; sex: $P = 0.507$). Finally, we tested the model including females only. Results were again consistent with those reported in Table 4 ($R^2 = 0.54$; antidepressant use: $P = 0.037$).

DISCUSSION

The major novel finding of this study of 110 morbidly obese patients referred for weight-loss treatment was a high prevalence

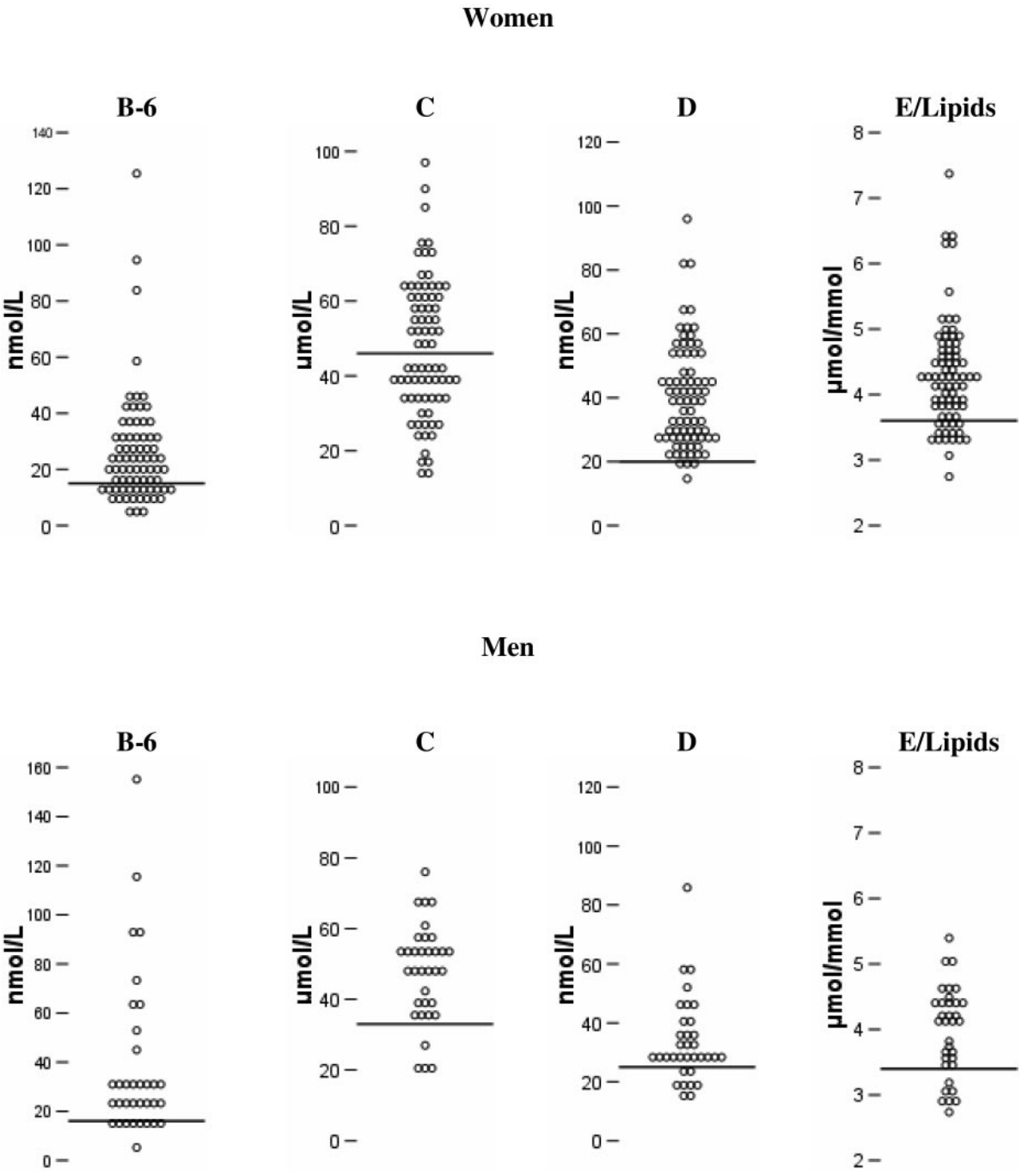

FIGURE 1. Vitamin status in morbidly obese women and men. Horizontal bars indicate the lower limit of the normal reference interval. Reference intervals were derived from the mean ± 2 SD of healthy individuals (n = 30 women and 28 men) who did not use multivitamin supplements. Vitamin concentrations were not normally distributed for vitamins B-6 and D; reference intervals for these vitamins were obtained by calculating the mean ± 2 SD of log-transformed values and backtransforming the result. For exact intervals, *see* Supplemental Table 1 under "Supplemental data" in the issue online at www.ajcn.org. B-6, serum pyridoxal-5'-phosphate; C, serum ascorbic acid; D, serum 25-hydroxyvitamin D; E/Lipids, serum α-tocopherol/(cholesterol + triacylglycerols). A female patient with an exceedingly high vitamin B-6 concentration (220 nmol/L) was omitted from the graph.

of low vitamin B-6, C, D, and E concentrations, ranging from 11% to 38% of patients. Furthermore, the most obese patients had the most pronounced reductions in vitamin concentrations, antidepressant medication use was associated with lower vitamin B-6 concentrations in women, and lower concentrations of 25-hydroxyvitamin D were associated with higher concentrations of parathyroid hormone. Few patients had low vitamin B-1 concentrations; rather, the patients had somewhat higher mean concentrations of vitamin B-1 than did the controls. The reason for this is not known.

The study included consecutive patients who visited 2 public healthcare institutions and whose characteristics were

TABLE 3
Vitamin concentrations in morbidly obese patients related to BMI, vitamin B-6 status, and C-reactive protein (CRP)[1]

	BMI			Vitamin B-6 status[2]			CRP[3]		
	<45 kg/m^2 (n = 55)	≥45 kg/m^2 (n = 55)	P[4]	Normal (n = 81)	Inadequate (n = 29)	P[4]	≤7 mg/L (n = 45)	8–69 mg/L (n = 65)	P[5]
Women [n (%)]	39 (71)	37 (67)	0.680[6]	54 (67)	22 (76)	0.358[6]	24 (53)	52 (80)	0.003[6]
Age (y)	41 ± 12[7]	41 ± 9	0.978	41 ± 11	41 ± 9	0.837	40 ± 10	42 ± 11	0.376
Vitamin									
A (μmol/L)	1.8 ± 0.5	1.7 ± 0.3	0.032	1.8 ± 0.4	1.6 ± 0.4	0.020	1.9 ± 0.4	1.6 ± 0.4	<0.007
B-1/Hb (pmol/g Hb)	980 ± 247	884 ± 232	0.038	965 ± 240	842 ± 236	0.019	962 ± 239	913 ± 247	0.542
B-2 (nmol/L)	20 ± 10	20 ± 7	0.919	21 ± 9	18 ± 7	0.097	19 ± 7	21 ± 9	0.070
B-6 (nmol/L)	37 ± 37	26 ± 22	0.021	39 ± 33	11 ± 3	—	37 ± 30	28 ± 31	0.036
Folic acid (nmol/L)[8]	16 ± 8	14 ± 5	0.382	15 ± 6	14 ± 8	0.463	13 ± 4	16 ± 9	0.678
B-12 (pmol/L)[8]	324 ± 76	310 ± 86	0.483	314 ± 74	322 ± 94	0.718	330 ± 94	307 ± 69	0.658
C (mmol/L)	49 ± 19	46 ± 15	0.331	50 ± 16	40 ± 17	0.004	52 ± 16	45 ± 17	0.018
D (nmol/L)	42 ± 18	34 ± 12	0.015	40 ± 16	34 ± 15	0.120	40 ± 19	37 ± 14	0.370
PTH (pmol/L)	6.1 ± 2.7	7.3 ± 2.8	0.016	6.8 ± 2.9	6.4 ± 2.3	0.555	6.5 ± 3.0	6.8 ± 2.6	0.499
E/lipids (μmol/mmol)	4.3 ± 0.8	4.1 ± 0.8	0.101	4.2 ± 0.8	4.3 ± 0.9	0.381	4.3 ± 0.8	4.2 ± 0.8	0.099

[1] Vitamins A, B-2, B-6, and D and folic acid were compared log transformed. Hb, hemoglobin; PTH, parathyroid hormone; lipids, cholesterol + triacylglycerols.
[2] Normal: ≥15 nmol/L; inadequate: <15 nmol/L.
[3] CRP ≤ 7 mg/L was used as a cutoff because it was the lower detection limit in one of the laboratories.
[4] Means were compared by t test.
[5] Means were compared by 2-factor ANOVA; P values were adjusted for sex. Significant effect of sex: vitamin A (P = 0.010), vitamin D (P = 0.018), and vitamin E/lipids (P = 0.019).
[6] Chi-square test.
[7] $\bar{x}$ ± SD (all such values).
[8] n = 24 and 27 (BMI), n = 33 and 18 (vitamin B-6 status), and n = 22 and 29 (CRP).

comparable with those of large populations of candidates for bariatric surgery (12). Vitamin concentrations in erythrocytes or serum are routinely used as indicators of tissue stores (vitamins A, B-1, B-6, D, and E) or recent intakes (vitamin C). Because obese persons often underreport their energy intakes (24), vitamin status was only assessed in blood. In general, care must be taken in interpreting vitamin status assessed by blood concentration measurements because external quality assurance schemes and control materials with defined values are not always available (25). Serum concentrations of 25-hydroxyvitamin D may be lower during the winter season at our latitude (60° N). In the present study, vitamin concentrations in the obese patients were compared with findings in individuals not different in age or ethnicity and examined in the same laboratory during the same season. If the reference interval normally used for 25-hydroxyvitamin D in our laboratory (37–131 nmol/L) had been applied, 55% of patients would have had inadequate vitamin D status (Figure 1). Patients and controls were examined 1 y apart and laboratory drift was therefore a potential confounder; however, we found no evidence of such a problem in reviewing the laboratory quality controls for the 2 periods.

TABLE 4
Multiple regression analysis of determinants of serum vitamin B-6 concentrations (log μmol/L) in 120 patients[1]

Variable	Adjusted effect (95% CI)	P
Alkaline phosphatase (U/L)	−0.010 (−0.005, −0.015)	<0.001
Vitamin C (mmol/L)	0.011 (0.005, 0.018)	0.001
Creatinine (μmol/L)	0.010 (0.003, 0.017)	0.005
Phosphate (mmol/L)	0.817 (0.228, 1.407)	0.007
Vitamin B-2 (nmol/L)	0.014 (0.001, 0.027)	0.029
Antidepressant medication (yes vs no)	−0.327 (−0.015, −0.638)	0.040

[1] R^2 = 0.42 with all variables included. Adjusted effects are β.

Most former studies of vitamin status in obesity have not focused on severe obesity but included subjects with a wide range of BMIs. Vitamin B-6 status was adequate in some (26, 27), but not all (28), reports. An increase in BMI was associated with lower concentrations of vitamins A, C, and E in the United States (29), Western Europe (30–33), and elsewhere (26, 27, 34). In contrast with 2 previous reports (10, 35), low concentrations of vitamin B-1 were rare in our study. However, a comparison of these findings is difficult because the analytic methods were not described. Vitamin B-2 status was normal in obese Polish (26) and Thai (27) subjects. Vitamin D deficiency and secondary hyperparathyroidism are known to be prevalent in obesity (5, 6). Thus, our findings are generally consistent with previous reports and add knowledge regarding several vitamins in patients with morbid obesity.

The low vitamin concentrations observed in the morbidly obese patients could have been caused by several mechanisms. Dietary and lifestyle habits may be the most important contributors. A low intake of fruit, vegetables, and alcohol has been described in severe obesity (11). Current smoking was more frequent in the obese patients than in the control group, and this might in part explain their lower concentrations of ascorbic acid in serum (30). Body composition may also influence vitamin status; low plasma vitamin C concentrations have been related to a central fat distribution independent of BMI (33), and the low serum concentrations of 25-hydroxyvitamin D in obesity may

partly be a result of sequestration in adipose tissue (36, 37). Obese individuals have elevated amounts of total body water, and the extracellular compartment is relatively more expanded than the intracellular compartment (38). Possibly, this leads to dilution effects on extracellular vitamin concentrations; the obese patients generally had lower serum vitamin concentrations, whereas blood (erythrocyte) concentrations were not different from, or higher than, those in controls (Table 2). No patient had a clinical condition associated with malabsorption, and only one patient currently used orlistat, which may induce malabsorption. Alkaline phosphatase hydrolyzes pyridoxal-5'-phosphate and is a major determinant of vitamin B-6 concentrations in serum (39). Elevated concentrations of alkaline phosphatase have also been reported in obese patients (40).

Importantly, systemic inflammation is associated with reduced serum concentrations of vitamins A (41), B-2 (42), B-6 (42, 43), and C (44), owing to reduced liver production of transport proteins (such as albumin), increased turnover of antioxidant vitamins, or a shift in tissue distribution. A low serum vitamin concentration in inflammation does not necessarily indicate that body stores are depleted (41). Obesity is associated with chronic low-grade inflammation (45). Previous studies have also found moderately elevated CRP concentrations to be more common in women with morbid obesity than in men (46). However, we also observed low concentrations of vitamins B-6, C, and D in patients with lower CRP concentrations. In an animal model, vitamin B-6–deficient rats had increased liver oxidative stress (47), and if also present in humans, such a mechanism could partly explain the relation between serum concentrations of vitamins B-6 and C in our patients.

Morbidly obese individuals have an increased prevalence of several diseases that have been related to a suboptimal status of vitamins B-6, C, and D in other populations, including cardiovascular disease (18, 48), colorectal neoplasia (16, 49), and depression (9, 14). The optimal serum concentration of 25-hydroxyvitamin D may be >75 nmol/L (49, 50). Such a concentration was found in only 4% of the morbidly obese patients (Figure 1). An inverse association between serum concentrations of 25-hydroxyvitamin D and diabetes has been reported (7). This agrees with our finding of a negative correlation between the magnitude of insulin resistance and 25-hydroxyvitamin D concentrations.

Future research

Cross-sectional studies such as the present study are important in generating knowledge for further research but cannot establish cause-and-effect relations, and most knowledge regarding vitamin status and disease risk comes from observational studies, which are often limited by confounders. Randomized trials are therefore necessary to evaluate whether morbidly obese patients with low vitamin concentrations benefit from supplementation. It is notable that vitamin B-6 concentrations were normal at baseline in a controlled trial in which supplementation provided no benefit (51). Possibly, patients with a low baseline concentration have a greater potential for benefit. After bariatric surgery, the low vitamin concentrations observed in our study may potentially be further reduced and cause severe adverse reactions (13). This highlights the importance of controlled trials to determine appropriate monitoring and supplementation for such patients.

Conclusion

We compared vitamin status in patients with clinically severe obesity with that in healthy controls. Selection bias was minimized by studying consecutive patients visiting public healthcare services. The limited size of the control group was a weakness of the study, and care should be taken in generalizing our findings to regions with differences in lifestyle and dietary habits. We conclude that morbidly obese Norwegian patients seeking weight loss may have low circulating concentrations of several vitamins, including 25-hydroxyvitamin D, vitamin B-6, vitamin C, and lipid-adjusted vitamin E.

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