Long-Term Ovo-Lacto Vegetarian Diet Impairs Vitamin B-12 Status in Pregnant Women

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A well-planned vegetarian diet has been stated to be adequate during pregnancy. The aim of the present study was to compare serum vitamin B-12 and homocysteine concentrations in pregnant women (n = 109) consuming vegetarian and Western diets and to evaluate the adequacy of current dietary reference intakes of vitamin B-12 for these women. Pregnant women adhering to vegetarian diets for at least 3 y, with subgroups of ovo-lacto vegetarians (OLVs; n = 27), low-meat eaters (LME, n = 43), and women eating an average Western diet (control group, n = 39), were recruited. Dietary vitamin B-12 intake, serum vitamin B-12, and plasma total homocysteine (tHcy) concentrations were measured in wk 9–12, 20–22, and 36–38 of pregnancy. During pregnancy serum vitamin B-12 concentrations of ovo-lacto vegetarians ($P < 0.001$) and low-meat eaters ($P = 0.050$) were lower than those of the control group. We observed the combination of low serum vitamin B-12 concentrations and elevated plasma tHcy in 22% of ovo-lacto vegetarians, in 10% of low-meat eaters, and in 3% of controls ($P = 0.003$). In OLVs, serum vitamin B-12 predicted 60% of the plasma tHcy variation ($P < 0.001$), but in LMEs and controls only <10% (NS). Serum vitamin B-12 concentrations increased and plasma tHcy decreased sharply with increasing dietary intake of vitamin B-12 toward a cutoff point of 3 µg/d. Pregnant women consuming a long-term predominantly vegetarian diet have an increased risk of vitamin B-12 deficiency. Current recommended dietary intakes urgently need reevaluation.

pregnancy, vitamin B-12, homocysteine, vegetarian diets

Topic:

pregnancy homocysteine diet meat plasma vegetarianism vitamin b12

Issue Section:

Nutrient Requirements

Natural sources of vitamin B-12 in human diets are restricted to foods of animal origin (1,2). Vegetarian diets, characterized by a reduced consumption of foods of animal origin and therefore lower intakes of vitamin B-12, are popular (3). One of the reasons for the increased popularity of vegetarian diets is the recent reports of lower morbidity and mortality rates from several chronic degenerative diseases in vegetarians relative to nonvegetarians (4–10). A well-planned lacto-ovo-vegetarian diet is often stated to be adequate during pregnancy, i.e., to meet the vitamin B-12 needs of both the mother and the fetus (11,12). Nevertheless, some studies have demonstrated a vitamin B-12 deficiency in infants born to mothers adhering to a strict vegetarian (vegan) diet (13–15). During pregnancy, low serum vitamin B-12 concentration is an independent risk factor for neural tube defects, pre-eclampsia, and other pregnancy-related complications (16–19). Low serum vitamin B-12 concentrations also lead to consequences for the mother such as macrocytic anemia, neurological complications, and cognitive disabilities (20). The breast-fed infant of a vitamin B-12-deficient mother is at risk for severe developmental abnormalities, growth failure, and anemia (21). Despite the increased risk of vitamin B-12 deficiency in utero, the intake of vitamin B-12 during pregnancy is not the focus of nutritional research because in mixed Western diets with high intake of foods of animal origin the intake of vitamin B-12 is well above the current recommended dietary intakes (22–24). There is evidence of increased intestinal absorption of vitamin B-12 during pregnancy, and fetal needs are thought to be very low (25,26). On the other hand, it has been speculated that vitamin B-12 from maternal tissue stores does not cross the placenta (13,14).

The dietary reference intakes for pregnant women are calculated mainly on the basis of data on fetal deposition and increased maternal absorption (24). The estimated average requirement (EAR)3 for vitamin B-12 intake is 2 µg/d for nonpregnant women. For pregnant women, the EAR is only increased by 0.2 µg/d (24). Despite this definition of EAR, convincing studies on vitamin B-12 requirements during pregnancy are lacking. Information about the consequences of long-term vegetarian and predominantly vegetarian diets, characterized by relatively low intake of vitamin B-12 (14), is therefore urgently needed.

The aim of the present study was to compare vitamin B-12 status in healthy pregnant women adhering to a predominantly vegetarian and plant-based diet for a long period with that of women consuming an average Western diet. In addition, the occurrence of vitamin B-12 deficiency, defined as low serum vitamin B-12 accompanied by elevated plasma total homocysteine (tHcy) concentrations, and their correlation to the dietary intake of vitamin B-12 was investigated.

SUBJECTS AND METHODS

Study design and subjects.

The present study was designed as a prospective cohort study in which pregnant women were monitored. Subject recruitment and selection has been described in detail elsewhere (27). Briefly, pregnant women of each trimester of pregnancy responding to announcements in health magazines and to study information handed out by their gynecologists were recruited for this study. Included were pregnant women who were apparently healthy, with <4 prior pregnancies. Participants could enter the study at any stage of pregnancy and were followed until delivery. Information on dietary intake and blood samples was collected in wk 9–12, 20–22, and 36–38 of gestation. A questionnaire regarding their nutritional behavior, food consumption (semi-quantitative food frequency list, considering the usual dietary intake before pregnancy), anthropometric and sociodemographic data, use of oral contraceptives, parity, smoking, and physical activities, as well as prevalent diseases, was sent to all participants.

From 1995 to 1997 pregnant women in each trimester of pregnancy were recruited and divided into 3 diet groups: women adhering to a plant-based diet, subdivided into ovo-lacto vegetarians and low-meat eaters, and women consuming an average Western diet (control group). Using a priori defined selection criteria subjects were assigned to diet groups. A total of 109 women participated in the study: 70 women consuming a plant-based diet (27 ovo-lacto vegetarians, 43 low-meat eaters) and 39 controls. Women consuming a plant-based diet were characterized by a high consumption of raw vegetables (>100 g/d), preference of whole-grain products (ratio of refined grain products/whole-grain products < 0.95) and limited meat consumption (meat < 300 g/wk; meat products < 105 g/wk). Of these, the subgroup of ovo-lacto vegetarians were defined as subjects who completely omitted meat and meat products from their diets. Participants in both of these groups were only included if they had not changed their diet substantially for at least 3 y. The diet of the control group was similar to that of the average German population as defined in the German National Consumption Study (28). Briefly, this diet consisted mainly of refined grain products (ratio of refined grain products/whole-grain products > 1.05) and of >300 g meat and 105 g meat products per week and <100 g unheated vegetables per day; participants in the control group were only included if they did not follow any special diet.

Due to pregnancy-related and organizational reasons, of 109 participants only 60 were assessed 3 times throughout pregnancy. After the first trimester 1 ovo-lacto vegetarian and 1 low-meat eater and after the second trimester 1 ovo-lacto vegetarian dropped out from the study after a miscarriage. After the second trimester 7 ovo-lacto vegetarians, 9 low-meat eaters, and 5 women from the control group were not assessed because of relocation or birth of the child before the last blood sampling date.

The study was approved by the Ethics Committee of the Division of Human Medicine, University of Giessen, Germany. Written informed consent was obtained from all participants.

Dietary assessment.

In wk 9–12, 20–22, and 36–38 of gestation, participants recorded their dietary intake for 4 d (Sunday through Wednesday) using a validated semi-quantitative food record. The validation procedure was described previously (27,29,30). Most participants recorded food intake preceding blood sampling, but for organizational reasons, 7 women recorded their food intake immediately after blood sampling. The food record included categories of 152 food items with descriptions and photographs of portion sizes estimated by typical household measures (27,29,30). Food supplement and medical drug intakes were assessed using a study diary during the whole study period.

At the time of this study, no systematic food fortification was carried out in Germany except for multivitamin-fortified juices. Calculation of dietary folate and vitamin B-12 intake was performed using the German Food Code and Nutrition Data Base BLS II.3 (31); vitamin B-12 concentrations in multivitamin-fortified juices were taken from producers' data.

Blood analyses.

In wk 9–12, 20–22, and 36–38 of gestation venous blood samples were drawn after an overnight fast into vacutainers with and without EDTA. Within 2 h of venepuncture, serum/plasma was separated from blood cells by centrifugation at 2000 × g for 10 min and then stored frozen (−18°C) until assayed. To quantify erythrocyte folate concentrations, a chemiluminescent competitive protein binding assay (ACS Folate Assay, Ciba Corning Diagnostics GmbH) was used (CV 3.9%). tHcy in plasma was determined according to Ubbink et al. (32) and Araki and Sako (33) (CV 4.0%). Serum vitamin B-12 concentrations were determined using the IMx cobalamin assay (Abbot Diagnostics Division), which incorporates microparticles coated with porcine intrinsic factor to bind cobalamin. Holotranscobalamin (holo-TC) was removed from whole serum by a modification of the method recommended by Das et al. (34), as described by Wickramasinghe and Fida (35). Briefly, a slurry containing synthetic amorphous precipitated silica (Sipernet 283 LS, PQ Corporation) in distilled water was prepared and stored at 4°C. Holo-TC was absorbed by adding silicea to serum, vortexing the mixture, and leaving it for 10 min. The mixture was centrifuged, and the supernatant fluid was assayed for holohaptocorrin (holo-HC) by the IMx cobalamin assay (CV serum vitamin B-12 4.2%; holo-HC 4.3%).

Statistical analyses.

All statistical analyses were performed using SAS 8.2 (SAS Institute). All values are arithmetic means ± SEM or medians with 25th and 75th percentiles in parentheses. BMI was calculated as reported pregravid weight/height (kg/m2).

The vitamin B-12 status of the diet groups was compared using the SAS generalized estimating equation (GEE) procedure. GEE models allow an appropriate analysis of longitudinal data with repeated measurement and missing values. For all models a dependent working correlation matrix was chosen. Data were log-transformed to normalize distribution of serum vitamin B-12. The risk of vitamin B-12 deficiency and the risk of elevated plasma tHCY levels were computed by a logistic regression analysis with repeated measurement design (GEE), and odds ratios and 95% confidence intervals are presented. Linear regression analysis was performed to calculate variance of tHCY explained by serum vitamin B-12 and erythrocyte folate. For this analysis, the mean blood concentrations during pregnancy were used. Cutoff values for low plasma vitamin B-12 concentrations were defined according to Koebnick et al. (36) as lower 95% confidence limits in omnivorous subjects for each trimester: <130 pmol/L in the first trimester, <120 pmol/L in the second trimester, and <100 pmol/L in the third trimester. Elevated plasma tHcy levels were defined as upper 95% confidence intervals in omnivorous healthy pregnant subjects based on data from Murphy et al. (37): >9.0 µmol/L in the first trimester and >7.8 µmol/L in the second and third trimesters.

RESULTS