

Dietary Factors of One-Carbon Metabolism in Relation to Non-Hodgkin Lymphoma and Multiple Myeloma in a Cohort of Male Smokers

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Abstract

Reported associations between genetic polymorphisms in folate-metabolizing enzymes and lymphoid malignancies suggest etiologic involvement of one-carbon metabolism and its related dietary exposures. We examined dietary factors of one-carbon metabolism in relation to non-Hodgkin lymphoma (NHL) and multiple myeloma (MM) among 27,111 healthy male smokers who completed baseline dietary questionnaires in the Alpha-Tocopherol Beta-Carotene Cancer Prevention Study cohort. During a follow-up of up to 16.4 years (1985-2001), 195 NHL and 32 MM cases were ascertained. Cox proportional hazard models were used to estimate multivariable-adjusted hazard ratios (HR) and 95% confidence intervals (95% CI). There was no significant association between

dietary folate and NHL (HR comparing fourth to first quartile, 1.03; 95% CI, 0.68-1.55). Dietary vitamin B₁₂ was inversely associated with NHL (HR, 0.61; 95% CI, 0.37-1.00; $P_{\text{trend}} = 0.06$). The inverse association of vitamin B₁₂ was evident for diffuse subtype but did not reach statistical significance. There were no significant associations of dietary vitamin B₆ or B₂, methionine, or alcohol with NHL. None of the dietary or supplemental one-carbon nutrients were associated with MM, although the power of these analyses was limited. Our results suggest that high intake of vitamin B₁₂ among heavy smokers may be protective against NHL but warrant further studies, including among nonsmokers. (Cancer Epidemiol Biomarkers Prev 2006;15(6):1109-14)

Introduction

Malignancies of lymphoid tissue origin, which are composed of non-Hodgkin lymphoma (NHL), Hodgkin lymphoma (HL), multiple myeloma (MM), and lymphoid leukemia (1), together rank fifth in U.S. cancer incidence with 93,420 new cases expected to occur during 2005 (2). NHL, the most common lymphoid malignancy, presented an estimated 50% increase in age-adjusted incidence from 1970 to 1990 (1) that is not well explained by established immunologic, viral, hereditary, or chemical risk factors, or changes in diagnostics and classification (3), warranting further investigation of historically underexplored common etiologic factors, such as nutrition.

Genetic polymorphisms in one-carbon metabolism pathways have been associated with risk for adult lymphomas (4-8) and leukemias (7, 9, 10). One-carbon metabolism, which refers to intracellular single-carbon transfer reactions mediated by folate and other nutrients (11), is essential for chromosomal stability and optimal DNA methylation patterns and, therefore, its impairment may be involved in lymphomagenesis (12). To date, epidemiologic data on specific nutrients involved in one-carbon metabolism and their associations with lymphoid malignancies are scarce. The only prospective study, to our knowledge, reported a null association between dietary

folate and NHL in U.S. women (13). Recently, we have reported inverse associations of dietary vitamin B₆ and methionine with overall NHL and of dietary folate with diffuse subtype of NHL in a case-control study (14). In the same study, we also found that alcohol consumption compared with no consumption was associated with a lower risk of NHL, which is consistent with most case-control studies of NHL to date (15) but is contradictory to the current understanding from epithelial cancer studies that alcohol interferes with one-carbon metabolism and, therefore, may increase the risk of cancer (16).

In the present study, we investigated the association of dietary factors related to one-carbon metabolism with the risk of NHL and MM among the Finnish male smokers of the Alpha-Tocopherol Beta-Carotene Cancer Prevention (ATBC) Study cohort.

Materials and Methods

Study Population. The ATBC Study was a double-blinded randomized trial that tested whether α -tocopherol or β -carotene supplements reduce the incidence of lung and other cancers in male smokers (17). Details of the trial have previously been described (17). Between 1985 and 1988, 29,133 Finnish men of ages 50 to 69 years, who smoked five or more cigarettes per day, were randomized to receive an intervention (supplements of α -tocopherol, 50 mg/d; β -carotene, 20 mg/d; or both) or placebo in a 2 \times 2 factorial design. Exclusion criteria included malignancies other than nonmelanoma skin cancer or carcinoma *in situ*, severe angina on exertion, chronic renal insufficiency, liver cirrhosis, chronic alcoholism, anticoagulant therapy, other medical problems that might limit long-term participation, and current use of supplements containing vitamin E (>20 mg/d), vitamin A (>20,000 IU/d), or β -carotene (>6 mg/d). The trial continued

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Note: L. Teerenhovi is deceased.

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until April, 1993, with the cohort followed up through national registers thereafter (17). The study was approved by the institutional review boards at the National Public Health Institute in Finland and the U.S. National Cancer Institute. All study participants provided written informed consent.

Data Collection on Baseline Characteristics, Lifestyle, and Dietary Intake. Study participants completed questionnaires at baseline, which provided information on general characteristics and medical, smoking, and dietary history. Diet was assessed by a self-administered dietary history questionnaire developed specifically for the ATBC Study and validated against food records in a pilot study conducted among middle-aged Finnish men (18). Frequency and usual portion size of 276 food items consumed over the previous 12 months were measured with aid of portion size picture guides, and nutrient intake was estimated through linkage with the food composition database of the National Public Health Institute in Finland (17).

Case Ascertainment. During the follow-up up to 16.4 years (median 13.2 years) through April 2001, incident lymphoid malignancies were identified from the Finnish Cancer Registry, which provides close to 100% case ascertainment in Finland (19, 20). Based on the pertinent case information (International Classification of Disease-Oncology second edition; ICD-O-2) and the WHO classification (21), we grouped all hematopoietic cancers into lymphoid and nonlymphoid origins and divided the lymphoid cancers into NHL (ICD-O-2 9590-9595, 9670-9677, 9680-9688, 9690-9698, 9700-9715, 9823),

MM (ICD-O-2 9732), and HL (ICD-O-2 9650, 9652-9655, 9657-9667). Using the scheme provided by Groves et al. (22) and considering the frequencies in the ATBC Study, we classified NHL further into four subtypes of diffuse lymphoma, follicular lymphoma, small lymphocytic lymphoma or chronic lymphocytic leukemia, and T-cell lymphoma.

Of the 29,133 men recruited in the trial, 27,111 individuals provided complete dietary and smoking history and were included in the current analysis. During the 323,232 person-years of follow-up until April 30, 2001, 293 men were diagnosed with incident hematopoietic cancers, including 238 lymphoid malignancies (195 NHL, 32 MM, and 11 HL). NHL was further classified into diffuse ($n = 54$), follicular ($n = 16$), small lymphocytic lymphoma or chronic lymphocytic leukemia ($n = 65$), T-cell lymphomas ($n = 17$), and other and unspecified types ($n = 43$).

Statistical Analyses. All nutrient and food group variables (except alcohol and supplements) were energy adjusted using the residual method (23), in which each nutrient or food variable was transformed to have a normal distribution to meet the assumption of linear regression for the residual adjustment. We conducted separate analyses for each one-carbon nutrient intake from food and supplements.

Baseline characteristics were examined by intake levels of each one-carbon nutrient (Table 1) and by case/noncase status (Table 2) to identify potential confounders. General linear model regression was used to estimate age-adjusted means of baseline characteristics for each quartile of energy-adjusted

Table 1. Baseline age-adjusted characteristics (means and proportions) by energy-adjusted dietary vitamin B₁₂ quartiles, ATBC Study cohort, 1985-2001 (N = 27,111)

Characteristics	Energy-adjusted dietary vitamin B ₁₂			
	Quartile 1 (1.68-8.05)	Quartile 2 (8.06-10.11)	Quartile 3 (10.12-12.87)	Quartile 4 (12.88-62.02)
Dietary vitamin B ₁₂ (μg/d)	6.56	9.06	11.37	16.03
Age (y)	57.3	57.4	57.0	57.0
Body mass index (kg/m ²)	26.0	26.2	26.4	26.5
Height (cm)	173.3	173.5	173.7	174.0
Smoking history				
Cigarettes/d	20.1	20.3	20.4	20.8
Years smoked	36.0	35.9	35.9	36.0
Education				
Elementary school or less (%)	83	81	78	72
Up to junior high school (%)	12	13	14	17
High school or more (%)	5	6	8	11
Serum HDL (mmol/L)	1.18	1.19	1.20	1.21
Dietary intake (per day)*				
Folate (μg)	313	318	328	345
Vitamin B ₆ (mg)	2.26	2.37	2.48	2.67
Vitamin B ₂ (mg)	2.42	2.78	2.94	3.14
Methionine (g)	1.76	1.96	2.07	2.19
Alcohol (g)	15.6	17.8	19.1	19.4
Calories (kcal)	2,788	2,828	2,844	2,800
Protein (g)	92	99	103	108
Animal protein (g)	58	68	72	78
Fat (g)	101	101	99	99
ω-3 fatty acids from fish	0.31	0.39	0.48	0.61
Vitamin D (μg)	3.5	4.5	5.3	6.5
Supplement use (%)				
Folate	5.6	5.8	5.9	6.4
Vitamin B ₁₂	6.8	7.5	7.2	7.9
Vitamin B ₆	11.9	12.6	13.1	14.1
Vitamin B ₂	10.3	10.7	11.6	12.2
Foods (g/d)*				
Dairy	599	739	767	744
Fish	17.4	27.7	35.9	46.7
Red meat	55	58	60	64

NOTE: General linear regression model was used to estimate age-adjusted means of variables for each quartile of energy-adjusted dietary vitamin B₁₂.

Abbreviation: HDL, high-density lipoprotein cholesterol.

*All nutrient and food variables, except alcohol and supplements, were energy adjusted using the residual method (23) to account for their significant correlations with energy.

Table 2. Baseline characteristics (medians and proportions) by case-noncase status for NHL and MM, ATBC Study cohort, 1985-2001 (N = 27,111)

Characteristics	Noncases* (n = 26,818)	NHL cases (n = 195)	MM cases (n = 32)
Age	57.0	58.0 [†]	60.0 [‡]
Body mass index (kg/m ²)	26.0	25.8	26.6
Height (cm)	174.0	174.0	174.0
Smoking history			
Cigarettes/d	20	20	20
Years smoked	36.0	40.0 [†]	39.5
Education (%)			
Elementary school or less	78	77	75
Up to junior high school	14	14	19
High school or more	8	9	6
Serum HDL (mmol/L)	1.14	1.08 [†]	1.03
Family history of cancer (%)	32	25 [†]	22
History of rheumatoid arthritis (%)	2.8	4.1	0
Foods/nutrients (per day) [§]			
Folate (μg)	326	331	324
Vitamin B ₁₂ (μg)	10.1	9.4 [†]	10.2
Vitamin B ₆ (mg)	2.4	2.4	2.5
Vitamin B ₂ (mg)	2.8	2.9	3.0
Methionine (g)	2.00	1.98	1.99
Alcohol (g)	11.0	8.3 [†]	10.9
Vitamin D (μg)	4.9	4.4 [†]	4.4
Fish (g)	32.7	28.6 [†]	28.3
ω-3 fatty acids from fish (g)	0.41	0.34 [†]	0.39

NOTE: Nonparametric Wilcoxon rank-sum tests were used for continuous variables, and χ^2 tests were done on categorical variables.

Abbreviation: HDL, high-density lipoprotein cholesterol.

*In total, 293 of 27,111 people with dietary data developed hematopoietic cancers: data on 11 Hodgkin lymphoma and 55 nonlymphoid cancers are not presented due to small numbers.

[†] $P < 0.05$.

[‡] $P < 0.01$.

[§]All nutrient and food variables, except alcohol, were energy adjusted using the residual method (23) to account for their significant correlations with energy.

dietary one-carbon nutrients: statistical significance of trends was determined by linear contrast in the general linear model (Table 1). Wilcoxon rank-sum tests (continuous variables) and χ^2 tests (categorical variables) were used to compare unadjusted baseline characteristics of cases and noncase cohort members (Table 2).

Cox proportional hazard regression models were used to calculate each nutrient-cancer association as a hazard ratio (HR) and 95% confidence interval (95% CI; Tables 3 and 4). Person-years during the follow-up were used as the time metric in the models. Energy-adjusted dietary one-carbon nutrients were analyzed as categorical variables based on the distribution in the entire cohort, quartiles for NHL (Table 3) and tertiles for NHL subtypes (Table 4) and MM; results on MM are not shown due to limited numbers. Each regression model was built from an age- and energy-adjusted model by adding potential confounding factors one at a time; confounders were determined if they changed the risk estimate of the nutrient-cancer association by $\geq 10\%$. As potential confounders, we evaluated all the variables in Tables 1 and 2, as well as the study interventions of α -tocopherol and β -carotene supplements, leisure and occupational physical activity, medical history, and other macro- and micronutrients. Linear trends between categorical nutrients and cancer were determined using the two-sided Wald test of a score variable that contained median nutrient values of the categories. Effect modification was evaluated using cross-product terms ($P < 0.10$ considered significant) and likelihood ratio test statistics comparing models with and without the product term. Statistical analyses were conducted using the SAS software systems (version 8, SAS Institute, Cary, NC). All P values were two sided and were considered statistically significant at $\alpha < 0.05$.

Results

Baseline characteristics, including putative risk factors of NHL, were examined in relation to dietary intake of one-carbon nutrients and by case/noncase status. Because we found an inverse association between dietary intake of vitamin B₁₂, but not of the other one-carbon nutrients, and NHL as described below, we present age-adjusted baseline characteristics of the cohort participants by quartile of energy-adjusted dietary vitamin B₁₂ in Table 1. Men with greater vitamin B₁₂ intake from food tended to be younger, taller; have higher body mass index and serum high-density lipoprotein cholesterol; smoked more cigarettes daily; and have higher education. Dietary vitamin B₁₂ was positively correlated with intake levels of all dietary and supplemental one-carbon nutrients and alcohol, protein, ω-3 fatty acids from fish, vitamin D, dairy, fish, and red meat. Use of vitamin supplements varied by vitamin and ranged from 6% (folate) to 13% (vitamin B₆).

Compared with cohort members without hematopoietic cancers, NHL cases were slightly older and smoked longer; had lower serum levels of high-density lipoprotein cholesterol; had lower prevalence of family history of any cancer; and had lower baseline intake of vitamin B₁₂, alcohol, vitamin D, fish, and ω-3 fatty acids from fish (Table 2). MM cases were older than noncases.

Table 3 shows the age- and energy-adjusted and multivariable-adjusted HRs and 95% CIs for the association between one-carbon nutritional factors and overall NHL. Dietary vitamin B₁₂ intake was inversely associated: subjects consuming the highest amounts had ~40% less risk of those consuming the lowest (age- and energy-adjusted HR, 0.58; 95% CI, 0.38-0.88; $P_{\text{trend}} = 0.01$). The inverse association remained after adjusting for multivariables (Table 3) and after excluding the first 2 years of follow-up ($n = 172$; HR, 0.59; 95% CI, 0.35-1.01; $P_{\text{trend}} = 0.05$). Dietary folate, vitamin B₆ or B₂, and methionine were not associated with the risk of overall NHL, nor was supplemental intake of any one-carbon nutrients. Compared with low-level consumption of alcohol, both nonuse (HR, 0.67; 95% CI, 0.40-1.14) and high-level consumption (HR, 0.76; 95% CI, 0.49-1.20) were associated with nonsignificantly lower NHL risk.

The inverse association between dietary vitamin B₁₂ and overall NHL was apparent in diffuse (Table 4; $P_{\text{trend}} = 0.13$) and small lymphocytic lymphoma ($P_{\text{trend}} = 0.44$) subtypes, but not in follicular or chronic lymphocytic leukemia subtype, although limited in power with small case numbers. Methionine was inversely associated only with T-cell lymphoma, although based only on 17 cases. The associations of other B vitamins remained similar for subtypes as for overall NHL.

The vitamin B₁₂-NHL association was not modified by alcohol intake, multivitamin use, other one-carbon nutrients, age, body mass index, smoking duration, education, or high-density lipoprotein cholesterol levels (data not shown). Potential confounders (Tables 1 and 2) other than the ones in the final models did not change the associations.

No associations of dietary or supplemental one-carbon nutrients were observed with MM (HR for vitamin B₁₂, 0.86; 95% CI, 0.31-2.39).

Discussion

We observed an inverse association between dietary intake of vitamin B₁₂ and the risk of NHL among the Finnish male smokers. Other nutrients involved in one-carbon metabolism, including folate, vitamins B₆ and B₂, methionine, and alcohol, were not associated with NHL or MM. The inverse association between high methionine intake and T-cell subtype in these data is inconclusive because of the small number of events.

Table 3. Adjusted HRs and 95% CIs for NHL (n = 195) according to baseline dietary intake of one-carbon-related nutrients (quartiles) and supplement use, ATBC Study cohort, 1985-2001 (N = 27,111)

	Cases	Person-years	Age- and energy-adjusted HR (95% CI)	Multivariable HR (95% CI)
Folate ($\mu\text{g}/\text{d}$)				
Q1 (128-290)	50	78,313	1.00 (—)	1.00 (—)
Q2 (291-325)	42	81,147	0.83 (0.55-1.25)	0.88 (0.58-1.32)
Q3 (326-362)	58	81,510	1.15 (0.79-1.68)	1.26 (0.86-1.85)
Q4 (363-703)	44	82,260	0.91 (0.61-1.36)	1.03 (0.68-1.55)
P_{trend}			0.98	0.54
Folate supplement				
No	186	304,533	1.00 (—)	1.00 (—)
Yes	9	18,699	0.77 (0.39-1.50)	0.76 (0.39-1.48)
Vitamin B ₁₂ ($\mu\text{g}/\text{d}$)				
Q1 (1.68-8.05)	62	81,300	1.00 (—)	1.00 (—)
Q2 (8.06-10.11)	50	80,182	0.81 (0.56-1.18)	0.81 (0.55-1.20)
Q3 (10.12-12.87)	48	80,607	0.80 (0.55-1.16)	0.81 (0.53-1.24)
Q4 (12.88-62.02)	35	81,143	0.58 (0.38-0.88)	0.61 (0.37-1.00)
P_{trend}			0.01	0.06
Vitamin B ₁₂ supplement				
No	184	300,054	1.00 (—)	1.00 (—)
Yes	11	23,177	0.76 (0.41-1.39)	0.74 (0.40-1.36)
Vitamin B ₆ (mg/d)				
Q1 (1.06-2.16)	61	78,574	1.00 (—)	1.00 (—)
Q2 (2.17-2.43)	51	80,463	0.83 (0.57-1.21)	0.88 (0.61-1.29)
Q3 (2.44-2.72)	38	81,743	0.63 (0.42-0.94)	0.70 (0.46-1.06)
Q4 (2.73-11.47)	45	82,452	0.76 (0.52-1.12)	0.91 (0.60-1.37)
P_{trend}			0.08	0.42
Vitamin B ₆ supplement				
No	173	282,287	1.00 (—)	1.00 (—)
Yes	22	40,944	0.87 (0.56-1.36)	0.85 (0.55-1.34)
Vitamin B ₂ (mg/d)				
Q1 (0.86-2.44)	49	82,513	1.00 (—)	1.00 (—)
Q2 (2.45-2.83)	45	81,439	0.91 (0.61-1.36)	1.05 (0.69-1.59)
Q3 (2.84-3.23)	58	80,167	1.18 (0.81-1.73)	1.50 (1.00-2.25)
Q4 (3.24-7.37)	43	79,112	0.88 (0.59-1.33)	1.25 (0.79-1.98)
P_{trend}			0.86	0.17
Vitamin B ₂ supplement				
No	175	287,618	1.00 (—)	1.00 (—)
Yes	20	35,614	0.92 (0.58-1.46)	0.90 (0.56-1.44)
Methionine (g/d)				
Q1 (0.55-1.78)	63	80,765	1.00 (—)	1.00 (—)
Q2 (1.79-1.99)	42	80,944	0.67 (0.45-0.99)	0.76 (0.51-1.13)
Q3 (2.00-2.20)	38	81,114	0.61 (0.41-0.91)	0.74 (0.48-1.14)
Q4 (2.21-5.10)	52	80,408	0.85 (0.59-1.23)	1.15 (0.75-1.76)
P_{trend}			0.30	0.66
Alcohol (g/d)				
None	19	34,645	0.70 (0.42-1.19)	0.67 (0.40-1.14)
Q1 (0.04-5.2)	55	70,951	1.00 (—)	1.00 (—)
Q2 (5.3-13.3)	43	73,597	0.80 (0.54-1.19)	0.83 (0.56-1.24)
Q3 (13.4-27.6)	46	72,913	0.90 (0.61-1.34)	0.97 (0.65-1.45)
Q4 (27.7-278.5)	32	71,125	0.67 (0.43-1.05)	0.76 (0.49-1.20)

NOTE: All nutrients, except alcohol and supplements, were energy adjusted using the residual method (23) to account for their significant correlations with energy. Multivariable models of one-carbon nutrients adjusted for age, calories, education (three categories: elementary school or less, up to junior high school, high school or more), smoking history (years smoked), and additionally for the following: folate model adjusted for vitamin B₁₂; vitamin B₁₂ model adjusted for methionine, vitamin D, and ω -3 fatty acids from fish; vitamin B₆ model adjusted for vitamin B₁₂; vitamin B₂ model adjusted for vitamin B₁₂; methionine model adjusted for vitamin B₁₂; alcohol model adjusted for serum high-density lipoprotein cholesterol.

To our knowledge, two epidemiologic studies of NHL have addressed specific one-carbon nutrients (13, 14). In a prospective investigation involving 199 incident cases after 14 years of follow-up, neither folate from foods nor total folate was associated with NHL, which is consistent with our observation (13). In a case-control study, we previously observed an ~50% reduction in risk of NHL associated with the highest quartile versus the lowest quartile of dietary vitamin B₆ and methionine (14). Dietary vitamin B₁₂ was inversely associated with NHL but did not reach statistical significance in the previous study. The vitamin B₆-NHL associations in these data were statistically nonsignificant yet consistently inverse with overall NHL and its main subtypes. Although the reported intake levels of one-carbon nutrients are higher in the current study, they may not reflect the status in the body because smoking is known to disturb the absorption and metabolism of one-carbon nutrients (24, 25). This may in part explain the

inconsistencies between the two studies. The higher NHL risk associated with nondrinking versus drinking alcohol in the previous case-control study may be due to recall bias and/or reverse causation.

In light of one-carbon metabolism, a protective effect of vitamin B₁₂ on NHL is plausible through its essential role as a cofactor to the enzyme methionine synthase, which catalyzes the initial supply of one-carbon units from circulatory folate (5-methyltetrahydrofolate) for intracellular DNA methylation and DNA synthesis/repair (26). In an animal study, moderately depleted vitamin B₁₂ in the presence of sufficient folate resulted in genomic hypomethylation and compromised DNA composition in the colonic DNA (27). In epidemiologic studies, vitamin B₁₂ status has been associated with lower risk of other cancers (28-30), and genetic polymorphisms of methionine synthase have been associated with altered risk of lymphoid cancers (5, 7, 8, 31).

In support of this finding, the inverse vitamin B₁₂-NHL association observed was not modified or confounded by one-carbon-related or general risk factors of NHL. We excluded the early follow-up years to determine whether the association might have been biased by recent dietary changes of NHL patients, such as reduced intake of animal products that are high in vitamin B₁₂, and found little difference. Data collection on dietary and lifestyle factors before disease and thorough ascertainment of histologically confirmed incident cases would have minimized recall bias and misclassification, respectively. We assessed past diet history using a validated and comprehensive food frequency questionnaire, which allowed us to carefully adjust for daily intake of energy and other dietary factors that could have confounded the associations of interest.

On the other hand, our findings based on dietary information among men with history of heavy smoking have limitations. Vitamin B₁₂ status may not coincide with vitamin B₁₂ intake. Nutritional status of vitamin B₁₂ among the elderly is commonly diminished regardless of dietary adequacy due to prevalent atrophic gastritis that reduces the intestinal absorption of vitamin B₁₂ (32). In addition, smoking disrupts vitamin B₁₂ status as mentioned above (24, 25). Past studies of serum vitamin B₁₂ in the ATBC Study cohort have shown a low correlation between dietary and circulating vitamin B₁₂ levels ($r = 0.06$; ref. 33) and only a small proportion of the participants with vitamin B₁₂ deficiency (34). Therefore, it is possible that dietary vitamin B₁₂ may be a marker for some other nutrient or lifestyle components that are causally associated with the risk of NHL.

We hypothesized that the association between one-carbon nutrition and lymphoid cancer may be modified by alcohol, as is the case for breast (28) and colorectal cancers (16), but we did

not find differential associations among drinkers versus nondrinkers or for increasing alcohol intake. The association between alcohol and NHL has been inconsistent in the literature, although the pooled analysis of nine case-control studies in the International Lymphoma Epidemiology Consortium (InterLymph) yielded a significant inverse association comparing any versus no alcohol consumption (odds ratio, 0.83; 95% CI, 0.76-0.89; ref. 15). We found nonsignificantly lower risk associated with both "nondrinking" and higher consumption; however, an earlier analysis of lung cancer in the same cohort suggested that nondrinkers may include ex-drinkers (35), in which case our findings may indicate an overall inverse association between alcohol and NHL. Considering the inconsistencies in the literature, no dose-response, potential confounding by other lifestyle factors, and potential misspecification of past alcohol intake (36-38), further prospective studies with detailed alcohol intake history are warranted to estimate the risk associated with true lifetime exposure to alcohol.

Despite the limited power and uncertainty in possible differential etiologic effects of one-carbon metabolism on NHL subtypes, we present subtype findings and their patterns of associations with one-carbon nutrients because previous observations on diet (14, 39) and demographics (22, 40) suggest that subtype-specific studies of NHL might be informative. We had a limited number of cases to examine MM, heterogeneity among NHL subtypes, or interactions among nutrients.

Our investigation of the potential causal link between one-carbon nutrition and lymphoid cancers should be replicated in other large prospective cohorts that have high-quality dietary data, power to examine subtypes on both smokers and nonsmokers, and biological markers of one-carbon nutrition.

Table 4. Multivariable HRs and 95% CIs for NHL subtypes according to baseline dietary intake of one-carbon-related nutrients (tertiles), ATBC Study cohort, 1985-2001 (N = 27,111)

	Diffuse lymphoma (n = 54)		Follicular lymphoma* (n = 16)		SLL (n = 24)		CLL (n = 41)		SLL/CLL (n = 65)		T-cell lymphoma* (n = 17)	
	n	HR (95% CI)	n	HR (95% CI)	n	HR (95% CI)	n	HR (95% CI)	n	HR (95% CI)	n	HR (95% CI)
Folate (µg/d)												
T1 (128-302)	16	1.0 (reference)	7	1.0 (reference)	8	1.0 (reference)	11	1.0 (reference)	19	1.0 (reference)	5	1.0 (reference)
T2 (303-348)	17	1.12 (0.56-2.22)	9	0.69 (0.25-1.90)	9	1.08 (0.42-2.83)	13	1.25 (0.56-2.80)	22	1.18 (0.63-2.20)	5	1.13 (0.33-3.93)
T3 (349-703)	21	1.46 (0.75-2.84)			7	0.84 (0.30-2.38)	17	1.73 (0.80-3.76)	24	1.34 (0.73-2.48)	7	1.74 (0.54-3.93)
<i>P</i> _{trend}		0.26		0.48		0.74		0.16		0.35		0.35
Vitamin B₁₂ (µg/d)												
T1 (1.68-8.72)	23	1.0 (reference)	7	1.0 (reference)	9	1.0 (reference)	13	1.0 (reference)	22	1.0 (reference)	9	1.0 (reference)
T2 (8.73-11.80)	18	0.75 (0.38-1.45)	9	0.79 (0.25-2.52)	6	0.49 (0.16-1.45)	16	1.45 (0.66-3.17)	22	0.98 (0.52-1.85)	8	0.95 (0.32-2.77)
T3 (11.81-62.02)	13	0.53 (0.24-1.21)			9	0.60 (0.19-1.92)	12	1.21 (0.48-3.08)	21	0.95 (0.46-1.95)		
<i>P</i> _{trend}		0.13		0.69		0.44		0.70		0.88		0.92
Vitamin B₆ (mg/d)												
T1 (1.06-2.26)	21	1.0 (reference)	7	1.0 (reference)	9	1.0 (reference)	15	1.0 (reference)	24	1.0 (reference)	9	1.0 (reference)
T2 (2.27-2.61)	19	0.96 (0.51-1.80)	9	0.72 (0.26-2.01)	9	0.94 (0.37-2.40)	12	0.85 (0.40-1.84)	21	0.89 (0.49-1.61)	8	0.57 (0.21-1.55)
T3 (2.62-11.47)	14	0.77 (0.37-1.57)			6	0.60 (0.20-1.81)	14	1.10 (0.51-2.40)	20	0.89 (0.47-1.68)		
<i>P</i> _{trend}		0.47		0.53		0.38		0.82		0.72		0.27
Vitamin B₂ (mg/d)												
T1 (0.86-2.59)	20	1.0 (reference)	7	1.0 (reference)	4	1.0 (reference)	17	1.0 (reference)	21	1.0 (reference)	9	1.0 (reference)
T2 (2.60-3.08)	15	0.86 (0.43-1.73)	9	0.79 (0.27-2.31)	10	2.55 (0.78-8.34)	10	0.59 (0.26-1.30)	20	0.95 (0.51-1.79)	8	0.73 (0.25-2.12)
T3 (3.09-7.37)	19	1.27 (0.63-2.57)			10	2.66 (0.76-9.24)	14	0.85 (0.39-1.88)	24	1.19 (0.63-2.28)		
<i>P</i> _{trend}		0.55		0.66		0.14		0.63		0.60		0.57
Methionine (g/d)												
T1 (0.55-1.85)	17	1.0 (reference)	6	1.0 (reference)	7	1.0 (reference)	16	1.0 (reference)	23	1.0 (reference)	12	1.0 (reference)
T2 (1.86-2.12)	20	1.41 (0.72-2.78)	10	1.07 (0.35-3.29)	9	1.24 (0.45-3.43)	10	0.67 (0.30-1.53)	19	0.85 (0.46-1.60)	5	0.28 (0.09-0.88)
T3 (2.13-5.10)	17	1.42 (0.66-3.05)			8	1.09 (0.35-3.36)	15	1.10 (0.49-2.48)	23	1.09 (0.56-2.10)		
<i>P</i> _{trend}		0.37		0.91		0.89		0.84		0.81		0.03
Alcohol (g/d)												
None	3	0.45 (0.14-1.49)	2	0.71 (0.16-3.26)	2	0.64 (0.14-2.86)	6	1.41 (0.55-3.65)	8	1.08 (0.49-2.38)	1	0.41 (0.05-3.22)
Low (0.04-13.3)	27	1.0 (reference)	10	1.0 (reference)	12	1.0 (reference)	15	1.0 (reference)	27	1.0 (reference)	9	1.0 (reference)
High (13.4-278.5)	24	0.95 (0.54-1.68)	4	0.55 (0.17-1.79)	10	1.04 (0.44-2.47)	20	1.84 (0.93-3.64)	30	1.48 (0.87-2.52)	7	0.93 (0.34-2.55)

NOTE: Multivariable models were adjusted for age and calories and additionally for vitamin B₁₂ (in the models of folate, vitamin B₆, vitamin B₂, and methionine), for methionine, vitamin D, and ω-3 fatty acids from fish (in the vitamin B₁₂ model), and for serum high-density lipoprotein cholesterol (in the alcohol model).

Abbreviations: SLL, small lymphocytic lymphoma; CLL, chronic lymphocytic leukemia.

*Cells with case counts <5 were combined with adjacent categories for follicular and T-cell lymphoma subtypes, except for alcohol.

With the main association of genetic and nutritional factors having been reported, gene-nutrient interaction may be considered in future studies to clarify any differential associations in the at-risk population.

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