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Dietary Choline and Betaine and the Risk of Distal Colorectal Adenoma in Women

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Abstract

Background—Choline and betaine are involved in methyl-group metabolism as methyl-group donors; thus, like folate, another methyl-group donor, they may be associated with a reduced risk of colorectal adenomas. No epidemiologic study has examined the association of intake of these nutrients and colorectal adenoma risk.

Methods—We investigated the relationship between intakes of choline and betaine and risk of colorectal adenoma in US women enrolled in the Nurses' Health Study. Dietary intake was measured by food-frequency questionnaires, and individual intakes of choline and betaine were calculated by multiplying the frequency of consumption of each food item by its choline and betaine content and summing the nutrient contributions of all foods. Logistic regression models were used to calculate adjusted odds ratios (as approximations for relative risks) and 95% confidence intervals (CIs) of colorectal adenoma. All statistical tests were two-sided.

Results—Among 39 246 women who were initially free of cancer or polyps and who had at least one endoscopy from 1984 through 2002, 2408 adenoma cases were documented. Increasing choline intake was associated with an elevated risk of colorectal adenoma; the multivariable relative risks (95% CIs) for increasing quintiles of intake, relative to the lowest quintile, were 1.03 (0.90 to 1.18), 1.01 (0.88 to 1.16), 1.23 (1.07 to 1.41), and 1.45 (1.27 to 1.67; P_{trend}<.001). Betaine intake had a nonlinear inverse association with colorectal adenoma; the multivariable relative risks (95% CIs) for increasing quintiles of intake were 0.94 (0.83 to 1.07), 0.85 (0.75 to 0.97), 0.86 (0.75 to 0.98), and 0.90 (95% CI = 0.78 to 1.04; P_{trend} = .09). Among individual sources of choline, choline from phosphatidylcholine and from sphingomyelin were each positively related to adenoma risk.

Conclusions—Our findings do not support an inverse association between choline intake and risk of colorectal adenoma. The positive association between choline intake and colorectal adenoma that we observed could represent effects of other components in the foods from which choline was derived and should be investigated further.

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Notes

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Choline and its oxidation product betaine are nutrients involved in one-carbon metabolism (1,2). Either folate (5-methyltetrahydrofolate) or betaine can donate a methyl group to homocysteine to create methionine. Methylation of homocysteine by betaine is confined to the liver and the kidney, but the pathway involving folate exists in all body cells (3). In addition to being methyl-group donors, choline and betaine have other biologic functions. Choline is a precursor for the cell membrane phospholipids phosphatidylcholine and sphingomyelin (1,4) and for the neurotransmitter acetylcholine, and it is essential in brain development and normal memory function (5–8). Prolonged reduction in choline intake promotes fatty liver, DNA hypomethylation, and tumor development in the liver (even in the absence of carcinogens) in animals (9–11). Betaine is an osmolyte that regulates cellular hydration and cell volume and, thus, protects the cell from environmental stress (2). Organ meats, eggs, and wheat germ are rich dietary sources of choline (12); wheat germ or bran, spinach, and shellfish are rich dietary sources of betaine.

Epidemiologic studies have found that intakes of dietary factors affecting one-carbon metabolism—lower folate, higher alcohol, and lower methionine—are associated with risk of colorectal adenoma (13,14). To our knowledge, however, there has been no epidemiologic study of the association of dietary choline and betaine with the risk of chronic diseases, including colorectal adenoma. The lack of such a study may be due in part to the fact that a food composition database for choline and betaine has not been available until recently (12). A few epidemiologic studies have examined the association of low choline and betaine intakes with risk of birth defects that had previously been associated with variations in one-carbon metabolism (15,16). We recently found that choline and betaine intake predicted plasma homocysteine levels independent of folate intake, confirming a biologic role of choline and betaine as methyl-group donors (17). In light of these findings, we examined the relationships between intakes of choline and betaine and the risk of colorectal adenoma among women in the Nurses' Health Study (18).

Methods

Study Population

The Nurses' Health Study enrolled 121 700 female registered nurses aged 30−55 years in 1976, who responded to a baseline questionnaire on lifestyle and major illnesses (18). Every 2 years, a follow-up questionnaire was sent to these women to update information regarding diet and lifestyle and to ascertain new diagnoses of major illnesses. Deaths in the cohort were ascertained by reports from family members or the postal service in response to questionnaire mailings and a search of the National Death Index. We estimate that more than 97% of deaths were ascertained through the National Death Index (19). The response rate to biennial questionnaire for this cohort has been close to 90%. In this analysis, we included only participants who provided dietary intake information in 1984 and who had no diagnosis of cancer (except nonmelanoma skin cancer), ulcerative colitis, or colorectal polyp before 1984 (n = 78 423). To reduce the potential for detection bias, we further restricted the analysis to women who reported having undergone a colonoscopy or sigmoidoscopy between 1984 and 2002 (n = 39 246). More than 90% of the adenomas recorded in this study were diagnosed during endoscopic procedures performed for screening for colorectal ademoma or because of gastrointestinal conditions that were unrelated to colorectal adenoma. The procedures and protocols of the study were approved by the Institutional Review Boards of the Brigham and Women's Hospital and the Harvard School of Public Health.

Dietary Assessment

As part of the biennial questionnaire, a semiquantitative food-frequency questionnaire (FFQ) with approximately 60 food items was sent to members of the cohort in 1980. Subsequently,

an expanded FFQ with approximately 130 food items was administered to the women in 1984, 1986, 1990, 1994, and 1998. Participants were asked how often, on average, they had consumed each food item during the past year. Serving sizes were specified for each food item in the FFQ. The questionnaire had nine possible responses for intake of each item, ranging from never or less than once per month to six or more times per day.

The choline and betaine composition of individual foods was added to the FFQ's nutrient database (Harvard University Food Composition Database) using values published by Zeisel et al. (12,20) and from the USDA's choline database (21). The average daily intake of choline and betaine was calculated by multiplying the frequency of consumption of each food item by its choline and betaine content and summing the contributions of choline and betaine from all foods. Total choline intake was calculated as the sum of choline intake from free choline, phosphocholine, glycerophosphocholine, phosphatidylcholine (lecithin), and sphingomyelin. For choline and betaine, as well as other nutrients, we used intake from food and supplements and the regression-residual method to adjust intakes for total energy intake (22). After examining values of choline and betaine calculated from successive FFQs, we decided to start follow-up from 1984 because similar and more comprehensive FFQs were used from then onward, making intake data more comparable across time.

CONTEXT AND CAVEATS

Prior knowledge

Epidemiologic studies have suggested that lower dietary intake of folate and methionine and higher intake of alcohol are associated with an increased risk of colorectal adenoma (polyps in the colon or rectum that may develop into colorectal cancer). All of these dietary factors are involved in a biochemical pathway(s) referred to as one-carbon metabolism. Choline and betaine in the diet also affect one-carbon metabolism, but their association with the risk of colorectal cancer was not known.

Study design

Dietary intake of choline and betaine and incidence of colorectal adenoma were assessed by a questionnaire that was sent to a large group of female nurses every 2 years, and statistical methods were used to assess the association between choline intake and the risk of colorectal adenoma.

Contribution

Increased dietary intake of choline was associated with an elevated risk of colorectal adenoma. The association with betaine intake was not clear.

Implications

Additional work will be needed to clarify the relationship between choline and risk of colorectal adenoma.

Limitations

Other components of the diet, the intakes of which are highly correlated with choline consumption, may be the source of the increased risk of colorectal adenoma that was observed.

In a previous study, we found that intakes of choline and betaine as measured by our FFQ predicted plasma total homocysteine levels in a cohort study of men and women in the Framingham Offspring Study (17), confirming the validity of the instrument and the biologic relevance of choline and betaine intake measured by the FFQ. For the lowest and highest quintiles of dietary choline plus betaine, the multivariable geometric means for homocysteine

were 10.9 and 9.9 μmol/L (P_{trend}<.001), respectively. The inverse association of choline and betaine intake with homocysteine levels was manifested primarily in participants with low folate intake ($P_{interaction}$<.001); among those with folate intake of ≤250 μg/day, the geometric mean homocysteine concentrations in the lowest and highest quintiles of choline plus betaine intake were 12.4 and 10.2 μmol/L (P_{trend}<.0001), respectively.

Assessment of Adenoma Cases

On each biennial questionnaire, we asked whether participants had undergone sigmoidoscopy or colonoscopy; what the indications for these procedures were; whether colon or rectal polyps had been diagnosed; and, if they had, the date of diagnosis. When a participant reported a diagnosis, we obtained her informed consent to acquire medical records and pathology reports. Study investigators who were blinded to exposure data reviewed all records and extracted data on the histologic type, anatomic location, and size of polyps. We did not ask participants to specify whether sigmoidoscopy or colonoscopy was performed. On the basis of secular trends, there was probably a gradual increase in the relative proportion of colonoscopies compared with sigmoidoscopies. Because we assumed that a substantial portion of all procedures were sigmoidoscopies, which encompass only examination of the distal colon and rectum, we analyzed only adenomas of the distal colon (descending and sigmoid colon) and rectum to prevent misclassification and potential detection bias. Women with adenomas proximal to the descending colon but without synchronous distal adenoma were included as non-case participants (n = 867). Subjects were defined as those with one or more pathology-verified adenomas less than 60 cm from the anus. If more than one adenoma was diagnosed, the subject was classified according to the adenoma of the largest size and most advanced histologic characteristics.

Statistical Analyses

To evaluate the correlations among choline compounds and betaine, we conducted a correlation analysis and calculated Spearman correlation coefficients among these nutrients. To represent participants' long-term dietary intake, women were grouped according to quintile of the cumulative averages of their choline and betaine intake from all available FFQs up to the time of their endoscopy. For example, for women who underwent endoscopy between 1984 and 1986, dietary data from the 1984 FFQ were used. For women who underwent endoscopy in 1992, the average of dietary data from 1984, 1986, and 1990 FFQs was used. For the primary analyses, we used relative risks as a measure of association; relative risks were defined as the incidence of adenoma among participants in a specific quintile of intake divided by the corresponding incidence among participants in the reference quintile.

Logistic regression models were used to calculate odds ratios (as approximations for relative risks) and 95% confidence intervals (CIs) of colorectal adenoma. Models were simultaneously adjusted for multiple known or suspected adenoma risk factors (age [continuous], smoking [never, ≤10 pack-years, ≤20 pack-years, and >20 pack-years], body mass index [<22, 22−24.9, 25.0−28.9, ≥29.0 kg/m^2], physical activity [quartiles], family history of colon cancer [yes, no], history of endoscopic screening [yes, no], year of endoscopy [continuous], aspirin use [yes, no], menopausal status and postmenopausal hormone use [premenopausal, postmenopausal and never user, postmenopausal and past user, postmenopausal and current user], and intakes of energy [continuous], alcohol [continuous], folate [continuous], fiber [continuous], and calcium [continuous]). We used women who underwent endoscopy and did not have a diagnosis of adenoma as control subjects. For all relative risks, 95% confidence intervals were calculated. Tests for trend were conducted using the median value for each quintile as a continuous variable. To examine whether the association between choline intake and colorectal adenoma risk was modified by intakes of alcohol, folate, and red meat, we included (one at a time) a cross-product term of choline intake and each of these dietary factors, both expressed

as continuous variables, in a multivariable model. The *P* value for the tests for interaction was obtained from a likelihood ratio test with one degree of freedom. Because some of the non-case participants included subjects with proximal colon adenoma, we also examined the risk for adenomas by excluding these subjects. We used the SAS statistical package, version 9.1 (SAS Institute, Cary, NC) for all analyses. All *P* values were two-sided.

Results

Among 39 246 US women in the Nurse's Health Study who were initially free of cancer or polyps and who underwent endoscopy from 1984 through 2002, 2408 were documented as having adenoma (1841 distal colon adenomas and 675 rectal adenomas).

We calculated the distribution of potential risk factors for adenomas by quintiles of choline and betaine intake in 1984 (Table 1). Women with higher choline intake were more likely to be past smokers, to exercise regularly, and to use aspirin on a regular basis, and they consumed more fiber, calcium, folate, methionine, vitamin B6, and vitamin B12. Similar trends were observed for those with higher betaine intake, except that aspirin use was not associated with betaine intake.

Table 2 presents energy-adjusted mean intakes and correlations with energy-adjusted choline and betaine in 1984. The energy-adjusted mean choline intake was 331 mg/day (standard deviation [SD] = 80). More than half of choline intake came from phosphatidylcholine. The energy-adjusted mean betaine intake was 189 mg/day (SD = 97). The correlation between choline and betaine intake was low, and food sources for these nutrients were different. Major food sources for choline were red meat (18% of intake), eggs (13%), poultry (9%), and milk (9%). Major food sources for betaine were spinach (34% of intake), white bread (8%), cold breakfast cereal (8%), pasta (7%), and dark bread (7%). Choline, betaine, and B vitamins that are related to one-carbon metabolism (folate and vitamins B6 and B12) share some food sources. However, the correlations between intakes of betaine and these vitamins were low (0.30 for folate, 0.18 for vitamin B6, and 0.15 for vitamin B12). The correlations between choline and these vitamins were modest (0.32 for folate, 0.34 for vitamin B6, and 0.47 for vitamin B12).

Higher intake of dietary choline was not, contrary to our hypothesis, inversely related to risk of distal colorectal adenoma (Table 3). On the contrary, choline intake was associated with an elevated risk of colorectal adenoma; the multivariable relative risks (95% CI) for increasing quintiles of intake were 1.00 (referent), 1.03 (0.90 to 1.18), 1.01 (0.88 to 1.16), 1.23 (1.07 to 1.41), and 1.45 (1.27 to 1.67; P_{trend}<.001). Betaine intake was inversely associated with colorectal adenoma risk in age-adjusted analyses, but the association was much attenuated in multivariable analyses, mainly due to adjustment for folate intake. Betaine intake had a nonlinear inverse association with colorectal adenomas; the multivariable relative risks for increasing quintiles of intake (95% CIs) were 1.00 (referent), 0.94 (0.83 to 1.07), 0.85 (0.75 to 0.97), 0.86 (0.75 to 0.98), and 0.90 (95% CI = 0.78 to 1.04; P_{trend} = .09). Additional adjustment for intakes of other nutrients related to methyl-group metabolism, including methionine and vitamins B6 and B12, did not substantially affect the associations (data not shown). Among individual sources of choline, choline from phosphatidylcholine and sphingomyelin was positively related to risk of colorectal adenoma (Table 3). The results were essentially the same when women with adenomas proximal to the descending colon but without synchronous distal adenoma were excluded from the non-case participants (data not shown). The positive association between choline intake and colorectal adenoma risk was consistent for larger and smaller adenomas (<1 versus ≥1 cm) and for distal versus rectal adenomas (data not shown).

Because choline is involved in methyl-group metabolism, we investigated whether the availability of other dietary factors related to methyl-group metabolism would modify the association between choline and adenoma risk. We examined choline intake and adenoma risk by levels of folate intake (<250, 250 to <400, and ≥400 μg/day) and alcohol intake (none, >0 to <10 g/day, and ≥10 g/day) (Table 4). The association between choline intake and adenoma risk was strongest among those with low folate intake and higher alcohol intake. However, none of the *P* values for interaction were statistically significant. We also examined beta-ine intake and adenoma risk by levels of folate and alcohol intakes. Betaine intake was not associated with adenoma risk at any levels of folate/alcohol intake (data not shown).

To address the possibility that the positive association between choline and colorectal adenoma is due to some particular foods that contribute to choline intake (e.g., red meat), we examined the association of major food sources of dietary choline with colorectal adenoma risk. Intakes of eggs and red meat were each positively related to colorectal adenoma risk. The relative risks for top versus bottom quintiles of intake were 1.25 (95% CI = 1.09 to 1.42) for eggs and 1.36 (95% CI = 1.15 to 1.60) for red meat. Next, we adjusted for intakes of these foods in a multivariable model with choline. The positive associations between choline intake and colorectal adenomas remained similar and statistically significant even after adjusting for the food sources (data not shown). Finally, we examined choline intake and adenoma risk by levels of red meat intake (<0.5, 0.5 to <1, and ≥1 servings/day) (Table 4). In this analysis, choline intake was positively associated with adenoma risk regardless of the level of red meat intake.

Discussion

In our study, higher choline intake was associated with an elevated risk of colorectal adenoma. The positive association between choline intake and colorectal adenoma persisted after adjustment for multiple dietary factors and when assessed for different sizes and sites of the adenoma. Betaine intake was weakly and inversely associated with colorectal adenoma risk.

Until recently, dietary choline and betaine have not been extensively investigated in epidemiologic studies due to a lack of food composition databases. Few epidemiologic studies have examined the association between choline and betaine and any endpoints; a case–control study found that higher maternal periconceptional choline intake was associated with somewhat reduced risk of neural tube defects, a disease that is related to one-carbon metabolism (15). The multivariable odds ratio for the highest versus lowest quartile of choline intake was 0.51 (95% CI = 0.25 to 1.07), independent of folate intake. Higher maternal choline intake has also been reported to be inversely associated with oro-facial clefts (16).

Because homocysteine can accept a methyl group from either betaine or folate, methylation of homocysteine through choline and betaine and through folate are interrelated. Therefore, disruption of one pathway may affect the other pathway. Depletion of choline intake in humans raised plasma homocysteine levels after oral methionine load (23), and high-dose supplementation of betaine has been used to lower homocysteine levels among participants with hyperhomocysteinemia (3). Supplementation with betaine also lowers fasting homocysteine levels in the general population (24). High-dose supplementation of choline in the form of phosphatidylcholine (2.6 g/day of choline) lowers fasting levels as well as post–methionine-loaded levels of plasma homocysteine in healthy men (25). However, given that choline and betaine are involved in methylation only in certain organs, these may have less impact on methylation status in the colon than folate.

Although choline is synthesized in the body de novo, humans require additional choline from dietary sources. The recommended daily intake was set by the Institute of Medicine, National Academy of Sciences, in 1998 at 550 mg/day for men and 425 mg/day for women (26). Our

data showed that mean intake of females in the Nurses' Health Study was lower than the recommended daily intake.

Intake of choline from phosphatidylcholine and from sphingomyelin accounted for the positive association of choline intake with colorectal adenomas, and phosphatidylcholine was the largest component of total choline intake. Although phosphatidylcholine and sphingomyelin are lipid soluble, other sources of choline are water soluble (12) and absorbed through different pathways, which may result in different bioavailabilities and fates in the body. Although in our previous examination of the relationship between choline intake and plasma homocysteine, choline from several sources predicted plasma homocysteine levels, choline from phosphatidylcholine was not associated with homocysteine levels (17).

Although our results were contrary to expectation based on choline's role as a methyl-group donor, there is a potential biologic basis for the positive association that we observed between choline intake and colorectal adenoma. Once a tumor is initiated, growth into a detectable adenoma depends in part on choline availability because choline is needed to make membranes in all rapidly growing cells. Supporting evidence for a critical role of choline in carcinogenesis comes from a number of divergent studies. For example, rats fed a choline-deficient diet for 3 or 6 months followed by a choline-supplemented diet had higher incidence of hepatocellular carcinoma and more amplification of the c-myc oncogene than animals fed continuously a diet deficient in choline (27,28). Furthermore, several cancers and cancer cell lines have altered membrane phospholipid metabolism with enhanced choline uptake and increased choline metabolite concentrations (29,30). Choline kinase, an enzyme that converts choline to phosphocholine, an intermediate in the generation of membrane phospholipids, is elevated in human cancers, including colon cancer (31,32). Consistent with this increase in enzymatic activity, concentrations of phosphocholine and total choline-containing phospholipid metabolites increased with progression of tumors in human mammary epithelial cells (33), and malignant colon and prostatic cells also exhibited higher levels of phosphocholine and glycerophosphocholine than normal cells (32,34). Finally, loss of p53 function in colon cancer cells resulted in increased phosphocholine and total choline (29). It is of interest to note that a similar tumor-promoting role of folate, another methyl-group donor that has been associated with a reduced risk of colorectal adenoma (13,14), has been raised recently (35,36).

Our study had several limitations. First, our observations may reflect the association of other dietary constituents (including the major source of choline, animal products) with adenoma risk. However, adjusting for several related nutrients and foods and stratification by red meat intake did not materially affect the association. Second, we only included participants who reported having undergone a colonoscopy or sigmoidoscopy. Although including participants who had not undergone such a procedure would have introduced a potential for bias, limiting the population to those who were screened may also have biased the results. For example, adenoma subjects with a high choline intake may be more likely to have an endoscopy than those with a low choline intake. However, because adenomas are largely asymptomatic, the effect of such bias is likely to be small.

In conclusion, we found that choline intake was associated with an elevated risk of colorectal adenoma in women. In contrast, betaine intake had a nonlinear inverse association with colorectal adenoma risk. It will be important to determine if epidemiologic studies from other populations and in men confirm our findings. Also, studies of choline and betaine in relation to colorectal cancer should be undertaken.

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Table 1

Characteristics of participants in the Nurses' Health Study according to energy-adjusted choline and betaine intake in 1984*

	Intake category (quintiles)					
	Choline			Betaine		
Variable	1	3	5	1	3	5
Number of participants	15328	15336	15323	15306	15339	15327
Current smokers, %	26	23	24	27	24	23
Past smokers, %	28	32	34	28	31	36
Regular physical activity, %	40	46	51	40	45	54
Regular aspirin use, %	36	38	39	37	37	37
Family history of colon cancer, %	21	21	20	21	21	21
Postmenopausal hormone use among postmenopausal women, %	20	22	22	21	21	22
Age (y), mean	49	50	52	50	50	51
Body mass index (kg/m^2), mean	25	25	26	25	25	25
Alcohol intake (g/day), mean	7	7	6	6	7	7
Red meat intake (servings/day), mean	1.0	1.2	1.1	1.2	1.2	0.9
Fiber intake (g/day), mean	15	16	18	14	16	20
Calcium intake (mg/day), mean	733	859	1086	829	863	975
Folate intake (μg/day), mean	317	374	486	335	366	473
Methionine intake (g/day), mean	1.3	1.7	2.0	1.6	1.7	1.8
Vitamin B6 intake (mg/day), mean	6	8	17	9	9	13
Vitamin B12 intake (μg/day), mean	7	11	18	11	11	14

*Except for the data on mean age, all data shown are standardized to the age distributions of the cohorts in 1984. For simplicity, only data for the first, third, and fifth quintiles of intakes of choline and betaine are shown.

Table 2

Mean intake (mg/day) and Spearman correlation coefficients of energy-adjusted choline compounds and betaine in women in the Nurses' Health Study in 1984

		Correlation coefficients						
	Mean intake ± standard deviation (mg/day)	**Total choline**	**Free choline**	**Choline from glycerophosphocholine**	**Choline from phosphocholine**	**Choline from phosphatidylcholine**	**Choline from sphingomyelin**	**Betaine**
Total choline	331 ± 80	1.00	0.52	0.48	0.58	0.84	0.71	0.20
Free choline	71 ± 16		1.00	0.51	0.54	0.17	0.16	0.28
Choline from glycerophosphocholine	49 ± 20			1.00	0.71	0.06	0.24	0.15
Choline from phosphocholine	14 ± 5				1.00	0.23	0.42	0.24
Choline from phosphatidylcholine	179 ± 64					1.00	0.71	0.12
Choline from sphingomyelin	18 ± 6						1.00	0.05
Betaine	189 ± 97							1.00

Table 3

Relative risks (with 95% confidence intervals) of colorectal adenoma according to quintile of energy-adjusted choline and betaine intake in women in the Nurses' Health Study*

Nutrient	Quintile of intake 1	2	3	4	5	P_{trend}†
Total choline						
Median intake (mg/day) and no. of cases	261	293	315	340	383	
No. of cases/controls	453/7395	448/7399	430/7422	504/7346	573/7276	
Age-adjusted RR (95% CI)	1.00	1.00 (0.87 to 1.14)	0.96 (0.83 to 1.09)	1.13 (0.99 to 1.29)	1.30 (1.14 to 1.48)	<.001
Multivariable‡ RR (95% CI)	1.00	1.03 (0.90 to 1.18)	1.01 (0.88 to 1.16)	1.23 (1.07 to 1.41)	1.45 (1.27 to 1.67)	<.001
Betaine						
Median intake (mg/day)	122	158	186	218	276	
No. of cases/controls	562/7287	502/7344	451/7402	442/7407	451/7398	
Age-adjusted RR (95% CI)	1.00	0.89 (0.79 to 1.01)	0.80 (0.70 to 0.91)	0.78 (0.69 to 0.89)	0.80 (0.71 to 0.91)	<.001
Multivariable‡ RR (95% CI)	1.00	0.94 (0.83 to 1.07)	0.85 (0.75 to 0.97)	0.86 (0.75 to 0.98)	0.90 (0.78 to 1.04)	.09
Free choline						
Median intake (mg/day)	58	66	72	78	89	
No. of cases/controls	470/7384	488/7355	468/7379	484/7372	498/7348	
Age-adjusted RR (95% CI)	1.00	1.06 (0.93 to 1.21)	1.02 (0.89 to 1.16)	1.06 (0.93 to 1.21)	1.10 (0.96 to 1.25)	.21
Multivariable‡ RR (95% CI)	1.00	1.07 (0.94 to 1.23)	1.06 (0.92 to 1.22)	1.12 (0.97 to 1.29)	1.16 (1.00 to 1.36)	.06
Choline from glycerophosphocholine						
Median intake (mg/day)	34	43	51	60	76	
No. of cases/controls	539/7310	495/7355	478/7371	433/7416	463/7386	
Age-adjusted RR (95% CI)	1.00	0.92 (0.81 to 1.05)	0.90 (0.79 to 1.02)	0.81 (0.71 to 0.92)	0.87 (0.77 to 0.99)	.01
Multivariable‡ RR (95% CI)	1.00	0.97 (0.85 to 1.10)	0.96 (0.84 to 1.10)	0.89 (0.77 to 1.02)	0.96 (0.83 to 1.11)	.38
Choline from phosphocholine						
Median intake (mg/day)	10	13	14	16	20	
No. of cases/controls	522/7327	489/7361	473/7378	471/7374	453/7398	
Age-adjusted RR (95% CI)	1.00	0.94 (0.83 to 1.07)	0.91 (0.80 to 1.04)	0.92 (0.80 to 1.04)	0.88 (0.77 to 1.01)	.06
Multivariable‡ RR (95% CI)	1.00	1.02 (0.90 to 1.17)	1.04 (0.91 to 1.19)	1.07 (0.93 to 1.24)	1.08 (0.92 to 1.27)	.30
Choline from phosphatidylcholine						
Median intake (mg/day)	121	141	156	172	205	
No. of cases/controls	428/7423	422/7425	422/7427	525/7320	611/7243	
Age-adjusted RR (95% CI)	1.00	0.98 (0.86 to 1.13)	0.98 (0.85 to 1.13)	1.24 (1.08 to 1.41)	1.45 (1.27 to 1.65)	<.001
Multivariable‡ RR (95% CI)	1.00	1.00 (0.87 to 1.15)	0.99 (0.86 to 1.14)	1.26 (1.10 to 1.44)	1.48 (1.30 to 1.69)	<.001
Choline from sphingomyelin						
Median intake (mg/day)	13	16	17	19	23	
No. of cases/controls	430/7418	446/7405	443/7407	514/7333	575/7275	
Age-adjusted RR (95% CI)	1.00	1.04 (0.90 to 1.19)	1.02 (0.89 to 1.17)	1.20 (1.05 to 1.37)	1.34 (1.18 to 1.53)	<.001
Multivariable‡ RR (95% CI)	1.00	1.05 (0.92 to 1.21)	1.05 (0.92 to 1.21)	1.23 (1.08 to 1.41)	1.37 (1.20 to 1.57)	<.001

*Women who underwent endoscopy and did not have a diagnosis of adenoma were used as control subjects. Women with adenomas proximal to the descending colon but without synchronous distal adenoma were included as non-case participants (n = 867). Odds ratio was used to approximate relative risk. RR = relative risk; CI = confidence interval.

†P_{trend} calculated with median intake of nutrient in each quintile as a continuous variable.

‡Multivariable model was simultaneously adjusted for age (continuous), pack-years of smoking (never, ≤10 pack-years, ≤20 pack-years, >20 pack-years), body mass index (<22, 22−24.9, 25.0−28.9, ≥29.0 kg/m^2), physical activity (quartiles), family history of colon cancer (yes, no), history of endoscopic screening (yes, no), year of endoscopy (continuous), aspirin use (yes, no), menopausal status and postmenopausal hormone use (premenopausal, postmenopausal and never user, postmenopausal and past user, postmenopausal and current user), and intakes of energy (continuous), alcohol (continuous), folate (continuous), total fiber (continuous), and calcium (continuous).

Table 4

Multivariable relative risk and 95% confidence intervals of colorectal adenoma according to quintile of energy-adjusted choline intake by intakes of folate and alcohol in women in the Nurses' Health Study*

Dietary factor	Quintile of intake 1	2	3	4	5	P_{trend}†	$P_{interaction}$
Folate							
<250 μg/day							
No. of cases/controls	104/1285	66/733	53/556	48/387	40/257		
RR (95% CI)	1.00	1.10 (0.79 to 1.53)	1.15 (0.81 to 1.65)	1.46 (1.00 to 2.13)	2.03 (1.35 to 3.07)	.0006	
250 to <400 μg/day							
No. of cases/controls	198/2962	194/2854	183/2735	204/2527	207/1972		
RR (95% CI)	1.00	0.99 (0.81 to 1.22)	0.97 (0.79 to 1.20)	1.15 (0.93 to 1.42)	1.43 (1.15 to 1.77)	.0004	
≥400 μg/day							
No. of cases/controls	151/3148	188/3812	194/4131	252/4432	326/5047		
RR (95% CI)	1.00	1.04 (0.84 to 1.30)	1.00 (0.80 to 1.25)	1.24 (1.00 to 1.53)	1.41 (1.14 to 1.73)	<.0001	>.99
Alcohol							
Nondrinker (n = 520)							
No. of cases/controls	130/1929	92/1447	73/1437	97/1370	128/1722		
RR (95% CI)	1.00	0.95 (0.72 to 1.26)	0.78 (0.58 to 1.05)	1.15 (0.87 to 1.53)	1.28 (0.97 to 1.68)	.04	
>0 to <10 g/day							
No. of cases/controls	229/4001	243/4343	264/4432	281/4470	327/4344		
RR (95% CI)	1.00	1.02 (0.85 to 1.23)	1.12 (0.93 to 1.35)	1.19 (0.99 to 1.44)	1.46 (1.21 to 1.77)	<.0001	
≥10 g/day							
No. of cases/controls	94/1465	113/1609	93/1553	126/1506	118/1210		
RR (95% CI)	1.00	1.20 (0.90 to 1.60)	1.03 (0.76 to 1.39)	1.48 (1.11 to 1.97)	1.71 (1.27 to 2.31)	<.0001	.48
Folate and alcohol							
Nondrinker and folate ≥250 μg/day							
No. of cases/controls	94/1537	73/1266	60/1296	89/1291	121/1652		
RR (95% CI)	1.00	0.92 (0.67 to 1.27)	0.75 (0.53 to 1.05)	1.16 (0.85 to 1.58)	1.29 (0.95 to 1.74)	.03	
Other‡							
No. of cases/controls	291/4965	328/5581	330/5711	375/5747	419/5437		
RR (95% CI)	1.00	1.05 (0.89 to 1.23)	1.05 (0.89 to 1.24)	1.21 (1.02 to 1.42)	1.44 (1.22 to 1.70)	<.0001	
Drinker and folate <250 μg/day							
No. of cases/controls	68/893	47/552	40/415	40/308	33/187		
RR (95% CI)	1.00	1.16 (0.78 to 1.73)	1.27 (0.83 to 1.94)	1.70 (1.10 to 2.63)	2.59 (1.61 to 4.17)	<.0001	.17
Red meat							
<0.5 servings/day							
No. of cases/controls	98/1816	87/1483	65/1449	77/1516	142/1969		
RR (95% CI)	1.00	1.20 (0.88 to 1.62)	0.97 (0.70 to 1.35)	1.09 (0.79 to 1.49)	1.65 (1.23 to 2.21)	.001	
0.5 to <1 servings/day							
No. of cases/controls	216/3457	202/3550	190/3394	216/3332	228/3020		
RR (95% CI)	1.00	0.93 (0.76 to 1.15)	0.93 (0.76 to 1.15)	1.11 (0.90 to 1.36)	1.32 (1.07 to 1.63)	.002	
≥1 servings/day							
No. of cases/controls	139/2122	159/2366	175/2579	211/2498	203/2287		
RR (95% CI)	1.00	1.06 (0.84 to 1.35)	1.06 (0.84 to 1.35)	1.37 (1.09 to 1.73)	1.41 (1.11 to 1.79)	.0006	.28

*Women who underwent endoscopy and did not have a diagnosis of adenoma were used as control subjects. Women with adenomas proximal to the descending colon but without synchronous distal adenoma were included as non-case participants (n = 867). Odds ratio was used to approximate relative risk. Multivariable model was adjusted for the same variables as in Table 3. RR = relative risk; CI = confidence interval.

†P_{trend} calculated with median intake of choline in each quintile as a continuous variable.

[≠] Nondrinker and folate <250 μg/day or drinker and folate ≥250 μg/day.