Selected micronutrient intake and the risk of colorectal cancer

M. Ferraroni[1], C. La Vecchia[1,2], B. D'Avanzo[2], E. Negri[2], S. Franceschi[3] & A. Decarli[1,4]

[1]Istituto di Statistica Medica e Biometria, Università di Milano, Via Venezian 1, 20133 Milan, Italy; [2]Istituto di Ricerche Farmacologiche 'Mario Negri', Via Eritrea 62, 20157 Milan, Italy; [3]Centro di Riferimento Oncologico, via Pedemontana occ., 33081 Aviano, Italy; [4]Istituto Nazionale Tumori, Via Venezian 1, 20133 Milan, Italy.

Summary The relationship between estimated intake of selected micronutrients and the risk of colorectal cancer was analysed using data from a case–control study conducted in northern Italy. The study was based on 828 patients with colon cancer, 498 with rectal cancer and 2,024 controls in hospital for acute, non-neoplastic, non-digestive tract diseases. Relative risks (RRs) of intake quintiles were computed after allowance for age, sex and other major potential confounding factors, including an estimate of total energy intake. No apparent trend in risk across intake quintiles was evident for retinol, vitamin D, methionine and calcium. For β-carotene, ascorbic acid, vitamin E and folate there was a trend of a protective effect with increasing consumption: the RR for the highest versus the lowest quintile was 0.32 for β-carotene, 0.40 for ascorbic acid, 0.60 for vitamin E and 0.52 for folate. These inverse associations were similar for colon and rectal cancer, and consistent across strata of sex and age. When simultaneous allowance was made for all these micronutrients, besides other covariates, the only persistent protective effects were for β-carotene (RR = 0.38 for the highest quintile) and ascorbic acid (RR = 0.52). Whether this reflects a specific, or stronger, effect of these micronutrients, rather than problems of collinearity between micronutrients or other limitations of the data, remains open to discussion. Still, this study suggests that specific micronutrients may exert an independent protective effect against colorectal carcinogenesis.

There are indications that several micronutrients may influence the process of colorectal carcinogenesis. These include a potential protective effect of folate (Benito et al., 1991; Freudenheim et al., 1991), a co-factor in the methylation of thymidylate for DNA synthesis and the production of S-adenosylmethionine, the primary methyl donor in the body (Cooper, 1983); of calcium, which may react with fatty acids to form insoluble soaps (Newmark et al., 1984; Garland et al., 1985; Sorenson et al., 1988); and of ascorbic acid, β-carotene and vitamin E, which may act as antioxidants (Iscovich et al., 1992; Longnecker et al., 1992). Two companion cohort studies (Giovannucci et al., 1993), including 564 women and 331 men with colorectal adenoma, have also suggested that folate may have a specific favourable effect on preneoplastic large bowel lesions. No convincing association for any micronutrients, however, has emerged from other studies (Peters et al., 1992), and the issue is therefore still unsettled, particularly since most studies did not make adequate allowance for various micronutrients.

To provide further data on the issue, we have considered the role of selected micronutrients on colorectal carcinogenesis using data from a case–control study conducted in the greater Milan area, previously considered with reference to intake of specific foods (La Vecchia et al., 1988). The analysis of the food items showed a protective effect of green vegetable consumption, and of a few selected types of fruits and vegetables. The question arises, therefore, whether specific micronutrients, such as β-carotene, retinol, ascorbic acid, vitamin D, vitamin E, folate, methionine and calcium, have an effect on colorectal cancer risk.

Subjects and methods

The data were derived from a case–control study of several digestive tract cancers, based on a network including the major teaching and general hospitals in the greater Milan area. The recruitment of cases of colorectal cancer started in January 1985, and this work is based on data collected up to

December 1992. The general design of this investigation has already been described (La Vecchia et al., 1988; Negri et al., 1990).

Briefly, the cases were 828 incident (i.e. diagnosed within the year prior to the interview) histologically confirmed colon cancers (423 males and 405 females) and 498 rectal cancers (288 males and 210 females). The age range was 20–74 years, and the median age was 62 years for both colon and rectal cancer.

The control group included patients admitted for a wide spectrum of acute, non-neoplastic, non-digestive tract conditions to the same network of hospitals where cases had been identified. Of these, 47% were admitted for traumatic conditions, 20% had non-traumatic orthopaedic diseases, 19% had acute surgical conditions, and 14% had other miscellaneous disorders. A total of 2,024 controls were included in the present analysis. The age range was 19–74 years, and the median age was 55 years. The catchment areas of cases and controls were comparable: over 80% of cases and controls resided in Lombardy, and over 90% came from northern Italy. Less than 3% of eligible subjects (cases and controls) refused to be interviewed.

Trained interviewers used a structured questionnaire to obtain information on general sociodemographic factors and lifestyle habits, weight and height, a problem-oriented medical history and family history of colorectal cancer. Further, information on the frequency of consumption per week of 29 indicator foods was collected. These included major sources of β-carotene, retinol, ascorbic acid, vitamins D and E, folate, methionine and calcium in the Italian diet. We computed nutrient intake by multiplying the consumption frequency of each unit of food by the nutrient content of the standard average portions, using composition values from the Italian composition tables (Fidanza & Verdiglioni, 1988), with the integration of other sources when these were not available (Paul & Southgate, 1980; Souci et al., 1989). The questionnaire was restricted to the frequency of consumption of a limited number of food items, with no quantitative indication of portion size. Thus, the measures obtained should be considered only approximations, and hence potential underestimates, of the real values. Subjects were categorised by quintiles of intake of each nutrient based on the distribution of controls.

Odds ratios [as estimators of relative risk (RR)], together with their 95% approximate confidence intervals (CIs), were derived from data stratified for sex and age in decades by the

Table I Distribution of 828 cases of colon cancer, 498 of rectal cancer and 2,024 controls according to sex, age group and education Milan, Italy, 1985–92.

	Colon cancer Number	%	Rectal cancer Number	%	Control Number	%
Sex						
Males	423	51.1	288	57.8	1189	58.7
Females	405	48.9	210	42.2	835	41.3
Age groups (years)						
<40	36	4.3	22	4.4	266	13.1
40–49	98	11.8	47	9.4	421	20.8
50–59	220	26.6	129	25.9	593	29.3
60–69	322	38.9	204	41.0	588	29.1
70–74	152	18.4	96	19.3	156	7.7
Education (years)						
<7	429	51.8	298	59.8	986	48.7
7–11	223	26.9	126	25.3	591	29.2
≥12	176	21.3	74	14.9	447	22.1

Mantel–Haenszel procedure (Mantel & Haenszel, 1959). Further, to control for several potentially confounding variables, multiple logistic regression (MLR) was used, with maximum likelihood fitting (Baker & Nelder, 1978; Breslow & Day, 1980). All the regression equations included terms of age in decades, sex, education, body mass index, family history of colorectal cancer, plus, whenever indicated, total energy intake. Further allowance for social class, alcohol and coffee consumption did not materially change any of the estimates. Nutrients significantly related to the risk of colorectal neoplasm were also analysed in separate strata of sex and age. Finally, simultaneous allowance for all nutrients significantly related to colorectal cancer after the previous analyses was made by fitting a single model with all significant factors included.

Results

The distribution of cases and controls with reference to sex, age group and education is given in Table I. Cases of colorectal cancer were older than the controls, and cases of rectal (but not colon) cancer tended to be less educated.

Table II gives the distribution of cases and controls according to quintiles of selected micronutrients and of total energy intake, and corresponding cut-off points. These values can be compared with the recommended daily allowance (RDA) of the Italian Society for Human Nutrition (SINU). The values were 60 mg day^{-1} for vitamin C, 2.5 μg day^{-1} for vitamin D, 8.0 mg day^{-1} for vitamin E, 200 μg day^{-1} for folate, 2,200 mg day^{-1} for methionine and 900 mg day^{-1} for calcium (Carnovale & Miuccio, 1989). No RDA was given for retinol and β-carotene. These values are in reasonable agreement with the estimates in the present data set. For β-carotene, ascorbic acid, and folate there was a general tendency for the frequency of cases to decline in the highest consumption quintiles. Cases were less frequent in the lowest quintile of total energy intake but in the absence of a trend in risk across higher quintiles.

The relative risk estimates for various micronutrients considered are given in Table III. The results are presented for colon and rectal cancer separately and, since no material difference was evident, for all colorectal cancers together. No apparent trend in risk across quintiles was evident for retinol, vitamin D, methionine, and calcium. Indeed, methionine was directly associated with risk in univariate analysis, but the apparent association was no longer evident after multiple logistic regression analysis including terms for total energy intake. For β-carotene, ascorbic acid, vitamin E and folate there was a significant trend for a protective effect with increasing consumption quintiles, considering both the model including only age and sex and that including all potential confounding factors. The protective effect was generally more evident after multivariate analysis: the estimated RR for the

Table II Distribution of 828 cases of colon cancer, 498 of rectal cancer and 2,024 controls according to quintiles (on the distribution of controls) of β-carotene, retinol, ascorbic acid, vitamin D, vitamin E, folate, methionine, calcium and total energy intake, Milan, Italy, 1985–92

	1° (low)	*2°*	Quintiles *3°*	*4°*	*5° (high)*
β-carotene (μg day^{-1})					
Upper limits	2268.0	2876.7	3608.7	5608.7	
Controls	398	387	436	415	388
Colon cancer	188	212	199	143	86
Rectal cancer	123	113	115	104	43
Retinol (μg day^{-1})					
Upper limits	1053.7	3990.0	4774.0	5368.0	
Controls	406	404	403	404	407
Colon cancer	166	181	170	171	140
Rectal cancer	109	104	101	92	92
Ascorbic acid (mg day^{-1})					
Upper limits	79.67	104.67	128.00	157.00	
Controls	403	407	405	406	403
Colon cancer	253	192	144	138	101
Rectal cancer	179	107	75	73	64
Vitamin D (μg day^{-1})					
Upper limits	0.79	1.14	1.47	1.97	
Controls	400	412	405	401	406
Colon cancer	154	186	173	186	129
Rectal cancer	107	119	104	97	71
Vitamen E (mg day^{-1})					
Upper limits	3.87	4.63	5.33	6.26	
Controls	405	405	405	403	406
Colon cancer	192	173	188	140	135
Rectal cancer	134	96	97	84	87
Folate (μg day^{-1})					
Upper limits	162.63	195.65	225.80	261.49	
Controls	405	404	405	406	404
Colon cancer	216	186	159	130	137
Rectal cancer	144	113	92	80	69
Methionine (mg day^{-1})					
Upper limits	1377.1	1594.3	1807.1	2039.0	
Controls	404	406	404	406	404
Colon cancer	160	137	176	166	189
Rectal cancer	89	83	127	91	108
Calcium (mg day^{-1})					
Upper limits	468.1	642.1	842.1	1029.7	
Controls	405	405	405	405	404
Colon cancer	156	162	179	171	160
Rectal cancer	90	86	105	113	104
Energy (kcal day^{-1})					
Upper limits	1603.6	1870.0	2132.7	2467.0	
Controls	404	406	404	404	406
Colon cancer	116	177	206	163	166
Rectal cancer	95	99	99	98	107

highest versus lowest quintile was 0.32 for β-carotene, 0.40 for ascorbic acid, 0.60 for vitamin E and 0.52 for folate when colon and rectal neoplasms were considered together. Some protection was also evident for retinol [RR for highest quintile = 0.74, $\chi^2_{1(trend)}$ = 5.3], and vitamin D [RR for highest quintile = 0.74, $\chi^2_{1(trend)}$ = 4.1], but only after allowance for total energy intake.

The relationship between significantly associated nutrients and colorectal cancer risk is further examined in separate strata of sex and age in Table IV. The trends in risk were consistent across strata. Several associations were apparently stronger in females than in males and in subjects less than 60 years than in the elderly, but the intereaction terms were not significant.

When we considered a model including all the nutrients significantly related to colorectal cancer (carotene, ascorbic acid, vitamin E and folate), besides other potential confounding factors, the only persisting protective effects were those of β-carotene and ascorbic acid (Table V). The RRs were 0.71 (95% CI = 0.57–0.88) and 0.38 (95% CI = 0.30–0.50) for the last two quintiles of β-carotene; the corresponding values

Table III   Relative risk estimates (and 95% confidence intervals) of colorectal cancer in relation to selected micronutrient intake[a], Milan, Italy, 1985–92

Quintile of intake	Colon cancer MH[b]	Colon cancer MLR[c]	Rectal cancer MH[b]	Rectal cancer MLR[c]	Colon and rectal cancer MH[b]	Colon and rectal cancer MLR[c]
β-carotene						
Second	1.06 (0.82–1.36)	0.99 (0.77–1.28)	0.89 (0.66–1.21)	0.87 (0.64–1.19)	1.00 (0.80–1.24)	0.95 (0.76–1.18)
Third	0.86 (0.67–1.10)	0.76 (0.58–0.98)	0.79 (0.58–1.06)	0.77 (0.57–1.04)	0.83 (0.67–1.03)	0.77 (0.61–0.96)
Fourth	0.70 (0.54–0.91)	0.55 (0.41–0.73)	0.81 (0.60–1.10)	0.73 (0.53–1.00)	0.74 (0.59–0.93)	0.61 (0.48–0.78)
Fifth (highest)	0.43 (0.32–0.58)	0.31 (0.23–0.43)	0.36 (0.25–0.53)	0.32 (0.22–0.49)	0.40 (0.31–0.52)	0.32 (0.24–0.42)
$\chi^2_{1(trend)}$	37.7[d]	63.0[d]	21.7[d]	25.6[d]	48.9[d]	71.3[d]
Retinol						
Second	1.08 (0.83–1.40)	0.93 (0.71–1.22)	0.97 (0.71–1.33)	0.94 (0.68–1.29)	1.03 (0.82–1.29)	0.92 (0.73–1.16)
Third	1.11 (0.85–1.44)	1.08 (0.82–1.41)	1.01 (0.74–1.38)	1.00 (0.73–1.38)	1.06 (0.84–1.32)	1.03 (0.82–1.30)
Fourth	1.05 (0.81–1.36)	0.92 (0.70–1.20)	0.87 (0.64–1.20)	0.86 (0.62–1.19)	0.99 (0.79–1.24)	0.90 (0.71–1.13)
Fifth (highest)	0.81 (0.62–1.06)	0.71 (0.53–0.94)	0.84 (0.60–1.14)	0.78 (0.56–1.08)	0.81 (0.65–1.03)	0.74 (0.58–0.93)
$\chi^2_{1(trend)}$	2.0	4.6[d]	1.7	2.4	2.9	5.3[d]
Ascorbic acid						
Second	0.76 (0.60–0.97)	0.73 (0.57–0.94)	0.62 (0.47–0.82)	0.60 (0.45–0.81)	0.70 (0.57–0.86)	0.68 (0.55–0.84)
Third	0.63 (0.49–0.81)	0.58 (0.45–0.75)	0.49 (0.36–0.67)	0.46 (0.34–0.64)	0.57 (0.45–0.71)	0.54 (0.43–0.67)
Fourth	0.62 (0.48–0.81)	0.56 (0.43–0.73)	0.50 (0.36–0.68)	0.49 (0.35–0.68)	0.57 (0.46–0.71)	0.53 (0.42–0.67)
Fifth (highest)	0.48 (0.36–0.63)	0.38 (0.28–0.51)	0.47 (0.34–0.66)	0.43 (0.30–0.61)	0.47 (0.37–0.60)	0.40 (0.31–0.51)
$\chi^2_{1(trend)}$	30.5[d]	45.8[d]	26.7[d]	28.3[d]	45.5[d]	58.9[d]
Vitamin D						
Second	1.18 (0.91–1.53)	1.10 (0.84–1.44)	1.13 (0.83–1.52)	1.13 (0.83–1.54)	1.16 (0.93–1.45)	1.11 (0.89–1.39)
Third	1.10 (0.85–1.44)	1.10 (0.76–1.31)	0.98 (0.72–1.34)	0.97 (0.70–1.33)	1.06 (0.84–1.33)	0.99 (0.78–1.24)
Fourth	1.28 (0.98–1.66)	1.15 (0.88–1.51)	1.01 (0.74–1.39)	1.03 (0.74–1.42)	1.18 (0.94–1.48)	1.11 (0.88–1.40)
Fifth (highest)	0.87 (0.66–1.15)	0.75 (0.56–1.01)	0.73 (0.52–1.03)	0.73 (0.51–1.03)	0.82 (0.64–1.04)	0.74 (0.58–0.95)
$\chi^2_{1(trend)}$	0.2	2.3	3.3	3.2	1.7	4.1[d]
Vitamin E						
Second	0.97 (0.75–1.25)	0.81 (0.62–1.06)	0.78 (0.58–1.06)	0.73 (0.53–1.00)	0.90 (0.73–1.13)	0.78 (0.62–0.98)
Third	1.18 (0.91–1.52)	0.84 (0.63–1.11)	0.89 (0.66–1.21)	0.76 (0.54–1.07)	1.06 (0.85–1.32)	0.80 (0.63–1.02)
Fourth	0.93 (0.71–1.21)	0.60 (0.44–0.81)	0.81 (0.59–1.11)	0.68 (0.47–0.98)	0.88 (0.70–1.10)	0.62 (0.48–0.81)
Fifth (highest)	0.97 (0.74–1.28)	0.58 (0.42–0.81)	0.89 (0.64–1.22)	0.67 (0.45–0.98)	0.93 (0.74–1.17)	0.60 (0.45–0.80)
$\chi^2_{1(trend)}$	0.1	12.9[d]	0.4	3.6	0.5	13.7[d]
Folate						
Second	0.94 (0.73–1.20)	0.82 (0.63–1.06)	0.90 (0.68–1.21)	0.84 (0.62–1.14)	0.93 (0.75–1.15)	0.83 (0.66–1.03)
Third	0.88 (0.68–1.14)	0.69 (0.53–0.91)	0.77 (0.57–1.05)	0.67 (0.48–0.93)	0.84 (0.67–1.05)	0.68 (0.54–0.86)
Fourth	0.76 (0.58–0.99)	0.56 (0.42–0.75)	0.72 (0.52–0.99)	0.59 (0.41–0.83)	0.74 (0.59–0.92)	0.56 (0.43–0.72)
Fifth (highest)	0.83 (0.64–1.08)	0.55 (0.41–0.75)	0.65 (0.46–0.90)	0.49 (0.33–0.71)	0.75 (0.60–0.94)	0.52 (0.40–0.68)
$\chi^2_{1(trend)}$	3.6	19.6[d]	8.8[d]	16.9[d]	9.7[d]	30.5[d]

Table III – *continued*

	Colon cancer		Rectal cancer		Colon and rectal cancer	
Quintile of intake	MH^b	MLR^c	MH^b	MLR^c	MH^b	MLR^c
Methionine						
Second	0.90	0.75	0.99	0.98	0.93	0.82
	(0.69–1.19)	(0.56–1.00)	(0.70–1.38)	(0.69–1.39)	(0.73–1.17)	(0.64–1.05)
Third	1.09	0.82	1.43	1.39	1.21	1.01
	(0.84–1.42)	(0.62–1.09)	(1.04–1.95)	(0.99–1.96)	(0.97–1.52)	(0.79–1.28)
Fourth	1.10	0.79	1.12	1.13	1.12	0.89
	(0.85–1.45)	(0.58–1.06)	(0.81–1.56)	(0.77–1.65)	(0.89–1.40)	(0.68–1.16)
Fifth (highest)	1.41	1.00	1.46	1.40	1.41	1.12
	(1.08–1.82)	(0.72–1.39)	(1.06–2.02)	(0.94–2.10)	(1.13–1.77)	(0.84–1.49)
$\chi^2_{1(trend)}$	8.5^d	0.0	5.7^d	3.1	11.3^d	0.9
Calcium						
Second	0.97	0.84	0.88	0.85	0.94	0.86
	(0.74–1.27)	(0.64–1.11)	(0.63–1.23)	(0.60–1.19)	(0.75–1.19)	(0.68–1.09)
Third	1.08	0.91	1.12	1.09	1.09	0.97
	(0.83–1.41)	(0.69–1.20)	(0.81–1.55)	(0.79–1.52)	(0.87–1.37)	(0.77–1.23)
Fourth	0.99	0.82	1.14	1.12	1.05	0.92
	(0.76–1.29)	(0.62–1.09)	(0.83–1.57)	(0.80–1.55)	(0.83–1.31)	(0.73–1.17)
Fifth (highest)	0.98	0.73	1.13	1.05	1.04	0.84
	(0.75–1.28)	(0.54–0.97)	(0.82–1.56)	(0.75–1.49)	(0.83–1.31)	(0.65–1.08)
$\chi^2_{1(trend)}$	0.0	3.9^d	1.8	1.1	0.5	0.9

[a]Reference category is the lowest quintile. [b]Mantel–Haenszel estimates adjusted for age in decades and sex. [c]Multiple logistic regression estimates adjusted for age, sex, education, family history of colorectal cancer, body mass index and total energy intake. [d]$P < 0.05$.

were 0.58 (95% CI = 0.44–0.75) and 0.52 (95% CI = 0.38–0.69) for ascorbic acid, and the trends in risk were significant. The relative risk estimates were 1.05 for the highest quintile of vitamin E and 1.1 for folate.

Discussion

This study suggests that carotene and ascorbic acid can have a protective effect on risk of colorectal cancer, while there was no evidence of protection by other micronutrients considered, such as retinol, vitamin D, methionine and calcium. There was also some evidence of a protective effect of vitamin E and folate, but this was no longer apparent after inclusion of these factors in a single model with β-carotene and ascorbic acid. Most results were similar when colon and rectal cancers were analysed separately.

Published data on micronutrients and colorectal cancer risk vary. Some studies have suggested protection by calcium (Newmark et al., 1984; Garland et al., 1985; Sorenson et al., 1988), ascorbic acid (Kune et al., 1987), β-carotene (Benito et al., 1991), folate (Freudenheim et al., 1991; Giovannucci et al., 1993) or vitamin E (Longnecker et al., 1992), but there have been no systematic efforts to allow for the potential effect of one micronutrient on that of others. This is of specific interest, since several of these micronutrients are highly correlated. In the present data set, for instance, the correlation of ascorbic acid was 0.49 with vitamin E and 0.45 with folate or β-carotene. This is not surprising, since these micronutrients are derived from similar sources, such as various types of fruits and vegetables, whose consumptions tend also to be correlated. In a cohort investigation (Giovannucci et al., 1993) of colorectal adenomas, folate was systematically adjusted for vitamins A, C, D and E and β-carotene: the protective effect of folate seemed to persist, while that of other micronutrients declined after allowance was made for simultaneous intake. The question of which specific micronutrients are protective on colorectal cancer risk is therefore still unsettled, and the issue of a more general protective effect of fresh fruit and vegetables (as opposed to that of specific micronutrients) remains open to discussion, at least in part because of the collinearity between

various micronutrients and between selected foods (such as fruits and vegetables) and micronutrients.

Some of the lack of association deserves comments too, particularly the absence of a protective effect of vitamin D and calcium, whose protective role has been indicated by several studies (Newmark et al., 1984; Garland et al., 1985; Sorenson et al., 1988). If not due to chance or bias, this may be related to the levels of intake suggested to have a protective effect (e.g. above 1,500–1,800 mg per day; Newmark & Lipkin, 1992), which were considerably higher than the cutoff points even of the highest quintile in this data set. This may be the result of an underestimate of vitamin D intake in this study. Further, apparent differences between various studies may be related to non-dietary sources of vitamin D (Garland et al., 1985) (and perhaps other micronutrients), including supplementation, although this is relatively uncommon in Italy.

This study was sufficiently large to obtain reasonably precise risk estimates and significant trends in risk for several micronutrients. Besides statistical power, potential sources of error or bias should be considered, starting with the reliability and validity of the estimated micronutrient intake. The limited number of foods on which estimates of micronutrient intake were based is likely to have caused some degree of underestimate for various values. Still, the comparison with the average levels of nutrient intake recommended for the Italian population (Carnovale & Miuccio, 1989) is reassuring in terms of reasonable validity of available information, although these estimates were based on the consumption of 29 food items only. Further, this study was able to find a number of significant associations with specific food items (direct with starchy foods and meats, and inverse with vegetables and coffee) (La Vecchia et al., 1988), generally consistent with our knowledge on dietary habits in colorectal carcinogenesis (Willett, 1989).

The fact that cases and controls came from comparable catchment areas, the almost complete participation rate and the absence of apparent confounding with reference to the issue of interest, including allowance for an estimate of total energy intake (Willett & Stampfer, 1986), indicate that selection, information or confounding bias is unlikely to have occurred.

Table IV Relative risk estimates (and 95% confidence intervals) of colorectal cancer in relation to selected micronutrient intake in separate strata of sex and age[a,b], Milan, Italy, 1985-92

Quintile of intake	Sex Males	Females	Age <60 years	≥60 years
β-carotene				
Second	1.33	0.60	0.93	0.97
	(1.00-1.78)	(0.42-0.86)	(0.68-1.28)	(0.71-1.34)
Third	0.87	0.59	0.74	0.79
	(0.65-1.18)	(0.41-0.84)	(0.54-1.02)	(0.58-1.09)
Fourth	0.65	0.50	0.62	0.60
	(0.48-0.89)	(0.34-0.74)	(0.44-0.86)	(0.43-0.85)
Fifth	0.46	0.17	0.28	0.33
	(0.32-0.67)	(0.11-0.26)	(0.19-0.42)	(0.22-0.49)
$\chi^2_{1(trend)}$	25.1[c]	60.0[c]	42.0[c]	33.0[c]
Ascorbic acid				
Second	0.90	0.49	0.57	0.81
	(0.67-1.20)	(0.35-0.68)	(0.41-0.78)	(0.61-1.07)
Third	0.72	0.38	0.44	0.64
	(0.53-0.98)	(0.27-0.54)	(0.31-0.61)	(0.46-0.87)
Fourth	0.73	0.32	0.42	0.66
	(0.54-0.99)	(0.22-0.46)	(0.30-0.58)	(0.47-0.92)
Fifth	0.54	0.24	0.29	0.54
	(0.38-0.76)	(0.16-0.35)	(0.21-0.41)	(0.37-0.78)
$\chi^2_{1(trend)}$	13.9[c]	62.5[c]	52.0[c]	15.0[c]
Vitamin E				
Second	0.98	0.63	0.83	0.73
	(0.70-1.37)	(0.46-0.86)	(0.58-1.18)	(0.54-0.99)
Third	0.88	0.72	0.72	0.86
	(0.62-1.23)	(0.50-1.03)	(0.50-1.04)	(0.62-1.20)
Fourth	0.82	0.43	0.53	0.70
	(0.58-1.17)	(0.28-0.64)	(0.36-0.78)	(0.49-1.02)
Fifth	0.77	0.40	0.52	0.75
	(0.53-1.13)	(0.25-0.64)	(0.34-0.79)	(0.50-1.14)
$\chi^2_{1(trend)}$	2.3	17.4[c]	13.0[c]	3.0
Folate				
Second	0.87	0.85	0.93	0.78
	(0.63-1.19)	(0.62-1.17)	(0.66-1.30)	(0.58-1.05)
Third	0.87	0.51	0.72	0.63
	(0.63-1.21)	(0.36-0.73)	(0.51-1.01)	(0.46-0.87)
Fourth	0.74	0.42	0.51	0.65
	(0.53-1.04)	(0.28-0.61)	(0.35-0.73)	(0.46-0.93)
Fifth	0.63	0.37	0.43	0.67
	(0.44-0.91)	(0.24-0.57)	(0.29-0.64)	(0.45-0.98)
$\chi^2_{1(trend)}$	6.5[c]	32.7[c]	26.0[c]	3.0

[a]Reference category is the lowest quintile. [b]Multiple logistic regression estimates adjusted for age, sex, education, family history of colorectal cancer, body mass index and total energy intake. [c]$P < 0.05$.

Table V Relative risk estimates (and 95% confidence intervals) of colorectal cancer in relation to selected micronutrient intake[a], Milan, Italy, 1985-92

Quintile of intake	Colon and rectal cancer MLR[b]	
Vitamin E		
Second	0.92	
	(0.74-1.15)	
Third	0.97	
	(0.76-1.23)	
Fourth	0.96	
	(0.74-1.26)	
Fifth (highest)	1.05	
	(0.78-1.41)	$\chi^2_{1(trend)} = 0.0$
Folate		
Second	1.09	
	(0.87-1.37)	
Third	1.06	
	(0.82-1.36)	
Fourth	1.00	
	(0.75-1.34)	
Fifth (highest)	1.10	
	(0.79-1.53)	$\chi^2_{1(trend)} = 0.0$
β-Carotene		
Second	1.00	
	(0.82-1.23)	
Third	0.83	
	(0.67-1.02)	
Fourth	0.71	
	(0.57-0.88)	
Fifth (highest)	0.38	
	(0.30-0.50)	$\chi^2_{1(trend)} = 52.0$[c]
Vitamin C		
Second	0.68	
	(0.55-0.83)	
Third	0.59	
	(0.47-0.75)	
Fourth	0.58	
	(0.44-0.75)	
Fifth (highest)	0.52	
	(0.38-0.70)	$\chi^2_{1(trend)} = 22.0$[c]

[a]Reference category is the lowest quintile. [b]Multiple logistic regression estimates adjusted for age, sex, education, family history of colorectal cancer, body mass index and total energy intake plus all the above variables. [c]$P < 0.05$.

In conclusion, this analysis of a large case-control study provides support for a protective effect of a few selected nutrients on colorectal cancer risk. The protective effect persisted after allowance for several covariates, including total energy intake. Only β-carotene and ascorbic acid, however, remained significantly protective after simultaneous allowance for various micronutrients. Whether this reflects a specific (or stronger) effect of these micronutrients, rather than problems of data analysis following collinearity between micronutrients or other limitations of available data, remains open for discussion.

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