

Association between nutrient intake related to the one-carbon metabolism and colorectal cancer risk: a case-control study in the Basque Country

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Supplementary Table S1. Descriptive socio-demographic and clinical data of the participants (cases vs controls)

Characteristics	Cases (n=308)		Controls (n=308)		<i>p</i>
Sex, % of men	66.2		66.2		
Age, y, mean SD	61.5	5.2	61.1	5.5	0.093
Smoking status, %					
Never	27.9		38.6		
Past/current	72.1		61.4		0.004
BMI, %					
Underweight	6.5		7.8		
Normal weight	26.0		34.1		
Overweight/obesity	67.5		58.1		0.033
PRM, % ¹					
L _{1.2}	15.6		12.3		
L _{3.4}	83.4		79.2		<0.001
DI, % ¹					
Q _{1.3}	47.1		65.6		
Q _{4.5}	18.8		29.5		<0.001

¹Valid percentages

BMI body mass index, *DI* deprivation index (this index was successfully assigned to 80.2% of the study sample), *L* level, *PRM* predictive risk modelling (this index was successfully assigned to 95.1% of the study sample), *Q* quintile, *SD* standard deviation

Supplementary Table S2. Daily intake of one-carbon metabolism nutrients in cases and control studied

Daily nutrient intake	Cases (n=308)					Controls (n=308)					<i>p</i> ^a
	Mean	SD	Median	P25	P75	Mean	SD	Median	P25	P75	
<i>Vitamins related to OCM</i>											
Vitamin B ₂ , mg/d	1.5	0.5	1.4	1.2	1.7	1.6	0.5	1.5	1.2	1.9	0.100
Vitamin B ₃ , mg/d	30.1	7.1	30.2	24.3	35.0	30.3	7.7	29.7	25.2	34.0	0.893
Vitamin B ₆ , mg/d	2.7	14.3	1.8	1.5	2.1	1.9	0.6	1.8	1.5	2.2	0.378
Folate, µg/d	267.2	80.1	261.0	209.0	307.0	273.3	76.5	261.5	217.3	325.0	0.406
Vitamin B ₁₂ , µg/d	6.6	28.5 ^b	4.8	3.8	6.0	4.9	1.7	4.5	3.7	5.8	0.094
<i>Other dietary methyl donors</i>											
Methionine, mg/d	1753.7	644.6	1717.5	1218.5	2146.3	1894.2	747.6	1810.5	1255.8	2473.00	0.012
Choline, mg/d	134.7	85.4	120.5	80.8	170.0	163.2	85.0	147.0	99.3	212.8	<0.001
Betaine, mg/d	113.6	55.2	101.5	71.4	144.8	151.0	62.9	141.5	102.0	187.8	<0.001
<i>Methyl donor index</i> ^c	-4.1	1.3	-4.1	-4.9	-3.2	-4.0	1.3	-4.0	-4.8	-3.1	0.653

^aDifferences between cases and controls. A value of $p < 0.05$ was considered significant. Significant results are highlighted in bold

^bThis high SD is due to the use of a nutritional supplement by one of the cases

^cThis variable follows a normal distribution (the rest of the variables follow a non-normal distribution). This index was derived by summing standardized intake values on the log scale [(value – mean)/SD] across the 8 individual nutrients

Met Methionine, *OCM* one-carbon metabolism, *P25* 25 percentile, *P75* 75 percentile, *SD* standard deviation

Supplementary Table S3. Top five food sources of total choline and betaine among control subjects

Food sources	Proportion
Total choline	
Eggs	24.9
Sausage	7.9
Lentils	7.9
Beef	5.8
Tuna	5.3
Betaine	
Pasta	48.1
Green beans	17.5
Potatoes	11.1
Beef	8.5
Lettuce	4.9

Supplementary Table S4. Association between the intake of nutrients related to one-carbon metabolism and colorectal cancer risk in men

Nutrients ^a	Case/control(n)	Model I ^b		Model II ^c		Model III ^d	
		OR (95% CI)	<i>p</i> ^e	OR (95% CI)	<i>p</i> ^e	OR (95% CI)	<i>p</i> ^e
Vitamin B ₂							
T1	89/82	1.00	-	1.00	-	1.00	-
T2	55/58	0.88(0.55-1.40)	0.586	0.61(0.33-1.14)	0.122	0.50(0.21-1.16)	0.108
T3	60/64	0.86(0.54-1.38)	0.535	0.47(0.22-0.99)	0.046	0.59(0.20-1.79)	0.355
Vitamin B ₃							
T1	72/73	1.00	-	1.00	-	1.00	-
T2	56/59	0.97(0.58-1.61)	0.897	1.09(0.54-2.21)	0.813	1.01(0.37-2.77)	0.986
T3	76/72	1.07(0.66-1.73)	0.779	1.00(0.55-1.83)	0.995	1.05(0.31-3.56)	0.938
Vitamin B ₆							
T1	69/70	1.00	-	1.00	-	1.00	-
T2	81/63	1.28(0.80-2.05)	0.308	0.96(0.51-1.81)	0.891	0.81(0.30-2.18)	0.680
T3	54/71	0.75(0.45-1.25)	0.266	0.61(0.30-1.25)	0.176	0.71(0.18-2.80)	0.625
Folate							
T1	84/73	1.00	-	1.00	-	1.00	-
T2	66/62	0.92(0.58-1.45)	0.704	0.88(0.49-1.56)	0.655	0.80(0.37-1.76)	0.585
T3	54/69	0.69(0.43-1.11)	0.123	0.67(0.36-1.26)	0.220	1.05(0.31-3.56)	0.955
Vitamin B ₁₂							
T1	54/68	1.00	-	1.00	-	1.00	-
T2	73/68	1.33(0.82-2.14)	0.248	1.28(0.70-2.33)	0.422	1.58(0.66-3.80)	0.302
T3	77/68	1.41(0.87-2.30)	0.162	1.85(0.85-4.01)	0.119	2.62(0.88-7.74)	0.082
Met							
T1	70/68	1.00	-	1.00	-	1.00	-
T2	66/69	0.93(0.58-1.50)	0.768	0.87(0.48-1.58)	0.646	0.86(0.42-1.78)	0.692
T3	68/67	0.98(0.60-1.61)	0.942	0.67(0.34-1.32)	0.251	0.72(0.32-1.64)	0.434

Choline								
T1	92/71	1.00	-	1.00	-	1.00	-	
T2	68/73	0.72(0.46–1.13)	0.155	0.77(0.43–1.38)	0.379	0.74(0.35–1.55)	0.421	
T3	44/60	0.55(0.33–0.92)	0.024	0.64(0.34–1.22)	0.174	0.75(0.34–1.64)	0.469	
Betaine								
T1	122/65	1.00	-	1.00	-	1.00	-	
T2	46/67	0.39(0.23–0.64)	<0.001	0.62(0.34–1.15)	0.128	0.58(0.28–1.21)	0.149	
T3	36/72	0.29(0.18–0.49)	<0.001	0.30(0.16–0.56)	<0.001	0.27(0.13–0.58)	<0.001	
<i>Methyl donor index^{f,g}</i>								
T1	98/61	1.00	-	1.00	-	-	-	
T2	75/71	0.63(0.40–1.02)	0.058	0.42(0.22–0.78)	0.006	-	-	
T3	31/72	0.28(0.16–0.48)	<0.001	0.19(0.09–0.41)	<0.001	-	-	

^aNutrients related to one-carbon metabolism were categorized into tertiles according to the distribution in controls; Tertiles of vitamin B₂ (mg/d): T1 < 1.4, T2 1.4–1.7, T3 > 1.7; vitamin B₃ (mg/d): T1 < 26.5, T2 26.5–32.4, T3 > 32.4; vitamin B₆ (mg/d): T1 < 1.6, T2 1.6–2.0, T3 > 2.0; folate (µg/d): T1 < 238.0, T2 238.0–298.0, T3 > 298.0; vitamin B₁₂ (µg/d): T1 < 4.0, T2 4.0–5.3, T3 > 5.3; methyl donor index: T1 < -4.46; T2 -4.46–-3.47, T3 > -3.47; Met (mg/d) for men: T1 < 1377.0, T2 1377.0–1985.0, T3 > 1985.0; Met (mg/d) for women: T1 < 1665.0, T2 1665.0–2531.0, T3 > 2531.0; total choline (mg/d): T1 < 117.0, T2 117.0–187.0, T3 > 187.0; betaine (mg/d): T1 < 119.0, T2 119.0–170.0, T3 > 170.0

^bModel I, analyses were performed using crude conditional logistic regression, without considering confounding factors

^cModel II, analyses were performed using conditional logistic regression analysis adjusted for age (50–59 years old, 60–69 years old), sex, body mass index (underweight/normal weight, overweight/obesity), energy intake (kcal/d), physical exercise level (< 15 min/d of cycling/sports, ≥ 15 min/d), smoking status and intensity of smoking (never; past: quit smoking ≥ 11 years ago, quit < 11 years ago; smoker: ≤ 15 cigarettes/d, > 15 cigarettes/d), alcohol intake (standard unit drinks/d, tertiles in controls: men: T1 < 0.36, T2 0.36–1.13, T3 > 1.13; women: T1 < 0.08, T2 0.8–0.69, T3 > 0.69), Deprivation Index (quintile 1–3, quintile 4–5) and Predictive Risk Modelling (level 1–2, level 3–4), including nutrient intakes separately; participants with missing data for the confounding variables were included as a separate category for these variables

^dModel III, model II including all the nutrients related to one-carbon metabolism

^eA value of $p < 0.05$ was considered significant. Significant results are highlighted in bold

^fModel III is not applicable because this index includes all nutrients in its construction

^gThis index was derived by summing standardized intake values on the log scale [(value – mean)/SD] across the 8 individual nutrients

CI Confidence interval, *Met* Methionine, *OR* Odds ratio, *T* Tertile

Supplementary Table S5. Association between the intake of nutrients related to one-carbon metabolism and colorectal cancer risk in women

Nutrients ^a	Case/control(n)	Model I ^b		Model II ^c		Model III ^d	
		OR (95% CI)	<i>p</i> ^e	OR (95% CI)	<i>p</i> ^e	OR (95% CI)	<i>p</i> ^e
Vitamin B ₂							
T1	29/26	1.00	-	1.00	-	1.00	-
T2	34/44	0.73(0.38-1.42)	0.356	0.88(0.37-2.05)	0.760	3.63(0.06-221.1)	0.087
T3	41/34	1.10(0.55-2.21)	0.794	1.38(0.50-3.81)	0.533	37.2(0.04-31674.4)	0.085
Vitamin B ₃							
T1	31/28	1.00	-	1.00	-	1.00	-
T2	27/46	0.49(0.23-1.05)	0.065	0.18(0.05-0.66)	0.010	0.02(0.00-0.62)	0.027
T3	46/30	1.36(0.66-2.80)	0.402	0.78(0.22-2.74)	0.693	1.77(0.07-41.72)	0.724
Vitamin B ₆							
T1	25/19	1.00	-	1.00	-	1.00	-
T2	46/50	0.74(0.38-1.43)	0.367	0.55(0.22-1.33)	0.182	0.28(0.00-20.53)	0.563
T3	33/35	0.74(0.36-1.55)	0.430	0.48(0.18-1.33)	0.158	0.82(0.01-55.58)	0.928
Folate							
T1	28/29	1.00	-	1.00	-	1.00	-
T2	37/42	0.90(0.45-1.80)	0.769	0.45(0.17-1.15)	0.094	0.07(0.00-2.60)	0.150
T3	39/33	1.18(0.61-2.28)	0.622	0.67(0.27-1.64)	0.381	0.01(0.00-1.13)	0.056
Vitamin B ₁₂							
T1	32/34	1.00	-	1.00	-	1.00	-
T2	35/35	1.07(0.54-2.13)	0.849	1.33(0.53-3.37)	0.542	0.04(0.00-1.39)	0.075
T3	37/35	1.13(0.57-2.25)	0.725	0.89(0.29-2.77)	0.844	0.01(0.00-1.94)	0.087
Met							
T1	41/34	1.00	-	1.00	-	1.00	-
T2	46/35	1.13(0.61-2.09)	0.708	1.14(0.54-2.42)	0.726	0.99(0.15-6.37)	0.992
T3	17/35	0.48(0.24-0.94)	0.034	0.31(0.11-0.86)	0.024	0.01(0.00-2.37)	0.098

Choline								
T1	55/32	1.00	-	1.00	-	1.00	-	
T2	31/29	0.70(0.35–1.43)	0.330	0.35(0.12–0.98)	0.046	0.13(0.01–2.47)	0.176	
T3	18/43	0.27(0.13–0.56)	<0.001	0.21(0.07–0.60)	0.004	1.57(0.09–26.34)	0.755	
Betaine								
T1	72/37	1.00	-	1.00	-	1.00	-	
T2	18/37	0.26(0.12–0.54)	<0.001	0.28(0.11–0.74)	0.011	0.01(0.00–1.59)	0.077	
T3	14/30	0.26(0.12–0.56)	<0.001	0.18(0.06–0.50)	0.001	0.00(0.00–0.78)	0.041	
<i>Methyl donor index^{f,g}</i>								
T1	57/41	1.00	-	1.00	-	-	-	
T2	37/32	0.92(0.46–1.86)	0.824	0.87(0.35–2.14)	0.762	-	-	
T3	10/31	0.27(0.12–0.61)	0.002	0.27(0.09–0.77)	0.015	-	-	

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