Published in final edited form as:
Int J Colorectal Dis. 2009 June ; 24(6): 647–654. doi:10.1007/s00384-009-0656-8.

Genetic polymorphisms in the cyclooxygenase-1 and cyclooxygenase-2 genes and risk of colorectal adenoma

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Abstract

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Conflict of interest The authors declare that they have no conflict of interest.

Purpose—Cyclooxygenase (COX) enzymes, COX1 and COX2, are key in converting arachidonic acid (AA) into prostaglandins that have been associated with colorectal carcinogenesis. The aim of our study was to investigate associations of polymorphisms in COX genes, alone and in interaction with exposures known to be related to inflammation and AA metabolism, with risk of colorectal adenomas.

Materials and methods—In a community-, colonoscopy-based case–control study with 162 incident, sporadic colorectal adenoma cases and 211 controls, we investigated associations of two promoter polymorphisms (−842 A >G in COX1 and −765 G>C in COX2) and two polymorphisms in the 3′-UTR of COX2 (8473 T>C and 9850 A>G) with risk of adenomas. Multiple logistic regression models were used to estimate odds ratios (OR) and 95% confidence intervals (CI) of colorectal adenoma after adjusting for potential confounders.

Results—Overall, there was no evidence for an association between any of the four polymorphisms and colorectal adenomas. However, we found a statistically significant interaction between the COX2 8473 T>C polymorphism and nonsteroidal anti-inflammatory drug (NSAIDs) use ($P_{\text{interaction}}$ =0.03): The greatest reduced risk was observed for individuals with the 8473 C variant allele who also regularly used NSAIDs (OR=0.35, 95% CI 0.16–0.75).

Conclusion—These results suggest that the C allele of COX2 8473 T>C polymorphism may interact with NSAIDs to reduce risk for colorectal adenoma.

Keywords

COX1; COX2; Polymorphisms; Inflammation; Colorectal adenoma

Introduction

Genetic susceptibility, environmental factors, and gene–environment interactions are related to the occurrence of colorectal cancer, though the risk attributable to each is unclear [1]. Evidence from experimental and epidemiological studies indicates that lipid metabolism, especially in the arachidonic acid (AA) pathway, plays a critical role in inflammation and colorectal carcinogenesis [2, 3]. AA is metabolized primarily by the cyclooxygenase (COX) and lipoxygenase enzymes. The COX enzymes, COX1 and COX2, can metabolize AA to produce eicosanoids such as prostaglandins (PGs) and thromboxanes [3]. The functions of these bioactive lipid metabolites have been linked to inflammation, mitogenesis, and differentiation [4].

The COX1 gene is constitutively expressed at relatively low levels in most tissues. COX2, on the other hand, is rapidly induced by growth factors, inflammatory cytokines, and tumor promoters [5]. Increased COX2 expression is observed in both sporadic colorectal adenomas and carcinomas compared to normal mucosa [6, 7]. Also, COX2-synthesized PGs (e.g., PGE2) are highly concentrated in colorectal tumors and have been shown to play roles in tumorigenesis [8-10]. Strong and consistent evidence from experimental, animal, and epidemiologic studies indicates that the use of aspirin or other nonsteroidal anti-inflammatory drugs (NSAIDs) that inhibit COX enzymes is associated with decreased risk of colorectal cancer and adenoma incidence and recurrence [11].

Polymorphisms in COX genes may affect the expression or activity of the two enzymes and consequently alter an individual's susceptibility to colorectal adenoma and cancer risk. One single-nucleotide polymorphism (SNP) in the COX1 gene and three SNPs in the COX2 gene were the focus of this study. The −842 A>G polymorphism, located in the promoter region of the COX1 gene, was found to differentially affect the response to aspirin [12]. The −765 G>C polymorphism is located in the promoter region of COX2, while polymorphisms of the 8473 T>C and 9850 A>G polymorphisms are located in exon 10 (3′-untranslated region: 3-

UTR). The −765 C variant allele, located within a putative transcription-factor binding site for Sp1, has significantly lower promoter activity than the −765 G allele [13]. The functional importance of 8473 T>C and 9850 A>G polymorphisms is unclear; however, the 3′-UTR of the human COX2 gene encoded by exon 10 contains multiple elements that regulate mRNA stability and translation efficiency [14, 15].

The purpose of this study is to (1) examine associations of the COX gene polymorphisms with risk of colorectal adenoma, (2) explore possible interactions of the polymorphisms with several inflammation- or AA-pathway related risk factors (e.g., dietary fat, fatty acids, and NSAID use) in relation to risk of adenoma, and (3) investigate whether associations differ by adenoma characteristics.

Materials and methods

Study design and study population

The design and population characteristics of this community-, colonoscopy-based case–control study of incident, sporadic colorectal adenoma was described previously [16, 17]. Briefly, participants were recruited through community gastroenterology practices in Winston-Salem and Charlotte, North Carolina. All patients who were scheduled for a colonoscopy between 1994 and 1997 were screened for specific study eligibility criteria and then recruited before colonoscopy. Eligibility criteria included 30–74 years of age, no previous adenoma, no individual history of cancer (except nonmelanoma skin cancer), no known genetic syndromes associated with predisposition to colonic neoplasia (i.e., no clinical history of familial adenomatous polyposis (FAP), Gardner's syndrome, or hereditary nonpolyposis colorectal cancer syndrome prior to colonoscopy and no findings at colonoscopy that were consistent with FAP), no history of ulcerative colitis or Crohn's disease, and resident of either of the above two North Carolina metropolitan areas. The study was approved by the Institutional Review Board of Wake Forest University.

Cases were identified as eligible colonoscopy patients who were determined to have study pathologist-confirmed incident adenomatous polyps, and controls were identified as those patients without adenomatous polyps. All polyps were removed at colonoscopy and examined by one index study pathologist using criteria from the National Polyp Study [18]. Information on polyp location, number, size, shape, histological type, and degree of dysplasia were collected.

Among all four clinical sites, 617 initially eligible participants were contacted, and 417 participants signed the consent form and participated in the study. Of the 417 participants, 179 had adenomatous polyps and 238 were adenoma free. Persons who had only hyperplastic polyps (n=80) were included as controls. We further excluded 15 participants (n=7 cases and 8 controls) who (1) reported an implausible total energy intake (<500 or >6,000 kcal/day) on their food frequency questionnaire (FFQ; n=2 cases and 6 controls), (2) left more than 15 blanks on their FFQ (n=3 cases and 1 control), or (3) provided incomplete medical history information (n=2 cases and 1 control). A total of 402 participants remained eligible for analysis on whom 373 participants (162 cases and 211 controls) had sufficient DNA for genotyping and thus for final data analyses.

Data collection

Self-administered questionnaires were sent to all participants before their colonoscopy visit. Information was collected on basic demographics (age, sex, education, marital status, and race), lifestyle factors, medical history, anthropometry (height, weight), and family history of colorectal adenoma or cancer. Dietary information was assessed using an adaptation of the Willett semi-quantitative FFQ (153 items), which was expanded to include additional

vegetables, fruit, and low-fat foods [19]. Physical activity data were obtained using a modified Paffenbarger questionnaire [20]. Information also was collected on smoking history, alcohol use, and regular use of aspirin and other NSAIDs. A person was defined as a regular aspirin or NSAID user if he or she consumed these drugs at least once a week.

SNP genotyping

Genomic DNA was extracted from stored white blood cells. The genomic DNA pellets (50–100μg) were dissolved in 300–800μl of TE buffer, of which about 1 μl was used for each polymerase chain reaction (PCR) reaction. Genetic polymorphisms were detected by the PCR-restriction fragment length polymorphism (RFLP) method. Laboratory personnel were blinded as to which samples were from cases or controls.

COX2 (−765 G>C: rs20417)—A 284-base-pair fragment of the COX2 gene was amplified using the following two primers: (forward) 5′-CCA TCA GAA GGC AGG AAA C-3′ and (reverse) 5′-GCT CTA TAT GCA GCA CAT AC-3′. The PCR was performed in a 25μl reaction mixture containing standard PCR buffer, 1.0 mM $MgCl_2$, 0.2 mM dNTP, 1 unit *Taq* polymerase (Gibco-Invitrogen), and 0.4μM of each oligonucleotide primer. The reactions were heated to 94°C for 2 min, followed by 35 cycles of 94°C for 30 s, 64°C for 30 s, 72°C for 45 s, and a final extension at 72°C for 7 min. PCR products were digested with the *Aci*I restriction enzyme overnight and separated by gel electrophoresis.

COX2 (8473 T>C: rs5275)—A 159-base-pair fragment of the COX2 gene was amplified using the following primers: (forward) 5′-GAA ATT TAA AGT ACT TTT GAT-3′ and (reverse) 5′-CCT ATG AAT TTA GAA TTT AGA AAT TTC-3′. The PCR was performed in a 25μl reaction mixture of containing standard PCR buffer, 1.5 mM $MgCl_2$, 0.2 mM dNTP, 1 unit *Taq* polymerase (Gibco-Invitrogen), and 0.4μM of each oligonucleotide primer. The reactions were then heated to 94°C for 2 min, followed by 35 cycles of 94°C for 30 s, 50°C for 30 s, 72°C for 40 s, and a final extension at 72°C for 7 min. PCR products were digested with the *Bcl*I restriction enzyme overnight and separated by gel electrophoresis.

COX2 (9850 A> G: rs4648298)—A 545-base-pair fragment of the COX2 gene was amplified using the following primers: (forward) 5′-CGT TCC CAT TCT AAT TAA TGC CCT T-3′ and (reverse) 5′-ATT AAA ACC CAC AGT GCT TGA CAC A-3′. The PCR was performed in a 25μl reaction mixture of containing standard PCR buffer, 1.0 mM $MgCl_2$, 0.2 mM dNTP, 1 unit *Taq* polymerase (Gibco-Invitrogen), and 0.4μM of each oligonucleotide primer. Cycling for this polymorphism was as follows: 94°C for 2 min, 35 cycles of 94°C for 30 s, 60°C for 30 s, 72°C for 40 s, and a final extension at 72°C for 7 min. PCR products were digested with the *AluI* restriction enzyme overnight and then separated by gel electrophoresis.

COX1 (−842 A>G [12])—A 190-base-pair fragment of the COX1 gene was amplified using the following two primers: (forward) 5′-CGA TAA CTG AGC ACC TAC ATG CTG G-3′ and (reverse) 5′-CCA GAC TCC ACA GCT TAC TG-3′. The PCR was performed in a 25μl reaction mixture containing standard PCR buffer, 1.5 mM $MgCl_2$, 0.2 mM dNTP, 1 unit *Taq* polymerase (Gibco-Invitrogen), and 0.4μM of each primer. Cycling for this polymorphism was as follows: 94°C for 2 min, 35 cycles of 94°C for 30 s, 60°C for 30 s, 72°C for 40 s, and a final extension at 72°C for 7 min. PCR products were digested with the *Bme*1580I restriction enzyme overnight and then separated by gel electrophoresis.

Statistical analysis

All statistical analyses were conducted using SAS 9.1 [21]. Baseline characteristics of cases and controls were calculated and compared using analysis of covariance for continuous variables and χ^2 tests for categorical variables. Each polymorphism was tested to ensure that it fit the Hardy–Weinberg equilibrium using χ^2 tests.

For investigating main effects, multiple logistic regression models were used to test for associations between genetic polymorphisms and colorectal adenoma while controlling for potential confounders. The odds ratio (OR) and 95% confidence interval (CI) for the association between each genotype and disease were calculated using the more common homozygous allele among controls as the reference group. In addition to examining each genotype separately, because of the low prevalence of the homozygous variant polymorphism, to increase statistical power, participants who were homozygous or heterozygous for the variant allele were combined into one group and compared to those in the common homozygote referent group. Several risk factors were examined as possible confounders of the polymorphism and colorectal adenoma association. Among these were age, sex, race, body mass index [BMI=weight (kg)/height $(m)^2$], family history of colorectal cancer in first-degree relatives, smoking, alcohol consumption, NSAID use, total physical activity, intake of dietary fat intake, polyunsaturated fat, energy, calcium, vitamin D, meat, fruit and vegetables, and various antioxidant micronutrients.

Statistical testing for multiplicative interactions between genotypes and exposures was performed by fitting an interaction term for each genotype–exposure combination in the final model. Multiplicative joint effects of genotypes and exposures also were calculated by including three-level dummy variables in the models [22]. The dummy variables were constructed using the most common homozygous genotype and the lowest level of exposure as the referent group. In these analyses, dietary fat and fatty acids intakes were dichotomized into low or high based on the sex-specific medians from the distributions of intakes among controls. Polytomous logistic regression models were used to examine whether associations of these polymorphisms with adenoma risk differed by adenoma characteristics (e.g., size, histological type) [23].

Results

Selected characteristics of cases and controls are shown in Table 1. On average, cases were older, more likely to be male, smokers, or drinkers. Controls were more likely to have a first-degree relative with colorectal cancer. Other factors were fairly well balanced between cases and controls.

Adjusted associations between genotypes and colorectal adenomas are shown in Table 2. All polymorphisms were in Hardy–Weinberg equilibrium (all $P>0.05$) in cases and controls. Overall, there was no statistically significant association between any of these COX gene polymorphisms and colorectal adenoma. For the COX2 9850 A>G polymorphism, the variant allele was rare (AG genotype, 4.2% among cases and 4.3% among controls; no GG genotype in either cases or controls), so no further analyses were performed related to this polymorphism. Associations of the other three polymorphisms and risk of colorectal adenomas according to adenoma characteristics also were examined. There were no statistically significant associations of genotypes with different adenoma subgroups (data not shown).

Adjusted associations of COX2 (−765 G>C, 8473 T>C) genotypes with risk of colorectal adenomas according to several inflammation- and AA-pathway-related factors are shown in Tables 3 and 4, respectively. There were no statistically significant interactions of COX2

−765 G>C genotypes with these factors (all $P_{interaction}$≥0.46). However, there was a significant interaction between COX2 8473 T>C genotypes and NSAID use ($P_{interaction}$=0.03). Having the 8473 C variant allele and regularly taking NSAIDs was associated with a 65% reduction in risk of colorectal adenoma (P=0.008). The association between high level of polyunsaturated fat intake and adenoma tended to be stronger among individuals with the TT wild genotype (OR, 1.83) than those with the variant C allele (OR, 1.01), but the P value for an interaction was not significant ($P_{interaction}$=0.15). We also examined the joint effect of the COX1 −842 A>G genotypes and these factors with risk of colorectal adenoma, but no significant interactions were found (data not shown).

Discussion

Overall, we found no statistically significant association between any of the four SNPs in the COX1 and COX2 genes and colorectal adenoma. However, our results suggest that there was an interaction between the COX2 8473 T>C polymorphism and NSAID use and risk of colorectal adenoma such that persons with at least one C allele who also regularly took an NSAID were at markedly reduced risk.

Several studies have investigated the COX2 −765 G>C polymorphism with risk of colorectal cancer or adenoma, but their results have been inconsistent. One study found no association between this polymorphism and colorectal cancer [24], while others reported either a suggestion that the C variant allele (GC or CC genotype) was associated with a modest, nonsignificant increase in risk of colon cancer in a Chinese population with high dietary intakes of *n*–6 polyunsaturated fatty acids [25] or that the GC genotype was associated with a significant increased risk of colorectal cancer [26]. However, one of two adenoma studies reported a suggestion of decreased risk with the GC genotype [27], and the other observed a statistically significant 74% lower risk in those with the CC genotype who did not take aspirin or other NSAIDs [28]. In our study, we found no significant association between the C variant allele (GC or CC genotype), alone or in interaction with NSAID use or intakes of dietary fat or specific fatty acids, and colorectal adenoma. Because of the low frequency of the variant C allele, studies that examined this polymorphism were limited by having small sample sizes in certain subgroups, which may partly explain the reported inconsistencies. Larger studies will be needed to establish the association between COX2 −765 G>C polymorphism and colorectal neoplasms.

Though results from studies that investigated the COX2 8473 T>C polymorphism are inconsistent, most found the C variant allele to be associated with reduced risk for lung, breast, and prostate cancers [29-33]. The results from four studies that reported on an association of this polymorphism with risk of colorectal neoplasms are also inconsistent. Two found no association between the COX2 8473 T>C polymorphism and colorectal cancer [24, 34]. One adenoma study found the TC genotype, but not the CC genotype, relative to the TT genotype, to be associated with a 47% increase in risk of colorectal adenoma [35]. In stratified analyses, high fish intake appeared to be associated with decreased risk of colorectal adenoma among TT genotype carriers. A nested case–control study of advanced colorectal adenoma (defined as villous or tubulovillous, large [≥1.0 cm], or having high-grade dysplasia) found a significant 31% increased risk with the TC genotype and a nonsignificant 40% increased risk with the CC genotype compared to TT genotype [36]. We did not find a significant association of the COX2 8473 T>C polymorphism with risk of colorectal adenoma in the present study; however, we did find a statistically significant interaction between the polymorphism and NSAID use such that individuals with the C variant allele who also took NSAIDs were at significantly lower risk. We know of no similar findings reported for colorectal neoplasms, but one study reported an analogous finding of the 8473 C variant allele being associated with a 30% reduction in risk

of hormone receptor-positive breast cancer among women who regularly took NSAIDs ($P_{interaction}$ =0.02) [29].

An interaction of COX2 8473 T>C genotypes with NSAIDs to affect risk for colorectal or other neoplasms is biologically plausible. The 8473 C variant is located downstream of the stop codon in the AU-rich 3′-UTR region and could affect the binding affinity of regulatory elements and result in decreased mRNA stability and expression [37], leading to reduced cellular COX2 activity and production. Also, since NSAIDs reduce PGE2 synthesis through COX2 inhibition, it is reasonable to speculate that lower COX2 production by this variant allows for more efficient inhibition by NSAIDs, thereby yielding a much stronger protective effect against colorectal adenoma.

The G allele of the COX2 9850 A>G polymorphism, which is located approximately 100 base pairs upstream of the first poly-A signal in the 3′-UTR of COX2, was associated with an increased risk for colorectal cancer in a study in Spain [24] and a suggestion of reduced risk in a study in the Netherlands [34]. We found no significant association in our study. The frequency of the G allele in our and other studies ranged from 4% to 6%; therefore, much larger studies will be needed to adequately address associations involving the COX2 9850 A>G polymorphism. In our study, there was a suggestion that the −842 G variant allele may be associated with reduced risk of colorectal adenoma. The −842 G variant creates a putative AP2 transcription factor binding site, which may act as a repressor element, thus decreasing transcription of COX1, which in turn could lower adenoma risk by decreasing enzyme activity [38]. We further examined a possible interaction between this polymorphism and NSAID use and found that the inverse association with the variant allele tended to be stronger among those who regularly took NSAIDs, but the test for interaction was not statistically significant.

Dietary fat or polyunsaturated fatty acids have been associated with colorectal carcinogenesis [39]. Polyunsaturated fatty acids, particularly *n*–6 polyunsaturated fatty acids, precursors of AA, promote colon carcinogenesis by up-regulating COX2 expression [40]. AA and linoleic acid are the most abundant polyunsaturated fatty acids in the Western diet. We found no associations for dietary fat or intakes of specific fatty acids and no interactions of these factors with COX gene polymorphisms. As with many epidemiological studies, an FFQ was used to assess dietary intake of foods and nutrients in this study. This self-reported measurement method can result in biases [41, 42], and since such errors are nondifferential with respect to outcome, they tend to bias results toward the null.

This study has several strengths. First, cases and controls were accurately defined by full colonoscopic examination, which minimized bias resulting from misclassification of adenoma status. Second, self-administered questionnaire data were collected prior to diagnosis, thus minimizing the probability of recall bias. Finally, detailed information on multiple important risk factors for the disease and adenoma characteristics was collected, allowing us to examine associations of genotypes with adenomas, controlling for other covariates, overall, and by adenoma characteristics. This study also has several limitations. First, although the colonoscopy-based design minimized possible misclassification bias, the study population may have been at higher risk for colorectal neoplasms than the general population. However, this would tend to attenuate observed associations. Second, small sample sizes in certain subgroups (especially for the low prevalence genetic variants) limited our power to detect main effect and interactive associations. Third, colorectal adenomas appear to share the same spectrum of risk factors seen with colon cancer, but not all adenomas progress to cancer.

In summary, this community-based case–control study provides evidence that persons with the COX2 8473 C variant allele who also regularly take an NSAID may be at particularly low risk for colorectal adenoma. Since our study was small and there is no direct laboratory evidence on the function of the COX2 8473 T>C polymorphism, future studies are needed to confirm the intriguing association raised by this study.

Acknowledgments

This study was supported by the Public Health Service grant R01CA-51932 from the National Cancer Institute, the National Center for Research Resources grant RR017698 from the National Institutes of Health, Department of Health and Human Services, the American Cancer Society Research Scholar grant RSG-06-122-01-CNE, and a Georgia Cancer Coalition Distinguished Scholar award (to R. Bostick), and National Cancer Institute, Center for Research and Cancer Health Disparities grant 1 U01 CA114601-01 (to JR Hebert).

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Table 1

Selected characteristics of cases and controls, Markers of Adenomatous Polyps (MAP) Study

Variable	Cases (*n*=162)	Controls (*n*=211)	*P* value[a]
Demographic and major risk factors			
Age (years)	58.5 (8.6)	56.0 (9.9)	0.02
Male (%)	58.6	34.6	<0.0001
White race (%)	87.7	90.0	0.48
College education (%)	18.5	24.2	0.31
Family history of colorectal cancer (%)	19.9	36.5	0.0005
Currently smoke cigarettes (%)	30.3	19.0	0.01
Currently drink alcohol (%)	54.4	43.1	0.02
Aspirin or NSAID use (%)[b]	47.5	54.0	0.21
Total physical activity (METs/day)[c]	50.0 (15.7)	51.3 (13.8)	0.25
Dietary intakes			
Total energy (kcal/day)	2,023 (760)	1,968 (720)	0.88
Total fat (g/day)	70.7 (33.5)	65.5 (29.8)	0.49
Total polyunsaturated fat (g/day)	13.7 (5.9)	13.0 (6.6)	0.77
Total linoleic acids (g/day)	11.9 (5.3)	11.2 (6.0)	0.26
Total red meat (servings/week)	4.6 (4.4)	4.0 (3.3)	0.16
Total fruits and vegetables (servings/day)	6.4 (3.5)	6.4 (3.8)	0.95
Body measurement			
Body mass index (kg/m^2)			
$\bar{X}$ ± SD	26.4 (5.9)	26.6 (5.3)	0.98
<25 (%)	35.8	38.9	0.83
25 to <30	37.7	36.0	
≥30	26.5	25.1	

Continuous variables presented as mean (SD) and categorical variables as proportions (percent)

[a] For categorical variables, based on χ^2 test, and for continuous variables, based on analysis of covariance (ANCOVA) for age- and sex-adjusted mean differences (exception: age variable adjusted only for sex)

[b] Nonsteroidal anti-inflammatory drug

[c] Metabolic equivalents=the equivalent of a person's energy expenditure at rest

Table 2

Frequencies of *COX* genotypes and associations with risk for incident, sporadic colorectal adenomas, MAP Study

Genotype	Cases		Controls		OR (95% CI)[a]
	n	%	*n*	%	
COX2 −765 G>C					
GG	108	66.7	127	60.2	1.00
GC	45	27.7	72	34.1	0.82 (0.49–1.36)
CC	9	5.6	12	5.7	0.60 (0.21–1.71)
GC+CC	54	33.3	84	39.8	0.78 (0.48–1.26)
					P_{trend}=0.25
COX2 8473 T>C					
TT	64	39.5	69	32.7	1.00
TC	70	43.2	109	51.7	0.78 (0.47–1.30)
CC	28	17.3	33	15.6	0.77 (0.39–1.52)
TC+CC	98	60.5	142	67.3	0.78 (0.49–1.26)
					P_{trend}=0.37
COX2 9850 A>G					
AA	155	95.7	202	95.7	1.00
AG	7	4.3	9	4.3	0.95 (0.32–2.85)
GG	0	0.0	0	0.0	N/A[b]
COX1 −842 A>G					
AA	142	88.2	179	85.3	1.00
AG	18	11.2	28	13.3	0.66 (0.33–1.33)
GG	1	0.6	3	1.4	0.37 (0.03–4.62)
AG+GG	19	11.8	31	14.7	0.64 (0.32–1.26)
					P_{trend}=0.17

[a]OR (odds ratio) and 95% CI (confidence interval) adjusted for age, race, sex, body mass index, smoking, alcohol intake, total energy intake, physical activity, and family history of colorectal cancer in a first-degree relative.

[b]Not done, only one allele was present in each group

Int J Colorectal Dis. Author manuscript; available in PMC 2012 October 01.

Table 3

Multivariable-adjusted joint effects of *COX2* (−765 G>C) genotypes, various risk factors for colorectal neoplasms, and risk for incident, sporadic colorectal adenomas, MAP Study

Risk Factors	Genotypes			
	GG		GC+CC	
	No. of cases/controls	OR (95% CI)[a]	No. of cases/controls	OR (95% CI)[a]
NSAID[b] use				
No	89/90	1.00	45/60	0.71 (0.41–1.22)
Yes	19/37	0.46 (0.24–0.92)	9/24	0.51 (0.21–1.22)
				$P_{interaction}$ =0.46
Total fat intake[c]				
Low	58/66	1.00	29/39	0.80 (0.41–1.54)
High	50/61	0.77 (0.39–1.55)	25/45	0.61 (0.29–1.31)
				$P_{interaction}$ =0.99
Total polyunsaturated fat[c]				
Low	49/65	1.00	28/43	0.84 (0.43-1.64)
High	59/62	1.26 (0.65–2.44)	26/41	0.91 (0.43–1.95)
				$P_{interaction}$ =0.76
Total linoleic acids[c]				
Low	52/64	1.00	28/45	0.77 (0.40–1.49)
High	56/63	1.06 (0.55–2.05)	26/39	0.84 (0.40–1.80)
				$P_{interaction}$ =0.59

[a] OR (odds ratio) and 95% CI (confidence interval) adjusted for age, race, sex, body mass index, smoking, alcohol intake, total energy intake, physical activity, and family history of colorectal cancer in a first-degree relative. Models for dietary fat and fatty acids intake are also adjusted for NSAID use.

[b] Nonsteroidal anti-inflammatory drug

[c] All dietary fat intakes classified into low and high based on sex-specific median values in controls

Table 4

Multivariable-adjusted joint effects of *COX2* (8473 T>C) genotypes, various risk factors for colorectal neoplasms, and risk for incident, sporadic colorectal adenomas, MAP study

Risk Factors	Genotypes			
	TT		TC+CC	
	No. of cases/controls	OR (95% CI)[a]	No. of cases/controls	OR (95% CI)[a]
NSAID[b] use				
No	50/54	1.00	84/96	1.04 (0.61–1.78)
Yes	14/15	1.19 (0.49–2.89)	14/46	0.35 (0.16–0.75)
				$P_{interaction}$ =0.03
Total fat intake[c]				
Low	34/39	1.00	53/66	0.92 (0.49–1.75)
High	30/30	0.96 (0.41–2.29)	45/76	0.64 (0.30–1.34)
				$P_{interaction}$ =0.49
Total polyunsaturated fat[c]				
Low	28/40	1.00	49/68	1.06 (0.55–2.06)
High	36/29	1.83 (0.80–4.18)	49/74	1.01 (0.49–2.08)
				$P_{interaction}$ =0.15
Total linoleic acids[c]				
Low	31/39	1.00	49/70	0.90 (0.47–1.73)
High	33/30	1.34 (0.58–3.06)	49/72	0.89 (0.43–1.83)
				$P_{interaction}$ =0.20

[a] OR (odds ratio) and 95% CI (confidence interval) adjusted for age, race, sex, body mass index, smoking, alcohol intake, total energy intake, physical activity, and family history of colorectal cancer in a first-degree relative. Models for dietary fat and fatty acids intake are also adjusted for NSAID use.

[b] Nonsteroidal anti-inflammatory drug

[c] All dietary fat intakes classified into low and high based on sex-specific median values in controls