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Genetic variants in COX-2, non-steroidal anti-inflammatory drugs, and breast cancer risk: the Western New York Exposures and Breast Cancer (WEB) Study

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Abstract

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Chronic inflammation has been consistently associated with cancers of several sites, including the breast, and inhibition of inflammation through the use of non-steroidal anti-inflammatory drugs (NSAIDs) has been inversely associated with risk. As NSAIDs bind with cyclooxygenase-2 (COX-2), genetic variation in *COX-2* may influence breast cancer risk by affecting inflammatory response and response to NSAID use. We identified eight single nucleotide polymorphisms (SNPs) for *COX-2* and examined their association with risk of breast cancer in a population-based case-control study in Western New York. Cases had incident, first primary, histologically confirmed breast cancer ($n = 1077$). Controls ($n = 1910$) were randomly selected from NY Department of Motor Vehicles records (<65) or Medicare rolls (≥ 65). Participants were queried on adult lifetime use of aspirin and recent use of ibuprofen. Unconditional logistic regression was used to estimate odds ratios (OR) and 95% confidence intervals (95% CI). One SNP, rs2745559, was associated with an increased risk of breast cancer (OR 1.23, 95% CI 1.03–1.46). Associations with other variants were not evident. Significant interaction (P interaction = 0.04) between recent aspirin use and rs4648261 was also observed. Variation in *COX-2* was modestly associated with breast cancer risk, indicating that *COX-2* may play a role in breast carcinogenesis. Better understanding of the role of *COX-2* genetic variation and interaction with NSAID use in breast carcinogenesis has potential to inform prevention strategies.

Keywords

Breast cancer; Cyclooxygenase-2; Non-steroidal anti-inflammatory drugs; Prostaglandinendoperoxide synthase 2; Single nucleotide polymorphism

Introduction

Chronic inflammation has been consistently associated with cancers of the liver and gastrointestinal tract; there is evidence that it is also associated with cancers of other sites including the breast [1]. Use of non-steroidal anti-inflammatory drugs (NSAIDs) has been associated with modest reductions in breast cancer risk in some, but not all studies [2]. Although reductions in risk have been reported for prospective and case-control studies, stronger associations were generally reported for case-control studies [2]. We previously reported that use of aspirin, but not ibuprofen, was inversely associated with breast cancer risk [3].

Non-steroidal anti-inflammatory drugs have an anti-inflammatory effect primarily because they bind with the inducible isoform of the cyclooxygenase enzymes, COX-2 (EC 1.14.99.1), thereby blocking the catalysis of arachidonic acid to pro-inflammatory prostaglandins. If the observed reduction in risk associated with NSAID use results from their inhibitory effects on prostaglandin synthesis, genetic variation in *COX-2* may influence inflammation, and thus also be associated with breast cancer risk. Genetic variation in *COX-2* may be important in modifying the association between NSAIDs and breast cancer risk.

The *COX-2* gene is on chromosome 1q25.2–25.3, 8.3 kb in size, and has 10 exons [4]. While expression of *COX-2* is nearly undetectable in normal breast tissue, the gene is over-expressed in 40% of invasive breast tumors [5]. Expression in tumor tissue is positively correlated with expression in adjacent normal and in situ tissue, suggesting that altered expression of the protein may be an early event in carcinogenesis [6–8]. Only some of the single nucleotide polymorphisms (SNPs) identified for *COX-2* have been studied with regard to breast cancer risk and findings have been inconsistent [9–18]. There is some evidence that polymorphisms in the 5' and 3' untranslated regions (UTR) may be positively

associated with breast cancer risk, however, associations did not reach statistical significance in most studies [9–13].

In a few studies, the interaction of *COX-2* genotypes and NSAID use in association with breast cancer has been examined, although few have studied the same SNP [11,14–16]. Only one study reported significant interactions [11]. However, some important dimensions have not been examined. In only one of the existing studies have the different types of NSAIDs (e.g., aspirin, ibuprofen) been investigated individually [15]. No study included assessment of use of these medications other than ever use (defined as 1/week for ≥ 6 months) [15] or recent use (1–8/month for ≥ 3 months–1 year) [11,14,16]. As aspirin and non-aspirin NSAIDs are chemically different and bind differently to the *COX-2* enzyme [19], they may differ in their interaction with *COX-2* and risk. Long-term use may have a greater impact on risk, particularly given that there is evidence that the critical period is early in carcinogenesis.

We report here on our investigation of the association of eight *COX-2* variants and breast cancer risk and the interaction of *COX-2* SNPs with recent and lifetime use of selected NSAIDs.

Materials and methods

The Western New York Exposures and Breast Cancer (WEB) Study is a population-based case–control study of women living in Erie or Niagara counties of western New York State [20–23]. Briefly, participants were English-speaking women, aged 35–79 years, with no prior malignancy other than non-melanoma skin cancer. Cases ($n = 1170$) were women with incident, first primary, histologically confirmed breast cancer, diagnosed in the period 1996–2001. Controls ($n = 2115$) were randomly selected from Department of Motor Vehicles records (< 65 years) or Medicare rolls (≥ 65 years) and frequency matched to cases on age and race. All participants provided informed consent; the Institutional Review Boards of the University at Buffalo and participating hospitals approved study protocols.

Data collection

Between 1996 and 2001, information on demographics, medical and reproductive history, anthropometrics, and other breast cancer risk factors was collected. Participants were asked to report the average monthly frequency of use of aspirin and ibuprofen during the period 12–24 months prior to interview. In addition, the frequency of aspirin use was queried for each decade of a participant's adult life, beginning at age 21. We did not collect data on lifetime use of ibuprofen because it was not available over the counter until 1984 [24].

SNP selection

We selected tag SNPs for *COX-2* using data from the International HapMap Project, phase II, NCBI Build 36.3 (<http://www.ncbi.nlm.nih.gov>), Caucasian population (CEU) [25]. Using the Tagger online program [26], we identified SNPs in the region including 5 kb up- and downstream of *COX-2* with minor allele frequencies (MAF) $\geq 5\%$ and pairwise r^2 with untyped SNPs ≥ 0.8 . We forced the inclusion of three SNPs; two, rs20417 and rs689466, which had been previously associated with inflammation [27,28], and another, rs5275, the most common *COX-2* polymorphism in Caucasians. We selected the following eight tag SNPs representing an additional nine SNPs: rs20417, rs274557, rs2206593, rs5275, rs5277, rs689466, rs4648261, rs12042763. Because of technical difficulties, we were unable to genotype rs20417 and rs274557, and we replaced them with SNPs for which they tagged ($r^2 = 1$), rs2143416 and rs2745559, respectively.

Genotyping

Blood or saliva samples were obtained from 94% ($n = 1099$) of cases and 92% ($n = 1945$) of controls; DNA was isolated from these. Samples were analyzed by real-time polymerase chain reaction (RT-PCR) Taqman allelic discrimination using pre-designed assays (Applied Biosystems, Foster City, CA, USA). Genotyping was performed for cases and controls together; laboratory technicians were blinded to case-control status. Blind duplicate samples were included to assess laboratory reliability. Genotyping call rates were $\geq 96.5\%$ (median = 97.4%) and concordance of blind duplicates was $\geq 96.2\%$ (median = 98.7%). Due to the small number of African-American participants in this study, we restricted our study sample to Caucasians, leaving 1,077 cases and 1,910 controls available for study.

Statistical analysis

For recent use of aspirin or ibuprofen, participants were classified as users or non-users. Total adult aspirin usage was estimated as the sum of average annual aspirin frequency for each decade of life. From these data, participants were classified based on their average monthly aspirin use throughout their adult life: lifetime aspirin non-users (0 days/month), irregular users (≤ 10 days/month), and regular users (> 10 days/month).

We used unconditional logistic regression models to estimate odds ratios (OR) and 95% confidence intervals (95% CI) for genetic models and to assess interaction with recent NSAID and lifetime aspirin use. We considered established risk factors for breast cancer (age at first birth, parity, body mass index [BMI, kg/m²], age at menarche and/or menopause, postmenopausal hormone use, family history of breast cancer, history of benign breast disease) as potential confounders in multivariable modeling. We forced in age and years of education into models and excluded from final models other variables that did not alter point estimates by $\geq 10\%$. Final models were adjusted for age (years), years of education, menopausal status (pre-/postmenopausal), parity (number of full-term births), hormone replacement therapy (ever/never), and history of benign breast disease (yes/no). In all analyses, the common genotype was the referent. We used the Bonferroni correction to adjust P values for multiple comparisons.

Interaction of recent NSAID use and *COX-2* genotypes was assessed by the creation of joint-classification variables with aspirin or ibuprofen non-users carrying the higher risk allele (as determined empirically from the SNP main effects analyses) as the referent. Observed joint-effects were compared to expected OR under multiplicative models. Differences of $\geq 10\%$ were considered possible interaction. Similarly, we assessed interaction of *COX-2* genotypes and adult lifetime aspirin. Interaction models for recent NSAID use were also adjusted for use of other NSAIDs (e.g., analysis of interaction with aspirin was adjusted for ibuprofen use). P values for interaction (P interaction) were calculated by including a multiplicative term in final multivariate models.

UNPHASED software was used to estimate haplotype frequencies and risk estimates [29]. Rare haplotypes ($< 2\%$ of cases and controls) were not analyzed due to small sample size. We tested for Hardy-Weinberg equilibrium (HWE) among controls using a χ^2 test.

Results

Minor allele frequencies for each SNP are given in Table 1. Genotype and minor allele frequencies among controls were similar to that of the CEU population in HapMap [25]. One SNP, rs5277, was out of HWE ($P = 0.02$) and was excluded from further analyses.

Associations of *COX-2* genotypes with breast cancer risk are given in Table 2. We observed an increase in risk of borderline significance associated with carriers of 1 or 2 rs2745559

minor alleles (CA + AA) compared to the major alleles (OR 1.23, 95% CI 1.03–1.46). The per-allele OR was 1.18, 95% CI 1.02–1.37 (data not shown). However, the association was no longer statistically significant after adjustment for multiple comparisons. There were suggestive inverse associations with the rs2143416-CC genotype (OR 0.79, 95% CI 0.48–1.31), the rs12042763-TT genotype (OR 0.78, 95% CI 0.56–1.10), and the rs5275-GG genotype (OR 0.81, 95% CI 0.62–1.06), compared to the common genotype. Variants of rs689466, rs4648261, and rs2206593 were not associated with breast cancer risk.

Haplotype results are given in Table 3. Haplotype alleles were designated in the table as common or variant (1 or 2, respectively) in the order of their position on the gene from 5' to 3' (i.e., rs2143416, rs2745559, rs12042763, rs689466, rs4648261, rs5275, rs2206593). Comparisons were made with the most prevalent haplotype, h1121111, as the referent. Two haplotypes that include rs2745559 rare alleles were associated with increases in risk, however, confidence intervals included the null (OR_{h1211111} 1.21, 95% CI 0.98–1.50; OR_{h1211112} 1.18, 95% CI 0.92–1.52). All other haplotypes were not associated with breast cancer.

We previously reported that recent use of aspirin (OR 0.81, 95% CI 0.70–0.94) but not ibuprofen (OR 1.13, 95% CI 0.97–1.32) was associated with breast cancer risk [3]. Analyses of *COX-2*-NSAID interactions are presented in Table 4. We observed multiplicative interaction of rs4648261 with recent aspirin use (P interaction = 0.04). Compared to non-users with the common (CC) genotype, aspirin users who were carriers of a T-allele had a reduced risk of breast cancer (OR 0.54, 95% CI 0.29–1.01). In a separate analysis, comparing aspirin users to non-users stratified by rs4648261 genotype, risk of breast cancer associated with aspirin use was greatly reduced among carriers of the variant allele (OR 0.35, 95% CI 0.16–0.77) but among women with the common genotype, the decrease did not reach statistical significance (OR 0.86, 95% CI 0.73, 1.01). Different from our findings with recent aspirin use, we did not observe interaction between adult lifetime aspirin use and rs4648261. There was a suggestion of interaction of recent and adult lifetime aspirin use and rs2745559 and rs2206593, however, findings did not reach statistical significance. No interactions were observed between recent ibuprofen use and *COX-2* SNPs (data not shown).

Discussion

In this population-based case-control study, there was evidence that *COX-2* genetic variation may be modestly associated with breast cancer risk. As *COX-2* plays an important role in inflammation and wound healing, it may be that an evolutionary disadvantage would be conferred to those with functional polymorphisms in this gene such that there are not large differences in function associated with genetic variation or that other factors are more important in terms of control of protein expression. We also found that there may be some interaction of at least one genetic variant with aspirin use.

Our findings were generally consistent with previous studies and extend those findings by assessing interaction with recent and adult lifetime aspirin use, and recent ibuprofen use. We observed a statistically significant increased risk of breast cancer with rs2745559. Consistent with our finding, Gallicchio et al. [11] reported an increased risk with the variant of rs2745557, a SNP in linkage disequilibrium with rs2745559 ($r^2 = 1$; OR 2.07, 95% CI 0.78–5.48). However, in another study, the Breast and Prostate Cancer Cohort Consortium (BPC3), a large pooled analysis of prospective studies including 6,292 breast cancer cases and 8,135 matched controls, the authors reported no association of breast cancer risk with rs2745557 [18].

We found a modest inverse association with breast cancer and the *COX-2* SNP, rs5275. This finding is consistent with that reported in a recent meta-analysis of this SNP in association with breast cancer risk (OR 0.84, 95% CI 0.70–1.00) [30]. Results of individual studies of this SNP with breast cancer have not been statistically significant [9–13,15,16]. Only in one study [13], a hospital-based case–control study in Austria was the association of rs5275 and breast cancer risk statistically significant (OR 2.03, 95% CI 1.30–2.25); however, in that study, genotype frequencies were not in HWE among controls.

The *COX-2* SNP, rs2143416, was inversely associated with risk in our study. In contrast, Gallicchio et al. [11] recently reported a non-significant increase in breast cancer risk with the homozygote variant of rs2143416 (OR 2.19, 0.74–6.46) in a small nested case–control study of women with benign breast disease. Others studied that a SNP in linkage disequilibrium with rs2143416, rs20417, reported no association with breast cancer [10,12,15,18].

We observed no association of rs2206593 and breast cancer risk, similar to BPC3 [18]. Two studies reported increases in risk (OR $\geq$ 2.3), but due to very small MAF in Caucasians, confidence intervals were wide [9,11]. Consistent with the BPC3 [18], we observed no association between rs689466 and breast cancer. In contrast, Gao et al. [12] reported an increase in breast cancer risk in a hospital-based case–control study conducted in Nanjing, China (OR 1.31, 95% CI 0.95–1.81). Only the BPC3 has reported on rs12042763 and rs4648261 in association with breast cancer risk [18]. Whereas we observed a suggestion of reduced risk among carriers of the homozygous variant of rs12042763, and no association among carriers of the rs4648261 variant allele, the authors of the BPC3 found no association with either SNPs [18].

Two haplotypes that included rs2745559 minor alleles were associated with increased risk of breast cancer. Although others have investigated haplotypes with risk of prostate or breast cancer, none have included this SNP [9,31].

Of studies examining *COX-2* polymorphisms in association with breast cancer, few have examined interactions with NSAID use [11,14–16]. Authors of only one previous study have included interaction analyses of aspirin and other NSAIDs separately [15]. We observed a significant interaction of recent aspirin use with rs4648261. To our knowledge, interaction with this SNP has not been previously reported.

Findings for rs2745559 and rs2206593 were suggestive of interaction with both recent and adult lifetime aspirin use, although results were not statistically significant. Gallicchio et al. [11] observed significant interaction between NSAID use and rs2745557 (a SNP in LD with rs2745559) in relation to breast cancer risk. Women carrying the homozygous variant of rs2745557 who were non-users of NSAIDs were at significantly increased risk of breast cancer compared to NSAID users carrying either the common genotype or heterozygotes [11].

In sum, our findings of increased risk of breast cancer among carriers of the rs2745559 variant, and inverse associations with several other *COX-2* SNPs, are largely consistent with published literature, including the recent findings of the BPC3. Although few have reported on interaction of *COX-2* variation with individual NSAIDs, our findings of generally small interactions with aspirin use are consistent with those of others, although different assessments and considerations in statistical analyses limits comparability of studies. While others have looked at all NSAIDs together, we were able to look at aspirin separate from ibuprofen. We did not see any interaction with ibuprofen. It may be that differences between mechanisms of action for aspirin and ibuprofen explain differences observed for interaction models.

Several mechanisms by which polymorphisms may alter *COX-2* function have been proposed, although little is known about their relation to phenotype. SNPs in the gene body may alter the *COX-2* enzyme conformation, and therefore alter enzymatic function or the affinity of NSAIDs to bind to the enzyme [32]. In murine models, polymorphisms in the 5' and 3' untranslated regions (UTR) are thought to alter gene promoter activity or alter mRNA stability, respectively [27,33,34]. The suggestion of a reduction in risk that we observed associated with rs2143416 and rs12042763, both in the 5'-UTR, may correspond to decreased *COX-2* expression. The increase in risk we observed with the variant of rs2745559 (5'-UTR) may be due to altered gene expression or due to some other mechanism—including one involving the SNP for which it tags, rs2745557, located on intron 1. Currently, the functional impact of neither SNP is known. Further, chance cannot be ruled out as an explanation for our findings in that the observation was not statistically significant after adjustment for multiple comparisons. It could be that the polymorphic variants of rs5275 and rs2206593 both in the 3'-UTR may affect the transcript's stability, therefore altering inflammation levels. The functional impact of rs4648261, located on intron 2, for which we observed significant interaction with aspirin use, is also not known. It may be that rs4648261 is in LD with a SNP in a translated region of the gene, or that intron 2 includes certain regulatory regions, or that the finding was a result of chance.

This study has several limitations. As very little is known about the function of these *COX-2* variants, at this time we can only hypothesize regarding their biologic role. Since we had no a priori hypotheses regarding function, for the analysis of interaction, we determined the referent group based on the findings from the analysis of main genetic effects. That is, the referent was determined as the group who were non-users and with apparently higher OR for the main effects. With more information regarding function, we might use a different group as referent. Further, our understanding of other factors influencing gene expression is limited. It may be that these genetic variations act in concert with other factors such as epigenetic changes, and that to understand this process, we need to know more about those dimensions as well.

One concern in a study of this kind is power. For most SNPs, we had 80% power to detect ORs of ≤ 0.79 or ≥ 1.25 using a dominant model, and interaction ORs of ≥ 0.62 . We cannot rule out associations among very rare SNPs that we did not have power to detect in our study. Inherent to genetic analyses such as these, we do not know if the SNPs studied are the true risk alleles, or rather linked to other polymorphisms of biologic significance. One SNP, rs5277, was out of HWE in our study population and we were therefore unable to study that haplotype block and cancer risk. While population stratification is a concern in genetic association studies, it is not likely to have had a large impact on our findings as our analysis was restricted to Caucasians [35].

It is possible that participants' recollection of NSAID use was dependent upon case-control status. Given the design of our study, we believe it is unlikely that the interviewers or participants were aware of NSAID use as a study hypothesis and probing of participants or over-/under-reporting of exposures is not likely to have been differential. Further, our results were similar to those of prospective studies where recall bias is not an issue [2]. In addition to possible recall bias in the report of NSAID use, there was likely non-differential error in the recall of participants' lifetime aspirin use and recent NSAID use. Non-differential measurement error would result in widened confidence limits and attenuated point estimates [36]. Although it is possible that participants could self-select based upon their use of NSAIDs, selection bias would not explain the observed gene-environment interaction [37].

This study has several strengths. It is among the largest to investigate associations of *COX-2* variation with breast cancer risk. Additionally, it is among the first to characterize this

variation across such a large area of the gene. Previous investigations of *COX-2* and cancer risk have captured variation across the gene body alone [9] or only a small (≤ 2 kb) area surrounding the gene [31]. As variation in the flanking region of *COX-2* is hypothesized to contribute to gene function [27,33,34], investigations including these regions are important. Only the BPC3 has reported on a larger area surrounding the gene [18]. Another strength is that we were able to investigate gene–environment interaction, taking into account possible differences between aspirin and ibuprofen. In addition, this study is the first to explore gene–environment interaction examining more than recent use; we had reports of adult lifetime aspirin use.

In this study, we found a modest increase in risk with the variant of rs2745559. There was some indication that aspirin use among carriers of the rs4648261 variant allele may have additional chemopreventive benefit with regard to breast cancer. Epidemiologic studies with large sample sizes and careful examination of exposure to NSAIDs are needed to further assess associations of variation in *COX-2* and other genes that impact inflammation with breast cancer risk and to investigate possible interaction with aspirin and NSAID use. Further study of the function of *COX-2* polymorphisms is warranted to better understand what may be an important mechanism for carcinogenesis in general and for breast cancer in particular. Better understanding of the role of inflammation in breast carcinogenesis has potential to inform prevention strategies.

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COX-2 SNPs on chromosome 1 (Caucasians only)

Table 1

SNP	Position	Location	Allele, Referent → Minor	MAF cases	MAF controls
rs2143416	184920388	5'UTR	A → C	0.16	0.17
rs2745559	184918625	5'UTR	C → A	0.20	0.17
rs12042763	184918499	5'UTR	G → T	0.25	0.26
rs689466	184917374	5'UTR	T → C	0.18	0.17
rs4648261	184915627	Intron 2	C → T	0.02	0.02
rs5277 ^a	184914820	Exon 3	G → C	0.15	0.16
rs5275	184909681	3'UTR	A → G	0.34	0.35
rs2206593	184909052	3'UTR	G → A	0.07	0.06

MAF minor allele frequency

^aRs5277 was out of Hardy-Weinberg equilibrium

Table 2Association of *COX-2* genotypes with breast cancer risk among WEB study participants (Caucasians only)

SNP	Genotype	Cases <i>n</i> = 1077, <i>n</i> (%)	Controls <i>n</i> = 1910, <i>n</i> (%)	OR (95% CI)	Adjusted OR (95% CI) ^a
rs2143416	AA	702 (71.63)	1206 (69.27)	1.00 referent	1.00 referent
	AC	250 (25.51)	482 (27.69)	0.89 (0.75–1.07)	0.88 (0.73–1.06)
	CC	28 (2.86)	53 (3.04)	0.91 (0.57–1.45)	0.79 (0.48–1.31)
rs2745559	AC + CC	278 (28.37)	535 (30.73)	0.89 (0.75–1.06)	0.87 (0.73–1.04)
	CC	622 (64.59)	1197 (69.07)	1.00 referent	1.00 referent
	CA	305 (31.67)	479 (27.64)	1.23 (1.03–1.46)	1.23 (1.03–1.47)
	AA	36 (3.74)	57 (3.29)	1.22 (0.79–1.87)	1.21 (0.77–1.89)
	CA + AA	341 (35.41)	536 (30.93)	1.22 (1.04–1.45)	1.23 (1.03–1.46)
	GG	530 (55.27)	947 (54.58)	1.00 referent	1.00 referent
rs12042763	GT	372 (38.79)	657 (37.87)	1.01 (0.86–1.19)	1.03 (0.86–1.22)
	TT	57 (5.94)	131 (7.55)	0.78 (0.56–1.08)	0.78 (0.56–1.10)
	GT + TT	429 (44.73)	788 (45.42)	0.97 (0.83–1.14)	0.99 (0.84–1.16)
rs689466	TT	660 (68.39)	1199 (69.55)	1.00 referent	1.00 referent
	TC	271 (28.08)	471 (27.32)	1.05 (0.88–1.25)	1.05 (0.87–1.26)
	CC	34 (3.52)	54 (3.13)	1.14 (0.74–1.78)	1.07 (0.67–1.68)
rs4648261	TC + CC	305 (31.61)	525 (30.45)	1.06 (0.89–1.25)	1.05 (0.88–1.26)
	CC	943 (95.16)	1671 (95.21)	1.00 referent	1.00 referent
	CT	48 (4.84)	84 (4.79)	1.01 (0.70–1.46)	1.04 (0.72–1.52)
rs5275	AA	432 (43.77)	732 (42.07)	1.00 referent	1.00 referent
	AG	447 (45.29)	782 (44.94)	0.97 (0.82–1.14)	0.97 (0.82–1.15)
	GG	108 (10.94)	226 (12.99)	0.81 (0.63–1.05)	0.81 (0.62–1.06)
rs2206593	AG + GG	555 (56.23)	1008 (57.93)	0.93 (0.80–1.09)	0.93 (0.79–1.10)
	GG	831 (85.85)	1529 (87.57)	1.00 referent	1.00 referent
	GA	133 (13.74)	212 (12.14)	1.15 (0.92–1.46)	1.14 (0.90–1.46)
	AA	4 (0.41)	5 (0.29)	1.47 (0.30–5.50)	1.30 (0.34–4.96)
	GA + AA	137 (14.15)	217 (12.43)	1.16 (0.92–1.46)	1.15 (0.90–1.46)

OR odds ratio, CI confidence interval

Adjusted for age, education, menopausal status, parity, postmenopausal hormone therapy, and benign breast disease

Table 3

Association of estimated *COX-2* haplotypes with breast cancer risk among WEB study participants (Caucasians only)

<i>COX-2</i> haplotype	Cases (%)	Controls (%)	Adjusted OR (95% CI) ^a
h1121111	453.60 (24.98)	880.90 (26.47)	1.00 referent
h1112111	319.80 (17.61)	554.50 (16.67)	1.10 (0.92–1.33)
h1111121	286.00 (15.75)	534.30 (16.05)	1.05 (0.87–1.27)
h2111121	268.60 (14.79)	553.90 (16.64)	0.93 (0.77–1.13)
h1211111	221.40 (12.19)	360.00 (10.82)	1.21 (0.98–1.50)
h1211112	133.80 (7.37)	216.80 (6.51)	1.18 (0.92–1.52)
h1111111	57.29 (3.16)	124.60 (3.75)	0.92 (0.65–1.31)
h1111221	45.00 (2.48)	79.43 (2.39)	1.12 (0.75–1.67)

OR odds ratio, CI confidence interval

^a Adjusted for age, education, menopausal status, parity, postmenopausal hormone therapy, and benign breast disease

Table 4

Odds ratios and 95% confidence intervals of the interaction of *COX-2* polymorphisms and recent or lifetime aspirin use in association with breast cancer risk among WEB study participants (Caucasians only)

			<i>P</i> interaction
rs2143416	AA	AC/CC	
Recent aspirin			0.65
Non-users	1.00 referent	0.89 (0.70–1.13)	
Users	0.84 (0.69–1.03)	0.69 (0.53–0.90)	
Lifetime aspirin			0.97
Non-users	1.00 referent	1.06 (0.69–1.61)	
Irregular users (≤10 days/month)	1.05 (0.80–1.37)	0.88 (0.65–1.19)	
Regular users (> 10 days/month)	0.53 (0.32–0.89)	0.76 (0.38–1.54)	
rs2745559	CA/AA	CC	
Recent aspirin			0.46
Non-users	1.00 referent	0.86 (0.68–1.08)	
Users	0.89 (0.67–1.18)	0.67 (0.52–0.85)	
Lifetime aspirin			0.61
Non-users	1.00 referent	0.81 (0.53–1.24)	
Irregular users (≤10 days/month)	0.93 (0.64–1.37)	0.78 (0.54–1.13)	
Regular Users (> 10 days/month)	0.47 (0.23–0.99)	0.53 (0.30–0.93)	
rs12042763	GG	GT/TT	
Recent aspirin			0.91
Non-users	1.00 referent	1.00 (0.80–1.25)	
Users	0.82 (0.65–1.03)	0.80 (0.63–1.02)	
Lifetime aspirin			0.87
Non-users	1.00 referent	0.90 (0.60–1.35)	
Irregular users (≤10 days/month)	0.91 (0.67–1.22)	0.94 (0.69–1.27)	
Regular users (> 10 days/month)	0.58 (0.33–1.03)	0.51 (0.28–0.92)	
rs689466	TT	TC/CC	
Recent aspirin			0.95
Non-users	1.00 referent	1.05 (0.83–1.33)	
Users	0.80 (0.65–0.98)	0.83 (0.63–1.09)	
Lifetime aspirin			0.75
Non-users	1.00 referent	1.07 (0.69–1.64)	
Irregular users (≤10 days/month)	0.95 (0.72–1.24)	1.02 (0.76–1.37)	
Regular users (> 10 days/month)	0.65 (0.40–1.04)	0.44 (0.19–1.03)	
rs4648261	CC	CT	
Recent aspirin			0.04
Non-users	1.00 referent	1.44 (0.89–2.34)	
Users	0.86 (0.72–1.02)	0.54 (0.29–1.01)	
Lifetime aspirin			0.85
Non-users	1.00 referent	0.95 (0.37–2.44)	

			<i>P</i> interaction
Irregular users (≤10 days/month)			0.97 (0.77–1.21) 1.07 (0.67–1.72)
Regular users (> 10 days/month)			0.59 (0.39–0.91) 0.67 (0.17–2.68)
rs5275	AA	AG/GG	
Recent aspirin			0.20
Non-users			1.00 referent 1.02 (0.82–1.28)
Users			0.91 (0.71–1.18) 0.75 (0.59–0.95)
Lifetime aspirin			0.75
Non-users			1.00 referent 0.99 (0.66–1.48)
Irregular users (≤10 days/month)			0.98 (0.69–1.38) 0.94 (0.67–1.32)
Regular users (> 10 days/month)			0.64 (0.33–1.22) 0.55 (0.31–0.96)
rs2206593	GA/AA	GG	
Recent aspirin			0.44
Non-users			1.00 referent 0.96 (0.70–1.32)
Users			0.94 (0.60–1.47) 0.75 (0.54–1.04)
Lifetime aspirin			0.22
Non-users			1.00 referent 0.68 (0.38–1.23)
Irregular users (≤10 days/month)			0.74 (0.41–1.35) 0.67 (0.39–1.17)
Regular users (> 10 days/month)			0.25 (0.06–1.01) 0.45 (0.23–0.88)

Adjusted for age, education, menopausal status, parity, postmenopausal hormone therapy, and benign breast disease