



***PTGS2* gene polymorphism -765G>C is associated with coronary artery disease: a meta-analysis**

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ABSTRACT. Previous studies focusing on the association of *PTGS2* polymorphism -765G>C with coronary artery disease (CAD) have failed to reach the same conclusion. In the present study, we performed a meta-analysis to systematically summarize the possible association between *PTGS2* polymorphism -765G>C and the risk of CAD. We conducted a search of case-control studies on the associations of *PTGS2* with susceptibility to CAD in PubMed, EMBASE, and Chinese National Knowledge Infrastructure databases. Data from eligible studies were extracted for meta-analysis. CAD risk associated with *PTGS2* -765G>C was estimated by pooled odds ratios (ORs) and 95% confidence intervals (95%CI) with the RevMan 5.2 software. Eleven independent case-control studies on *PTGS2* -765G>C were included in our meta-analysis. Our results showed that *PTGS2* -765G>C was associated with a decreased risk of CAD (OR = 0.66, 95%CI = 0.56-0.79; P < 0.001). This meta-analysis suggests that *PTGS2* -765G>C is associated with a decreased risk of CAD.

Key words: *PTGS2*; Coronary artery disease; Meta-analysis

INTRODUCTION

Coronary artery disease (CAD) is a complex multifactorial and polygenic disorder in which multiple environmental and genetic factors are simultaneously involved (Xie et al., 2011). Therefore, the identification of genetic determinants associated with CAD risk has tremendous importance from a public health perspective. Recently, a number of case-control studies have suggested that -765G>C polymorphism in the *PTGS2* gene is associated with the risk of CAD (Cipollone et al., 2004, 2009; Hegener et al., 2006; Lee et al., 2008; Szczeklik et al., 2008; Kohsaka et al., 2008; Huuskonen et al., 2008; Li et al., 2009; Rudock et al., 2009; Sanak et al., 2010; McGettigan et al., 2011; Ol et al., 2011).

Cyclooxygenase (COX) is the key enzyme for the production of prostaglandins (PGs) from free arachidonic acid and two isoforms of COX have been identified. COX-1 is constitutively expressed in most normal tissues and is considered to be a housekeeping enzyme that is responsible for various physiological functions. In contrast, COX-2 is rarely expressed in normal tissues, but is rapidly induced in response to cytokines, growth factors and tumor promoters (Dubois et al., 1998; Smith et al., 2000). COX-2-derived prostaglandins participate in angiogenesis, inhibition of apoptosis, and immune response suppression, which have been associated with cardiovascular disease. COX-2 has been found to be upregulated in atherosclerotic plaques (Kuge et al., 2007), and the polymorphisms of its gene, designated as *PTGS2*, have previously been associated with ischemic heart disease and risk of stroke (Colaizzo et al., 2006; Orbe et al., 2006). However, several studies indicated that there may be no association between *PTGS2* polymorphism and CAD (Hegener et al., 2006; Lee et al., 2008). Therefore, we systematically reviewed the data published to date on the relationship between *PTGS2* polymorphism and CAD risk and quantitatively summarized the available evidence by performing a formal meta-analysis.

MATERIAL AND METHODS

Search strategy, eligibility criteria, and data extraction

A systematic review of original articles, reviews, and meta-analyses analyzing the association between *PTGS2* locus polymorphisms and CAD risk was performed by two independent investigators. The search was carried out in the PubMed, Embase, and Chinese National Knowledge Infrastructure (CNKI) databases with the following search terms: “*PTGS2* or COX-2 or rs20417” AND “CAD” or “Myocardial infarction” or “Coronary artery disease” or “Acute coronary syndrome” or “ACS”. Disagreements were resolved by iteration, discussion, and consensus. To unravel potential systematic biases, a third investigator performed a concordance study by independently reviewing all eligible studies; complete concordance (100%) was reached for all variables assessed.

Publication date and publication language were not restricted in our search. Reference lists were examined manually to further identify potentially relevant studies. The following data were extracted from eligible studies: the first author’s last name, year of publication, country of origin, the numbers of genotyped cases and controls, and genotyping methods. All studies matching the inclusion criteria were retrieved for further examination and data extraction. All investigators had received training in literature search, statistics, and evidence-based medicine.

Statistical analysis

Meta-analysis was performed by using the RevMan 5.2 software provided by the Cochrane Collaboration. We directly used Q-test and I^2 test to examine the heterogeneity between each study. In the heterogeneity test, $P > 0.05$, we selected the Fixed Effect Model to merge OR. To test the publication bias, we used the RevMan 5.2 statistical software to make the funnel plot. The statistical significance of the pooled OR was determined with the Z test, and a $P < 0.05$ was considered to be significant.

RESULTS

Study characteristics

A total of 386 articles were retrieved after first search in the databases above. After our selection, 11 case-control studies fulfilled the inclusion criteria. The qualities of the studies were considered to be acceptable for our meta-analysis. Characteristics of studies included are summarized in Table 1. A total of 11 studies involving 22,584 subjects were ultimately analyzed in our meta-analysis. In these 11 studies, four genotyping methods were employed including TaqMan, Illumina Human550 BeadChips, PCR-RFLP and sequencing. The genotype distribution in the controls was in agreement with Hardy-Weinberg equilibrium (HWE) in all of the studies included.

Table 1. Characteristics of studies included in the meta-analysis.

Author	Year	Country	Genotyping methods	No. (Cases/controls)	Genotype case (%)		Genotype control (%)	
					GG	GC+CC	GG	GC + CC
Cipollone	2004	Italy	N/A	864/864	700	164	441	423
Hegener	2006	USA	Taqman	600/600	474	126	463	137
Kohsaka	2008a	Turkey	Chips	1153/8814	825	329	6391	2423
Kohsaka	2008b	Australia	Taqman	334/3128	160	174	1411	1717
Lee	2008a	USA	Taqman	776/607	546	230	425	182
Lee	2008b	USA	Taqman	224/255	113	111	111	144
McGettigan	2011	Canada	Taqman	460/640	336	124	478	162
Morgan	2007	USA	Sequenom MALDI-TOF	811/650	576	217	447	195
Ol	2011	USA	PCR-RFLP	118/80	48	70	27	53
Xie	2009	China	PCR-RFLP	356/350	234	122	245	105
Xie	2011	China	PCR-RFLP	430/470	364	66	368	102

Meta-analysis

The association between *PTGS2* -765G>C and susceptibility to CAD was analyzed in 11 independent studies. Results of the meta-analysis are shown in Figure 1. The Q-test showed no significant heterogeneity ($P = 0.68$, $I^2 = 0\%$). Therefore, the fixed-effects model was used to analyze the association. In our analysis, significant differences were observed for the comparison of GG vs GC+CC (OR = 0.66, 95%CI = 0.56-0.79; $P < 0.0001$; Figure 1).

Publication bias

The funnel plot and Egger test were performed to assess the publication bias of the literature. Symmetrical funnel plots were obtained in the SNP tested. The Egger test further confirmed the absence of publication bias in this meta-analysis ($P > 0.05$) (Figure 2).

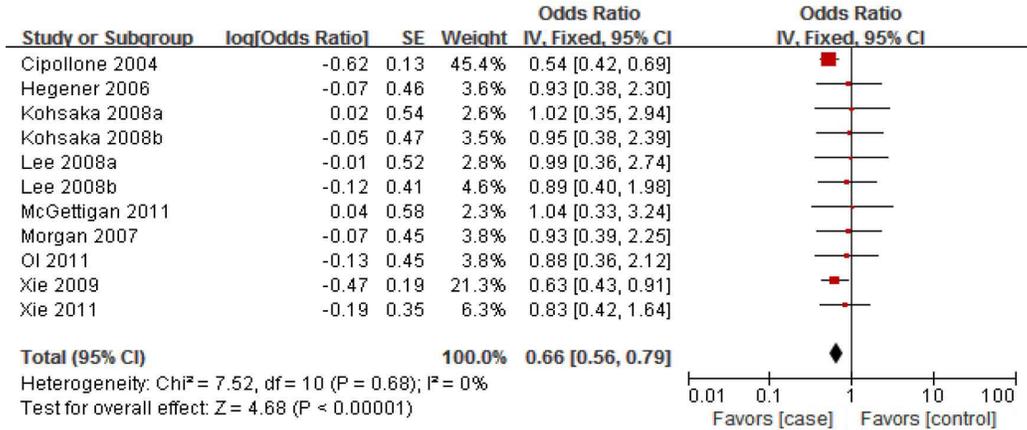


Figure 1. Forest plot of MI risk associated with COX-2. The squares and horizontal lines correspond to the study-specific OR and 95%CI, respectively. The area of the squares reflects the study-specific weight. The diamond represents the pooled results of OR and 95%CI. In this analysis, fixed-effects model was used.

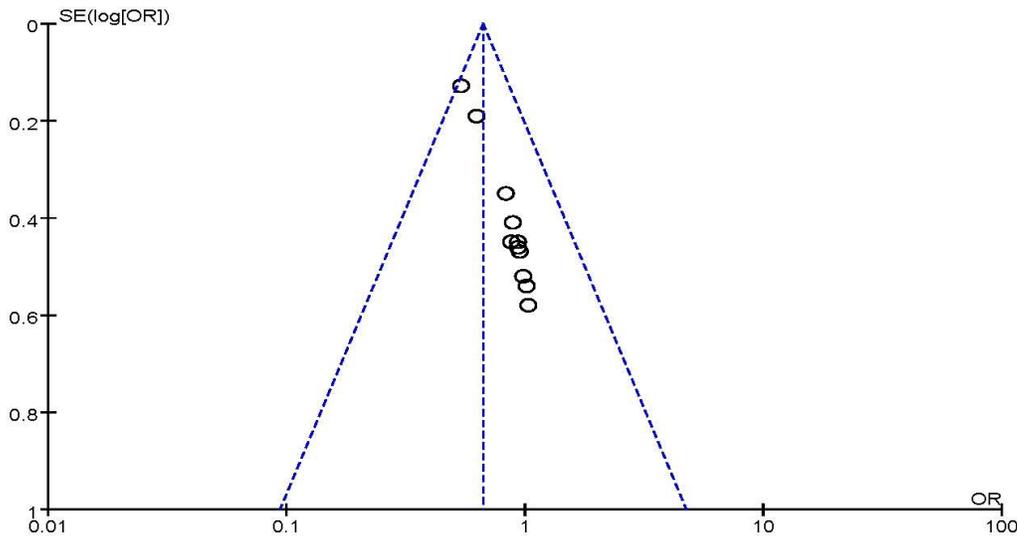


Figure 2. Begg's funnel plot for publication bias test. Each circle denotes an independent study for the indicated association. $\text{Log}[\text{OR}]$, natural logarithm of OR. Horizontal line stands for mean effect size.

Sensitivity analysis

We deleted one single study from the overall pooled analysis each time to check the influence of the removed data set on the overall ORs. The pooled ORs and 95%CIs were not significantly altered when any part of the study was omitted, which indicated that any single study had little impact on the overall ORs.

DISCUSSION

We investigated the influence of the *PTGS2* polymorphism on the risk of CAD by conducting a meta-analysis. We found the -765G>C polymorphism in the promoter region of the *PTGS2* gene was associated with a significantly decreased risk of CAD. These findings suggest that the *PTGS2* -765G>C polymorphism could be used as a marker for genetic susceptibility to CAD.

A -765G>C polymorphism in the promoter region of the *COX-2* gene disrupts the Sp1 binding site (Papafili et al., 2002), which may alter the susceptibility to develop CAD. A previous study has shown that the -765C allele may provide protective effects against myocardial infarction (Cipollone et al., 2004). The C allele may also be associated with lower levels of inflammatory markers such as C-reactive protein and interleukin-6 in cardio/cerebrovascular and hypercholesterolemic patients (Sowers et al., 2005). In contrast to these prior data, Hegener et al. (2006) found no evidence for an association of the polymorphism with risk of incident CAD (Delgado et al., 2004). Furthermore, Kohsaka et al. (2008) have recently reported that the -765C allele is in fact a risk factor for incident stroke in African-Americans (Bresalier et al., 2005).

In this meta-analysis, a total of 11 case-control studies were analyzed to provide a comprehensive assessment of the association between *PTGS2* -765G>C polymorphism and CAD. All the studies checked genotypes for quality control. Genotype distribution of controls in all studies was consistent with HWE. In addition, exploring heterogeneity is one of the important goals of a meta-analysis. In the present study no significant heterogeneity was found between the studies included. Sensitivity analysis also showed that omission of any single study did not have significant impact on the combined ORs. Furthermore, a funnel plot did not reveal obvious asymmetry, and the Egger test further indicated no considerable publication bias in this meta-analysis. This made the results of this meta-study more reliable to some extent.

There are some limitations in the present meta-analysis. In the studies included, the genotyping methods used were not the same. Besides, other clinical factors such as age, gender and different chemotherapies in each study might have led to bias. Determining whether or not these factors influence the results of this meta-analysis would need further investigation.

In conclusion, our study suggests that *PTGS2* -765G>C is associated with a significantly decreased risk of CAD. Larger well-designed epidemiological studies with ethnically diverse populations and functional evaluations are warranted to confirm our findings.

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