Original Article
Peroxisome proliferator-activated receptor-γ (PPARγ) Pro12Ala polymorphism and colorectal cancer (CRC) risk

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Abstract: Background: The association between the peroxisome proliferator-activated receptor-γ (PPARγ) Pro12Ala polymorphism and colorectal cancer (CRC) risk was inconclusive. We conducted a meta-analysis to evaluate the association between PPARγ Pro12Ala polymorphism and CRC risk. Material and Method: We searched Pubmed, EMBASE, and China National Knowledge Infrastructure databases. Data were extracted and pooled odds ratios (OR) with 95% confidence intervals (CI) were calculated. Results: A total of 17 case-control studies with 12635 and 15803 controls were included in this meta-analysis. Overall, PPARγ Pro12Ala polymorphism was associated with CRC risk (OR = 0.84, 95% CI 0.75-0.94, \( P = 0.003 \), \( I^2 = 35\% \)). In the subgroup analysis by ethnicity, a significant association was found among Caucasians (OR = 0.85, 95% CI 0.75-0.96, \( P = 0.007 \), \( I^2 = 38\% \)) but not among Asians (OR = 0.76, 95% CI 0.51-1.12, \( P = 0.17 \), \( I^2 = 28\% \)). In the subgroup analysis by CRC site, a significant association was found among colon cancer (OR = 0.81, 95% CI 0.66-0.98, \( P = 0.03 \), \( I^2 = 16\% \)) but not among rectal cancer (OR = 0.83, 95% CI 0.57-1.21, \( P = 0.34 \), \( I^2 = 63\% \)). The sensitivity analysis did not influence the result by omitting low-quality studies (OR = 0.76, 95% CI 0.63-0.93, \( P = 0.006 \), \( I^2 = 51\% \)). Conclusions: In conclusion, this meta-analysis suggested that PPARγ Pro12Ala polymorphism was significant associated with CRC risk.

Keywords: Colorectal cancer, peroxisome proliferator-activated receptor, meta-analysis, polymorphism

Introduction
Colorectal cancer (CRC) was diagnosed in 1.2 million persons worldwide in 2008, and it accounted for close to 10% of all cancers. Risk factors for CRC include advanced age, medical history of benign adenomatous polyps and inflammatory bowel diseases, family history of CRC, low intake of vegetables and fruits and high intake of dietary fat (particularly animal fat) and processed meat [1, 2]. Several lines of evidence indicate that inherited genetic factors influence the development and progress of CRC [3].

Peroxisome proliferator-activated receptor-γ (PPARγ) is part of a family of transcription factors. The PPARγ gene is expressed in many tissues, including high levels of expression in normal colonic mucosa, colorectal adenocarcinomas, and colon cancer cell lines [4, 5]. PPARγ activation also may provide a molecular link between a high-fat diet and increased risk of CRC, since studies in mice have shown that mice treated with a PPARγ ligand had greater number of polyps in the colon [6]. A relatively common variant of the PPARγ gene (substitution of Ala for Pro at codon 12) has been found, which was associated with improved insulin sensitivity, smaller body size, and reduced risk of type-2 diabetes [7]. Several studies investigated the association between PPARγ Pro12Ala polymorphism and CRC risk. However, the results remained inconclusive [8-24]. Meta-analysis is an useful method for investigating associations between genetic factors and diseases, because a quantitative approach is used to combine the results from different studies on the same topic, thereby providing more reliable conclusions. Thus, we performed a
meta-analysis to assess the association of PPARγ Pro12Ala polymorphism with CRC risk.

Methods

Publication search

In this meta-analysis, we searched the articles using the search terms “Colorectal cancer”, “Peroxisome proliferator-activated receptor-γ” and “polymorphism” in the PubMed, EMBASE and Chinese National Knowledge Infrastructure (CNKI) databases, and the last search updated on October 2014. Additional studies were identified by a hand search of references of original studies or review articles on the association between PPARγ Pro12Ala polymorphism with CRC risk. No publication date or language restriction were imposed.

Inclusion and exclusion criteria

The following inclusion criteria were used: (1) evaluation of the PPARγ Pro12Ala polymorphism with CRC risk, (2) using a case-control or cohort design, and (3) genotype distributions in both cases and controls should be available for estimating an odds ratio (OR) with 95% confidence interval (CI).

Studies were excluded if one of the following existed: (1) not relevant to CRC or PPARγ polymorphism, (2) not designed as case-control or cohort design studies, (3) genotype frequencies or number not offered, (4) animal studies, and (5) editorials, reviews and abstracts. If more than one study used the same cases, the one with the most comprehensive population were included.

Data extraction and quality assessment

The following data were collected from each study: first author’s surname, year of publication, ethnicity, CRC location, sample size, and source of controls. To assess the quality of the included studies, the Newcastle-Ottawa Scale was adopted. The studies were judged by 8 items of 3 aspects. The highest quality studies were awarded a maximum of one star of each item, except that the item of comparability allowed a maximum of two stars. Studies that controlled for age received one star, whereas studies that controlled for other important factors received an additional star. The Newcastle-Ottawa Scale score ranged from zero up to nine stars. And the high-quality study was defined ≥ 7 stars.

Statistical analysis

The strength of the associations between the PPARγ Pro12Ala polymorphism and CRC risk was measured by ORs and 95% CIs. The random-effects model was used. The statistical
### Table 2. Methodological quality of the included studies

<table>
<thead>
<tr>
<th>Author</th>
<th>Year</th>
<th>Adequate definition of cases</th>
<th>Representative-ness of cases</th>
<th>Selection of controls</th>
<th>Definition of controls</th>
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significance of summary OR was determined with Z test. The Q statistic and the I² statistic were used to assess the degree of heterogeneity among the studies included in the meta-analysis. The source of heterogeneity was detected by using Galbraith plot. Subgroup analyses were carried out by ethnicity and CRC location. Sensitivity analysis was performed by excluding low-quality studies. The potential publication bias was examined visually in a funnel plot of log [OR] against its standard error (SE), and the degree of asymmetry was tested using Egger’s test [25]. All statistical tests were performed using STATA 11.0 software (Stata Corporation, College Station, TX, USA) and Reviewer Manager 5.1. A P value < 0.05 was considered statistically significant.

### Results

#### Study characteristics

A total of 17 case-control studies with 12635 and 15803 controls on the association between PPARγ Pro12Ala polymorphism and CRC risk were included for this meta-analysis. There were 3 studies of Asians and 14 studies of Caucasians. The characteristics of each case-control study are listed in Table 1. Quality scores of each study were summarized in Table 2. The study scores ranged from 5 to 9 stars.

#### Overall and subgroup meta-analysis results

The results suggested that PPARγ Pro12Ala polymorphism was associated with CRC risk (OR = 0.84, 95% CI 0.75-0.94, P = 0.003, I² = 35%, Figure 1). In the subgroup analysis by ethnicity, a significant association was found among Caucasians.
Caucasians (OR = 0.85, 95% CI 0.75-0.96, \( P = 0.007, I^2 = 38\% \)) but not among Asians (OR = 0.76, 95% CI 0.51-1.12, \( P = 0.17, I^2 = 28\% \)). In the subgroup analysis by CRC site, a significant association was found among colon cancer (OR = 0.81, 95% CI 0.66-0.98, \( P = 0.03, I^2 = 16\% \)) but not among rectal cancer (OR = 0.83, 95% CI 0.57-1.21, \( P = 0.34, I^2 = 63\% \)). The sensitivity analysis did not influence the result by omitting low-quality studies (OR = 0.76, 95% CI 0.63-0.93, \( P = 0.006, I^2 = 51\% \)). The Galbraith plot was used to find the source of the heterogeneity. As shown in Figure 2, two studies were the outliers. After excluding these studies, the between-study heterogeneity effectively decreased and there was no obvious heterogeneity among the remaining studies (\( I^2 = 0\% \), \( P = 0.65 \)). Besides, the result was still statistically significant (OR = 0.92, 95% CI 0.86-0.99, \( P = 0.04 \)).

Funnel plot and Egger’s test were both performed to access the publication bias of this meta-analysis. The shape of the funnel plot seemed symmetrical (Figure 3). Egger’s test showed no evidence of publication bias (\( P = 0.110 \)).

Discussion

The main finding of this meta-analysis was that PPARγ Pro12Ala polymorphism was a potential protective factor for developing CRC. In the subgroup analysis of ethnicity, no significant association was found in Asians, while a significant association was found in Caucasians. It was possible that different lifestyles, diets, and environments may account for this apparent discrepancy. These issues should be investigated in the future studies. Only three studies with Asians were included in this meta-analysis. Thus, more studies with Asians should be conducted to determine the association between PPARγ Pro12Ala polymorphism and risk of CRC. In the subgroup analysis by CRC site, we found that there was a significant association between PPARγ Pro12Ala polymorphism and risk of colon cancer, suggesting that PPARγ Pro12Ala polymorphism might influence the etiology of colon cancer.

Laboratory studies have indicated a complex role of PPARγ at the cellular level through regulation of cell growth, differentiation, and apoptosis [26]. A persistent state of inflammation might lead to colon cancer, as exemplified by the high risk of colon cancer associated with ulcerative colitis [27]. Chemically induced colonic inflammation and aberrant crypt foci have been diminished in animal models by administration of PPARγ ligands [28]. The Pro12Ala polymorphism of PPARγ gene has been associated with altered lipid profiles, lower fasting insulin concentrations, improved insulin sensitivity and a reduced risk of type II diabetes and the metabolic syndrome [29]. This amino acid was located in the PPARγ domain that enhanced ligand-independent activation [30]. The Pro to Ala change may cause a conformational change in the protein, thus affecting its activity and CRC risk.

Some limitations should be acknowledged. First, only published studies that were included in the selected electronic databases were identified. It was possible that some relevant published or unpublished studies may have been missed. Second, the effects of gene-gene and gene-environment interactions were not addressed in this meta-analysis, because of limited available data. Third, there was moderate heterogeneity in this meta-analysis. However, when the main source of heterogeneity was
References


