Osteoprotegerin SNP associations with coronary artery disease and ischemic stroke risk: a meta-analysis

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Short title: OPG SNP associations with CAD and ischemic stroke
Abstract

Osteoprotegerin (OPG) is involved in the development of atherosclerosis and cardio-cerebrovascular disease. The goal of this meta-analysis was to evaluate the association of OPG single nucleotide polymorphisms (SNPs) with coronary artery disease (CAD) and ischemic stroke. A total of 15 eligible studies were extracted from electronic databases. Odds ratios (ORs) were presented, with 95% confidence intervals (CIs), to assess the associations. Meta-analysis was conducted using MetaGenyo, STATA, and Comprehensive Meta-Analysis. Meta-analysis of our data showed that the OPG SNP T950C was significantly associated with increased CAD risk among Asians via recessive (OR 1.55, 95% CI 1.18-2.04, p=0.002), CC vs TT (OR 1.57, 95% CI 1.16-2.11, p=0.003) and allelic (OR 1.21, 95% CI 1.05-1.38, p=0.007) models. No strong associations were observed for the OPG SNP G1181C, T245G and G209A with CAD risk. When evaluating the OPG SNP T245G and T950C associations with ischemic stroke, we found the OPG SNP T245G to be significantly associated with increased risk of ischemic stroke among Chinese via recessive (OR 1.53, 95% CI 1.02-2.29, p=0.039) and CC vs AA (OR 1.61, 95% CI 1.07-2.42, p=0.021) models. Our results suggested that the OPG SNP T950C was associated with increased risk of CAD among Asians, and the OPG SNP T245G was associated with enhanced ischemic stroke risk among Chinese.

Key words: Coronary artery disease; Ischemic Stroke; Meta-analysis; Osteoprotegerin; SNP
**Introduction**

Osteoprotegerin (OPG) is a member of the tumour necrosis factor (TNF) receptor superfamily, also termed as TNF receptor superfamily member 11B (TNFRSF11B) [1]. OPG acts as a decoy receptor, binding to receptor activator of nuclear factor kappa-B ligand (RANKL) and blocking its interaction with RANK. Multiple tissues and cells have been described as producing OPG, including bone, the heart, endothelial cells, and smooth muscle cells [2]. Although OPG was originally identified as a molecular regulator of bone metabolism, many studies have shown evidence of its involvement in atherosclerosis development. OPG protein and mRNA levels were elevated in human atherosclerotic plaques [3-5]. Additionally, high circulating OPG levels were positively correlated with atherosclerosis progression and the presence of coronary artery disease (CAD) and stroke [6-12]. Increased levels of OPG were also strongly predictive of long-term mortality of CAD patients [13,14]. Based on these observations, OPG was thought to be a potential new biomarker for cardio-cerebrovascular disease. In humans, there are several common single nucleotide polymorphisms (SNPs) identified in the OPG gene, including T950C (T→C, promoter), T245G (T→G, promoter), G209A (G→A, promoter), and G1181C (G→C, exon 1). These SNPs affect circulating OPG levels or protein function [15]. For example, the T950C SNP is significantly associated with increased serum OPG levels, whereas the nonsynonymous polymorphism G1181C can affect cellular secretion of OPG [15]. Numerous studies have assessed their association with CAD and ischemic stroke, but inconclusive results were obtained. To overcome the small sample size
problem, we performed an up-to-date meta-analysis to investigate the association between these OPG SNPs and the risk for CAD and ischemic stroke.

**Methods**

Search strategy

A systematic literature search was performed in March 2020 using the following databases: PubMed, Embase, China Science and Technology Journal Database (http://qikan.cqvip.com), and China National Knowledge Infrastructure (http://www.cnki.net). The search terms used in literature search were limited to the following: “osteoprotegerin or osteoclastogenesis inhibitory factor or tumour necrosis factor receptor superfamily member 11B” and “coronary heart disease or coronary artery disease or ischemic stroke and “polymorphism or gene or risk”. We did not apply any geographical restrictions or time restrictions. Reference lists of included studies and of previous related reviews were searched for additional titles. Unpublished data were not considered in this meta-analysis.

Eligibility criteria

Studies were eligible for inclusion upon meeting the following criteria: (1) case-control studies or cohort studies or retrospective studies; (2) studies evaluated the association of OPG SNPs with CAD or ischemic stroke risk; and (3) studies contained applicable data on allele or genotype distribution in both cases and controls. Exclusion criteria were as follows: (1) familial-based studies or studies using siblings; (2) studies that recruited only cases; (3) published abstracts; and (4) allele or genotype distribution can not be extracted. When studies used similar sources of data, only the
study with the largest sample size, or with the most detailed information was selected.

Data extraction

A standardised data-extraction form was developed for use in our meta-analysis to extract key information from the included articles. Specifically, one reviewer (JW) extracted data from the included articles and a second independent reviewer (XL) validated the data. The following information was collected: the last name of the first author, country, year of publication, ethnicity, the number of cases and controls, diagnosis method, the mean/range of participants’ age, percentage of male subjects, methods for genotyping OPG SNP, genotype frequency, and HWE. We did not email the corresponding authors of the eligible studies for additional information.

Quality assessment

Following the selection of final studies, the Newcastle-Ottawa Scale (NOS) was used to assess study quality [16]. It is widely accepted that the NOS is a reliable quality assessment tool for observational studies. The included studies were evaluated on 8 items across three key areas: selection of the participants, comparability of the participants and outcomes. NOS scores of 1-3, 4-6, 7-9 indicated low, intermediate and high quality, respectively.

Statistical analysis

The odds ratios (ORs) and 95% confidence intervals (CIs) were used to compare distributions of alleles and genotype contrasts between cases and controls. Allelic, dominant, recessive and co-dominant models were applied to assess the association. Pooled ORs and 95% CIs were calculated using the Mantel-Haenszel fixed-effects
model and the DerSimonian-Laird random-effects model [17,18]. Forest plots were
generated to visually show the individual study ORs and pooled ORs. Heterogeneity
across studies was assessed using the I² statistic and interpreted based on the study by
Higgins et al. [19], where 25%, 50%, and 75% represented low, moderate, and high
heterogeneity, respectively. We performed subgroup analyses according to ethnicity.
Sensitivity analysis was conducted to assess the stability of the results. Finally,
publication bias was evaluated using funnel plots and Begg’s test. All analyses were
performed using Stata, MetaGenyo [20], and Comprehensive Meta-Analysis version
2.

Results

Study characteristics

The initial electronic database search identified 622 citations. The reference lists
of the review articles were hand-searched; one related article was found. Figure 1
shows the identification, screening, and eligibility selection process. After removing
duplicates, we evaluated article title and abstracts for the remaining 357 articles and
selected 20 articles for full-text review. Following the full-text review, a total of 15
studies were included in the meta-analysis [21-35]. The countries in which the studies
had been conducted include China, Japan, Poland, Italy and Germany. The number of
studies with respect to the relationship between the OPG SNPs and CAD risk was 9
[21-28,35]. Six studies provided genotype data for ischemic stroke [29-34].
Descriptive summaries of study characteristics are shown in Tables 1 and 2.

OPG SNPs and CAD
Focusing on the OPG SNP T950C, a total of 6 studies including 1444 patients with CAD and 1023 control subjects were quantitatively analyzed (Table 1). The overall meta-analysis indicated that the OPG SNP T950C was associated with increased CAD risk in recessive (OR 1.46, 95% CI 1.15-1.85, p=0.002), CC vs TT (OR 1.54, 95% CI 1.18-2.01, p=0.001) and allelic (OR 1.21, 95% CI 1.07-1.39, p=0.002) models (Figure 2, Figure 3 and Table 3). Further subgroup analysis according to ethnicity revealed that the OPG SNP T950C was associated with increased CAD risk among Asians (OR 1.55, 95% CI 1.18-2.04, recessive model, p=0.002; OR 1.57, 95% CI 1.16-2.11, CC vs TT, p=0.003; OR 1.21, 95% CI 1.05-1.38, allelic model, p=0.007) (Table 3).

Six studies involving 1126 CAD patients and 813 controls provided results on the association of the OPG SNP G1181C with CAD risk (Table 1). The pooled analyses showed that the SNP G1181C was associated with increased CAD risk in dominant and allelic models, but not in recessive and CC vs GG models (Table 3). However, when removing two studies that was not in line with HWE [21,24], we found no association between this SNP and CAD risk (OR 1.12, 95% CI 0.88-1.43, dominant model, p=0.367; OR 1.45, 95% CI 0.99-1.78, recessive model, p=0.060; OR 1.33, 95% CI 0.88-2.01, CC vs GG model, p=0.176; OR 1.17, 95% CI 0.97-1.40, allelic model, p=0.105) (Table 3). Stratification analysis based on ethnicity did not find a significant association between CAD risk and the OPG SNP G1181C in Asians or Caucasians (Table 3).

Four studies including 754 cases and 517 controls examined the associations of
the SNP T245G with CAD risk, while 3 studies with 576 cases and 205 control subjects performed evaluation of the association between the OPG SNP G209A and CAD (Table 1). In the overall meta-analysis and stratification analysis based on ethnicity, we found no association between these SNPs and CAD risk (Table 3).

OPG SNPs and ischemic stroke

The OPG SNP T245G associations with ischemic stroke risk was evaluated in 5 studies involving 4917 cases and 4250 controls, whereas 4 studies including 2082 cases and 2031 controls focused on the T950C polymorphism (Table 2). The SNP T245G was significantly associated with ischemic stroke in recessive (OR 1.53, 95% CI 1.02-2.29, \( p=0.039 \)) and CC vs AA (OR 1.61, 95% CI 1.07-2.42, \( p=0.021 \)) models (Figure 4 and Table 4) among Chinese subjects. No significant association was found between the OPG SNP T950C and ischemic stroke when combining all studies together or performing subgroup analysis for Chinese (Table 4). Subgroup analysis could not be performed for Caucasians, as there was only one small Caucasian study.

Heterogeneity and publication bias

When evaluating the association between OPG polymorphisms and CAD, there was no significant heterogeneity (\( I^2<50\% \)) in most pooled estimates (Table 3). Significant heterogeneity was found between studies evaluating OPG polymorphisms and ischemic stroke (Table 4). However, when subgroup analysis was performed, heterogeneity was greatly reduced in the 4 genetic models for the SNP T950C and in recessive and CC vs AA models for the SNP T245G (Table 4). The number of included study for each SNP was less than 10, precluding us from creating funnel
plots. So we used the Begg’s test to evaluate publication bias. The data did not suggest the presence of publication bias (p>0.10). Sensitivity analysis was performed to evaluate the robustness of the observed outcomes. The relevant pooled ORs were not considerably altered after excluding any study (Supplementary file).

**Discussion**

CAD is the major cause of fatality and disability for both men and women in the world. It accounts for more than 7 million deaths each year. Due to its high incidence, prevalence and mortality, the identification of a specific biomarker that can be used to estimate an individual person’s risk of developing CAD would be invaluable. OPG is a cytokine belonging to the TNF receptor superfamily. Initial studies found that OPG was an important mediator of bone remodeling, but recent experimental and observational studies demonstrated that OPG may also contribute to the development of atherosclerosis and CAD. In atherosclerotic plaques, elevated OPG mRNA and protein levels were observed [3-5]. Several observational studies found that serum levels of OPG was independently correlated with the presence and severity of CAD after adjusting for traditional risk factors such as hypertension and smoking in logistic regression models [8,10,11]. Moreover, increased OPG levels strongly predicted adverse clinical outcomes in patients with CAD. These findings raised the possibility that genetic polymorphisms in the OPG might affect an individual person’s risk of CAD.

We evaluated the association between CAD and several OPG SNPs, including T950C, G1181C, T245G, and G209A in this present study. These are common SNPs...
within the OPG gene and potentially affect the OPG protein’s structural or functional properties. Through a comprehensive meta-analysis using 9 studies, we found that the OPG SNP T950C was associated with increased risk of CAD (OR 1.46 for recessive model; OR 1.54 for CC vs TT model; OR 1.21 for allelic model), especially in Asian subjects. Studies of the OPG SNP G1181C, T245G, and G209A showed no significant association with CAD risk.

Jia et al. [36] meta-analyzed the association of the OPG SNP T950C, G1181C, T245G, and G209A with CAD risk, including 2 Chinese-language and 4 English-language studies in 2017. Their results showed an association of the OPG SNP T950C with CAD risk, in line with our findings. The sample size of their meta-analysis was not large; only 3 studies were pooled to estimate the association. In our study, we increased the sample size by including 3 new studies published since the previous meta-analysis and confirmed that the OPG SNP T950C was an important genetic risk factor for CAD. Although Jia et al. also reported an association between the OPG SNP G1181C and CAD risk, this association was not confirmed by our meta-analysis. We did not detect any significant effect of the SNP G1181C on CAD among either Asians or Caucasians. Additionally, sensitivity analysis by excluding the studies that did not follow HWE showed no association. With respect to the OPG SNP G209A and T245G, the results obtained by Jia et al. [36] were in agreement with the present meta-analysis, indicating no effect of them on CAD risk.

The OPG SNP T950C lies in the OPG gene promoter (129 bp upstream from the TATA box). Previous studies reported that the SNP was significantly and
independently associated with increased circulating OPG levels [37]. It was also shown to affect atherosclerotic plaque stability [38,39]. The role of the SNP T950C in atherosclerosis development and plaque stability may help explain its association with increased CAD risk, but further functional assays are needed to investigate the precise mechanisms linking the SNP T950C and CAD. Moreover, the SNP T950C may be linked to other functional polymorphisms in the RANKL/RANK/OPG signaling pathway. A recent meta-analysis of GWAS identified new significant loci on chromosome 17q11.2 as well as chromosome 14q21.2 that associated with circulating OPG levels [40]. Future research is required to explore potential gene-gene interactions between OPG SNPs and other functional variants that affect circulating OPG levels.

Ischemic stroke is the most common form of stroke. Previous studies have observed a relation between a higher level of OPG and ischemic stroke severity and outcome [6,9,12]. To our knowledge, no published meta-analyses in the literature have evaluated the effect of OPG polymorphisms on ischemic stroke risk. Therefore, we meta-analyzed the OPG SNP T245G and T950C and their relationships with ischemic stroke in this study. The analysis showed the OPG SNP T245G to be associated with increased risk of ischemic stroke among Chinese via recessive and CC vs AA models. The SNP T245G located in the promoter region is functional for the OPG gene. It has been shown to affect OPG expression level and might be involved in atherosclerotic lesion progression [32]. Wang et al. [41] showed that the SNP T245G was correlated with a worse outcome in patients with large artery atherosclerosis.
stroke. Our meta-analysis did not evaluate the OPG SNP effects among Caucasians, due to limited published studies in the literature. However, it is noteworthy that one small Caucasian study performed in an Italian population found significant effects of the SNP T245G and T950C in ischemic stroke risk. This interesting finding needed to be confirmed in other Caucasian populations.

Our meta-analysis has some limitations. First, for some studies, the selection of CAD patients might bias the results. These studies recruited hospital patients undergoing diagnostic coronary angiography for suspected CAD, who were generally a highly selected population \[21,22,26\]. Second, the relation between the SNP T245G and T950C and ischemic stroke was only evaluated among Italian and Chinese. It needs to be further assessed in more ethnic groups. Third, heterogeneity was not fully eliminated by subgroup analysis when evaluating the relation between the SNP T245G and ischemic stroke. Heterogeneity could also be related to other factors such as study design and patients’ characteristics. Fourth, the pooled ORs were not adjusted for patient characteristics including body mass index, arterial hypertension and hypercholesterolemia, due to a small size or insufficient information. Finally, subgroup analysis by subtype of CAD or ischemic stroke was unavailable.

In summary, the present meta-analytic study identified a significant association between the OPG SNP T950C and the risk of CAD among Asians via recessive, CC vs TT and allelic models. Additionally, we showed the OPG SNP T245G to be significantly associated with increased ischemic stroke risk among Chinese via recessive and CC vs AA models. We are hopeful that our findings would be further


evaluated by adequately powered and well-designed studies, especially in non-Asian populations.

**Abbreviations**

CAD, coronary artery disease; CIs, confidence intervals; HWE, Hardy-Weinberg equilibrium; NOS, Newcastle-Ottawa Scale; OPG, osteoprotegerin; OR, odds ratio; RANKL, receptor activator of nuclear factor kappa-B ligand; SNP, single nucleotide polymorphism; TNF, tumour necrosis factor; TNFRSF11B, TNF receptor superfamily member 11B.

**Authors’ contributions**

JL and HQ were involved in the conception and design of the meta-analysis. JW and XL contributed to study selection, data extraction, statistical analysis and writing of manuscript. FG and SG contributed to the preparation of the meta-analysis and literature review. JL and HQ critically revised the manuscript and all authors read and approved the manuscript.

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**Ethics approval**

Not applicable.

**Disclosure statement**

The authors declare that they have no competing interests.

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### Table 1 Characteristics of included studies that evaluated OPG polymorphisms and CAD risk in the meta-analysis.

| First author | Year | Country | Ethnic Group | CAD diagnosis | Sample size | Male subjects% | Controls’ MAF | In HWE | NOS | Genotyping method |
|--------------|------|---------|--------------|---------------|-------------|----------------|--------------|--------|-----|-------------------|
| Soufi        | 2004 | Germany | Caucasian (German) | Coronary angiography | 361 | 107 | 100 | 44.9% | Yes | 8 | DNA sequencing |
| Ohnori       | 2006 | Japan   | Asian (Japanese) | Coronary angiography | 405 | 126 | NR | 33.3% | Yes | 8 | PCR-RFLP |
| Xu           | 2009 | China   | Asian (Chinese) | NR | 48 | 102 | NR | 37.7% | Yes | 7 | PCR-RFLP |
| Fang         | 2010 | China   | Asian (Chinese) | Coronary angiography | 150 | 150 | 70.7 | 63.3 | 45.0% | Yes | 8 | PCR-RFLP |
| Guo          | 2013 | China   | Asian (Chinese) | NR | 178 | 312 | 64.0 | 62.5 | 41.0% | Yes | 7 | PCR-RFLP |
| Zhao         | 2019 | China   | Asian (Chinese) | Coronary angiography | 302 | 226 | 61.6 | 45.1 | 36.9% | Yes | 8 | PCR-RFLP |
| Soufi        | 2004 | Germany | Caucasian (German) | Coronary angiography | 361 | 107 | 100 | 27.6% | No | 8 | DNA sequencing |
| Fang         | 2010 | China   | Asian (Chinese) | Coronary angiography | 150 | 150 | 70.7 | 63.3 | 27.0% | No | 8 | PCR-RFLP |
| Celczyński a Bajew | 2011 | Poland  | Caucasian (Poles) | Elective coronary arteriography | 31 | 30 | 0 | 0 | 56.7% | Yes | 8 | PCR |
| Hong         | 2012 | China   | Asian (Chinese) | Coronary angiography | 222 | 146 | 62.5 | 59.6 | 27.1% | Yes | 7 | PCR |
| Luo          | 2012 | China   | Asian (Chinese) | Coronary angiography | 184 | 68 | NR | NR | 18.3% | Yes | 6 | DNA sequencing |
| Guo          | 2013 | China   | Asian (Chinese) | NR | 178 | 312 | 64.0 | 62.5 | 33.2% | Yes | 7 | PCR-RFLP |
| Soufi        | 2004 | Germany | Caucasian (German) | Coronary angiography | 361 | 107 | 100 | 6.0% | Yes | 8 | DNA sequencing |
| Celczyński a Bajew | 2011 | Poland  | Caucasian (Poles) | Elective coronary arteriography | 31 | 30 | 0 | 0 | 8.0% | Yes | 8 | PCR |
| Luo          | 2012 | China   | Asian (Chinese) | Coronary angiography | 184 | 68 | NR | NR | 11.2% | Yes | 6 | DNA sequencing |
| Guo          | 2013 | China   | Asian (Chinese) | NR | 178 | 312 | 64.0 | 62.5 | 11.5% | Yes | 7 | PCR-RFLP |
| Soufi        | 2004 | Germany | Caucasian (German) | Coronary angiography | 361 | 107 | 100 | 6.0% | Yes | 8 | DNA sequencing |
| Celczyński a Bajew | 2011 | Poland  | Caucasian (Poles) | Elective coronary arteriography | 31 | 30 | 0 | 0 | 8.0% | Yes | 8 | PCR |
| Luo  | 2012 | China | Asian (Chinese) | Coronary angiography | 184 | 68 | NR | NR | 13.2% | Yes | 6 | DNA sequencing |

Abbreviations: MAF, minor allele frequency; NOS, Newcastle-Ottawa Scale; NR, not reported; PCR-RFLP, polymerase chain reaction-restriction fragment length polymorphism.
| First author | Year | Country | Ethnic Group | Diagnosis of ischemic stroke | Sample size | Male subjects% | Controls% | In HWE | NOS | Genotyping method |
|--------------|------|---------|--------------|------------------------------|-------------|----------------|-----------|--------|-----|------------------|
| **T245G**    |      |         |              |                              |             |                |           |        |     |                  |
| Sun          | 2016 | China   | Asian (Chinese) | CT or MRI scan              | 372         | 62.2           | 43.3      | Yes    | 8   | PCR-RFLP         |
| Biscetti     | 2016 | Italy   | Caucasian     | CT or MRI scan              | 487         | 49.7           | 51.0      | No     | 7   | PCR-RFLP         |
| Xiong        | 2018 | China   | Asian (Chinese) | CT or MRI scan              | 2835        | 56.4           | 65.8      | Yes    | 8   | High-resolution method |
| Wang         | 2018 | China   | Asian (Chinese) | CT or MRI scan              | 1010        | 74.1           | 63.1      | Yes    | 8   | SNPscan          |
| Fan          | 2018 | China   | Asian (Chinese) | CT or MRI scan              | 213         | 65.7           | 56.9      | Yes    | 7   | PCR              |
| **T950C**    |      |         |              |                              |             |                |           |        |     |                  |
| Sun          | 2016 | China   | Asian (Chinese) | CT or MRI scan              | 372         | 62.2           | 43.3      | Yes    | 8   | PCR-RFLP         |
| Biscetti     | 2016 | Italy   | Caucasian     | CT or MRI scan              | 487         | 49.7           | 51.0      | Yes    | 7   | PCR-RFLP         |
| Wang         | 2018 | China   | Asian (Chinese) | CT or MRI scan              | 1010        | 74.1           | 63.1      | Yes    | 8   | SNPscan          |
| Fan          | 2018 | China   | Asian (Chinese) | CT or MRI scan              | 213         | 65.7           | 56.9      | Yes    | 7   | PCR              |

**Abbreviations:** MAF, minor allele frequency; HWE, Hardy-Weinberg equilibrium; NOS, Newcastle-Ottawa Scale; PCR-RFLP, polymerase chain reaction-restriction fragment length polymorphism.

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| OPG SNP | Population | Number of studies | Dominant model |  | Recessive model |  | CC vs TT model |  | Allelic model |  |
|---------|------------|------------------|----------------|---|----------------|---|----------------|---|----------------|---|
|         |            |                  | OR (95% CI)    | p | I² | OR (95% CI)    | p | I² | OR (95% CI)    | p | I² |
| T950C   | Overall    | 6                | 1.45 (0.94-2.24) | 0.092 | 82.6 | 1.46 (1.15-1.85) | 0.082 | 0 | 1.54 (1.18-2.01) | 0.001 | 0 | 1.21 (1.07-1.38) | 0.002 | 0 |
|         | Asian      | 5                | 1.45 (0.96-2.45) | 0.166 | 36.1 | 1.55 (1.18-2.04) | 0.082 | 0 | 1.57 (1.16-2.11) | 0.003 | 0 | 1.21 (1.09-1.38) | 0.007 | 0 |
|         | Overall    | 6                | 1.23 (1.02-1.50) | 0.034 | 7.9 | 1.20 (0.99-1.42) | 0.239 | 49.4 | 1.31 (0.98-1.74) | 0.057 | 34.5 | 1.22 (1.05-1.41) | 0.009 | 37.2 |
|         | Asian      | 4                | 1.21 (0.96-1.51) | 0.082 | 11.7 | 1.11 (0.77-1.60) | 0.566 | 62.4 | 1.18 (0.86-1.62) | 0.385 | 80.9 | 1.26 (0.94-1.70) | 0.170 | 61.4 |
|         | Caucasian  | 2                | 1.50 (0.96-2.37) | 0.286 | 48.2 | 1.40 (0.82-2.39) | 0.212 | 78.9 | 1.29 (0.72-2.32) | 0.396 | 0 | 1.28 (0.95-1.74) | 0.100 | 59.6 |
|         | Pelican HWE| 4                | 1.12 (0.80-1.54) | 0.367 | 21.3 | 1.11 (0.75-1.68) | 0.600 | 70.5 | 1.33 (0.88-2.01) | 0.176 | 59.8 | 1.37 (0.97-1.94) | 0.105 | 59.9 |
| T245G   | Overall    | 4                | 0.98 (0.71-1.44) | 0.479 | 0 | 1.50 (0.52-4.29) | 0.454 | 6.3 | 1.47 (0.51-4.24) | 0.474 | 3.5 | 1.02 (0.73-1.40) | 0.019 | 95.0 |
|         | Asian      | 2                | 0.96 (0.66-1.39) | 0.624 | 0 | 1.50 (0.52-4.29) | 0.454 | 6.3 | 1.47 (0.51-4.24) | 0.474 | 3.5 | 1.05 (0.74-1.43) | 0.847 | 95.0 |
|         | Caucasian  | 2                | 0.96 (0.55-1.74) | 0.103 | 0 | NA | NA | NA | NA | NA | 0.96 (0.54-1.71) | 0.887 | 0 |
| G209A   | Overall    | 3                | 0.95 (0.69-1.31) | 0.740 | 0 | 4.99 (0.26-89.76) | 0.276 | NA | 4.64 (0.26-83.84) | 0.299 | NA | 1.01 (0.67-1.51) | 0.978 | 0.0 |
|         | Caucasian  | 2                | 1.01 (0.56-1.82) | 0.987 | 0 | NA | NA | NA | NA | NA | 1.01 (0.57-1.78) | 0.988 | 0.0 |

**Abbreviations:** CAD, coronary artery disease; CI, confidence interval; NA, not applicable; OPG, osteoprotegerin; OR, odds ratio; SNP, single-nucleotide polymorphism.
Table 4 Summary of comparative study outcomes for ischemic stroke and OPG polymorphisms.

| OPG SNP | Population | Number of studies | Dominant model | Recessive model | CT vs AA model | Allelic model |
|---------|------------|-------------------|----------------|----------------|----------------|--------------|
|         |            |                   | OR (95% CI)    | OR (95% CI)    | OR (95% CI)    | OR (95% CI)  |
|         |            |                   | p              | p              | p              | p            |
|         |            |                   | I²             | I²             | I²             | I²           |
| T245G   | Overall    | 5                 | 1.32 (0.97-1.79)| 1.88 (0.86-4.06)| 1.93 (0.78-4.81)| 1.29 (0.95-1.80)|
|         | Chinese    | 4                 | 1.18 (0.97-1.43)| 1.53 (1.02-2.28)| 1.61 (1.07-2.42)| 1.15 (0.95-1.39)|
|         | Follow HWE | 4                 | 1.18 (0.97-1.43)| 1.53 (1.02-2.28)| 1.61 (1.07-2.42)| 1.15 (0.95-1.39)|
| T950C   | Overall    | 4                 | 1.17 (0.79-1.78)| 1.21 (0.57-2.89)| 1.28 (0.84-3.11)| 1.14 (0.76-1.71)|
|         | Chinese    | 3                 | 0.97 (0.83-1.12)| 0.98 (0.57-1.39)| 0.91 (0.53-1.56)| 0.96 (0.78-1.17)|

Abbreviations: CI, confidence interval; NA, not applicable; OPG, osteoprotegerin; OR, odds ratio; SNP, single-nucleotide polymorphism.
Figure 1  Flow diagram for included studies.
### Figure 2

Forest plot demonstrating the association between the OPG SNP T950C and coronary artery disease risk via recessive model.
Figure 3  Forest plot demonstrating the association between the OPG SNP T950C and coronary artery disease risk via CC vs TT model.
Figure 4  Forest plots demonstrating the association between the OPG SNP T245G and ischemic stroke risk among Chinese. (A) recessive model; (B) CC vs AA model.
Supplementary file 1  Sensitivity analysis of the association between the SNP T950C and CAD risk in recessive model.
Supplementary file 2  Sensitivity analysis of the association between the SNP T245G and ischemic stroke risk in dominant model.
