Effects of Tumor Necrosis Factor-β (TNF-β) 252A>G Polymorphism on the Development of Migraine: A Meta-Analysis

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Abstract

Background and Objective: Genetic factors including TNF-β have been considered as important components in the aetiology of migraine. Many studies have investigated the association between TNF-β 252A>G polymorphism and migraine risk, with debatable results. This study was designed to examine whether the TNF-β 252A>G polymorphism confers genetic susceptibility to migraine in diverse populations.

Method: Studies eligible for this meta-analysis were searched in the PubMed, Embase, and CNKI by using the keywords “tumor necrosis factor”, “TNF”, “252A>G”, “rs909253”, “polymorphism”, “polymorphisms”, “variant”, “SNP”, combined with “migraine” or “migraine with aura (MA)” or “migraine without aura (MO)”. Pooled ORs and 95% CI were appropriately calculated using the fixed-effect model.

Results: We finally included a total of seven studies, providing 5,557 migraineurs and 20,543 unrelated healthy controls. Meta-analysis results showed no statistical evidence of a significant association between TNF-β 252A>G polymorphism and overall migraine risk. Stratified analyses by type of migraine and gender revealed similar results. Interestingly, an OR with 95% CI representing an increased migraine risk was indicated in Asians under the recessive model (GG vs. AG + AA: OR, 1.38; 95%CI, 1.04–1.84; P for heterogeneity, 0.665).

Conclusions: Our findings appear to support the hypothesis that genetic variability of 252A>G polymorphism in TNF region may modulate risk of migraine in the population of Asian ancestry.

Introduction

Migraine is a chronic neurological disorder accompanied with digestive system and autonomic nervous system symptoms composed of nausea, vomiting and extreme photophobia, and phonophobia [1,2]. It is estimated that 8.4% of the general population (10% of men and 24% of women) are afflicted by the common disease [3–5]. With the characteristics of severity and intense throbbing pain in the head, migraine has been ranked as one of the top 20 most debilitating diseases worldwide [6]. Despite the currently incomplete definition of the exact pathophysiology of migraine, cytokine-related genes, such as tumor necrosis factor (TNF), interleukin-1β (IL-1β) and IL-10, are known genetic candidates contributing to the predisposition towards migraine development [7–9].

TNF (TNF-α and lymphotoxin-α or LT-α or TNF-β) are cytokines implicated to influence the intensity and duration of local inflammation. TNF could act as an inflammatory mediator in the activation and sensitization of meningeal nociceptors, threshold brain excitability, and propagation of neuronal hyperexcitability, consequently leading to persistent pulsating headache characterized in migraine [10,11]. Single nucleotide polymorphisms (SNPs) within this locus are likely to be genetically associated with a variety of autoimmune and infectious diseases, due to the major histocompatibility complex and biological activities in the location of this gene [12].

TNF-β mapped on chromosome 6 is a member of the TNF cytokine superfamily, playing an important role in immune and inflammatory responses [13]. Polymorphisms of this gene have been found to have regulatory function in cytokine levels of TNF-β [14]. 252A>G polymorphism (rs909253) is such a variant with silent point mutation capable of modifying gene expression and has been investigated in migraine risk research in multiple populations. The susceptibility of 252A>G polymorphism was
primarily reported in a population of Italian ancestry, with a
discovery suggesting that carriage of the A allele conferred high
risk for the development of migraine without aura (MO) [15].
Since then, several follow-up studies have been continuously
carried out in an effort to replicate the initial finding. Some were
successful [16,17], whereas others failed [18,19]. Multiple factors
including selection of diverse populations, different control
sources, and misclassification of genotypes may individually or
jointly result in the controversy, but the main source can attribute
to the various numbers of subjects used in each study.

The aim of this study was to determine whether 252A>G
polymorphism in the TNF-β region is linked with the risk of
migraine. We hypothesized that the 252A>G polymorphism may
be associated with migraine risk. The hypothesis was examined
though a meta-analysis of 5,557 migraineurs and 20,543 unrelated
healthy controls from seven publications.

Materials and Methods

Literature source and search strategy

Studies examining the association between TNF-β 252A>G
polymorphism and migraine were systematically searched using
the keywords “tumor necrosis factor”, “TAP”, “252A>G”,
“rs909253”, “polymorphism”, “polymorphisms”, “variant”,
“SNP”, combined with “migraine”, or “migraine with aura
(MA)” or “migraine without aura (MO)” in the PubMed, Embase,
and China national knowledge infrastructure (CNKI) by two
independent investigators. A manual search for potentially
relevant studies was also conducted by reviewing the literature
identified in the electronic databases. The last search was updated
in June, 2013.

Inclusion criteria

The meta-analysis only included the articles i) that evaluated the
association of TNF-β 252A>G polymorphism and migraine risk
between patients and matched controls; ii) that contained
genotype frequency in full detail allowing for calculation of crude
risks for migraine. We excluded the studies reporting the
association between TNF-β 252A>G polymorphism and survival
from migraine and/or response to any therapy. If more than one
article was published by the same author sharing the same cases
series, we selected the study with the most subjects.

Data extraction

Usable data were independently extracted from each paper by
two investigators using a structured sheet and then pooled into a
database. The items recorded were first author, publication year,
location of the study, racial descent of the studied subjects, type of
controls, mean age of the cases, total number of cases and controls
for migraine, MA and MO, and the corresponding genotype
distributions.
Statistical analysis

Statistical analysis was performed by using STATA version 12.0 (Stata Corporation, College Station, TX). The significance threshold was defined as P < 0.05 unless specially stated. Deviation from Hardy-Weinberg equilibrium (HWE) was tested using the goodness-of-fit $\chi^2$ test for each control group of the included studies. An odd ratio (OR) with 95% confidence interval (CI) was measured to estimate the association between 252A $\rightarrow$ G polymorphism and migraine risk. The significance for pooled ORs was evaluated by Z test. The ORs of homozygous model (GG vs. AA), dominant model (GG + AG vs. AA), recessive model (GG vs. AG + AA), allele model (G vs. A), and heterozygous model (AG vs. AA) were calculated by the fixed effects model or the random effects models according to the outcome of between-study heterogeneity detected by the Chi square based Q test [20]. The fixed effects model with the use of Mantel-Haenszel (M–H) method [21] was performed for estimates of the pooled ORs when heterogeneity was indicated to be non-significant (P > 0.05); otherwise, the random effects model based on DerSimonian and Laird (D–L) method [22] was more appropriate.

Stratified analyses were performed by racial descent, type of migraine, and gender. The leave-one-out sensitivity analysis was carried out to detect the extent of influence from the single studies on the combined results. The funnel plot and the Egger’s test were applied to evaluate publication bias across studies [23].

Results

Study inclusion and characteristics

The systematically electronic along with manual searches supplied 137 papers that may have usable data for this meta-analysis. However, 126 of them were removed after preliminary review of titles and abstracts. Among the remaining 11 papers, we screened the full texts to examine their eligibility based on the inclusion criteria. As a result, 4 studies were excluded (one was carried out without control population; one focused on other polymorphisms rather than the 252A $\rightarrow$ G polymorphism; two presented insufficient genotype data for risk estimate) [24–27] and thus 7 studies [15–19,28,29] were considered eligible for the present analysis (Figure 1), including four Caucasian studies and three Asian studies. All of the included studies were case-control designed, comprising 5 557 migraineurs and 20 543 unrelated healthy controls. In addition, five studies provided available data for MA and seven for MO. None of the genotype distributions in each control group was significantly deviated from HWE (Table 1 and 2).

Quantitative synthesis

Table 3 lists the main meta-analysis results calculated with the fixed effects model. By pooling the 7 selected studies, non-significant association between 252A $\rightarrow$ G polymorphism and migraine was observed under all genetic models. Strikingly, the meta-analysis provided an OR of 1.38 (95%CI, 1.04–1.84; P for heterogeneity, 0.665) for migraine associated with 252A $\rightarrow$ G.

![Table 1. General characteristics of the studies included in the present meta-analysis.](image)

| First Author | Publication Year | No. of cases/controls | MAF (cases/control) | Racial Descent | Study Country | Type of Controls | Mean Age |
|--------------|------------------|-----------------------|---------------------|----------------|---------------|-----------------|----------|
| Trabace      | 2002             | 77/101                | 0.253/0.386         | Caucasian      | Italy         | Unrelated healthy controls | 39.7±7.4, 36.7±6.9 |
| Lee          | 2007             | 439/382               | 0.468/0.425         | Asian          | Korea         | Healthy controls | 47.8±11.2     |
| Asuni        | 2009             | 299/278               | 0.183/0.133         | Caucasian      | Italy         | Healthy Sardinian controls | 35.28±10.22 |
| Schurks      | 2009             | 4332/19269            | 0.337/0.336         | Caucasian      | USA           | NS              | NS        |
| Ghosh        | 2010             | 216/216               | 0.241/0.241         | Asian          | India         | Healthy controls | 32.08±12.28  |
| Pappa        | 2010             | 103/178               | 0.189/0.216         | Caucasian      | Greece        | Unrelated healthy controls | 10.5±0.7 |
| Ishii        | 2012             | 91/119                | 0.390/0.328         | Asian          | Japan         | Non-headache healthy volunteer | 42.4±10.2 |

NS: not specified in original paper.
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![Table 2. Available data for meta-analysis of MA and MO risk in association with TNF-$\beta$ 252A $\rightarrow$ G polymorphism.](image)

| First Author | Publication Year | MA No. of Cases | MA No. of Controls | MO No. of Cases | MO No. of Controls |
|--------------|------------------|-----------------|-------------------|-----------------|-------------------|
| Trabace      | 2002             | 30              | 101               | 47              | 101               |
| Lee          | 2007             | 65              | 382               | 327             | 382               |
| Asuni        | 2009             | 1213            | 19269             | 1824            | 19269             |
| Schurks      | 2010             | 84              | 216               | 132             | 216               |
| Pappa        | 2010             | 103             | 178               |                 |                   |
| Ishii        | 2012             | 24              | 119               | 67              | 119               |

MA: migraine with aura, MO: migraine without aura.
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polymorphism under the recessive model (GG vs. AG + AA) in Asian populations (Figure 2). None of the contrast models showed a significant association in Caucasian populations.

When stratifying the data by type of migraine, we did not observe either an increased risk or decreased risk of MA or MO. Subgroup analysis based on gender provided similar results that as compared with men, women had no higher risk of developing migraine or MO (Table 3).

To explore the potential sources of heterogeneity on the overall results, we conducted sensitivity analysis and the initial ORs were not significantly influenced by sequential removal of each study from the total analysis. Egger’s test and the Begg’s funnel plots performed in all contrast models provided no statistical evidence of publication bias (P, 0.900 and P, 1.000, respectively for the two tests). However, if the two factors are obviously indicated in related tests. Considering the number of Caucasian and Asian subjects is much larger relative to the Asian subjects (24,637 vs. 1,463), which assures an adequate detection power in Caucasian population. And we cannot exclude the possibility that the current positive association revealed in Asians may be derived by chance. The other explanation may be the different frequency of the G allele between the two populations: for Asians, the frequency is 37.4% and 22.6% for Caucasians. This implies that the genetic background is a potential factor influencing the genetic predisposition to migraine.

Heterogeneity and publication bias are two inevitable confounding factors which could distort the true associations between SNPs and diseases under research when performing meta-analysis. In our study, we observed statistical evidence for a significant association of the 252A>G polymorphism with migraine in Asian subjects, but not in Caucasians. Multiple possibilities may result in the differential relation, but there are two most likely explanations. One refers to the study size. Notably, the number of Caucasian subjects is much larger relative to the Asian subjects (24,637 vs. 1,463), which assures an adequate detection power in Caucasian population. And we cannot exclude the possibility that the current positive association revealed in Asians may be derived by chance. The other explanation may be the different frequency of the G allele between the two populations: for Asians, the frequency is 37.4% and 22.6% for Caucasians. This implies that the genetic background is a potential factor influencing the genetic predisposition to migraine.

Table 3. Main results of the pooled data in the present meta-analysis.

| Subgroups | GG vs. AA | GG + AG vs. AA | GG vs. AG + AA | G vs. A | AG vs. AA |
|-----------|-----------|----------------|----------------|--------|-----------|
|           | OR (95%CI) | P (Q-test)     | OR (95%CI)     | P (Q-test) | OR (95%CI) | P (Q-test) | OR (95%CI) | P (Q-test) | OR (95%CI) | P (Q-test) |
| Total     | 1.01 (0.92, 1.12) | 0.246 | 1.01 (0.96, 1.06) | 0.538 | 1.01 (0.92, 1.11) | 0.160 | 1.01 (0.97, 1.06) | 0.139 | 1.01 (0.96, 1.07) | 0.658 |
| Ethnicity |           |         |                |        |            |         |            |         |            |         |
| Caucasian | 0.98 (0.89, 1.09) | 0.204 | 1.01 (0.96, 1.07) | 0.179 | 0.97 (0.88, 1.08) | 0.340 | 1.00 (0.96, 1.05) | 0.047 | 1.02 (0.96, 1.08) | 0.259 |
| Asian     | 1.29 (0.95, 1.75) | 0.717 | 1.03 (0.87, 1.21) | 0.944 | 1.38 (1.04, 1.84) | 0.665 | 1.09 (0.95, 1.25) | 0.765 | 0.98 (0.81, 1.19) | 0.985 |
| Subtype   |           |         |                |        |            |         |            |         |            |         |
| MA        | 1.04 (0.87, 1.24) | 0.440 | 1.02 (0.93, 1.12) | 0.952 | 1.02 (0.87, 1.21) | 0.174 | 1.02 (0.95, 1.10) | 0.735 | 1.03 (0.93, 1.13) | 0.887 |
| MO        | 1.03 (0.90, 1.18) | 0.284 | 1.01 (0.94, 1.08) | 0.540 | 1.05 (0.92, 1.20) | 0.248 | 1.01 (0.95, 1.07) | 0.120 | 1.00 (0.93, 1.08) | 0.711 |
| Gender    |           |         |                |        |            |         |            |         |            |         |
| Migraine  | 0.76 (0.35, 1.67) | 0.569 | 0.91 (0.65, 1.29) | 0.910 | 0.80 (0.37, 1.72) | 0.552 | 0.90 (0.67, 1.22) | 0.679 | 0.92 (0.64, 1.33) | 0.933 |
| MO        | 0.53 (0.20, 1.36) | 0.934 | 0.89 (0.61, 1.30) | 0.971 | 0.54 (0.22, 1.38) | 0.903 | 0.85 (0.61, 1.19) | 0.988 | 0.92 (0.62, 1.38) | 0.933 |

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Discussion

Migraine is a primary headache disorder with its aetiology being incompletely understood. In recent years, a great deal of attention has been directed to the field of migraine genetics [30]. Migraine is recognized as a polygenic disorder induced by a wide range of candidate genes that confer a minor nonetheless significant impact on the susceptibility [31]. Evidence from twin studies showed that approximately 40% of total migraine can be attributable to genetic factors is of substantial importance in risk estimate. Such association and migraine. Further, influence from environmental factors is of substantial importance in risk estimate. Such exploration was precluded owing to the inadequate data. The other explanation may be the different frequency of the G allele between the two populations: for Asians, the frequency is 37.4% and 22.6% for Caucasians. This implies that the genetic background is a potential factor influencing the genetic predisposition to migraine.

Heterogeneity and publication bias are two inevitable confounding factors which could distort the true associations between SNPs and diseases under research when performing meta-analysis. Considerable attention should be paid in interpreting the findings if the two factors are obviously indicated in related tests. Fortunately, as no significant heterogeneity and publication bias were revealed in our meta-analysis, the results for the association assessed in the present study are relatively convincing by using a rigorous analytic approach. However, the possibility that the effect size of risk estimate is derived by chance cannot be excluded, because the total number of cases in general analysis, not to mention the subgroup analyses, is not large enough to obtain a precise assessment of the association between 252A>G polymorphism and migraine. Further, influence from environmental factors is of substantial importance in risk estimate. Such exploration was precluded owing to the inadequate data.

To sum up, these data provided further evidence that 252A>G polymorphism in the TNF region may represent a risk factor for the development of migraine in the population of Asian ancestry. No statistical evidence was indicated in subgroup analyses by type of migraine and gender. Further investigations using a much larger sample with interplay between environment and gene taken into account are clearly needed to validate the findings in this meta-analysis.
Figure 2. Forest plot of migraine risk associated with TNF-β 252A>G polymorphism stratified by ethnicity under GG vs. AG + AA model. The boxes and horizontal lines represent the OR and the corresponding 95% CI. The area of the boxes indicates the weight (inverse of the variance). The diamond correspond to the summary OR and 95% CI.
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Figure 3. Begg’s funnel plot for TNF-β 252A>G polymorphism. Log OR is plotted versus standard error of Log OR for each included study. Each circle dot represents a separate study for the indicated association between TNF-β 252A>G polymorphism and migraine risk under GG vs. AA model.
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Supporting Information

Checklist S1  PRISMA Checklist.

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