Original Article

Angiotensinogen polymorphism and ischemic stroke risk

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Abstract: The angiotensinogen M235T polymorphism was associated with ischemic stroke risk. However, the results were controversial. Thus, a meta-analysis was conducted. NCBI, Medline, Web of Science and Embase databases were systematically searched. Summary odds ratios (ORs) and corresponding 95% confidence intervals (CIs) were estimated using random-effects models. There was a significant association between angiotensinogen M235T polymorphism and ischemic stroke risk (OR = 1.69; 95% CI, 1.35-2.11; \(P\) < 0.001). In the stratified analysis by ethnicity, we found that this polymorphism was significantly associated with ischemic stroke in Asian (OR = 1.85; 95% CI, 1.45-2.35; \(P\) < 0.001). In the age subgroup, we found that angiotensinogen M235T polymorphism could increase both early-onset ischemic stroke risk (OR = 1.88; 95% CI, 1.33-2.43; \(P\) < 0.001) and late-onset ischemic stroke risk (OR = 1.20; 95% CI, 1.01-1.39; \(P\) = 0.04). This meta-analysis suggested that angiotensinogen M235T polymorphism was associated with ischemic stroke.

Keywords: Ischemic stroke, angiotensinogen, meta-analysis, genetic

Introduction

Stroke is the fourth leading cause of death in the United States. One in 6 people worldwide will have a stroke during their lifetime and one third of the survivors will experience a recurrence [1, 2]. In the past 4 decades, the incidence rates of stroke decreased by 42% in the economically developed nations, while they increased in the developing countries [3]. Recently, some genetic variants, such as single nucleotide polymorphisms (SNPs) of several genes, are shown to be associated with an increased risk of ischemic stroke [4], suggesting that genetic factors may play an important role in ischemic stroke development.

The renin-angiotensin-aldosterone system (RAAS) plays a major role in maintaining salt-water balance and controlling blood pressure [5]. Angiotensinogen is an upstream member of the RAAS encoded by the AGT gene on chromosome 1. It is primarily synthesized in the liver and then cleaved by renin from the kidney to form angiotensin I. Angiotensin I is further cleaved by the angiotensin-converting enzyme (ACE) to form the biologically active form called angiotensin II. Didion et al. found that acetylcholine-induced relaxation of carotid artery is impaired selectively in mice made hypertensive by expression of human renin and human angiotensinogen [6]. Maeda et al. suggested that in angiotensinogen-knockout mice the more efficient collateral blood supply delays ischemic injury despite the lower blood pressure [7].

The human angiotensinogen gene is located on chromosome 1q42-43. Some studies investigated the association between the angiotensinogen M235T polymorphism and susceptibility of ischemic stroke [8-28]. However, the results were controversial and inconsistent. In this meta-analysis, we evaluated the correlation between angiotensinogen M235T polymorphism and ischemic stroke risk.

Methods

Publication search

Relevant studies were systematically searched by using the NCBI, Medline, Web of Science and...
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![Flow chart of included studies for this metaanalysis.](image)

**Statistical analysis**

The strength of association between angiotensinogen M235T polymorphism and ischemic stroke risk was assessed by calculating OR with 95% CI. A statistical test for heterogeneity was performed based on the Q statistic. The $P > 0.10$ of the Q-test indicated a lack of heterogeneity among studies. The random effects model was used to calculate the pooled ORs. A goodness-of-fit $\chi^2$ test was used to check whether the frequencies of genotypes deviate from the Hardy-Weinberg equilibrium (HWE). Stratified analysis was performed by ethnicity and age. Cumulative meta-analysis was conducted. The one-way sensitivity analyses were performed to assess the stability of the results, namely, a single study in the meta-analysis was deleted each time to reflect the influence of the individual data set to the pooled ORs. Potential publication bias was examined by Egger’s test. All statistical tests were performed with the software STATA version 11.0 (Stata Corporation, College station, TX, USA). A $P$ value $< 0.05$ was considered statistically significant.

**Results**

**Study characteristics**

As shown in Figure 1, 21 studies met the inclusion criteria and were included in the final analysis [8-28]. Only 4 case-control study included Caucasians; while 17 studies were performed in Asian population. Only 2 studies were not in HWE. The characteristics of included studies summarized in Table 1.

**Results of meta-analysis**

There was a significant association between angiotensinogen M235T polymorphism and ischemic stroke risk ($OR = 1.69; 95\% CI, 1.35-2.11; P < 0.001; Figure 2$). In the stratified analysis by ethnicity, we found that this polymorphism was significantly associated with

Embase databases (The last retrieval date was Dec 1, 2014, using the search terms: “ischemic stroke” and “angiotensinogen” and “polymorphism”). All searched studies were retrieved and only published studies with full-text articles were included. When more than publications with duplicate samples, only the newest study was used in this research.

**Inclusion and exclusion criteria**

The following inclusion criteria were used: (1) evaluate the association between angiotensinogen M235T polymorphism and ischemic stroke risk; (2) a case-control or cohort design; (3) sufficient data should have been provided in order to calculate odds ratios (OR) and 95% confidence interval (CI). Studies were excluded if any of the following conditions applied: (1) not relevant to angiotensinogen or ischemic stroke; (2) only case population; (3) studies were repeated or publications overlapped.

**Data extraction**

The following data were recorded from each article: first author, years of publication, ethnicity of participants, age, numbers of cases and controls, and genotype numbers of angiotensinogen M235T polymorphism. The data were extracted by two of the authors independently. Discrepancies between these two authors were resolved by discussion.
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Table 1. Characteristics of the studies

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<th>Ethnicity</th>
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<th>No. of control</th>
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HWE, Hardy-Weinberg equilibrium.

Figure 2. Meta-analyses of the angiotensinogen M235T polymorphism and ischemic stroke risk.

ischemic stroke in Asian (OR = 1.85; 95% CI, 1.45-2.35; P < 0.001). In the age subgroup, we found that angiotensinogen M235T polymorphism could increase both early-onset ischemic stroke risk (OR = 1.88; 95% CI, 1.33-2.43; P < 0.001) and late-onset ischemic stroke risk (OR = 1.20; 95% CI, 1.01-1.39; P = 0.04).

Table 2 listed the results of the meta-analysis.

As shown in Figure 3, the results showed that the pooled ORs tended to be stable. Statistically similar results were obtained after sequentially excluding each study and the corresponding pooled ORs were not materially altered (Figure 4), suggesting stability and liability of this meta-analysis.
Table 2. Results of this meta-analysis

<table>
<thead>
<tr>
<th>No. of study</th>
<th>OR (95% CI)</th>
<th>P</th>
<th>I² (%)</th>
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<td>Overall</td>
<td>1.69 (1.35-2.11)</td>
<td>&lt; 0.001</td>
<td>77</td>
</tr>
<tr>
<td>Asian</td>
<td>1.85 (1.45-2.35)</td>
<td>&lt; 0.001</td>
<td>68</td>
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<tr>
<td>Caucasian</td>
<td>1.23 (0.82-1.83)</td>
<td>0.32</td>
<td>82</td>
</tr>
<tr>
<td>Early-onset</td>
<td>1.88 (1.33-2.43)</td>
<td>&lt; 0.001</td>
<td>32</td>
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<tr>
<td>Late-onset</td>
<td>1.20 (1.01-1.39)</td>
<td>0.04</td>
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</table>

Egger’s test was used to provide statistical evidence of funnel plot symmetry (Figure 5) and did not detect evidence of publication bias ($P = 0.11$).

Discussion

Many studies indicated that genetic factors played important roles in the development of ischemic stroke. Cui found that MTHFR C677T mutation increased the risk of ischemic stroke in adults, especially in large-artery atherosclerosis [29]. Türkanoğlu Özcelik et al. suggested that NOS3 genetic polymorphisms are the risk of development of ischemic stroke the Turkish Population [30]. van Goor et al. indicated that PAI-1 4G/5G polymorphism is a strong risk factor for ischemic stroke [31]. Furthermore, Han and coworkers suggested that both rs1711503 and rs2479408 of PCSK9 genes were associated with cerebral ischemic stroke in the Han population of China [32].

This meta-analysis of 21 studies evaluated the association between angiotensinogen M235T polymorphism and ischemic stroke risk. The results indicated that angiotensinogen M235T polymorphism was a risk factor for ischemic stroke. In the stratified analysis by ethnicity, this polymorphism was significantly associated with ischemic stroke in Asians. However, no significant association between this polymorphism and ischemic stroke risk in Caucasian was found. In the age subgroup, we found that this polymorphism could increase both early-onset ischemic stroke risk and late-onset ischemic stroke risk.
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This result suggested that angiotensinogen M235T polymorphism might play important roles in the development of early-onset ischemic stroke risk and late-onset ischemic stroke.

Angiotensinogen M235T polymorphism was also associated with some disease risks. Mao et al. found that angiotensinogen M235T polymorphism might be a protective factor against the Henoch-Schönlein purpura risk in adult [33]. Wang et al. suggested that M235T polymorphism in the angiotensinogen gene might be related to the increased risk of atrial fibrillation in Asians [34]. Jiang et al. found that angiotensinogen M235T polymorphism is a low-penetrant risk factor for the development of heart failure among Asians [35].

Our meta-analysis had some limitations. First, the numbers of published studies were not sufficient for a comprehensive analysis, particularly for Africans. Second, other than M235T polymorphism, there are other variants in the angiotensinogen gene. We did not carry out meta-analysis on these polymorphisms due to limited data. Third, lacking of the original data of the eligible studies limited the evaluation of the effects of the gene-gene interactions in ischemic stroke.

In conclusion, this meta-analysis suggested that angiotensinogen M235T polymorphism is significantly associated with the risk of ischemic stroke.

Figure 5. Funnel plot between the angiotensinogen M235T polymorphism and ischemic stroke risk.

Disclosure of conflict of interest

None.

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References

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