Association Between SLC30A8 Gene rs3802177 Polymorphism and T2DM in Asian Population-a Meta-Analysis

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**Keywords:** Type 2 diabetes mellitus, Genetic polymorphism, Meta-analysis.

**Abstract.** To further confirm the association between SLC30A8 gene rs3802177 polymorphism and T2DM in Asian population, we collect data from multiple relevant papers and performed Meta-analysis. We constructed allele model, dominant model and recessive model by fixed effects model or random effects models which were determined according to the $Q$ value and $I^2$ value calculated by heterogeneity test. OR with 95\% CI was selected as evaluation index to discuss the influences of SLC30A8 gene rs3802177 polymorphisms on T2DM. The most reasonable genetic model was determined by comparisons between every two genotypes. According to the inclusion and exclusion criteria, a total of five qualified articles were used for data retrieval, which include 6234 cases and 6169 controls. Significant association were found in all the genetic models (C vs T: OR=1.110, 95\% CI 1.053-1.169, $P<0.001$, CC+CT vs TT: OR=1.124, 95\% CI 1.018-1.240, $P=0.020$, CC vs CT +TT: OR=1.153, 95\% CI 1.071-1.241, $P<0.001$), suggesting that the polymorphism of rs3802177 in SLC30A8 gene is associated with the occurrence of T2DM, and the C allele is a risk gene. The results of genotype comparison showed that the most suitable genetic model was recessive model, in which individuals with CC genotypes were 1.153 times more likely to have T2DM than those with CT and TT genotypes. The results suggested SLC30A8 gene rs3802177 polymorphism is associated with type 2 diabetes in Asian population.

**Introduction**

Type 2 diabetes mellitus (T2DM) is a chronic non infectious disease characterized by glucose metabolism disorder caused by dysregulation of multiple genes and environmental factors [1]. The current studies found that more than one gene were associated with T2DM [2, 3]. Solute carrier family 30 member 8 gene (SLC30A8) is expressed in pancreatic beta cells, and encodes a protein named zinc encoding member 8 transporter (ZnT8). ZnT8 is involved in the transportation and accumulation of zinc in intracellular vesicles, so as to provide zinc for the maturation and/or storage of pancreatic beta cells [4]. Mutated alleles at some loci in SLC30A8 may increase susceptibility to T2DM. A number of studies showed that rs13266634 polymorphism is associated with the occurrence of T2DM [5-7]. According to Dimas [8] and
Voight [3], rs3802177 in SLC30A8 is in strong linkage disequilibrium with rs13266634. Similar results were also obtained from the study of 1000 Genomes project. Despite the strong linkage disequilibrium, there are inconsistent results of rs13266634 and rs3802177 in terms of insulin sensitivity and insulin secretion [4, 8]. In addition, conclusions about the correlation between rs3802177 polymorphism and T2DM remains controversial, likely due to small sample size used in the research. Therefore, we collected data from multiple relevant papers to create a larger sample size and performed Meta-analysis based on this pooled data to further confirm the association between SLC30A8 gene rs3802177 polymorphism and T2DM.

Materials and Methods

Search Strategy

Two reviewers performed this search using the terms (rs3802177 and (zinc transporter protein member 8 or ZnT8 or ZnT-8 or SLC30A8)) to search PubMed, Ovid, springer, CNKI, Wanfang database, ChongqingVIP. Articles were restricted on those written in English or Chinese. Eligible articles were included and irrelevant literatures were excluded through reading the title and/or abstracts and/or reading through the full text.

Inclusion and Exclusion Criteria

Inclusion criteria: ① Association study was case-control or cohort study of Asian populations; ② The study involved the association of rs3802177 polymorphism with T2DM in SLC30A8 gene, ③ The study contained results about frequency of each genotype or odds ratio (OR) with 95% confidence interval (CI) of each genotype.

Exclusion criteria: ① abstract, comment, review or editorial; ② study was conducted by other ethnic groups other than the Asian population, such as Europe, Africa, etc.; ③ subjects were not T2DM patients, such as T1DM, gestational diabetes mellitus, or animals; ④ duplication articles from the same research; ⑤ lack enough data about the frequency of each genotype or OR with 95% CI of each genotype from articles for meta-analysis.

Data Extraction

Information was extracted from eligible articles by two reviewers independently after searching each database. Data extraction included the following information: first author's name, the year of publication, study design, sample size, age, gender, frequency of each genotype or OR with 95% CI of each genotype, ethnicity. Discrepant results were discussed or judged by the third reviewer until consensus was reached.

Statistical Analyses

Meta-analyses were conducted by Stata 14 software. We constructed allele model (C vs T), dominant model (CC+CT vs TT) and recessive model (CC vs CT+TT) by fixed effects model or random effects model which were determined according to the Q-test result and I2 value calculated by heterogeneity test: if Q-test result was not significant (P>0.05) or I2<50%, the fixed effects model was used, otherwise, the random effects model was selected. Egger method and Begg funnel plot were used to judge the publication bias of each model. OR with 95% CI were selected as the evaluation index to explore the effect of SLC30A8 gene rs3802177 polymorphism on T2DM. According to the model selection method developed by Thakkinstian

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[9], the optimal model was constructed by comparing the genotypes (CC vs TT, OR1; CT vs TT, OR2; CC vs CT OR3).

Results

Screening Process of Included Articles

Fig. 1 showed the screening process according to the search strategy. According to the terms, a total of 21 papers were retrieved, which included 9 Chinese papers and 11 English papers. Eleven papers were excluded through reading the title and/or abstracts (4 do not meet the inclusion criteria and 7 were duplication articles). 5 articles were excluded after reading the full text (genotype frequency or OR with 95%CI of each genotype was not provided). A total of 5 articles that met the criteria were used for the meta-analysis, which included 3 Chinese papers and 2 English papers [10-14].

Figure 1. Schematic diagram of the screening process.

Characteristics of Eligible Articles

Five case-control studies included 6234 cases and 6169 controls were enrolled. Table 1 showed the characteristics of 5 eligible articles, including first author's name, the year of publication, sample size, gender, frequency of each genotype.

Table 1. Characteristics of 5 case-control studies of T2DM and rs3802177 polymorphism.

<table>
<thead>
<tr>
<th>First author</th>
<th>Publication year</th>
<th>Sample size</th>
<th>Male/female (%)</th>
<th>CC/CT/TT (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Yukio Horikawa</td>
<td>2008</td>
<td>1840/1572</td>
<td>56.9/43.1/43.7</td>
<td>35.3/48.1/16.6</td>
</tr>
<tr>
<td>Fu LL</td>
<td>2012</td>
<td>727/650</td>
<td>48.0/52.0/46.2</td>
<td>40.6/44.2/15.3</td>
</tr>
<tr>
<td>Wassim Y</td>
<td>2013</td>
<td>995/1076</td>
<td>42.8/57.2/31.2</td>
<td>59.2/33.4/7.4</td>
</tr>
<tr>
<td>Zhao LJ</td>
<td>2015</td>
<td>1740/1939</td>
<td>58.8/41.2/47.0</td>
<td>35.3/48.2/16.6</td>
</tr>
<tr>
<td>Su YX</td>
<td>2016</td>
<td>932/932</td>
<td>60.9/39.1/62.8</td>
<td>48.6/37.2/14.2</td>
</tr>
</tbody>
</table>
Meta-analysis of the association between SLC30A8 gene rs3802177 polymorphism and T2DM

According to the information retrieved from 5 eligible studies, we constructed allele model (C vs T), dominant model (CC+CT vs TT) and recessive model (CC vs CT +TT). Results of heterogeneity test showed that there was homogeneity among the studies (C vs T: $Q=1.76$, $P=0.780$, $I^2=0.000$, CC+CT vs TT: $Q=1.07$, $P=0.900$, $I^2=0.000$, CC vs CT +TT: $Q=3.36$, $P=0.499$, $I^2=0.000$). Therefore, the fixed effect model analysis was applied.

The pooled analysis showed that there were significant overall association among all genetic models (C vs T: $OR=1.110$, 95%CI 1.05-1.17, $P<0.001$, CC+CT vs TT: $OR=1.124$, 95%CI 1.018-1.240, $P=0.020$, CC vs CT +TT: $OR=1.153$, 95%CI 1.071-1.241, $P<0.001$). (Table 2 and Fig. 2).

Publication bias was judged through Egger method and Begg funnel plot. There was no obvious asymmetry in Begg funnel plot (Fig. 3). Results of Egger’s test also showed that there was not significant publication bias among genetic models (C vs T: $P=0.408$, CC+CT vs TT: $P=0.656$, CC vs CT +TT: $P=0.516$).

Table 2. Meta-analysis results of three genetic models.

<table>
<thead>
<tr>
<th>genetic models</th>
<th>OR</th>
<th>OR 95%CI</th>
<th>$P$</th>
<th>heterogeneity test</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Lower bound</td>
<td>Upper bound</td>
<td>$\chi^2$</td>
</tr>
<tr>
<td>C vs T</td>
<td>1.110</td>
<td>1.053</td>
<td>1.169</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>CC+CT vs TT</td>
<td>1.124</td>
<td>1.018</td>
<td>1.240</td>
<td>0.020</td>
</tr>
<tr>
<td>CC vs CT +TT</td>
<td>1.153</td>
<td>1.071</td>
<td>1.241</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>
Figure 2. The association between SLC30A8 gene rs3802177 polymorphism and T2DM.
Figure 3. Begg’s publication bias funnel plot of the ORs for the association between SLC30A8 gene rs3802177 polymorphism and T2DM.
Selection of Genetic Model

As suggested by Thakkinstian [9], the optimal model was selected through pairwise comparison among genotypes (CC vs TT, OR₁; CT vs TT, OR₂; CC vs CT OR₃). If results of genotypes comparison showed that OR₁ and OR₃ are significant and OR₂ is not, then the optimal model is the recessive model; if OR₁ and OR₂ are significant and OR₃ is not, then the optimal model is the dominant model; if all the results are significant, then codominant model is the optimal model. Pairwise comparison among genotypes showed that OR₁=1.215, 95%CI: 1.091-1.354, P<0.001; OR₂=1.058, 95%CI: 0.953-1.175, P=0.289; OR₃=1.137, 95%CI: 1.051-1.299, P=0.001, indicating that the most suitable model was recessive model. (Table 3).

Table 3. Pairwise comparison among SLC30A8 gene rs3802177 genotypes.

<table>
<thead>
<tr>
<th>genetic models</th>
<th>OR</th>
<th>OR 95%CI</th>
<th>P</th>
<th>heterogeneity test</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Lower bound</td>
<td>Upper bound</td>
<td></td>
</tr>
<tr>
<td>CC vs TT OR₁</td>
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<td>1.091</td>
<td>1.354</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>CT vs TT OR₂</td>
<td>1.058</td>
<td>0.953</td>
<td>1.175</td>
<td>0.289</td>
</tr>
<tr>
<td>CC vs CT OR₃</td>
<td>1.137</td>
<td>1.051</td>
<td>1.299</td>
<td>0.001</td>
</tr>
</tbody>
</table>

Discussion

T2DM is a disease caused by multiple genes and environmental factors that contribute to insulin resistance or insufficient insulin secretion [15]. SLC30A8 encodes the Zn²⁺ transporter ZnT8, which is responsible for the Zinc supply in pancreatic β cells [4], therefore mutations in this gene sequence may affect the function of β cells and thus increase the risk of T2DM. So far, many association studies have been carried out using polymorphism loci located in this gene, such as rs13266634, which has been confirmed to increase the risk of T2DM [6]. Locating in the same exon (exon 13 of SLC30A8) as the rs13266634, rs3802177 has also been studied for potential association with T2DM occurrence. However, different groups have reached inconsistent conclusions regarding the correlation between rs3802177 and T2DM.

In the present study, we performed a meta-analysis to further confirm the association between rs3802177 and T2DM. A total of 5 articles were included in the meta-analysis with 6234 cases of T2DM and 6169 cases of control enrolled. Allele model (C vs T), dominant model (CC+CT vs TT) and recessive model (CC vs CT +TT) were constructed to investigate the association between rs3802177 polymorphism and T2DM. At the same time, pairwise comparison among SLC30A8 gene rs3802177 genotypes was performed to determine the most suitable gene model.

Results of meta analysis showed that significant overall association was found in all the genetic models (significance level of P<0.05, OR>1), suggesting that the polymorphism of rs3802177 in SLC30A8 gene is associated with the occurrence of T2DM, and the C allele is a risk gene. Pairwise comparison among genotypes showed that OR₁ and OR₃ were statistically significant, but OR₂ was not, which means the most suitable model was recessive model (OR₁=1.153, 95%CI 1.071-1.241, P<0.001). Individuals with CC genotypes were 1.153 times more likely to have T2DM than those with CT and TT genotypes.

Compared to previous studies, our analysis exhibited several advantages: First, heterogeneity was examined by heterogeneity test before constructing genetic models. Egger method and Begg funnel plot were applied to judge the publication bias, which made our results more credible.
Second, we constructed three different genetic models and determined the most reasonable genetic model of rs3802177 polymorphism on T2DM through pairwise comparison among genotypes. Therefore, the results of this paper can provide a reference for the follow-up study.

However, limitations also exist in this study. First, due to the limited number of qualified articles, the pooled sample size in our analysis is still not very large. Moreover, the targeted population was limited to Asian populations, not involving other ethnic groups. Subsequent studies could be carried out for different regions and ethnic groups. Second, the occurrence of T2DM involved in the dysregulation of multiple genes and particular environmental risk factors. In this study, we only discussed the association between SLC30A8 rs3802177 polymorphism and T2DM. To obtain a comprehensive view of the T2DM pathogenesis, interactions between different genes and environment factors should also be included. Third, due to language limitations, we are not able to include related articles not written in English or Chinese into our meta-analysis, leading to certain publication bias.

Acknowledgement

This study was supported by scientific fund from National Natural Science Fund in China (No.81360445)

References


