Genetic polymorphisms of methylenetetrahydrofolate reductase (MTHFR) gene and susceptibility to depression in Asian population: a systematic meta-analysis

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Abstract

Genetic association studies on MTHFR C677T polymorphism and depression have been repeatedly performed over the last two decades and results are generally inconsistent. The aim of the present study was to assess the risk of MTHFR C677T polymorphism for depression in Asian population. The author performed a meta-analysis and pooled data from individual case-control studies that examined the association between C677T polymorphism and depression (meta-analysis: 13 studies, 1,120 cases and 1,688 controls). The pooled Odds Ratios (OR) were estimated by both fixed effects and random effects models. Overall, there was an association between MTHFR C677T polymorphism and increased risk of depression under five genetic models (OR T vs. C=1.44, 95% CI= 1.56-1.78, P<0.001; OR TT vs. CC= 1.78, 95% CI 1.17–2.69, P=0.006; OR CT vs CC=1.102, 95% CI=0.91-1.32, P=0.31; OR TT vs. CT+CC=1.73, 95% CI=0.87-3.41, P=0.11; OR TT+CT vs. CC=1.26, 95% CI=0.96-1.64, P=0.08). Sensitivity analysis suggested exclusion of any single study did not alter the overall pooled ORs. In conclusion results of present meta-analysis supports that there is a significant association between MTHFR C677T polymorphism and depression risk, and MTHFR 677T allele contributes to increased risk of depression in Asian individuals.

Key words: Meta-analysis, Depression, MTHFR, C677T, Genotype, Polymorphism, Asian population.

Introduction

Neuropsychiatric diseases are complex genetic disorders and family, twin and adoption studies indicate that simple Mendelian genetics with one gene–one phenotype are not applicable to psychiatric diseases. Mood disorders are among the most prevalent forms of mental illness. Epidemiologic studies show that roughly 40%-50% of the risk for depression is genetic (1). This makes depression a highly heritable disorder, at least as heritable as several common complex medical conditions like type II diabetes, hypertension, asthma etc. Although a possible role of nutritional factors in the pathogenesis of neuropsychiatric disorders has long been debated, clinical studies have shown an inverse relationship between folate status and depression (2-4). Such a relationship has been inferred from studies showing increased frequency of folate deficiency among depressed patients, more severe and prolonged (5) depressive episodes and weaker treatments with low folate status (5-7) and enhanced antidepressed response with folic acid supplementation (5,7,8).

The principal biochemical role of folate in mammals is in mediating the transfer of single-carbon molecules for various biological reactions (9). Folate plays an integral role in DNA synthesis and methylation, and as an epigenetic regulator of gene expression, DNA integrity and stability (9). The intracellular coenzymatic form of folate is 5, 10-methylenetetrahydrofolate which is required for de novo purine synthesis (9) and also in the remethylation of homocysteine to methionine. Methionine is the precursor of S-adenosylmethionine (SAM), which is the primary methyl group donor for most biological methylation reactions, including DNA methylation (10). Methylene tetrahydrofolate reductase (MTHFR) is one of the enzymes involved in this metabolic pathway. MTHFR catalyzes the irreversible conversion of 5,10-methylenetetrahydrofolate to 5-methyltetrahydrofolate and directs the flux of intracellular folate toward the conversion of homocysteine to methionine at the expense of nucleotide synthesis (11). C677T (Ala222Val) is clinically important and common polymorphism which has been inferred from studies showing increased frequency of C677T allele between individuals with depression and psychiatric disorders etc. (12-20). There is marked variation in the frequency of C677T allele between populations (21-23). The highest frequency (>20%) is found among US Hispanics, Colombians and Amerindians in Brazil, conversely in Black populations, less than 2 percent have the variant genotype (24). Case–control studies that investigated the association between depression and the C677T polymorphism so far provided controversial or inconclusive results. Each study involved small numbers of cases and controls, and data interpretation was complicated by the fact that different populations and sampling strategies were used. To shed some light on these controversial results, as well as to decrease the uncertainty of the effect size of estimated risk, a meta-analysis of all available Asian studies relating the C677T polymorphism of the MTHFR gene to the risk of developing depression was carried out.

Methods

Study identification

A literature search for all studies reporting on the association between MTHFR C677T genotype and de-
pression was conducted using the electronic databases PubMed, Google Scholar and Springer link up to October, 2013. The search strategy included the keywords “depression”, “polymorphism”, “methylenetetrahydrofolate reductase” and “MTHFR”. Eligible studies had to meet all of the following criteria: (1) published in a peer-reviewed journal, (2) contained independent data, (3) presented sufficient data to calculate the odds ratio (OR) with a confidence interval and a P-value, (4) were association studies, (5) described the relevant genotyping protocols, (6) they properly diagnosed patients according to DSM-IV criteria and (7) they used healthy individuals as controls.

**Data extraction**

Following informations were extracted from each study- first authors name, journal name, year of publication, country name, number of cases and controls. When a study reported results on different subpopulations according to ethnicity, author considered each subpopulation as a separate study in the meta-analysis.

**Meta-analysis**

The meta-analysis examined the overall association of the 677T allele with the risk of depression relative to allele C. The associations were indicated as odds ratios (OR) with the corresponding 95% confidence interval (CI). Then, based on the individual OR, a pooled OR was estimated. Heterogeneity between studies was tested using the Q-statistic, which is a weighted sum of squares of the deviations of individual study OR estimates from the overall (pooled) estimate (25) and quantified with the I2 metric. I2 takes values between 0 and 100% with higher values denoting greater degree of heterogeneity (26,27). The pooled OR was estimated using fixed effects (FE) (28) and random effects (29) models. Random effects modeling assume a genuine diversity in the results of various studies, and it incorporates to the calculations a between-study variance. Hence, when there is heterogeneity between studies, then the pooled OR is preferably estimated using the random effects model.

**Publication bias**

Publication bias was investigated with the funnel plot. Funnel plot asymmetry was further assessed by the method of Egger's linear regression test (30). All analyses were performed using the computer program MIX version 1.7 (31). A p value less than 0.05 was considered statistically significant, and all the p values were two sided.

**Results**

**Characteristics of included studies**

Thirteen studies, summarized in Table 1, reported the association of SNP C677T polymorphism in the MTHFR gene with depression and included in the present meta-analysis (32-44). The studies were carried out in Japan (32,33), Singapore (34), Taiwan (35), China (36-39,41-44), and Korea (40). In all thirteen studies, total cases were 1120 with CC (379), CT (500) and TT (241). In all thirteen studies, the studies were carried out in Japan (32,33), Singapore (34), Taiwan (35), China (36-39,41-44), and Korea (40). In all thirteen studies, total cases were 1120 with CC (379), CT (500) and TT (241).

**Table 1. Characteristics of thirteen studies included in the present meta-analysis.**

<table>
<thead>
<tr>
<th>S. No.</th>
<th>Study ID</th>
<th>Country</th>
<th>Case</th>
<th>Control</th>
<th>Year</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Arinami et al.,1997</td>
<td>Japan</td>
<td>32</td>
<td>419</td>
<td>1997</td>
<td>Am J Med Genet 74:526-528</td>
</tr>
<tr>
<td>3</td>
<td>Tan et al.,2004</td>
<td>Singapore</td>
<td>88</td>
<td>120</td>
<td>2004</td>
<td>Psychiatr Genet 14:227-231</td>
</tr>
<tr>
<td>4</td>
<td>Chen et al.,2005</td>
<td>China</td>
<td>39</td>
<td>20</td>
<td>2005</td>
<td>Am J Geriatr Psychiatry 13:872-875</td>
</tr>
<tr>
<td>5</td>
<td>Yuan et al.,2007</td>
<td>China</td>
<td>60</td>
<td>71</td>
<td>2007</td>
<td>Chin J Geriatrics 26:767-9.</td>
</tr>
<tr>
<td>6</td>
<td>Yuan et al.,2008</td>
<td>China</td>
<td>116</td>
<td>80</td>
<td>2008</td>
<td>Acta Neuropsychiatria 20: 251-255</td>
</tr>
<tr>
<td>8</td>
<td>Hong et al.,2009</td>
<td>China</td>
<td>178</td>
<td>85</td>
<td>2009</td>
<td>Am J Geriatr Psychiatry 17:847-55.</td>
</tr>
<tr>
<td>9</td>
<td>Kim et al.,2009</td>
<td>South Korea</td>
<td>63</td>
<td>458</td>
<td>2009</td>
<td>Psychosom. 71, 286-291</td>
</tr>
</tbody>
</table>

**Table 2. The distributions of MTHFR C677T genotypes and allele frequencies in depression cases and controls.**

<table>
<thead>
<tr>
<th>Study ID</th>
<th>Genotype</th>
<th>Alleles</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Case</td>
<td>Control</td>
</tr>
<tr>
<td>Arinami,1997</td>
<td>9</td>
<td>154</td>
</tr>
<tr>
<td>Kunugi,1998</td>
<td>30</td>
<td>95</td>
</tr>
<tr>
<td>Tan,2004</td>
<td>49</td>
<td>80</td>
</tr>
<tr>
<td>Chen,2005</td>
<td>22</td>
<td>11</td>
</tr>
<tr>
<td>Yuan,2007</td>
<td>22</td>
<td>27</td>
</tr>
<tr>
<td>Yuan,2008</td>
<td>46</td>
<td>27</td>
</tr>
<tr>
<td>Zhao,2008</td>
<td>12</td>
<td>21</td>
</tr>
<tr>
<td>Hon,2009</td>
<td>75</td>
<td>32</td>
</tr>
<tr>
<td>Kim,2009</td>
<td>16</td>
<td>84</td>
</tr>
<tr>
<td>Yang,2009</td>
<td>33</td>
<td>52</td>
</tr>
<tr>
<td>Cao,2010</td>
<td>9</td>
<td>24</td>
</tr>
<tr>
<td>Feng,2010</td>
<td>32</td>
<td>51</td>
</tr>
<tr>
<td>Quiao,2012</td>
<td>24</td>
<td>36</td>
</tr>
</tbody>
</table>
TT (241), and controls were 1688 with CC (694), CT (994), and TT (326). In controls genotypes percentage of CC, CT, and TT were 41.11%, 58.88% and 19.31% respectively. In total cases genotype percentage of CC, CT, and TT was 33.84%, 44.64% and 21.52% respectively. Frequency of CC genotype was highest in both cases and controls (Table 2). Except three studies (33,39,40), OR was above one in all included studies. Author has assessed whether the frequencies of CC, CT and TT genotypes among controls in individual studies were consistent with the expected distribution (that is in Hardy-Weinberg equilibrium) by using the X2 test. Genotypes were in Hardy-Weinberg equilibrium in all controls.

Meta-analysis

The main analysis for investigating the association of the C677T allele T and the risk of developing depression relative to the allele C showed significant heterogeneity (Phet=0.0001, I2= 68.68%) between the 13 studies; the fixed and random effects pooled OR were significant (ORFE=1.4, 95% CI=1.25-1.58, p<0.0001; OR RE=1.44, 95%CI=1.56-1.78, p=0.001) (Table 3; Figure 1). The genotype differences for the homozygote (TT vs. CC) revealed significant heterogeneity (Phet= 0.02; I2= 60.83%) and a significant association in both fixed and random effects pooled OR (ORFE = 1.75, 95% CI=1.37-2.23, p<0.0001; ORRE= 1.79, 95%CI= 1.17-2.69, p=0.006) (Figure 2). The recessive model for allele T (TT vs CT+CC) produced the same pattern of genotypic association as found for the homozygote frequencies, and found significant heterogeneity, (Phet=0.0001; I2=90.38), overall fixed effects OR=1.19(95%CI=1.00-1.43;p=0.04) and random effect OR= 1.73 (95%CI=0.87-3.41; p=0). The dominant model for the effect of T allele in the main analysis

Table 3. Summary estimates for the odds ratio (OR) of MTHFR C677T in various allele genotype contrasts, the significance level (p value) of heterogeneity test (Q test), and the F metric and publication bias p-value (Egger Test).

<table>
<thead>
<tr>
<th>Genetic Models</th>
<th>Fixed effect OR (95% CI), p</th>
<th>Random effect OR (95% CI), p</th>
<th>Heterogeneity p-value (Q test)</th>
<th>Publication Bias (p of Egger’s test)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Allele Contrast (T vs C)</td>
<td>1.41(1.251-1.585),&lt;0.0001</td>
<td>1.44(1.56-1.783),0.001</td>
<td>0.0001</td>
<td>68.68</td>
</tr>
<tr>
<td>Co-dominant (Ct vs CC)</td>
<td>1.10(0.92-1.326),0.3013</td>
<td>1.10 (0.878-1.381),0.4025</td>
<td>0.14</td>
<td>30.16</td>
</tr>
<tr>
<td>Homozygote (TT vs CC)</td>
<td>1.75(1.372-2.235),&lt;0.0001</td>
<td>1.78(1.174-2.692),0.006</td>
<td>0.002</td>
<td>60.83</td>
</tr>
<tr>
<td>Dominant (TT+CT vs CC)</td>
<td>1.25(1.052-1.490),0.011</td>
<td>1.26(0.969-1.640),0.084</td>
<td>0.01</td>
<td>53.45</td>
</tr>
<tr>
<td>Recessive (TT vs CT+CC)</td>
<td>1.19(1.00-1.431),0.049</td>
<td>1.73(0.871-3.416),0.117</td>
<td>&lt;0.001</td>
<td>90.38</td>
</tr>
</tbody>
</table>

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Figure 1. A. Forest plots for the association between MTHFR C677T polymorphism and depression for additive model (T vs C) with fixed effect model, B, for homozygote model (TT vs. CC).
Figure 2. A. Forest plots for the association between MTHFR C677T polymorphism and depression for dominant model (TT+CT vs. CC), B. codominant model (CT vs. CC) and C. recessive model (TT vs. CT+TT).
Publication bias

Funnel plot and Egger’s test were used to assess the publication bias in this meta-analysis. Funnel plots’ shape of all contrasts did not reveal obvious evidence of asymmetry, and all the P values of Egger’s test were showed significant marginal association with fixed effects model (OR= 1.25; 95%CI= 1.05-1.49; p=0.01) and insignificant association with random effects (OR= 1.26; 95% CI= 0.96-1.64; p=0.08) with moderate heterogeneity (Phet=0.01; I²=53.45) (Figure 3).

Figure 3. Funnel plots A. precision versus OR (T vs C), B. standard error versus OR (T vs C), C. precision versus OR (TT vs CC) D. standard error versus OR (TT vs CC), E. precision versus OR (TT+CT vs CC) and F. standard error versus OR (TT+CT vs CC).
more than 0.05, providing statistical evidence for the funnel plots’ symmetry (Table 3; Figure 4).

Discussion

Folate deficiency due to low dietary, or impaired absorption and metabolism or due to dysfunctional MTHFR enzyme, may result in decreased DNA synthesis, increased DNA strand breaks, impaired DNA repair, enhanced mutagenesis and alterations in DNA methylation patterns. All of these events have been implicated in neurodevelopment (45-47). It has also been reported that disturbances in the folate-dependent one carbon metabolism may contribute to neurodegenerative diseases including depression (48). A low concentration of SAM is associated with reduced activity of SAM-dependent methyltransferases, including DNA methyltransferase and COMT (49). MTHFR plays a central role in balancing DNA synthesis (which involves methylenetetrahydrofolate) and DNA methylation (which involves methyltetrahydrofolate). Heterozygosity and homozygosity for MTHFR C677T polymorphism resulted in a lower MTHFR activity (50) leads to hyperhomocysteinemia. High concentrations of homocysteine are toxic not only for vascular endothelial cells but also to neuronal cells (51). An adequate amount of folate is required to maintain low levels of homocysteine in the central nervous system and brain especially developing brain may be particularly vulnerable to high level of homocysteine in the blood because it lacks two major metabolic pathways for its elimination: betaine remethylation and transsulfuration (52). The possible mechanism underlying MTHFR C677T polymorphism and depression association is that the genetic and environmental factors elevate homocysteine levels, which cause vascular disease of the brain, and/or transmitter alterations, which causes depression (53).

Meta-analysis is a powerful tool for analyzing cumulative data of studies where the individual sample sizes are small and the statistical power low. Present meta-analysis has included data for the C677T MTHFR polymorphism from over 1120 subjects who were suffering with depression, along with a 1688 controls. Author found an association of this polymorphism with depression, but there was statistically significant heterogeneity in the results of different studies. Several meta-analysis studies were published to illustrate the utility of the technique in identifying genes of small effects like MTHFR with phenotypes like cancer (54,55), myocardial infarction (56), NTD (57), Down syndrome (58,59), Stroke (60), Migraine (61), Schizophrenia (20,62), bipolar disorder (63), Alzheimers disease (64), recurrent pregnancy loss (65) and oro-facial cleft (66) etc.

Author identified four meta-analysis (20,49,67,68) published between 2006 to 2013 concerning similar topic during the literature search; all four examined the effect of MTHFR C677T on depression risk. In 2006, Zintzaras (67) performed a meta-analysis based on five studies and did not find significant association between MTHFR polymorphisms and depression risk (OR=1.15; 95% CI=0.97–1.36). In 2007, another meta-analysis aggregated with ten studies and found slight significant relationship (OR=1.14; 95%CI=1.04-1.26) (49). Peerbooms et al (20) included seventeen studies of depression in their combined meta-analysis of major psychiatric disorder including 26 studies of schizophreni, bipolar and depression. In a recent meta-analysis (68), authors included twenty six studies in their meta-analysis on the association between MTHFR gene polymorphisms and depression, including 4992 depression cases and 17,082 controls and found statistical association with additive dominant model: OR = 1.19, 95%CI = 1.07–1.32. There are several newly published studies available but not included in the previous meta-analyses. In addition these meta-analyses included mixed population. So author conducted a meta-analysis with single ethnic population i.e Asian population and found strong significant association between C677T polymorphism and depression risk in Asian population. Depression risk in Asian population could be ascribed to the folate metabolism profile and dietary structure of different regions.

There were few limitations in the present meta-analysis: i) used crude ORs in the pooled analysis without adjustment; ii) relatively small sample sizes of some studies included in the analysis. Present meta-analysis had some strength. First, only Asian studies were included, second substantial number of cases and controls were pooled from different studies, which significantly increased the statistical power of the analysis and third, no publication biases were detected; indicating that the whole pooled results may be unbiased.

In conclusion, pooled analysis of data from thirteen separate studies indicates that the MTHFR 677TT genotype is associated with significant risk of depression in Asian population. This association was complicated by between study heterogeneity. Future large-scale, population-based association studies are required to investigate potential gene–gene and gene–environment interactions involving the MTHFR C677T polymorphism in determining depression risk in Asian population.

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References


