RESEARCH ARTICLE

# The MTHFR C677T Polymorphism and Risk of Acute Lymphoblastic Leukemia: an Updated Meta-analysis Based on 37 Case-control Studies

**Yuan Jiang[1], Jing Hou[1], Qiang Zhang[1], Shu-Ting Jia[2], Bo-Yuan Wang[3], Ji-Hong Zhang[2], Wen-Ru Tang[2]*, Ying Luo[2]***

## Abstract

**Background: The C677T polymorphism of the methylenetetrahydrofolate reductase (MTHFR) has been associated with acute lymphoblastic leukemia (ALL). However, results were conflicting. The aim of this study was to quantitatively summarize the evidence for the MTHFRC677T polymorphism and ALL risk. Methods: Electronic searches of PubMed and the Chinese Biomedicine database were conducted to select case-control studies containing available genotype frequencies of C677T and the odds ratio (OR) with 95% confidence interval (CI) was used to assess the strength of any association. Results: Case-control studies including 6,371 cases and 10,850 controls were identified. The meta-analysis stratified by ethnicity showed that individuals with the homozygous TT genotype had decreased risk of ALL (OR= 0.776, 95% CI: 0.687~0.877, *p*< 0.001) in Caucasians (OR= 0.715, 95% CI: 0.655~0.781, *p*= 0.000). However, results among Asians (OR=0.711, 95% CI: 0.591~1.005, *p*= 0.055) and others (OR=0.913, 95% CI: 0.656~1.271, *p*= 0. 590) did not suggest an association. A symmetric funnel plot, the Egger's test (*P*=0.093), and the Begg- test (*P*=0.072) were all suggestive of the lack of publication bias. Conclusion: This meta-analysis supports the idea that the MTHFR C677T genotype is associated with risk of ALL in Caucasians. To draw comprehensive and true conclusions, further prospective studies with larger numbers of participants worldwide are needed to examine associations between the MTHFRC677T polymorphism and ALL.**

**Keywords:** MTHFR C677T - meta-analysis - acute lymphoblastic leukemia - risk factor

*Asian Pac J Cancer Prev,* **14 (11)**, 6357-6362

## Introduction

Acute lymphoblastic leukaemia (ALL) is the maximum common pediatric leukemia accounting for 25-30% of all cases of childhood malignancies (Krajinovic et al., 1999). The disease-free survival of childhood ALL has surpassed 80% in the developed countries over the last years. Nevertheless, almost 20% of the children with ALL either revert or do not respond to treatment (Karathanasis et al., 2011). Even though the scientific, pathological and immunophenotypic types of the disease are well acknowledged, about leukemogenesis is little known (Krajinovic et al., 1999). A range of factors might be related to the biologic mechanisms and etiology of ALL. It is generally considered that the development of ALL is a comprehensive result of environment, genetic risk factors, and gene-environment interactions (Robien et al., 2003; Scelo et al., 2009). Folate deficiency and aberrant metabolism have been reported to be associated with ALL (Yang et al., 2011). Poly-morphisms in genes involved in folate transport, metabolism, and distributionin vivo drew widespread attention in the last decade (McNeer 2011). The polymorphisms in the methylenetetrahydrofolate reductase ( MTHFR) gene C677T, has been associated with acute lymphoblastic leukemia .because they reduced MTHFR enzyme activity, leading to enhanced availability of 5,10-methylenetetrahydrofolate in the DNA synthesis pathway and reduced uracil misincorporation into DNA (Skibola et al., 1999).

The human MTHFR gene contains 11 exons, located on chromosome 1p36.3, and encodes methylenetetrahydrofolate reductase (MTHFR) a key enzyme in folate and homocysteine metabolism. MTHFR catalyzes the biologically irreversible reduction of 5,10-methylenetetrahydrofolate to 5-methyltetrahydrofolate, which provides the methyl group for the remethylation of homocysteine to methionine (Bailey et al., 1999). In the MTHFR enzyme, several

*[1]Lab of Molecular Genetics of Aging & Tumor, Faculty of Life Science and Technology, [2]Lab of Molecular Genetics of Aging & Tumor, Medical Faculty, [3]Lab of Molecular Genetics of Aging & Tumor, Faculty of Life Science and Technology & Faculty of Environmental Science and Engineering, Kunming University of Science and Technology, Kunming, Yunnan, China *For correspondence: yingluo@kmust.edu.cn, twr@sina.com*

**Table 1. The Distribution of the MTHFR C677T Variant for Cases and Controls**

| Author, year | Case | | | Control | | | Ethnicity | Country | $P^a$ |
|---|---|---|---|---|---|---|---|---|---|
| | CC | CT | TT | CC | CT | TT | | | |
| Azhar et al., 2012 | 35 | 31 | 6 | 65 | 34 | 10 | Other | Iran | 0.089 |
| Chan et al., 2011 | 140 | 43 | 2 | 122 | 51 | 4 | Asian | Indonesia | 0.620 |
| Yang et al., 2011 | 96 | 180 | 96 | 84 | 168 | 115 | Asian | china | 0.136 |
| Sood et al., 2010 | 54 | 38 | 3 | 173 | 71 | 11 | other | India | 0.290 |
| Yeoh et al., 2010 | 184 | 111 | 23 | 163 | 150 | 32 | Asian | Singapore | 0.765 |
| Lightfoot et al., 2010 | 374 | 341 | 90 | 359 | 314 | 84 | Caucasian | UK | 0.223 |
| Damnjanovic et al., 2010 | 45 | 28 | 5 | 163 | 190 | 59 | Caucasian | Serbia | 0.762 |
| Tong et al., 2010 | 135 | 192 | 34 | 173 | 257 | 73 | Asian | China | 0.152 |
| Lv et al., 2010 | 38 | 65 | 24 | 72 | 83 | 27 | Asian | China | 0.700 |
| Jonge et al., 2009 | 130 | 93 | 22 | 219 | 223 | 54 | Caucasian | New Zealand | 0.805 |
| Kim et al., 2009 | 29 | 51 | 27 | 540 | 863 | 297 | Asian | Korea | 0.133 |
| Kantar et al., 2009 | 8 | 9 | 3 | 11 | 5 | 1 | other | Turkey | 0.679 |
| Alcasabas et al., 2008 | 145 | 41 | 3 | 322 | 66 | 6 | Asian | Philippines | 0.227 |
| Liu et al., 2008 | 34 | 23 | 26 | 38 | 36 | 9 | Asian | China | 0.914 |
| Giovannetti et al., 2008 | 51 | 11 | 3 | 26 | 6 | 0 | Asian | Indonesia | 0.558 |
| Giovannetti et al., 2008 | 26 | 6 | 0 | 51 | 11 | 3 | Asian | Indonesia | 0.039 |
| Giovannetti et al., 2008 | 224 | 234 | 45 | 47 | 31 | 8 | Caucasian | Surabaya | 0.391 |
| Kamel et al., 2007 | 39 | 42 | 7 | 156 | 135 | 20 | other | Egypt | 0.195 |
| Petra et al., 2007 | 30 | 33 | 5 | 112 | 110 | 36 | Caucasian | Slovenia | 0.287 |
| Oh et al., 2007 | 49 | 55 | 14 | 138 | 229 | 60 | Asian | Korea | 0.023 |
| Kim et al., 2006 | 17 | 38 | 11 | 24 | 55 | 21 | Asian | Korea | 0.313 |
| Reddy et al., 2006 | 51 | 77 | 7 | 79 | 58 | 5 | other | India | 0.148 |
| Yu, 2006 | 30 | 14 | 7 | 20 | 23 | 10 | Asian | China | 0.466 |
| Zanrosso et al., 2006 | 43 | 35 | 8 | 59 | 50 | 10 | mixed | Brazil | 0.897 |
| Zanrosso et al., 2006 | 53 | 21 | 5 | 37 | 32 | 10 | mixed | Brazil | 0.462 |
| Chatzidakis et al., 2006 | 31 | 18 | 3 | 32 | 47 | 9 | Caucasian | Greece | 0.169 |
| Oliveira et al., 2005) | 48 | 50 | 5 | 45 | 57 | 9 | Caucasian | Portugal | 0.120 |
| Schnakenberg et al., 2005 | 195 | 201 | 47 | 184 | 152 | 43 | Caucasian | Germany | 0.179 |
| Thirumaran et al., 2005 | 199 | 195 | 59 | 600 | 681 | 167 | Caucasian | Germany | 0.210 |
| Gemmati et al., 2004 | 52 | 53 | 9 | 78 | 128 | 51 | Caucasian | Italy | 0.908 |
| Balta et al., 2003 | 71 | 60 | 11 | 90 | 87 | 8 | other | Turkey | 0.020 |
| Krajinovic et al., 2004 | 112 | 127 | 31 | 126 | 128 | 46 | Caucasian | Canada | 0.159 |
| Jiang et al., 2004 | 15 | 14 | 0 | 18 | 41 | 8 | Asian | China | 0.039 |
| Deligezer et al., 2003 | 27 | 31 | 4 | 74 | 73 | 14 | other | Turkey | 0.501 |
| Franco et al., 2001 | 36 | 28 | 6 | 22 | 36 | 13 | mixed | Brazil | 0.796 |
| Wiemels et al., 2001 | 98 | 91 | 27 | 89 | 79 | 32 | Caucasian | UK | 0.047 |
| Skibola et al., 1999 | 35 | 29 | 5 | 61 | 39 | 14 | Caucasian | England | 0.061 |

[a]*p* value for Hardy–Weinberg equilibrium in control group

single nucleotide polymorphisms including the two most important, C677T and A1298C, can affect folate and total homocysteine (tHcy) status. The MTHFR C677T, which involves a cytosine (C) to a thymine (T) substitution at position 677, changes an alanine to a valine in the enzyme. The C677T increases thermolability of MTHFR and causes impaired folate binding and reduced activity of the MTHFR enzyme (Frosst et al., 1995). MTHFR C677T is associated with decreased concentrations of folate in serum, plasma, and red blood cells , and mildly increased plasma total homocysteine (tHcy) concentration (Frosst et al., 1995).Base on its biological functions, MTHFR C677T can be seen as a candidate gene for Acute lymphoblastic leukaemia. Accumulating studies have investigated the association between this polymorphism and Acute lymphoblastic leukaemia. However, the results were inconsistent. Therefore, we conducted a meta-analysis to quantitatively assess the effect of the MTHFRC677T polymorphism on the risk of Acute lymphoblastic leukaemia.

## Materials and Methods

*Publication Search*

We searched the PubMed and Chinese biomedicine databases for all articles on the association between MTHFR C677T and acute lymphoblastic leukemia risk (last search update, march 1, 2013). The following key words were used : "MTHFR", "polymorphism" and "Acute lymphoblastic leukaemia" or "leuki". Case-control studies containing available genotype frequencies of C677T were chosen. Of the studies with overlapping data published by the same author, only the most recent or complete study was included in this meta-analysis.

*Statistic analysis*

For control group of each study, the observed genotype frequencies of the MTHFR C677T polymorphism were evaluated for Hardy Weinberg-Equilibrium (HWE) using the $\chi^2$ test. The strength of association between MTHFR C677T gene and Acute lymphoblastic leukaemia was

**Table 2. ORs and 95% CI for ALL and the MTHFR C677T Polymorphism under Different Genetic Models**

| Genetice model (population) | pooled OR [95% CI] p | Heterogeneity p-value | Publication Bias p-value | |
|---|---|---|---|---|
| | | | Begg | Egger |
| Additive (T vs. C) | | | | |
| Caucasian | 0.715[0.655~0.781]0.000 | 0.260 | 0.493 | 0.218 |
| Asian | 0.711[0.591~1.005]0.055 | 0.000 | 0.392 | 0.580 |
| Others | 0.913[0.656~1.271]0.590 | 0.000 | 0.061 | 0.275 |
| overall | 0.776[0.687~0.877]0.000 | 0.000 | 0.072 | 0.093 |
| Dominant (T-carriers vs.CC) | | | | |
| Caucasian | 0.863[0.718~1.036]0.114 | 0.001 | 0.273 | 0.197 |
| Asian | 0.939[0.769~1.148]0.540 | 0.025 | 0.815 | 0.584 |
| Others | 1.122[0.776~1.622]0.541 | 0.001 | 0.297 | 0.574 |
| overall | 0.944[0.828~1.076]0.386 | 0.000 | 0.289 | 0.447 |
| Recessive (TT vs. C-carriers) | | | | |
| Caucasian | 0.794[0.644~0.980]0.031 | 0.143 | 0.131 | 0.002 |
| Asian | 1.009[0.723~1.409]0.956 | 0.005 | 0.392 | 0.464 |
| Others | 0.861[0.593~1.251]0.433 | 0.648 | 0.532 | 0.564 |

accessed by calculating crude odds ratios (ORs) and 95% confidence intervals (CIs). The pooled ORs were performed for additive genetic model (T vs. C), dominant model (TT+CT vs. CC), and recessive model (TT vs. CT+ CC) respectively. Heterogeneity assumption was evaluated by a chi-square based Q-test. A P-value of <0.05 for the Q-test indicated heterogeneity among the studies, the summary OR estimate of each study was calculated by the random effects model (DerSimonian et al., 1986; Mantel et al., 1959). The potential for publication bias was examined by a Begg's test (funnel plot method) and Egger's linear regression test ($P$<0.05 considered representative of statistical significance) (Egger et al., 1997). All analyses were performed using Stata software (version 8.2; Stata Corporation, College Station, TX).

## Results

*Eligible studies*

We identified a total of 37 relevant publications that association between MTHFR C677T and Acute lymphoblastic leukaemia, including 6371 Acute lymphoblastic leukaemia cases and 10850 controls in our meta-analysis (Table 1). Since C677T genotypes in the control group by Giovannetti (Giovannetti et al., 2008), Oh (Oh et al., 2007), Balta (Balta et al., 2003), Jiang (Jiang et al., 2004), Wiemels (Wiemels et al., 2001), were not in HWE, these data (537 cases and 944 controls) were excluded from our meta-analysis. Therefore, our final data pooling consisted of 32 publications, including 5834 cases and 9906 controls.

*Meta-analysis*

Differences in allelic distribution by ethnicity could be partially responsible for the observed differences in the association between MTHFR C677T and acute lymphoblastic leukemia. The results of the association between the MTHFR C677T polymorphism and Acute lymphoblastic leukaemia and the heterogeneity test are shown in Table 2. The association was most pronounced for carriers of the T allelic gene (additive model: R= 0.776, 95% CI: 0.687~0.877, $p$ = 0.000). The positive association was driven by a Caucasian additive model: (OR= 0.715, 95% CI: 0.655~0.781, $p$ = 0.000, $P$ = 0.260 for heterogeneity, Figure 1). No significant association was found in Asian (OR= 0.711, 95% CI: 0.591~1.005, $p$= 0.055) and other (OR=0. 913, 95% CI: 0.656~1.271, $p$= 0.590).

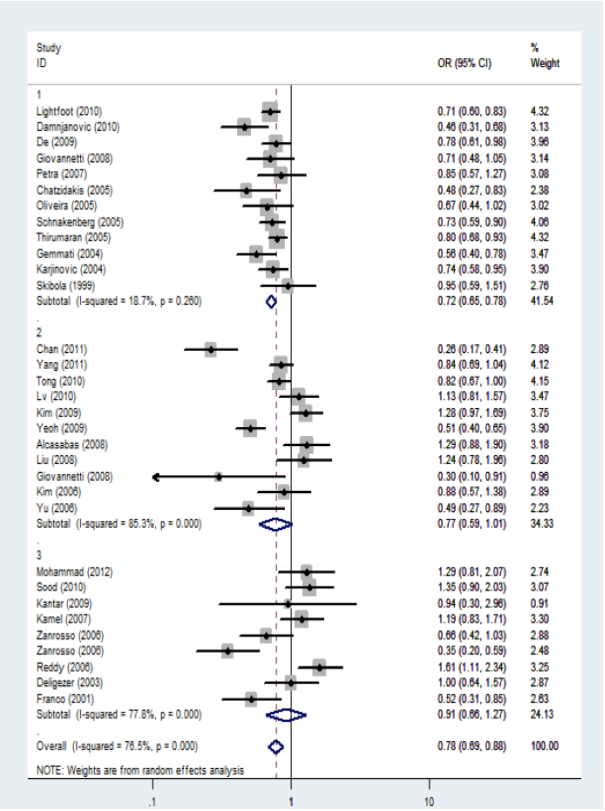


**Figure 1. Forest Plot of ORs of Recurrent Pregnancy Loss for T Allele When Compared to the C Allele.** The squares and horizontal lines correspond to the study-specific OR and 95% CI. The area of the squares reflects the study-specific weight. The diamond represents the pooled OR and 95% CI

*Publication bias*

Funnel plot and Egger's test were performed to quantitatively evaluate the publication bias of literatures on Acute lymphoblastic leukaemia. The results of Egger's test provided statistical evidence for funnel plot symmetry ($P$=0.285) in overall results, suggesting the absence of publication bias.

## Discussion

Acute lymphoblastic leukaemia, an important clinical problem, has been well-studied but the mechanism of ALL is still relatively unclear. Single nucleotide polymorphisms (SNPs) can be used as a implement in investigating genetic variations and disease susceptibility. ALL is speculated to be associated with inherited thrombophilias that encompass diverse conditions including the thermolabile variant of the MTHFR (Jilma et al., 2003). Methylenetetrahydrofolate reductase is an enzyme in homocysteine metabolism. The MTHFR C677T, which is found within the enzyme catalytic domain, result in both a thermolabile protein and increased tHcy. Through its effect

on tHcy levels, MTHFR C677T has been implicated as a risk factor in the pathogenesis of Acute lymphoblastic leukaemia.

The frequency of homozygous TT genotypes was found significantly higher among the childhood with Acute lymphoblastic leukaemia than the other two groups indicated that MTHFR C677T polymorphism would be expected to play a major role to bring about ALL. Some studies reported significantly decreased prevalence of MTHFR C677T among cases in distinct ethnicity (Wang et al., 2012; Yan et al., 2012). In contrast, other studies reported an insignificant association between the MTHFR C677T and ALL (Li et al., 2011; Wang et al., 2010). The variances in ethnicity may be one major reason for the controversy. A meta-analysis summarizing studies on association between the MTHFR C677T polymorphism and Acute lymphoblastic leukaemia until march 2011 found the TT genotype marginally significantly associated with reduced ALL risk in Children. However, five new studies have been published increasing the number of Acute lymphoblastic leukaemia and controls with genetic information. We excluded five studies in this meta-analysis after evaluating the articles in their entirety.

The summary OR from our meta-analyses revealed that individuals with the T allelic gene had decreased risk of Acute lymphoblastic leukaemia (p= 0.000), especially in Caucasian (p= 0.000). However, our data do not support the association for Asian (p= 0.055).

The TT genotype of C677T polymorphism in MTHFR gene was associated with elevated plasma homocysteine level, increased risk of arterial stiffness (Trabetti 2008). Women with elevated total homocysteine concentrations showed a significant association with defective chorionic villous vascularization (Nelen et al., 2000). independently of hyperhomocysteinemia, was due to interference with red blood cell folate metabolism, as has been suggested (Golbahar et al., 2005). It is also possible that association of the MTHFR 677TT genotype with Acute lymphoblastic leukaemia. In the Govindaiah study, MTHFR genotype impacts more prominent in men than women (Govindaiah et al., 2009). However, the mechanism by which homocysteine causes DNA damage needs to be elucidated further.

There are limitations that are present in this analysis, which mainly relate to the lack of other risk factors between the subjects in the available studies. In these cases, few investigators reported results from subgroup analysis for other risk factors, such as age, environmental pollution, sexuality and so on; therefore, the association in these factors could not be assessed. There is a need for larger and wider case-control studies to explore the role of other factors that are likely to cause Acute lymphoblastic leukaemia.

This is a meta-analysis with sufficient individual data to stratify results by both ethnicity and the number of Acute lymphoblastic leukaemia, including a total of 5834 cases and 9906 controls from 32 independent publications, we examined the association between C677T of the MTHFR gene and Acute lymphoblastic leukaemia risk. We demonstrated that the T allele of the C677T polymorphism was associated with a significantly reduced risk of Acute lymphoblastic leukaemia on Caucasians, whereas did not appear to have an effect in both Asians and Others. Future well designed large studies might be necessary to validate this association in different populations incorporated with environmental factors in the susceptibility of Acute lymphoblastic leukaemia.

## Acknowledgements

This work was financially supported by the Science and Technology Innovation Project of Yunnan Province (No. 2011DH011). The author (s) declare that they have no competing interests.

## References

Alcasabas P, Ravindranath Y, Goyette G, et al (2008). 5, 10-methylenetetrahydrofolate reductase (MTHFR) polymorphisms and the risk of acute lymphoblastic leukemia (ALL) in Filipino children. *Pediatr Blood Cancer*, **51**, 178-82.

Azhar MR, Rahimi Z, Vaisi-Raygani A, et al (2012). Lack of association between MTHFR C677T and A1298C polymorphisms and risk of childhood acute lymphoblastic leukemia in the Kurdish population from Western Iran. *Genet Test Mol Biomarkers*, **16**, 198-202.

Bailey LB, Gregory JF, 3rd (1999). Folate metabolism and requirements. *J Nutr*, **129**, 779-82.

Balta G, Yuksek N, Ozyurek E, et al (2003). Characterization of MTHFR, GSTM1, GSTT1, GSTP1, and CYP1A1 genotypes in childhood acute leukemia. *Am J Hematol*, **73**, 154-60.

Chan JY, Ugrasena DG, Lum DW, et al (2011). Xenobiotic and folate pathway gene polymorphisms and risk of childhood acute lymphoblastic leukaemia in Javanese children. *Hematol Oncol*, **29**, 116-23.

Chatzidakis K, Goulas A, Athanassiadou-Piperopoulou F, et al (2006). Methylenetetrahydrofolate reductase C677T polymorphism: association with risk for childhood acute lymphoblastic leukemia and response during the initial phase of chemotherapy in greek patients. *Pediatr Blood Cancer*, **47**, 147-51.

Damnjanovic T, Milicevic R, Novkovic T, et al (2010). Association between the methylenetetrahydrofolate reductase polymorphisms and risk of acute lymphoblastic leukemia in Serbian children. *J Pediatr Hematol Oncol*, **32**, e148-50.

de Jonge R, Tissing WJ, Hooijberg JH, et al (2009). Polymorphisms in folate-related genes and risk of pediatric acute lymphoblastic leukemia. *Blood*, **113**, 2284-9.

Deligezer U, Akisik E, Dalay N (2003). Genotyping of the MTHFR gene polymorphism, C677T in patients with leukemia by melting curve analysis. *Mol Diagn*, **7**, 181-5.

DerSimonian R, Laird N (1986). Meta-analysis in clinical trials. *Control Clin Trials*, **7**, 177-88.

Egger M, Davey Smith G, Schneider M, Minder C (1997). Bias in meta-analysis detected by a simple, graphical test. *BMJ*, **315**, 629-34.

Franco RF, Simoes BP, Tone LG, et al (2001). The methylenetetrahydrofolate reductase C677T gene polymorphism decreases the risk of childhood acute lymphocytic leukaemia. *Br J Haematol*, **115**, 616-8.

Frosst P, Blom HJ, Milos R, et al (1995). A candidate genetic risk factor for vascular disease: a common mutation in methylenetetrahydrofolate reductase. *Nat Genet*, **10**, 111-3.

Gemmati D, Ongaro A, Scapoli G L, et al (2004). Common gene polymorphisms in the metabolic folate and methylation

pathway and the risk of acute lymphoblastic leukemia and non-Hodgkin's lymphoma in adults. *Cancer Epidemiol Biomarkers Prev*, **13**, 787-94.

Giovannetti E, Ugrasena DG, Supriyadi E, et al (2008). Methylenetetrahydrofolate reductase (MTHFR) C677T and thymidylate synthase promoter (TSER) polymorphisms in Indonesian children with and without leukemia. *Leuk Res*, **32**, 19-24.

Golbahar J, Aminzadeh MA, Sharifkazemi MB, Rezaian GR (2005). Association of red blood cell 5-methyltetrahydrofolate and severity of coronary artery disease: a cross-sectional study from Shiraz, southern Iran. *Heart Vessels*, **20**, 203-6.

Govindaiah V, Naushad SM, Prabhakara K, et al (2009). Association of parental hyperhomocysteinemia and C677T Methylene tetrahydrofolate reductase (MTHFR) polymorphism with recurrent pregnancy loss. *Clin Biochem*, **42**, 380-6.

Jiang H, Gu LJ, Xue HL (2004). Methylenetetrahydrofolate reductase gene polymorphism of childhood acute lymphocytic leukaemia. *Zhonghua Xue Ye Xue Za Zhi*, **25**, 439-40.

Jilma B, Kamath S, and Lip GY (2003). ABC of antithrombotic therapy: Antithrombotic therapy in special circumstances. II-In children, thrombophilia, and miscellaneous conditions. *BMJ*, **326**, 93-6.

Kamel AM, Moussa HS, Ebid GT, et al (2007). Synergistic effect of methyltetrahydrofolate reductase (MTHFR) C677T and A1298C polymorphism as risk modifiers of pediatric acute lymphoblastic leukemia. *J Egypt Natl Canc Inst*, **19**, 96-105.

Kantar M, Kosova B, Cetingul N, et al (2009). Methylenetetrahydrofolate reductase C677T and A1298C gene polymorphisms and therapy-related toxicity in children treated for acute lymphoblastic leukemia and non-Hodgkin lymphoma. *Leuk Lymphoma*, **50**, 912-7.

Karathanasis NV, Stiakaki E, Goulielmos GN, and Kalmanti M (2011). The role of the methylenetetrahydrofolate reductase 677 and 1298 polymorphisms in Cretan children with acute lymphoblastic leukemia. *Genet Test Mol Biomarkers*, **15**, 5-10.

Kim HN, Kim YK, Lee IK, et al (2009). Association between polymorphisms of folate-metabolizing enzymes and hematological malignancies. *Leuk Res*, **33**, 82-7.

Kim NK, Chong SY, Jang MJ, et al (2006). Association of the methylenetetrahydrofolate reductase polymorphism in Korean patients with childhood acute lymphoblastic leukemia. *Anticancer Res*, **26**, 2879-81.

Krajinovic M, Labuda D, Richer C, et al (1999). Susceptibility to childhood acute lymphoblastic leukemia: influence of CYP1A1, CYP2D6, GSTM1, and GSTT1 genetic polymorphisms. *Blood*, **93**, 1496-501.

Krajinovic M, Lamothe S, Labuda D, et al (2004). Role of MTHFR genetic polymorphisms in the susceptibility to childhood acute lymphoblastic leukemia. *Blood*, **103**, 252-7.

Li X L, Yu F, Zhang Y, et al (2011). [Study on the association between 5, 10-methylenetrahydrofolate reductase C677T polymorphism and acute lymphoblastic leukemia risk: a Meta-analysis]. *Zhonghua Liu Xing Bing Xue Za Zhi*, **32**, 1030-6.

Lightfoot TJ, Johnston WT, Painter D, et al (2010). Genetic variation in the folate metabolic pathway and risk of childhood leukemia. *Blood*, **115**, 3923-9.

Liu JX, Chen JP, Tan W, Lin D X (2008). [Association between mthfr gene polymorphisms and toxicity of HDMTX chemotherapy in acute lymphocytic leukemia]. *Zhongguo Shi Yan Xue Ye Xue Za Zhi*, **16**, 488-92.

Lv L, Wu C, Sun H, et al (2010). Combined 677CC/1298AC genotypes of methylenetetrahydrofolate reductase (MTHFR) reduce susceptibility to precursor B lymphoblastic leukemia in a Chinese population. *Eur J Haematol*, **84**, 506-12.

Mantel N, Haenszel W (1959). Statistical aspects of the analysis of data from retrospective studies of disease. *J Natl Cancer Inst*, **22**, 719-48.

McNeer JL (2011). The complex interplay between folate metabolism and risk of acute lymphoblastic leukemia. *Leuk Lymphoma*, **52**, 1621-2.

Nelen WL, Bulten J, Steegers EA, et al (2000). Maternal homocysteine and chorionic vascularization in recurrent early pregnancy loss. *Hum Reprod*, **15**, 954-60.

Oh D, Kim NK, Jang MJ, et al (2007). Association of the 5, 10-methylenetetrahydrofolate reductase (MTHFR C677T and A1298C) polymorphisms in Korean patients with adult acute lymphoblastic leukemia. *Anticancer Res*, **27**, 3419-24.

Oliveira E, Alves S, Quental S, et al (2005). The MTHFR C677T and A1298C polymorphisms and susceptibility to childhood acute lymphoblastic leukemia in Portugal. *J Pediatr Hematol Oncol*, **27**, 425-9.

Petra BG, Janez J, Vita D (2007). Gene-gene interactions in the folate metabolic pathway influence the risk for acute lymphoblastic leukemia in children. *Leuk Lymphoma*, **48**, 786-92.

Reddy H, Jamil K (2006). Polymorphisms in the MTHFR gene and their possible association with susceptibility to childhood acute lymphocytic leukemia in an Indian population. *Leuk Lymphoma*, **47**, 1333-9.

Robien K, Ulrich C M (2003). 5, 10-Methylenetetrahydrofolate reductase polymorphisms and leukemia risk: a HuGE minireview. *Am J Epidemiol*, **157**, 571-82.

Scelo G, Metayer C, Zhang L, et al (2009). Household exposure to paint and petroleum solvents, chromosomal translocations, and the risk of childhood leukemia. *Environ Health Perspect*, **117**, 133-9.

Schnakenberg E, Mehles A, Cario G, et al (2005). Polymorphisms of methylenetetrahydrofolate reductase (MTHFR) and susceptibility to pediatric acute lymphoblastic leukemia in a German study population. *BMC Med Genet*, **6**, 23.

Skibola CF, Smith MT, Kane E, et al (1999). Polymorphisms in the methylenetetrahydrofolate reductase gene are associated with susceptibility to acute leukemia in adults. *Proc Natl Acad Sci U S A*, **96**, 12810-5.

Sood S, Das R, Trehan A, et al (2010). Methylenetetrahydrofolate reductase gene polymorphisms: association with risk for pediatric acute lymphoblastic leukemia in north Indians. *Leuk Lymphoma*, **51**, 928-32.

Thirumaran RK, Gast A, Flohr T, et al (2005). MTHFR genetic polymorphisms and susceptibility to childhood acute lymphoblastic leukemia. *Blood*, **106**, 2590-1; author reply 1-2.

Tong N, Fang Y, Li J, et al (2010). Methylenetetrahydrofolate reductase polymorphisms, serum methylenetetrahydrofolate reductase levels, and risk of childhood acute lymphoblastic leukemia in a Chinese population. *Cancer Sci*, **101**, 782-6.

Trabetti E (2008). Homocysteine, MTHFR gene polymorphisms, and cardio-cerebrovascular risk. *J Appl Genet,* **49**, 267-82.

Wang H, Meng L, Zhao L, et al (2012). Methylenetetrahydrofolate reductase polymorphism C677T is a protective factor for pediatric acute lymphoblastic leukemia in the Chinese population: a meta-analysis. *Genet Test Mol Biomarkers*, **16**, 1401-7.

Wang J, Zhan P, Chen B, et al (2010). MTHFR C677T polymorphisms and childhood acute lymphoblastic leukemia: a meta-analysis. *Leuk Res*, **34**, 1596-600.

Wiemels JL, Smith RN, Taylor GM, et al (2001). Methylenetetrahydrofolate reductase (MTHFR) polymorphisms and risk of molecularly defined subtypes

of childhood acute leukemia. *Proc Natl Acad Sci U S A*, **98**, 4004-9.

Yan J, Yin M, Dreyer ZE, et al (2012). A meta-analysis of MTHFR C677T and A1298C polymorphisms and risk of acute lymphoblastic leukemia in children. *Pediatr Blood Cancer*, **58**, 513-8.

Yang L, Liu L, Wang J, et al (2011). Polymorphisms in folate-related genes: impact on risk of adult acute lymphoblastic leukemia rather than pediatric in Han Chinese. *Leuk Lymphoma*, **52**, 1770-6.

Yeoh AE, Lu Y, Chan JY, et al (2010). Genetic susceptibility to childhood acute lymphoblastic leukemia shows protection in Malay boys: results from the Malaysia-Singapore ALL Study Group. *Leuk Res*, **34**, 276-83.

Yu H, Jin RM, Bai Y, et al (2006). The relationship between the methylenetetrahydrofolate reducase C677T gene polymorphism and acute lymphocytic leukemia in childrn. *J Clin Hematol*, **19**, 205-6, 9.

Zanrosso CW, Hatagima A, Emerenciano M, et al (2006). The role of methylenetetrahydrofolate reductase in acute lymphoblastic leukemia in a Brazilian mixed population. *Leuk Res*, **30**, 477-81.