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*Original Article*

# Methylenetetrahydrofolate reductase C677T polymorphism and susceptibility to lung cancer as well as response to chemotherapy: a meta-analysis based on 31 studies

Wenfang Tang[1*], Ranwei Li[2*], Jingjin Tan[1], Yaqin Yu[3], Lijing Zhao[4], Xingxing Li[1], Shan Jiang[1], Rihui Liu[3], Ke Wang[1]

*[1]Department of Respiratory Medicine, Second Affiliated Hospital of Jilin University, Changchun 130041, Jilin, China; [2]Department of Urinary Surgery, The Second Affiliated Hospital of Jilin University, Changchun 130041, Jilin, China; [3]Department of Epidemiology and Health Statistics, School of Public Health, Jilin University, Changchun 130041, Jilin, China; [4]Department of Pathophysiology, College of Basic Medicine, Jilin University, Changchun 130021, Jilin, China. [*]Equal contributors.*

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**Abstract:** Background: Conflicting results have been reported regarding the correlation of the methylenetetrahydrofolate reductase (MTHFR) C677T polymorphism and the risk of lung cancer as well as response to chemotherapy. Objective: To estimate the correlations of the MTHFR C677T polymorphism and risk of lung cancer as well as response to chemotherapy by conducting a meta-analysis. Methods: A comprehensive literature search was conducted in PubMed, Embase, Web of Science, CKNI, VIP, and Wanfang database through January 2015. Only case-control studies evaluating the MTHFR C677T polymorphism and lung cancer risk or response to chemotherapy were eligible. Results: Thirty-one case-control studies involving 12,878 cases of lung cancer and 12,447 controls were analyzed. The MTHFR C677T gene polymorphism was associated with lung cancer susceptibility in three genetic models (TT vs. CT+CC: odds ratio [OR] = 1.250, 95% confidence interval [CI] 1.076-1.452; TT vs. CC: OR = 1.324, 95% CI 1.095-1.601; and T vs. C: OR = 1.126, 95% CI 1.024-1.239). Stratified analyses indicated significantly increased lung cancer risks were consistently indicated in Asian patients and for non-small cell lung cancer (NSCLC) but not in European patients or for small cell lung cancer. Patients carrying the TT+CT genotype had a 47.9% lower response rate (OR = 0.521, 95% CI 0.361-0.753) to chemotherapy than the CC allele carriers. However, the differences were not significant for the other genetic models. Conclusions: The MTHFR C677T polymorphism is associated with an increased risk of lung cancer in Asian patients and with the NSCLC subtype. Moreover, the MTHFR 677T mutation might lower the response to chemotherapy.

**Keywords:** Methylenetetrahydrofolate reductase, gene polymorphism, lung cancer, chemotherapy, meta-analysis

## Introduction

Lung cancer is the leading cause of cancer-related mortality, with approximately 160,000 newly diagnosed cases and 130,000 deaths occurring annually worldwide [1]. In China, lung cancer has become the most frequent malignant tumor and the leading cause of cancer death [2]. Lung cancer is generally divided into non-small-cell lung cancer (NSCLC) and small-cell lung cancer according to its pathologic types. However, the underlying mechanisms of lung cancer are not fully understood. As lung cancer is a multi-factorial disease, genetic factors contribute greater to the occurrence of lung cancer [3]. Therefore, identification of possible genetic factors may help identify individuals who are at increased risk for lung cancer.

Folate metabolism plays an important role in carcinogenesis by involving both DNA methylation and repair. Methylenetetrahydrofolate reductase (MTHFR) is a crucial regulatory enzyme in folate metabolism that irreversibly converts 5,10-methylenetetrahydrofolate to 5-methyltetrahydrofolate [4]. The MTHFR gene is lo-

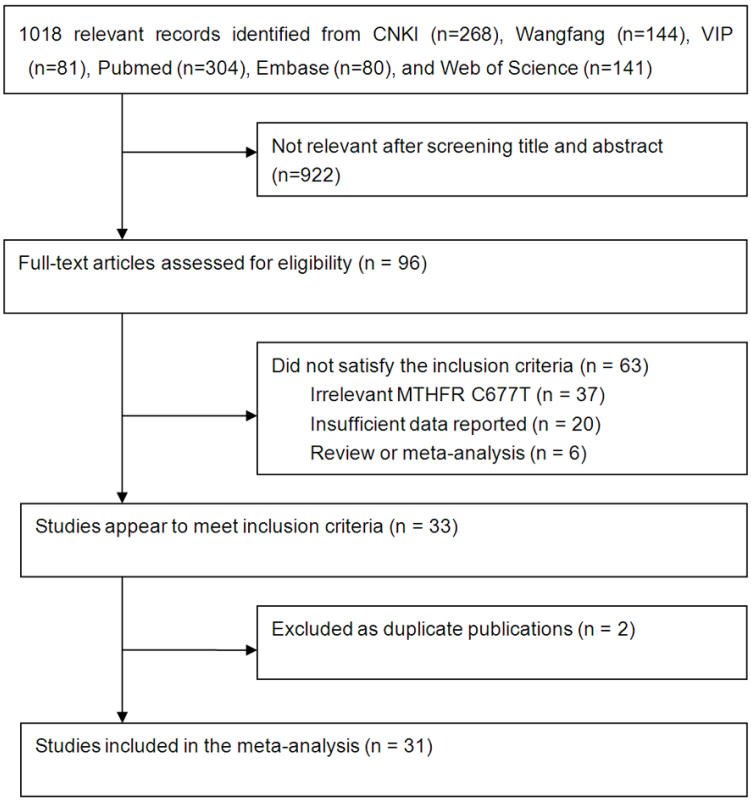


**Figure 1.** Flowchart of study selection process.

cated at the end of the short arm of chromosome 1 (1p36.3) [5]. The most common mutation in the MTHFR gene is the C to T transition at nucleotide 677, which results in an amino acid sequence change of an alanine to valine and thereby affects enzyme activity [6]. The MTHFR 677C>T variant is associated with a significant decrease in plasma levels of folate, which likely induces and accelerates lung cancer [7].

The relationship between the MTHFR C677T gene polymorphism and susceptibility to lung cancer has been widely investigated in many studies, but with conflicting findings [8]. Small sample sizes, diverse ethnic origins, different dietary habits, different characteristics of controls, and varying methods of genotyping may partly explain the discrepancies. Several meta-analyses of the relationship between the MTHFR 677T genotype and susceptibility to lung cancer have been reported. The first meta-analysis by Mao et al [9] was based on eight studies and suggested no evidence for a major role of the MTHFR C677T gene polymorphism in carcinogenesis of lung cancer. Similarly, the next two meta-analyses [10, 11] supported that the MTHFR C677T polymorphism was not a susceptibility gene for lung cancer. However, an updated meta-analysis by Hou et al [12] concluded that the MTHFR 677T null genotype increased the NSCLC risk. A more recently published meta-analysis indicated that the MTHFR C677T polymorphism is associated with an increased risk of lung cancer in Asians [13], particularly in Chinese patients [14]. Chemotherapy is one of the principal methods for treating lung cancer. However, drug resistance remains a serious problem experienced by lung cancer patients. The MTHFR C677T gene polymorphism has been investigated as a determinant for the chemotherapy response in lung cancer patients with inconsistent findings. The recent publication of new studies instigated our interest in conducting an updated meta-analysis using all of the available accumulated clinical evidence.

Here, we conducted a comprehensive, updated meta-analysis of all the relevant case-control studies to investigate the association of the MTHFR C677T gene polymorphism with the risk of lung cancer as well as response to chemotherapy.

## Methods

### *Search strategy*

We conducted a comprehensive literature search of PubMed, Embase, Web of Science, China National Knowledge Infrastructure, VIP, and Wanfang database from their inception to January 2015. The search keywords were methylenetetrahydrofolate reductase OR MTHFR AND lung cancer OR lung carcinoma OR lung neoplasm AND polymorphism OR variant OR mutation OR genetic susceptibility AND platinum AND chemotherapy. The literature search was restricted to papers published in

**Table 1.** Basic characteristic of the included studies analyzing lung cancer susceptibility

| Study | Study region | Source of control | Score | HWE | No. lung cancer/Control | Lung cancer group | | | Control group | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | | CC | CT | TT | CC | CT | TT |
| Jeng YL 2003 [18] | Taiwan | Hospital | 7 | Yes | 59/232 | 36 | 22 | 1 | 123 | 95 | 14 |
| Zhang XM 2005 [19] | Mainland China | Healthy | 7 | Yes | 505/500 | 120 | 230 | 155 | 160 | 231 | 109 |
| Shen M 2005 [20] | Mainland China | Healthy | 7 | Yes | 116/111 | 33 | 65 | 18 | 53 | 42 | 16 |
| Zou QF 2006 [21] | Mainland China[#,*] | Healthy | 8 | - | 100/100 | 24 | 52 | 24 | 39 | 48 | 13 |
| Suzuki T 2007 [22] | Japan | Hospital | 7 | Yes | 515/1030 | 182 | 256 | 77 | 379 | 474 | 177 |
| Liu HL 2008 [23] | Mainland China | Hospital | 6 | Yes | 500/517 | 157 | 245 | 98 | 149 | 265 | 103 |
| Liu CS 2009 [24] | Taiwan | Healthy | 7 | Yes | 358/716 | 205 | 124 | 29 | 362 | 291 | 63 |
| Yao QF 2010 [25] | Mainland China | Healthy | 8 | Yes | 93/106 | 27 | 46 | 20 | 36 | 51 | 19 |
| Yang XX 2010 [26] | Mainland China | Healthy | 7 | Yes | 120/165 | 49 | 52 | 19 | 62 | 75 | 28 |
| Ma QL 2012 [27] | Mainland China[*] | Hospital | 7 | Yes | 120/60 | 20 | 54 | 46 | 22 | 28 | 10 |
| Cheng Z 2011 [28] | Mainland China[*] | Healthy | 7 | Yes | 178/180 | 49 | 58 | 71 | 47 | 88 | 45 |
| Cui LH 2011 [29] | Korea | Healthy | 8 | Yes | 3938/1700 | 1361 | 1909 | 668 | 540 | 862 | 298 |
| Kiyohara C 2011 [30] | Japan | Hospital | 7 | - | 462/379 | 153 | 201 | 108 | 158 | 170 | 51 |
| Cui LH 2011 [31] | Mainland China[*,^] | Healthy | 6 | Yes | 438/641 | 58 | 240 | 140 | 121 | 325 | 195 |
| Cai ZX 2014 [32] | Mainland China | Healthy | 7 | Yes | 202/202 | 54 | 102 | 46 | 69 | 112 | 21 |
| Shen H 2001 [33] | USA | Healthy | 8 | Yes | 550/554 | 241 | 252 | 57 | 245 | 252 | 57 |
| Siemianowicz K 2003 [34] | Poland[#] | Healthy | 8 | Yes | 146/44 | 38 | 60 | 48 | 18 | 20 | 6 |
| Heijmans BT 2003 [35] | Poland | Healthy | 9 | Yes | 44/793 | 23 | 17 | 4 | 399 | 329 | 65 |
| Shi QL 2005 [36] | USA | Healthy | 9 | Yes | 1051/1141 | 483 | 468 | 100 | 498 | 519 | 124 |
| Hung R 2007 [37] | France | Hospital | 7 | Yes | 2169/2803 | 1009 | 929 | 231 | 1397 | 1147 | 259 |
| Gemignani F 2007 [38] | Europe | Hospital | 7 | Yes | 247/259 | 104 | 107 | 36 | 131 | 103 | 25 |
| Arslan S 2011 [39] | Turkey[#] | Healthy | 8 | Yes | 64/61 | 30 | 27 | 7 | 29 | 29 | 3 |
| Cavic M 2014 [40] | Serbia | Healthy | 7 | - | 55/53 | 34 | 18 | 3 | 13 | 33 | 7 |
| Yilmaz M 2014 [41] | Turkey[#] | Healthy | 8 | Yes | 100/100 | 55 | 38 | 7 | 51 | 43 | 6 |

Abbreviations: NSCLC, Non-small cell lung cancer; SCLC, Small cell lung cancer. Note: [#]Reported SCLC and NSCLC; [*]Reported squamous cell carcinoma and adenocarcinoma; [^]Simulatenously reported data on lung cancer susceptibility and response to chemotherapy.

English and Chinese. In addition, we manually reviewed the reference lists of the articles to identify additional eligible articles.

*Study selection*

The following inclusion criteria were applied: 1) case-control study evaluating the association between the MTHFR C677T gene polymorphism and lung cancer susceptibility; 2) patients had pathologically and cytology confirmed diagnosis and typing of lung cancer; 3) study provided odds ratios (ORs) and 95% confidence intervals (CIs) for genotype distributions or sufficient genotyping data were presented to calculate ORs and 95% CIs; 4) genotype distributions of the control population were in Hardy-Weinberg equilibrium (HWE); and 5) for response to chemotherapy analysis, platinum-based chemotherapy served as the sole therapeutic regimen without any other adjuvant therapy. Patient responses to the therapeutic regimens were divided into four categories: complete response (CR), partial response (PR), stable disease (SD), and progressive disease (PD) according to the World Health Organization (WHO) criteria or the Response Evaluation Criteria in Solid Tumors. For data analysis, patients who showed CR and PR were considered as good responders. A study was excluded if: 1) it investigated the interaction between MTHFR C677T and other factors; 2) the methodological quality of the article was poor or there were obvious data abnormalities; 3) it was a duplicate publication; or 4) if it was an abstract, case report, letter, or review.

*Data extraction*

Two reviewers (WF Tang and XX Li) independently extracted the data using a standardized data extraction form. The following data were extracted from the individual studies: first author's surname, year of publication, country/region, source of controls, number of cases and controls, number of genotypes, HWE of controls, type of histology, treatment protocol, and clinical stage. Where discrepancies were detected, the reviewers resolved these by discussion. The methodological quality of the includ-

**Table 2.** Basic characteristics of the included studies analyzing treatment response to chemotherapy

| Study | Study region | Type of histology | Clinical stage | Treatment protocol | No. lung cancer | No. complete+partial responce | | | No. stable+progressive disease | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | | CC | CT | TT | CC | CT | TT |
| Cui LH 2011 [31] | Mainland China | NSCLC | IIIB-IV | Platinum-based | 101 | 3 | 14 | 14 | 11 | 43 | 16 |
| Shi MQ 2006 [42] | Mainland China | NSCLC | II-IV | Platinum-based | 97 | 12 | 18 | 8 | 21 | 31 | 7 |
| Li WJ 2013 [43] | Mainland China | NSCLC | IIIB-IV | Platinum-based | 45 | 8 | 5 | 2 | 9 | 16 | 5 |
| Hong W 2013 [44] | Mainland China | NSCLC | IIIB-IV | Gemcitabine-based | 135 | 21 | 14 | 4 | 30 | 40 | 26 |
| Dong CM 2014 [45]^ | Mainland China | Lung adenocarcinoma | IIIB-IV | Platinum-based | 92 | 20 | 0 | 2 | 36 | 23 | 11 |
| Kou JY 2014 [46] | Mainland China | NSCLC | IIIB-IV | Gemcitabine-based | 100 | 16 | 13 | 4 | 20 | 31 | 16 |
| Jung M 2013 [47]^ | Korea | Lung adenocarcinoma | - | Platinum-based | 88 | 5 | 9 | 0 | 18 | 37 | 19 |
| Alberola V 2004 [48] | Spain | NSCLC | IIIB-IV | Platinum-based | 191 | 67 | | 11 | 98 | | 15 |

Note: ^Simulatenously reported data on lung cancer susceptibility and response to chemotherapy.

ed studies was assessed using the Newcastle-Ottawa Quality Assessment Scale (NOS) [15]. We excluded studies that had a NOS score less than 5 in the analysis of lung cancer susceptibility.

*Statistical analyses*

All statistical analyses were conducted using the STATA statistical package (version 10, STATA, College Station, TX). We first examined whether the genotype distribution in the control group was in HWE using the chi-square test. The relationship between the MTHFR gene polymorphism and risk of lung cancer was estimated by OR and 95% CI. The OR and corresponding 95% CI were pooled by calculating those for TT versus CT+CC, TT+CT versus CC, TT versus CC, and T versus C. The heterogeneity was tested by the Q-statistic and $I^2$ test. When a $P$-value<0.1 in Q statistic or $I^2$>50%, we pooled the risk estimate using the random-effects model; otherwise, a fixed-effect model was used. Subgroup analyses were conducted based on the type of pathology (squamous cell carcinoma or adenocarcinoma), general histological type (NSCLC or SCLC), and geographical region (Asia or Europe). Publication bias was examined using Begg's test [16] and Egger's test [17], and a $P$-value<0.1 was considered statistically significant.

## Results

After application of our predefined inclusion criteria, a total of 31 case-control studies [18-48] involving 12,878 cases of lung cancer and 12,447 controls were identified. Twenty-four studies reported data on the MTHFR C677T polymorphism [18-41] and lung cancer susceptibility, of which, 8 studies [21, 22, 27, 30, 34, 39, 41] analyzed data according to the type of histology; 4 studies [21, 34, 39, 41] reported data on SCLC and NSCLC separately; and 5 studies [21, 22, 27, 28, 30] reported data on squamous cell carcinoma and adenocarcinoma. Eight studies [30, 42-48] analyzed the MTHFR C677T polymorphism and response to chemotherapy. Controls in seven studies [18, 22, 23, 27, 31, 37, 38] were hospital-based populations, and those in the other studies were healthy populations. A flow chart showing the study selection is presented in **Figure 1**. The basic characteristics of the included studies are listed in **Tables 1** and **2**.

*The MTHFR C677T polymorphism and lung cancer susceptibility*

As shown in **Table 3**, compared with the genotype CT+CC, the genotype TT was associated with an increased risk of lung cancer in the overall analysis (OR = 1.250; 95% CI 1.076-1.452). Subgroup analyses showed that increased lung cancer susceptibility was observed in the Asian population (OR = 1.300, 95% CI 1.065-1.588) and for the NSCLC type (OR = 1.300; 95% CI 1.065-1.588).

As shown in **Table 4**, compared with the genotype CC, the genotype TT+CT was associated with an increased risk of lung cancer in the Asian population (OR = 1.191; 95% CI 1.001-1.416), for the NSCLC type (OR = 1.417; 95% CI 1.015-1.978), and for adenocarcinoma (OR = 1.258; 95% CI 1.005-1.575), which was not found in the overall analysis.

**Table 5** indicates that the genotype TT was associated with an increased risk of lung cancer in the overall analysis (OR = 1.324; 95% CI 1.095-1.601) more than the genotype CC. Subgroup analyses indicated that increased lung cancer susceptibility was found in the Asian group (OR = 1.416; 95% CI 1.095-1.831) and for the NSCLC type (OR = 2.816; 95% CI 1.645-4.818).

As shown in **Table 6**, the genotype T was associated with an increased risk of lung cancer in the overall analysis (OR = 1.126; 95% CI 1.024-1.239) compared with the genotype C. Subgroup analyses revealed that increased lung cancer susceptibility was observed in the Asian population (OR = 1.178; 95% CI 1.035-1.340), for the NSCLC type (OR = 1.511; 95% CI 1.190-1.920), and for squamous cell carcinoma (OR = 1.427; 95% CI 1.160-1.745).

*The MTHFR C677T polymorphism and response to chemotherapy*

Eight studies [30, 42-48] were eligible to be included in the analysis of the association between the MTHFR C677T polymorphism and response to chemotherapy. Meta-analysis indicated the pooled OR for the genotype TT+CT was 0.521 (95% CI 0.361-0.753) compared with the CC allele. However, MTHFR 677T polymorphisms were not associated with the treatment response under the following three

**Table 3.** MTHFR C677T polymorphism and lung cancer risk (TT/CT+CC)

| Group/subgroup | No. of studies | No. lung cancer/Control | Heterogeneity | | Pooled results | | | Publication bias (*P*-value) | |
|---|---|---|---|---|---|---|---|---|---|
| | | | *P*-value | $I^2$ | OR | 95% CI | *P*-value | Begg's test | Egger's test |
| Overall | 24 | 12130/12447 | <0.001 | 63.9% | 1.250 | 1.076-1.452 | 0.004 | 0.472 | 0.840 |
| Asia | 15 | 7704/6639 | <0.001 | 72.0% | 1.300 | 1.065-1.588 | 0.010 | 0.322 | 0.632 |
| Mainland China | 10 | 2372/2852 | 0.005 | 62.1% | 1.461 | 1.146-1.863 | 0.002 | 0.245 | 0.910 |
| No-Mainland China | 5 | 5332/4057 | 0.003 | 75.5% | 1.052 | 0.759-1.459 | 0.760 | 1.000 | 0.992 |
| Europe | 9 | 4426/5808 | 0.095 | 40.9% | 1.115 | 0.976-1.274 | 0.110 | 0.917 | 0.847 |
| SCLC | 4 | 87/305 | 0.915 | 0% | 1.636 | 0.784-3.413 | 0.190 | 0.308 | 0.101 |
| NSCLC | 4 | 323/305 | 0.592 | 0% | 2.476 | 1.517-4.043 | <0.001 | 1.000 | 0.461 |
| Squamous cell carcinoma | 5 | 382/2011 | 0.158 | 39.5% | 1.129 | 0.844-1.509 | 0.414 | 0.806 | 0.617 |
| Adenocarcinoma | 5 | 583/2011 | 0.005 | 73.0% | 1.308 | 0.777-2.202 | 0.312 | 0.221 | 0.793 |

Abbreviations: NSCLC, non-small cell lung cancer; SCLC, small cell lung cancer; OR, odds ratio; CI, confidence interval.

**Table 4.** MTHFR C677T polymorphism and lung cancer risk (TT+CT/CC)

| Group/subgroup | No. of studies | No. lung cancer/Control | Heterogeneity | | Pooled results | | | Publication bias (*P*-value) | |
|---|---|---|---|---|---|---|---|---|---|
| | | | *P*-value | $I^2$ | OR | 95% CI | P | Begg's test | Egger's test |
| Overall | 24 | 12130/12447 | <0.001 | 72.3% | 1.113 | 0.983-1.260 | 0.092 | 0.472 | 0.725 |
| Asia | 15 | 7704/6639 | <0.001 | 75.2% | 1.191 | 1.001-1.416 | 0.048 | 0.138 | 0.088 |
| Mainland China | 10 | 2372/2852 | 0.002 | 65.0% | 1.377 | 1.092-1.735 | 0.007 | 0.180 | 0.598 |
| No-Mainland China | 5 | 5332/4057 | 0.006 | 72.6% | 1.035 | 0.840-1.276 | 0.746 | 0.624 | 0.605 |
| Europe | 9 | 4426/5808 | 0.001 | 70.0% | 1.005 | 0.8221.229 | 0.96 | 0.754 | 0.319 |
| SCLC | 4 | 87/305 | 0.161 | 41.7% | 1.064 | 0.639-1.779 | 0.814 | 0.308 | 0.386 |
| NSCLC | 4 | 323/305 | 0.219 | 32.2% | 1.417 | 1.015-1.978 | 0.041 | 1.000 | 0.397 |
| Squamous cell carcinoma | 5 | 382/2011 | 0.519 | 0% | 1.123 | 0.840-1.501 | 0.434 | 0.308 | 0.363 |
| Adenocarcinoma | 5 | 583/2011 | 0.113 | 49.8% | 1.258 | 1.0051.575 | 0.045 | 0.734 | 0.960 |

Abbreviations: NSCLC, non-small cell lung cancer; SCLC, *small* cell lung cancer; OR, odds ratio; CI, confidence.

**Table 5.** MTHFR C677T polymorphism and lung cancer risk (TT/CC )

| Group/subgroup | No. of studies | No. lung cancer/Control | Heterogeneity | | Pooled results | | | Publication bias (*P*-value) | |
|---|---|---|---|---|---|---|---|---|---|
| | | | *P*-value | $I^2$ | OR | 95% CI | P | Begg's test | Egger's test |
| Overall | 24 | 12130/12447 | <0.001 | 72.5 | 1.324 | 1.095-1.601 | 0.004 | 0.503 | 0.753 |
| Asia | 15 | 7704/6639 | <0.001 | 77.7% | 1.416 | 1.095-1.831 | 0.008 | 0.235 | 0.377 |
| Mainland China | 10 | 2372/2852 | 0.003 | 64.6% | 1.682 | 1.247-2.270 | 0.001 | 0.180 | 0.903 |
| No-Mainland China | 5 | 5332/4057 | 0.001 | 79.2% | 1.033 | 0.703-1.519 | 0.868 | 1.000 | 0.863 |
| Europe | 9 | 4426/5808 | 0.008 | 61.3% | 1.17 | 0.859-1.593 | 0.319 | 0.917 | 0.629 |
| SCLC | 4 | 87/305 | 0.692 | 0% | 1.813 | 0.812-4.048 | 0.146 | 0.734 | 0.100 |
| NSCLC | 4 | 323/305 | 0.427 | 0% | 2.816 | 1.645-4.818 | <0.001 | 0.734 | 0.490 |
| Squamous cell carcinoma | 5 | 382/2011 | 0.117 | 49.1% | 1.096 | 0.731-1.643 | 0.658 | 0.308 | 0.333 |
| Adenocarcinoma | 5 | 583/2011 | 0.217 | 32.5% | 1.188 | 0.876-1.612 | 0.268 | 0.734 | 0.815 |

Abbreviations: NSCLC, non-small cell lung cancer; SCLC, small cell lung cancer; OR, odds ratio; CI, confidence interval.

comparison models (TT vs. CT+CC: OR = 0.829; 95% CI 0.425-1.616; TT vs. CC: OR = 0.555; 95% CI 0.224-1.378; and T vs. C: OR = 0.642; 95% CI 0.378-1.089) (**Table 7**).

*Publication bias*

Both Begg's rank correlation test and Egger's linear regression test were performed to access

**Table 6.** MTHFR C677T polymorphism and lung cancer risk (T/C)

| Group/subgroup | No. of studies | No. lung cancer/ Control | Heterogeneity | | Pooled results | | | Publication bias (*P*-value) | |
|---|---|---|---|---|---|---|---|---|---|
| | | | *P*-value | $I^2$ | OR | 95% CI | *P*-value | Begg's test | Egger's test |
| Overall | 24 | 12130/12447 | <0.001 | 77.5% | 1.126 | 1.024-1.239 | 0.015 | 0.535 | 0.727 |
| Asia | 15 | 7704/6639 | <0.001 | 80.5% | 1.178 | 1.035-1.340 | 0.013 | 0.276 | 0.204 |
| Mainland China | 10 | 2372/2852 | 0.001 | 68.3% | 1.305 | 1.119-1.523 | 0.001 | 0.180 | 0.724 |
| No-Mainland China | 5 | 5332/4057 | <0.001 | 81.7% | 0.984 | 0.845-1.146 | 0.901 | 1.000 | 0.770 |
| Europe | 9 | 4426/5808 | <0.001 | 73.7% | 1.043 | 0.888-1.224 | 0.610 | 1.000 | 0.478 |
| SCLC | 4 | 87/305 | 0.165 | 41.1% | 1.151 | 0.793-1.672 | 0.459 | 0.308 | 0.097 |
| NSCLC | 4 | 323/305 | 0.107 | 50.8% | 1.511 | 1.190-1.920 | 0.001 | 1.000 | 0.966 |
| Squamous cell carcinoma | 5 | 382/2011 | 0.526 | 0% | 1.427 | 1.160-1.745 | 0.001 | 1.000 | 0.510 |
| Adenocarcinoma | 5 | 583/2011 | 0.513 | 0% | 1.083 | 0.940-1.248 | 0.270 | 0.734 | 0.994 |

Abbreviations: NSCLC, non-small cell lung cancer; SCLC, small cell lung cancer; OR, odds ratio; CI, confidence interval.

**Table 7.** MTHFR C677T polymorphism and treatment response to chemotherapy

| Genotype | No. of studies | Heterogeneity | | Pooled results | | | Publication bias (*P*-value) | |
|---|---|---|---|---|---|---|---|---|
| | | *P*-value | $I^2$ | OR | 95% CI | *P*-value | Begg's test | Egger's test |
| TT/CT+CC | 8 | 0.025 | 56.2% | 0.829 | 0.425-1.616 | 0.581 | 0.266 | 0.134 |
| TT+CT/CC | 7 | 0.069 | 48.8% | 0.521 | 0.361-0.753 | 0.001 | 0.999 | 0.746 |
| TT/CC | 7 | 0.023 | 59.2% | 0.555 | 0.224-1.378 | 0.205 | 0.999 | 0.595 |
| T/C | 7 | 0.001 | 73.8% | 0.642 | 0.378-1.089 | 0.100 | 0.999 | 0.289 |

Abbreviations: OR, Odds ratio; CI, Confidence interval.

the publication bias of the included studies. No evidence of publication bias was observed in all pooled analysis. The detailed results of Begg's rank correlation test and Egger's linear regression test are listed in **Tables 3**-**6**.

## Discussion

This updated meta-analysis was designed to investigate the role of MTHFR C677T gene polymorphisms in the susceptibility to lung cancer and response to chemotherapy among lung cancer patients. We have added all newly eligible articles, which were not included in the previously published meta-analyses. Overall, the current meta-analysis revealed that the MTHFR C677T gene polymorphism was associated with an increased risk of lung cancer in Asian patients and poor response to chemotherapy.

Stratified analysis based on the geographical region showed significantly increased lung cancer risk in the Asian population but did not find such an association in the European population. This discrepancy might be explained by the different genetic backgrounds and environmental exposures of the diverse populations. For example, lack of sufficient folic acid intake has been reported in the Chinese population [49], which supports lung cancer susceptibility in the Asian population. Subgroup analysis by the general histological type suggested that the MTHFR C677T polymorphism showed a significant relationship with an increased risk of the NSCLC subtype. Similarly, in the stratified analysis by the type of pathology, compared with two C alleles, the TT+CT genotype was associated with a 25.8% higher risk of adenocarcinoma, whereas the one C allele conferred a 42.7% higher risk of squamous cell carcinoma. The pathogenesis of squamous cell carcinoma is different from that of adenocarcinoma. Therefore, whether the effect of the MTHFR C677T polymorphism differs between squamous cell carcinoma and adenocarcinoma needs to be further investigated.

Most participants in the included studies were advanced NSCLC patients. Currently, platinum-based chemotherapy remains the standard chemotherapy for the treatment of advanced NSCLC [44]. However, the response to chemotherapy has remarkable inter-individual variability. The anticancer activity of platinum agents is mainly achieved via the formation of DNA

adducts that block DNA replication and hinder cell division [50]. Genetic factors may contribute to the differing response to chemotherapy [51]. The MTHFR 677T gene polymorphism may influence the treatment outcome of chemotherapy in lung cancer patients by altering the methylation pattern, which may modify DNA conformation. In our pooled analysis, lung cancer patients carrying the TT+CT genotype had a 52.1% lower rate of CR+PR to chemotherapy compared with the CC allele carriers, suggesting that these variants may be predictors for treatment response to chemotherapy in lung cancer patients. Apart from response to chemotherapy, patients with NSCLC carrying the MTHFR 677CC genotype had a longer overall survival time with cisplatin/gemcitabine [48] and platinum/pemetrexed therapy [52]. In contrast, the MTHFR TT genotype was associated with increased progression-free survival in a pemetrexed-based therapy [53]. Moreover, the MTHFR TT genotype also was associated with sensitivity to fluorouracil-based chemotherapy in other types of cancer [54]. Taking these findings together, the MTHFR 677 polymorphism may determine the response to chemotherapy and prognosis.

The current meta-analysis included 31 eligible studies involving 12,878 cases and 12,447 controls, which made our meta-analysis more comprehensive than previously published meta-analyses. There was no evidence of publication bias in the pooled analysis, suggesting our results were robust and unbiased. We demonstrated a significant relationship between the MTHFR C677T polymorphism and lung cancer susceptibility in an Asian population. Moreover, we further confirmed that the MTHFR C677T polymorphism was associated with treatment response to a chemotherapy regimen, indicating individualized management of lung cancer patients based on MTHFR genotype may improve the response to chemotherapy.

Several potential limitations in this study should be mentioned: first, we only included studies published in English and Chinese, and thus, some relevant studies might be not included in the meta-analysis. Second, conclusions drawn from subgroup analyses might be unreliable due to the small numbers of patients analyzed. Moreover, the selection of control subjects from a hospital-based population may result in selection bias. Third, the role of the MTHFR polymorphisms in modulating cancer risk is associated with folate status, and folate deficiency and smoking are important factors for the development of lung cancer [55]. However, this meta-analysis did not make adjustments for differences in folic acid intake, smoking status, or other potential confounding factors. A more accurate analysis could be achieved by adjusting for these covariates. Finally, due to a lack of sufficient data in the literature, the effects of gene-gene and gene-environment interactions were not addressed in this meta-analysis.

In conclusion, this meta-analysis suggests that the MTHFR C677T polymorphism may be a genetic marker for lung cancer susceptibility, especially in Asian populations. The MTHFR 677 TT+CT genotype was associated with a lower response to chemotherapy in lung cancer patients. However, more well-designed studies with large samples are needed to confirm our findings. Specifically, *clarifying the* interaction *of the* MTHFR C677T genotype and smoking status in lung cancer susceptibility is required.

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## Disclosure of conflict of interest

None.

**Address correspondence to:** Dr. Ke Wang, Department of Respiratory Medicine, Second Affiliated Hospital of Jilin University, No. 218 Ziqiang Street, Nanguan District, Changchun 130021, Jilin, China. Tel: +86-186 4311 1766; Fax: +86-0431-8879-6820; E-mail: kewangm1@hotmail.com

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