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MTHFR C677T and A1298C polymorphisms and risk of lung cancer: a comprehensive evaluation

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ABSTRACT. Results from previous studies on the association between methylenetetrahydrofolate reductase (*MTHFR*) polymorphisms C677T and A1298C and lung cancer have been conflicting. The aim of this meta-analysis was to clarify the effect of *MTHFR* polymorphisms on the risk of lung cancer. An electronic search of PubMed, EMBASE, the Cochrane library, and the China Knowledge Resource Integrated Database for papers on C677T and A1298C and susceptibility to lung cancer was performed. The STATA software (Version 13.0) was used for statistical analysis. Statistical heterogeneity, tests of publication bias, and a sensitivity analysis were performed. Twenty-six studies on C677T (12,324 cases and 12,532 controls) and thirteen studies on A1298C (6773 cases and 8207 controls) were included in the meta-analysis. The *MTHFR* C677T polymorphism showed significant pooled ORs for the homozygote comparison (TT versus CC: OR = 1.518, 95%CI = 1.220-1.890), heterozygote comparison (CT versus CC: OR = 1.053, 95%CI = 0.940-1.179), dominant model (CT +

TT versus CC: OR = 1.143, 95%CI = 1.013-1.291), recessive model (TT versus CT + CC: OR = 1.435, 95%CI = 1.190-1.730), and additive model (T versus C: OR = 1.176, 95%CI = 1.066-1.298). In summary, our meta-analysis showed that the *MTHFR* C677T polymorphism is associated with a significant increase in lung cancer risk in Asian and overall populations, but not in Caucasian populations. However, no significant association between the *MTHFR* A1298C polymorphism and lung cancer risk was found in either the Caucasian or Asian group with any genetic models.

Key words: MTHFR; C677T; A1298C; Polymorphisms; Lung cancer; Meta-analysis

INTRODUCTION

Approximately 14.1 million new cancer cases and 8.2 million deaths occurred in 2012 worldwide. Lung cancer is the leading cause of cancer death among males in both developed and developing countries, and has surpassed breast cancer as the leading cause of cancer death among females in developed countries (Torre et al., 2015). Lung cancer risk is not fully understood, but it has been commonly accepted to be related to many factors, including genetic and/or environmental factors. Among these, smoking is regarded as an important risk factor for lung cancer (relative risk = 10-30 compared with nonsmokers). However, lung cancer occurs in less than 20% of people who smoke throughout their life, suggesting that genetic factors may play a very important role in the development of lung cancer (Shields, 2002).

Previous epidemiological studies have demonstrated that consumption of fruits and vegetables is associated with a decreased risk of cancer, including lung cancer. Numerous constituents of fruits and vegetables are thought to contribute to this protective effect. One is folate, often in the form of folic acid (Shen et al., 2003). The enzyme MTHFR plays a critical role in the folate metabolism pathway by regulating the intracellular folate pool for synthesis and methylation of DNA. The MTHFR gene is located on chromosome 1 at the end of the short arm (1p36.6) and is 2.2 kb in length with a total of 11 exons (Goyette et al., 1994).

Several single nucleotide polymorphisms (SNP) in the MTHFR gene have been characterized, including C677T and A1298C. The C-to-T transition at nucleotide 677 in exon 4 is a point mutation that results in an amino acid substitution of valine for alanine, which decreases the thermal stability and reduces the activity of MTHFR. A reduced level of MTHFR substrate could lead to uracil misincorporation into DNA, diminished DNA repair, and increased frequency of chromosomal breaks and damage (Krajinovic et al., 2004). The MTHFR polymorphism A1298C (rs1801131) in exon 7, resulting in the replacement of glutamate with alanine at amino acid 429 (E429A), may increase serum folate levels, possibly influencing cancer risk (Parle-McDermott et al., 2006).

A number of studies have investigated the association between the two common MTHFR polymorphisms and the risk of lung cancer. However, the results remain controversial and previous studies have generally been small in size. In 2008, a meta-analysis based on eight studies reported the absence of associations between MTHFR C677T and A1298C and lung cancer (Mao et al., 2008). In 2009, a meta-analysis based on ten studies concluded that a possible increased risk exists for subjects carrying the MTHFR 677 TT genotype, but not for MTHFR 1298 CC (Boccia et al., 2009). To clarify the effect of the MTHFR C677T and A1298C polymorphisms on the risk of lung cancer, we have performed a meta-analysis including twenty-six studies on C677T (12,324 cases and 12,532 controls) and thirteen studies on A1298C (6773 cases and 8207 controls).

MATERIAL AND METHODS

Search strategy

This meta-analysis was performed according to the standard MOOSE guideline (Stroup et al., 2000). PubMed, EMBASE, the Cochrane library, and the China Knowledge Resource Integrated Database (until January 1, 2015) were searched using the search terms "(C677T or A1298C or MTHFR) and (polymorphism or variants) and (lung cancer)". Only studies published in English or Chinese were included. Related reference articles were also searched to identify other relevant publications. Unpublished data and further information were also obtained from the corresponding authors.

Inclusion and exclusion criteria

Potential studies were selected based on the following inclusion criteria: 1) *MTHFR* C677T or A1298C polymorphisms and lung cancer were assessed; 2) human case-control design; 3) frequency of the *MTHFR* C677T or A1298C polymorphism was reported as number of cancer cases and controls according to the three variant genotypes of either polymorphism; and 4) published in English or Chinese. The criteria for exclusion were as follows: 1) not related to the *MTHFR* C677T or A1298C polymorphisms and lung cancer; 2) not a primary case-control study; 3) no usable or sufficient genotype data reported; 4) studies whose allele frequency in the control population deviated from the Hardy-Weinberg equilibrium at $P \leq 0.01$; 5) case reports, letter to Editor, book chapters, or reviews. The study inclusion and exclusion procedures are summarized in Figure 1.

We contacted the corresponding authors by e-mail in order to obtain the absolute number of homozygous and heterozygous individuals in case and control groups for those papers reporting only the allele frequencies. If more than one article was published by the same author using the same case series, we selected one paper and excluded all others.

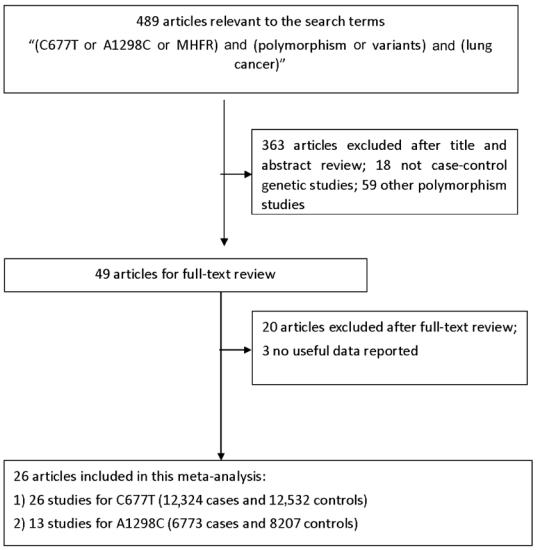

Figure 1. Inclusion and exclusion procedures.

Data extraction

Two investigators independently extracted the data from all qualified studies using the selection standards listed above. Discrepancies were resolved through discussion until agreement was reached. We extracted the following information for each study: the first author's name, year of publication, the country in which the study was conducted, the source of the control group, evidence of Hardy-Weinberg equilibrium in controls, the sample size, the number of cases and controls with the CC/CT/TT or AA/AC/CC genotypes.

Statistical analysis

The STATA software (Version 13.0) was used for all statistical analyses. Two-sided P values $\leq$0.05 were considered statistically significant. For the control groups for each study, the observed genotype frequencies of the *MTHFR* C677T and A1298C polymorphisms were evaluated for Hardy-Weinberg equilibrium. The strength of the association between the *MTHFR* C677T and A1298C polymorphisms and lung cancer risk was assessed by the odds ratios (ORs) with 95%CIs. The pooled ORs were calculated for the homozygote model, heterozygote model, dominant model, recessive model, and additive model. Cochran's Q-statistic and the I^2 metric were conducted to assess heterogeneity between studies, where $P < 0.10$ and $I^2 > 50\%$ were considered to indicate the existence of significant heterogeneity (Higgins and Thompson, 2002). If the heterogeneity test result returned $P > 0.1$, the pooled ORs were analyzed using the random-effect model; otherwise, the fixed-effect model was used. Sensitivity analyses were also performed after sequential removal of each study. Lastly, Begg's funnel plot and the Egger test were used to statistically examine publication biases.

RESULTS

Characteristics of included studies

Overall, twenty-six case-control studies with 12,324 lung cancer cases and 12,532 controls were retrieved based on the search criteria for lung cancer susceptibility related to the *MTHFR* C677T polymorphism. Thirteen studies also provide data on A1298C with 6773 lung cancer cases and 8207 controls. All studies included in the meta-analysis are summarized in Table 1 (Heijmans et al., 2003; Jeng et al., 2003; Siemianowicz et al., 2003; Shen et al., 2001, 2005; Shi et al., 2005; Zhang et al., 2005; Zou et al., 2006; Gemignani et al., 2007; Hung et al., 2007; Jing et al., 2007; Suzuki et al., 2007; Liu et al., 2008, 2009; Yang et al., 2010; Yao et al., 2010; Arslan et al., 2011; Cui et al., 2011a,b; Kiyohara et al., 2011; Cheng et al., 2011, 2012; Ma et al., 2012; Cai et al., 2014; Cavic et al., 2014; Yilmaz et al., 2014). No overlap occurred between the studies based on case or control participation. The genotype distributions in the controls for all studies were consistent with the Hardy-Weinberg equilibrium (Norton and Neel, 1965), with the exception of three studies.

Results of the overall meta-analysis

Our main results on the association between the *MTHFR* C677T and A1298C

polymorphisms and lung cancer are listed in Table 2. The *MTHFR* C677T polymorphism showed significant pooled ORs for the homozygote comparison (TT versus CC: OR = 1.518, 95%CI = 1.220-1.890), heterozygote comparison (CT versus CC: OR = 1.053, 95%CI = 0.940-1.179), dominant model (CT + TT versus CC: OR = 1.143, 95%CI = 1.013-1.291), recessive model (TT versus CT + CC: OR = 1.435, 95%CI = 1.190-1.730), and additive model (T versus C: OR = 1.176, 95%CI = 1.066-1.298). The *MTHFR* A1298C polymorphism showed non-significant pooled ORs for the homozygote comparison (CC versus AA: OR = 1.073, 95%CI = 0.943-1.221), heterozygote comparison (AC versus AA: OR = 0.992, 95%CI = 0.925-1.064), dominant model (AC + CC versus AA: OR = 1.004, 95%CI = 0.940-1.074), recessive model (CC versus AC + AA: OR = 1.073, 95%CI = 0.948-1.214), and additive model (C versus A: OR = 1.015, 95%CI = 0.964-1.070). We found no association between the *MTHFR* A1298C polymorphism and lung cancer risk, whereas a significant correlation existed between the *MTHFR* C677T polymorphism and lung cancer risk (Figure 2).

Table 1. Characteristics of studies included in this meta-analysis.

Author	Year	Region	Ethnicity			Case				Control		Hardy Weinberg (P)
MTHFR C677T				Total	CC	CT	TT	Total	CC	CT	TT	
Shen et al., 2001	2001	USA	Caucasian	550	241	252	57	554	245	252	57	0.508
Jeng et al., 2003	2003	China	Asian	59	36	22	1	232	123	95	14	0.438
Siemianowicz et al., 2003	2003	Poland	Caucasian	146	38	60	48	44	18	20	6	0.906
Heijmans et al., 2003	2003	Netherlands	Caucasian	44	23	17	4	793	399	329	65	0.806
Shi et al., 2005	2005	USA	Caucasian	1051	483	468	100	1141	498	519	124	0.516
Zhang et al., 2005	2005	China	Asian	505	120	230	155	500	160	231	109	0.138
Shen et al., 2005	2005	China	Asian	116	33	65	18	111	53	42	16	0.117
Zou et al., 2006	2006	China	Asian	100	24	52	24	100	39	48	13	0.767
Jing et al., 2007	2007	China	Asian	100	24	52	24	100	39	48	13	0.767
Suzuki et al., 2007	2007	Japan	Asian	515	182	256	77	1030	379	474	177	0.170
Gemignani et al., 2007	2007	European	Caucasian	247	104	107	36	259	131	103	25	0.473
Hung et al., 2007	2007	France	Caucasian	2169	1009	929	231	2803	1397	1147	259	0.288
Liu et al., 2008	2008	China	Asian	500	157	245	98	424	149	265	10	P < 0.05
Liu et al., 2009	2009	Taiwan	Asian	358	205	124	29	716	362	291	63	0.679
Yang et al., 2010	2010	China	Asian	120	49	52	19	165	62	75	28	0.516
Yao et al., 2010	2010	China	Asian	93	27	46	20	106	36	51	19	0.899
Kiyohara et al., 2011	2011	Japan	Asian	462	153	201	108	379	158	170	51	0.624
Arslan et al., 2011	2011	Turkey	Caucasian	64	30	27	7	61	29	29	3	0.206
Cui et al., 2011b	2011	China	Asian	438	58	240	140	641	121	325	195	0.483
Cheng et al., 2011	2011	China	Asian	178	49	58	71	180	47	88	45	0.767
Cui et al., 2011a	2011	Korean	Asian	3938	1361	1909	668	1700	540	862	298	0.148
Cheng et al., 2012	2012	China	Asian	94	26	33	35	78	21	39	18	0.990
Ma et al., 2012	2012	China	Asian	120	20	54	46	60	22	28	10	0.830
Cavic et al., 2014	2014	Serbia	Caucasian	55	34	18	3	53	13	33	7	0.057
Yilmaz et al., 2014	2014	Turkey	Caucasian	100	55	38	7	100	51	43	6	0.433
Cai et al., 2014	2014	China	Asian	202	54	102	46	202	69	112	21	P < 0.05
MTHFR A1298C				Total	AA	AC	CC	Total	AA	AC	CC	
Shen et al., 2001	2001	USA	Caucasian	550	261	246	43	554	265	249	40	0.072
Siemianowicz et al., 2003	2003	Poland	Caucasian	146	32	76	38	44	12	24	8	0.507
Shi et al., 2005	2005	USA	Caucasian	1051	480	462	109	1141	554	496	91	0.168
Zhang et al., 2005	2005	China	Asian	505	355	141	9	500	345	150	5	P < 0.05
Shen et al., 2005	2005	China	Asian	114	71	41	2	109	69	34	6	0.509
Jing et al., 2007	2007	China	Asian	100	70	28	2	100	68	30	2	0.528
Suzuki et al., 2007	2007	Japan	Asian	512	341	149	22	1019	652	322	45	0.515
Hung et al., 2007	2007	France	Caucasian	2209	1031	960	218	2865	1285	1268	312	0.976
Liu et al., 2008	2008	China	Asian	500	341	141	18	517	364	142	11	0.509
Liu et al., 2009	2009	Taiwan	Asian	358	228	115	15	716	467	226	23	0.491
Kiyohara et al., 2011	2011	Japan	Asian	462	278	154	30	379	239	122	18	0.633
Arslan et al., 2011	2011	Turkey	Caucasian	64	29	29	6	61	28	29	4	0.543
Cai et al., 2014	2014	China	Asian	202	55	106	41	202	65	102	35	0.642

Table 2. Results of the overall meta-analysis.

MTHFR C677T			
Contrast	OR, 95%CI	Heterogeneity	Z and P
TT versus CC	1.518, [1.220, 1.890]	chi-squared = 120.84 (d.f. = 25) P < 0.001, I-squared = 79.3%	z = 3.74 P = 0.000
CT versus CC	1.053, [0.940, 1.179]	chi-squared = 65.32 (d.f. = 25) P < 0.001, I-squared = 61.7%	z = 0.89 P = 0.372
CT+TT versus CC	1.143, [1.013, 1.291]	chi-squared = 86.41 (d.f. = 25) P < 0.001, I-squared = 71.1%	z = 2.16 P = 0.031
TT versus CT+CC	1.435, [1.190, 1.730]	chi-squared = 108.37 (d.f. = 25) P < 0.001, I-squared = 76.9%	z = 3.78 P = 0.000
T versus C	1.176, [1.066, 1.298]	chi-squared = 120.43 (d.f. = 25) P < 0.001, I-squared = 79.2%	z = 3.23 P = 0.001
MTHFR A1298C			
Contrast	OR, 95% CI	Heterogeneity	Z and P
CC versus AA	1.073, [0.943, 1.221]	chi-squared = 15.07 (d.f. = 12) P = 0.238, I-squared = 20.4%	z = 1.07 P = 0.283
AC versus AA	0.992, [0.925, 1.064]	chi-squared = 4.96 (d.f. = 12) P = 0.959, I-squared = 0.0%	z = 0.22 P = 0.829
AC+CC versus AA	1.004, [0.940,1.074]	chi-squared = 8.12 (d.f. = 12) P = 0.775, I-squared = 0.0%	z = 0.13 P = 0.898
CC versus AC+AA	1.073, [0.948,1.214]	chi-squared = 13.01 (d.f. = 12) P = 0.368, I-squared = 7.7%	z = 1.12 P = 0.263
C versus A	1.015, [0.964,1.070]	chi-squared = 12.17 (d.f. = 12) P = 0.432, I-squared = 1.4%	z = 0.57 P = 0.565

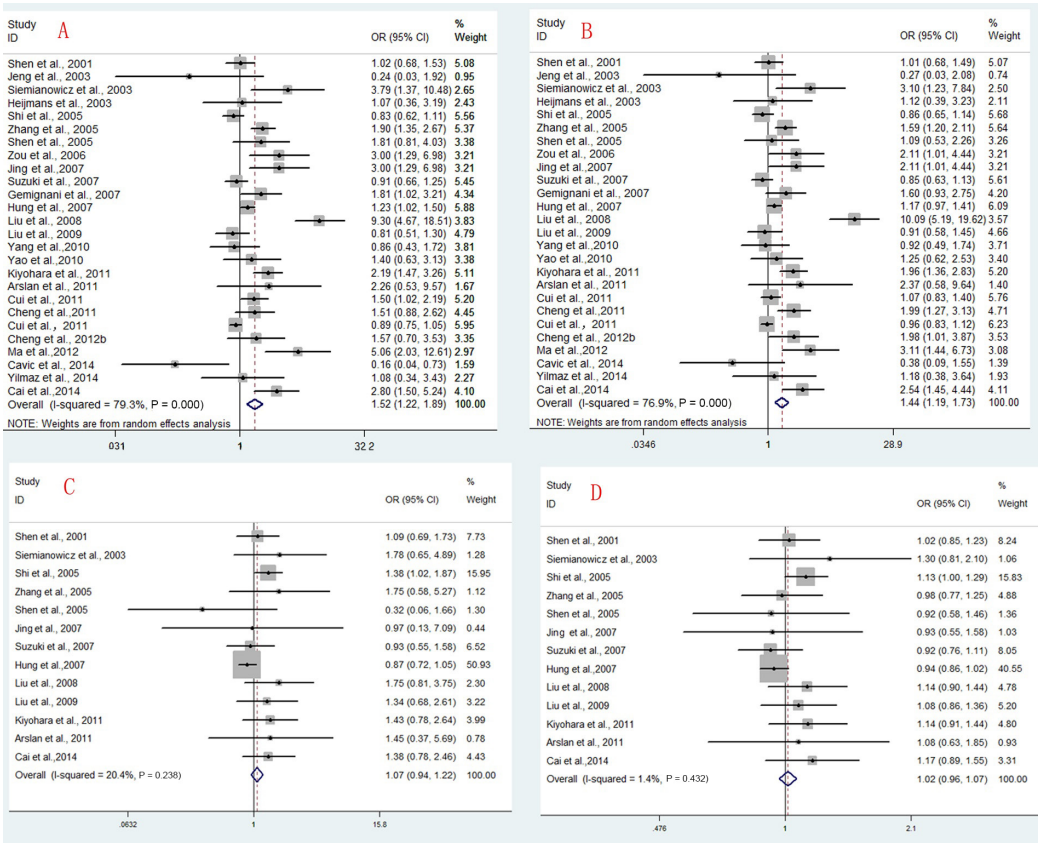

Figure 2. Overall meta-analysis for C677T (A, TT *vs* CC; B, TT *vs* CT + CC) and for A1298C (C, CC *vs* AA; D, C *vs* A) polymorphisms.

Subgroup analysis

We performed a subgroup analysis stratified by ethnicity. We found that the *MTHFR* C677T polymorphism was associated with a significantly increased lung cancer risk in the Asian and overall populations. In the homozygote comparison (TT versus CC), the pooled OR was 1.722 (95%CI = 1.271-2.334, P < 0.001) for the Asian population. Under the recessive model (TT versus CT + CC), the pooled OR was 1.572 (95%CI = 1.215-2.033, P = 0.001) for the Asian population. However, we did not find an association between the A1298C polymorphism and lung cancer risk in the Asian group under any genetic model. Similarly, we found no significant association between either the C677T or A1298C polymorphism and lung cancer risk in the Caucasian group under any genetic model. The meta-analysis results for all genetic models are listed in detail in Table 3.

Table 3. Subgroup analysis stratified by ethnicity.

MTHFR C677T						
Ethnicity	Comparisons	TT versus CC (OR, 95%CI, Z, P)	CT versus CC (OR, 95%CI, Z, P)	CT+TT versus CC (OR, 95%CI, Z, P)	TT versus CT+CC (OR, 95%CI, Z, P)	T versus C (OR, 95%CI, Z, P)
Caucasian	9	1.170, [0.859, 1.593] z = 1.00 P = 0.319	0.978, [0.815, 1.174] z = 0.24 P = 0.813	1.005, [0.822, 1.229] z = 0.05 P = 0.960	1.146, [0.910, 1.444] z = 1.16 P = 0.247	1.043, [0.888, 1.224] z = 0.51 P = 0.610
Asian	17	1.722, [1.271, 2.334] z = 3.51 P < 0.001	1.107, [0.948, 1.292] z = 1.28 P = 0.201	1.235, [1.046, 1.458] z = 2.49 P = 0.013	1.572, [1.215, 2.033] z = 3.45 P = 0.001	1.251, [1.096, 1.429] z = 3.31 P = 0.001
Overall	26	1.518, [1.220, 1.890] z = 3.74 P < 0.001	1.053, [0.940, 1.179] z = 0.89 P = 0.372	1.143, [1.013, 1.291] z = 2.16 P = 0.031	1.435, [1.190, 1.730] z = 3.78 P < 0.001	1.176, [1.066, 1.298] z = 3.23 P = 0.001
MTHFR A1298C						
Ethnicity	Comparisons	CC versus AA (OR, 95%CI, Z, P)	AC versus AA (OR, 95%CI, Z, P)	AC+CC versus AA (OR, 95%CI, Z, P)	CC versus AC+AA (OR, 95%CI, Z, P)	C versus A (OR, 95%CI, Z, P)
Caucasian	5	1.021, [0.879, 1.185] z = 0.27 P = 0.789	0.987, [0.903, 1.079] z = 0.29 P = 0.774	0.993, [0.912, 1.081] z = 0.17 P = 0.865	1.030, [0.893, 1.187] z = 0.40 P = 0.686	1.002, [0.939, 1.068] z = 0.05 P = 0.957
Asian	8	1.247, [0.964, 1.612] z = 1.68 P = 0.092	1.001, [0.895, 1.119] z = 0.01 P = 0.990	1.023, [0.919, 1.139] z = 0.42 P = 0.674	1.215, [0.949, 1.556] z = 1.54 P = 0.123	1.043, [0.953, 1.141] z = 0.91 P = 0.361
Overall	13	1.073, [0.943, 1.221] z = 1.07 P = 0.283	0.992, [0.925, 1.064] z = 0.22 P = 0.829	1.004, [0.940, 1.074] z = 0.13 P = 0.898	1.073, [0.948, 1.214] z = 1.12 P = 0.263	1.015, [0.964, 1.070] z = 0.57 P = 0.565

Ideally, we should perform a subgroup analysis stratified by type of lung cancer. However, we were unable to do this, as most of the studies included mixed types of lung cancer among the cases. Similarly, a subgroup analysis stratified by gender is also desirable, as gender may play a role in the development of lung cancer. However, only a few studies reported the absolute number of homozygous and heterozygous individuals in case and control groups stratified by gender.

Test for heterogeneity

For the C677T polymorphism, there was significant heterogeneity for the homozygote comparison (TT versus CC, χ^2 = 120.84, d.f. = 25, P < 0.001, I^2 = 79.3%) and for the recessive model (TT versus CT + CC, χ^2 = 108.37, d.f. = 25, P < 0.001, I^2 = 76.9%). We assessed the source of the heterogeneity by region, publication year, control source, and sample size. However, we did not observe any sources that contributed to the substantial heterogeneity.

For the A1298C polymorphism, there was no significant heterogeneity for either the homozygote comparison (CC versus AA, χ^2 = 15.07, d.f. = 12, P = 0.238, I^2 = 20.4%) or the recessive model (CC versus AC + AA, c^2 = 13.01, d.f. = 12, P = 0.368, I^2 = 7.7%).

Sensitivity analysis

We conducted sensitivity analyses to ascertain the primary origin of the heterogeneity. The sensitivity analysis showed that no individual study had a marked effect on the pooled ORs (Figure 3A and B).

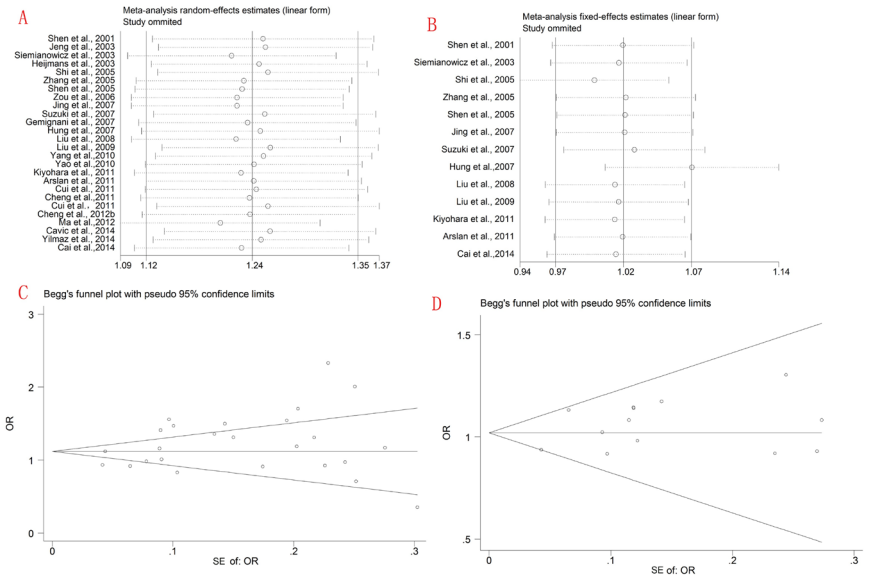

Figure 3. Sensitivity analysis and tests for publication bias.

Publication bias

A funnel plot was generated to assess publication bias (Figure 3C and D). Begg and Egger tests were performed to evaluate the funnel plot's symmetry statistically. The results showed no publication bias (Begg test Pr > |z| = 0.440 for C677T and Pr > |z| = 0.669 for A1298C).

DISCUSSION

Lung cancer is the leading cause of cancer death among males in both developed and developing countries and has surpassed breast cancer as the leading cause of cancer death among females in developed countries. Despite rapid advances in treatment over recent decades, the prognosis has not greatly improved: the 5-year survival rates for surgically resectable non-small cell lung cancer are still unsatisfactory and range from 19% for stage IIIA to 63% for stage IA (van Rens et al., 2000). Efforts toward primary prevention in addition to early detection have become more important. Lung cancer risk is commonly accepted to be multi-factorial, with genetic and/or environmental contributors. Although it is well known that cigarette smoking is a major cause of lung cancer, only 10-20% of lifetime smokers are known to develop lung cancer.

Folate is one of the micronutrients that provides protection against lung carcinogenesis (Voorrips et al., 2000). Shen et al. (2001) first examined the association between MTHFR gene polymorphisms and the risk of lung cancer. Subsequent case-control studies have provided controversial results. Small sample sizes, various ethnic groups, diets, environments, and methodologies may be responsible for the discrepancies. Two meta-analyses have been published, and neither detected any significant associations (Mao et al., 2008; Boccia et al., 2009). To clarify the effect of MTHFR polymorphisms C677T and A1298C on the risk of lung cancer, we performed a meta-analysis of twenty-six studies concerning C677T (12,324 cases and 12,532 controls) and thirteen studies concerning A1298C (6773 cases and 8207 controls). Our results

showed no association between the *MTHFR* A1298C polymorphism and lung cancer risk, whereas a significant correlation between the *MTHFR* C677T polymorphism and lung cancer risk existed in the overall population. In addition, we found that the C677T polymorphism was associated with a significantly increased lung cancer risk in Asian populations, but not in Caucasian populations. However, no significant association between the *MTHFR* A1298C polymorphism and lung cancer risk was found in Caucasian or Asian populations under any genetic models.

Although we performed a subgroup analysis stratified by ethnicity, we failed to perform a subgroup analysis stratified by type of lung cancer or by gender since most of the studies included mixed types of lung cancer in the case group and only a few studies reported the absolute number of homozygous and heterozygous individuals in case and control groups stratified by gender. As we know, the reliability of a meta-analysis based on small sample sizes is not high. Future studies stratified by type of lung cancer or by gender are therefore needed.

Through meta-analysis, a statistical technique for combining the results from independent studies, we were able to draw more reliable conclusions on the influence of the *MTHFR* C677T and A1298C polymorphisms on lung cancer risk. However, as lung cancer is believed to involve many factors, genetic and/or environmental, future research should investigate not only individual genes, but also gene-gene interactions, genetic-nutritional interactions, and other SNPs.

Several potential limitations of this meta-analysis should be mentioned. Although the funnel plot and the Begg test showed no publication bias, selection bias may have occurred because only studies in English or Chinese were selected. There was also significant heterogeneity detected for the C677T polymorphism. Despite these limitations, our meta-analysis has some clear advantages. Our meta-analysis contains the largest sample size to date to assess the association between the *MTHFR* C677T and A1298C polymorphisms and lung cancer risk. We were able to perform a subgroup analysis stratified by ethnicity, and the sensitivity analysis showed that no single study strongly affected the combined results. The well-designed search and selection method significantly increased the statistical power of this meta-analysis, and no publication bias was detected, indicating that our pooled results are likely to be reliable.

In summary, our meta-analysis showed that the *MTHFR* C677T polymorphism was associated with a significantly increased lung cancer risk in Asian and overall populations, but not in Caucasian populations. However, no significant association between the *MTHFR* A1298C polymorphism and lung cancer risk was found in Caucasian or Asian populations under any genetic models. Future research should investigate not only individual genes, but also gene-gene interactions, genetic-nutritional interactions, and other SNPs.

Conflicts of interest

The authors declare no conflict of interest.

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