BMJ Open

Relevance of *MTHFR* polymorphisms with response to fluoropyrimidine-based chemotherapy in oesophagogastric cancer: a meta-analysis

Lei Zhong,[1] Qi Fu,[2] Shu Zhou,[3] Lu Chen,[1] Qian Peng[4]

To cite: Zhong L, Fu Q, Zhou S, *et al*. Relevance of *MTHFR* polymorphisms with response to fluoropyrimidine-based chemotherapy in oesophagogastric cancer: a meta-analysis. *BMJ Open* 2018;**8**:e020767. doi:10.1136/bmjopen-2017-020767

► Prepublication history and additional material for this paper are available online. To view these files, please visit the journal online (http://dx.doi.org/10.1136/bmjopen-2017-020767).

Received 22 November 2017
Revised 10 February 2018
Accepted 16 April 2018

[1]Personalized Drug Therapy Key Laboratory of Sichuan Province, Sichuan Academy of Medical Sciences, Sichuan Provincial People's Hospital, Chengdu, China
[2]State Key Laboratory of Biotherapy and Cancer Center, West China Medical School, West China Hospital, Sichuan University, Chengdu, China
[3]School of Life Sciences and Key Laboratory of Bio-resources and Eco-environment, Ministry of Education, Sichuan University, Chengdu, China
[4]Cancer Center, Sichuan Academy of Medical Sciences, Sichuan Provincial People's Hospital, Chengdu, China

Correspondence to
Dr Qian Peng;
pengqian0522@163.com

ABSTRACT

Objective To evaluate the association between methylenetetrahydrofolate reductase (*MTHFR*) polymorphisms and the response to fluoropyrimidine-based chemotherapy in oesophagogastric cancer.

Design Meta-analysis.

Methods We searched PubMed, Embase and Web of Science databases from inception up to October 2017 for relevant studies. The statistical analysis was performed using STATA V.12.0 software. The pooled ORs and 95% CIs were used to assess the strength of the association under the allele, dominant and recessive models. We also conducted subgroup analysis stratified by cancer type, ethnicity and study design. Additionally, the sensitivity analysis was performed by sequential omission of individual studies, and the publication bias was detected using both Begg's test and Egger's test.

Results A total of 2020 patients from 12 studies were included in this meta-analysis. The results showed that there was no significant association between *MTHFR* C677T (rs1801133) and A1298C (rs1801131) polymorphisms and the clinical response to fluoropyrimidine-based chemotherapy under all of the three genetic models (T vs C: OR 0.93, 95% CI 0.76 to 1.15; C vs A: OR 0.88, 95% CI 0.56 to 1.40. CT+TT vs CC: OR 0.94, 95% CI 0.72 to 1.23; AC+CC vs AA: OR 0.80, 95% CI 0.47 to 1.35. TT vs CC+CT: OR 1.02, 95% CI 0.74 to 1.39; CC vs AA+AC: OR 1.15, 95% CI 0.50 to 2.67). When stratified by cancer type, ethnicity or study design, the association was still not significant in all subgroups.

Conclusions This meta-analysis suggested that *MTHFR* polymorphisms could not be considered as reliable factors for predicting the response to fluoropyrimidine-based chemotherapy in oesophagogastric cancer.

Strengths and limitations of this study

- ► We adopted the random effects model to analyse the pooled data to allow for a different effect in each population, and conducted stratified analysis to avoid heterogeneity.
- ► This study was limited by some variables, such as age, gender, diet, living habits, environmental exposure and pathological type of patients.
- ► This study was also limited by the small sample size in some subgroup analysis.

INTRODUCTION

Fluorouracil (5-FU) is the backbone of treatments for gastric and oesophageal cancers. Oral fluoropyrimidines including capecitabine and tegafur show similar efficacy to 5-FU.[1–4] Fluoropyrimidine drugs themselves have no antitumour activity, but they are converted to 5-fluoro-dUMP, which can further form a ternary complex with 5, 10-methylene tetrahydrofolate (5, 10-MTHF) and thymidylate synthase (TS). Formation of this ternary complex results in sustained inhibition of TS; it prevents the conversion of 2′ -deoxyuridine-5′-monophosphate into 2′-deoxythymidine-5′-monophosphate, thereby restraining the synthesis of DNA.[5] This is considered as the predominant mechanism of the antitumour effect of fluoropyrimidines.

Folate metabolism is an important factor influencing the antitumour activity of fluoropyrimidines. Increased 5, 10-MTHF could produce tighter ternary complexes and improve the efficacy of fluoropyrimidine drugs. Methylenetetrahydrofolate reductase (MTHFR) is a critical enzyme in folate-metabolising pathway. It catalyses the irreversible conversion of 5, 10-MTHF to 5-methyltetrahydrofolate, and reduces the amount of 5, 10-MTHF available for binding to FdUMP and TS.[5 6] Therefore, MTHFR plays a key role in the anabolism of fluoropyrimidines to the active metabolites. *MTHFR* gene locates in chromosome 1p36.3, and is highly polymorphic.[7] Two common functional polymorphisms of *MTHFR*, C677T (rs1801133) and A1298C (rs1801131), have been identified, the main variants that could decrease the activity of MTHFR.[8 9] Thus, *MTHFR* C677T and A1298C polymorphisms may contribute

greatly to the clinical response of fluoropyrimidine-based chemotherapy.

Theoretically, *MTHFR* gene polymorphisms are closely related to the efficacy of fluoropyrimidines for the treatment of gastric cancer and oesophageal cancer. However, the available evidence from the gene polymorphism studies in the clinic was weak, and the published results were inconsistent among studies.[10–13] Therefore, further assessment is needed. In this account, a systematic review and meta-analysis were carried out on the published data in order to comprehensively estimate the association of *MTHFR* C677T and A1298C polymorphisms with the clinical response to fluoropyrimidine-based chemotherapy in patients with oesophagogastric cancer.

METHODS

Literature search

We conducted a comprehensive search of PubMed, Embase and Web of Science databases from inception up to October 2017 using a combination of the following terms: "methylenetetrahydrofolate reductase" or "MTHFR", "polymorphism" or "pharmacogenetic" or "genotype" or "variant", "fluoropyrimidine" or "fluorouracil" or "5-Fu" or "capecitabine" or "tegafur", and "gastric cancer" or "esophageal cancer" or "esophagogastric cancer". The search was limited to articles reported in English. We have included the full search strategy for PubMed as an example in the online supplementary file. To identify more potentially relevant studies, a manual search for references cited in the eligible articles was also performed.

Selection criteria

The included literature in this study met the following criteria: (1) studies involving gastric cancer and oesophageal cancer; (2) chemotherapy regimens containing 5-FU, capecitabine or tegafur; (3) studies using validated molecular methods for genotyping and (4) studies providing information on *MTHFR* polymorphism or estimated genetic effects on response to treatment. No restrictions were imposed on the design of the studies, which could have been prospective or retrospective studies. Studies investigating susceptibility, progression or severity, and the case reports, letters, conference abstracts, meta-analysis and reviews were excluded.

Data extraction

The data were independently extracted by two researchers (LZ and QF). For each included study, the following information was collected: first author, publication year, ethnicity of the study population, study design, distribution of gender and age in patients, cancer type, chemotherapy regimen, clinical response, genotype distribution of *MTHFR* and genotyping methods, and the Hardy-Weinberg equilibrium examination result. Any discrepancies in data extraction were resolved by consensus.

Assessment of study quality

The quality of the included studies was evaluated independently by two reviewers according to the Newcastle-Ottawa Scale (NOS). The NOS includes three parameters of quality for studies: selection of the study population, comparability of subjects and exposure assessment, with scores ranging from 0 to 9. NOS scores of 0–4 and 5–9 were considered as low-quality and high-quality studies, respectively.

Statistical analysis

The OR and corresponding 95% CI were used to assess the strength of the association between *MTHFR* C677T and A1298C polymorphisms and clinical response. Three genetic models including the allele model (C677T: T vs C; A1298C: C vs A), dominant model (C677T: CT +TT vs CC; A1298C: AC +CC vs AA) and recessive model (C677T: TT vs CC +CT; A1298C: CC vs AA +AC) were compared. The pooled OR and 95% CIs were assessed by the random effects model. The heterogeneity among studies was evaluated by the Q-test. P<0.1 was considered significant heterogeneity. I^2 statistic was also calculated to quantify the heterogeneity: I^2 <25%, I^2=25%–50%, I^2=50%–75% and I^2 >75%, indicated no heterogeneity, moderate heterogeneity, large heterogeneity and extreme heterogeneity, respectively. Subgroup analysis was carried out based on cancer type (gastric cancer and oesophageal cancer), ethnicity (Caucasians and Asians) and study design (prospective and retrospective). The sensitivity analysis was performed by the sequential omission of individual studies to assess the stability of the results. The publication bias was detected using Begg-Mazumdar adjusted rank correlation test and Egger's regression test. All statistical analyses were conducted with the software STATA V.12.0.

RESULTS

Characteristics of the included studies

As shown in figure 1, a total of 113 relevant publications were retrieved from the databases. According to the inclusion/exclusion criteria, data from 12 studies that investigated the association between the *MTHFR* C677T and A1298C polymorphisms and response to fluoropyrimidine-based chemotherapy in oesophagogastric cancer were collected for the meta-analysis.[12–23] The eligible studies were published between 2006 and 2017, and sample sizes ranged from 52 to 369 (table 1). Among these publications, four studies (33.3%) were conducted prospectively; nine studies were in Caucasians, and three in Asians; seven were reports on gastric cancer, four on oesophageal cancer and one on oesophagogastric cancer (table 1). In the studies, responders were defined as patients with complete response, partial response or no recurrence, and non-responders were defined as patients with stable disease, progressive disease or early recurrence. Of the eligible studies, 12 reports including 2020 patients reported tumour response events associated with

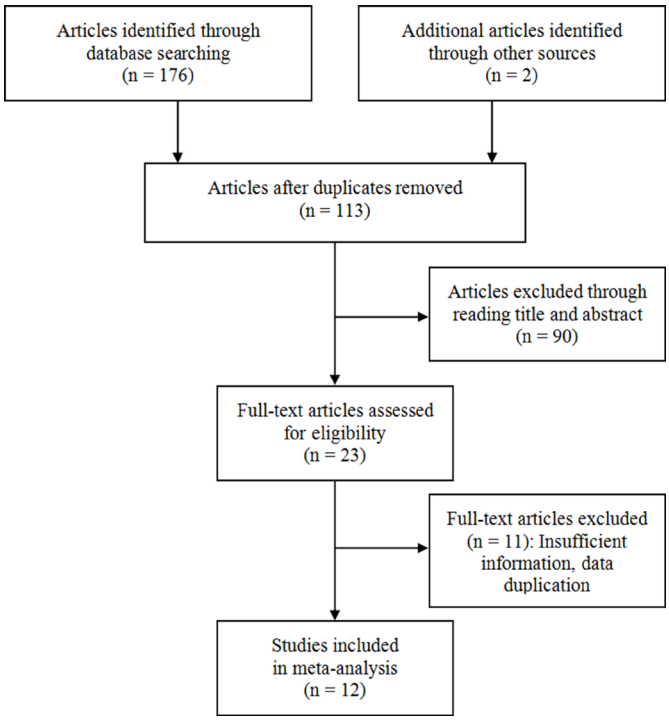

Figure 1 Flow diagram of study selection.

MTHFR C677T polymorphism, and 5 studies provided 1183 patients for testing the association of *MTHFR* A1298C variant with response to chemotherapy (table 1). The quality of each eligible article was assessed by the NOS, and all studies received a high NOS score (≥5, data not shown).

Meta-analysis results

The main results of meta-analysis and heterogeneity test for *MTHFR* C677T were summarised in table 2. No significant correlation was found between *MTHFR* C677T polymorphism and response to fluoropyrimidine-based chemotherapy in all of the three genetic models: allele model (OR 0.93, 95% CI 0.76 to 1.15) (figure 2A), dominant model (OR 0.94, 95% CI 0.72 to 1.23) (online supplementary figure S1A) and recessive model (OR 1.02, 95% CI 0.74 to 1.39) (online supplementary figure S1B). The results of Q-test and I^2 statistic indicated moderate heterogeneity in allele and dominant models (P_Q >0.1, 25% < I^2<50%), and no significant heterogeneity under the recessive model (P_Q=0.356, I^2=9.4%).

In the stratified analysis by cancer type, seven studies were used to evaluate the association of *MTHFR* C677T polymorphism with response to fluoropyrimidine-based chemotherapy in gastric cancer, and four studies in oesophageal cancer. As shown in table 2, no significant association was observed in both gastric and oesophageal cancer under all genetic models. The similar results were obtained in the stratified analysis according to ethnicity or study design. The association was still not significantly altered between *MTHFR* C677T polymorphism and response to fluoropyrimidine-based chemotherapy in all subgroups (table 2).

For the association between *MTHFR* A1298C polymorphism and response to fluoropyrimidine-based chemotherapy, the pooled results indicated no significant association in all genetic models (table 3, figure 2B, and online supplementary figure S1C,D). Large heterogeneity was observed in allele and dominant contrasts (P_Q <0.1, I^2 >50%; table 3). Moreover, as indicated in table 3, when stratified by cancer type, ethnicity or study design, there was no significant association in all subgroups.

Sensitivity analysis

The influence of any single study on the overall results was analysed by gradual deletion of individual studies. As shown in figure 3A,B and online supplementary figure S2A–D, no significant difference was observed when any of the studies was excluded in all of the three genetic models, indicating the reliability and stability of the results.

Publication bias

The Egger's regression test and Begg's test were performed to evaluate the publication bias. As shown in figure 4A,B and online supplementary figure S3A–S3D, the shape of the funnel plot was symmetrical, and the p values were all greater than 0.05 in both Begg's test and Egger's test under all genetic models (tables 2 and 3), suggesting the absence of significant publication bias in the overall meta-analysis.

DISCUSSION

There are many factors influencing the chemosensitivity to fluoropyrimidine drugs; among them, the polymorphism of metabolism-related genes of fluoropyrimidine is one of the most pivotal factors.[24–27] Despite the biological rationale suggesting a role of *MTHFR* polymorphisms in affecting the efficacy of fluoropyrimidines, the results of genetic polymorphism studies related to the response to fluoropyrimidine-based chemotherapy in patients with gastric and oesophageal cancer are still conflicting. Zhang *et al* has conducted a retrospective comparative exploratory study on *MTHFR* polymorphisms in gastric cancer, and concluded that the homozygous genotypes rs2274976G/G and rs1801131A/A were over-represented in responsive patients; carriers of the rs2274976A allele genotypes (G/A and A/A) and of the rs1801131C allele genotypes (A/C and C/C) were prevalent in non-responsive patients.[19] These results suggested that polymorphisms of the *MTHFR* gene could be used as predictors for the response to fluorouracil-based chemotherapy in gastric cancer. However, the studies performed by several other research groups in oesophagogastric cancer found no significant correlation between them.[22 23] To further comprehensively evaluate the effect of *MTHFR* C677T and A1298C polymorphisms on fluoropyrimidine-based chemotherapy in patients with oesophagogastric cancer, a meta-analysis including 12 studies was performed. The results of pooled data suggested that there was no

Table 1 Characteristics of the studies included in the meta-analysis

Study (year)	Ethnicity	Clinical data gathering	Patients, n (male%)	Age, mean (range)	Cancer type	Chemotherapy regimens	Definition of responders	Definition of non- responders	MTHFR SNP	Method of MTHFR SNP analysis	Hardy-Weinberg equilibrium reported and in equilibrium?
Ott *et al*[12] (2006)	Caucasian	Retrospective	135 (71.8)	56 (23–70)	Advanced GC	PLF, E-PLF, paclitaxel-PLF	CR, PR	SD, PD	C677T	TaqMan assay	Not reported
Sarbia *et al*13 (2006)	Caucasian	Retrospective	68 (–)	_	Oesophageal squamous cell cancer	FLEP	CR, PR	SD, PD	C677T	PCR-HRM	Not reported
Goekkurt *et al*[14] (2006)	Caucasian	Retrospective	52 (65.4)	56 (27–82)	Advanced GC	5-FU+ cisplatin+FA	CR, PR	SD, PD	C677T	PCR-RFLP	Not reported
Ruzzo *et al*[15] (2006)	Caucasian	Prospective	175 (56.6)	61 (38–79)	Advanced GC	Fluorouracil/ cisplatin	CR, PR	SD, PD	C677T	PCR-RFLP	Not reported
Wu *et al*[16] (2006)	Caucasian	Retrospective	210 (86.67)	61 (32–79)	Oesophageal cancer	Fu+cisplatin+ paclitaxel	No recurrence	Recurrence	C677T A1298C	TaqMan assay	Not reported
Goekkurt *et al*[17] (2009)	Caucasian	Prospective	134 (68.6)	64 (27–86)	Advanced GC	FLO, FLP	CR, PR	SD, PD	C677T A1298C	PCR-RFLP	Yes
Chen *et al*[18] (2010)	Asian	Retrospective	98 (70.4)	_	Oesophageal squamous cell cancer	Cisplatin/ fluorouracil	CR, PR	SD, PD	C677T	Sequencing	Yes
Zhang *et al*[19] (2014)	Asian	Retrospective	362 (77.3)	57.5 (18–82)	GC	F, FP, FT, TPF, EOF and others	CR, PR	SD, PD	C677T A1298C rs2274976 GA	MALDI-TOF-MS	Yes
Blank *et al*[20] (2014)	Caucasian	Retrospective	369 (83.7)	_	Oesophagogastric cancer	OLF/PLF, EOX, FLOT	CR, PR	SD, PD	C677T A1298C	PCR-based KASP genotyping chemistry	Yes
Liu *et al*[21] (2016)	Asian	Retrospective	108 (59.2)	_	mGC	EOF	CR, PR	SD, PD	C677T A1298C	TaqMan assay	Yes
Meulendijks *et al*[22] (2017)	Caucasian	Prospective	185 (73)	59 (27–77)	Advanced GC	Cisplatin+ capecitabine	CR, PR	SD, PD	C677T	Sequencing/PCR-RFLP	Yes
Gusella *et al*[23] (2017)	Caucasian	Prospective	124 (83.9)	60 (42–74)	Advanced oesophageal cancer	Fluorouracil+ docetaxel+ cisplatin	No recurrence	Recurrence	C677T	PCR-RFLP	Yes

5-FU, 5-fluorouracil; CR, complete response; EOF, 5-FU/capecitabine/S-1+cisplatin/oxaliplatin+epirubicin; EOX, epirubicin+oxaliplatin+capecitabin; E-PLF, epirubicin+cisplatin+leucovorin+5-FU; F, 5-FU/capecitabine/S-1; FA, folinic acid; FLEP, 5-FU+folinic acid+etoposide+cisplatin; FLP, 5-FU+leucovorin+cisplatin;FLO, 5-FU+leucovorin+oxaliplatin; FP, 5-FU/capecitabine/S-1+c isplatin/oxaliplatin; FT, 5-FU/capecitabine/S-1+d ocetaxel/paclitaxel; FLOT, 5-fluoruracil+folinic acid+oxaliplatin+docetaxel; GC, gastric cancer; HRM, High Resolution Melting; KASP, a competitive allele-specific PCR genotyping system; MALDI-TOF-MS, matrixassisted laser desorption/ionization time-of-flight mass spectrometry; MGC, metastatic gastric cancer; MTHFR, methylenetetrahydrofolate reductase; OLF, oxaliplatin/cisplatin+folinic acid+fluorouracil; PD, progressive disease; PR, partial response; PLF, cisplatin+leucovorin+5-FU; RFLP, restriction fragment length polymorphism; TPF, 5-FU/capecitabine/S-1+c isplatin/oxaliplatin+docetaxel/paclitaxel; SD, stable disease; SNP, single nucleotide polymorphisms.

Table 2 OR with the corresponding 95% CI, heterogeneity results, Begg' test and Egger' test for genetic contrasts of methylenetetrahydrofolate reductase C677T

Models	Population	No studies	Random effects OR (95% CI)	P values (Q-test)	I^2 (%)	Begg' test	Egger' test
T versus C	All	9	0.93 (0.76 to 1.15)	0.109	38.9	0.251	0.355
	GC	6	0.85 (0.61 to 1.17)	0.058	53.3	0.452	0.495
	EC	2	1.00 (0.66 to 1.53)	0.226	31.7	1.000	–
	Caucasians	7	0.99 (0.78 to 1.25)	0.167	34.2	0.548	0.404
	Asians	2	0.72 (0.37 to 1.41)	0.081	67.1	1.000	–
	Prospective	3	1.06 (0.74 to 1.51)	0.185	40.7	1.000	0.711
	Retrospective	6	0.86 (0.65 to 1.14)	0.105	45.1	0.452	0.190
Dominant model	All	11	0.94 (0.72 to 1.23)	0.131	33.4	0.533	0.836
	GC	6	0.75 (0.46 to 1.22)	0.043	56.4	1.000	0.835
	EC	4	1.15 (0.78 to 1.71)	0.878	0.0	1.000	0.939
	Caucasians	8	1.02 (0.76 to 1.37)	0.278	19.2	0.108	0.400
	Asians	3	0.78 (0.39 to 1.52)	0.097	57.1	1.000	0.862
	Prospective	3	1.31 (0.84 to 2.04)	0.442	0.0	0.296	0.231
	Retrospective	8	0.83 (0.61 to 1.14)	0.155	34.2	0.902	0.588
Recessive model	All	10	1.02 (0.74 to 1.39)	0.356	9.4	1.000	0.929
	GC	7	1.05 (0.75 to 1.47)	0.454	0.0	0.764	0.944
	EC	2	0.93 (0.21 to 4.19)	0.047	74.6	1.000	–
	Caucasians	8	0.98 (0.67 to 1.44)	0.368	8.1	0.386	0.408
	Asians	2	0.95 (0.39 to 2.29)	0.147	52.5	1.000	–
	Prospective	4	0.87 (0.56 to 1.36)	0.405	0.0	0.734	0.768
	Retrospective	6	1.12 (0.71 to 1.77)	0.298	17.8	1.000	0.924

EC, oesophageal cancer; GC, gastric cancer.

significant association between *MTHFR* C677T and A1298C polymorphism and the clinical response to fluoropyrimidine-based chemotherapy in sufferers with gastric and oesophageal cancer under all three genetic models. In the subgroup analysis based on cancer type, ethnicity or study design, the correlation was still not detected. This result was similar to the meta-analysis performed by Zintzaras *et al* in colorectal cancer, in which it showed that *MTHFR* C677T and A1298C gene polymorphisms could not be considered as reliable predictors of response to fluorouracil chemotherapy in patients with colorectal cancer.[28]

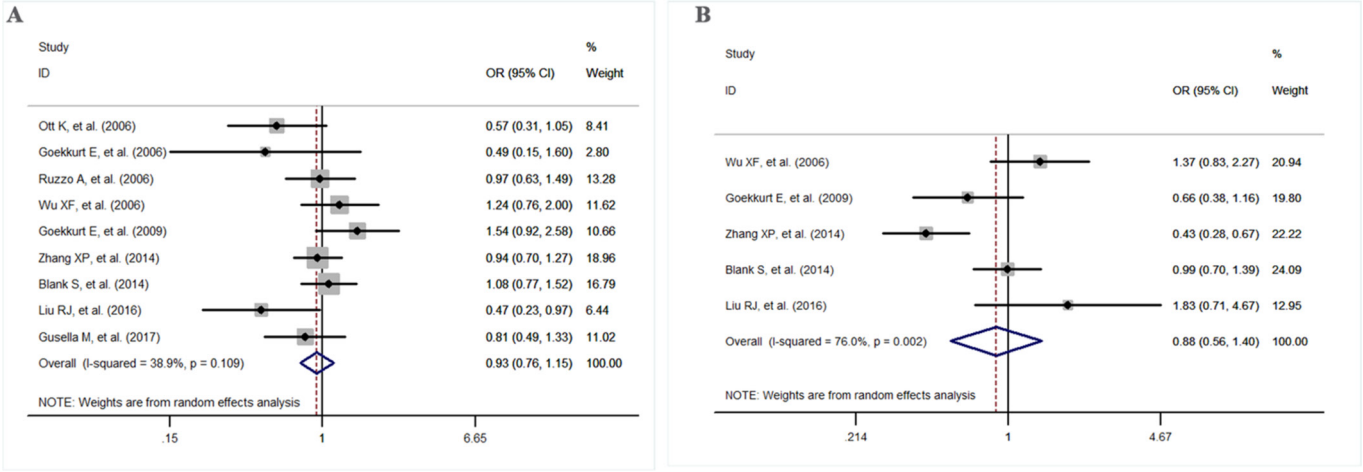

Figure 2 Forest plot. (A) Forest plot for the allele contrast of methylenetetrahydrofolate reductase (*MTHFR*) C677T variant and response to fluoropyrimidine-based chemotherapy; (B) Forest plot for the allele contrast of *MTHFR* A1298C variant and response to fluoropyrimidine-based chemotherapy.

Table 3 OR with the corresponding 95% CI, heterogeneity results, Begg' test and Egger' test for genetic contrasts of methylenetetrahydrofolate reductase A1298C

Models	Population	No studies	Random effects OR (95% CI)	P values (Q-test)	I^2 (%)	Begg' test	Egger' test
C versus A	All	5	0.88 (0.56 to 1.40)	0.002	76.0	0.806	0.501
	GC	3	0.72 (0.36 to 1.44)	0.022	73.7	0.296	0.070
	EC	1					
	Caucasians	3	0.98 (0.69 to 1.40)	0.162	45.1	1.000	0.958
	Asians	2	0.84 (0.21 to 3.40)	0.007	86.5	1.000	–
	Prospective	1					
	Retrospective	4	0.96 (0.54 to 1.70)	0.001	81.1	0.308	0.464
Dominant model	All	5	0.80 (0.47 to 1.35)	0.007	71.8	0.462	0.332
	GC	3	0.63 (0.30 to 1.35)	0.038	69.5	0.296	0.310
	EC	1					
	Caucasians	3	0.86 (0.50 to 1.45)	0.091	58.4	1.000	0.854
	Asians	2	0.83 (0.19 to 3.63)	0.011	84.4	1.000	–
	Prospective	1					
	Retrospective	4	0.92 (0.50 to 1.69)	0.007	75.5	0.308	0.218
Recessive model	All	5	1.15 (0.50 to 2.67)	0.207	32.2	0.462	0.516
	GC	3	0.71 (0.14 to 3.59)	0.138	49.5	1.000	0.955
	EC	1					
	Caucasians	3	1.40 (0.74 to 2.64)	0.489	0.0	0.296	0.290
	Asians	2	0.43 (0.03 to 5.73)	0.146	52.6	1.000	–
	Prospective	1					
	Retrospective	4	1.08 (0.31 to 3.79)	0.120	48.6	0.308	0.606

EC, oesophageal cancer; GC, gastric cancer.

Several potential limitations of the present meta-analysis should be acknowledged. First, this study was based on the reported data of the eligible study without adjustment for other covariates such as age and gender, which may result in relatively low power to estimate the real association. This is also a general problem of meta-analysis when pooling data from primary studies.[29 30] Second, the treatment of oesophagogastric cancer could also be influenced by diet, living habits, environmental exposure and pathological type of patients, while these factors were not considered in this study. Third, some stratified analysis in this account was not sufficiently large (contain only

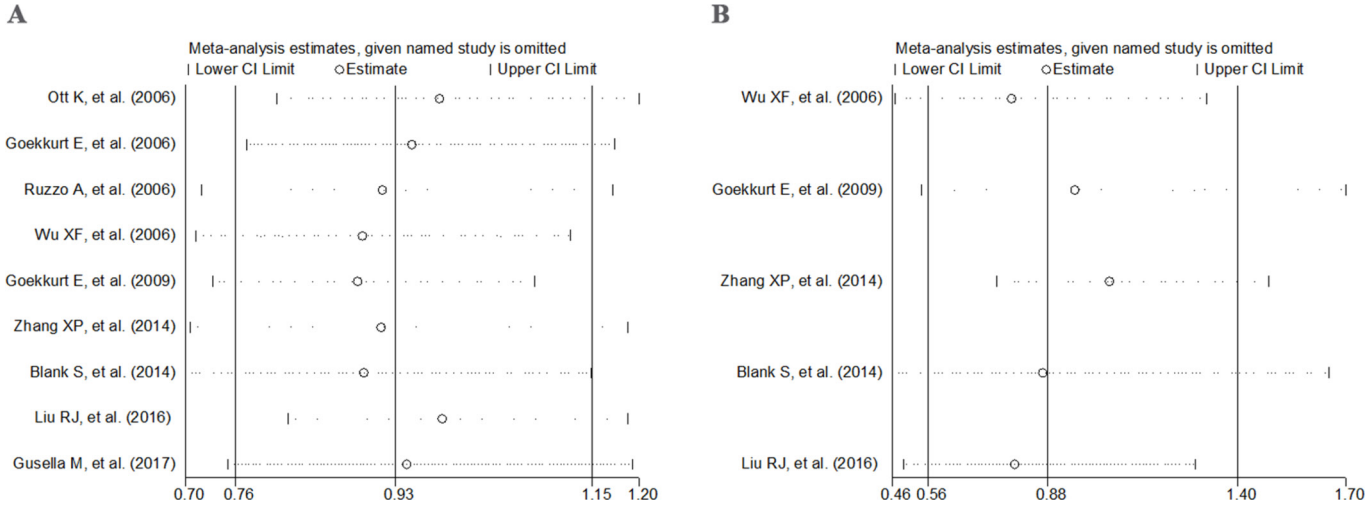

Figure 3 Sensitivity analysis. (A) Sensitivity analysis for the allele contrast of *MTHFR* C677T polymorphism. (B) Sensitivity analysis for the allele contrast of *MTHFR* A1298C polymorphism.

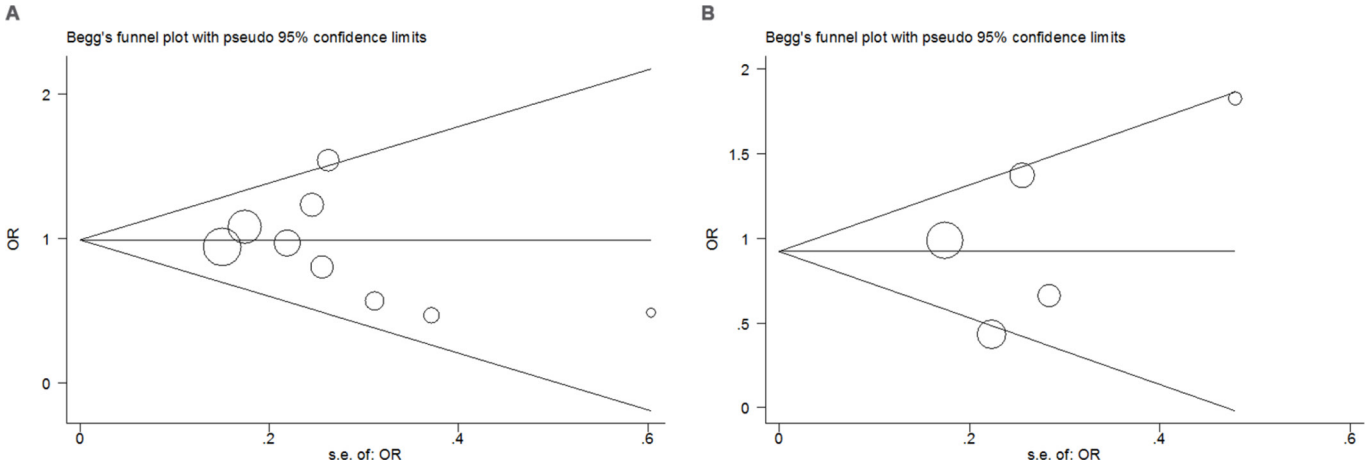

Figure 4 Publication bias. (A) Begg's funnel plot of the publication bias in the allele model of *MTHFR* C677T polymorphism. (B) Begg's funnel plot of the publication bias in the allele model of *MTHFR* A1298C polymorphism.

two studies). Therefore, the association in the relevant subgroup analysis was unconvincing, and needed to be further estimated. Finally, heterogeneity was a noticeable problem in this meta-analysis, and we found moderate or large heterogeneity in most of the comparison. Potential sources of heterogeneity were not found by the sensitivity analysis. When stratified by cancer type, ethnicity and study design, the heterogeneity just greatly decreased in partial subgroups (tables 2 and 3).

Multiple factors may contribute to the heterogeneity in this study. Treatment setting may be one the most pivotal influence factors. The eligible studies covered all stages of management in oesophagogastric cancer, including neoadjuvant (preoperative), adjuvant (postoperative) and palliative therapy. Meanwhile, in the chemotherapy regimens, fluoropyrimidines were all combined with other agents. The difference in treatment type and combination regimen may cause the diversities in efficacy, thus contributing to the heterogeneity among studies. Folate intake status is also a factor influencing the efficacy of fluoropyrimidine drugs.[31 32] MTHFR is a critical enzyme in folate-metabolising pathway, and folate status may affect the association of *MTHFR* polymorphisms with response to fluoropyrimidine-based treatment through gene–nutrition interaction. However, this effect cannot be assessed unless specifically sought and accounted for in the individual studies. In addition, the administration mode of fluoropyrimidines may also be one of the causes of heterogeneity. Fluoropyrimidines act in two different ways (bolus/infusion administration). Bolus fluoropyrimidines may incorporate into RNA and preclude protein synthesis, whereas continuous infusion exerts its major effect on TS.[33] The eligible studies in this meta-analysis used both modes of fluorouracil administration.

CONCLUSION

In summary, we demonstrate that *MTHFR* C677T and A1298C polymorphisms cannot be considered as reliable factors for predicting the clinical response to fluoropyrimidine-based chemotherapy in patients with oesophagogastric cancer. However, the results in present meta-analysis should be interpreted cautiously due to the existence of heterogeneity. Therefore, well-designed prospective studies based on larger sample sizes are warranted to validate the present findings. Additionally, in view of the fact that fluoropyrimidines exert their effects through a multistep, multigenic cascade, hence, composite pharmacogenomics analysis may be more precise for efficacy prediction of fluoropyrimidine-based regimens.

Contributors LZ and QF contributed equally to this work, performed the research and drafted the manuscript; LZ, QF and QP designed the research; LC, SZ and QP interpreted the results and revised the manuscript.

Funding This work was supported by the Scientific Research Subject of Health and Family Planning Commission of Sichuan Province (16PJ483), the Special Foundation for Young Scientists of Sichuan Academy of Medical Sciences & Sichuan Provincial People's Hospital (2016QN11) and the Fundamental Research Funds of Science & Technology Department of Sichuan Province (2017YSKY0001).

Competing interests None declared.

Patient consent Not required.

Provenance and peer review Not commissioned; externally peer reviewed.

Data sharing statement No additional data are available.

REFERENCES

1. Qu JL, Li X, Qu XJ, *et al*. Optimal duration of fluorouracil-based adjuvant chemotherapy for patients with resectable gastric cancer. *PLoS One* 2013;8:e83196.
2. Li W, Zhao X, Wang H, *et al*. Maintenance treatment of Uracil and Tegafur (UFT) in responders following first-line fluorouracil-based chemotherapy in metastatic gastric cancer: a randomized phase II study. *Oncotarget* 2017;8:37826–34.

3. Liu Y, Ren Z, Yuan L, *et al*. Paclitaxel plus cisplatin vs. 5-fluorouracil plus cisplatin as first-line treatment for patients with advanced squamous cell esophageal cancer. *Am J Cancer Res* 2016;6:2345–50.
4. Stein A, Arnold D, Thuss-Patience PC, *et al*. Docetaxel, oxaliplatin and capecitabine (TEX regimen) in patients with metastatic gastric or gastro-esophageal cancer: results of a multicenter phase I/II study. *Acta Oncol* 2014;53:392–8.
5. Longley DB, Harkin DP, Johnston PG. 5-fluorouracil: mechanisms of action and clinical strategies. *Nat Rev Cancer* 2003;3:330–8.
6. Bueno O, Molloy AM, Fernandez-Ballart JD, *et al*. Common Polymorphisms That Affect Folate Transport or Metabolism Modify the Effect of the MTHFR 677C > T Polymorphism on Folate Status. *J Nutr* 2016;146:1–8.
7. Goyette P, Sumner JS, Milos R, *et al*. Human methylenetetrahydrofolate reductase: isolation of cDNA, mapping and mutation identification. *Nat Genet* 1994;7:195–200.
8. Pereira AC, Schettert IT, Morandini Filho AA, *et al*. Methylenetetrahydrofolate reductase (MTHFR) c677t gene variant modulates the homocysteine folate correlation in a mild folate-deficient population. *Clin Chim Acta* 2004;340:99–105.
9. Friedman G, Goldschmidt N, Friedlander Y, *et al*. A common mutation A1298C in human methylenetetrahydrofolate reductase gene: association with plasma total homocysteine and folate concentrations. *J Nutr* 1999;129:1656–61.
10. Zhao Y, Li X, Kong X. MTHFR C677T polymorphism is associated with tumor response to preoperative chemoradiotherapy: a result based on previous reports. *Med Sci Monit* 2015;21:3068–76.
11. Wang Z, Chen JQ, Liu JL, *et al*. Polymorphisms in ERCC1, GSTs, TS and MTHFR predict clinical outcomes of gastric cancer patients treated with platinum/5-Fu-based chemotherapy: a systematic review. *BMC Gastroenterol* 2012;12:137.
12. Ott K, Vogelsang H, Marton N, *et al*. The thymidylate synthase tandem repeat promoter polymorphism: A predictor for tumor-related survival in neoadjuvant treated locally advanced gastric cancer. *Int J Cancer* 2006;119:2885–94.
13. Sarbia M, Stahl M, von Weyhern C, *et al*. The prognostic significance of genetic polymorphisms (Methylenetetrahydrofolate Reductase C677T, Methionine Synthase A2756G, Thymidilate Synthase tandem repeat polymorphism) in multimodally treated oesophageal squamous cell carcinoma. *Br J Cancer* 2006;94:203–7.
14. Goekkurt E, Hoehn S, Wolschke C, *et al*. Polymorphisms of glutathione S-transferases (GST) and thymidylate synthase (TS)--novel predictors for response and survival in gastric cancer patients. *Br J Cancer* 2006;94:281–6.
15. Ruzzo A, Graziano F, Kawakami K, *et al*. Pharmacogenetic profiling and clinical outcome of patients with advanced gastric cancer treated with palliative chemotherapy. *J Clin Oncol* 2006;24:1883–91.
16. Wu X, Gu J, Wu TT, *et al*. Genetic variations in radiation and chemotherapy drug action pathways predict clinical outcomes in esophageal cancer. *J Clin Oncol* 2006;24:3789–98.
17. Goekkurt E, Al-Batran SE, Hartmann JT, *et al*. Pharmacogenetic analyses of a phase III trial in metastatic gastroesophageal adenocarcinoma with fluorouracil and leucovorin plus either oxaliplatin or cisplatin: a study of the arbeitsgemeinschaft internistische onkologie. *J Clin Oncol* 2009;27:2863–73.
18. Chen J, Yf H, Cs J, *et al*. Prognostic value of the ERCC1 and TS genetic polymorphisms in advanced esophageal cancer treated with cisplatin/fluorouracil chemotherapy. *Tumor* 2010;30:314–21.
19. Zhang X, Bai Z, Chen B, *et al*. Polymorphism of methylenetetrahydrofolate reductase gene is associated with response to fluorouracil-based chemotherapy in Chinese patients with gastric cancer. *Chin Med J* 2014;127:3562–7.
20. Blank S, Rachakonda S, Keller G, *et al*. A retrospective comparative exploratory study on two methylentetrahydrofolate reductase (MTHFR) polymorphisms in esophagogastric cancer: the A1298C MTHFR polymorphism is an independent prognostic factor only in neoadjuvantly treated gastric cancer patients. *BMC Cancer* 2014;14:58.
21. Liu R, Zhao X, Liu X, *et al*. Influences of ERCC1, ERCC2, XRCC1, GSTP1, GSTT1, and MTHFR polymorphisms on clinical outcomes in gastric cancer patients treated with EOF chemotherapy. *Tumour Biol* 2016;37:1753–62.
22. Meulendijks D, Rozeman EA, Cats A, *et al*. Pharmacogenetic variants associated with outcome in patients with advanced gastric cancer treated with fluoropyrimidine and platinum-based triplet combinations: a pooled analysis of three prospective studies. *Pharmacogenomics J* 2017;17:441–51.
23. Gusella M, Giacopuzzi S, Bertolaso L, *et al*. Genetic prediction of long-term survival after neoadjuvant chemoradiation in locally advanced esophageal cancer. *Pharmacogenomics J* 2017;17:252–7.
24. Hur H, Kang J, Kim NK, *et al*. Thymidylate synthase gene polymorphism affects the response to preoperative 5-fluorouracil chemoradiation therapy in patients with rectal cancer. *Int J Radiat Oncol Biol Phys* 2011;81:669–76.
25. Terrazzino S, Cargnin S, Del Re M, *et al*. DPYD IVS14+1G>A and 2846A>T genotyping for the prediction of severe fluoropyrimidine-related toxicity: a meta-analysis. *Pharmacogenomics* 2013;14:1255–72.
26. Meulendijks D, Henricks LM, Sonke GS, *et al*. Clinical relevance of DPYD variants c.1679T>G, c.1236G>A/HapB3, and c.1601G>A as predictors of severe fluoropyrimidine-associated toxicity: a systematic review and meta-analysis of individual patient data. *Lancet Oncol* 2015;16:1639–50.
27. Di Francia R, Cimino L, Berretta M. Genetic variants influencing fluoropyrimidine based-therapy and available methods to detect them. *Eur Rev Med Pharmacol Sci* 2012;16:285–98.
28. Zintzaras E, Ziogas DC, Kitsios GD, *et al*. MTHFR gene polymorphisms and response to chemotherapy in colorectal cancer: a meta-analysis. *Pharmacogenomics* 2009;10:1285–94.
29. Lu SC, Zhong JH, Tan JT, *et al*. Association between COX-2 gene polymorphisms and risk of hepatocellular carcinoma development: a meta-analysis. *BMJ Open* 2015;5:e008263.
30. Yokoyama A, Kato H, Yokoyama T, *et al*. Genetic polymorphisms of alcohol and aldehyde dehydrogenases and glutathione S-transferase M1 and drinking, smoking, and diet in Japanese men with esophageal squamous cell carcinoma. *Carcinogenesis* 2002;23:1851–9.
31. Petrelli N, Herrera L, Rustum Y, *et al*. A prospective randomized trial of 5-fluorouracil versus 5-fluorouracil and high-dose leucovorin versus 5-fluorouracil and methotrexate in previously untreated patients with advanced colorectal carcinoma. *J Clin Oncol* 1987;5:1559–65.
32. Roy Moulik N, Kumar A, Agrawal S, *et al*. Role of folate status and methylenetetrahydrofolate reductase genotype on the toxicity and outcome of induction chemotherapy in children with acute lymphoblastic leukemia. *Leuk Lymphoma* 2015;56:1379–84.
33. Sobrero AF, Aschele C, Bertino JR. Fluorouracil in colorectal cancer--a tale of two drugs: implications for biochemical modulation. *J Clin Oncol* 1997;15:368–81.