Clinical and Experimental Obstetrics & Gynecology

Clin. Exp. Obstet. Gynecol. **2023**; 50(12): 266
https://doi.org/10.31083/j.ceog5012266

Systematic Review

MTHFR A1298C Polymorphism and Risk of Preeclampsia: A Meta-Analysis

Yong Hu[1,2], Ao Wang[2], Ke Yi[2,*]

[1]Department of Pediatrics, Key Laboratory of Birth Defects and Related Diseases of Women and Children of the Ministry of Education, West China Second University Hospital, Sichuan University, 610041 Chengdu, Sichuan, China
[2]Key Laboratory of Obstetrics & Gynecologic and Pediatric Diseases and Birth Defects of the Ministry of Education, West China Second University Hospital, Sichuan University, 610041 Chengdu, Sichuan, China
*Correspondence: 341749797@qq.com (Ke Yi)
Academic Editor: George Daskalakis
Submitted: 31 August 2023 Revised: 5 October 2023 Accepted: 12 October 2023 Published: 15 December 2023

Abstract

Background: Published research findings regarding the relationship between the methylenetetrahydrofolate reductase (*MTHFR*) A1298C polymorphism and the risk of preeclampsia (PE) have generated conflicting results. A meta-analysis was conducted to investigate whether the *MTHFR* A1298C polymorphism is associated with preeclampsia. **Methods**: We conducted a systematic search across several databases, including PubMed, Embase, Web of science, China National Knowledge Infrastructure, and Chinese Biomedicine Databases, to identify relevant studies. We then calculated pooled odds ratios (ORs) and their corresponding 95% confidence intervals (CIs) to assess the association between the *MTHFR* A1298C polymorphism and preeclampsia (PE) risk. **Results**: A total of 11 studies were enrolled in this meta-analysis. The pooled analyses revealed that *MTHFR* A1298C polymorphism significantly decreased the risk of PE (allele contrast (A (alanine) *vs.* C (glutamate)): OR, 0.81; 95% CI, 0.71–0.93, $p = 0.207$; homozygote (AA *vs.* CC): OR, 0.57; 95% CI, 0.40–0.79, $p = 0.056$; heterozygote (AC *vs.* CC): OR, 0.62; 95% CI, 0.45–0.87, $p = 0.010$; dominant model (AA + AC *vs.* CC): OR, 0.59; 95% CI, 0.43–0.81, $p = 0.031$; recessive model (AA *vs.* AC + CC): OR, 0.83; 95% CI, 0.70–0.98), $p = 0.817$. **Conclusion**: Present meta-analysis reveals that *MTHFR* A1298C variant may serve as genetic biomarkers of PE. The study was registered on PROSPERO (https://www.crd.york.ac.uk/prospero/), registration number: CRD42023459681.

Keywords: *MTHFR*; polymorphisms; preeclampsia; meta-analysis

1. Introduction

Preeclampsia (PE) is the leading contributor to maternal and perinatal mortality globally [1]. It is responsible for an estimated 46,000 maternal deaths and roughly 500,000 fetal and newborn fatalities yearly [2]. Women in low- and middle-income countries bear a disproportionate burden of this disease [3]. The World Health Organization (WHO) has reported significantly higher numbers in developing countries than in developed ones, with approximately seven times higher figures. In Asian regions, gestational hypertensive disorders account for 9.1% of maternal deaths [4,5]. A variety of environmental factors, including smoking habits, hypertension, diabetes mellitus, advanced maternal age, and obesity, have been linked to PE [6,7]. However, a great deal of PE patients manifest without the presence of any of these identified risk factors, which suggests that environmental factors are not the only contributors. Genetic factors also appear to play a significant role in PE onset and progression [8–10].

Single-nucleotide polymorphisms (SNPs) represent variations in the genome that can change biological reactions and disease susceptibility. Several SNPs have been investigated in relation to PE [11–14]. The enzyme methylenetetrahydrofolate reductase (*MTHFR*) plays a key role in the metabolism of homocysteine (HCy). It facilitates 5,10-MTHF conversion to 5-MTHF, which leads to the vitamin B12-mediated transformation of HCy to methionine [15,16]. Decreased *MTHFR* levels or activity as a result of certain gene mutations can cause slight to moderate plasma HCy level elevations [17]. Several *MTHFR* gene mutations have been identified as potential links to PE, with the C677T and A1298C the most frequently observed. The most frequently observed mutation is the C-to-T change at nucleotide 677 (C677T) in exon 4, which leads to a substitution ofalanine with valine in the catalytic domain of the enzyme, and decreases its activity. Another mutation at position 1298 in exon 7 (A1298C) replaces glutamate with alanine at codon 429. This mutation also diminishes enzyme function in a similar way [18,19]. The *MTHFR* A1298C variant shows a difference in the prevalence of its allelic distribution among different populations and ethnicities. Studies on the association between preeclampsia and this condition have been conducted among Asians, Caucasians, Africans, and Americans. Correlation with the Asian and Caucasian populations was found to be significant [20,21].

MTHFR A1298C is linked to diminished levels of folate in serum, plasma, and red blood cells, as well as a

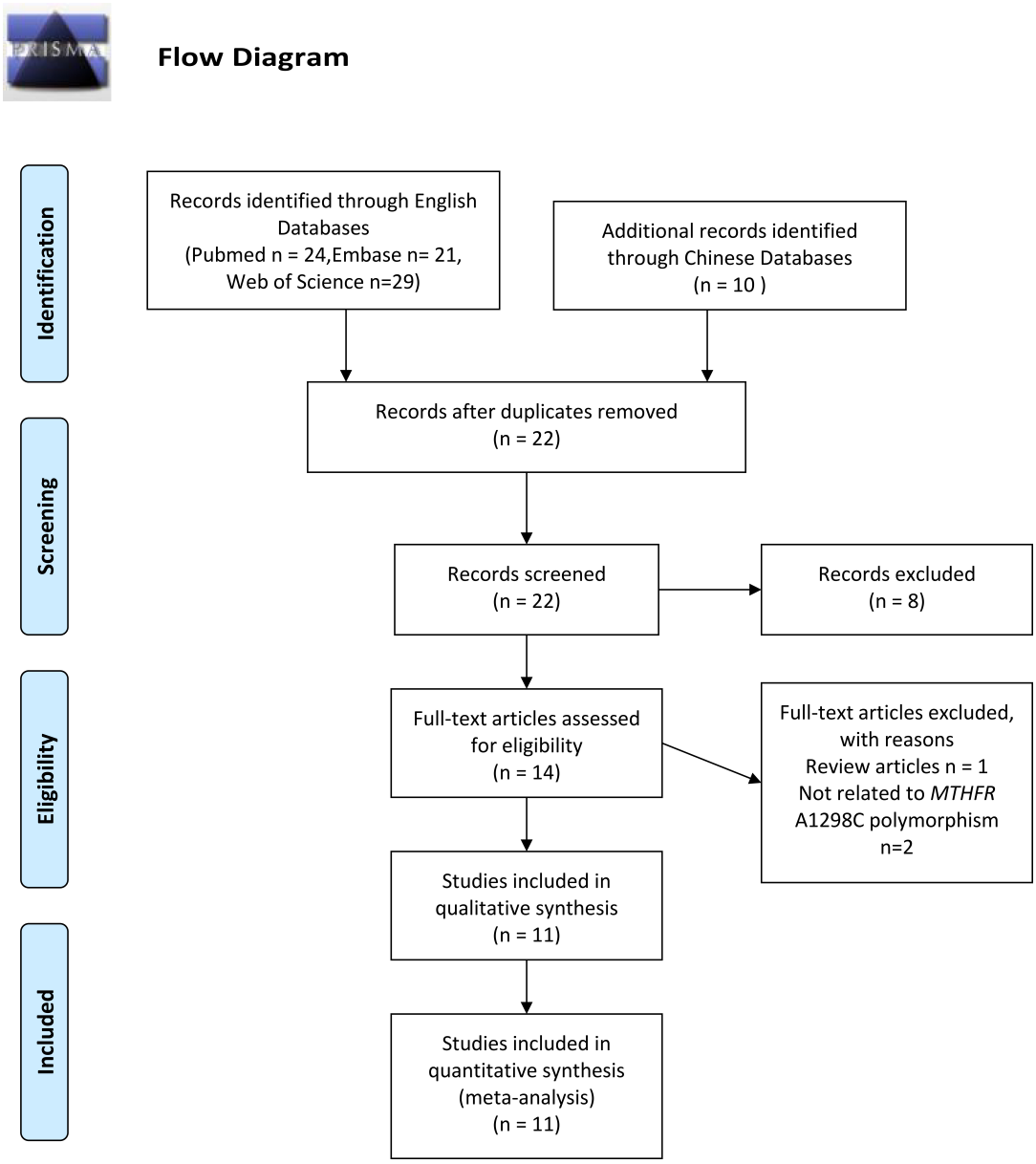

Fig. 1. Literature search and study selection procedures used for the meta-analysis of *MTHFR* A1298C genetic polymorphism and preeclampsia (PE). *MTHFR*, methylenetetrahydrofolate reductase

slight elevation in plasma total homocysteine (tHcy) concentration [22]. Considering the presence of mild hyperhomocysteinemia in women with preeclampsia, the *MTHFR* A1298C polymorphism could potentially be a genetic factor contributing to the pathophysiology of preeclampsia [23,24]. In cases of preeclampsia, there is a confirmed hereditary element: women born from preeclamptic pregnancies face an elevated risk of developing the condition during their own pregnancies [10]. Although genetic predisposition is a significant factor in the development of preeclampsia, efforts to establish associations between *MTHFR* A1298C and preeclampsia have generated inconsistent findings [25–28]. As a consequence, a meta-analysis was conducted on the published case-control studies to more closely examine the relationship between the *MTHFR* A1298C polymorphism and PE susceptibility.

2. Materials and Methodology

2.1 Publication Search

A comprehensive search was conducted to find articles that have explored the association between *MTHFR* polymorphisms and PE risk. Databases that were consulted include PubMed, Embase, Web of science, the China Biomedical Database, and China National Knowledge Infrastructure (CNKI). The search used keywords including "*MTHFR* A1298C", "variant", "polymorphism", and "preeclampsia". The most recent update to this search was made on January 10th, 2023.

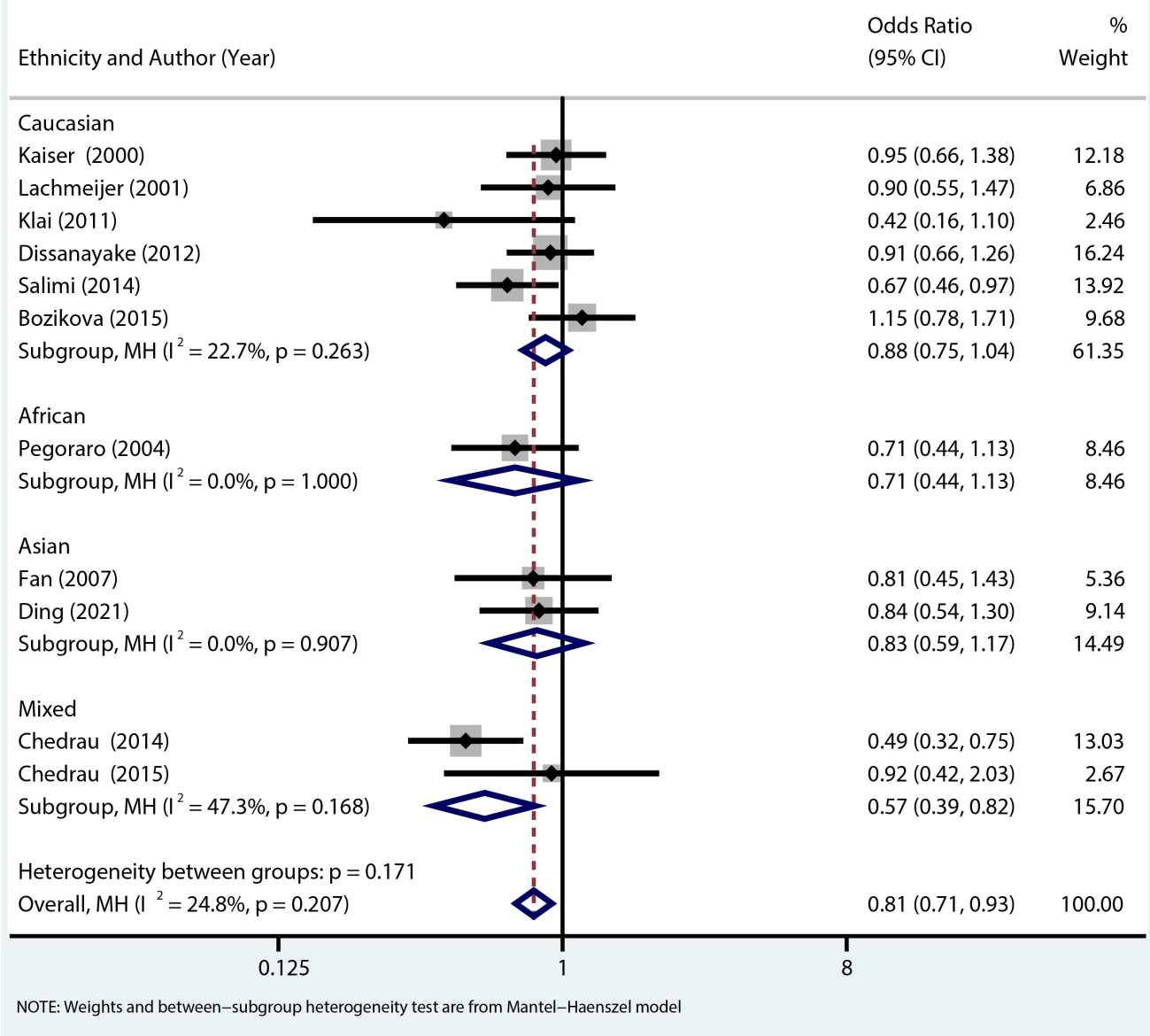

Fig. 2. Forest plots of odds ratios (ORs) with 95% confidence intervals (CIs) for *MTHFR* A1298C polymorphism and PE risk stratified by ethnicity. Note: weights and between-subgroup heterogeneity test are from Mantel-Haenszel model. MH, Mantel-Haenszel model.

2.2 Inclusion and Exclusion Criteria

Preeclampsia is defined as high blood pressure (140/90 mmHg or higher) and proteinuria (0.3 g or higher in 24-hour urine collection) in pregnant women at least 20 weeks into gestation. The control group comprised unrelated healthy pregnant women. Pregnant individuals with twin pregnancies, diabetes, systemic lupus erythematosus, liver dysfunction, renal disease, or any other systemic diseases were excluded.

In order to be considered for inclusion, every study, regardless of sample size, needed to satisfy the following criteria: (i) evaluating the association between the *MTHFR* A1298C polymorphism and PE risk; (ii) being case-control research; and (iii) providing sufficient data for calculating a 95% confidence interval (95% CI) for the odds ratio (OR). A study was excluded if it fell within the following categories: (i) abstracts, reviews, overviews, or editorials; or (ii) lacking sufficient data.

2.3 Data Extraction

Based on the aforementioned inclusion criteria, two authors (YH and AW) independently extracted information from all eligible and qualified publications. Any discrepancies they encountered were resolved by consulting with corresponding author (KY).

The following information was obtained from all eligible publications: surname of first author, date of publication, country and race of participants, genotyping meth-

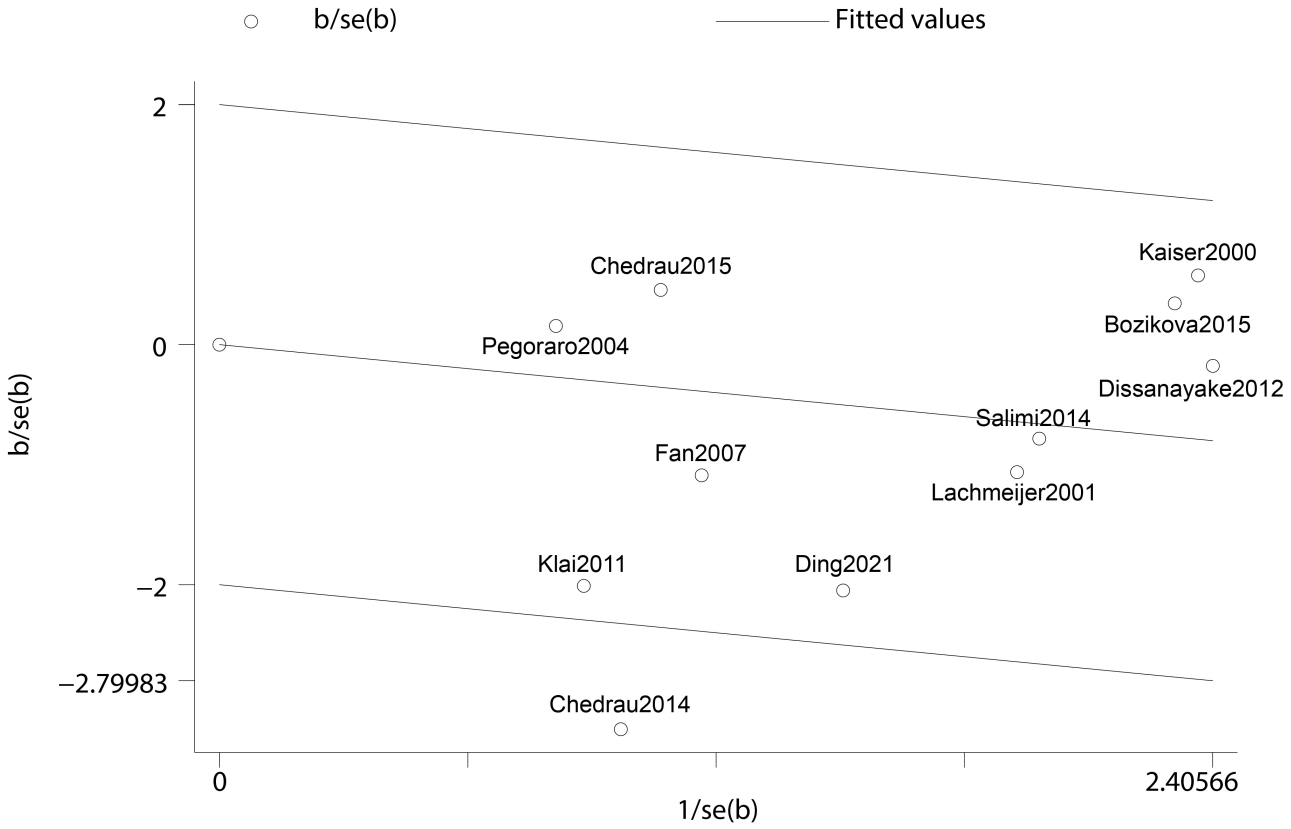

Fig. 3. Galbraith plots for the heterogeneity test of *MTHFR* A1298C polymorphism.

ods, minor allele frequencies (MAF), and Hardy-Weinberg equilibrium (HWE). Ethnic groupings were classified as African, Asian, Caucasian, or Mixed.

2.4 Statistical Analysis

Odds ratios (ORs) and corresponding 95% confidence intervals (CIs) were used for evaluating the strength of the association between the *MTHFR* A1298C polymorphism and PE risk. In addition, stratified analyses were conducted based on ethnicity.

The Cochran Q statistic and I^2 index were employed for assessing and validating heterogeneity in the studies. A *p*-value of more than 0.05 for the Q statistic indicated a lack of significant between-study heterogeneity [29]. In such cases, a fixed-effects model (utilizing the Mantel-Haenszel method) was deemed appropriate [30]. Conversely, with evidence of heterogeneity, the random effects model (employing the DerSimonian and Laird method) was employed [31].

Publication bias was investigated by performing a visual inspection of funnel plots, utilizing Egger's power-weighted regression method and Begg's rank correlation method [32,33]. A *p*-value of below 0.05 was considered to be an indicator of statistical significance [32,33]. All statistical analyses were conducted using version 13.0 of STATA software (STATA Corp., College Station, TX, USA).

3. Results

3.1 Characteristics of Studies

Following the literature review, 22 publications that were deemed worthy of thorough examination were shortlisted. After reviewing the titles and abstracts, eight publications were identified that did not satisfy the criteria for this study and they were excluded. After a close examination of the full texts of the remaining 14 articles, one article was excluded as it predominantly focused on a literature review [34]. In addition, two articles were excluded because they were unrelated to the *MTHFR* A1298C polymorphism [35,36]. Ultimately, 11 case-control studies related to the *MTHFR* A1298C polymorphism and PE were identified [17,25–28,37–42], all of them conforming to the Meta-analysis of Observational Studies in Epidemiology (MOOSE) guidelines [43]. The process of document retrieval and research selection can be seen in Fig. 1.

The detailed characteristics of the selected studies are shown in Table 1 (Ref. [17,25–28,37–42]). Of the chosen studies, six focused on Caucasian participants, two on Asian individuals, two on Mixed-race groups, and one investigated individuals of African descent. The research was conducted in Australia, China, Ecuador, Iran, South Africa, and Tunisia.

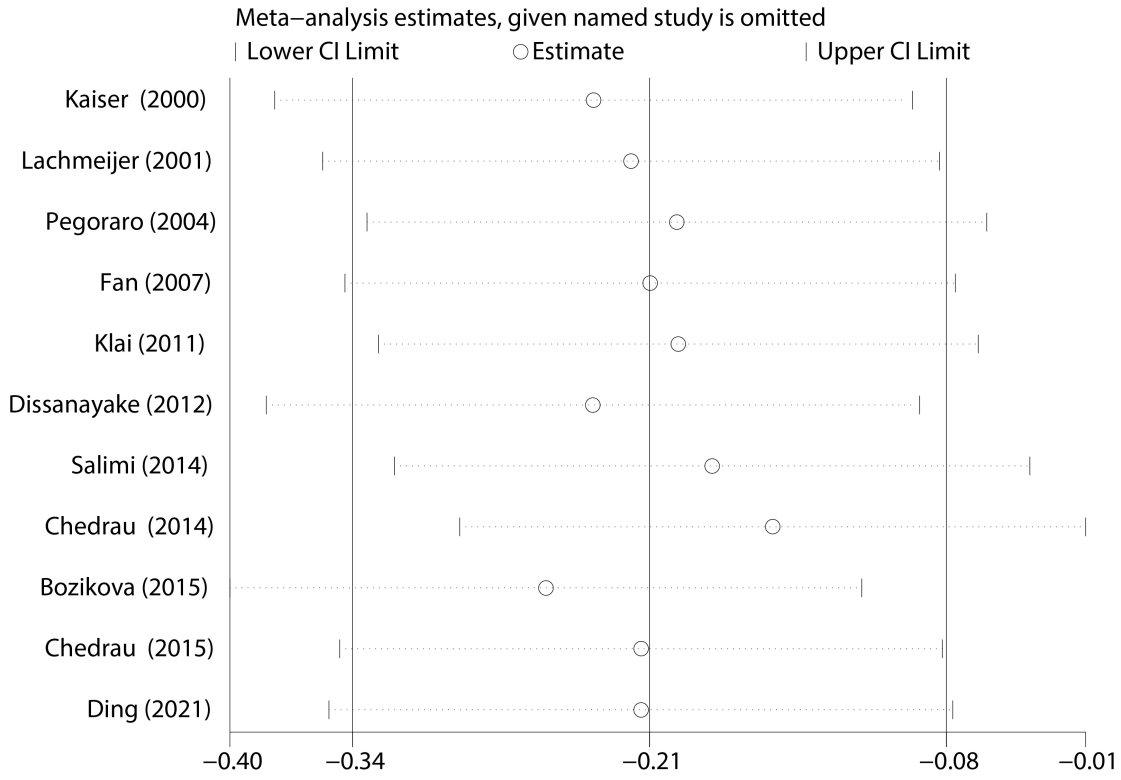

Fig. 4. Sensitivity analysis of associations between *MTHFR* A1298C polymorphism and PE risk.

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

Author	Year	Country	Ethnicity	Sample	Genotyping methods	MAF in controls	HWE
Kaiser, *et al.* [37]	2000	Australia	Caucasian	147/109	PCR-RFLP	0.353	0.502
Lachmeijer, *et al.* [38]	2001	Australia	Caucasian	204/338	PCR-RFLP	0.391	0.080
Pegoraro, *et al.* [39]	2004	South Africa	African	204/338	AS-PCR	0.062	0.421
Fan, *et al.* [25]	2007	China	Asian	64/62	PCR-RFLP	0.255	0.711
Klai, *et al.* [17]	2011	Tunisia	Caucasian	44/100	MPCR	0.045	0.061
Dissanayake, *et al.* [26]	2012	Australia	Caucasian	175/171	PCR-RFLP	0.312	0.091
Salimi, *et al.* [28]	2015	Iran	Caucasian	192/196	PCR-RFLP	0.145	0.101
Chedraui, *et al.* [27]	2014	Ecuador	Mixed	150/150	PCR-RFLP	0.136	0.212
Bozikova, *et al.* [40]	2015	China	Caucasian	120/105	TaqMan	0.357	0.553
Chedrau, *et al.* [41]	2015	Ecuador	Mixed	50/50	PCR-RFLP	0.140	0.017
Ding, *et al.* [42]	2021	China	Asian	150/150	PCR-RFLP	0.150	0.810

Abbreviations: PCR-RFLP, polymerase chain reaction-restriction fragment length polymorphism; AS-PCR, allele-specific polymerase chain reaction; MPCR, multiplex polymerase chain reaction; MAF, minor allele frequency; HWE, Hardy-Weinberg equilibrium.

3.2 Quantitative Synthesis

11 case-control studies were included, which encompassed 1500 patients and 1769 controls. The results of the meta-analysis can be seen in Table 2. The forest plots evaluating the association between the *MTHFR* A1298C polymorphism and PE risk are shown in Fig. 2.

The *MTHFR* A1298C polymorphism was found to be significantly associated with the risk of PE in all models (allele contrast (A (alanine) *vs.* C (glutamate)): OR, 0.81; 95% CI, 0.71–0.93, $p = 0.207$; homozygote (AA *vs.* CC): OR, 0.57; 95% CI, 0.40–0.79, $p = 0.056$; heterozygote (AC *vs.* CC): OR, 0.62; 95% CI, 0.45–0.87, $p = 0.010$; dominant model (AA + AC *vs.* CC): OR, 0.59; 95% CI, 0.43–0.81, $p = 0.031$; recessive model (AA *vs.* AC + CC): OR, 0.83; 95% CI, 0.70–0.98), $p = 0.817$.

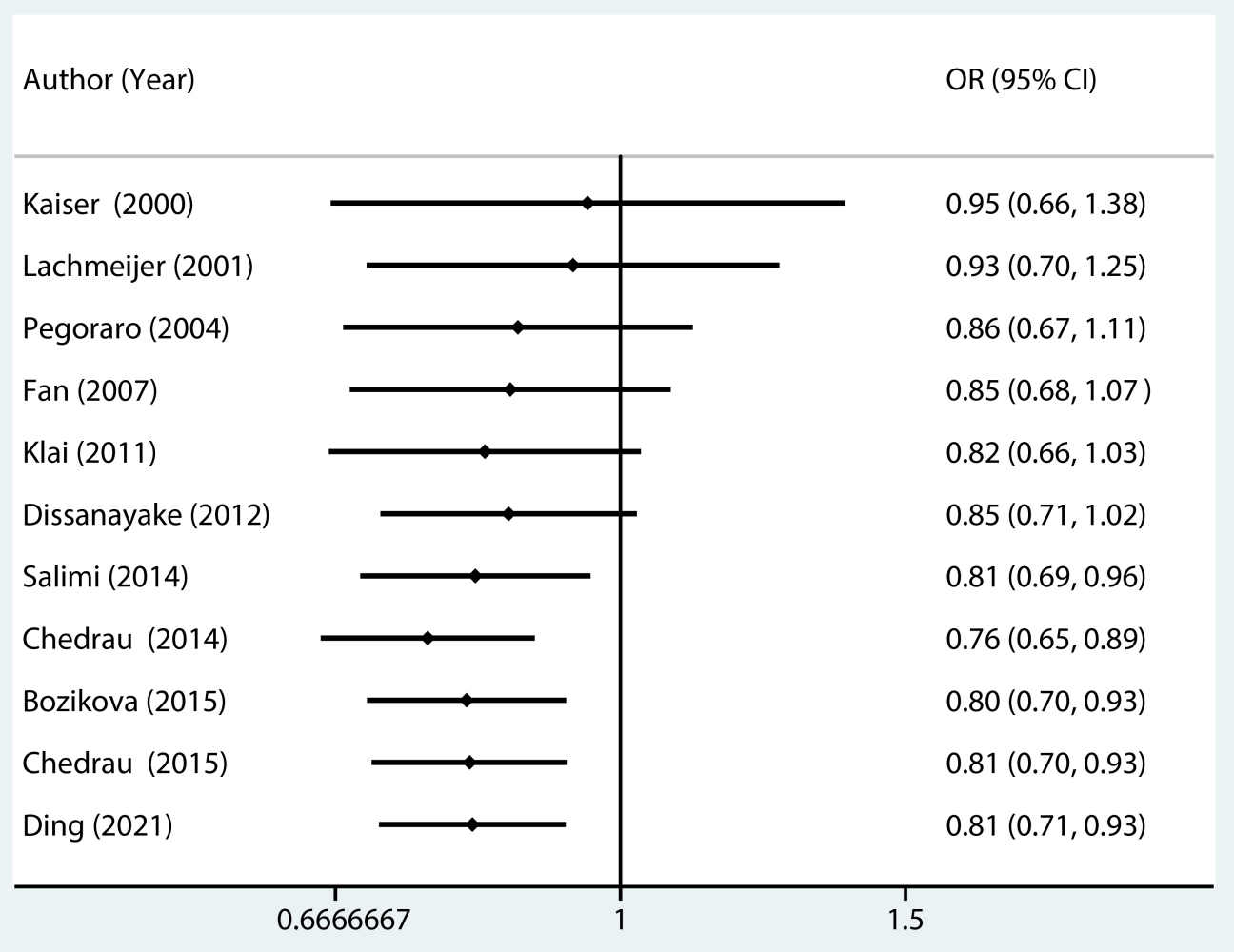

Fig. 5. Cumulative meta-analysis of associations between *MTHFR* A1298C polymorphism and PE risk.

The Newcastle-Ottawa Scale (NOS) scores for the selected studies are shown in Table 3 (Ref. [17,25–28,37–42]). The quality scores for the studies that were included ranged from 7 to 8 points.

3.3 Heterogeneity Analysis

Significant heterogeneity was observed in the heterozygote and dominant models (heterozygote model, $p_{\text{heterogeneity}} = 0.010$; dominant model, $p_{\text{heterogeneity}} = 0.031$). Galbraith plot analysis was used for probing heterogeneity sources in different researches. It was noted that a single study was the source of heterogeneity for the *MTHFR* A1298C polymorphism [27] (Fig. 3). After the removal of the outlier study, heterogeneity was found to markedly decrease (heterozygote model, $p_{\text{heterogeneity}} = 0.185$; dominant model, $p_{\text{heterogeneity}} = 0.340$).

3.4 Sensitivity Analysis and Cumulative Analysis

The sensitivity analyses (Fig. 4) and the cumulative meta-analysis (Fig. 5) demonstrated the robustness and stability of the results.

3.5 Publication Bias

Begg's and Egger's [32,33] tests were conducted as a means of assessing potential publication bias in the literature. As can be seen in Fig. 6, the Begg's funnel plot showed no signs of asymmetry [32]. Furthermore, the statistical results indicated there to be an absence of publication bias. Results from the Begg's and Egger's test were (allele contrast 0.35 and 0.36, homozygote 0.12 and 0.19, heterozygote 0.18 and 0.20, dominant model 0.17 and 0.16, recessive model 0.64 and 0.72).

4. Discussion

Following a meta-analysis of 11 case-control studies [17,25–28,37–42], a significant association was found between the *MTHFR* A1298C polymorphism and PE risk. In a subgroup analysis stratified by ethnicity, all the studies examined showed a significant decrease in PE risk. After the analysis was stratified by ethnicity, a notably decreased risk was identified among Asian populations for the homozygote, heterozygote, and dominant models. For mixed populations, there was also found to be a significant decrease

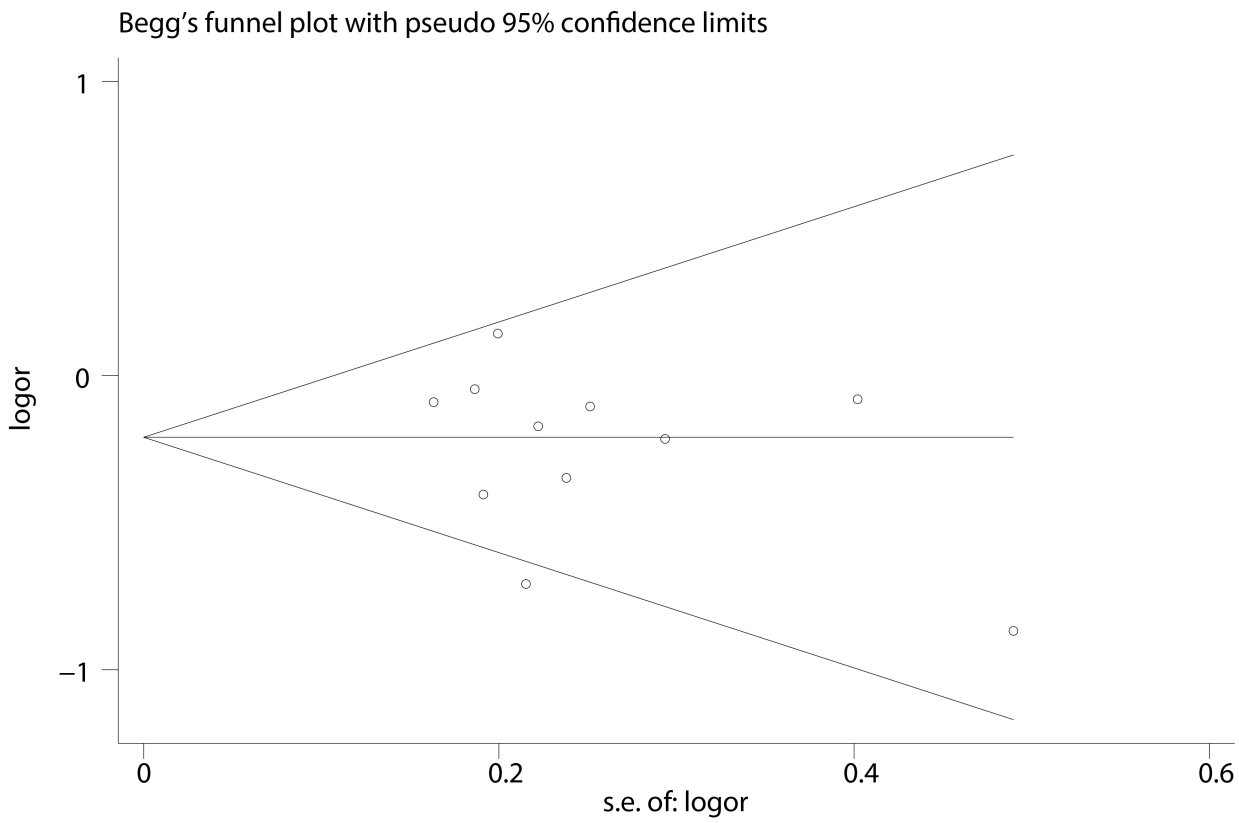

Fig. 6. Begg's funnel plot for publication bias test.

in risk in the allele contrast, homozygote, heterozygote, and dominant models. In contrast, no significant risk was detected in Caucasian populations, there reason for this being unclear. A potential reason may be the variance in the genotype distribution of *MTHFR* A1298C CC among Caucasian, Mixed, and Asian populations. The studies found that the prevalence of the *MTHFR* A1298C CC alleles in the 3′−untranslated region stood at 12.4% for Caucasian patients [17,26,28,37,38,40], while it was 7.4% for Asian patients [25,42], and 15.3% for those of Mixed ethnicity [27,41]. The variation in genotype distribution may play a role in the diverse associations that were observed between *MTHFR* A1298C polymorphism and PE risk in different ethnicities. Another potential factor is the restricted sample size of Asian and mixed patients in the meta-analysis. The limited representation of these Mixed patients may have diminished the statistical power, which potentially concealed the genuine associations.

In this meta-analysis, the studies included in the Asian population category are predominantly from China [25,42]. We acknowledge that this limited representation of Asian populations, encompassing only one country, could potentially restrict the generalizability of our findings. Therefore, it is imperative to exercise caution when interpreting our results and consider the possibility of regional variations when applying these findings to other Asian populations.

The findings of this study relating to the *MTHFR* A1298C polymorphism deviate from those of Wu *et al.* [34]. In their meta-analysis, they could not successfully demonstrate a connection between PE risk and the *MTHFR* A1298C polymorphism in any of their analyzed models. One potential reason for this inconsistency may be the limited sample size of the earlier study. The meta-analysis of Wu *et al.* [34] included just six studies that examined the *MTHFR* A1298C polymorphism and PE risk, resulting in a less refined risk assessment.

With meta-analysis, addressing heterogeneity is a major concern, as high variability among studies can potentially cause biased outcomes. In this research, heterogeneity was identified across the studies in the heterozygote and dominant models. The Galbraith plot analysis identified one study as being the primary contributor to this heterogeneity [27]. Following the removal of this outlier study, a marked reduction in heterogeneity was noticed, although the primary findings remained consistent.

Another major concern in meta-analysis is publication bias potential as a result of the selective representation of studies. In the meta-analysis of this study, Egger's and Begg's [32,33] tests were both used for evaluating this concern. The statistical outcomes, together with the symmetry

Table 2. Quantitative analyses of the *MTHFR* A1298C polymorphism on PE risk.

Genetic model			Allele contrast		Homozygote		Heterozygote		Dominant model		Recessive model	
Variables	Sample size		A *vs.* C		AA *vs.* CC		AC *vs.* CC		AA + AC *vs.* CC		AA *vs.* AC + CC	
	N[a]	Case/control	OR (95% CI)	*p*-value[b]	OR (95% CI)	*p*-value[b]	OR (95% CI)	*p*-value[b]	OR (95% CI)	*p*-value[b]	OR (95% CI)	*p*-value[b]
Total	11	1500/1769	0.81 (0.71, 0.93)	0.207	0.57 (0.40, 0.79)	0.056	0.62 (0.45, 0.87)	0.010	0.59 (0.43, 0.81)	0.031	0.83 (0.70, 0.98)	0.817
Caucasian	6	882/1019	0.88 (0.75, 1.04)	0.263	0.81 (0.54, 1.21)	0.382	0.91 (0.61, 1.36)	0.264	0.85 (0.58, 1.25)	0.331	0.85 (0.68, 1.06)	0.521
Asia	2	214/212	0.83 (0.59, 1.17)	0.907	0.31 (0.11, 0.87)	0.752	0.27 (0.09, 0.77)	0.566	0.30 (0.11, 0.83)	0.696	0.98 (0.65, 1.47)	0.700
Mixed	2	200/200	0.57 (0.39, 0.82)	0.168	0.15 (0.05, 0.45)	0.007	0.16 (0.05, 0.47)	0.003	0.12 (0.05, 0.43)	0.005	0.74 (0.48, 1.15)	0.853
African	1	204/338	0.71 (0.44, 1.13)	NA[c]	1.14 (0.10, 12.6)	NA[c]	0.77 (0.66, 0.90)	NA[c]	1.21 (0.11, 13.4)	NA[c]	0.67 (0.41, 1.10)	NA[c]

[a] Number of comparisons.

[b] *p*-value of Q-test for heterogeneity test; Random effects model was used when the *p*-value for heterogeneity test <0.05; otherwise, fixed effects model was used.

[c] NA – not available.

OR, odds ratios; CI, confidence intervals; A, alanine; C, glutamate.

Table 3. Quality assessment of case–control studies that were included in the meta-analysis[a].

Study	Adequate definition of cases	Representativeness of cases	Selection of control	Definition of control	Control for important factor or additional factor[b]	Exposure assessment	Same method of ascertainment for cases and controls	Nonresponse rate[c]	Total quality scores[d]
Kaiser *et al.* [37], 2000	★	★	★	★	★	-	★	★	7
Lachmeijer *et al.* [38], 2001	★	★	★	★	★★	-	★	★	8
Pegoraro *et al.* [39], 2004	★	★	★	★	★	-	★	★	7
Fan *et al.* [25], 2007	★	★	★	★	★★	-	★	★	8
Klai *et al.* [17], 2011	★	★	★	★	★★	-	★	★	8
Dissanayake *et al.* [26], 2012	★	★	★	★	★★	-	★	★	8
Salimi *et al.* [28], 2015	★	★	★	★	★★	-	★	★	8
Chedraui *et al.* [27], 2014	★	★	★	★	★★	-	★	★	8
Bozikova *et al.* [40], 2015	★	★	★	★	★★	-	★	★	8
Chedrau *et al.* [41], 2015	★	★	★	★	★	-	★	★	7
Ding *et al.* [42], 2021	★	★	★	★	★★	-	★	★	8

[a] A study was awarded a maximum of one star for each numbered item, except for the item ‘Control’.

[b] A maximum of two stars were awarded for ‘Control’ – one for most important factor or two for second most important factor; studies that controlled for maternal age received one star, while studies that controlled for high risk factor (diabetes or pre-pregnancy body mass index or family history of hypertension) received one additional star.

[c] One star was awarded if no significant difference was observed in the response rate between control subjects and cases in the chi-square test ($p > 0.05$).

[d] A study was considered low-quality if there were fewer than six stars in the quality assessment.

of the funnel plot, indicated an absence of notable publication bias. More importantly, the results were further bolstered by sensitivity analysis, which affirmed their consistency and robustness. It should be noted that one of the studies that was considered deviated from the Hardy-Weinberg equilibrium (HWE) [41]. However, following the removal of this anomalous study and a data reassessment, the conclusions remained consistent.

There are several limitations with this study: (i) due to the limited sample size and the small number of researches that were included in the meta-analysis, the results may not adequately reflect the true associations; (ii) the analysis was reliant on unadjusted OR estimates as not all the included studies provided adjusted ORs, and where adjusted ORs were provided, the adjustments varied based on factors such as ethnicity, age, or smoking habits; (iii) the genotype distribution of the control group in one study deviated from the Hardy-Weinberg equilibrium (HWE); and (iv) in this meta-analysis, the representation of the Asian population came from two studies that were conducted in China and due to the lack of research data from other Asian populations, the findings may have limited generalizability; (v) compared to our study, the latest Whole Genome Sequencing (WGS) research has utilized a much larger study population, including participants from diverse geographical regions and ethnic backgrounds. These studies possess higher statistical power and encompass broader genetic diversity, thus enabling them to provide more accurate and comprehensive analysis results. Therefore, we anticipate these large-scale studies will offer deeper insights into the association between specific genetic variations and preeclampsia.

5. Conclusion

In conclusion, our meta-analysis suggests that the *MTHFR* A1298C variant holds potential as a genetic marker for PE. Nonetheless, additional comprehensive, multicenter studies are essential to solidify and validate our findings.

Availability of Data and Materials

All data generated or analysed during this study are included in this article. Further enquiries can be directed to the corresponding author.

Author Contributions

KY conceived and designed the meta-analysis. YH and AW performed the literature search. AW analyzed the data. YH wrote the paper. All authors contributed to editorial changes in the manuscript. All authors read and approved the final manuscript. All authors have participated sufficiently in the work and agreed to be accountable for all aspects of the work.

Ethics Approval and Consent to Participate

Not applicable.

Acknowledgment

Thanks to all the peer reviewers for their opinions and suggestions.

Funding

This study was supported by the Key Research and Development Program of Sichuan Province (grant no. 2022YFS0079).

Conflict of Interest

The authors declare no conflict of interest.

Supplementary Material

Supplementary material associated with this article can be found, in the online version, at https://doi.org/10.31083/j.ceog5012266.

References

[1] Fishel Bartal M, Sibai BM. Eclampsia in the 21st century. American Journal of Obstetrics and Gynecology. 2022; 226: S1237–S1253.

[2] Dimitriadis E, Rolnik DL, Zhou W, Estrada-Gutierrez G, Koga K, Francisco RPV, *et al*. Pre-eclampsia. Nature Reviews. Disease Primers. 2023; 9: 8.

[3] GBD 2015 Maternal Mortality Collaborators. Global, regional, and national levels of maternal mortality, 1990-2015: a systematic analysis for the Global Burden of Disease Study 2015. Lancet (London, England). 2016; 388: 1775–1812.

[4] Magee LA, Nicolaides KH, von Dadelszen P. Preeclampsia. The New England Journal of Medicine. 2022; 386: 1817–1832.

[5] Wu P, Green M, Myers JE. Hypertensive disorders of pregnancy. BMJ (Clinical Research Ed.). 2023; 381: e071653.

[6] Steegers EAP, von Dadelszen P, Duvekot JJ, Pijnenborg R. Pre-eclampsia. Lancet (London, England). 2010; 376: 631–644.

[7] Kanayama N. Trophoblastic injury: new etiological and pathological concept of preeclampsia. Croatian Medical Journal. 2003; 44: 148–156.

[8] Chappell S, Morgan L. Searching for genetic clues to the causes of pre-eclampsia. Clinical Science (London, England: 1979). 2006; 110: 443–458.

[9] Haram K, Mortensen JH, Nagy B. Genetic aspects of preeclampsia and the HELLP syndrome. Journal of Pregnancy. 2014; 2014: 910751.

[10] Jung E, Romero R, Yeo L, Gomez-Lopez N, Chaemsaithong P, Jaovisidha A, *et al*. The etiology of preeclampsia. American Journal of Obstetrics and Gynecology. 2022; 226: S844–S866.

[11] Eskandari F, Teimoori B, Rezaei M, Mohammadpour-Gharehbagh A, Narooei-Nejad M, Mehrabani M, *et al*. Relationships between Dicer 1 polymorphism and expression levels in the etiopathogenesis of preeclampsia. Journal of Cellular Biochemistry. 2018; 119: 5563–5570.

[12] Wang T, Lian Y. The relationship between Fas and Fas ligand gene polymorphism and preeclampsia risk. Bioscience Reports. 2019; 39: BSR20181901.

[13] Pan X, Wei B, Wang H, Ma L, Du Z, Chen Y. Novel association between FOXO3 rs2232365 polymorphism and late-onset preeclampsia: a case-control candidate genetic study. BMC Pregnancy and Childbirth. 2020; 20: 779.

[14] Zheng Y, Ma C, Liu X, Wu S, Zhang W, Zhao S. Association between HLA-A gene polymorphism and early-onset preeclampsia

in Chinese pregnant women early-onset. BMC Pregnancy and Childbirth. 2020; 20: 656.
[15] Jacques PF, Bostom AG, Williams RR, Ellison RC, Eckfeldt JH, Rosenberg IH, *et al*. Relation between folate status, a common mutation in methylenetetrahydrofolate reductase, and plasma homocysteine concentrations. Circulation. 1996; 93: 7–9.
[16] Barbosa PR, Stabler SP, Machado ALK, Braga RC, Hirata RDC, Hirata MH, *et al*. Association between decreased vitamin levels and MTHFR, MTR and MTRR gene polymorphisms as determinants for elevated total homocysteine concentrations in pregnant women. European Journal of Clinical Nutrition. 2008; 62: 1010–1021.
[17] Klai S, Fekih-Mrissa N, El Housaini S, Kaabechi N, Nsiri B, Rachdi R, *et al*. Association of MTHFR A1298C polymorphism (but not of MTHFR C677T) with elevated homocysteine levels and placental vasculopathies. Blood Coagulation & Fibrinolysis: an International Journal in Haemostasis and Thrombosis. 2011; 22: 374–378.
[18] Goyette P, Sumner JS, Milos R, Duncan AM, Rosenblatt DS, Matthews RG, *et al*. Human methylenetetrahydrofolate reductase: isolation of cDNA, mapping and mutation identification. Nature Genetics. 1994; 7: 195–200.
[19] Frosst P, Blom HJ, Milos R, Goyette P, Sheppard CA, Matthews RG, *et al*. A candidate genetic risk factor for vascular disease: a common mutation in methylenetetrahydrofolate reductase. Nature Genetics. 1995; 10: 111–113.
[20] van der Put NM, Gabreëls F, Stevens EM, Smeitink JA, Trijbels FJ, Eskes TK, *et al*. A second common mutation in the methylenetetrahydrofolate reductase gene: an additional risk factor for neural-tube defects? American Journal of Human Genetics. 1998; 62: 1044–1051.
[21] Mou AD, Barman Z, Hasan M, Miah R, Hafsa JM, Das Trisha A, *et al*. Prevalence of preeclampsia and the associated risk factors among pregnant women in Bangladesh. Scientific Reports. 2021; 11: 21339.
[22] Chen M, Xia B, Rodriguez-Gueant RM, Bigard M, Gueant JL. Genotypes 677TT and 677CT+1298AC of methylenetetrahydrofolate reductase are associated with the severity of ulcerative colitis in central China. Gut. 2005; 54: 733–734.
[23] Yuan X, Wang T, Gao J, Wang Y, Chen Y, Kaliannan K, *et al*. Associations of homocysteine status and homocysteine metabolism enzyme polymorphisms with hypertension and dyslipidemia in a Chinese hypertensive population. Clinical and Experimental Hypertension (New York, N.Y.: 1993). 2020; 42: 52–60.
[24] Chango A, Boisson F, Barbé F, Quilliot D, Droesch S, Pfister M, *et al*. The effect of 677C–>T and 1298A–>C mutations on plasma homocysteine and 5,10-methylenetetrahydrofolate reductase activity in healthy subjects. The British Journal of Nutrition. 2000; 83: 593–596.
[25] Fan LP, Fu F. Study on the correlation between hypertensive disorder complicating pregnancy and the MTHFR gene polymorphism Nanchang. Nanchang University. 2007; 1–27. (In Chinese)
[26] Dissanayake VHW, Sirisena ND, Weerasekera LY, Gammulla CG, Seneviratne HR, Jayasekara RW. Candidate gene study of genetic thrombophilic polymorphisms in pre-eclampsia and recurrent pregnancy loss in Sinhalese women. The Journal of Obstetrics and Gynaecology Research. 2012; 38: 1168–1176.
[27] Chedraui P, Salazar-Pousada D, Villao A, Escobar GS, Ramirez C, Hidalgo L, *et al*. Polymorphisms of the methylenetetrahydrofolate reductase gene (C677T and A1298C) in nulliparous women complicated with preeclampsia. Gynecological Endocrinology: the Official Journal of the International Society of Gynecological Endocrinology. 2014; 30: 392–396.
[28] Salimi S, Saravani M, Yaghmaei M, Fazlali Z, Mokhtari M, Naghavi A, *et al*. The early-onset preeclampsia is associated with MTHFR and FVL polymorphisms. Archives of Gynecology and Obstetrics. 2015; 291: 1303–1312.
[29] Higgins JPT, Thompson SG, Deeks JJ, Altman DG. Measuring inconsistency in meta-analyses. BMJ (Clinical Research Ed.). 2003; 327: 557–560.
[30] MANTEL N, HAENSZEL W. Statistical aspects of the analysis of data from retrospective studies of disease. Journal of the National Cancer Institute. 1959; 22: 719–748.
[31] DerSimonian R, Laird N. Meta-analysis in clinical trials. Controlled Clinical Trials. 1986; 7: 177–188.
[32] Begg CB, Mazumdar M. Operating characteristics of a rank correlation test for publication bias. Biometrics. 1994; 50: 1088–1101.
[33] Egger M, Davey Smith G, Schneider M, Minder C. Bias in meta-analysis detected by a simple, graphical test. BMJ (Clinical Research Ed.). 1997; 315: 629–634.
[34] Wu X, Yang K, Tang X, Sa Y, Zhou R, Liu J, *et al*. Folate metabolism gene polymorphisms MTHFR C677T and A1298C and risk for preeclampsia: a meta-analysis. Journal of Assisted Reproduction and Genetics. 2015; 32: 797–805.
[35] Mishra J, Talwar S, Kaur L, Chandiok K, Yadav S, Puri M, *et al*. Differential global and MTHFR gene specific methylation patterns in preeclampsia and recurrent miscarriages: A case-control study from North India. Gene. 2019; 704: 68–73.
[36] Ding G, Li Y, Gao J, Wang W, Wang H, Bai G. Associations between AGT, MTHFR, and VEGF gene polymorphisms and preeclampsia in the Chinese population. Placenta. 2022; 118: 38–45.
[37] Kaiser T, Brennecke SP, Moses EK. Methylenetetrahydrofolate reductase polymorphisms are not a risk factor for preeclampsia/eclampsia in Australian women. Gynecologic and Obstetric Investigation. 2000; 50: 100–102.
[38] Lachmeijer AM, Arngrímsson R, Bastiaans EJ, Pals G, ten Kate LP, de Vries JI, *et al*. Mutations in the gene for methylenetetrahydrofolate reductase, homocysteine levels, and vitamin status in women with a history of preeclampsia. American Journal of Obstetrics and Gynecology. 2001; 184: 394–402.
[39] Pegoraro RJ, Chikosi A, Rom L, Roberts C, Moodley J. Methylenetetrahydrofolate reductase gene polymorphisms in black South Africans and the association with preeclampsia. Acta Obstetricia et Gynecologica Scandinavica. 2004; 83: 449–454.
[40] Bozikova A, Gabrikova D, Pitonak J, Bernasovska J, Macekova S, Lohajova-Behulova R. Ethnic differences in the association of thrombophilic polymorphisms with obstetric complications in Slovak and Roma (Gypsy) populations. Genetic Testing and Molecular Biomarkers. 2015; 19: 98–102.
[41] Chedraui P, Andrade ME, Salazar-Pousada D, Escobar GS, Hidalgo L, Ramirez C, *et al*. Polymorphisms of the methylenetetrahydrofolate reductase gene (C677T and A1298C) in the placenta of pregnancies complicated with preeclampsia. Gynecological Endocrinology: the Official Journal of the International Society of Gynecological Endocrinology. 2015; 31: 569–572.
[42] Ding ZY, Chen YY, Deng QX, Dong J. Correlation between MTHFR gene polymorphism and preeclampsia. Maternal and Child Health Care of China. 2021; 36: 3828–3831. (In Chinese)
[43] Stroup DF, Berlin JA, Morton SC, Olkin I, Williamson GD, Rennie D, *et al*. Meta-analysis of observational studies in epidemiology: a proposal for reporting. Meta-analysis Of Observational Studies in Epidemiology (MOOSE) group. JAMA. 2000; 283: 2008–2012.