Systematic Review and Meta-Analysis

Medicine®

OPEN

Association of β-fibrinogen polymorphisms and venous thromboembolism risk

A PRISMA-compliant meta-analysis

Da Li, MM, Xiaosong Zhang, MM, He Huang, MM, Honggang Zhang, MD*

Abstract

Background: Venous thromboembolism (VTE) is a multifactorial disease in which genetic and acquired risk factors may contribute to disease pathogenesis. Several studies have demonstrated that β-fibrinogen (FGB) polymorphisms are associated with the risk of VTE. However, the results of these studies were not totally consistent. In this paper, we performed a meta-analysis to further investigate the relationship between FGB polymorphisms and susceptibility to VTE.

Methods: To identify studies pertinent to the focused question, the following databases were systematically searched: PubMed, EMBASE, Web of Science, China National Knowledge Infrastructure, and Wanfang Data. The strength of correlations was evaluated by calculating pooled odds ratios (ORs) and 95% confidence intervals (95% CIs). Subgroup analyses stratified by ethnicity, type of disorders, and source of control were also performed.

Results: Overall, A total of 18 relevant case-control studies met the inclusion criteria and were incorporated in this meta-analysis, involving 3033 VTE cases and 4547 healthy controls. FGB -455G>A polymorphism and -148C>T polymorphism were not significantly associated with susceptibility to VTE in overall populations. However, results of stratified analysis demonstrated that among Caucasian population, the -455G>A mutation was negatively associated with the risk of VTE under all genetic comparison models (A:G OR = 0.80 95% CI = 0.70–0.91; GA + AA:GG OR = 0.80 95% CI = 0.68–0.93; GA:GG OR = 0.84 95% CI = 0.71–0.98; AA:GG + GA OR = 0.61 95% CI = 0.43–0.87; AA:GG OR = 0.57 95% CI = 0.40–0.82), which indicates FGB -455G>A polymorphism may be a protective factor for VTE. There was no correlation between -148C>T polymorphism and susceptibility to VTE in all subgroup analyses.

Conclusion: FGB -455G>A polymorphism was associated with a decreased risk of VTE among the Caucasian population.

Abbreviations: CI = confidence interval, DVT = deep venous thrombosis, FGB = β-fibrinogen, HWE = Hardy–Weinberg equilibrium, OR = odds ratio, PE = pulmonary embolism, SNP = single nucleotide polymorphism, VTE = venous thromboembolism.

Keywords: β-fibrinogen (FGB), gene polymorphisms, meta-analysis, venous thromboembolism

1. Introduction

Venous thromboembolism (VTE), which consists of deep venous thrombosis (DVT) and pulmonary embolism (PE), is described as the functional venous outflow obstruction attributed to previous deep vein clot and the pathophysiological changes and clinical manifestations of the pulmonary artery occlusion followed by thrombus detachment and migration. VTE is a fairly common disease with high morbidity and mortality.[1] Due to the high incidence of complications, the intense tendency to relapse and the significantly lower survival rate, VTE imposed a heavy burden both on patients and society.[2] The pathogenesis of VTE is complex and multifactorial, including genetic or acquired predisposition to thrombosis as well as a variety of risk factors. Over the last decades, a number of genetic mutations have been reported to be associated with the susceptibility to VTE.[3–5]

Fibrinogen (Fg; also known as coagulation factor I) serves as a key protein in the coagulation process. It is converted to fibrin monomer by thrombin in the final stage of the coagulation cascade, and finally forms an insoluble fibrin clot to exert blood coagulation. Fibrinogen is comprised of 2 symmetric half molecules, each consisting of 3 polypeptide chains termed Aα, Bβ, and γ, which are joined together by 5 symmetrical disulfide bridges.[6,7] Many prospective studies and case-control studies have reported that plasma fibrinogen concentration is an independent risk factor for cardiovascular and thrombotic diseases.[8,9] Fibrinogen gene family is comprised of 3 genes coding fibrinogen γ, α, and β, clustered in a region of approximately 50 kb on the long arm of chromosome 4. Transcription of β-fibrinogen gene (*FGB*) is the rate-limiting step in fibrinogen production. Therefore, the *FGB* gene is thought to obviously affect plasma fibrinogen levels, and has been

Editor: Nikhil Jain.

The authors have no funding and conflicts of interest to disclose.

Department of Vascular Surgery, The First People's Hospital of Lianyungang, Lianyungang, Jiangsu Province, China.

** Correspondence: Honggang Zhang, Department of Vascular Surgery, The First People's Hospital of Lianyungang, NO.182 North Tongguan Road, Lianyungang, Jiangsu Province 222002, China (e-mail: 540660190@qq.com).*

How to cite this article: Li D, Zhang X, Huang H, Zhang H. Association of β-fibrinogen polymorphisms and venous thromboembolism risk: a PRISMA-compliant meta-analysis. Medicine 2019;98:48(e18204).

Received: 14 May 2019 / Received in final form: 8 October 2019 / Accepted: 31 October 2019

http://dx.doi.org/10.1097/MD.0000000000018204

confirmed by following researches.[10] A large number of studies have reported the correlation between FGB gene -455G>A and -148C>T polymorphisms with increased risk of arterial thrombotic disease.[11–15] Several meta-analysis studies have also demonstrated these 2 polymorphisms of FGB gene are associated with increased risk of ischemic stroke and coronary heart disease.[16,17] Koster first conducted a case-control study aimed to elucidate the association between FGB -455G>A polymorphism and VTE susceptibility, and demonstrated a significantly decreased risk of VTE in carriers of the A-allele heterozygote variant.[18] However, several subsequent studies reported that the FGB polymorphisms did not contribute to VTE susceptibility or decrease the risk of VTE. [19–21] Given the inconsistent and even contradictory findings of previous studies, we conducted a comprehensive meta-analysis to further investigate the association between *FGB* gene polymorphisms and susceptibility to VTE.

2. Materials and methods

2.1. Literature search

The databases of PubMed, EMBASE, Web of Science, Chinese National Knowledge Infrastructure, and Wan Fang were searched in a comprehensive and systematic approach to collect eligible case-control studies investigating the association between *FGB* gene polymorphisms and VTE risk. The retrieval time was up to March 2019. To avoid omission of potential relevant literatures, references of the incorporated literatures were also traced and manually searched. The search terms were as follows: ("FGB" OR "beta-fibrinogen" OR "fibrinogen beta" OR "β-fibrinogen" OR "fibrinogen β" OR "HaeIII") and ("VTE" OR "Venous thromboembolism" OR "Venous Thrombosis" OR "pulmonary embolism") and ("mutation" OR "variant" OR "polymorphism" OR "genotype" OR "allele" OR "gene" OR "snp").

2.2. Inclusion and exclusion criteria

Strict inclusion and exclusion criteria were set and performed in this current meta-analysis. The inclusion criteria of the search included the following: case-control study evaluating the relationship between *FGB* gene polymorphisms and risk of VTE; the enrolled patients in the case group were confirmed by the diagnostic criteria of VTE, PE, or DVT. The exclusion criteria were mainly based on the following: articles published neither in English nor in Chinese; repeated publications or studies published by the identical author, only retained and incorporated the study with the largest sample size and the most comprehensive data; studies with insufficient information about genotype distribution; reviews, animal experiments, case reports or conference abstracts.

2.3. Data extraction

Two reviewers (DL and XZ) independently screened the potential eligible literature and extracted data. First, the title and abstract of citations obtained through the search strategy were screened for eligibility. Next, the full text of all potentially eligible trials was retrieved and then further evaluated to exclude studies not relevant to the topic. In case of disagreement, a third reviewer (HZ, MD) was consulted to decide. To obtain missing data, corresponding authors of the included studies were contacted by email. The data extraction content includes: first author, publication year, nationality, ethnicity, thrombotic disorder category, source of control, genotyping method, and genotype distribution.

2.4. Quality assessment

Newcastle–Ottawa scale is a extensively adopted quality-assessment tool in observational and nonrandomized trials and was applied in this current meta-analysis to evaluate the quality of included studies. The scoring of this scale tool contains 3 aspects: the choice of the research group; comparability between groups; result determination. The "star system" was conducted in the scoring system with a maximum score of 9 points. Higher scores indicate the better quality of design and methodology of the study.

2.5. Statistical analysis

Meta-analysis was performed with Revman5.3 software from the Cochrane Collaboration. The strength of association between FGB polymorphisms and VTE was evaluated by calculating odds ratios (ORs) and its 95% confidence interval (CI) under 5 gene comparison models, for example, the correlation between -455G>A polymorphism and VTE was measured and assessed under the following 5 gene comparison models: allele model (A vs G), homozygous model (AA vs. GG), heterozygous model (GA vs AA), dominant model (GA + AA vs. GG), and recessive model (AA vs GA + GG). The heterogeneity was quantitatively determined by I^2. If there was no statistical heterogeneity among the results (I^2 statistic $<50\%$), the fixed-effect model was performed for meta-analysis. If statistical heterogeneity existed (I^2 statistic $>50\%$), the source of heterogeneity would be further analyzed, and the random effect model will be adopted. In addition to overall analysis, subgroup analyses stratified by ethnicity, type of thrombotic disorders, and source of control were also performed.

The possible publication bias was first evaluated by observing the symmetry of the funnel plots. The Stata14.0 software (STATA Corporation, College Station, TX) was further used to perform the Egger test operational procedure which was adopted to assess the existence of potential publication bias. The genotype distribution of the control group was calculated by χ^2 test to estimate whether the control group was consistent with Hardy–Weinberg equilibrium (HWE). Sensitivity analyses were also carried out to examine the stability of synthetic results.

Trial sequential analysis (TSA) was conducted by TSA v0.9.5.10 Beta software developed by Copenhagen clinical trial center. The odds ratio reduction (OR reduction) was set as 20%. Type 1 error $=0.05$ and type 2 error $=0.2$ were adopted to calculate the required information size (RIS). When the cumulative Z value crosses the TSA threshold, the result was considered statistically significant. At the same time, the sample size of cumulative evidence was judged to be sufficient based on RIS.

2.6. Ethical approval

The current meta-analysis was performed on the base of previous studies. Thus, the ethical approval was not required.

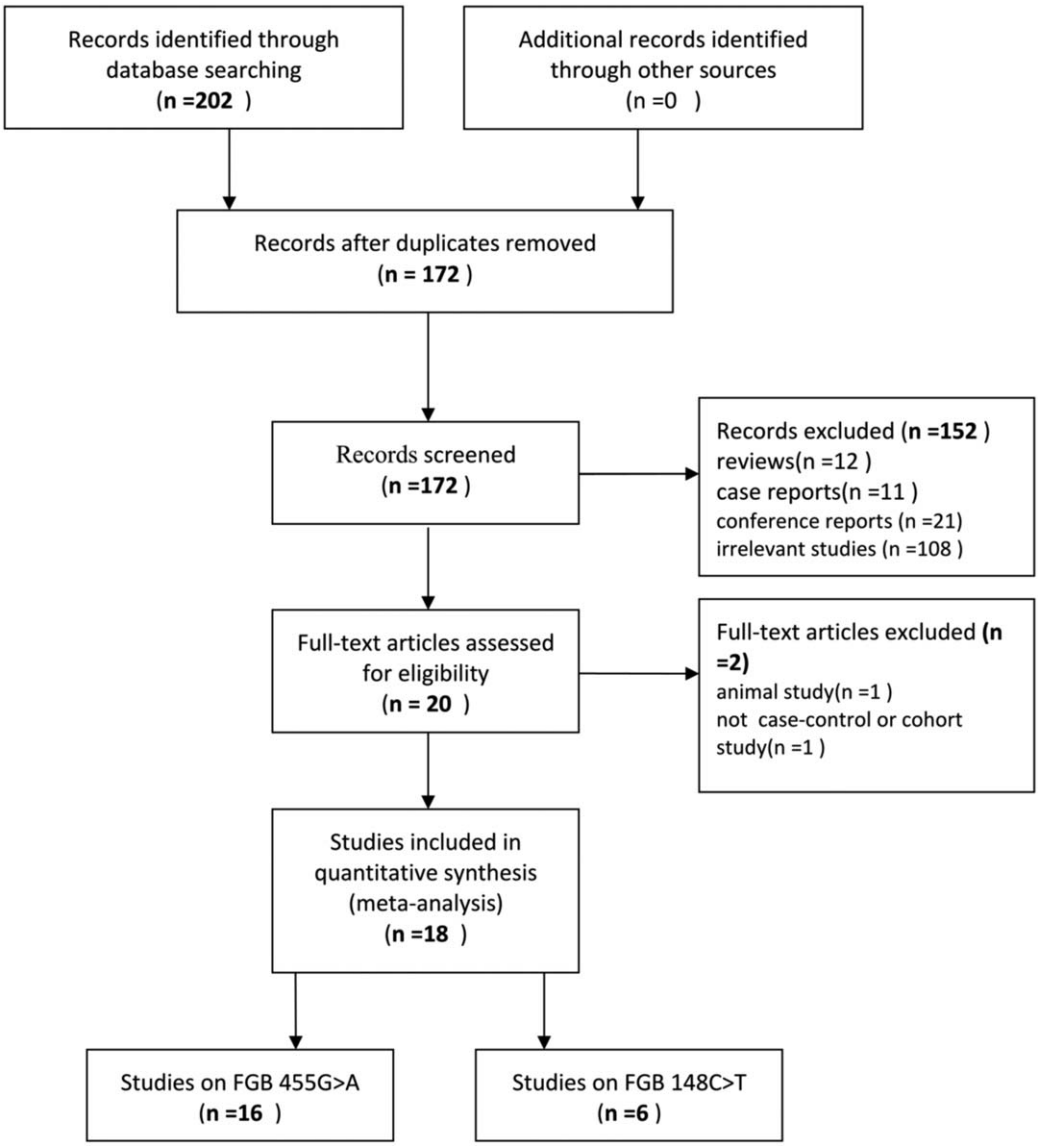


Figure 1. Flow diagram of selection and inclusion process.

3. Results

3.1. Study characteristics

A total of 202 articles were identified through systematic search. After removing 30 duplicated articles, 152 articles were further excluded by browsing the title and abstract. Through particularly screening the left remaining 20 articles in full text, a total of 18 studies accorded with the inclusion criteria were ultimately enrolled in this current meta-analysis,[18–35] including 3033 VTE cases and 4547 healthy controls overall. Among the incorporated literatures, 16 investigated -455G>A polymorphism and 6 inspected -148C>T polymorphism. Among the studies on -455G>A polymorphism, 6 were based on the Caucasian population and 9 were based on the Asian population. It should be mentioned that 2 of these studies, Camilleri and Cushman's studies were mixed population consisting, the former provided specific genotype distribution of each ethnicity, and Cushman's study provided the genotype distribution of the Caucasian population. Therefore, genotype distribution of independent ethnicity in the 2 literatures was included in the ethno-based stratified subgroup analysis. In the literature on -148C>T polymorphism, there were 3 studies based on the Asian population and 2 based on the Latino population. Among all included eligible studies, 3 studies were not consistent with HWE. Figure 1 shows the flowchart of selection and inclusion process. Table 1 summaries the general characteristics of all included literatures. Table 2 shows the detailed -455G>A polymorphism and -148C>T polymorphism genotype distributions and the *P*-value of HWE in each control group.

3.2. Quantitative synthesis

3.2.1. Meta-analysis of association between FGB polymorphisms and VTE susceptibility. Table 2 lists the main results of the association between the -455G>A,-148C>T polymorphisms and VTE risk. Figure 2 displays the forest plot of the association

Table 1

Characteristics of included studies.

Author	Year	Region	Ethnicity	Type of disease	Control source	Genotyping method	Sample size		NOS
							Case	Control	
Austin	2000	USA	African-Americans	VTE	Hospital-based	PCR	91	185	7
Bezgin	2018	Turkey	Asian	VTE	Population-based	PCR	310	289	7
Blake	2001	USA	Caucasian	VTE	Population-based	PCR	156	751	7
Bozic	2004	Slovenia	Caucasian	DVT	Population-based	RT-PCR	114	244	8
Camilleri	2005	UK	Mixed	VTE	Population-based	PCR	339	190	8
Chen	2013	China	Asian	DVT	Hospital-based	PCR	132	155	7
Cushman	2007	USA	Mixed	VTE	Population-based	PCR	511	1028	8
Guzman	2010	Chile	Latino	DVT	Population-based	PCR-RFLP	60	112	8
Han	2012	China	Asain	DVT	Hospital-based	PCR-RFLP	120	120	8
Harrington	2003	Russia	Caucasian	PE	Population-based	PCR	58	60	6
Hidalgo	2010	Costa Rica	Latino	VTE	Population-based	PCR	57	178	7
Koster	1994	Netherland	Caucasian	DVT	Population-based	PCR-RFLP	199	199	8
Kumari	2014	India	Asain	VTE	Population-based	PCR-RFLP	93	102	7
Lin	2009	China	Asain	DVT	Hospital-based	PCR-RFLP	153	262	7
Qin	2017	China	Asain	DVT	Population-based	PCR-RFLP	100	100	6
Renner	2002	Austria	Caucasian	DVT	Hospital-based	AS-PCR	307	316	7
Zhai	2011	China	Asain	PE	Population-based	PCR-RFLP	101	101	7
Zhang	2012	China	Asain	DVT	Population-based	PCR	132	155	7

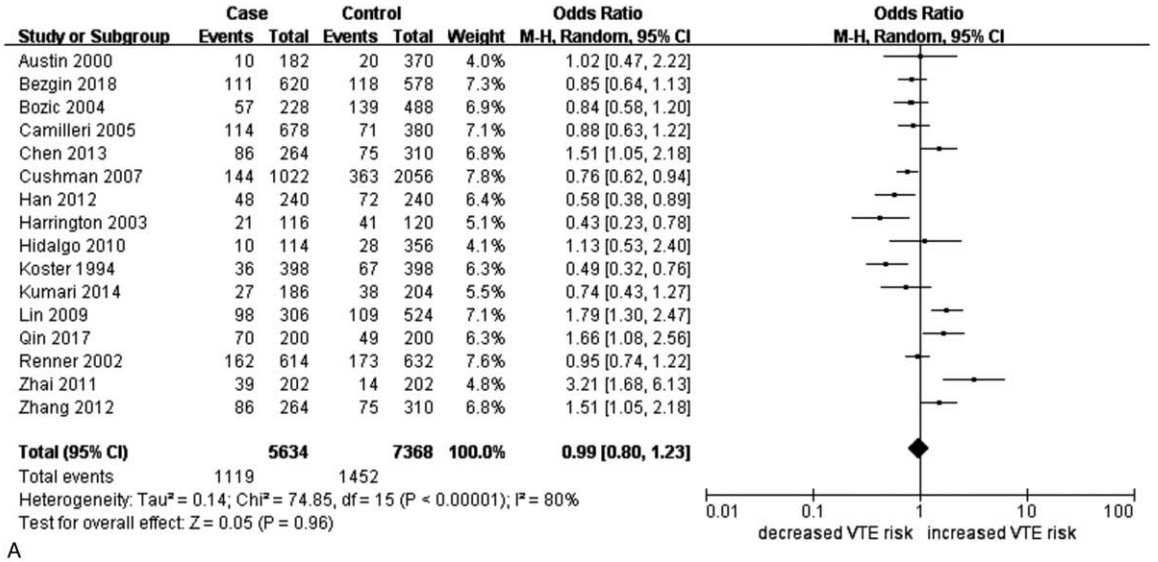
AS-PCR = allele specific polymerase chain reaction, DVT = deep venous thrombosis, NOS = Newcastle–Ottawa scale, PCR = polymerase chain reaction, PE = pulmonary embolism, RFLP = restriction fragment length polymorphism, RT-PCR = real time polymerase chain reaction, VTE = venous thromboembolism.

Table 2

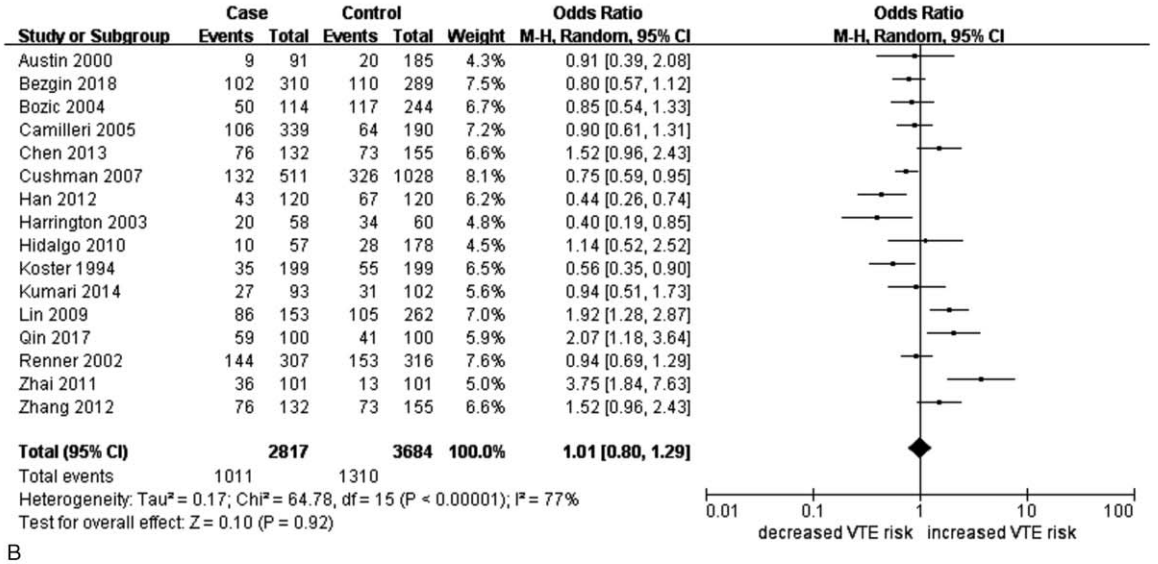
Genotype distribution and HWE.

SNP	Author	Ethnicity	Case genotype			Control genotype			HWE
			GG	GA	AA	GG	GA	AA	(*P*)
455G>A (rs1800790)	Austin	African-American	82	8	1	165	20	0	.437
	Bezgin	Asian	208	93	9	179	102	8	.143
	Bozic	Caucasian	64	43	7	127	95	22	.489
	Camilleri	Total	233	98	8	126	57	7	.861
		Caucasian	175	81	8	118	55	7	.852
		Asian	8	4	0	2	1	0	.729
		Afro-Caribbean	30	3	0	6	1	0	.839
	Chen	Asian	56	66	10	82	71	2	.002
	Cushman	Total	375	120	12	701	289	37	.291
		Caucasian	249	109	12	463	256	35	.959
		Non-Caucasian	126	11	0	238	33	2	.475
	Han	Asian	77	38	5	53	62	5	.017
	Harrington	Caucasian	38	19	1	26	27	7	.998
	Hidalgo	Latino	47	10	0	150	28	0	.255
	Koster	Caucasian	164	34	1	144	43	12	.001
	Kumari	Asian	62	27	0	71	24	7	.024
	Lin	Asian	67	74	12	157	101	4	.006
	Qin	Asian	41	48	11	59	33	8	.280
	Renner	Caucasian	163	126	18	163	133	20	.298
	Zhai	Asian	65	33	3	88	12	1	.427
	Zhang	Asian	56	66	10	82	71	2	.002
SNP	Author	Ethnicity	Case genotype			Control genotype			HWE
			CC	CT	TT	CC	CT	TT	(*P*)
148C>T (rs1800787)	Blake	Caucasian	108	43	5	468	243	40	.255
	Guzman	Latino	50	10	0	94	18	0	.355
	Han	Asian	85	32	3	109	10	1	.179
	Hidalgo	Latino	37	20	0	156	22	0	.380
	Kumari	Asian	53	38	2	52	45	5	.225
	Zhai	Asian	62	29	10	66	25	10	.004

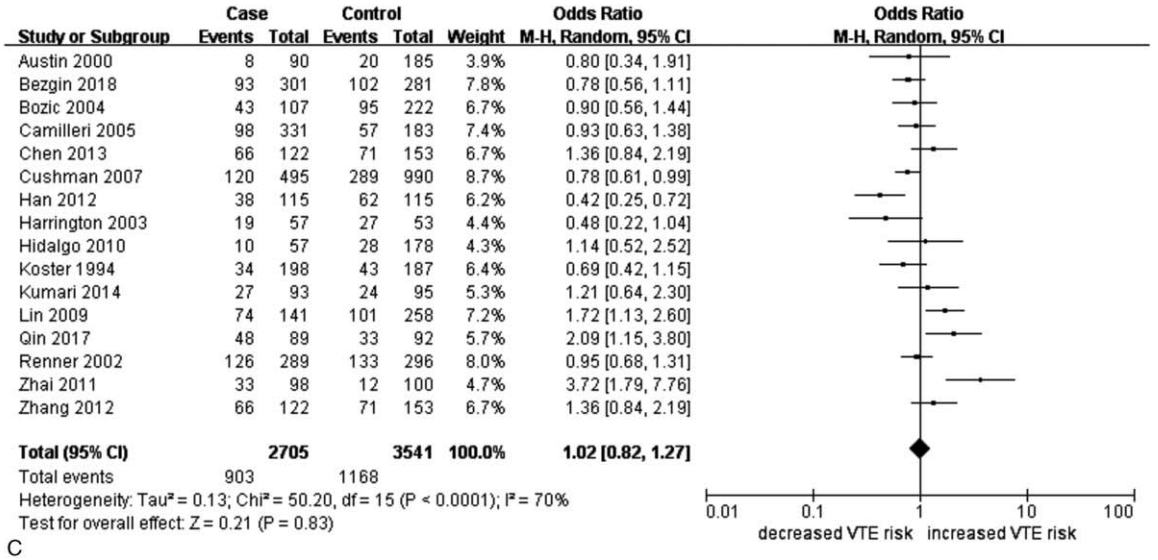
HWE = Hardy–Weinberg equilibrium.



A

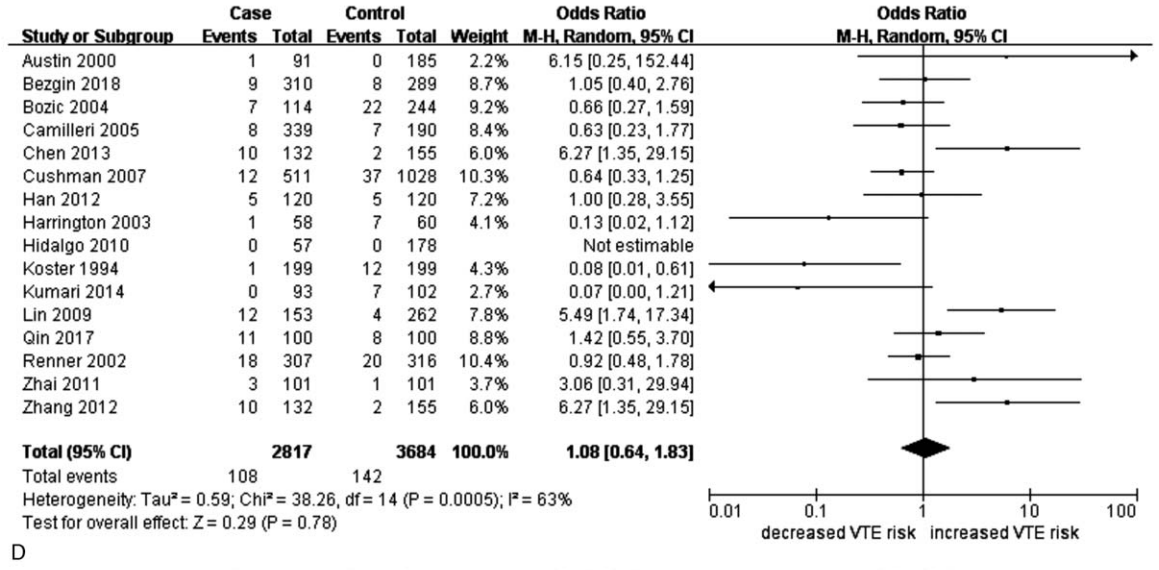


B

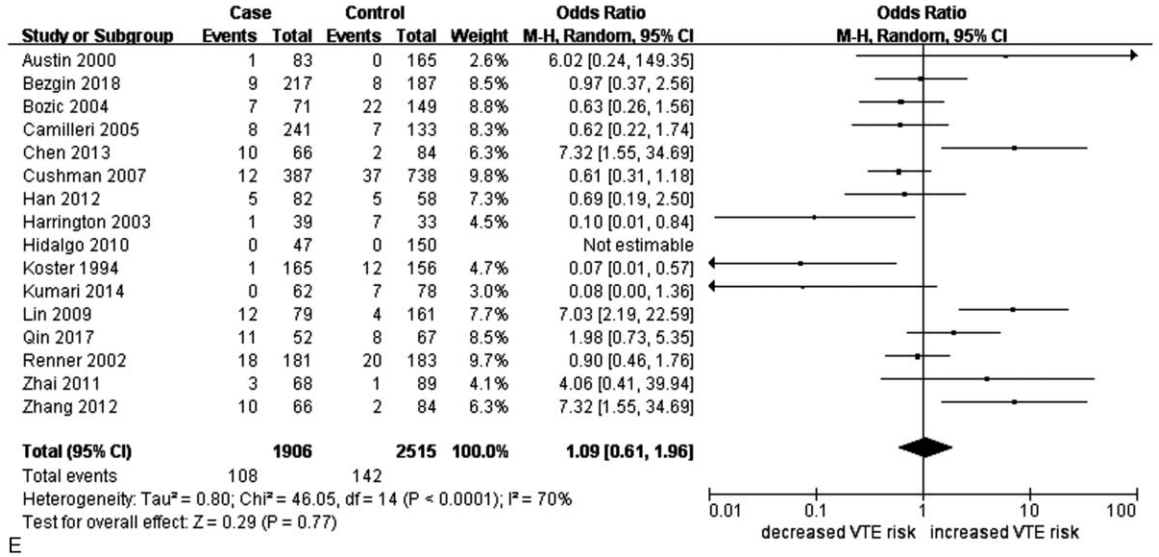


C

Figure 2. Forest plots for association between FGB -455G>A polymorphism and VTE in different genetic models (A: allele model; B: heterozygous model; C: dominant model; D: homozygous model; E: recessive model). FGB = β-fibrinogen, VTE = venous thromboembolism.



D



E

Figure 2. (*continued*).

between FGB 455G>A polymorphism and VTE. Overall, the pooled results based on the whole population revealed no significant correlation between FGB -455G>A variation and VTE susceptibility under all 5 gene comparison models (A:G OR=0.99 95% CI=0.80–1.23 *P*=.96; GA + AA:GG OR=1.01 95% CI=0.80–1.29 *P*=.92; GA:GG OR=1.02 95% CI=0.82–1.27 *P*=.83; AA:GG + GA OR=1.08 95% CI=0.64–1.83 *P*=.78; AA:GG OR=1.09 95% CI=0.67–1.96 *P*=.77). Then subgroup analyses were performed stratified based on ethnicity, source of control, and specific thrombotic disorders. When stratified by ethnicity, significantly statistical decreased risk of VTE was observed among Caucasian population under all 5 gene comparison models (A:G OR=0.80 95% CI=0.70–0.91 *P*=.0006; GA + AA:GG OR=0.80 95% CI=0.68–0.93 *P*=.004; GA:GG OR=0.84 95% CI=0.71–0.98 *P*=.03; AA:GG + GA OR=0.61 95% CI=0.43–0.87 *P*=.007; AA:GG OR=0.57 95% CI=0.40–0.82 *P*=.003),at the mean time a conspicuous decrease in heterogeneity was also observed. Figure 3 displays the forest plot of the association between FGB -455G>A polymorphism and VTE among Caucasian ethnicity. However, no significant association was found between -455G>A polymorphism and VTE susceptibility in light of Asian ethnicity or other stratified subgroup analyses. In terms of association between -148C>T polymorphism and VTE risk, there was no correlation between -148C>T polymorphism and risk of VTE, whether based on aggregate analysis of all populations or subgroup analyses based on different ethnic and thrombotic disease types. Figure 4 displays the forest plot of the association between FGB -148C>T polymorphism and VTE. Table 3 summarizes the main results of overall and subgroup analyses for FGB polymorphisms and VTE.

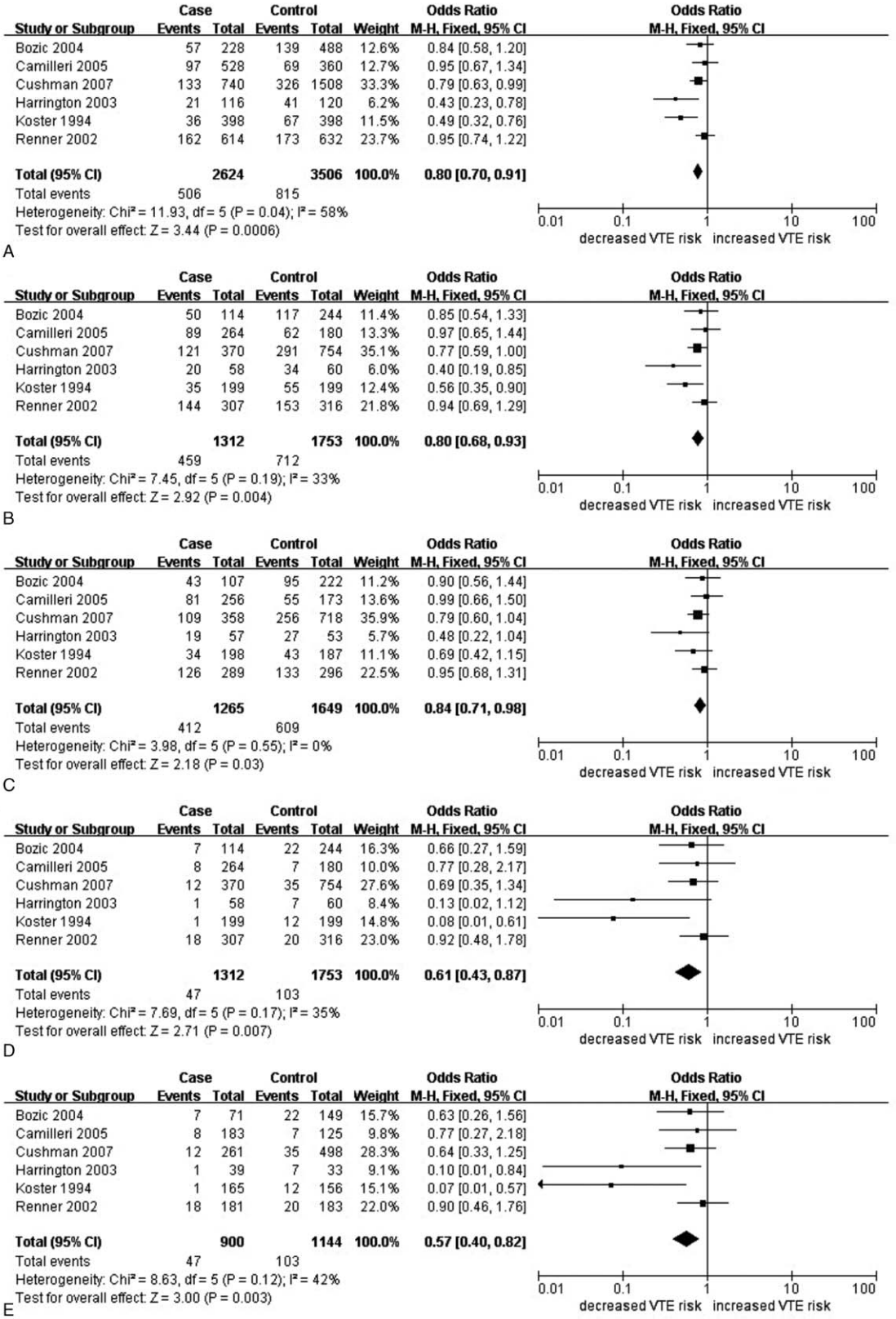


Figure 3. Forest plots for association between FGB -148C>T polymorphism and VTE in different genetic models (A: allele model; B: heterozygous model; C: dominant model; D: homozygous model; E: recessive model). FGB = β-fibrinogen, VTE = venous thromboembolism.

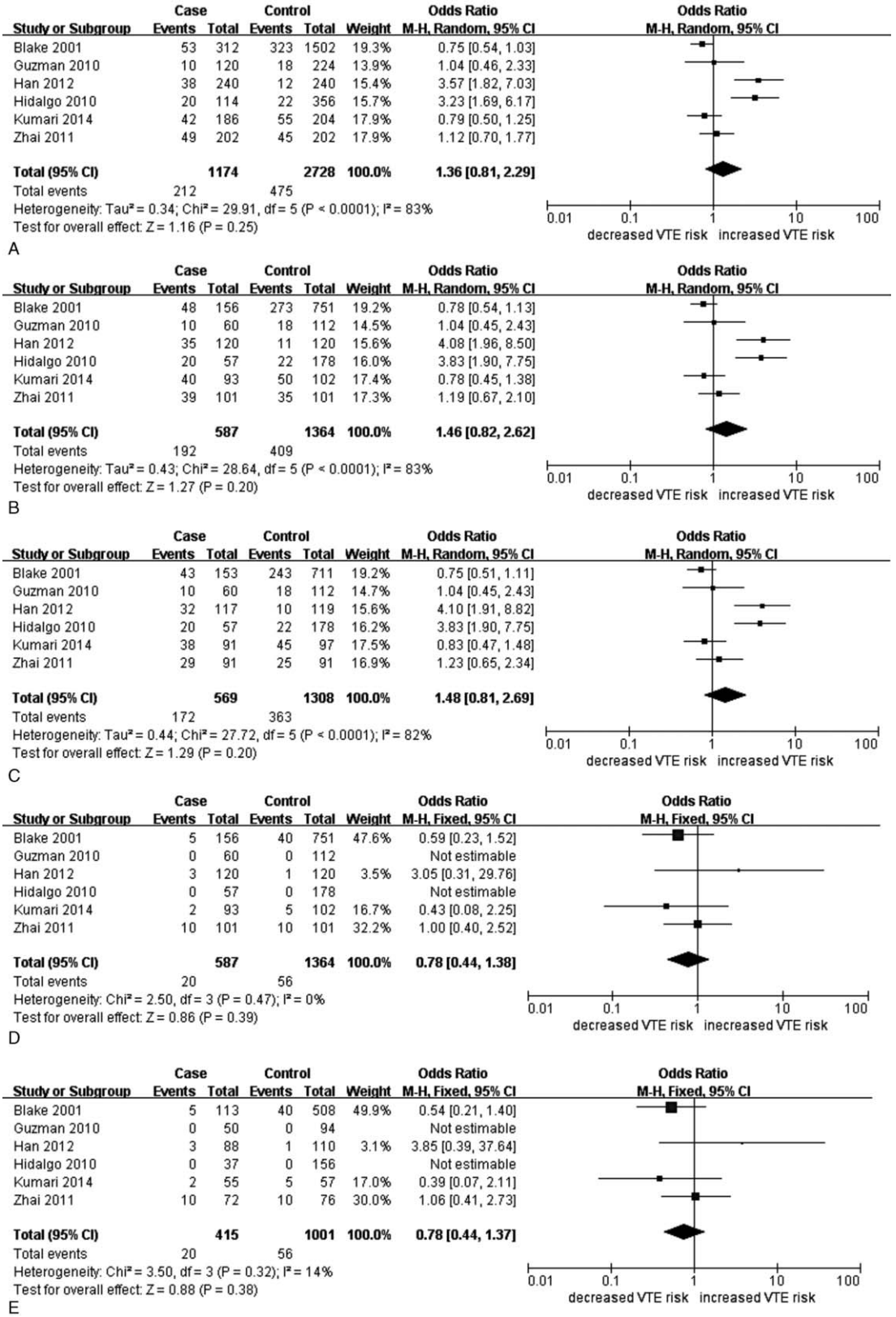


Figure 4. Forest plots for association between FGB -455G>A polymorphism and VTE among Caucasian ethnicity in different genetic models (A: allele model; B: heterozygous model; C: dominant model; D: homozygous model; E: recessive model). FGB = β-fibrinogen, VTE = venous thromboembolism.

Table 3

Results of overall and subgroup analyses for FGB polymorphisms and VTE.

Group/subgroup	Sample size (case/control)	Allele model			Dominant model			Heterozygote model			Recessive model			Homozygote model		
		OR (95% CI)	*P*	I^2	OR (95% CI)	*P*	I^2	OR (95% CI)	*P*	I^2	OR (95%CI)	*P*	I^2	OR (95%CI)	*P*	I^2
455G>A																
Total	2817/3684	0.99 [0.80–1.23]	.96	80%	1.01 [0.80–1.29]	.92	77%	1.02 [0.82–1.27]	.83	70%	1.08 [0.64–1.83]	0.78	63%	1.09 [0.61–1.96]	0.77	70%
Caucasian	1312/1753	0.80 [0.70–0.91]	**.0006**	58%	0.80 [0.68–0.93]	**.004**	33%	0.84 [0.71–0.98]	**.03**	0%	0.61 [0.43–0.87]	**0.007**	35%	0.57 [0.40–0.82]	**0.003**	42%
Asian	1153/1287	1.27 [0.91–1.76]	.15	80%	1.32 [0.88–1.98]	.18	81%	1.29 [0.87–1.91]	.21	78%	2.03 [0.96–4.26]	0.06	56%	1.27 [0.70–3.56]	0.28	66%
HB	803/1038	1.11 [0.74–1.64]	.62	82%	1.04 [0.64–1.70]	.88	82%	0.97 [0.61–1.54]	.90	78%	2.32 [0.87–6.15]	0.09	65%	2.41 [0.77–7.50]	0.13	73%
PB	2014/2646	0.95 [0.73–1.23]	.68	79%	1.00 [0.75–1.32]	.98	75%	1.04 [0.80–1.35]	.75	68%	0.75 [0.41–1.39]	0.36	58%	0.75 [0.38–1.49]	0.41	65%
DVT	1257/1551	1.12 [0.82–1.52]	.48	83%	1.08 [0.75–1.55]	.69	81%	1.06 [0.77–1.47]	.70	74%	1.47 [0.69–3.13]	0.32	72%	1.54 [0.66–3.60]	0.32	77%
PE	159/161	1.17 [0.16–8.46]	.88	95%	1.23 [0.14–10.98]	.85	94%	1.34 [0.18–9.98]	.77	93%	0.62 [0.03–13.57]	0.76	74%	0.62 [0.02–24.00]	0.80	82%
148C>T																
Total	587/1364	1.36 [0.81–2.29]	.25	83%	1.46 [0.82–2.62]	.20	83%	1.48 [0.81–2.69]	.20	82%	0.78 [0.44- 1.38]	0.39	0%	0.78 [0.44–1.37]	0.38	14%
Asian	314/323	1.41 [0.65–3.08]	.39	85%	1.52 [0.62–3.71]	.36	84%	1.57 [0.65–3.80]	.32	82%	0.95 [0.46–1.98]	0.90	0%	1.01 [0.48–2.11]	0.98	21%
Latino	117/290	1.88 [0.62- 5.72]	.26	78%	2.05 [0.57- 7.33]	.27	81%	2.05 [0.57–7.33]	.27	81%	NA			NA		
DVT	180/232	1.97 [0.59–6.61]	.27	81%	2.10 [0.55–7.99]	.28	82%	2.10 [0.55–8.02]	.28	82%	3.05 [0.31–29.76]	0.34	NA	3.85 [0.39–37.64]	0.25	NA

The bold values represent statistically significant differences.

CI = confidence interval, DVT = deep vein thrombosis, FGB = β-fibrinogen, NA = not available, OR = odds ratio, PE = pulmonary embolism, VTE = venous thromboembolism.

3.3. Sensitivity analysis

Sensitivity analysis was performed to assess the impact of each individual study on the overall study. By removing individual studies seriatim, no statistical variation of pooled OR were observed, which indicated that the results of this current meta-analysis were stable. (Figs. 5 and 6).

3.4. Publication bias

Funnel plots showed no significant visual asymmetry in all gene comparison models (Figs. 7 and 8). Egger test was applied to further assess potential publication bias. The results of the Egger test also revealed no significant evidence of publication bias. Table 4 summarizes the main results of the Egger test.

3.5. TSA

On account of the stratified analysis revealed that -455G>A variation was significantly associated with decreased VTE susceptibility among Caucasian population, TSA was performed based on the 5 genetic comparison models of this subgroup to verify the reliability of the synthetic analysis results and the adequacy of the included samples. Under the allele model and the dominant model, the cumulative *Z* curve passed through the traditional boundary threshold and also crossed the trial sequential monitoring boundary, the result confirmed a reliable conclusion in advance although the actually accrued number of participants did not transcend the RIS, enlarging the sample size for subsequent study was not necessary. Under the heterozygous model, although the cumulative *Z* curve did not intersect with the trial sequential monitoring boundary, the cumulative *Z*-statistic crossed above the conventional threshold for statistical significance, and the cumulative samples exceeded the RIS, thereby indicated that the current sample size was sufficient and the pooled calculating result was credible. Under both the recessive model and the homozygous model, the cumulative *Z* curve did not cross with the sequential trial monitoring boundary, and the sample size has not reached RIS, which suggested that more large sample studies may be needed to get a solid conclusion. (Fig. 9A and B)

4. Discussion

Fibrinogen is a 340 KDa plasma glycoprotein synthesized and secreted mainly by liver cells. As the coagulation factor with the highest content, it is not only an crucial component of the coagulation system but also an important acute reactive protein, participate in a variety of physiological and pathological processes.[6,36] The structural and functional abnormalities of fibrinogen caused by single nucleotide polymorphisms (SNPs) in the coding region of fibrinogen gene are associated with a variety of clinical diseases.[9,37–39]

The 3 polypeptide chains of fibrinogen are encoded by 3 independent genes: fibrinogen alpha (*FGA*), fibrinogen beta (*FGB*), and fibrinogen gamma (*FGG*), all of which are located at chromosome 4 (4q23 ~ q32). The *FGB* gene contains 8 exons with a total length of 1476 kb and is responsible for encoding the β chain which is composed of 461 amino acid residues.[6,7,40]

The synthesis of fibrinogen β chain is the dominating speed limiting step in fibrinogen production, so the *FGB* gene is

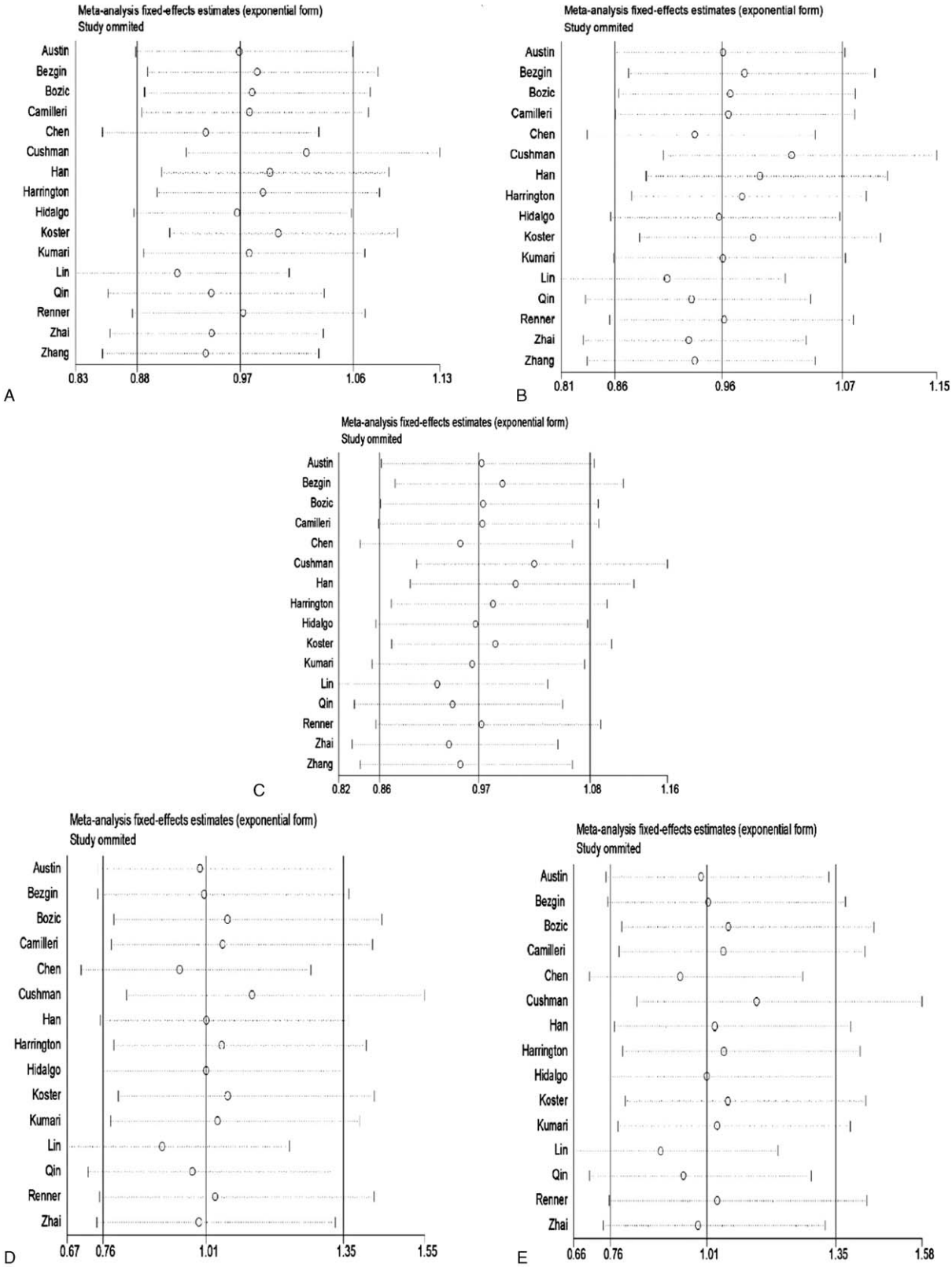


Figure 5. Sensitivity analyses results between FGB -455G>A polymorphism and VTE (A: allele model; B: heterozygous model; C: dominant model; D: homozygous model; E: recessive model). FGB = β-fibrinogen, VTE = venous thromboembolism.

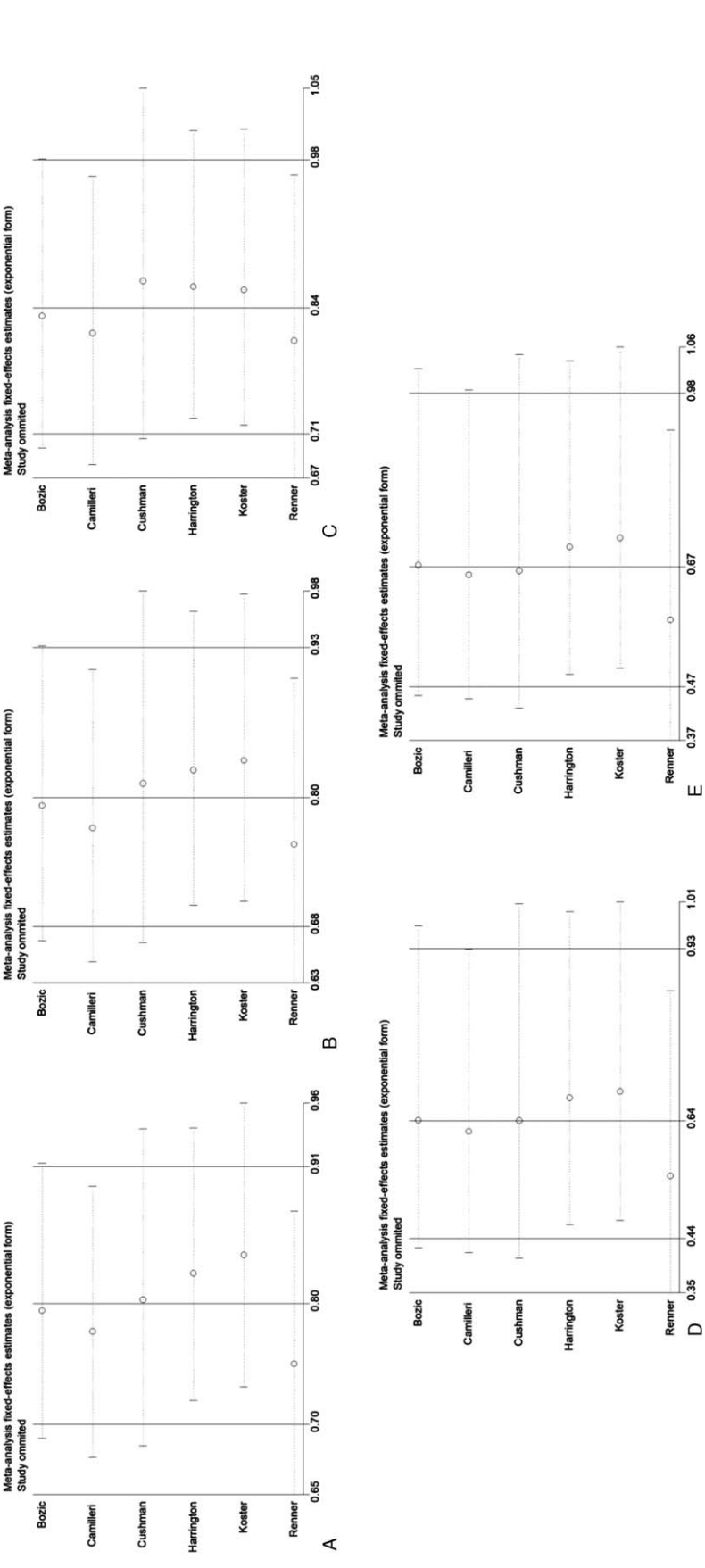


Figure 6. Sensitivity analyses results between FGB -455G>A polymorphism and VTE among Caucasian ethnicity (A: allele model; B: heterozygous model; C: dominant model; D: homozygous model; E: recessive model). FGB = β-fibrinogen, VTE = venous thromboembolism.

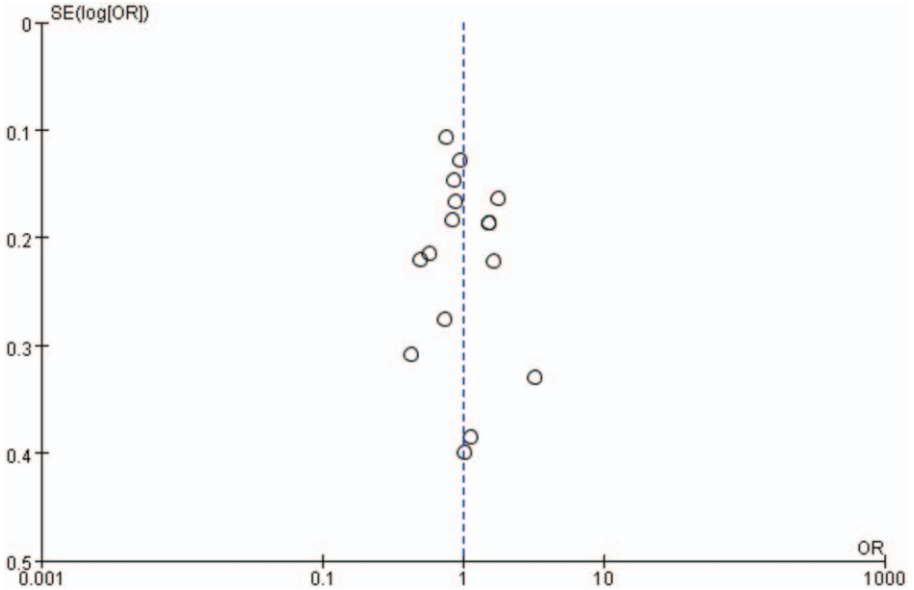


Figure 7. Funnel plot of publication bias for the association between FGB -455G>A polymorphism and VTE under allele model. FGB = β-fibrinogen, VTE = venous thromboembolism.

considered to be associated with changes in plasma fibrinogen levels. At present, more than 10 kinds of SNPs of *FGB* gene have been found, among which 2 SNPs, -455G>A and -148C>T, have been studied most extensively and intensively. -455G>A is the earliest and most studied locus of *FGB* gene polymorphisms. A subsequent case-control study confirmed this inference and suggested that A-allele of -455G>A mutation was significantly associated with elevated fibrinogen levels.[10] Most subsequent studies reached similar conclusions, suggesting that the A allele was positively related to the increase of plasma fibrinogen concentration. Comprehensive assessment of all common gene polymorphisms involving FGA, FGB, and FGG genes suggested that approximately 2% of plasma fibrinogen levels are affected by inherent fibrinogen gene polymorphisms.[41] Another large-

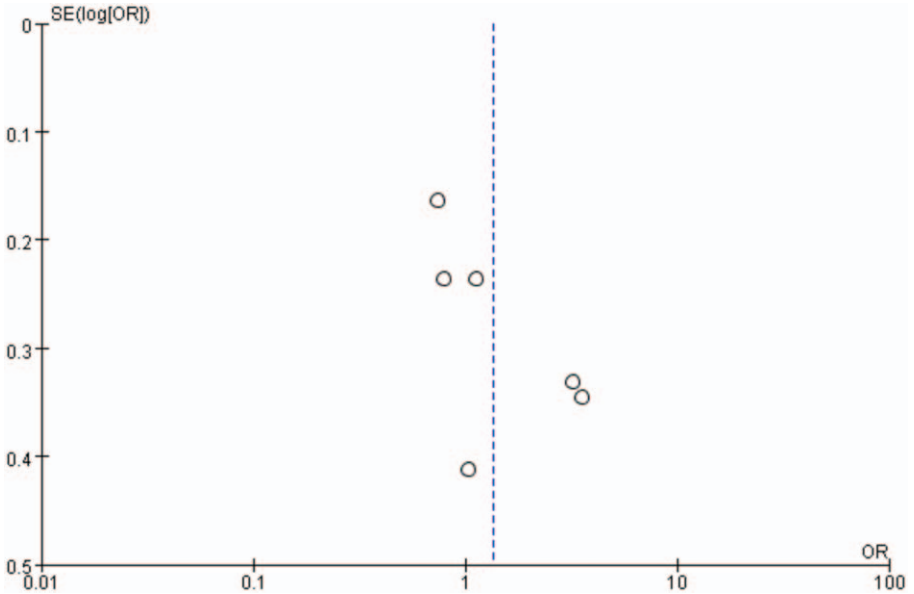


Figure 8. Funnel plot of publication bias for the association between FGB -148C>T polymorphism and VTE under allele model. FGB = β-fibrinogen, VTE = venous thromboembolism.

Table 4

Summary of publication bias tests for association between FGB polymorphisms and VTE.

SNP	Comparison model type	Egger text	
		t	*P*-value
455G>A	Allele model	1.00	.335
	Heterozygous model	1.41	.181
	Dominant model	1.53	.149
	Homozygous model	1.61	.132
	Recessive model	1.55	.145
148C>T	Allele model	1.88	.134
	Heterozygous model	1.59	.187
	Dominant model	1.57	.190
	Homozygous model	1.09	.391
	Recessive model	1.18	.359

FGB = β-fibrinogen, SNP = single nucleotide polymorphism, VTE = venous thromboembolism.

sample study also demonstrated that the FGB -455G>A polymorphism had the strongest influence on increasing fibrinogen levels, and this polymorphism impact on approximately 1% of fibrinogen level variation.[42] In addition, the FGB -455G>A mutation has different effects on increasing fibrinogen in different ethnic groups. Study revealed that FGB -455G>A had the most intense correlation with elevated fibrinogen levels among the European-American, while no similar significant correlation was observed among Asian Americans.[43] Cook et al tested and compared the -455G>A and -148C>T polymorphisms of white, black, and Indian people living in London, and found that there were differences in allele frequency and linkage imbalance among the 3 ethnic groups, which caused changes in plasma fibrinogen level, suggesting that genetic polymorphism and disease susceptibility were different due to diverse ethnicities.[44] The relationship between these 2 SNPs of *FGB* gene with the susceptibility of arteriosclerotic diseases and arterial thrombotic diseases has been extensively explored. Maat et al revealed that the progression of coronary atherosclerosis was rapid in patients with genotype AA, which was speculated to lead to the progression of coronary artery disease by increasing the plasma fibrinogen level.[11] Schmidt et al conducted -148C> T polymorphism analysis on 399 middle-aged and elderly individuals and demonstrated that TT genotype was more advanced than CT and CC genotype in carotid atherosclerosis, suggesting that TT genotype was a genetic risk factor for carotid atherosclerosis in middle-aged and elderly people.[13] However, some studies suggested that the -148C> T polymorphism has no positive significance, a large-scale prospective study showed that -148C> T mutation does not increase the risk of cardiovascular events.[24] Recently, a number of meta-analyses summarized the data of previous studies and concluded that FGB -455G>A and -148C>T polymorphisms were significantly correlated with arterial thrombotic disorders such as acute myocardial infarction, acute coronary syndrome, and ischemic stroke, pooled results indicated these 2 SNPs significantly increase the risk of these diseases.[17,45,46]

The strength of this present meta-analysis lists as follows: By setting and performing rigorous inclusion and exclusion criteria, a total of 18 studies were incorporated in this current meta-analysis study, including 3033 VTE cases and 4547 healthy controls, covering 5 ethnicities among 10 countries. We systematically evaluated the contribution of *FGB* gene polymorphisms to the occurrence of VTE. The pooled results illuminate FGB -455G>A polymorphism is negative correction with the susceptibility to VTE in the Caucasian ethnicity subtype, which participates as a protective factor in the occurrence of VTE. After evaluated by sensitivity analysis and TSA, the ultimate pooled results showed high quality and reliability.

Potential limitations of the present meta-analysis study should be mentioned. First, in addition to the subgroup analysis of the Caucasian population, there was significant heterogeneity in both the aggregate analysis including all the population groups and the subgroup analysis classified by race, source of control group, and disease type, suggesting that the pooled results obtained in these analyses were uncertainly stable. The main sources of heterogeneity may root in the following aspects: on the one hand, VTE includes P and DVT of lower limbs, and there may be differences in the etiology of specific thrombotic disease, several articles enrolled in this present meta-analysis are studies of mixed diseases; On the other hand, a variety of high risk factors for venous thrombosis, such as gender, age, history of traumatic surgery, tumor history, and other confounding factors also affect the heterogeneous results. These potential interfering factors were not corrected due to the lack of data in the original literature. Second, the protective effect of FGB -455G>A in the occurrence of venous thrombotic disease among Caucasians were not observed in the calculation results of other subgroup analyses, suggesting that more multi-population studies based on large samples may be needed to explore whether the FGB gene variation participate different roles in different ethnicity. Third, the limitation of retrieval language to Chinese and English may result in relevant eligible studies not being included in the analysis. Finally, VTE is a kind of disease with complex multi-factor etiology and pathogenesis, its morbidity is closed associated with age, the most recent study also pointed out that the function of fibrinogen in the human body also changes along with the age growth.[47] Restricted to the original research materials, the present meta-analysis did not conduct subgroups analysis based on aging stratification.

Despite some potential limitations, this meta-analysis still has several luminescent spots. So far, this is the first known meta-analysis study aimed to clarify the vague correlation between *FGB* gene polymorphisms and VTE risk. Since many previous studies have suggested that the SNPs of FGB gene are associated with increased risk of arterial thrombotic disease, and subsequently confirmed by several meta-analyses, the results of this current meta-analysis revealed FGB polymorphisms have a distinct different impact on the pathogenesis of venous thrombosis in contrast to its role in the pathogenesis of arterial thrombosis, which generate novel insights into the association of FGB polymorphisms with thrombotic disease. Of course, this conclusion requires more relevant investigations with larger sample size, more ethnic groups and strict protocols to further validate and more relevant experimental research to clarify the potential specific mechanism.

In summary, the results of this current meta-analysis demonstrated that FGB -455G>A polymorphism contributes to reduced risk of VTE among Caucasian population, but no similar association was observed in other stratified analyses, and there was no significant correlation between FGB -148C>T polymorphism and VTE susceptibility.

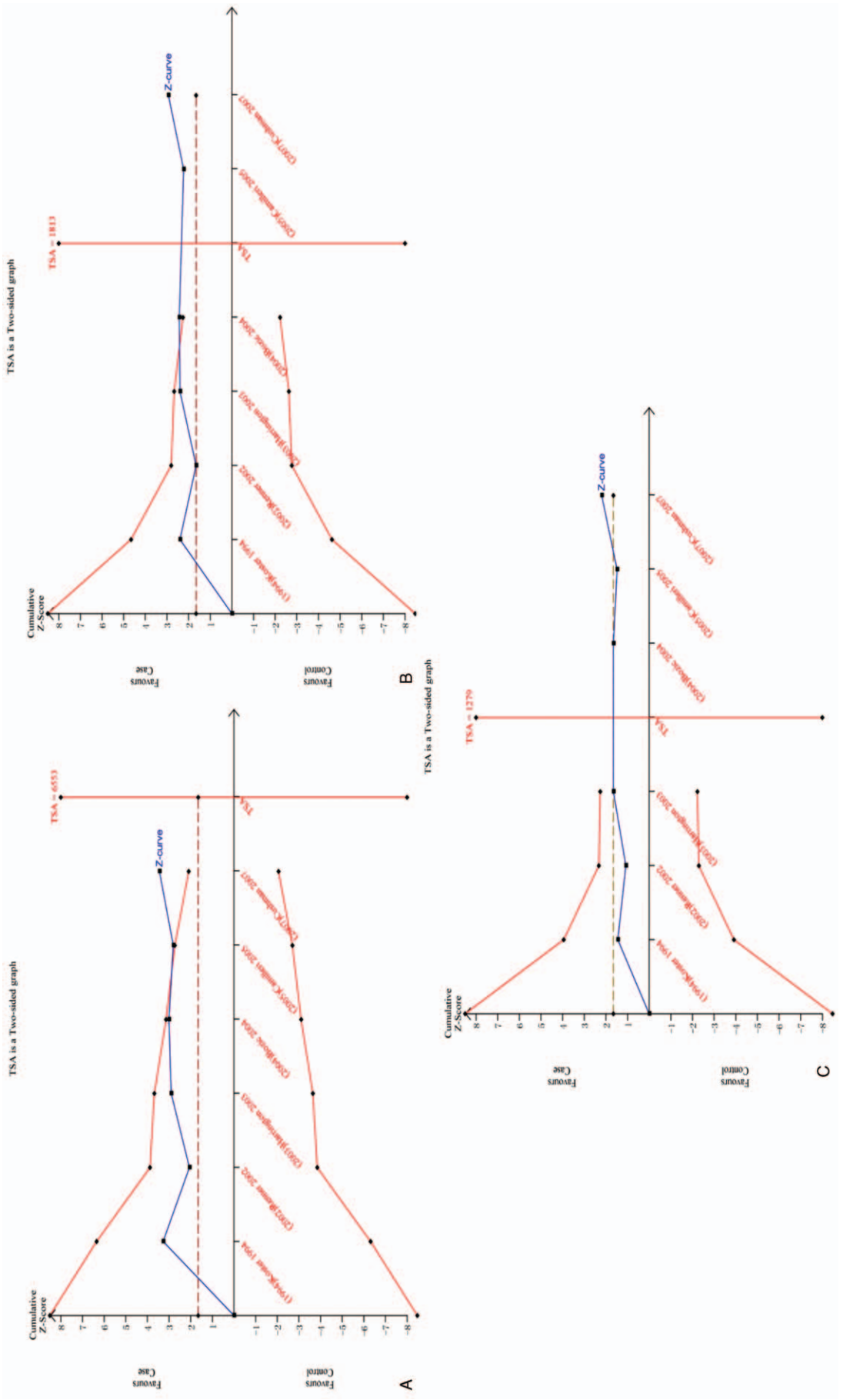


Figure 9. (A and B) Trial sequential analysis results of the association between FGB -455G>A polymorphism and VTE among Caucasian ethnicity (A: allele model; B: heterozygous model; C: dominant model; D: homozygous model; E: recessive model). FGB = β-fibrinogen, VTE = venous thromboembolism.

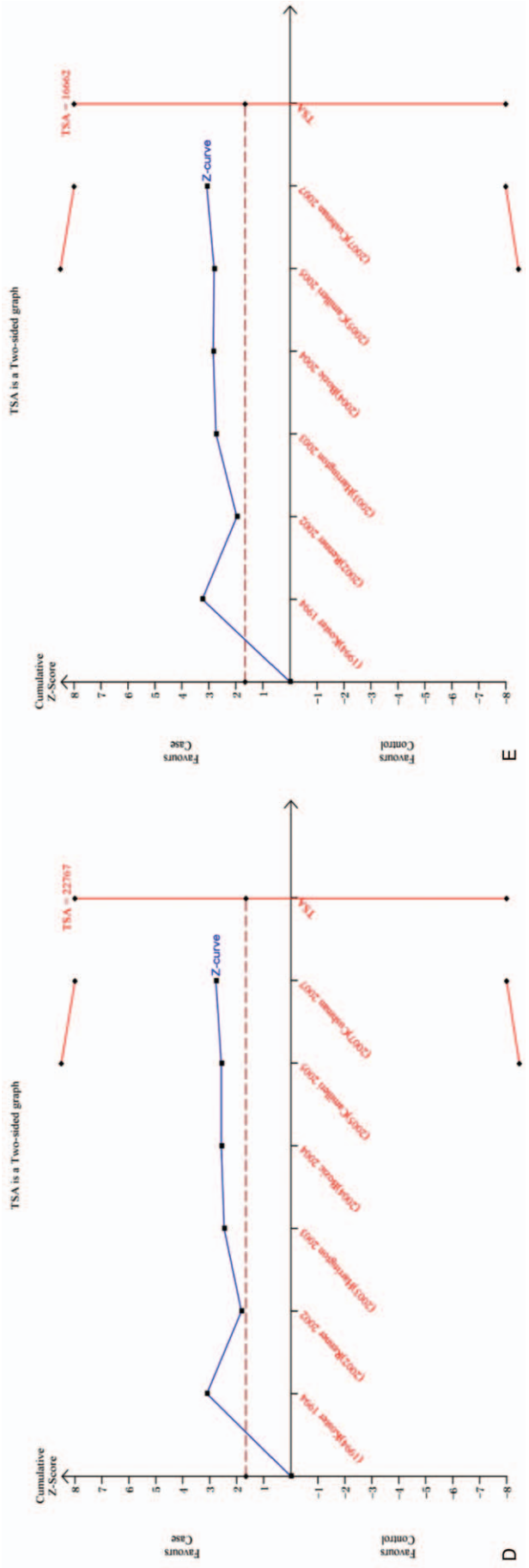


Figure 9. (*continued*).

Acknowledgments

The authors sincerely thank all the colleagues working in the Department of Vascular Surgery of the First People's Hospital of Lianyungang.

Author contributions

Conceptualization: Honggang Zhang.
Data curation: Da Li.
Formal analysis: He Huang.
Investigation: Da Li, Xiaosong Zhang.
Methodology: Honggang Zhang.
Project administration: Honggang Zhang.
Resources: Da Li.
Software: Da Li, Xiaosong Zhang, He Huang.
Supervision: Honggang Zhang.
Visualization: He Huang.
Writing – original draft: Da Li.
Writing – review and editing: Honggang Zhang.

References

[1] Tagalakis V, Patenaude V, Kahn SR, et al. Incidence of and mortality from venous thromboembolism in a real-world population: the Q-VTE study cohort. Am J Med 2013;126:832.e13–21.
[2] Heit JA. Venous thromboembolism: disease burden, outcomes and risk factors. J Thromb Haemost 2005;3:1611–7.
[3] Crous-Bou M, Harrington LB, Kabrhel C. Environmental and genetic risk factors associated with venous thromboembolism. Semin thromb Hemost 2016;42:808–20.
[4] de Haan HG, Bezemer ID, Doggen CJ, et al. Multiple SNP testing improves risk prediction of first venous thrombosis. Blood 2012;120:656–63.
[5] Lindstrom S, Brody JA, Turman C, et al. A large-scale exome array analysis of venous thromboembolism. Genet Epidemiol 2019;43:449–57.
[6] Mosesson MW. Fibrinogen and fibrin structure and functions. J Thromb Haemost 2005;3:1894–904.
[7] Mosesson MW, Siebenlist KR, Meh DA. The structure and biological features of fibrinogen and fibrin. Ann N Y Acad Sci 2001;936:11–30.
[8] Koenig W. Fibrin(ogen) in cardiovascular disease: an update. Thromb Haemost 2003;89:601–9.
[9] Tousoulis D, Papageorgiou N, Androulakis E, et al. Fibrinogen and cardiovascular disease: genetics and biomarkers. Blood Rev 2011;25:239–45.
[10] van 't Hooft FM, von Bahr SJ, Silveira A, et al. Two common, functional polymorphisms in the promoter region of the beta-fibrinogen gene contribute to regulation of plasma fibrinogen concentration. Arterioscler Thromb Vasc Biol 1999;19:3063–70.
[11] de Maat MP, Kastelein JJ, Jukema JW, et al. -455G/A polymorphism of the beta-fibrinogen gene is associated with the progression of coronary atherosclerosis in symptomatic men: proposed role for an acute-phase reaction pattern of fibrinogen. REGRESS group. Arterioscler Thromb Vasc Biol 1998;18:265–71.
[12] Kumar A, Misra S, Kumar P, et al. Association between beta-fibrinogen C148T gene polymorphism and risk of ischemic stroke in a North Indian population: a case-control study. Pulse 2017;4:165–71.
[13] Schmidt H, Schmidt R, Niederkorn K, et al. Beta-fibrinogen gene polymorphism (C148–>T) is associated with carotid atherosclerosis: results of the Austrian Stroke Prevention Study. Arterioscler Thromb Vasc Biol 1998;18:487–92.
[14] Carter AM, Mansfield MW, Stickland MH, et al. Beta-fibrinogen gene-455 G/A polymorphism and fibrinogen levels. Risk factors for coronary artery disease in subjects with NIDDM. Diabetes Care 1996;19:1265–8.
[15] Hu X, Wang J, Li Y, et al. The beta-fibrinogen gene 455G/A polymorphism associated with cardioembolic stroke in atrial fibrillation with low CHA2DS2-VaSc score. Sci Rep 2017;7:17517.
[16] Gu L, Liu W, Yan Y, et al. Influence of the beta-fibrinogen-455G/A polymorphism on development of ischemic stroke and coronary heart disease. Thromb Res 2014;133:993–1005.

[17] Luo H, Li X, Jiang A, et al. Associations of beta-fibrinogen polymorphisms with the risk of ischemic stroke: a meta-analysis. J Stroke Cerebrovasc Dis 2019;28:243–50.
[18] Koster T, Rosendaal FR, Reitsma PH, et al. Factor VII and fibrinogen levels as risk factors for venous thrombosis. A case-control study of plasma levels and DNA polymorphisms–the Leiden Thrombophilia Study (LETS). Thromb Haemost 1994;71:719–22.
[19] Renner W, Cichocki L, Forjanics A, et al. G-455A polymorphism of the fibrinogen beta gene and deep vein thrombosis. Eur J Clin Invest 2002;32:755–8.
[20] Camilleri RS, Cohen H. No association between pulmonary embolism or deep vein thrombosis and the -455G/A beta-fibrinogen gene polymorphism. Blood Coagul Fibrinolysis 2005;16:193–8.
[21] Bozic M, Teran N, Peterlin B, et al. Fibrinogen polymorphisms TaqI, HaeIII and BclI are not associated with a higher risk of deep vein thrombosis. Pathophysiol Haemost Thromb 2003;33:164–9.
[22] Austin H, Hooper WC, Lally C, et al. Venous thrombosis in relation to fibrinogen and factor VII genes among African-Americans. J Clin Epidemiol 2000;53:997–1001.
[23] Bezgin T, Kaymaz C, Akbal O, et al. Thrombophilic gene mutations in relation to different manifestations of venous thromboembolism: a single tertiary center study. Clin Appl Thromb Hemost 2018;24:100–6.
[24] Blake GJ, Schmitz C, Lindpaintner K, et al. Mutation in the promoter region of the beta-fibrinogen gene and the risk of future myocardial infarction, stroke and venous thrombosis. Eur Heart J 2001;22:2262–6.
[25] Chen YS, Hou XH, Zhang XH. Association between fibrinogen gene 455G/A and PAI gene 4G/5G polymorphisms and deep venous thrombosis. Chin J Gerontol 2013;33:2278–80.
[26] Cushman M, Cornell A, Folsom AR, et al. Associations of the beta-fibrinogen Hae III and factor XIII Val34Leu gene variants with venous thrombosis. Thromb Res 2007;121:339–45.
[27] Guzman N, Lanas F, Salazar LA. Influence of Amerindian mitochondrial DNA haplogroups on thrombosis susceptibility and frequency of four genetic prothrombotic variants in Southern Chilean subjects. Clin Chim Acta 2010;411:444–7.
[28] Han S. Association Between Idiopathic Deep Venous Thrombosis and Genetic Polymorphisms in the Promoter Region of β-Fibrinogen Gene (Doctoral Dissertation). 2013;Kunming Medical University,
[29] Harrington DJ, Malefora A, Schmeleva V, et al. Genetic variations observed in arterial and venous thromboembolism–relevance for therapy, risk prevention and prognosis. Clin Chem Lab Med 2003;41:496–500.
[30] Hidalgo A, Salazar-Sanchez L, Cartin M, et al. Prevalence of the fibrinogen β-chain, angiotensin-converting enzyme and plasminogen activator inhibitor-1 polymorphisms in Costa Rican young adults with thrombotic disease. Cell Biochem Funct 2010;28:367–73.
[31] Kumari B, Srivastava S, Chatterjee T, et al. Study of associated genetic variants in Indian subjects reveals the basis of ethnicity related differences in susceptibility to venous thromboembolism. Thrombosis 2014;2014: 182762.
[32] Lin W. Relationship Between Fibrinogen Gene β-455G/A,Arg448Lys Polymorphisms and Plasma Fibrinogen Concentration, Lower Extremity Deep Venous Thrombosis (Master Dissertation). 2009;Fujian Medical University,
[33] Qin S, Tian H, Zhang J, et al. Correlation between fibrinogen B β-455G/A polymorphism and deep vein thrombosis. South China J Cardiovasc Dis 2017;23:435–8.
[34] Zhai ZG, Wang C, Yang YH, et al. Study on the relationship between polymorphisms of susceptible genes in coagulation pathway related to pulmonary thromboembolism in Chinese Han population. Chin J Epidemiol 2006;27:165–9.
[35] Zhang XH, Chen YS, Hou XH, et al. Correlation between fibrinogen gene 455G/A, PAI gene 4G/5G polymorphisms and deep venous thrombosis of lower limbs. Pract J Cardiac Cereb Pneumal Vasc Dis 2012;20:665–6.
[36] Blomback B. Fibrinogen and fibrin–proteins with complex roles in hemostasis and thrombosis. Thromb Res 1996;83:1–75.
[37] Green FR. Fibrinogen polymorphisms and atherothrombotic disease. Ann N Y Acad Sci 2001;936:549–59.
[38] Siegerink B, Rosendaal FR, Algra A. Genetic variation in fibrinogen; its relationship to fibrinogen levels and the risk of myocardial infarction and ischemic stroke. J Thromb Haemost 2009;7:385–90.
[39] Tiscia GL, Margaglione M. Human fibrinogen: molecular and genetic aspects of congenital disorders. Int J Mol Sci 2018;19:1597.
[40] Fish RJ, Neerman-Arbez M. Fibrinogen gene regulation. Thromb Haemost 2012;108:419–26.
[41] Reiner AP, Carty CL, Carlson CS, et al. Association between patterns of nucleotide variation across the three fibrinogen genes and plasma fibrinogen levels: the Coronary Artery Risk Development in Young Adults (CARDIA) study. J Thromb Haemost 2006;4:1279–87.
[42] Kathiresan S, Yang Q, Larson MG, et al. Common genetic variation in five thrombosis genes and relations to plasma hemostatic protein level and cardiovascular disease risk. Arterioscler Thromb Vasc Biol 2006;26:1405–12.
[43] Carty CL, Cushman M, Jones D, et al. Associations between common fibrinogen gene polymorphisms and cardiovascular disease in older adults. The Cardiovascular Health Study. Thromb Haemost 2008;99:388–95.
[44] Cook DG, Cappuccio FP, Atkinson RW, et al. Ethnic differences in fibrinogen levels: the role of environmental factors and the beta-fibrinogen gene. Am J Epidemiol 2001;153:799–806.
[45] Chen X, Xu M, Jin L, et al. Association of beta-fibrinogen gene -148C/T and -455G/A polymorphisms and coronary artery disease in Chinese population: a meta analysis. Sci China C Life Sci 2008;51:814–20.
[46] Cheng SY, Zhao YD, Zeng JW, et al. A polymorphism (-455G>A) in the beta-fibrinogen gene is associated with an increased risk of cerebral infarction in the Chinese population: a meta-analysis. J Renin Angiotensin Aldosterone Syst 2015;16:399–408.
[47] Gligorijevic N, Zamorova Krizakova M, Penezic A, et al. Structural and functional changes of fibrinogen due to aging. Int J Biol Macromol 2018;108:1028–34.