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GENETICS

Associations of *ACE I/D, AGT M235T* gene polymorphisms with pregnancy induced hypertension in Chinese population: a meta-analysis

Ming Zhu · Jie Zhang · Shaofa Nie · Weirong Yan

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Abstract
Purpose There have been many studies concerning the associations of angiotensin-converting enzyme (ACE) *I/D*, angiotensinogen (AGT) *M235T* polymorphisms with pregnancy induced hypertension (PIH) among Chinese populations. However, the results were inconsistent, prompting the necessity of meta-analysis.
Methods Studies published in English and Chinese were mainly searched in EMbase, PubMed and CBM up to January 2012.
Results Twenty-three studies with 3,551 subjects for *ACE I/D* and seven studies with 1,296 subjects for *AGT M235T* were included. Significant associations were found between *ACE I/D* and PIH under dominant, recessive and allelic models. A separate analysis confined to preeclampsia suggested that *ACE I/D* was associated with preeclampsia under recessive model and allelic model, but not dominant model. Stratified analyses were conducted as meta-regression analysis indicated that the sample size of case group was a significant source of heterogeneity, which suggested no significant association between *ACE I/D* and PIH in the subgroup of more than 100 cases. Associations were found between *AGT M235T* and PIH under dominant genetic model (OR=1.59; 95 %CI: 1.04–2.42), recessive genetic model (OR=1.60; 95 %CI: 1.07–2.40), and allelic model (OR=1.40; 95 %CI: 1.17–1.68). No publication bias was found in either meta-analysis.
Conclusions The present meta-analysis suggested significant associations between *ACE I/D, AGT M235T* and PIH in Chinese populations. However, no significant association was found between *ACE I/D* and PIH in the subgroup of more than 100 cases. Studies with larger sample sizes are necessary to investigate the associations between gene polymorphisms and PIH in Chinese populations.

Keywords Angiotensin-converting enzyme (ACE) · Angiotensinogen (AGT) · Meta-analysis · Polymorphism · Pregnancy induced hypertension (PIH)

Capsule *ACE I/D* and *AGT M235T* were associated with PIH in Chinese, however the association became insignificant for *ACE I/D* in subgroup with a large sample size

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Introduction

Pregnancy induced hypertension (PIH), defined as a condition in pregnant women with elevated systolic (≥140 mmHg) or diastolic (≥90 mmHg) blood pressure on at least two occasions 6 h apart, generally occurs after 20 weeks of gestation and returns to normal 12 weeks postpartum. PIH is the most common obstetrical complication of pregnancy, as well as a leading cause of maternal and perinatal mortality and morbidity. The incidence of PIH is 9 %, and the mortality is 3.3/100,000 in China [1], that exerts a great burden on medical and social expenditures.

Research supports the etiology and pathogenesis of PIH being due to the interaction of genetic and environmental factors. The renin-angiotensin system (RAS) plays a pivotal

role in the regulation of blood pressure and electrolyte balance [2]. RAS consists of renin, angiotensinogen (AGT), angiotensin converting enzyme (ACE), two major types of angiotensin receptors (AT1R, AT2R), etc. Studies have demonstrated that RAS components experience major changes during a normal pregnancy: the excretion of AGT and angiotensin II (ANG II) increases while the level of serum ACE decreases. Comparing with normotensive pregnant women, the ACE level of PIH patients is approximately at the same level as before pregnancy, while their renin and ANG II level become lower [3]. The alterations of these vasoactive elements indicated that RAS might play a crucial role in the development of PIH [4]. Thus, extensive studies have been carried out to investigate the associations between RAS related gene polymorphisms and PIH.

AGT, the precursor of angiotensin peptides, is cleaved to angiotensin I by renin, and then Angiotensin I is converted into bioactive angiotensin II by ACE, which is the major enzyme of RAS [5]. *ACE I/D* (rs4646994) and *AGT M235T* (rs699) are two of the most frequently investigated RAS related gene polymorphisms. *ACE I/D* corresponds to either the *presence* (insertion, I) or *absence* (deletion, *D*) of a *287 bp Alu* repeat in *intron* 16 on chromosome 17; the polymorphism of *AGT M235T* is a replacement of methionine by threonine at amino-acid position 235. Although extensive investigations had been taken to evaluate the associations of *ACE I/D* and/or *AGT M235T* with PIH in Chinese populations, the results were inconsistent with each other.

A meta-analysis, which was based on an ethnically mixed population, reported there were significant associations between preeclampsia and *ACE I/D, AGT M235T* in 2006 [6]. However, this analysis included only one study of Chinese population. Evidence has shown the distributions of genes polymorphism are various in different ethnic groups, for example the frequency of the *T235* allele in Australians and Chinese population are of big difference [7]. Studies also found that the incidence of ACE gene deletion varies among different study populations and geographic regions [8]. Results drawn from the previous meta-analysis may not be appropriate for Chinese populations, thus we aimed to conduct a meta-analysis investigating the associations between the gene polymorphisms of *ACE I/D, AGT M235T* and PIH in Chinese populations in the current study.

Methods

Data sources

PubMed, EMbase, and CBM (China Biological Medicine), as the main sources used, were searched up to January 2012. The preliminary terms used for searching were "pregnancy induced hypertension", "preeclampsia", "eclampsia", "ACE", "angiotensin-converting enzyme", "AGT", "angiotensinogen", "gene polymorphism", and "Chinese" in MeSH terms, and language searched was limited to English and Chinese. Additional searching was performed using CNKI (National Knowledge Infrastructure), WANFANG databases, and Google Scholar to avoid inadvertent omissions. Further, relevant researches and references cited in these publications were also searched to complement the analysis.

Study selection

Inclusion criteria were set as follows: (1) Articles reported associations between PIH and *ACE I/D* and/or *AGT M235T*; (2) Studies based on case–control design with clear diagnostic criteria. PIH was diagnosed for blood pressure ≥140/90 mmHg, and returned to normal 12 weeks postpartum; mild preeclampsia was defined for BP≥140/90 mmHg with proteinuria ≥300 mg/24 h, and severe preeclampsia was diagnosed as BP≥160/110 mmHg and proteinuria ≥2.0 g/24 h with additional symptoms and medical signs; eclampsia was diagnosed for tonic–clonic seizures in pregnant women with high blood pressure and proteinuria; (3) Genotype frequencies were reported in both case and control groups. Methods used for genotyping were validated, and subjects were unrelated individuals.

Exclusion criteria were defined as follows: (1) studies with only allele frequencies reported; (2) studies with smaller data sets were excluded for duplicate publications; (3) the distributions of genotype frequency were deviated from Hardy-Weinberg Equilibrium (HWE) in control groups; (4) subjects enrolled in the studies having other diseases (cardiovascular disease, diabetes mellitus, renal disease, or other pregnancy complications).

Data extraction

The following information were extracted from each publication including the name of first author, year of publication, location, age, gestational weeks, parity, sample size, and distributions of genotype and allele frequencies in both case and control groups. All information was checked and collected by two researchers independently, and the inconsistencies were examined and discussed until a unanimous interpretation was reached. The citations were ordered by the year of publications in tables.

Statistical analysis

Genetic model statistic analysis

HWE test was calculated with chi-square goodness of fit by two researchers when the original studies did not report this information. If the distributions of genotype frequency were inconsistent with HWE in control group, the study was excluded from further analysis. The associations of the two gene polymorphisms with PIH were evaluated under the dominant

genetic model, recessive genetic model, and allelic model. Heterogeneity in meta-analysis was estimated with the I^2 test (when I^2>50 %, it was considered that there was a significant heterogeneity between studies) and chi-square-based Q statistic (p value <0.05 was used). A random effect model (DerSimonian-Laird method) was applied instead of a fixed effect model (Mantel-Haenszel method) when heterogeneity was evident.

Cumulative meta-analysis and sensitivity analysis

Cumulative meta-analysis was conducted according to the year of publication in order to identify the influence of the first published study on the subsequent publications, and evaluate the stability of the effect estimations over time. To identify potential influential studies, sensitivity analysis was carried out to check whether any of these studies would bias the overall estimation under either genotypic models or the allelic model.

Meta-regression and subgroup analysis

Meta-regression was applied to explore the source of between-study heterogeneity when the number of recruited studies was more than ten [9, 10]. Detailed information which could be retrieved from all original articles, such as the year of publication, sample size of case group, sample size of total study subjects, parity and geographic region (southern versus northern: demarcating by the Qinling Mountains of China's Huaihe River), were taken into consideration for explaining the heterogeneity. Further, stratified analysis was conducted if there were any variables found as significant sources of heterogeneity (P_{Het}=0.05).

Publication bias

Evidence of publication bias was examined by a visual inspection of funnel plots, and Egger's test was used to measure the asymmetry of funnel plot.

All the above mentioned analyses were performed by STATA 11.0, P value less than 0.05 was considered to be statistically significant.

Results

ACE I/D

The literature search generated two English papers and 42 Chinese papers for the association studies between *ACE I/D*

Table 1 Characteristics of included studies for *ACE I/D* and PIH

Author	Year	Location	Enrollment	Cases	Controls	Parity	Comments
Zhu [18]	1998	Shanghai	1994–1995	35	25	primiparity	Singleton, excluded other pathologies
Zhou [19]	1999	Shandong	1995.9–1998.8	60	76	primiparity	Singleton, excluded other pathologies
Wang [20]	1999	Heibei	1996.12–1997.8	61	70	N	Excluded other pathologies
Hong [21]	2000	Zhejiang	1997.12–1998.6	52	100	N	Excluded other pathologies
Bai [22]	2002	Sichuan	1998.8–2000.5	81	205	N	Han ethnic, no family history, excluded other pathologies
Gao [23]	2002	Heilongjiang	2000.12–2001.3	110	81	primiparity	Singleton, excluded other pathologies
Wu [24]	2002	Jiangxi	1998.5–2000.12	56	52	primiparity	Singleton, excluded other pathologies
Mao [25]	2004	Heilongjiang	1998.1–2002.12	62	120	primiparity	Singleton, excluded other pathologies
Wang [26]	2004	Jiangxi	2001.9–2003.4	100	100	N	Han ethnic, excluded other pathologies
Wang# [27]	2004	Tianjin	2001–2003	41	50	primiparity	Singleton, excluded other pathologies
Li [11]	2005	Shandong	1998.8–2002.10	103	76	primiparity	Singleton, excluded other pathologies
Chen [28]	2006	Zhejiang	2002–2004	92	85	N	Excluded other pathologies
Li [29]	2006	Guangdong	2000.3–2004.3	54	100	N	Excluded gestational diabetes and medical complications.
Li [12]	2007	Shandong	2003.10–2006.9	133	105	M	Han ethnic, excluded other pathologies
Song [13]	2007	Hubei	2004.7–2005.2	45	45	N	Exclude cardiovascular disease, nephropathy, et al.
Huang [30]	2008	Yunan	2004.12–2007.6	90	120	primiparity	Singleton, excluded other pathologies
Ren [31]	2008	Shanxi	2006.8–2007.2	60	54	primiparity	Excluded other pathologies
Zhan [14]	2008	Jiangxi	2006.9–2007.9	120	60	N	Excluded other pathologies
Cui [15]	2008	Tianjin	2005.8–2006.2	63	40	N	Singleton, excluded other pathologies.
Deng [16]	2010	Henan	2009–8–2010.5	50	100	N	Excluded other pathologies
Yue [17]	2011	Sichuan	2007.3–2009.5	43	44	N	Excluded other pathologies
Yan [32]	2011	Henan	2008.1–2009.12	113	113	M	Han ethnic, excluded other pathologies

N: Not mentioned M: Cases and controls were matched. #: To distinguish the two studies with the same last name of first author

and PIH. Among these 44 articles, six articles were excluded for the deviation from HWE; eight studies were duplicated publications with the same population; and another seven papers were excluded for non-compliance with the design, such as studying the association of fetal genotype and PIH, testing the genotype using the tissue of placenta maternal site, etc. Finally, 23 articles, involving 1,684 patients and 1,867 controls, met the selection criteria. In the 23 studies, patients in seven studies were of preeclampsia [11–17]. Cases in the other 16 studies were the mixture of gestational hypertension, preeclampsia, and eclampsia patients, which we failed to retrieve genotype data for different diseases in the original studies. The characteristics of the included studies and women were shown in Tables 1 and 2, and the geographical distributions of the subjects from 13 provinces in China were shown in Fig. 1.

Genetic model statistic analysis

Figure 2 presented the forest plots of the meta-analyses. (a) Dominant genetic model (DD+ID versus II): The overall odds ratio under the random-effect model was 1.74 (95 %CI: 1.29–2.33), and between-study heterogeneity was significant (I^2=72.6 %). (b) Recessive genetic model (DD versus ID+II): The pooled odds ratio under the random-effect model (I^2=78.7 %) was 2.31 (95 %CI: 1.61–3.31). (c) Allelic model (D versus I): The pooled odds ratio under the random effect model (I^2=86.6 %) was 1.77 (95 %CI: 1.34–2.33).

A separate analysis confined to cases of preeclampsia was carried out. Significant associations were found under recessive genetic model (OR=1.81, 95 %CI: 1.34–2.44) and allelic model (OR=1.31, 95 %CI: 1.09–1.57), but not dominant genetic model (OR=1.23, 95 % CI: 0.94–1.60). Between-study heterogeneity and publication bias were not found in the above models.

Cumulative meta-analysis and sensitivity analysis

In cumulative meta-analyses, the pooled odds ratio showed stable following the year 2004 for both genotypic models and the allelic model, and the associations reached significance with the accumulation of data over time (figures not shown). Sensitivity analysis showed none of the single

Table 2 Characteristics of included women for *ACE I/D* and PIH

Author	Age (years)		Gestational weeks (weeks)		Genotype distributions	
	Cases	Controls	Cases	Controls	Cases(DD/ID/II)	Controls(DD/ID/II)
Zhu [18]	26–32	26–32	N	N	23/7/15	21/10/13
Zhou [19]	24–32	24–32	N	N	39/12/9	8/32/38
Wang [20]	27.5±3.7	27.8±8.9	38.8±4.6	39.5±2.7	15/31/15	13/24/33
Hong [21]	23–32	23–32	N	N	34/10/8	16/34/50
Bai [22]	28±4	28±3	32–42	36–42	8/38/35	31/83/85
Gao [23]	26.7±3.2	27.4±2.8	38.2±2.8	39.4±1.2	66/34/10	57/18/6
Wu [24]	25±5	25±4	36–40	36–40	21/23/12	12/22/28
Mao [25]	27.8±3.4	26.8±1.8	38.4±2.3	39.2±1.4	25/24/13	12/46/62
Wang [26]	26.3	25.7	38.4±2.9	39.2±1.9	14/43/43	14/46/40
Wang# [27]	26±4	25±6	37.2±2.0	36.4±3.4	13/18/7	9/19/22
Li [11]	M	M	38.6±2.6	39.3±1.4	34/34/35	14/33/29
Chen [28]	M	M	M	M	41/31/20	16/34/35
Li [29]	26.8±5.4	26.8±5.4	35.8±3.6	N	24/14/16	19/39/42
Li [12]	29	28	35.7	38.6	37/46/50	25/31/49
Song [13]	30.5±3.7	28.7±3.2	35.3±3.6	38.9±1.2	17/21/7	13/23/9
Huang [30]	29.4±5.4	28.7±4.4	35.3±3.2	37.6±2.4	46/32/12	21/47/52
Ren [31]	25±5	25±5	36±3	39±3	27/20/13	13/17/24
Zhan [14]	N	N	N	N	32/41/47	10/24/26
Cui [15]	N	29.64	N	N	11/33/19	4/19/17
Deng [16]	30.8±5.2	29.6±3.3	N	N	20/16/14	20/57/23
Yue [17]	M	M	N	N	12/8/23	6/14/24
Yan [32]	30.41±5.4	30.14±5.0	37.6±2.2	38.9±1.4	21/57/35	23/52/38

N: Not mentioned #: To distinguish the two studies with the same last name of first author. M: Cases and controls were matched

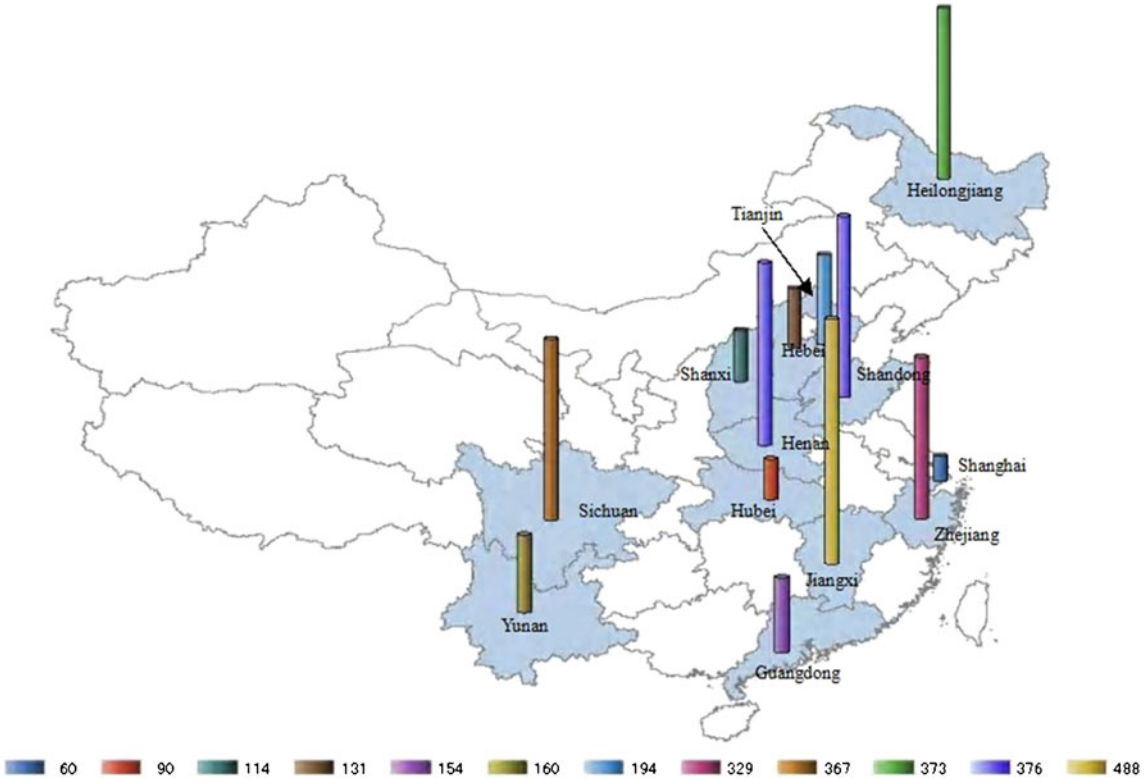

Fig. 1 Distributions of the sample source in the studies of association between *ACE I/D* and PIH. The study subjects were selected from 13 provinces and study size was marked by cylinder of different colors and heights, the number of participants from each province was shown below the map

studies influenced the overall odds ratio significantly (figures not shown).

Meta-regression and subgroup analysis

In meta-regression analysis, only the sample size of the case group was identified as a significant source of heterogeneity (P_{Het}=0.18, 0.02, and 0.09 under the dominant genetic model, recessive genetic model and allelic model, respectively). Stratified analyses were conducted by dividing the studies into three groups according to the sample size of case group (cases<50, 50<=cases<100 and cases>=100). The summarized results were presented in Table 3. The results revealed that the pooled odds ratio decreased as the study size of case group increased under both genotypic models and allelic model. No significant association was found between *ACE I/D* and PIH in the subgroup with more than 100 cases.

Publication bias

There was no publication bias since funnel plots didn't show asymmetric distribution and Egger's test was statistical insignificance (P=0.13, 0.19, and 0.22 under the dominant genetic model, recessive genetic model and allelic model, respectively).

AGT M235T

Regarding *AGT M235T* and PIH, the literature search resulted in two English papers and 11 Chinese papers, among which seven articles met the selection criteria. Six studies were excluded, two due to duplicated publications, one for inappropriate control group, one for incorrect data, and two for studying the other polymorphisms (*T174M*, *6G-A*) within AGT. In the seven studies, two were focusing on preeclampsia [13, 33], and the other five were on pregnancy induced hypertension without detailed genotype data for gestational hypertension, preeclampsia and eclampsia respectively. Studies were re-calculated by the first and second author to examine HWE in control groups. The characteristics of the seven studies with 551 cases and 745 controls were shown in Tables 4 and 5, and the distributions of the participants from seven provinces in China were shown in Fig. 3.

Genetic model statistic analysis

Figure 4 showed the forest plots of meta-analyses, (d) Dominant genetic model (TT+MT versus MM): the pooled odds ratio under the fixed-effects model was 1.59 (95 %CI:

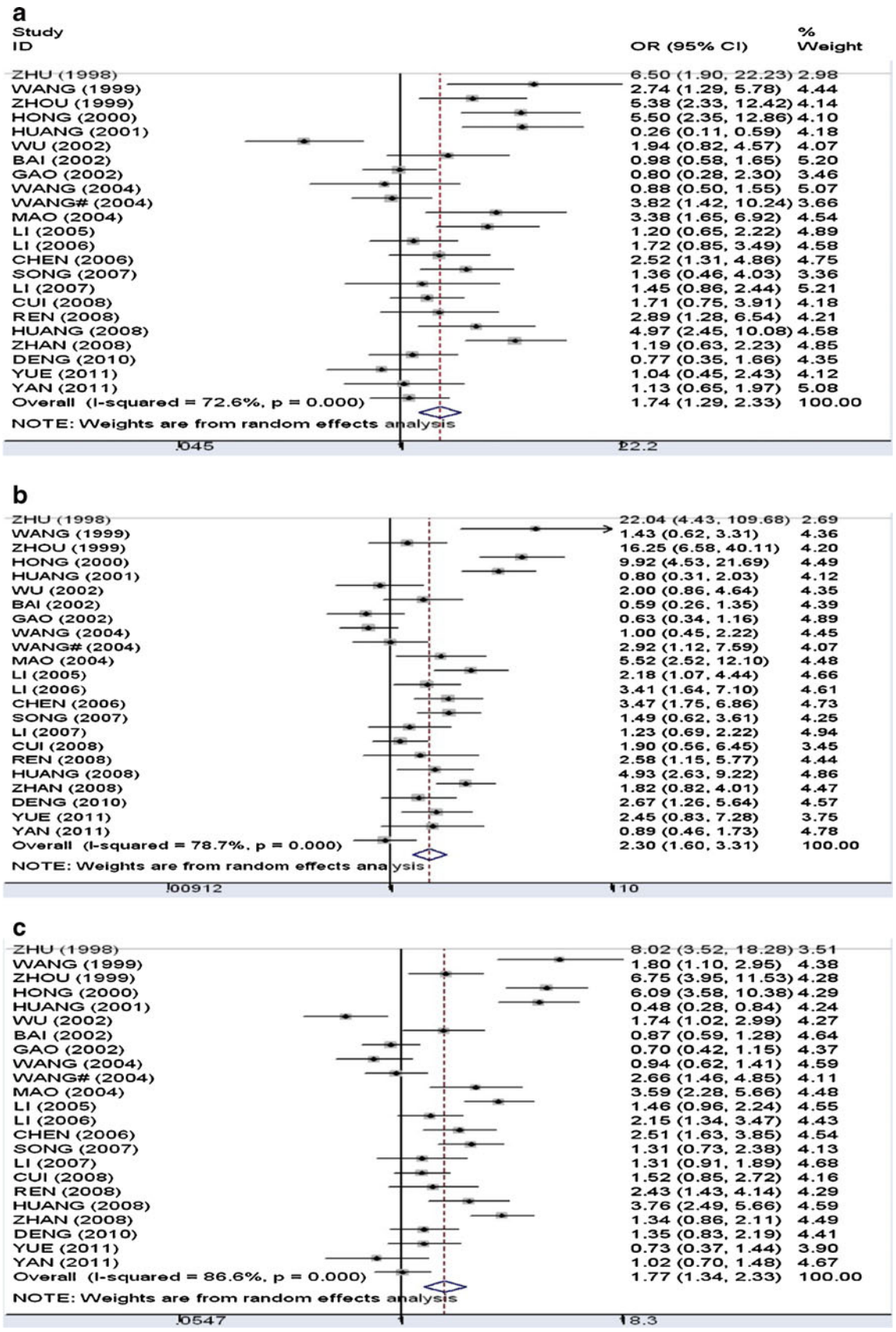

Fig. 2 Forest plots of meta-analysis of the association between *ACE I/D* and PIH. **a** Dominant genetic model (the random effect model); **b** Recessive genetic model (the random effect model); **c** Allelic model (the random effect model)

Table 3 Subgroup analyses results of different study size of case group for *ACE I/D* with PIH

Models	OR, 95 %CI (cases<50) *n*=4	OR, 95 %CI (50<=cases<100) *n*=13	OR, 95 %CI (cases>=100) *n*=6
Dominant genetic model	2.30 (1.00–5.30)	2.04 (1.29–3.21)	1.14 (0.88–1.46)
Recessive genetic model	3.30 (1.32–8.26)	2.93 (1.82–5.26)	1.14 (0.87–1.50)
Allelic model	2.01 (0.84–7.56)	2.14 (1.45–3.16)	1.12 (0.94–1.32)

*n denoted the number of studies in each group

1.04–2.42). The test for heterogeneity was not significant (I^2=0); (e) Recessive genetic model (TT versus MT+MM): the pooled odds ratio under the random-effects model was 1.60 (95 %CI: 1.07–2.40). The overall data indicated significant heterogeneity (I^2=62 %); (f) Allelic model (T versus M): the pooled odds ratio was 1.40 (95 %CI: 1.17–1.68) under the fixed-effects model (I^2=46 %). Due to the limited studies of *AGT M235T* and preeclampsia, we failed to conduct a separate analysis in this regard.

Cumulative meta-analysis and sensitivity analysis

In cumulative meta-analyses, the pooled ORs for both genotypic models and the allelic model were stable following the year 2005 (figures not shown). Sensitivity analysis and meta-regression were not performed since less than ten studies were included in the current analysis.

Publication bias

Asymmetric distribution was not found in funnel plots, and Egger's test showed statistical insignificance under the dominant genetic model, recessive genetic model and allelic model (*P*=0.68, 0.34, and 0.31, respectively) (figures not shown).

Discussion

Many studies investigating the etiology and pathogenesis of PIH suggested an inherited susceptibility. Renin, AGT and ACE are three important components in RAS, each playing a critical role in the regulation of blood pressure and electrolyte balance. Renin enzymatically cleaves AGT to angiotensin I, which is the rate-limiting step of the RAS cascade. However, very few studies have been carried out to investigate the relationship for gene polymorphisms of renin with PIH in Chinese populations so that we failed to conduct a meta-analysis in this aspect. *ACE I/D* and *AGT M235T* are the most frequently studied ones of RAS associated with PIH. However, studies had inconsistent conclusions, which suggested a meta-analysis necessary to assess the genetic effects of *ACE I/D* and *AGT M235T* on PIH.

Previous meta-analyses in this regard were mainly based on mixed populations. Studies showed that the distribution of gene polymorphisms differed among different ethnic populations. The result of a meta-analysis is likely to be affected by the population sources of study subjects, and thus the conclusions drawn from studies based on other populations may not be appropriate for a specific population [38]. It is necessary to conduct a meta-analysis to evaluate the associations between *ACE I/D*, *AGT M235T*, and PIH among Chinese populations.

Significant associations were found in meta-analysis of *ACE I/D* and PIH under the genotypic models and allelic model in the present study. A separate analysis for *ACEI/D* with preeclampsia was carried out, and significant associations were found under recessive genetic and allelic models, but not dominant genetic model. Further, meta-regression analyses revealed that sample size of case group was a

Table 4 Characteristics of the included studies for *AGT M23T* and PIH

Author	Year	Location	Enrollment	Cases	Controls	Parity	Comments
Bai [22]	2002	Sichuan	1998.8–2000.5	81	205	N	Han ethnic, no family history, excluded other pathologies
Wu [34]	2002	Shandong	2000.2–2001.5	60	40	Primiparity	Singleton, excluded other pathologies
Fu [35]	2003	Jiangxi	2001.9–2002.12	100	102	N	Excluded other pathologies
Hu [36]	2005	Heilongjiang	1998–2003	93	140	N	N
Huang [33]	2007	Yunan	2004.7–2005.12	58	102	primiparity	Singleton, excluded other pathologies
Song [13]	2007	Hubei	2004.7–2005.2	45	45	N	Excluded other pathologies
Xiang [37]	2011	Henan	2008.1–2009.12	113	113	M	Han ethnic, excluded other pathologies

N: Not mentioned. M: Cases and controls were matched

Table 5 Characteristics of the included women for *AGT M23T* and PIH

Author	Age (years)		Gestational weeks (weeks)		Genotype distribution	
	Cases	Controls	Cases	Controls	Cases (TT/MT/MM)	Controls (TT/MT/MM)
Bai [22]	28±3	28±4	32–42	36–42	58/21/1	148/41/6
Wu [34]	27.9±3.4	28.1±3.2	36.7±4.5	37.2±4.2	34/22/4	12/23/5
Fu [35]	26.3±2.1	25.7±2.5	N	N	53/42/7	30/57/13
Hu [36]	M	M	M	M	69/22/2	81/56/3
Huang [33]	29±4	29±3	28–36	28–36	14/31/13	25/50/27
Song [13]	30.5±3.7	28.7±3.2	35.32±3.6	38.96±1.2	15/23/7	7/25/13
Xiang [37]	30.41±5.4	30.14±5.0	37.56±2.2	38.90±1.3	59/48/6	63/43/7

N: Not mentioned. M: Cases and controls were matched

significant source of heterogeneity in current analysis. A stratified analysis showed a diminished effect as the number of cases increased, and no significant association was found in the subgroup with large case numbers either under genotypic models or allelic model. Previous meta-analysis based on mixed populations concluded that *ACE I/D* was associated with PIH only under the recessive genetic model, which was inconsistent with the current findings. The finding from current study demonstrated the conclusion drawn by Norma C. Serrano and his colleagues, which indicated the small increased risk of preeclampsia associated with ACE-D allele might be due to the small study bias [39]. Small sample sized studies were more likely to exaggerate the genetic effect, which was actually small [40]. Most studies conducted in Asian populations with the number of cases less than 100 tended to have larger ORs, and Chinese studies typically suggested even a stronger genetic effect than non-Chinese studies [41].

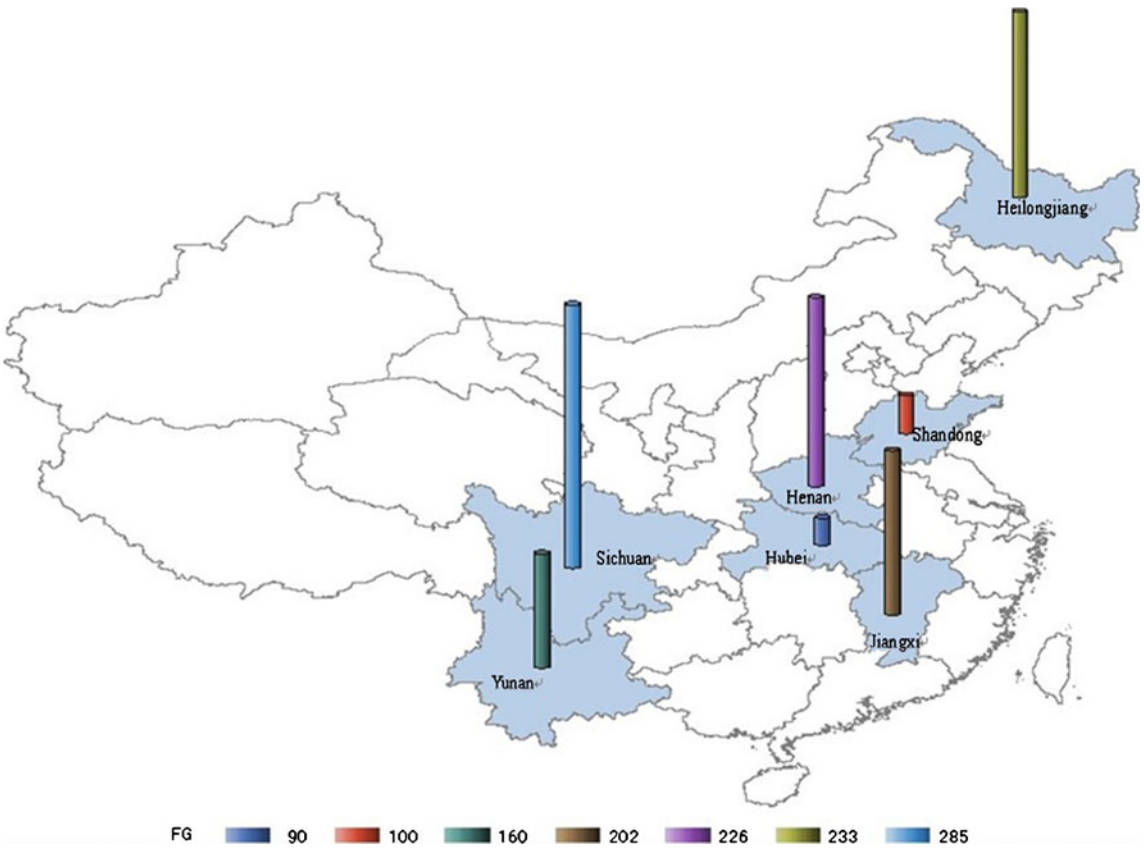

Fig. 3 Distributions of the sample source in the studies of the association between *AGT M235T* and PIH. The participants were selected from seven provinces and study size was marked by cylinders of different colors and heights, and the number of participants from each province was shown below the map

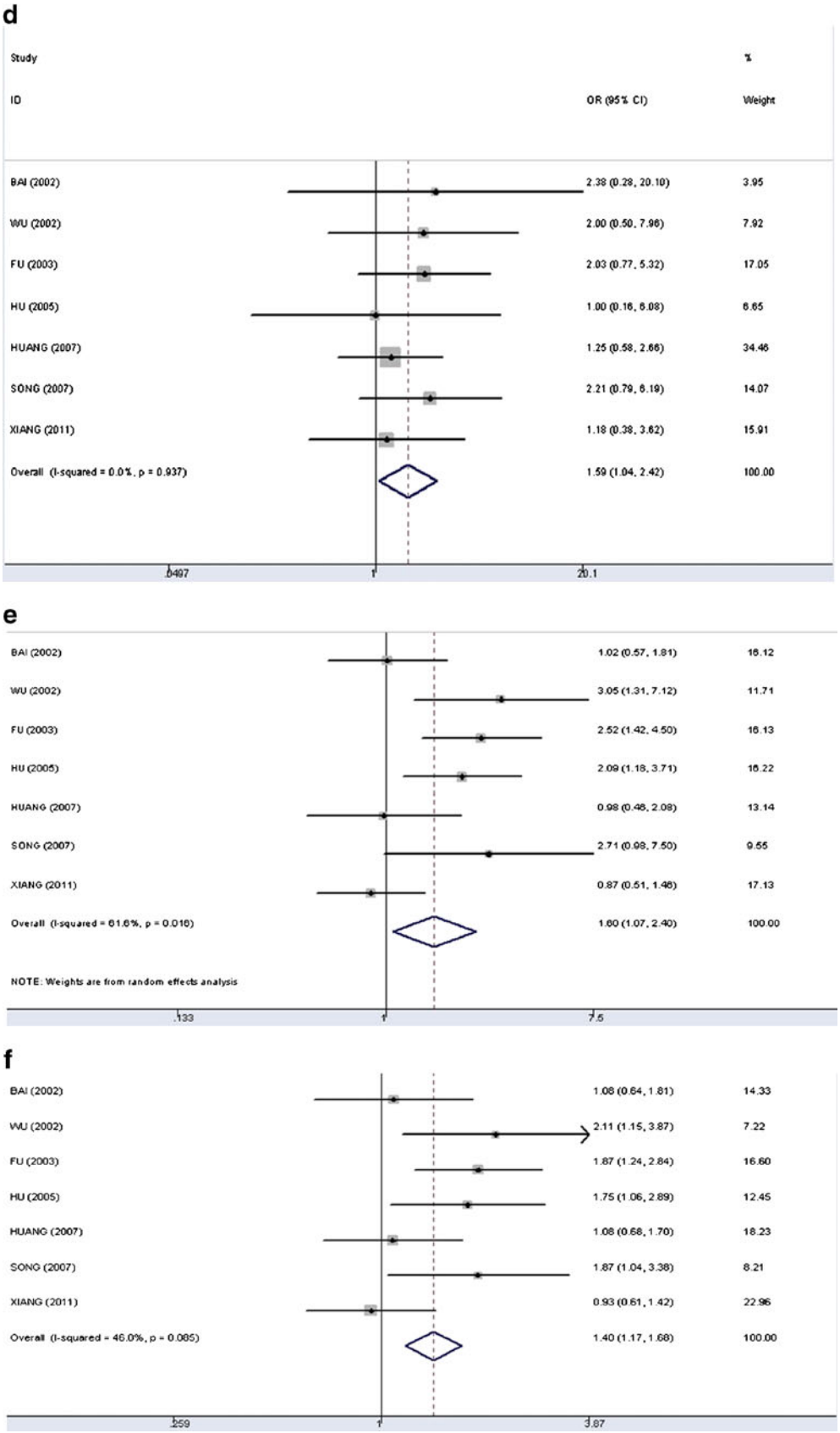

Fig. 4 Forest plots of meta-analysis of the association between *AGT M235T* and PIH. **d** Dominant genetic model (the fixed-effect model); **e** Recessive genetic model (the random-effect model); **f** Allelic model (the fixed-effect model)

Significant heterogeneity was presented in the current meta-analysis of association between *ACE I/D* and PIH, which several reasons may account for: (1) Disease severity may serve as a confounder in the studies of the association between gene polymorphisms and PIH, and recent studies reported that a stronger genetic effect was found in severe cases of PIH in comparison with mild cases [42]. The effects of genetic and environmental varied in the development of preeclampsia and gestational hypertension. Studies suggested that the two diseases should be analyzed separately for their different gene markers [43]. However, most studies carried out in China on associations between gene polymorphisms and PIH did not differentiate preeclampsia and gestational hypertension in analysis. (2) Environmental factors were not taken into analysis although the occurrence of PIH is the result of the interaction of genetic and environmental factors. In the present meta-analysis, the distribution of study subjects ranged from northeast to southwest in China. Environmental factors, living habits, and economic situations varied a lot among those populations, which could influence the evaluation of the associations. (3) Early-onset preeclampsia and late-onset preeclampsia, which were found to be associated with different biomarkers, genetic and environmental risk factors, should also be considered for interpreting the source of heterogeneity [44, 45].

Significant associations of *AGT M235T* with PIH were found under both the dominant genetic model and recessive genetic model, individuals with allele T would be at a higher risk of developing PIH in the present meta-analysis, which was consistent with the previous meta-analysis conducted by Medica I and his colleagues, based on mixed populations. A meta-regression analysis with 1,446 cases and 3,829 controls in 2008 revealed that individuals carrying TT were more likely to develop preeclampsia or eclampsia compared to those with MM in Caucasians, but not in East Asian population. It might be due to this study having very limited numbers of studies of East Asian population [46]. A large scale study with 1,068 Caucasian women concluded that *AGT M235T* might contribute to the pathogenesis of gestational hypertension and preeclampsia [47]. However, a literature review by Elke Knyrim in 2008 [48] reported that most of the studies, including a British multi-centre GOPEC study [49], did not support this association. The current results from studies of Chinese populations should be viewed cautiously and need to be further investigated with a larger sample size in the future.

The current study was the first meta-analysis studying the associations between *ACE I/D*, *AGT M235T* and PIH specifically on a Chinese population. Although no evidence of publication bias was found in the current analysis, conclusions should be drawn cautiously. The sample size in the current study was relatively small. We have over 90 % power to detect an increased odds ratio of 0.3 for *ACE I/D* (MAF: D=0.399) and over 80 % power to detect the odds ratio of 1.40 for *AGT M235T* in the present study. Studies with a small sample size usually lack sufficient power to detect the real associations [50], and evidence also showed that some significant gene-disease associations revealed by meta-analysis based on small sample sized studies had been refuted by later large-scale studies [51]. Thus, it is necessary to conduct large-scale researches in the future study of PIH. Furthermore, recent studies supported that uteroplacental renin-angiotensin system also plays an important role in the development of PIH, and that RAS related gene polymorphisms of fetuses as well as fetal-maternal genotype incompatibility might be one of the potential mechanisms of pregnancy complications [52–54]. It is suggested that fetal genotypes should also be examined in the future genetic studies of PIH, besides maternal genotypes.

The current meta-analysis suggested significant associations between *ACE I/D, AGT M235T*, and PIH in Chinese populations. However, the results of stratified analysis of *ACE I/D* and PIH suggested that the apparent significance of the result may be due to small sample size. It is necessary to conduct large sample sized studies in the future; in addition, the genetic polymorphisms in both mother and fetus need to be investigated together.

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Conflict of interest The authors declare that they have no conflict of interest.

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