Association of angiotensin-converting enzyme gene I/D polymorphism with chronic obstructive pulmonary disease: a meta-analysis

Guodong Xu*, Guohui Fan*, Yingtong Sun, Lili Yu, Sinan Wu and Wenquan Niu (iD)

Abstract

Objectives: We conducted a meta-analysis of published studies on the angiotensin-converting enzyme (*ACE*) gene insertion/deletion (I/D) polymorphism associated with the risk of chronic obstructive pulmonary disease, as well as with pulmonary function and circulating angiotensin-converting enzyme changes.

Methods: A literature search, quality assessment and data extraction were completed independently and in duplicate.

Results: A total of 16 articles were meta-analysed, including 12 articles (2113 patients and 8786 controls) for chronic obstructive pulmonary disease risk and eight articles (11,664 subjects) for pulmonary and circulating phenotypes. In overall and subgroup analyses, no significance was noted between the I/D polymorphism and chronic obstructive pulmonary disease risk under all genetic models ($P>0.05$), without heterogeneity or publication bias. Carriers of II, ID and II plus ID genotypes had significantly lower levels of circulating angiotensin-converting enzyme than those with the DD genotype (weighted mean difference -13.35, -8.13 and -10.74 U/L, respectively, $P<0.001$). For forced expiratory volume in one second (FEV_1)/forced vital capacity, carriers of the DD genotype had marginally lower levels than those with the DD genotype (weighted mean difference -1.66, $P=0.034$). Furthermore in the case of FEV_1 of 50% or greater of predicted FEV_1, FEV_1 was marginally lower in ID genotype carriers than DD genotype carriers (weighted mean difference -3.50, $P=0.056$).

Conclusions: Our meta-analytical findings demonstrated that the *ACE* gene I/D polymorphism was not associated with the risk of chronic obstructive pulmonary disease.

Introduction

Chronic obstructive pulmonary disease (COPD) is characterised by persistent respiratory symptoms and airflow limitation, and it has ranked as the third killer in the world as per the latest statistics.[1,2] Although several modifiable risk factors such as cigarette smoking and exposure to ambient air pollution have been identified, there is substantial evidence for the contribution of a heritable component to COPD susceptibility. For instance, a twin study estimated that inherited genetic susceptibility contributed to 60% of COPD risk.[3] Genome-wide association studies have succeeded in showing convincingly that COPD is a multifactorial polygenic disease.[4,5] Despite tremendous advances gained in heritable insights, it still remains a challenge to unlock the genetic basis of COPD, because of apparent inconsistency from underpowered individual studies.

Institute of Clinical Medical Sciences, China-Japan Friendship Hospital, China

* GX and GF share first authorship.

Corresponding author:
Wenquan Niu, Institute of Clinical Medical Sciences, China-Japan Friendship Hospital, No. 2 Yinghuayuan East Street, Chaoyang District, Beijing 100029, China.
Email: niuwenquan_shcn@163.com

It is widely recognised that the gene encoding angiotensin-converting enzyme (*ACE*) is a COPD susceptibility candidate.[6–9] Low ACE activity was identified to play a beneficial role in the development of COPD. The genomic sequence of the *ACE* gene is polymorphic, and thereof an insertion/deletion (I/D) polymorphism (rs4646994) in intron 16 is found to be highly associated with the circulating ACE level.[10] In particular, carriers of the DD genotype of this polymorphism had higher cellular and circulating levels of ACE, as well as power/sprint performance related to those with the II genotype.[11] It is thus reasonable to hypothesise that the *ACE* gene I/D polymorphism is a promising candidate locus in susceptibility to COPD. In fact, the association of the *ACE* gene I/D polymorphism with COPD risk has been extensively studied in the medical literature, while the findings are inconsistent and inconclusive.[6–9] A comprehensive evaluation of this inconsistency is obviously needed for such a conclusion to be drawn more firmly. To test this hypothesis and yield more information, we conducted a systematic meta-analysis of published data on the *ACE* gene I/D polymorphism in association with COPD. In addition, we attempted to interrogate the association of this polymorphism with pulmonary function and circulating ACE changes.

Methods

The conduct of this meta-analysis conformed to the guidelines of the preferred reporting items for systematic reviews and meta-analyses (PRISMA) statement.[12] The PRISMA checklist and flow diagram are presented in Supplementary Table 1 and Supplementary Figure 1, respectively.

Search strategy

We searched major online public databases including Medline/PubMed, EMBASE (Excerpta Medica database) and Web of Science for studies published before 25 August 2017. We used heading search strategy with the terms: ('ACE' or 'angiotensin-converting enzyme') and ('COPD' or 'chronic obstructive pulmonary disease') and ('polymorphism' or 'SNP' or 'variant' or 'mutation'). We also checked the reference lists of identified reports for the other potentially relevant studies. The search strategy was completed by two authors (Guodong Xu and Guohui Fan). A final reference list was determined after combining search results and removing duplicated hits.

Selection criteria

Articles were included if the genotype or allele counts of the *ACE* gene I/D polymorphism were provided in both COPD patients and healthy controls, and if COPD was diagnosed by a standard method, and if the I/D polymorphism was genotyped by a validated method. Only articles published in the English language were included in this meta-analysis. An article was excluded if it was published in forms of conference abstract, letter to the editor, case reports or case series, or if it lacked the control group.

As per the selection criteria formulated above, two authors (Guodong Xu and Guohui Fan) independently assessed the eligibility of each potential article for inclusion, and the results were compared and disagreement was solved by consensus.

Data extraction

The following data were extracted into a uniform design table from each eligible study: the first author's surname, year of publication, country where the study was conducted, race of the study population, study design, diagnosis of COPD, source of controls, age, gender, body mass index, forced expiratory volume in one second (FEV_1), FEV_1/FVC (forced vital capacity), smoking status, pack years of smoking, the number of subjects with different genotypes of the *ACE* gene I/D polymorphism in COPD patients and controls. Data abstracted by the two authors were checked for coherence, and any divergence was resolved by resorting to original context until a consensus was reached.

Statistical analysis

Statistical analyses were conducted using STATA/SE software (version 11.2 for Windows; Stata Corp, College Station, TX, USA).

The primary outcome was the weighted odds ratio (OR) with 95% confidence interval (CI) for different genotypes and alleles of the *ACE* gene I/D polymorphism in patients with COPD compared with controls. The second outcome of this meta-analysis was the discrepancies of FEV_1 and circulating ACE between different genotypes, as expressed as weighted mean difference (WMD) with 95% CI. The summary OR and WMD were calculated with a fixed effects model. Heterogeneity was tested using the inconsistency index (I^2) statistic (ranging from 0.0% to 100.0%), which is defined as the percentage of observed between-study variability that is due to heterogeneity rather than chance. If the I^2 exceeds 50.0%, it indicates statistically significant heterogeneity.

Funnel plots were used to assess the probability of publication bias and the small study effect. If funnel shape was asymmetric inverted, it might suggest an association between the pooled estimate and study size (publication bias or small study bias). The Egger's test was used to assess funnel asymmetry objectively.

Univariate random-effects meta-regressions with log-transformed ORs were used to assess potential confounding of some risk factors, including age, gender and proportion of smokers.

Results

Qualifying studies

Our initial search of three public databases identified 42 articles published in the English language, and only 16 of them were eligible for analysis.[6–9, 13–24] There were 12 articles (12 studies including 2113 patients and 8786 controls) that incorporated data on the risk of the *ACE* gene I/D polymorphism for COPD, and eight articles (14 studies including 11,664 subjects) that incorporated data on the changes of either FEV_1 or FEV_1/FEV or circulating ACE across the genotypes of this polymorphism.

Baseline characteristics

The baseline characteristics of eligible studies are presented in both Table 1 and Supplementary Table 2.

I/D polymorphism and COPD risk: overall analysis

The association of the *ACE* gene I/D polymorphism with COPD risk was examined under the allelic, homozygous genotypic and dominant models, respectively. Figure 1 shows the corresponding forest plots. Overall, no hint of statistical significance was noted between this polymorphism and COPD risk under all genetic models ($P>0.05$). There was no evidence of heterogeneity between studies ($I^2=0.0\%$, 0.0% and 10.4%, respectively). In addition, there was a low likelihood of publication bias (Egger's test $P=0.811$, 0.994 and 0.894, respectively). The Begg's funnel plot in Figure 2 seemed symmetrical, and no study was reported to be missing.

I/D polymorphism and COPD risk: subgroup analysis

Table 2 shows the subgroup analysis of the *ACE* gene I/D polymorphism in the prediction of COPD risk under the allelic, homozygous genotypic and dominant models, respectively. Factors under stratification included continent, race, study design, matched condition, COPD diagnosis, control source and sample size. Still, there was no detectable significance across all subgroups under all genetic models ($P>0.05$), and there was low evidence of heterogeneity for a majority of comparisons ($I^2<50\%$).

I/D polymorphism and COPD risk: meta-regression analysis

Other sources of heterogeneity were explored through meta-regression analysis, including age, gender, smoking and pack years of smoking, and no factors exhibited a significant contribution ($P>0.05$).

I/D polymorphism and phenotypes: overall analysis

Table 3 shows the changes in FEV_1, FEV_1/FVC and circulating ACE across genotypes of the *ACE* gene I/D polymorphism. Carriers of the II genotype, ID genotype and the combination of II and ID genotypes had significantly lower levels of circulating ACE than those with the DD genotype (WMD −13.35, −8.13 and −10.74 U/L, respectively, all $P<0.001$). For FEV_1/FVC, carriers of the ID genotype had a marginally lower level than those with the DD genotype (WMD −1.66, $P=0.034$). There was no evidence of heterogeneity and publication bias, as reflected by the I^2 and Egger's test, respectively.

I/D polymorphism and phenotypes: subgroup analysis

Shown in Table 4 is the subgroup analysis of relevant phenotypes across genotypes of the *ACE* gene I/D polymorphism under various genetic models. According to the degree of FEV_1/predicted FEV_1 in DD genotype carriers, no significant association was found in COPD patients and controls, respectively. When FEV_1 of 50% or greater predicted FEV_1, FEV_1 was marginally lower in the ID genotype carriers than the DD genotype carriers (WMD −1.73, $P=0.056$).

In the case of circulating ACE, changes were consistently significant in both COPD patients and controls, and it was more obvious for the comparison of the II genotype with the DD genotype (WMD −14.03 U/L, $P<0.001$ in patients and WMD −13.12, $P<0.001$ in controls).

Discussion

This meta-analysis was designed to examine the association of the *ACE* gene I/D polymorphism with COPD risk. After a comprehensive analysis of 16 articles, our findings demonstrated that the *ACE* gene I/D polymorphism was not associated with the risk of COPD. In addition, we observed that the ID genotype carriers of this polymorphism tended to have reduced FEV_1 (% predicted FEV_1) in COPD patients with GOLD stages I–II. To our knowledge, this is the first meta-analytical report that has evaluated the association of the *ACE* gene I/D polymorphism with pulmonary function.

COPD is a complex multifactorial disease, and its development is thought to be largely under genetic control. A considerable number of genes and polymorphisms have been identified as susceptible candidates for COPD risk.[25–27] The gene encoding ACE is one such candidate. ACE is a key element of the renin–angiotensin–aldosterone system, and its plasma levels are modulated by common defects of the gene.[28] In particular, the *ACE* gene I/D polymorphism has received great attention, as the D allele of this

Table 1. Baseline characteristics of eligible studies for the association of *ACE* gene I/D polymorphism with COPD risk.

First author	Year	Country	Race	Design	Match	Diagnosis	Source	Sample size		Age (years)		Gender (male)	
								Cases	Controls	Cases	Controls	Cases	Controls
Mlak et al.	2016	Poland	Caucasian	Cross-sectional	n.a.	GOLD	Population	206	165	63.0	64.0	0.6796	0.6303
Ayada et al.	2014	Turkey	Turkish	Cross-sectional	Yes	GOLD	Hospital	47	64	n.a.	n.a.	n.a.	n.a.
Ulasli et al.	2013	Turkey	Caucasian	Cross-sectional	n.a.	GOLD	Hospital	50	49	64.0	54.6	0.9400	0.7959
Simsek et al.	2013	Turkey	Turkish	Cross-sectional	n.a.	GOLD	Hospital	66	40	61.2	59.7	0.6364	0.7250
Kuzubova et al.	2013	Russia	Caucasian	Cross-sectional	n.a.	GOLD	Population	63	95	60.4	57.3	1.0000	1.0000
Pabst et al.	2009	Germany	Caucasian	Cross-sectional	n.a.	ATS/ERS	Population	152	158	62.8	63.9	0.6842	0.3924
Lee et al.	2009	Denmark	Caucasian	Prospective	n.a.	GOLD	Population	1259	7775	n.a.	n.a.	0.5060	0.4319
Zhang et al.	2008	China	Chinese	Cross-sectional	Yes	ATS/ERS	Hospital	61	57	63.0	61.6	0.8361	0.7719
Busquets et al.	2007	Spain	Caucasian	Cross-sectional	n.a.	ATS/ERS	Population	74	159	74.0	56.0	1.0000	1.0000
Tkacova et al.	2005	Slovakia	Caucasian	Cross-sectional	n.a.	GOLD	Hospital	66	118	65.4	63.5	0.7424	0.6780
Ahsan et al.	2004	India	Indian	Cross-sectional	Yes	ATS/ERS	Hospital	27	66	n.a.	n.a.	1.0000	1.0000
Yildiz et al.	2003	Turkey	Caucasian	Cross-sectional	Yes	ATS/ERS	Hospital	42	40	62.0	60.0	0.9762	1.0000

BMI (kg/m²)		Smoking (%)		Smoking (pack years)		FEV₁ (% predicted)		FEV₁/FVC (%)		Cases			Controls		
Cases	Controls	Cases	Controls	Cases	Controls	Cases	Controls	Cases	Controls	DD	ID	II	DD	ID	II
27.0	28.4	0.9126	0.9333	41.50	43.30	45.40	n.a.	48.30	n.a.	60	99	47	51	73	41
n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	13	26	8	28	28	8
25.8	28.9	n.a.	0.0000	57.20	n.a.	45.10	100.00	44.20	79.00	28	11	11	33	10	6
n.a.	n.a.	n.a.	0.0000	55.12	n.a.	36.20	8.50	62.80	82.44	31	20	15	13	19	8
n.a.	n.a.	1.0000	0.5700	32.80	n.a.	45.00	n.a.	n.a.	n.a.	17	29	17	22	49	24
n.a.	n.a.	0.4803	0.3354	31.10	18.70	52.60	81.50	58.60	96.80	33	76	43	50	69	39
n.a.	n.a.	0.8682	0.7307	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	336	609	314	2038	3876	1861
23.9	25.1	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	13	27	21	9	28	20
25.0	27.0	1.0000	n.a.	49.00	n.a.	44.00	n.a.	44.00	n.a.	27	40	7	53	79	27
n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	20	31	15	31	68	19
n.a.	n.a.	1.0000	1.0000	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	5	12	10	10	33	23
n.a.	n.a.	0.2381	0.3000	34.00	29.00	42.00	101.00	55.00	86.00	14	21	7	12	18	10

ACE: angiotensin-converting enzyme; BMI: body mass index; FEV₁: forced expiratory volume in one second; FVC: forced vital capacity; DD: deletion/deletion genotype; ID: insertion/deletion genotype; II: insertion/ insertion genotype; n.a.: not available.
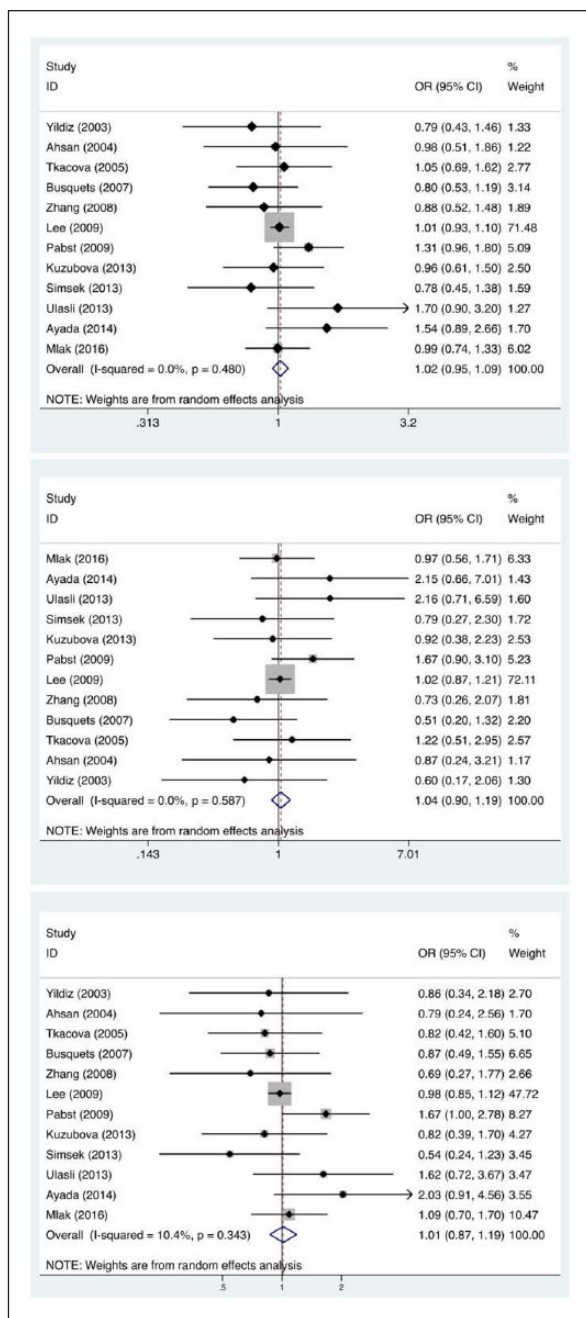
Figure 1. Forest plots of the angiotensin-converting enzyme (*ACE*) gene insertion/deletion (I/D) polymorphism in association with chronic obstructive pulmonary disease under allelic (the upper), homozygous genotypic (the middle) and dominant (the lower) models.

Figure 2. Begg's and filled funnel plots of the angiotensin-converting enzyme (*ACE*) gene insertion/deletion (I/D) polymorphism in association with chronic obstruction pulmonary disease under the allelic model.

polymorphism correlated with a higher plasma ACE activity, the finding being further validated by our current analysis on different genetic models and diverse disease conditions. It has recently been increasingly recognised that the *ACE* gene I allele, interacting with the endothelial nitric oxide synthase (*eNOS*) gene 894G allele, can cause less vasoconstriction and increase vasodilation that may be
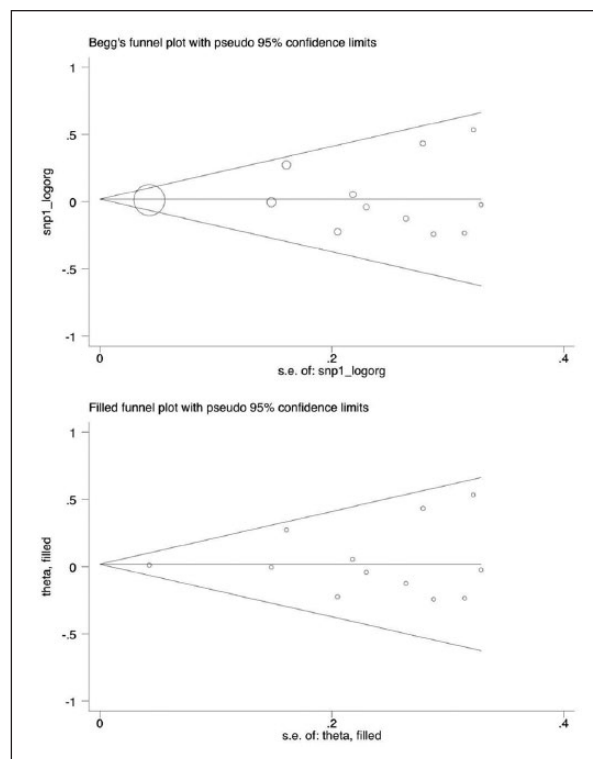
advantageous in the improvement of COPD.[14] However, in the present meta-analysis, although the *ACE* gene I allele was associated with a slightly increased risk of COPD, there was no detectable significance under different genetic models of inheritance, even in various subgroups by many study characteristics. This association was robust and solid, as this meta-analysis is currently based on the largest number of eligible studies relative to previously published meta-analyses,[29–31] and importantly our findings remain unperturbed by between-study heterogeneity and publication bias. On the other hand, it is widely recognised that the increased risk attributable to a single allele is small in genetically susceptible individuals, and such a small effect may also be exacerbated by locus heterogeneity across different ethnic/racial groups.[6,8,18] Wide coverage of the genetic variability of the *ACE* gene and other COPD susceptibility genes is required to decipher the genetic basis of COPD.

In addition, our findings also dropped a hint of a probable contribution of the *ACE* gene I/D polymorphism to pulmonary function, as carriers of the ID genotype had a significantly lower level of FEV_1/FVC than carriers of the DD genotype. Moreover, the ID genotype was associated with reduced FEV_1 in COPD patients with GOLD stages I–II, while this reduction was non-significant in patients

Table 2. Subgroup analyses of ACE gene I/D polymorphisms in association with COPD.

Subgroups	Studies (cases/controls)	Allele model (I vs. D)					Homozygous genotype model (II vs. DD)					Dominant model (II + ID vs. DD)				
		OR	95% CI lower	95% CI upper	P value	I² (%)	OR	95% CI lower	95% CI upper	P value	I² (%)	OR	95% CI lower	95% CI upper	P value	I² (%)
Continent																
Asia	6 (293/316)	1.05	0.83	1.33	0.678	22.9	1.06	0.66	1.69	0.814	0.0	1.00	0.64	1.56	0.986	31.3
Europe	6 (1820/8470)	1.02	0.94	1.09	0.678	0.0	1.03	0.89	1.20	0.662	0.0	1.00	0.89	1.12	0.962	0.0
Race																
Caucasian	8 (1912/8559)	1.02	0.95	1.10	0.606	2.9	1.04	0.89	1.22	0.618	2.0	1.00	0.89	1.13	0.938	0.0
Chinese	1 (61/57)	0.98	0.51	1.86	0.631	n.a.	0.87	0.24	3.21	0.551	n.a.	0.79	0.24	2.56	0.443	n.a.
Indian	1 (27/66)	0.88	0.52	1.48	0.941	n.a.	0.73	0.26	2.07	0.834	n.a.	0.69	0.27	1.77	0.689	n.a.
Turkish	2 (113/104)	1.11	0.75	1.64	0.596	64.9	1.26	0.47	3.38	0.644	34.7	1.05	0.29	3.84	0.937	80.2
Study design																
Cross-sectional	11 (854/1011)	1.04	0.91	1.19	0.559	4.2	1.07	0.82	1.40	0.628	0.0	1.04	0.83	1.31	0.714	15.9
Nested	1 (1259/7775)	1.01	0.93	1.10	0.805	n.a.	1.02	0.87	1.21	0.785	n.a.	0.98	0.85	1.12	0.722	n.a.
Matched																
Yes	4 (177/227)	1.02	0.77	1.36	0.867	5.9	0.95	0.52	1.71	0.856	0.0	1.05	0.62	1.79	0.845	19.7
No	8 (1936/8559)	1.02	0.95	1.10	0.621	5.1	1.04	0.90	1.20	0.585	0.0	1.01	0.85	1.20	0.908	17.2
COPD diagnosis																
ATS/ERS	5 (356/480)	1.01	0.82	1.23	0.946	17.1	0.90	0.54	1.51	0.683	29.1	1.05	0.74	1.51	0.776	15.0
GOLD	7 (1757/8306)	1.02	0.95	1.10	0.595	0.0	1.04	0.90	1.21	0.582	0.0	1.00	0.82	1.22	0.974	17.8
Control source																
Hospital	7 (359/434)	1.05	0.86	1.29	0.632	7.5	1.09	0.72	1.65	0.675	0.0	0.97	0.67	1.39	0.855	21.0
Population	5 (1754/8352)	1.01	0.94	1.09	0.705	0.0	1.03	0.83	1.27	0.781	12.6	1.03	0.86	1.22	0.773	13.9
Sample size																
<158	6 (293/320)	1.05	0.83	1.33	0.678	22.9	1.06	0.66	1.69	0.814	0.0	1.00	0.64	1.56	0.986	31.3
≥158	6 (1820/8470)	1.02	0.94	1.09	0.678	0.0	1.04	0.90	1.19	0.662	0.0	1.00	0.89	1.12	0.962	0.0

ACE: angiotensin-converting enzyme; COPD: chronic obstructive pulmonary disease; OR: odds ratio; 95% CI: 95% confidence interval; I^2: inconsistency index; I: insertion allele; D: deletion allele; DD: deletion/deletion genotype; ID: insertion/deletion; II: insertion/insertion genotype; n.a.: not available.

Table 3. Mean changes of FEV$_1$, FEV$_1$/FVC and circualting ACE between carriers of different genotypes of *ACE* gene I/D polymorphism.

Comparison	Studies	WMD	95% CI lower	95% CI upper	*P* value	I^2	Egger's *P* value
II vs. DD							
FEV$_1$	7	0.03	−0.77	0.83	0.937	0.0%	0.974
FEV$_1$/FVC	2	0.04	−2.43	2.50	0.978	0.0%	n.a.
Circulating ACE	5	−13.35	−14.58	−12.12	<0.001	0.0%	0.272
ID vs. DD							
FEV$_1$	7	0.01	−0.65	0.68	0.971	0.0%	0.641
FEV$_1$/FVC	2	−1.66	−3.19	−0.13	0.034	0.0%	n.a.
Circulating ACE	5	−8.13	−10.16	−6.09	<0.001	24.5%	0.182
II + ID vs. DD							
FEV$_1$	7	−0.04	−0.59	0.66	0.911	0.0%	0.879
FEV$_1$/FVC	2	−0.74	−2.23	0.76	0.334	0.0%	n.a.
Circulating ACE	5	−10.74	−12.32	−9.15	<0.001	15.1%	0.215

ACE: angiotensin-converting enzyme; WMD: weighted mean difference; 95% CI: 95% confidence interval; I^2: inconsistency index; DD: deletion/deletion genotype; ID: insertion/deletion; II: insertion/insertion genotype; FEV$_1$: forced expiratory volume in one second; FVC: forced vital capacity; n.a.: not available.

Table 4. Subgroup analyses of FEV$_1$/predicted FEV$_1$ and circulating ACE between carriers of different genotypes of *ACE* gene I/D polymorphism.

Subgroup	Studies	II vs. DD					ID vs. DD					II + ID vs. DD				
		WMD	95% CI lower	95% CI upper	*P* value	I^2 (%)	WMD	95% CI lower	95% CI upper	*P* value	I^2 (%)	WMD	95% CI lower	95% CI upper	*P* value	I^2 (%)
FEV$_1$/predicted FEV$_1$																
Patients	6	−0.39	−2.08	1.30	0.652	0.0	−0.81	−2.15	0.52	0.234	0.0	−0.44	−1.71	0.82	0.495	0.0
FEV$_1$ ≥50% predicted	3	−1.05	−3.07	0.96	0.305	0.0	−1.73	−3.50	0.04	0.056	0.0	−1.38	−3.03	0.27	0.101	0.0
FEV$_1$ <50% predicted	3	1.18	−1.92	4.28	0.455	0.0	0.39	−1.64	2.43	0.705	0.0	0.91	−1.07	2.89	0.368	0.0
Controls	1	−2.70	−12.77	7.37	0.599	n.a.	−8.60	−18.62	1.42	0.092	n.a.	−5.65	−14.36	3.06	0.203	n.a.
Circulating ACE																
Patients	2	−14.03	−16.47	−11.59	<0.001	0.0	−9.00	−11.51	−6.49	<0.001	0.0	−11.52	−13.73	−9.31	<0.001	0.0
Control	3	−13.12	−14.54	−11.69	<0.001	37.7	−7.20	−8.63	−5.77	<0.001	47.4	−10.14	−11.49	−8.79	<0.001	43.6

ACE: angiotensin-converting enzyme; WMD: weighted mean difference; 95% CI: 95% confidence interval; DD: deletion/deletion genotype; ID: insertion/deletion; II: insertion/insertion genotype; FEV$_1$: forced expiratory volume in one second; n.a.: not available.

with GOLD stages III–IV. In view of limited studies in subgroup analysis, our findings gave rise to speculation that the *ACE* gene I/D polymorphism may serve as a predictive marker for the early development of COPD. We agree that further validation of our findings in large, well-designed studies is necessary.

Our analyses have several limitations. The first limitation was that our findings were based on cross-sectional observational data, which precluded comment on causality between circulating ACE and COPD risk. The second limitation lay in the analysis of only one polymorphism in the *ACE* gene. The third limitation was that we only retrieved articles published in the English language, as it is estimated that the exclusion of grey literature from meta-analysis may

result in an overestimate of an association impact by an average of 12%.[32] The fourth limitation was although our analyses indicated low probabilities of heterogeneity and publication bias, we cannot fully exclude their potential confounding impact. The fifth limitation was that residual confounding due to unadjusted divergence in baseline characteristics of eligible studies cannot be completely assessed or ruled out using study-level data.

Taken together, our meta-analytical findings demonstrated that the *ACE* gene I/D polymorphism was not associated with the risk of COPD. Large-scale validation is necessary to confirm our findings. For practical reasons, a single well-designed study is warranted to interrogate gene–gene and gene–environment interactions further on

the association of *ACE* gene multiple polymorphisms with COPD risk and related pulmonary phenotypes.

Author contribution

WN planned and designed the study; GX and WN searched the literature, selected articles and abstracted data; GX, GF, YS and LY conducted the data preparation, quality control and data analyses; WN, GX and SW wrote and revised the manuscript.

Declaration of conflicting interest

The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

Funding

ORCID iD

Wenquan Niu (iD) https://orcid.org/0000-0003-1715-3372

References

1. Kressel G, Trunz B, Bub A, et al. Systemic and vascular markers of inflammation in relation to metabolic syndrome and insulin resistance in adults with elevated atherosclerosis risk. *Atherosclerosis* 2009; 202: 263–271.

2. Mascitelli L and Pezzetta F. Role of systemic inflammation in the relationship between metabolic syndrome and cigarette smoking. *Intern Med* 2007; 46: 333.

3. Qasim AN, Metkus TS, Tadesse M, et al. Resistin gene variation is associated with systemic inflammation but not plasma adipokine levels, metabolic syndrome or coronary atherosclerosis in nondiabetic Caucasians. *Clin Endocrinol (Oxf)* 2009; 70: 698–705.

4. Marsland AL, McCaffery JM, Muldoon MF, et al. Systemic inflammation and the metabolic syndrome among middle-aged community volunteers. *Metabolism* 2010; 59: 1801–1808.

5. Watz H, Waschki B, Kirsten A, et al. The metabolic syndrome in patients with chronic bronchitis and COPD: frequency and associated consequences for systemic inflammation and physical inactivity. *Chest* 2009; 136: 1039–1046.

6. Kon SS, Jolley CJ, Shrikrishna D, et al. ACE and response to pulmonary rehabilitation in COPD: two observational studies. *BMJ Open Respir Res* 2017; 4: e000165.

7. Simsek S, Tekes S, Oral D, et al. The insertion/deletion polymorphism in the ACE gene and chronic obstructive pulmonary disease. *Genet Mol Res* 2013; 12: 1392–1398.

8. Pabst S, Theis B, Gillissen A, et al. Angiotensin-converting enzyme I/D polymorphism in chronic obstructive pulmonary disease. *Eur J Med Res* 2009; 14 (Suppl. 4): 177–181.

9. van Suylen RJ, Wouters EF, Pennings HJ, et al. The DD genotype of the angiotensin converting enzyme gene is negatively associated with right ventricular hypertrophy in male patients with chronic obstructive pulmonary disease. *Am J Respir Crit Care Med* 1999; 159: 1791–1795.

10. Gorges J. First-year students' initial motivational beliefs at university: predicted by motivational beliefs derived from within and out-of-school experience and malleable regardless of the extent of students' out-of-school experience. *Front Psychol* 2017; 8: 1258.

11. Teramoto S, Yamamoto H, Yamaguchi Y, et al. Obstructive sleep apnea causes systemic inflammation and metabolic syndrome. *Chest* 2005; 127: 1074–1075.

12. Moher D, Liberati A, Tetzlaff J, et al. Preferred reporting items for systematic reviews and meta-analyses: the PRISMA statement. *BMJ* 2009; 339: b2535.

13. Kanazawa H, Tateishi Y and Yoshikawa J. Acute effects of nifedipine administration in pulmonary haemodynamics and oxygen delivery during exercise in patients with chronic obstructive pulmonary disease: implication of the angiotensin-converting enzyme gene polymorphisms. *Clin Physiol Funct Imaging* 2004; 24: 224–228.

14. Ahsan A, Ram R, Baig MA, et al. ACE I allele and eNOS G allele crosstalk may have a role in chronic obstructive pulmonary disease. *Clin Biochem* 2004; 37: 1037–1040.

15. Kanazawa H, Hirata K and Yoshikawa J. Influence of oxygen administration on pulmonary haemodynamics and tissue oxygenation during exercise in COPD patients with different ACE genotypes. *Clin Physiol Funct Imaging* 2003; 23: 332–336.

16. Yildiz P, Oflaz H, Cine N, et al. Gene polymorphisms of endothelial nitric oxide synthase enzyme associated with pulmonary hypertension in patients with COPD. *Respir Med* 2003; 97: 1282–1288.

17. Tkacova R, Joppa P, Stancak B, et al. The link between angiotensin-converting enzyme genotype and pulmonary artery pressure in patients with COPD. *Wien Klin Wochenschr* 2005; 117: 210–214.

18. Lee J, Nordestgaard BG and Dahl M. Elevated ACE activity is not associated with asthma, COPD, and COPD comorbidity. *Respir Med* 2009; 103: 1286–1292.

19. Kuzubova NA, Chukhlovin AB, Morozova EB, et al. Common intronic D variant of ACE gene is associated with endothelial dysfunction in COPD. *Respir Med* 2013; 107: 1217–1221.

20. Mlak R, Homa-Mlak I, Powrozek T, et al. Impact of I/D polymorphism of ACE gene on risk of development and course of chronic obstructive pulmonary disease. *Arch Med Sci* 2016; 12: 279–287.

21. Busquets X, MacFarlane NG, Heine-Suner D, et al. Angiotensin-converting-enzyme gene polymorphisms, smoking and chronic obstructive pulmonary disease. *Int J Chron Obstruct Pulmon Dis* 2007; 2: 329–334.

22. Zhang X, Wang C, Dai H, et al. Association between angiotensin-converting enzyme gene polymorphisms and exercise performance in patients with COPD. *Respirology* 2008; 13: 683–688.

23. Ayada C, Toru U, Genc O, et al. Evaluation of whether the ACE gene I/D polymorphism constitutes a risk factor for chronic obstructive pulmonary disease in the Turkish population. *Genet Mol Res* 2014; 13: 10427–10433.

24. Ulasli SS, Eyuboglu FO, Verdi H, et al. Associations between endothelial nitric oxide synthase A/B, angiotensin converting enzyme I/D and serotonin transporter L/S gene polymorphisms with pulmonary hypertension in COPD patients. *Mol Biol Rep* 2013; 40: 5625–5633.

25. Egli A, Schumann DM, Mandal J, et al. P142 IFNL3 polymorphisms and circulating levels are associated with COPD severity and outcomes. *Chest* 2017; 151: A39.

26. Mizuno S, Ishizaki T, Kadowaki M, et al. p53 Signaling pathway polymorphisms associated with emphysematous changes in patients with COPD. *Chest* 2017; 152: 58–69.

27. Qin R and Wang P. Association of ADAM33 gene polymorphisms with chronic obstructive pulmonary disease in Han population of Northern China. *Chest* 2016; 149: A246.

28. Cox R, Bouzekri N, Martin S, et al. Angiotensin-1-converting enzyme (ACE) plasma concentration is influenced by multiple ACE-linked quantitative trait nucleotides. *Hum Mol Genet* 2002; 11: 2969–2977.

29. Kang SW, Kim SK, Chung JH, et al. Genetic polymorphism of angiotensin-converting enzyme and chronic obstructive pulmonary disease risk: an updated meta-analysis. *Biomed Res Int* 2016; 2016: 7636123.

30. Li W, Lan F, Yan F, et al. Angiotensin-converting enzyme I/D polymorphism is associated with COPD risk in Asian population: evidence from a meta-analysis. *COPD* 2013; 10: 35–39.

31. Li X, Wei N, Wu Z, et al. The D/I polymorphism in the angiotensin-converting enzyme gene and chronic obstructive pulmonary disease risk: a meta-analysis. *COPD* 2012; 9: 485–491.

32. McAuley L, Pham B, Tugwell P, et al. Does the inclusion of grey literature influence estimates of intervention effectiveness reported in meta-analyses? *Lancet* 2000; 356: 1228–1231.