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Interleukin-18 Polymorphisms Deficiency Association with Asthma Risk: An Update Meta-analysis

Huan Liu[1,2], Yu Feng[3], Wei Zhang[2], Xiao-Dong Deng[2], Ying Ma[4], and Yun Liu[2]

[1] *Department of Preventive Medicine, North Sichuan Medical College, Nanchong, Sichuan, China*
[2] *Department of Forensic Medicine, North Sichuan Medical College, Nanchong, Sichuan, China*
[3] *Department of Criminal Science and Technology, Chongqing Police College, Chongqing, China*
[4] *Department of Neurology, Affiliated Hospital of North Sichuan Medical College, Nanchong, Sichuan, China*

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ABSTRACT

Growing evidence indicated conflicting results that Interleukin-18 (IL-18) promoter polymorphisms rs1946518 (A-607C), rs187238 (G-137C) and rs549908 (A-105C) were associated with asthma risk. The aim of this study is to comprehensively evaluate the IL-18 polymorphisms and asthma by a systematic review and meta-analysis.

A total of 12 studies testing the association between these polymorphisms and asthma were examined (8 studies for A-607C, 8 studies for G-137C, and 4 studies for A-105C) in the update meta-analysis, up to Dec 30, 2017. Summary odds ratios (ORs) and 95% confidence intervals (CI) were used to estimate the strength of association between each polymorphism and asthma using fixed- and random-effects models when appropriate. Heterogeneity and publication bias were evaluated.

The meta-analysis results indicated that any allele frequencies of the IL-18 polymorphisms (A-607C, G-137C and A-105C) was not associated with asthma risk (p>0.05). And no statistically significant association was observed between genotype frequencies of these polymorphisms and asthma under different genetic models (p>0.05). Subgroup analysis results were similar to the main analysis by ethnicity, sample size, genotyping methods, matching criteria and quality score. There was no evidence of publication bias.

The present meta-analysis suggests that IL-18 polymorphisms (A-607C, G-137C and A-105C) were unlikely to be associated with asthma risk.

Keywords: Asthma; Genetic susceptibility; Interleukin-18; Meta-analysis; Single nucleotide polymorphism

INTRODUCTION

Asthma is a common, complex, chronic medical condition, which is characterized by lung inflammation, reversible airflow obstruction, and enhanced airway responsiveness to a diversity of environmental stimuli.[1] It was caused by a combination of genetic and environmental factors,[2,3] and which is associated with the imbalance of different types of helper T cells (Th), especially the imbalance of Th1/Th2 cells,[4,5] Interleukin-18 (IL-18) is a pleiotropic cytokine

Corresponding Author: Yun Liu, PhD;
Department of Forensic Medicine, North Sichuan Medical College, NO.234 Fujiang Road, Nanchong, 637000, Sichuan, China. Tel/Fax: (+86 817) 2240 083, E-mail: xyun2005@163.com

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involved in regulating the immune response of Th1/Th2 cells.[6] So the IL-18 gene is considered as a candidate asthma susceptibility gene. IL-18 was initially isolated from the serum of Mycobacterium bovis Bacillus Calmette-Guerin (BCG)-infected mice challenged with lipopolysaccharide (LPS) which is a member of the Interleukin-1 (IL-1) family and described as IFN-γ-inducing factor (IGIF).[7,8] The gene for human IL-18 is located on chromosome 11q22.2-22.3, a region that has been previously linked to atopy-related traits.[9]

Recently, IL-18 promoter polymorphisms at positions A-607C (rs1946518), G-137C (rs187238) and A-105C (rs549908) had been extensively studied. However, the results of these studies were incoherent. Many studied suggested that the IL-18 polymorphisms were associated with asthma,[10-14] whereas some studies indicated that lack of an association between IL-18 polymorphisms and asthma.[6,15-18] The inconsistent results may result in a single study or small sample size makes the lower statistical power. Ma and colleagues'[19] conducted a meta-analysis to evaluate the association between these polymorphisms (A-607C, G-137C) and the risk of asthma in 2012. In their study, A-607C polymorphism was associated with asthma risk, but G-137C did not. In the past few years, more studies concerning the association between the IL-18 polymorphisms (A-607C, G-137C) and the risk of asthma have been published in different regions.[12,13,17,18] We thought these studies should also be included, and we added a new polymorphism (A-105C) of IL-18.[14,15,20,21] Thus, the aim of our study is to use a systematic review and update meta-analysis to estimate the association between IL-18 polymorphisms (A-607C, G-137C, and A-105C) and asthma according to all published case-control studies.

MATERIALS AND METHODS

Publication Search

We searched the PubMed, Medline, Web of Science, Wanfang and China National Knowledge Infrastructure (CNKI) databases for all articles on the association between IL-18 polymorphisms and asthma up till December 30, 2017. The search terms were as followed: ("IL-18" or "interleukin-18" or "interleukin 18" or "IGIF") and ("genetic variation" or "genetic variant" or "polymorphism" or "snps") and ("asthma" or "bronchial asthma" or "allergic asthma"). All of the identified articles and their references were checked as well for other relevant studies. We evaluated review and previous meta-analysis articles in order to found additional eligible studies.

Inclusion Criteria and Exclusion Criteria

Eligible studies should cater all the followed criteria: 1. Studies on people were associated between the IL-18 gene polymorphisms (A-607C, G-137C, and A-105C) and asthma, but not animal experiments. 2. Studies should cater case-control or cross-sectional research, and excluded review, systematic review or meta-analysis. 3. Data provided within the studies was useful for odds ratios (ORs) and 95% credibility interval (CI), or ORs and 95% CI can be calculated. Studies without extractable or credible data should be excluded.[22] 4. For overlapping studies, only the most complete or the latest literature will be included. 5. The meta-analysis is not limited sample size of the literature, no matter how many samples can be included.

Data Extraction

Data was extracted from each eligible literature according to the above-mentioned inclusion and exclusion criteria listed by 2 investigators (Liu H and Zhang W) independently, who were blinded to each other. Extractive data were checked by 1 investigator (Deng XD), and disputed data were resolved through negotiation. Characteristics of the studies were extracted as followed: first author, year of publication, country, Ethnicity, numbers of cases and controls, genotype distribution, allele frequency, evidence of hardy-weinberg equilibrium (HWE), genotyping methods, and matching variables. The basic information of eligible studies was showed in Table 1.

Quality Score Assessment

Quality of each eligible study was evaluated independently by 3 investigators (Liu H, Zhang W and Liu Y) who were blinded to each other according to a set of predetermined criteria (Supplementary Data) which was modified from previous research,[23,24] and disputes were settled by consensus among the three authors. Total scores were ranged from 0 (worst) to 10 (best), and quality scores≥6 were identified as high-quality studies, whereas as low-quality one (Table 1).

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Table 1. Characteristics of the studies included in the meta-analysis on interleukin-18 polymorphisms and asthma risk

Author	Year	Country	Ethnicity	Sample size (case/control)	genotype distribution (case/control)		
A-607C					AA	AC	CC
Lachheb J[10]	2008	Tunisian	African	105/112	23/8	45/58	37/46
Pawlik A[6]	2007	Poland	Caucasian	231/305	46/53	95/45	90/107
Shin HD[15]	2005	Korea	Asian	435/140	114/30	185/70	94/24
Heinzmann A[16]	2004	Germany	Caucasian	321/270	46/45	121/134	63/90
Harada M[20]	2009	Japan	Asian	453/719	166/234	197/362	89/119
Shaaban HH[17]	2014	Egypt	African	40/20	9/9	21/5	10/6
Wu SQ[13]	2012	China	Asian	120/120	29/41	59/49	32/30
Yi jinping[18]	2013	China	Asian	146/106	35/32	76/46	35/28
G-137C					GG	GC	CC
Pawlik A[6]	2007	Poland	Caucasian	231/305	124/162	85/112	22/31
Imboden M[11]	2006	Switzerland	Caucasian	530/5204	296/2804	209/2012	25/388
Shin HD[15]	2005	Korea	Asian	435/140	337/114	93/34	8/1
Heinzmann A[16]	2004	Germany	Caucasian	321/270	117/133	93/118	21/17
Harada M[20]	2009	Japan	Asian	453/719	341/542	104/167	8/10
Birbian N[12]	2013	India	Asian	410/414	287/290	109/91	14/33
Wu SQ[13]	2012	China	Asian	120/120	81/83	33/32	6/5
Yi jinping[18]	2013	China	Asian	146/106	104/77	38/27	4/2
A-105C					AA	AC	CC
Lee CC[14]	2008	chinese	Asian	201/60	156/31	45/27	0/2
Higa S[21]	2003	Japan	Asian	497/85	394/58	98/23	5/4
Shin HD[15]	2005	Korea	Asian	435/140	307/101	99/36	9/1
Harada M[20]	2009	Japan	Asian	453/719	342/542	102/165	7/11

Author	Allele frequency (case/control)		hardy-weinberg equilibrium (HWE)	Genotyping method	Matching criteria	Quality score
A-607C	**A**	**C**				
Lachheb J[10]	91/74	119/150	Yes	PCR-RFLP	Age, gender	6
Pawlik A[16]	187/251	275/359	Yes	AS-PCR	NA	6
Shin HD[15]	413/130	373/118	Yes	SNaPshot	Age, gender	7
Heinzmann A[16]	213/224	247/314	Yes	PCR-RFLP	age	8
Harada M[20]	529/830	375/600	Yes	TaqMan	age	5
Shaaban HH[17]	39/23	41/17	No	PCR-RFLP	age	4.5
Wu SQ[13]	117/131	123/109	Yes	PCR-SSP	Age, gender	6
Yi jinping[18]	146/110	146/102	Yes	PCR-SSP	Age, gender	6
G-137C	G	C				
Pawlik A[16]	333/436	129/174	Yes	AS-PCR	NA	6
Imboden M[11]	801/7620	259/2788	Yes	TaqMan	Age, gender	7
Shin HD[15]	767/262	109/36	Yes	SNaPshot	Age, gender	7
Heinzmann A[16]	327/384	135/152	Yes	PCR-RFLP	age	8
Harada M[20]	786/1251	120/187	Yes	TaqMan	age	5
Birbian N[12]	683/671	137/157	Yes	PCR-ARMS	age, same area	7
Wu SQ[13]	195/198	45/42	Yes	PCR-SSP	Age, gender	6
Yi jinping[18]	246/181	46/31	Yes	PCR-SSP	Age, gender	6
A-105C	A	C				
Lee CC[14]	357/89	45/31	No	PCR-RFLP	NA	4.5
Higa S[21]	886/139	108/31	Yes	PCR-SSP	NA	6
Shin HD[15]	713/238	117/38	Yes	SNaPshot	Age, gender	7
Harada M[20]	786/1249	116/187	Yes	TaqMan	age	5

PCR-RFLP: polymerase chain reaction-restriction fragment length polymorphism
PCR-SSP: PCR with sequence-specific primers
PCR-ARMS: PCR with amplification refractory mutation system
AS-PCR: allele specific-PCR

Statistical Analysis

We first checked the control subjects in each study if there was significant deviation from the HWE by a web-based program (http://ihg2 helmholtz-muenchen.de/cgi-bin/hw/hwa1.pl), and considered $p<0.05$ was significant.

The STATA software (version 12, Stata Corporation) was applied for data calculation and analysis. Summary ORs with corresponding 95% CI were used to estimate the association between genotypes, allele and asthma genetic risk for each polymorphism in different comparison genetic models and allele, including dominant model (-607A/C: CC + CA vs AA, -137G/C: CC + GC vs GG, and -105A/C: CC + CA vs AA), and recessive model (-607A/C: CC vs CA + AA, -137G/C: CC vs GC + GG, and -105A/C: CC vs CA + AA), heterozygote comparison (-607A/C: CA vs AA, -137G/C: GC vs GG, and -105A/C: CA vs AA), homozygote comparison (-607A/C: CC vs AA, -137G/C: CC vs GG, and -105A/C: CC vs AA), and allele comparison (-607A/C: C vs A, -137G/C: C vs G, and -105A/C: C vs A), respectively.

The statistical heterogeneity among eligible studies was estimated using the Q-test and I^2 statistics.[25,26] The fixed-effect model (Mantel-Haenszel method) was used to estimate the pooled ORs if the heterogeneity among studies was not significant ($p>0.10$ and $I^2<50\%$). Otherwise, random-effect model should be considered.[27,28] To explore sources of heterogeneity in studies, we used a galbraith plot to estimate the heterogeneity. Besides, logistic meta-regression analysis and subgroup analysis were performed to investigate potential sources of heterogeneity among studies. The logistic meta-regression analysis was conducted by year of publication, ethnicity, sample size (≥500 and＜500 subjects), genotyping methods, matching criteria and HWE. Subgroup analysis included the following variables: ethnicity (Asian and No Asian), Sample size (≥500 and＜500 subjects), genotyping methods (polymerase chain reaction-restriction fragment length polymorphism (PCR-RFLP) and no PCR-RFLP), matching criteria and quality score. Since the number of studies of -105A/C locus was only 4, logistic meta-regression analysis and subgroup analysis were not performed.

Sensitivity analysis was applied to evaluate the reliability of this meta-analysis. A Begg's funnel plot and Egger's regression asymmetry test were used to evaluate publication bias of eligible studies, if $p<0.05$, publication bias was considered statistically significant.[29,30]

RESULTS

Characteristics of Studies

In total, 12 literatures fulfilled the criteria (Figure 1).

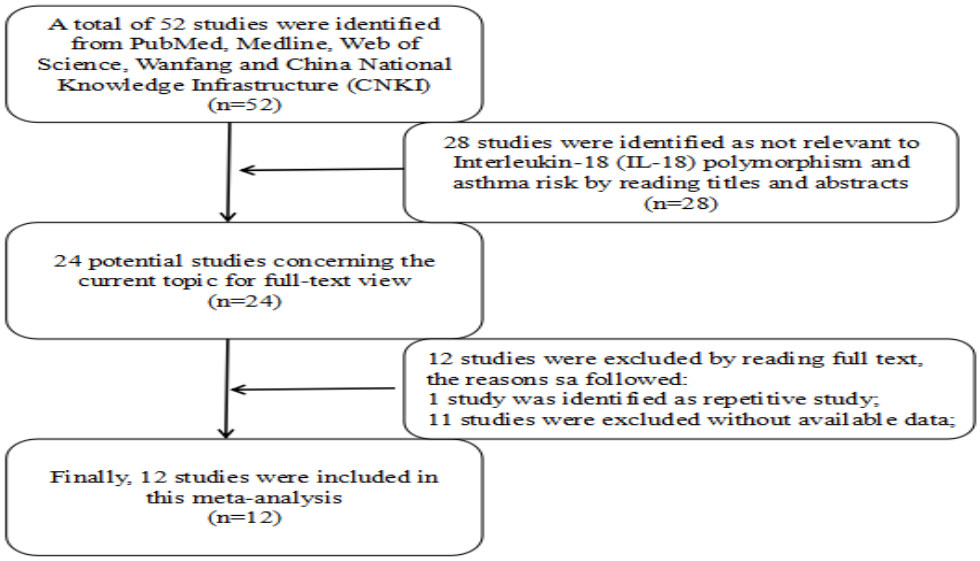

Figure 1. Flow diagram of included/excluded studies in meta-analysis on interleukin-18 polymorphisms and asthma risk

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In some literatures, two or three gene loci have been studied.[6,13,15,16,18,20] The characteristics of the literatures are summarized in Table 1. For IL-18 polymorphisms, all the 8 studies (1851 cases and 1792 controls) for -607 A/C,[6,10,13,15-18,20] 8 studies (2646 cases and 7278 controls) for -137 G/C[6,11-13,15,16,18,20] and 4 studies (1586 cases and 1004 controls) for -105 A/C[14,15,20,21] were conducted in the meta-analysis. Four studies were conducted in Asian,[13,15,18,20] tow in Caucasian,[6,16] and tow in African[10,17] for -607A/C; Five studies were conducted in Asian,[12,13,15,18,20] three in Caucasian[6,11,16] for 137 G/C; Four studies were conducted in Asian[14,15,20,21] for 105 A/C. The sample size varied greatly in the eligible literatures, ranging from 60 to 5734. SNaPShot, TaqMan, AS-PCR,PCR-RFLP, PCR-SSP and PCR-ARMS genotyping methods were used among the eligible literatures. Except for Lee CC[14] and Shaaban HH,[17] the genotype distributions among the controls of all eligible literatures were consistent with HWE. All studies have presented matching information, such as age and gender, except for the studies of Pawlik A,[6] Lee CC[14] and Higa S.[21]

Main Results of Meta-Analysis

The meta-analysis suggested that the A-607C polymorphism, G-137C polymorphism and A-105C polymorphism were not associated with asthma in all comparison allele and genotypes genetic models (dominant, recessive, heterozygote and homozygote; Table 2, Table 3, and Table 4). However, we failed to find out any significant association between the A-607C polymorphism, G-137C polymorphism and asthma in all comparison allele and genotypes models through subgroup analysis according to ethnicity, sample size, genotyping methods, matching criteria and quality score (Table 2, Table 3). Since the number of studies of -105A/C locus was only 4, subgroup analysis were not performed.

Table 2. Stratified analysis of the interleukin-18 (IL-18) -607A/C polymorphism on asthma risk

	Studies(cases/controls)	CC vs AA (homozygote)		CA vs AA (heterozygote)	
		OR (95%CI)[a]	I^2（%）	OR (95%CI)	I^2（%）
		P	P_Q[b]	P	P_Q[b]
Total	8（1851/1792）	0.951(0.781-1.158)	39.0	0.936(0.672-1.304)	66.4
		0.617	0.119	0.695	0.004
Ethnicity					
Asian	4（1154/1085）	1.114(0.866 -1.434)	0.0	1.048(0.714-1.539)	62.6
		0.402	0.822	0.810	0.045
No Asian	4（697/707）	0.715(0.413-1.239)	57.0	0.816(0.411-1.619)	74.4
		0.231	0.073	0.561	0.008
Sample size					
≥500	4（1440/1434）	0.951(0.758-1.193)	0.0	0.813(0.672-0.985)	0.0
		0.662	0.583	0.034	0.868
＜500	4（411/358）	0.934(0.432-2.018)	68.5	1.242 (0.498-3.095)	81.1
		0.862	0.023	0.643	0.001
Genotyping methods					
PCR-RFLP	3（466/402）	0.619(0.274-1.395)	60.4	0.907(0.272-3.019)	82.9
		0.247	0.080	0.873	0.003
No PCR-RFLP	5（1385/1390）	1.081(0.864-1.353)	0.0	0.971(0.714-1.320)	54.5
		0.493	0.884	0.850	0.067
Matching criteria					
Yes	7（1620/1487）	0.931(0.667-1.300)	47.7	0.976(0.662-1.440)	70.4
		0.674	0.075	0.904	0.002
No		0.969(0.597-1.573)	NA	0.755(0.471-1.210)	NA
	1（231/305）	0.899	NA	0.243	NA
Quality score					
≥6	6（1358/1053）	0.877(0.608-1.265)	51.2	0.869(0.578-1.308)	68.0
		0.482	0.069	0.501	0.008
＜6	2（493/739）	1.083(0.779-1.504)	0.0	1.642(0.352-7.651)	80.8
		0.635	0.525	0.528	0.022

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Table 2. Continue

	CC + CA vs AA (dominant)		CC vs CA + AA (recessive)		C vs A	
	OR (95%CI) [a]	I^2 (%)	OR (95%CI) [a]	I^2 (%)	OR (95%CI) [a]	I^2 (%)
	P	P_Q [b]	P	P_Q [b]	P	P_Q [b]
Total	0.928 (0.692-1.243)	62.4	1.040(0.888-1.219)	0.0	0.970(0.879-1.070)	25.6
	0.614	0.010	0.628	0.445	0.542	0.225
Ethnicity						
Asian	1.035 (0.743-1.441)	56.0	1.165(0.936-1.450)	0.0	1.027 (0.905-1.165)	0.0
	0.838	0.078	0.172	0.729	0.679	0.639
No Asian	0.792 (0.431-1.454)	71.9	0.917(0.729-1.155)	12.1	0.888(0.760-1.038)	47.4
	0.451	0.014	0.463	0.332	0.137	0.127
Sample size						
≥500	0.825 (0.690 -0.987)	0.0	1.095(0.911-1.315)	39.0	0.959(0.858-1.073)	0.0
	0.035	0.993	0.334	0.178	0.469	0.621
＜500	1.128 (0.497-2.560)	80.0	0.894(0.652-1.225)	0.0	1.017 (0.725-1.428)	59.9
	0.773	0.002	0.485	0.863	0.920	0.058
Genotyping methods						
PCR-RFLP	0.801 (0.280-2.290)	80.9	0.761(0.561-1.032)	0.0	0.804 (0.656-0.985)	41.4
	0.679	0.005	0.079	0.993	0.035	0.182
No PCR-RFLP	0.930(0.781 -1.108)	43.2	1.169(0.971-1.409)	0.0	1.027(0.918 -1.149)	0.0
	0.416	0.134	0.100	0.861	0.639	0.792
Matching criteria						
Yes	0.946 (0.668-1.339)	67.6	1.007(0.844-1.203)	3.7	0.959(0.862-1.068)	34.4
	0.754	0.005	0.936	0.398	0.447	0.165
No	0.846(0.546-1.311)	NA	1.181(0.829-1.683)	NA	1.028(0.804-1.315)	NA
	0.454	NA	0.356	NA	0.825	NA
Quality score						
≥6	0.879(0.607-1.273)	65.6	0.984(0.815-1.187)	2.5	0.955(0.845-1.079)	40.2
	0.494	0.013	0.864	0.400	0.459	0.137
＜6	1.342(0.421-4.272)	75.4	1.194(0.889-1.604)	0.0	0.998 (0.846-1.177)	0.0
	0.619	0.044	0.238	0.468	0.980	0.352

[a] 95% confidence intervals. [b] P_Q value of Q test for heterogeneity test and associated I^2 are shown. Random-effects model was used when I^2>50% and/or P_Q<0.1; otherwise, fixed-effects model was used. NA, not applicable.

Heterogeneity Analysis

A-607C polymorphism

For dominant and heterozygote genetic models, the values of I^2 and P_Q were greater than 50% and less than 0.10, which indicated statistically significant heterogeneity among eligible literatures. Logistic meta-regression analysis was performed to explore the sources of heterogeneity among the eligible publications, including year of publication, ethnicity, sample size, genotyping methods, matching criteria and HWE. However, no data could explain the heterogeneity in allele and different comparison genetic models (p≥0.501). Subgroup analysis based on ethnicity, sample size, genotyping methods, matching criteria and quality score showed that no asian, PCR-RFLP and small sample size might be the sources of heterogeneity (Table 2). The galbraith plots showed that the Lachheb J[10] and Shaaban HH[17] were the outlier studies, which were in accordance with subgroup analysis belong the no asian, PCR-RFLP and small sample size publications. The heterogeneity was greatly dropped under allele and different comparison genetic models by omitting these two literatures (allel: I^2=0.0%, P_Q=0.539; dominant: I^2=32.3%, P_Q=0.194;

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recessive: I^2=8.0%, P_Q=0.365; heterozygote: I^2=43.2%, P_Q=0.117; homozygote: I^2=0.0%, P_Q=0.601). And summary ORs and 95%CI were not significantly influenced (allel: OR=0.991, 95%CI=0.895-1.098, *p*=0.861; dominant: OR=0.913, 95%CI=0.776-1.074, *p*=0.273; recessive: OR=1.074, 95%CI=0.909-1.270, *p*=0.402; heterozygote: OR=0.912, 95%CI=0.766-1.085, *p*=0.298; homozygote: OR=1.007, 95%CI=0.820-1.237, *p*=0.946). The results indicated that the Lachheb J[10] and Shaaban HH[17] studies were the major sources of the heterogeneity in the meta-analysis.

G-137C Polymorphism

For recessive genetic models, the values of P_Q was less than 0.10, which indicated statistically heterogeneity among eligible literatures. Logistic meta-regression analysis was performed to explore the sources of heterogeneity among the eligible publications, including year of publication, ethnicity, sample size, genotyping methods, matching criteria. However, no data could explain the heterogeneity in allele and different comparison genetic models ($p \geq 0.728$).

Table 3. Stratified analysis of the IL-18 -137G/C polymorphism on asthma risk

		CC vs GG (homozygote)		GC vs GG (heterozygote)		CC / GC vs GG (dominant)	
	Studies(cases/ controls)	OR (95%CI)[a]	I^2（%）	OR (95%CI)	I^2（%）	OR (95%CI)	I^2（%）
		P	P_Q[b]	P	P_Q	P	P_Q
Total	8（2646/7278）	0.867(0.605-1.244)	37.5	1.003(0.895-1.124)	0.0	0.970 (0.870 - 1.082)	0.0
		0.439	0.130	0.953	0.966	0.590	0.998
Ethnicity							
Asian	5（1564/1499）	0.770(0.497-1.191)	40.8	1.049(0.883-1.247)	0.0	1.013 (0.858 -1.195)	0.0
		0.240	0.149	0.584	0.874	0.880	0.998
No Asian	3（1082/5779）	0.789(0.585-1.063)	54.7	0.970(0.834-1.128)	0.0	0.939(0.812 -1.086)	0.0
		0.119	0.110	0.691	0.898	0.397	0.951
Sample size							
≥500	6（2380/7052）	0.835(0.552-1.263)	50.7	1.000 (0.888-1.126)	0.0	0.962 (0.859 -1.078)	0.0
		0.394	0.071	0.997	0.871	0.503	0.996
＜500	2（266/226）	1.311(0.484-3.551)	0.0	1.049(0.699-1.575)	0.0	1.076(0.729 -1.589)	0.0
		0.594	0.863	0.816	0.973	0.711	0.985
PCR-RFLP	1（321/270）	1.404(0.707 -2.788)	NA	0.896(0.620-1.295)	NA	0.960 (0.675-1.365)	NA
		0.332	NA	0.559	NA	0.820	NA
No PCR-RFLP	7（2325/7008）	0.719(0.550-0.939)	26.1	1.016(0.901-1.145)	0.0	0.972(0.866 -1.090)	0.0
		0.016	0.230	0.799	0.960	0.622	0.995
Yes	7（2415/6973）	0.880(0.565-1.371)	44.8	1.005(0.892-1.133)	0.0	0.970(0.864 -1.088)	0.0
		0.573	0.092	0.937	0.930	0.599	0.995
No	1（231/305）	0.927(0.512-1.680)	NA	0.992(0.687-1.430)	NA	0.978(0.694-1.377)	NA
		0.803	NA	0.964	NA	0.897	NA
≥6	7（2193/6559）	0.756(0.585 -0.977)	40.9	1.006(0.888 -1.140)	0.0	0.964(0.856-1.086)	0.0
		0.033	0.118	0.921	0.930	0.544	0.996
＜6	1（453/719）	1.272(0.497-3.254)	NA	0.990(0.748-1.309)	NA	1.006(0.766-1.321)	NA
		0.616	NA	0.943	NA	0.967	NA

[a] 95% confidence intervals. [b] Q value of Q test for heterogeneity test and associated I^2 are shown.Random-effects model was used when I^2>50% and/or P_Q<0.1;otherwise, fixed-effects model was used. NA, not applicable.

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Table 3. Continue

	Studies(cases/ controls)	CC vs GC/GG (recessive)		C vs G	
		OR (95%CI)	I^2(%)	OR (95%CI)	I^2(%)
		P	P_Q	P	P_Q
Total	8（2646/7278）	0.881(0.603-1.288)	44.2	0.946 (0.864-1.036)	0.0
		0.514	0.084	0.232	0.869
Ethnicity					
Asian	5（1564/1499）	0.747(0.485-1.152)	45.7	0.977 (0.845-1.130)	0.0
		0.188	0.118	0.752	0.803
No Asian	3（1082/5779）	0.897(0.543-1.480)	60.6	0.926(0.824-1.040)	0.0
		0.669	0.079	0.195	0.541
Sample size					
≥500	6（2380/7052）	0.848(0.548-1.310)	56.7	0.935(0.851-1.027)	0.0
		0.457	0.041	0.163	0.785
＜500	2（266/226）	1.292(0.481-3.475)	0.0	1.090 (0.777 -1.529)	0.0
		0.611	0.859	0.619	0.992
Genotyping methods					
PCR-RFLP	1（321/270）	1.476(0.759-2.872)	NA	1.043 (0.792-1.373)	NA
		0.251	NA	0.764	NA
No PCR-RFLP	7（2325/7008）	0.714(0.549-0.929)	31.2	0.934(0.848-1.028)	0.0
		0.012	0.190	0.165	0.855
Matching criteria					
Yes	7（2415/6973）	0.896(0.562-1.429)	50.9	0.942(0.856-1.038)	0.0
		0.645	0.057	0.226	0.793
No	1（231/305）	0.930(0.523-1.654)	NA	0.971(0.742-1.270)	NA
		0.806	NA	0.828	NA
Quality score					
≥6	7（2193/6559）	0.848(0.561-1.281)	47.9	0.934(0.847-1.030)	0.0
		0.433	0.074	0.171	0.842
＜6	1（453/719）	1.275(0.499-3.254)	NA	1.021(0.799-1.306)	NA
		0.612	NA	0.866	NA

Table 4. Main analysis of the IL-18-105A/C polymorphism on asthma risk

	Studies (cases/controls)	CC vs AA (homozygote)		CA vs AA (heterozygote)		CC / CA vs AA (dominant)	
		OR (95%CI)[a]	I^2（%）	OR (95%CI)	I^2（%）	OR (95%CI)	I^2（%）
		P	P_Q[b]	P	P_Q	P	P_Q
Total	4（1586/1004）	0.484(0.112-2.094)	68.4	0.693(0.451-1.066)	72.8	0.668(0.415-1.074)	78.9
		0.331	0.023	0.095	0.012	0.096	0.003

	Studies (cases/controls)	CC vs CA/ AA (recessive)		C vs A	
		OR (95%CI)	I^2(%)	OR (95%CI)	I^2(%)
		P	P_Q	P	P_Q
total	4（1586/1004）	0.538(0.137-2.106)	63.9	0.692(0.441-1.084)	81.7
		0.373	0.040	0.108	0.001

[a] 95% confidence intervals. [b]P_Q value of Q test for heterogeneity test and associated I^2 are shown. Random-effects model was used when I^2>50% and/or P_Q<0.1;otherwise, fixed-effects model was used.

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Subgroup analysis (Table 3) and galbraith plots showed that the Imboden M[11] maybe the source of heterogeneity. The heterogeneity was greatly dropped under allele and different comparison genetic models by omitting the literature (allel: I^2=0.0%, P_Q=0.936; dominant: I^2=0.0%, P_Q=1.000; recessive: I^2=40.7%, P_Q=0.119; heterozygote: I^2=0.0%, P_Q=0.935; homozygote: I^2=31.9%, P_Q=0.184). And summary ORs and 95%CI were not significantly influenced (allel: OR=0.987, 95%CI=0.879-1.109, *p*=0.827; dominant: OR=0.999, 95%CI=0.871-1.146, *p*=0.987; recessive: OR=0.920, 95%CI=0.678-1.248, *p*=0.590; heterozygote: OR=1.015, 95%CI=0.879-1.173, *p*=0.834; homozygote: OR=0.919, 95%CI=0.673-1.255, *p*=0.594). The results indicated that the Imboden M[11] publication was the major source of the heterogeneity in the meta-analysis.

Sensitivity Analysis

Sensitivity analysis was performed by sequentially deleting one study at a time. For each polymorphism, the significance of pooled ORs were not fundamentally altered under allele and different comparison genetic models (data not shown) that indicated our meta-analysis results were statistically reliable.

Publication Bias

Begg's funnel plot and Egger's test were used to estimate the publication bias. The shapes of the Begg's funnel plot did not indicate any evidence of obvious asymmetry under different genetic models. We did not find obvious publication bias for any of the polymorphisms under different genetic models (A-607C, Egger's test $p \geq 0.806$; G-137C, Egger's test $p \geq 0.662$; A-105C, Egger's test $p \geq 0.176$).

DISCUSSION

I L-18 is a unique cytokine that enhances innate immunity and both Th1 and Th2 driven immune responses.[17] In human beings, the secretion of IL-18 by peripheral blood mononuclear cells and level of serum IL-18 were increased in asthma.[31,32] So far, the polymorphisms of IL-18 genetic susceptibility to asthma have been widely studied, but the results were not consistent. Therefore, we performed a meta-analysis to clarify the relationship between the three polymorphisms (-607A/C, -137G/C, -105A/C) and susceptibility to asthma.

In current meta-analysis, we did not find a genetic association between the polymorphisms (-607A/C, -137G/C, -105A/C) and asthma risk in overall populations. Interestingly, for -607A/C polymorphism, our result was inconsistent with Ma and colleagues' meta-analysis,[19] it might be that we added three studies which might have effectively altered the overall results. But for -137G/C polymorphism, our result was consistent with Ma and colleagues' meta-analysis[19], three case-control studies were added, which have no effect on the result. For -105A/C polymorphism, as we have seen, this study was the first comprehensive meta-analysis to evaluate the relationship with asthma susceptibility. Perhaps, the results showed that the three polymorphisms were of no role in asthma risk was premature due to limited number of studies included in this study.

Heterogeneity is a crucial issue when conducting a meta-analysis. There existed significant heterogeneity in dominant and heterozygote genetic models comparisons in A-607C polymorphism. We failed to find any characteristics that could explain the heterogeneity using the meta-regression analysis. Subgroup analysis revealed that no asian, PCR-RFLP and small sample size might be the sources of heterogeneity. The galbraith plots showed that the Lachheb J[10] and Shaaban HH[17] were the outlier studies, which were in accordance with subgroup analysis belong the no asian, PCR-RFLP and small sample size publications. The heterogeneity was greatly dropped under allele and different comparison genetic models by omitting these two literatures, and summary ORs and 95%CI were not significantly influenced. In addition, the results indicated that the study of Lachheb J[10] and Shaaban HH[17] were the sources of heterogeneity. For -137G/C polymorphism, significant heterogeneity was found in the recessive genetic models. We failed to find any characteristics that could explain the heterogeneity using the meta-regression analysis. Subgroup analysis and galbraith plots showed that the Imboden M[11] maybe the source of heterogeneity. The heterogeneity was greatly dropped under allele and different comparison genetic models by omitting the literature, and summary ORs and 95%CI were not significantly influenced. So, we think the study of Imboden M[11] was the source of heterogeneity.

Begg's test and Egger's test showed no publication bias in this meta- analysis, and sensitivity analysis

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showed that the current meta-analysis results were stable and reliable.

There were some limitations to this meta-analysis need to be taken into account when interpreting the results. First, in this meta-analysis, all eligible studies were published in English and Chinese indexed by the selected databases, the lack of data published in other languages might contribute to some publication bias. Second, The ethnicity of studies (A-607C: African, Caucasian, Asian; G-137C: Caucasian, Asian; A-105C: Asian) were too limited to be extended to all races. Third, we excluded some studies that cannot acquire genotype numbers or frequencies from the original studies, which may result in a selection bias. Fourth, asthma is a complex disease, we studied only three polymorphisms of one gene. But we did not conduct the gene-gene and gene-environment interactions analysis because of the unavailability of raw data from the original studies. Finally, elimination of these studies which the genetic distributions of the controls in some studies were deviated from the HWE from analysis did not influence the meta-analysis results significantly. So, we did not exclude these studies from our analysis. Due to these limitations, the results of this meta-analysis should be further verified by prospective trials or full-gene sequencing of random, multicenter, and large samples.

This meta-analysis suggests that the IL-18 gene polymorphisms (A-607C, G-137C and A-105C) may not contribute to asthma susceptibility.

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REFERENCES

1. Zhen Che, Xiaoxia Zhu, Cijiang Yao, Ying Liu, Yanyan Chen, Jiyu Cao, et al .The association between the C-509T and T869C polymorphisms of TGF-b1 gene and the risk of asthma: A meta-analysis. Hum Immunol 2014, 75(2):141-50.
2. Holgate ST, Davies DE, Powell RM, Howarth PH, Haitchi HM, Holloway JW. Local genetic and environmental factors in asthma disease pathogenesis: chronicity and persistence mechanisms. Eur Respir J 2007; 29(4):793-803.
3. Vercelli D. Discovering susceptibility genes for asthma and allergy. Nat Rev Immunol .2008, 8:169-182.
4. Tattersfield AE, Knox AJ, Britton JR , Hall IP. Asthma. Lancet 2002; 360(9342):1313-22.
5. Huang XZ, Zhuang JH, Ren YG, Zhou LJ, Zhou Q. Association of interleukin-6 and interleukin-18 gene polymorphism with rheumatoid arthritis in Guangdong Han population. Nan Fang Yi Ke Da Xue Xue Bao 2007; 27(11):1661-4.
6. Pawlik A, Kaminski M, Kuśnierczyk P, Kurzawski M, Dziedziejko V, Adamska M, et al. Interleukin-18 promoter polymorphism in patients with atopic asthma. Tissue Antigens 2007; 70(4):314-8.
7. Nakamura K, Okamura H, Wada M, Nagata K, Tamura T. Endotoxin- induced serum factor that stimulates gamma interferon production. Infect Immun 1989; 57(2):590-5.
8. Chang JT, Segal BM, Nakanishi K, Okamura H, Shevach EM. The costimulatory effect of IL-18 on the induction of antigen-specific IFN-gamma production by resting T cells is IL-12 dependent and is mediated by up-regulation of the IL-12 receptor beta2 subunit. Eur J Immunol 2000; 30(4):1113-9.
9. Blumenthal MN, Langefeld CD, Beaty TH, Bleecker ER, Ober C, Lester L, et al. A genome-wide search for allergic response (atopy) genes in three ethnic groups: Collaborative Study on the Genetics of Asthma. Hum Genet 2003; 114(2):157–64.
10. Lachheb J, Chelbi H, Ammar J, Hamzaoui K, Hamzaoui A. Promoter polymorphism of the IL-18 gene is associated with atopic asthma in Tunisian children. Int J Immunogenet 2008; 35(1):63-68.
11. Imboden M, Nicod L, Nieters A, Glaus E, Matyas G, Bircher AJ, et al.The common G-allele of interleukin-18 single-nucleotide polymorphism is a genetic risk factor for atopic asthma. The SAPALDIA Cohort Study. Clin Exp Allergy 2006; 36(2):211-8.
12. Birbian N, Singh J, Jindal SK. Protective role of IL-18 -137G/C polymorphism in a North Indian population with asthma: a pilot study. Cytokine 2013; 61(1):188-93.
13. Wu SQ, Liang W, Wang GL, Li LY, Wang DL, Chen C. Polymorphisms of the IL-18 promoter and bronchial asthma. Mol Med Rep 2012; 6(6):1385-8.
14. Lee CC, Lin WY, Wan L, Tsai Y, Tsai CH, Huang CM, et al. Association of interleukin-18 gene polymorphism with asthma in Chinese patients. J Clin Lab Anal 2008;

22(1):39-44.
15. Shin HD, Kim LH, Park BL, Choi YH, Park HS, Hong SJ, et al. Association of interleukin 18 (IL18) polymorphisms with specific IgE levels to mite allergens among asthmatic patients. Allergy 2005; 60(7):900-6.
16. Heinzmann A, Gerhold K, Ganter K, Kurz T, Schuchmann L, Keitzer R, et al. Association study of polymorphisms within interleukin-18 in juvenile idiopathic arthritis and bronchial asthma. Allergy 2004 59(8):845-9.
17. Shaaban HH, Mohy AM, Abdel-Razek AR, Wahab AA. Interleukin-18 -607C/A gene polymorphism in Egyptian asthmatic children. Mol Diagn Ther 2014; 18(4):427-34.
18. Yi Jingping, CHEN Xiaowen, LI Jianchun,Deng qiong, Association study of interleukin-18 gene polymorphisms and asthma in Jiangxi province. Experimental and Laboratory Medicine 2013, 31(4):309-310.
19. Ying Ma, Bo Zhang, Ren-Kuan Tang, Yun Liu, Guo-Guang Peng. Interleukin-18 promoter polymorphism and asthma risk: a meta-analysis. Mol Biol Rep 2012; 39(2):1371-6.
20. Harada M, Obara K, Hirota T, Yoshimoto T, Hitomi Y, Sakashita Met al. A functional polymorphism in IL-18 is associated with severity of bronchial asthma. Am J Respir Crit Care Med 2009; 180(11):1048-55.
21. Higa S, Hirano T, Mayumi M, Hiraoka M, Ohshima Y, Nambu M, et al. Association between interleukin-18 gene polymorphism 105A/C and asthma. Clin Exp Allergy 2003; 33(8):1097-1102.
22. Aker S, Bantis C, Reis P, Kuhr N, Schwandt C, Grabensee B, et al. Influence of interleukin-6 G-174C gene polymorphism on coronary artery disease, cardiovascular complications and mortality in dialysis patients. Nephrol Dial Transplant 2009; 24(9):2847-2851.
23. Ma Y, Tang RK, Yang X, Peng GG, Liu Y, Wang XM, et al. Lack of an association between interleukin-6 gene promoter polymorphisms (-174G/C, -572G/C) and ischemic heart disease and/or ischemic stroke: a meta-analysis. Hum Immunol 2011; 72(8):641-51.
24. Thakkinstian A, McEvoy M, Minelli C, Gibson P, Hancox B, Duffy D, et al. Systematic review and meta-analysis of the association between {beta}2-adrenoceptor polymorphisms and asthma: a HuGE review. Am J Epidemiol 2005; 162(3):201-11.
25. Higgins JP, Thompson SG. Quantifying heterogeneity in a meta-analysis. Stat Med 2002; 21(11):1539-58.
26. Higgins JP, Thompson SG, Deeks JJ, Altman DG. Measuring inconsistency in meta-analyses. BMJ 2003, 327(7414):557-60.
27. Mantel N, Haenszel W. Statistical aspects of the analysis of data from retrospective studies of disease. J Natl Cancer Inst 1959; 22(4):719-48.
28. Dersimonian R, Laird N. Meta-analysis in clinical trials. Control Clin Trials. 1986; 7(3):177-86.
29. Egger M, Davey Smith G, Schneider M, Minder C. Bias in meta-analysis detected by a simple, graphical test. BMJ 1997; 315(7109):629-34.
30. Begg CB, Mazumdar M. Operating characteristics of a rank correlation test for publication bias. Biometrics 1994; 50(4):1088-101.
31. Tanaka H, Miyazaki N, Oashi K, Teramoto S, Shiratori M, Hashimoto M, et al. IL-18 might reflect disease activity in mild and moderate asthma exacerbation. J Allergy Clin Immunol 2001; 107(2):331-336.
32. El-Mezzein RE, Matsumoto T, Nomiyama H, Miike T. Increased secretion of IL-18 in vitro by peripheral blood mononuclear cells of patients with bronchial asthma and atopic dermatitis. Clin Exp Immunol 2001; 126(2):193-8.