Int J Clin Exp Med 2013;6(8):621-631
www.ijcem.com /ISSN:1940-5901/IJCEM1307012

Original Article

Signal transducer and activator of transcription 6 polymorphism and asthma risk: a meta-analysis

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Received July 17, 2013; Accepted August 13, 2013; Epub September 1, 2013; Published September 15, 2013

Abstract: Background: The polymorphism in the signal transducer and activators of transduction 6 (*STAT6*) gene has been implicated in susceptibility to asthma and aetiology of asthma, but a number of studies have reported inconclusive and ambiguous results of the association between polymorphism in *STAT6* gene and asthma risk in different populations. The aim of this study is to further investigate the association between the *STAT6* gene polymorphism and asthma susceptibility. Methods: Pubmed, EMBASE, China National Knowledge Infrastructure (CNKI) Weipu Database and Wanfang Database were searched to find relevant studies. Odds ratios (ORs) with 95% confidence intervals (CIs) were used to assess the strength of association. Results: Included in this meta analysis were 14 studies involving 2875 cases and 3227 controls for STAT6 2964G/A polymorphism and six studies involving 1431 cases and 2027 controls for 2892C/T polymorphism. Overall, there was no significant association between 2964G/A polymorphism of STAT6 and asthma susceptibility for GA+AA vs. GG (OR = 0.96, 95% CI 0.85-1.07, P = 0.39). Except TT vs. CT+CC and TT vs. CC, no significant association was observed between 2892C/T polymorphism and asthma risk under other different contrast models. However, the result was instable. Conclusions: This meta-analysis suggests that the 2964G/A polymorphism of STAT6 gene is not a risk factor of asthma. For 2892C/T, it contributes to the aetiology of or susceptibility to asthma. More studies are needed to validate this result.

Keywords: Asthma, signal transducer and activator of transcription 6, meta-analysis, polymorphism

Introduction

Asthma is a common complex inflammatory disease characterized by intermittent inflammation of the small airways of the lung with symptoms of wheezing and shortness of breath, affecting hundreds of millions of individuals worldwide [1]. The prevalence of asthma has increased considerably over the past three decades. Currently, it is recognized that both gene-gene as well as gene-environment interactions involving more than 100 susceptibility genes contribute to asthma. A growing number of published studies have focused on the association between genes and asthma, and among these genes, the signal transducer and activators of transduction 6 (*STAT6*) gene have been extensively studied.

STAT6 is an important member of *STAT* family and is crucial for the signaling of many cytokines. It acts as a mediator of allergic inflammation affecting cells that are critical to asthma [2]. It has been showed that *STAT6* plays an important role in Il-13 and Il-4 pathways on *STAT6*$^{-/-}$ mice. Kuperman et al. [3] reported that *STAT6*$^{-/-}$ mice failed to develop airway hyperresponsiveness after allergen provocation, lacked a typical Th2-cytokine response and produced no detectable levels of serum IgE. The result of Kuperman et al. is consistent with Miyata et al. [4] and Akimoto et al. [5], who reported that *STAT6*$^{-/-}$ mice were protected against bronchial eosinophilia and antigen-induced mucus production. These findings suggest that *STAT6* may contribute to the pathogenesis of asthma.

The human *STAT6* gene, containing spans 19 kb of genomic DNA, consists of 23 exons and 22 introns. It is located at 12q13.3-q14.1 where a locus related to serum IgE levels has

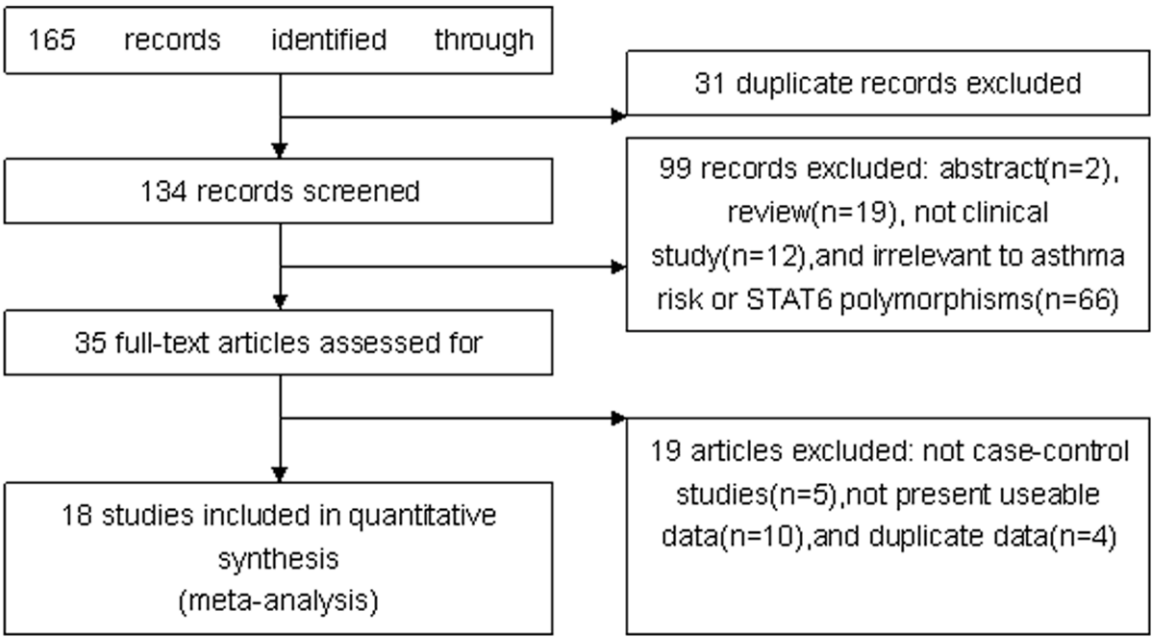

Figure 1. Flow of study identification, inclusion, and exclusion.

been mapped. The 2964G/A (dbSNP rs324015) and 2892C/T (dbSNP rs324011) polymorphism characterized by a single nucleotide alternate variation are located in the 3′ untranslated region and intron region of the *STAT*-6, respectively. Some researchers have studied the association of the polymorphisms in the 3′ untranslated region and intron of the *STAT*-6 gene with asthma [6-11]. However, the results remain controversial.

Considering that these controversial results may be due to the small effect of the polymorphism on asthma risk or the relatively small sample size in each of these published studies, we performed this meta-analysis in an attempt to derive a more precise estimation of associations between 2964G/A and 2892C/T variants of *STAT*6 gene and asthma. To the best of our knowledge, this is the first meta-analysis on the association between *STAT*6 polymorphism and asthma risk or susceptibility.

Methods

Publication search

Pubmed, EMBASE, China National Knowledge Infrastructure (CNKI), Wanfang Database and Weipu Database were all searched with the end-point date on November, 2012. The following terms were used in searching (asthma or asthmatic) and (*STAT*6 or *STAT*-6) and (polymorphism or mutation or variant). No publication date or language restrictions were imposed. All the searched studies were retrieved, and their references were checked as well for other relevant publications. We also searched review articles related to *STAT*6 polymorphism and asthma risk to find additional eligible studies.

Inclusion and exclusion criteria

Studies fulfilling the following selection criteria were included in this meta-analysis: (1) evaluation of the SNP in *STAT*6 gene and asthma risk; (2) using a case-control design where control subjects were unrelated individuals, chosen randomly from the same geographic region; and (3) genotype distributions in both cases and controls were available for estimating an odds ratio (OR) with 95% confidence interval (CI). Studies were excluded if one of the following existed: (1) not relevant to *STAT*6 or asthma; (2) the design was based on sibling pairs; (3) genotype frequencies or numbers were not reported; and (4) reviews and abstracts. For overlapping studies, only the one with the largest sample numbers was included.

Data extraction

Four investigators (Bin Li, Wei Nie, Qiong Li and Hongchao Liu) independently reviewed full manuscripts of eligible studies, and the rele-

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

Author	Year	Country	Ethnicity	Age group	Atopic status	Case (n)	Control (n)	Genotyping method	*STAT6* Polymorphism
Li [15]	2007	China	Asian	Adults	Atopic	95	95	PCR-SSCP	G2964A
Tamura [8]	2003	Japan	Asian	Children	Atopic	73	66	PCR-RFLP	G2964A
Daley [16]	2009	Australia	Caucasian	Adults	NA	617	735	PCR-SSCP	G2964A, C2892T
Murk [17]	2011	USA	Caucasian	Children	Atopic	100	474	PCR-RFLP	G2964A
Pykalainen [10]	2005	Finland	Caucasian	Adults	NA	199	161	PCR-RFLP	G2964A, C2892T
Undarmaa1 [14]	2010	Japan	Asian	Children	Atopic	325	336	PCR-SSCP	G2964A
Undarmaa2 [14]	2010	Japan	Asian	Adults	Atopic	367	676	PCR-RFLP	G2964A
Gao1 [7]	2000	UK	Asian	Adults	Mixed*	400	100	PCR-SSCP	G2964A
Gao2 [7]	2000	UK	Caucasian	Adults	Mixed*	181	119	PCR-SSCP	G2964A
Ding [18]	2010	China	Asian	Adults	NA	108	115	PCR-RFLP	G2964A
Wu [19]	2006	China	Asian	Adults	Atopic	42	42	PCR-SSCP	G2964A
Lin [21]	2012	China	Asian	Children	NA	113	87	PCR-RELP	G2964A
Hu1 [20]	2005	China	Asian	Adults	Atopic	135	109	PCR-RELP	G2964A
Hu2 [22]	2005	China	Asian	Adults	NA	120	112	PCR-RELP	G2964A
Kavalar [11]	2012	Slovenia	Caucasian	Children	NA	154	71	PCR-SSCP	C2892T
Wu2 [23]	2010	China	Asian	Children	NA	252	227	PCR-RFLP	C2892T
Kabesch [24]	2006	Germany	Caucasian	Children	NA	73	773	PCR-SSCP	C2892T
Godava [25]	2012	Czech	Caucasian	Children	Atopic	109	45	PCR-RFLP	C2892T

*Data for atopic or non-atopic asthma patients could be separately extracted. PCR, polymerase chain reaction; RFLP, restriction fragment length polymorphism; NA, not available.

Table 2. Distribution of *STAT6 2964A/G* genotype among patients and controls included in the meta-analysis

Studies	Asthma		Control		HWE
STAT6-G2964A	GG	GA+AA	GG	GA+AA	
Li	35	60	38	57	Yes
Tamura	32	41	22	44	Yes
Delay	362	255	413	322	Yes
Murk	68	32	287	187	Yes
Pykalainen	115	84	89	72	Yes
Undarmaa1	45	280	43	293	Yes
Undarmaa2	50	317	85	591	Yes
Gao1	62	338	9	91	Yes
Gao2	111	70	68	51	Yes
Ding	19	89	18	97	Yes
Wu	16	26	18	24	Yes
Lin	18	95	19	68	Yes
Hu1	20	115	18	91	No
Hu2	15	105	17	95	No

HWE: Hardy-Weinberg equilibrium.

vant data were extracted into designed data collection forms. The accuracy of data was verified by comparing collection forms from each investigator. Any discrepancy was resolved by discussion or another author (Shiyuan Liu) would assess these articles. The following variables were collected from each study: first author's name, year of publication, original country, ethnicity, sample size, genotyping method, atopic status, and genotype numbers in cases and controls.

Statistical analysis

Meta-analysis was performed when data from at least 2 similar studies were available. The strength of the association between the SPNs of *STAT6* in 2964G/A, 2892C/T and asthma risk was measured by OR and 95% CI. The statistical significance of OR was analyzed by Z test, and $P < 0.05$ was considered statistically significant. Due to G2964A variation was expressed by GG *vs.* GA+AA in some studies, we estimated the association risks of dominant model (GA+AA *vs.* GG). For 2892C/T, meta-analyses were performed using (1) allelic contrast model (T *vs.* C), (2) additive model (TT *vs.* CC), (3) recessive (TT *vs.* CC+CT), and (4) dominant model (TT+CT *vs.* CC).

The Chi square-test was used to determine whether observed genotype frequencies conformed to Hardy-Weinberg (H-W) equilibrium. The heterogeneity between the studies was assessed by the Chi square-test based

Table 3. Distribution of *STAT6 2892C/T* genotype among patients and controls included in the meta-analysis

Studies	Asthma			Control			HWE
STAT6-C2892T	CC	CT	TT	CC	CT	TT	
Kavalar	53	75	26	23	37	11	Yes
Wu2	147	91	14	115	100	12	Yes
Pykalainen	63	98	38	59	77	25	Yes
Delay	201	317	126	266	362	122	Yes
Kabesch	28	32	13	306	355	112	Yes
Godava	40	46	23	14	25	6	Yes

HWE: Hardy-Weinberg equilibrium.

Cochrane Q-test. I^2 was also used to test the heterogeneity between the included studies. When P value was greater than 0.10, the pooled OR of each study was calculated by the fixed-effects model; otherwise, the random-effects model was used, as this was more appropriate when there was significant heterogeneity.

To evaluate the ethnic-specific, age-specific and atopic-specific effect and then explore sources of heterogeneity, subgroup analyses were performed by ethnicity, age and the atopic status. Sensitivity analysis was performed by two ways: excluding the studies without showing HWE and sequentially excluding individual study to assess the stability of the results. Asymmetry funnel plots were used to assess potential publication bias. The Begg's test [12] and Egger's test [13] were also used to assess publication bias statistically.

All statistical tests were performed by using the Revman 5.1 software (Nordic Cochrane Center, Copenhagen, Denmark) and STATA 12.0 software (Stata Corporation, College Station, TX).

Results

Literature search and study characteristics

Figure 1 outlines our study selection process. Briefly, a total of 165 articles were identified after an initial search. After removing duplications, 31 articles were excluded. After reading the abstracts, 99 articles were excluded for not getting the full text, review, or clinical study irrelevant to asthma risk or *STAT6* polymorphisms. After reading full texts of the remaining 35 articles, 19 articles were then excluded and 16 remained. Two articles reported two cohorts [7, 14], and each cohort was considered as a separate case-control study. Finally, a total of 18 case-control studies in 16 articles were identified as meeting our inclusion criteria [7, 8, 10, 14-20]. This meta-analysis included 2875 cases and 3227 controls for STAT6 2964G/A and 1431 cases and 2027 controls for 2892C/T. Twelve studies only involve in 2964G/A and four only about 2892C/T; Two studies refer to both polymorphisms. For 2964G/A, there were 10 studies of Asians [7, 8, 14, 15, 18-22] and 4 studies of Caucasians [10, 16, 17]. Ten studies were performed in adults [7, 10, 14-16, 18-20, 22] and four in children [8, 14, 17, 21]. Seven studies [8, 14, 15, 17, 19, 20] included only atopic asthma patients. Others except two studies [7] included both atopic and non-atopic asthma patients but the data for these patients could not be separately extracted. Regarding to 2892C/T, there were 1 study of Asians [23] and 5 studies of Caucasians [10, 11, 16, 24, 25]. Two studies were performed in adults [10, 16] and four in children [11, 23-25]. One study [25] included only atopic asthma patients and other studies included both atopic and non-atopic asthma patients but the data for these patients could be separately extracted [11, 23-25]. The characteristics of each study included in this meta-analysis are presented in **Table 1**. Genotype numbers and HWE examination results are listed in **Tables 2** and **3**.

Quantitative data synthesis

The STAT6 2964G/A polymorphism: As shown in **Table 2**, 14 studies determined the association between 2964A/G polymorphism and asthma [7, 8, 10, 14-18, 20-22]. All studies involved 2964A/G polymorphism were included in pooling. Total sample sizes for asthma and control groups were 2875 and 3227, respectively. We analyzed the heterogeneity of GA+AA vs. GG for all the 14 studies about *STAT6* 2964G/A, and the value of χ^2 was 10.01 with 13 of freedom and P = 0.69 in a random-effects model. In addition, the I^2 value was another index of the test of heterogeneity. For this study, I^2 was 0%, suggesting no significant heterogeneity. Therefore, the fixed-effects model was used for synthesis of the data. The overall OR was 0.95 (95% CI 0.85-1.07) and the Z test for overall effect was 0.81 (P = 0.42) (**Figure 2**) suggesting that asthma risk for individuals of the A allele carriers (GA+AA) did not decrease compared with those with the GG homozygote.

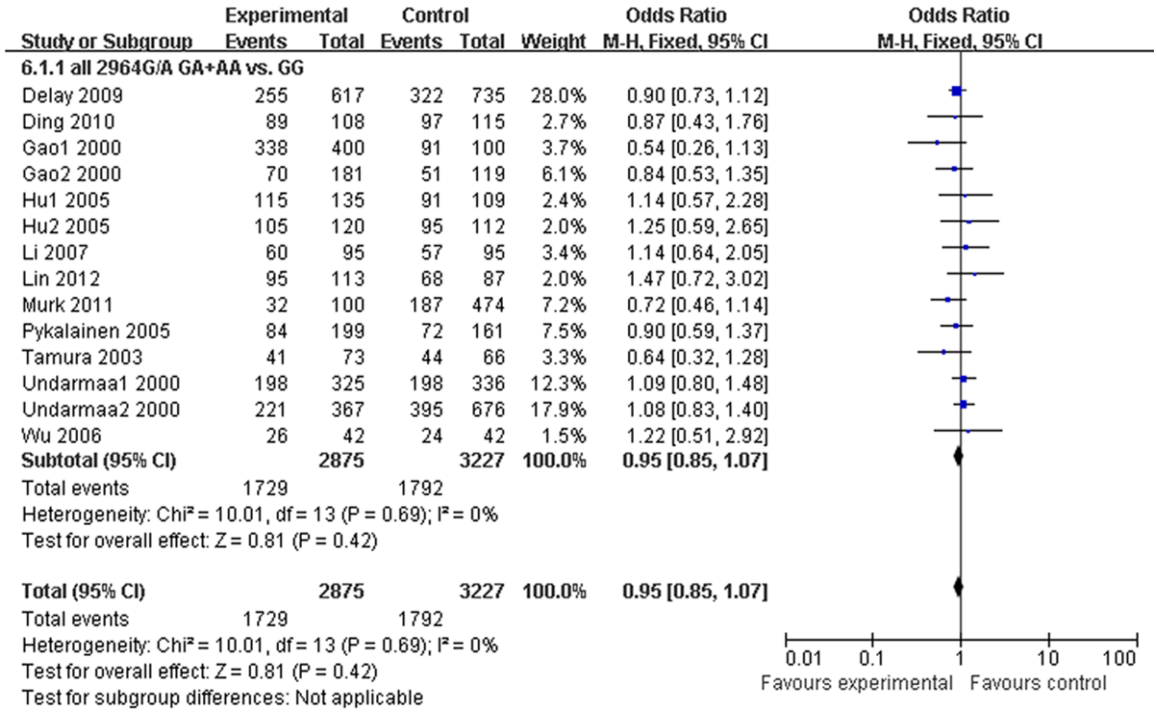

Figure 2. Meta-analysis for the association between asthma risk and the *STAT6* 2964G/A polymorphism (GA+AA vs. GG).

We performed the sensitivity analysis by two means. Firstly, because there were two studies not in HWE, we excluded these two studies [20, 22]. The result was similar (OR = 0.95, 95% CI 0.84-1.06, P = 0.35). Then we sequentially excluded individual studies. Statistically similar results were obtained after sequentially excluding each study, which suggesting stability of the result. In the subgroup analyses by ethnicity, no significant associations were found among Asians (OR 1.04, 95% CI 0.89-1.21, P = 0.66) and among Caucasians (OR 0.87, 95% CI 0.74-1.03, P = 0.10). In the subgroup analyses by age, there were also no significant associations found among adults (OR 0.94, 95% CI 0.82-1.08, P = 0.40) and children (OR 0.93, 95% CI 0.68-1.28, P = 0.67). A negative outcome was observed in atopic asthma patients (OR 0.96, 95% CI 0.87-1.05, P = 0.32). Publication bias was assessed by Begg's funnel plot and Egger's test. The shape of the funnel plot showed symmetric, suggesting there was no publication bias (**Figure 3**). Then, the Egger's test was conducted to provide statistical evidence of funnel plot symmetry. The results indicated an absence of publication bias of the current meta-analysis (t = 0.53, P = 0.606).

The STAT6 2892C/T polymorphism: As shown in **Table 3**, six studies involved 1431 cases and 2027 controls identified the association between 2892C/T polymorphism and asthma [10, 11, 16, 23-25]. Overall, there was no association between asthma and *STAT6* 2892C/T polymorphism using the contrast of alleles (T vs. C) or dominant (TT+CT vs. CC) model. Stratifying subjects by ethnicity indicated an association between the *STAT6* 2892 T allele and asthma in Caucasian (OR = 1.14, 95% CI = 1.02-1.28, p = 0.03) under contrast of alleles (T vs. C) model. There also was an association between the *STAT6* 2892 T allele and asthma in adults (OR = 1.17, 95% CI = 1.02-1.34, p = 0.02) under contrast of alleles (T vs. C) model. Only one study [25] provided data about 2892C/T polymorphism in atopic asthma, so the subgroup analyses of atopic status couldn't be performed. However, there was an association between asthma and the STAT6 2892C/T polymorphism using the recessive (TT vs. CT+CC) (OR = 1.26, 95% CI = 1.02-1.55, p = 0.03) (**Figure 4**, **Table 4**). Furthermore, stratification by ethnicity indicated an association between the *STAT6* 2892 TT genotype and asthma in Caucasian (**Table 4**). Analyses using the additive model (TT vs. CC) revealed associ-

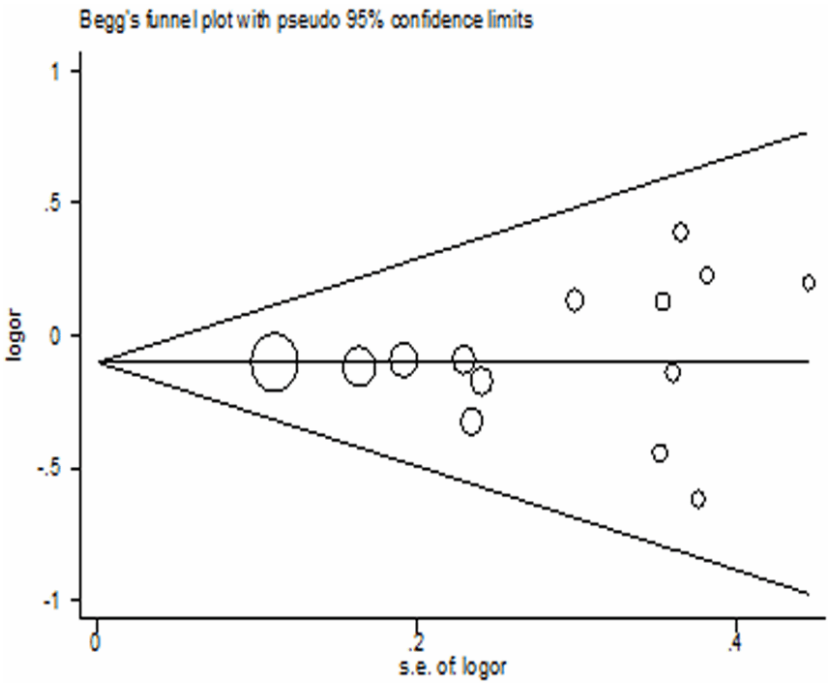

Figure 3. Funnel plot for publication bias in selection of studies on the *STAT6* 2964G/A polymorphism (GA+AA vs. GG).

ations between the *STAT6* 2892 TT genotype and asthma in overall population (OR = 1.29, 95% CI = 1.03-1.63, p = 0.03) (**Figure 5**; **Table 4**), in Caucasian (OR = 1.13, 95% CI = 1.05-1.69, p = 0.02) (**Table 4**) and in adults (OR = 1.38, 95% CI = 1.05-1.82, p = 0.02) (**Table 4**).

While sensitivity analysis was performed by sequentially excluding each study, the results were materially affected for all models. Statistically different results were obtained. After excluding a large-sample study [16], the difference became insignificant statistically. Begg's funnel plot and Egger's test showed no publication bias in this meta-analysis. A summary of the findings concerning the association between the *STAT6* 2892C/T polymorphism and asthma is shown in **Table 4**.

Discussion

Common variants in genes involved in the pathway of pathogenesis may disturb relevant protein function and the predisposition of an individual to disease. By using both the candidate gene and genome-wide screening approaches, multiple loci of the human genome have been implicated in the etiology of asthma and asthma-associated phenotypes. In recent years, the mechanism underlying the regulation of cytokines on the gene transcription, especially signal transducers and activators of transcription (STATs), has been studied extensively. *STAT6* is a characteristic membership of *STAT*s related to allergic asthma and other inflammatory and allergic diseases [2]. It is involved in the interleukin 4 (IL-4) and interleukin 13 (IL-13) signaling pathway [26, 27]. Recent studies suggested that *STAT6* may promote the development of asthma by leveling up IgE in serum and facilitating AHR. For instance, Kuperman et al. [28] reported that *STAT6*$^{-/-}$ mice failed to develop airway hyper-responsiveness after allergen provocation, lacked a typical Th2-cytokine response and produced no detectable levels of serum IgE. Miyata et al [4] and Akimoto et al. [5] reported that *STAT6*$^{-/-}$ mice were protected from bronchial eosinophilia and antigen-induced mucus production. These evidences supported that *STAT6* may play an important role in the pathogenesis of asthma. To our knowledge, this study is the first comprehensive meta-analysis to assess the relationship between the *STAT6* polymorphism (2964G/A and 2892C/T) and asthma susceptibility.

This current meta-analysis including 14 studies with 2875 cases and 3227 controls systematically evaluated the association between 2964G/A polymorphism in the *STAT6* gene and asthma risk. For *STAT6* 2892C/T polymorphism, a total of 6 studies with 1431 cases and 2027 controls were included. The results for 2892C/T polymorphism indicate that TT homozygote carriers had a 26% increase in asthma risk compared with (CC+CT) in recessive model. The subgroup analysis by ethnicity showed that asthma risk was increased in Caucasian (P = 0.03), suggesting that environmental exposures and different genetic backgrounds may be factors influencing the asthma risk. In addi-

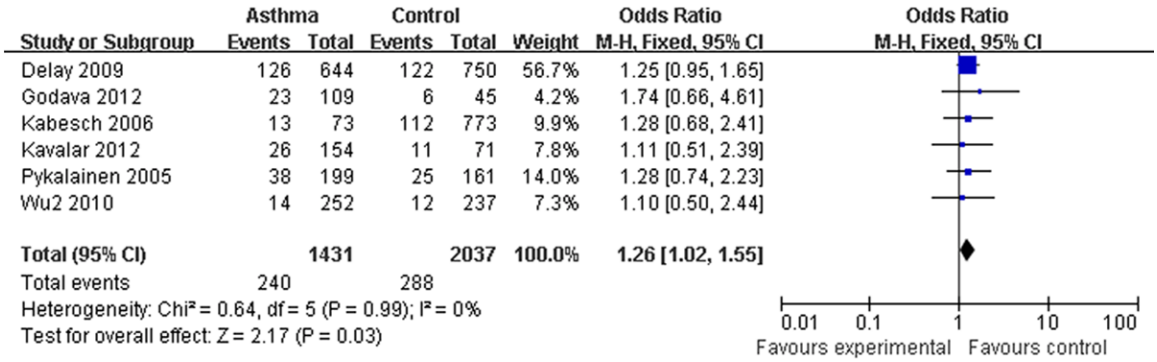

Figure 4. Meta-analysis with a random-effects model for the association between asthma risk and the *STAT6 2892* C/T polymorphism (TT vs. CC+CT).

Table 4. Summary of different comparative results

Polymorphism	Population	No.	Test of association			Test of heterogeneity		
STAT6			OR	95% CI	*p* value	Model	*p* value	I2
2892C/T T vs. C (alleles)	Overall	6	1.09	0.98-1.21	0.12	F	0.42	0%
	Asian	1	NA	NA	NA	NA	NA	NA
	Caucasian	5	1.14	1.02-1.28	**0.03**	F	0.93	0%
	Adult	2	1.17	1.02-1.34	**0.02**	F	0.90	0%
	Children	4	0.95	0.80-1.15	0.62	F	0.61	0%
	Atopic	1	NA	NA	NA	NA	NA	NA
2892C/T TT vs. CC (additive)	Overall	6	1.29	1.03-1.63	**0.03**	F	0.94	0%
	Asian	1	NA	NA	NA	NA	NA	NA
	Caucasian	5	1.13	1.05-1.69	**0.02**	F	0.98	0%
	Adult	2	1.38	1.05-1.82	**0.02**	F	0.91	0%
	Children	4	1.12	0.74-1.69	0.60	F	0.92	0%
	Atopic	1	NA	NA	NA	NA	NA	NA
2892C/T TT vs. CC+CT (recessive)	Overall	6	1.26	1.02-1.55	**0.03**	F	0.99	0%
	Asian	1	NA	NA	NA	NA	NA	NA
	Caucasian	5	1.27	1.02-1.57	**0.03**	F	0.97	0%
	Adult	2	1.26	0.98-1.61	0.07	F	0.94	0%
	Children	4	1.25	0.85-1.83	0.25	F	0.89	0%
	Atopic	1	NA	NA	NA	NA	NA	NA
2892C/T TT+CT vs. CC (dominant)	Overall	6	1.02	0.84-1.24	0.84	R	0.24	26%
	Asian	1	NA	NA	NA	NA	NA	NA
	Caucasian	5	1.14	0.96-1.36	0.13	F	0.72	0%
	Adult	2	1.22	1.00-1.49	0.05	F	0.90	0%
	Children	4	0.87	0.68-1.11	0.27	R	0.82	0%
	Atopic	1	NA	NA	NA	NA	NA	NA
2964A/G GA+AA vs. GG	Overall	14	0.95	0.85-1.07	0.42	F	0.69	0%
	Asian	10	1.06	0.90-1.23	0.49	F	0.68	0%
	Caucasian	4	1.11	0.94-1.31	0.20	F	0.59	0%
	Adult	10	0.96	0.84-1.09	0.52	F	0.81	0%
	Children	4	0.93	0.68-1.28	0.67	F	0.19	37%
	Atopic	9	0.96	0.82-1.11	0.56	F	0.39	5%

F fixed model, R random model, NA not available; the bold values mean that their association is significant.

tion, TT homozygote carriers would have a 29% increase in asthma risk compared with CC homozygote carriers in adults (P = 0.02) in the addictive model, suggesting that age was also

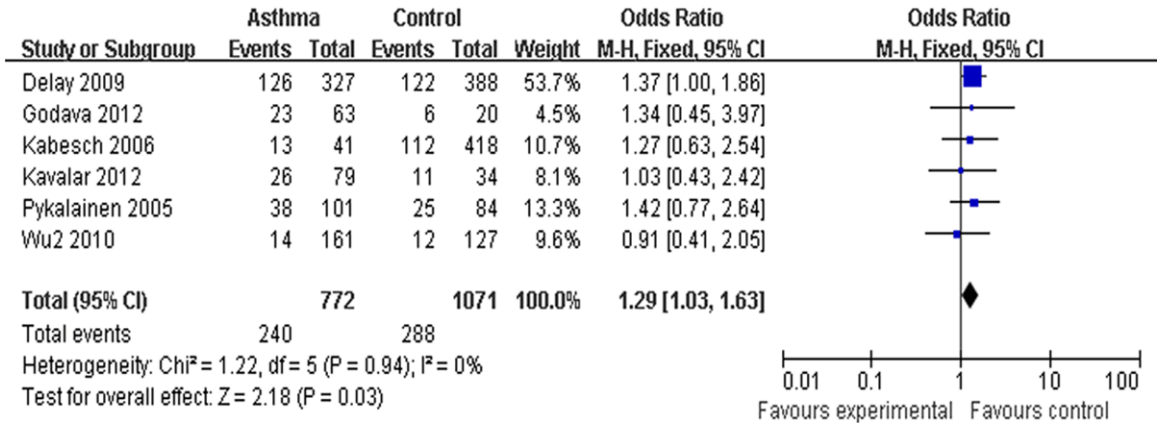

Figure 5. Meta-analysis with a random-effects model for the association between asthma risk and the *STAT6 2892* C/T polymorphism (TT vs. CC).

a possible factor influencing susceptibility to asthma, though no significant association was found in the dominant model and allele comparison. Since only one study offered the data for 2892C/T polymorphism that could be separately extracted in atopic asthma, the subgroup analysis could not be performed and the association between atopic status and asthma could not be ruled out. Additional future studies should be performed by focusing on atopic status with asthma.

From above results we could find that *STAT6* 2892C/T polymorphism may be a conspicuous mild risk factor in the pathogenesis of asthma in the overall study populations. However, this finding showed instability. When all studies meeting our inclusion criteria were included, the statistically significant association between *STAT6* 2892C/T polymorphism and asthma risk in the recessive and additive model could be observed. In sensitivity analysis performed by sequentially excluding each study, it was found that the result was different after excluding the study of Delay et al. [16]. As only six studies were included and the results were instable, it would be prudent to draw the conclusion that the association between *STAT6* 2892C/T polymorphism and asthma risk was definite and determined. Further studies are warranted to validate our finding.

It is difficult to explain the inconsistency or instability of the meta-analysis on the 2892C/T polymorphism. On the one hand, it seems possible that this difference was affected by exposure to various environmental factors [29]. However, no reported article assessed the effect of *STAT6*-environment interactions in different ethnicities. More efforts should be made to analyze these associations in future studies. On the other hand, ethnic difference in the genotype distribution of *STAT6* 2892C/T gene could also lead to this ethnic difference in asthma risk [7]. Furthermore, since the sample size of the study which caused instability of results is the most weighted and only 6 studies were included in this meta-analysis, the positive association between this polymorphism and asthma could not be ruled out. Studies with small sample size may have insufficient statistical power to detect a slight effect, so more large-sample studies are required to assess the association between 2892C/T polymorphism and asthma risk. In the last years, researchers have pointed out that environmental influences and epigenetic mechanisms could pose important modifying factors shaping the phenotype at a clinical level. Methylation and Histone acetylation belong to the important epigenetic mechanisms of gene expression [30]. Gene-gene and gene-environment interactions are involve in the pathogenesis of asthma. The interactions are more often present in patients with asthma or those with increased total IgE levels [9]. It is possible that that effect may be affected by several polymorphisms, rather than one individual polymorphism. This implies that the 2892C/T polymorphism might influence IgE levels but not in a substantial or direct way.

But it is biologically plausible that this polymorphism could influence the tendency, susceptibility and development of asthma, which is supported by the findings of Schedel et al [31, 32],

who demonstrated that 2892C/T polymorphism had an influence on the elevation of total IgE level. Nuclear factor κB (NF- κB) in the *STAT6* second intron sequence and the *STAT6* binding to target sequence, two putative transcription factor binding (TFB) sites, is responsible for inducing IgE production. The two TFB sites are very close to each other in the second intron and the T allele of 2892C/T (rs324011) gives rise to the distal one. In a functional study, Schedel et al confirmed that the T allele of rs324011 increased *STAT6* promoter activity by creating a new site for NF- κB specific binding. They also discovered two new *STAT6* isoforms: *STAT6*d and *STAT6*e. Patients with asthma bearing the TT genotype in rs324011 had a mild, though not significant, increase in the known *STAT6* form, which significantly increased the level of *STAT6*d and *STAT6*e mRNA. However, the role of these two isoforms on IgE production remains unclear. Duetsch et al [6] observed a weakly positive effect of 2892C/T (rs324011) on total IgE (P = 0.0200). The association between polymorphism 2892C/T in *STAT6* and increased IgE levels or atopic asthma was also replicated by Duetsch, Weidinger and Daley et al [6, 16, 33]. Further investigations on gene-gene interaction or interaction of SNPs in genes implicated in asthma or IgE level are needed to analyze associations between the 2892C/T polymorphism and asthma.

The results of our meta-analysis did not show significant associations between this polymorphism and risk of asthma with respect to 2964G/A polymorphism, for this SNP did not reach a significant level; however, the result should be interpreted with caution. The *STAT6* 2964G/A polymorphism in the 3'-UTR was shown to be associated with adult mild asthma in Japanese population but this association was not detected in English people in the study of Gao et al [7]. Similar results were reported by Deutsch et al. [6] and Tamura et al [8]. The functional role of 2964G/A variant remains unknown. It may be in linkage disequilibrium with so far unidentified but functional variants in the regulating or coding parts of *STAT6* or variants of the immediately adjacent genes [8]. Although this present meta-analysis did not show significant associations between 2964G/A polymorphism and risk of asthma, it was unable to thoroughly rule out a positive association between 2964G/A polymorphisms and asthma. This may be related to the fact that the functionality of the polymorphism has not been definitively established or genetic heterogeneity among and within ethnic groups in different studies imposed on the disease.

As the publication of findings often depends on the expectation of researchers, false-negative results may be suppressed or false-positive results magnified. The results of this meta-analysis did not show significant publication bias. Furthermore, there was no significant heterogeneity in overall comparisons for both polymorphisms. Therefore, publication bias and heterogeneity have not influenced the results, suggesting the reliability of our results.

This meta-analysis has some limitations. First, the number of the studies available that could be included in this meta-analysis was not large enough, and therefore the results could be influenced by factors like random error. Second, this study was unable to address gene-gene and gene-environment interactions due to insufficient information extracted from the primary publications. Finally, because only studies that were indexed by the selected databases were included for data analysis, some relevant published studies or unpublished studies with null results were missed, which may have biased our results.

In conclusion, this meta-analysis demonstrated that the 2964G/A polymorphism of STAT6 gene is not a risk factor of asthma. For 2892C/T, it contributes to aetiology or susceptibility to asthma. Future large-scale studies are still needed to validate our findings. Moreover, gene-gene and gene-environment interactions should also be focused on in the future.

Acknowledgements

This study was supported by grant No. 81171333 and No. 81230030 from the National Natural Science Foundation of China.

Disclosure of conflict of interest

None.

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References

[1] Masoli M, Fabian D, Holt S and Beasley R. The global burden of asthma: executive summary of the GINA Dissemination Committee report. Allergy 2004; 59: 469-478.

[2] Chen W and Khurana Hershey GK. Signal transducer and activator of transcription signals in allergic disease. J Allergy Clin Immunol 2007; 119: 529-541.

[3] Kuperman D, Schofield B, Wills-Karp M and Grusby MJ. Signal transducer and activator of transcription factor 6 (Stat6)-deficient mice are protected from antigen-induced airway hyperresponsiveness and mucus production. J Exp Med 1998; 187: 939-948.

[4] Miyata S, Matsuyama T, Kodama T, Nishioka Y, Kuribayashi K, Takeda K, Akira S and Sugita M. STAT6 deficiency in a mouse model of allergen-induced airways inflammation abolishes eosinophilia but induces infiltration of CD8+ T cells. Clin Exp Allergy 1999; 29: 114-123.

[5] Akimoto T, Numata F, Tamura M, Takata Y, Higashida N, Takashi T, Takeda K and Akira S. Abrogation of bronchial eosinophilic inflammation and airway hyperreactivity in signal transducers and activators of transcription (STAT) 6-deficient mice. J Exp Med 1998; 187: 1537-1542.

[6] Duetsch G, Illig T, Loesgen S, Rohde K, Klopp N, Herbon N, Gohlke H, Altmueller J and Wjst M. STAT6 as an asthma candidate gene: polymorphism-screening, association and haplotype analysis in a Caucasian sib-pair study. Hum Mol Genet 2002; 11: 613-621.

[7] Gao PS, Mao XQ, Roberts MH, Arinobu Y, Akaiwa M, Enomoto T, Dake Y, Kawai M, Sasaki S, Hamasaki N, Izuhara K, Shirakawa T and Hopkin JM. Variants of STAT6 (signal transducer and activator of transcription 6) in atopic asthma. J Med Genet 2000; 37: 380-382.

[8] Tamura K, Suzuki M, Arakawa H, Tokuyama K and Morikawa A. Linkage and association studies of STAT6 gene polymorphisms and allergic diseases. Int Arch Allergy Immunol 2003; 131: 33-38.

[9] Shao C, Suzuki Y, Kamada F, Kanno K, Tamari M, Hasegawa K, Aoki Y, Kure S, Yang X, Endo H, Takayanagi R, Nakazawa C, Morikawa T, Morikawa M, Miyabayashi S, Chiba Y, Karahashi M, Saito S, Tamura G, Shirakawa T and Matsubara Y. Linkage and association of childhood asthma with the chromosome 12 genes. J Hum Genet 2004; 49: 115-122.

[10] Pykalainen M, Kinos R, Valkonen S, Rydman P, Kilpelainen M, Laitinen LA, Karjalainen J, Nieminen M, Hurme M, Kere J, Laitinen T and Lahesmaa R. Association analysis of common variants of STAT6, GATA3, and STAT4 to asthma and high serum IgE phenotypes. J Allergy Clin Immunol 2005; 115: 80-87.

[11] Kavalar MS, Balantic M, Silar M, Kosnik M, Korosec P and Rijavec M. Association of ORMDL3, STAT6 and TBXA2R gene polymorphisms with asthma. Int J Immunogenet 2012; 39: 20-25.

[12] Begg CB and Mazumdar M. Operating characteristics of a rank correlation test for publication bias. Biometrics 1994; 50: 1088-1101.

[13] Egger M, Davey Smith G, Schneider M and Minder C. Bias in meta-analysis detected by a simple, graphical test. BMJ 1997; 315: 629-634.

[14] Undarmaa S, Mashimo Y, Hattori S, Shimojo N, Fujita K, Miyatake A, Doi S, Kohno Y, Okamoto Y, Hirota T, Tamari M, Hata A and Suzuki Y. Replication of genetic association studies in asthma and related phenotypes. J Hum Genet 2010; 55: 342-349.

[15] Li Y, Wu B, Xiong H, Zhu C and Zhang L. Polymorphisms of STAT-6, STAT-4 and IFN-gamma genes and the risk of asthma in Chinese population. Respir Med 2007; 101: 1977-1981.

[16] Daley D, Lemire M, Akhabir L, Chan-Yeung M, He JQ, McDonald T, Sandford A, Stefanowicz D, Tripp B, Zamar D, Bosse Y, Ferretti V, Montpetit A, Tessier MC, Becker A, Kozyrskyj AL, Beilby J, McCaskie PA, Musk B, Warrington N, James A, Laprise C, Palmer LJ, Pare PD and Hudson TJ. Analyses of associations with asthma in four asthma population samples from Canada and Australia. Hum Genet 2009; 125: 445-459.

[17] Murk W, Walsh K, Hsu LI, Zhao L, Bracken MB and Dewan AT. Attempted replication of 50 reported asthma risk genes identifies a SNP in RAD50 as associated with childhood atopic asthma. Hum Hered 2011; 71: 97-105.

[18] Ding YP, Wu HH, Xia YH, Lin L and Shi HF. Relationship between STAT6 Gene Polymorphism and Bronchial Asthma in Li Nationality People in Hainan. Chin General Practice 2010; 13: 1765-1767.

[19] Wu B, Li YF, Xiong HY and Zhang L. Case-control study of STAT6 gene polymorphisms for asthma in Chongqing. Immunol J 2006; 22: 308-310.

[20] Hu JH, Wu JM, Cui TP, Li YR and Lou DD. Study ontlle correlation between STAT6 gene polymorphism and atopic asthma in Chinese Hail population of Hubei province. Chin J Micmbiol Immuno 2005; 25: 243-247.

[21] Lin RJ, Liu DX and Sui AH. Relationship between Signal transducers and activators of transcription 6 gene polymorphism and genetic susceptibility of bronchial asthma in children. J Applied Clin Pediatrics 2012; 27: 1258-1260.

[22] Hu JH, Wu JM, Cui TP and Li YR. Correlation of the gene polymorphism at position 2964G/A in 3 untranslated region of STAT6 gene with asthma and serum IgE in Chinese Hail population of Hubei. Chin J Clini Lab Scienc 2005; 23: 9-12.
[23] Wu X, Li Y, Chen Q, Chen F, Cai P, Wang L and Hu L. Association and gene-gene interactions of eight common single-nucleotide polymorphisms with pediatric asthma in middle china. J Asthma 2010; 47: 238-244.
[24] Kabesch M, Schedel M, Carr D, Woitsch B, Fritzsch C, Weiland SK and von Mutius E. IL-4/IL-13 pathway genetics strongly influence serum IgE levels and childhood asthma. J Allergy Clin Immunol 2006; 117: 269-274.
[25] Godava M, Kopriva F, Bohmova J, Vodicka R, Dusek L, Cvanova M, Muzik J, Markova M, Schneiderova E and Vrtel R. Association of STAT6 and ADAM33 single nucleotide polymorphisms with asthma bronchiale and IgE level and its possible epigenetic background. Biomed Pap Med Fac Univ Palacky Olomouc Czech Repub 2012; 156: 236-247.
[26] McKenzie AN. Regulation of T helper type 2 cell immunity by interleukin-4 and interleukin-13. Pharmacol Ther 2000; 88: 143-151.
[27] Takeda K, Kamanaka M, Tanaka T, Kishimoto T and Akira S. Impaired IL-13-mediated functions of macrophages in STAT6-deficient mice. J Immunol 1996; 157: 3220-3222.
[28] Kuperman DA and Schleimer RP. Interleukin-4, interleukin-13, signal transducer and activator of transcription factor 6, and allergic asthma. Curr Mol Med 2008; 8: 384-392.
[29] Zhang G, Candelaria P, Khoo S, Hayden CM, Makela M, Goldblatt J, Haahtela T and LeSouef PN. Eastern vs. western environments: Different genetic profiles associated with asthma and allergy. Am J Respir Crit Care Med 2010; 181.
[30] Kim EG, Shin HJ, Lee CG, Park HY, Kim YK, Park HW, Cho SH, Min KU, Cho ML, Park SH and Lee CW. DNA methylation and not allelic variation regulates STAT6 expression in human T cells. Clin Exp Med 2010; 10: 143-152.
[31] Schedel M, Carr D, Woitsch B, Schmid I, Fritzsch C, Weiland SK, Von Mutius E and Kabesch M. Polymorphisms in the IL-4/IL-13 pathway influence IgE regulation and the development of asthma. Allergologie 2005; 28: 307-314.
[32] Schedel M, Frei R, Bieli C, Cameron L, Adamski J, Lauener R and Kabesch M. An IgE-associated polymorphism in STAT6 alters NF-(kappa)B binding, STAT6 promoter activity, and mRNA expression. J Allergy Clin Immunol 2009; 124: 583-9, 589.e1-6.
[33] Weidinger S, Klopp N, Wagenpfeil S, Rummler L, Schedel M, Kabesch M, Schafer T, Darsow U, Jakob T, Behrendt H, Wichmann HE, Ring J and Illig T. Association of a STAT 6 haplotype with elevated serum IgE levels in a population based cohort of white adults. J Med Genet 2004; 41: 658-663.