Association of TLR4 (896A/G and 1196C/T) Gene Polymorphisms with Asthma Risk: A Meta-Analysis

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Background: Conflicting data have been reported on the association between Toll-like receptor 4 (TLR4) +896A/G and +1196C/T polymorphisms and the risk of asthma. Therefore, we conducted this meta-analysis to clarify the effect of TLR4 +896A/G and +1196C/T polymorphisms on the risk of asthma.

Material/Methods: An electronic literature search was performed using PubMed, Embase, Web of Science, Chinese National Knowledge Infrastructure (CNKI), and Wanfang Data to find relevant studies. Odds ratios (ORs) and 95% confidence intervals (CIs) were used to assess the strength of the associations. All statistical analyses were conducted using STATA software version 12.0.

Results: A total of 14 studies with 2873 asthma cases and 3110 controls were included. The pooled results indicated a significant association between TLR4 +1196C/T polymorphism and the risk of asthma (T vs. C: OR=0.79, 95%CI=0.63–0.99, P=0.04; TT+CT vs. CC: OR=0.76, 95%CI=0.59–0.96, P=0.03; CT vs. CC: OR=0.74, 95%CI=0.58–0.95, P=0.02). In subgroup analysis by ethnicity, TLR4 +1196C/T polymorphism was significantly associated with asthma risk in Asians (T vs. C: OR=0.73, 95%CI=0.54–0.98, P=0.04; TT+CT vs. CC: OR=0.70, 95%CI=0.51–0.96, P=0.03; CT vs. CC: OR=0.69, 95%CI=0.50–0.96, P=0.03), but not in whites. For TLR4 +896A/G polymorphism, no significant association was found between TLR4 +896A/G polymorphism and asthma risk under any genetic models.

Conclusions: The results of this meta-analysis suggest that T allele of the TLR4 +1196C/T might act as a protective factor against the development of asthma.

MeSH Keywords: Asthma • Meta-Analysis • Polymorphism, Genetic • Toll-Like Receptor 4

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META-ANALYSIS

Background

Asthma is a chronic inflammatory airway disorder characterized by airway hyper-responsiveness, reversible airway obstruction, and airway remodeling [1,2]. It is commonly thought that asthma is a multi-factorial disease resulting from complex interactions between genetic predisposition and environmental factors [3]. Studies have shown that genetic influences on asthma are substantial, with heritability estimates ranging between 35% and 95% [4]. Notably, a large number of polymorphisms in genes positioned throughout the genome have been implicated in asthma causation, including the gene encoding Toll-like receptor 4 (TLR4) [5].

Toll-like receptors (TLRs) are a class of evolutionarily conserved membrane-bound pattern recognition receptors (PRRs) presented on the cell surface of innate immune cells. TLRs recognize pathogen-associated molecular patterns (PAMPs) exclusively expressed by microbial pathogens, and activate cellular signaling pathways to induce immune-response genes, including inflammatory cytokines [6]. Ten different human TLRs have been identified. TLR4, the best-studied TLR, is expressed on macrophages, dendritic cells, and other cell types, and mainly recognizes lipopolysaccharide (LPS) of gram-negative bacteria [7,8]. Interaction of LPS with TLR4 can activate the nuclear factor kappa B (NF-kB) signaling pathway, increase expression of inflammatory cytokines and disturb the Th1/Th2 balance in asthma [9]. TLR4 also regulates innate immune responses to respiratory syncytial virus infection, which is a risk factor for the development of asthma [10].

Human TLR4 gene is located on chromosome 9q32-q33 [11]. Two single-nucleotide polymorphisms (SNPs), TLR4 +896A/G (rs4986790, also known as Asp299Gly) and TLR4 +1196C/T (rs4986791, also known as Thr399Ile), have been demonstrated to modify the receptor’s response to endotoxin, which is an important trigger of asthma [12]. This genetically determined alteration in endotoxin responsiveness may be involved in the development of asthma. Several studies have evaluated the association of TLR4 +896A/G and +1196C/T polymorphisms with asthma [13–31]. However, due to the limitation of subjects, the results were inconsistent and controversial. A previous meta-analysis has shown a marginal association of TLR4 +896A/G with asthma, and no association between TLR4 +1196C/T polymorphism and asthma has not been entirely established. Therefore, we performed a meta-analysis including all eligible case-control studies to clarify and quantify the authentic effect of TLR4 +896A/G and +1196C/T polymorphisms on the risk of asthma.

Material and Methods

This study was conducted according to the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines.

Literature search strategy

A literature search was performed using PubMed, Embase, Web of Science, Chinese National Knowledge Infrastructure (CNKI), and Wanfang Data from inception to July 2015. The search strings was: (“asthma” or “asthmatic”) and (“toll-like receptor 4” or “TLR4”) and (“mutation” or “polymorphism” or “variant”). The reference lists of the identified articles were also examined. No restrictions were applied for language, population, sample size, publication date, or type of report.

Inclusion and exclusion criteria

Studies were included in the meta-analysis that met the following criteria: 1) case-control study design, 2) evaluation of the association between the TLR4 +896A/G (Asp299Gly) and +1196C/T (Thr399Ile) polymorphisms and risk of asthma, 3) genotype frequency was available or sufficient data could be extracted to calculate odds ratios (ORs) and 95% confidence intervals (CIs), and 4) not animal studies. For overlapping studies, the most recent article or the one with the largest sample size was selected. Studies were excluded if they did not meet these inclusion criteria. Unpublished data were not considered.

Data extraction

The following data were extracted: year of publication, first author, country, ethnicity, age, atopic status, detection methods, sample size, and genotype frequencies in cases and controls. Two investigators independently extracted data. Disagreements were resolved by discussion and consensus.

Statistical analysis

All the statistical analyses were conducted using STATA software version 12.0 (STATA Corporation, College Station, TX, USA). We first evaluated Hardy-Weinberg equilibrium (HWE) in the control group for each study using the chi-square test, and it was considered statistically significant when P<0.05. A statistical test was performed based on the Q statistic to assess heterogeneity. The P>0.10 of the Q test indicated a lack of heterogeneity among studies. If heterogeneity was observed among studies, the random-effects model was used. Otherwise, the fixed-effects model was adopted. The strength of association between TLR4 +896A/G and +1196C/T polymorphisms and the risk of asthma was assessed by calculating ORs with 95% CIs. Stratified analysis was performed by ethnicity, if possible.
The main results of the relationship between TLR4 +896A/G and +1196C/T polymorphisms and asthma risk are listed in Table 3. Overall, no significant association between TLR4 +896A/G polymorphism and asthma risk was found in the allelic model (G vs. A: OR=1.05, 95% CI=0.90–1.23, P=0.51), the dominant model (GG + AG vs. AA: OR=1.05, 95% CI=0.89–1.24, P=0.56), or the codominant models (AG vs. AA: OR=1.04, 95% CI=0.88–1.23, P=0.64). After categorizing subjects into different subgroups on the basis of ethnicity, the results remained non-significant (Figure 2). The recessive model (GG vs. AG+AA) and codominant model (GG vs. AA) were not performed due to the low frequency of the GG genotype in cases and controls.

As for TLR4 +1196C/T polymorphism, a protective association was found between TLR4 +1196C/T polymorphism and asthma in the allele model (T vs. C: OR=0.79, 95% CI=0.63–0.99, P=0.04), the dominant model (TT+CT vs. CC: OR=0.76, 95% CI=0.59–0.96, P=0.03), and the codominant model (CT vs. CC: OR=0.74, 95% CI=0.58–0.95, P=0.02). This association was not examined via the recessive model (TT vs. CT+CC) due to the low frequency of the TT genotype in cases and controls. The subgroup analysis by ethnicity showed that the T allele of TLR4 +1196C/T polymorphism was a significant protective gene for the development of asthma in Asians (T vs. C: OR=0.95, 95% CI=0.70–1.29, P=0.00; TT+CT vs. CC: OR=0.70, 95% CI=0.51–0.96, P<0.03; CT vs. CC: OR=0.69, 95% CI=0.50–0.96, P=0.03), but there was no statistically significant difference in whites (T vs. C: OR=0.87, 95% CI=0.65–1.17, P=0.29; TT+CT vs. CC: OR=0.87, 95% CI=0.57–1.32, P=0.39; CT vs. CC: OR=0.83, 95% CI=0.56–1.22, P=0.33) (Figures 3–5).

Test of heterogeneity, sensitivity analysis, and publication bias

There was no significant heterogeneity between any studies when analyzing the association of TLR4 +896A/G and +1196C/T polymorphisms and asthma risk in all genetic models, so we used fixed-effects models. Sensitivity analyses were conducted by altering the statistical models. No material alterations were detected, indicating that our results were statistically robust. Exclusion of the HWE-deviated studies did not meaningfully change the pooled estimates (data not shown). The funnel plot was used to evaluate publication bias, and there was no obvious asymmetry. Furthermore, no significant publication bias was detected by Begg’s test and Egger’s test (all P>0.05).

Discussion

Asthma is a chronic inflammatory airway disorder with complex etiologies involving both genetic and environmental factors. The association between TLR4 +896A/G and +1196C/T polymorphisms and asthma risk suggests a potential role of these genetic variants in the pathogenesis of asthma. Further research is needed to elucidate the underlying mechanisms and to identify potential therapeutic targets for asthma treatment.
conclusions. Several candidate genes, such as TLR4, CD14, STAT6, ADAM33, and IL-13, have been reported to be associated with asthma susceptibility [18,33–36]. TLR4 is a principal receptor for LPS. Recognition of LPS by TLR4 plays a crucial role in the activation of subsequent immune and inflammatory responses against invaders. Numerous studies have indicated the role of TLR4 in the pathogenesis of asthma [37–40]. TLR4 is up-regulated in patients with asthma/allergic rhinitis [37]. Bortolatto et al. demonstrated that LPS impair the development of Th2 immunity, signaling via TLR4 and MyD88 molecules and via the IL-12/IFN-γ axis, and the synthetic TLR4 agonists protect against allergic asthma development [41].

Genetic polymorphisms of TLR4 gene have been demonstrated to be associated with diminished airway responsiveness to inhaled LPS [42], and to be closely involved in the susceptibility to many diseases, including asthma, juvenile spondyloarthritis, inflammatory bowel disease, and systemic lupus erythematosus [18,43–45].

Two co-segregating single-nucleotide polymorphisms, +896 A/G and +1196 C/T, in human TLR4 gene that result in amino acid changes in the extracellular domain of the TLR4 protein have been widely studied. Environmental endotoxins are important triggers of asthma. These 2 variants have been reported to be associated with asthma risk. The characteristics of the studies included in the meta-analysis are shown in Table 1.

| Study     | Year | Country | Ethnicity | Age | Atopic status | Cases | Controls | Genotyping method |
|-----------|------|---------|-----------|-----|---------------|-------|----------|-------------------|
| +896A/G   |      |         |           |     |               |       |          |                   |
| Yang      | 2004 | UK      | Caucasian | Adults | Mixed       | 185   | 179      | ARMS-PCR          |
| Adjers    | 2005 | Finland | Caucasian | Adults | Mixed       | 243   | 401      | TaqMan            |
| Liu       | 2005 | China   | Asian     | Mixed | Atopic       | 197   | 156      | PCR-RFLP          |
| Larocca   | 2006 | Venezuela | American | Mixed | Mixed       | 100   | 100      | PCR-RFLP          |
| Smit      | 2007 | Denmark | Caucasian | Adults | Mixed       | 100   | 87       | TaqMan            |
| Smit      | 2009 | Denmark | Caucasian | Adults | Mixed       | 100   | 87       | PCR-SSP           |
| Voronko   | 2011 | Russia  | Caucasian | Mixed | Atopic       | 283   | 227      | MALDI-TOF-MS      |
| Zaborowski| 2011 | Poland  | Caucasian | Adults | Mixed       | 106   | 159      | PCR-RFLP          |
| Hussein   | 2012 | Egypt   | Caucasian | Children | Mixed | 500 | 251      | ARMS-PCR          |
| Sahin     | 2014 | Turkey  | Caucasian | Adults | Mixed       | 131   | 75       | RT-PCR            |
| Sinha     | 2014 | India   | Asian     | Mixed | Mixed       | 481   | 483      | PCR-RFLP          |
| Bahrami   | 2015 | Iran    | Caucasian | Adults | Mixed       | 99    | 120      | PCR-RFLP          |
| +1196C/T  |      |         |           |     |               |       |          |                   |
| Liu       | 2005 | China   | Asian     | Mixed | Atopic       | 197   | 156      | PCR-RFLP          |
| Larocca   | 2006 | Venezuela | American | Mixed | Mixed       | 100   | 100      | PCR-RFLP          |
| Smit      | 2007 | Denmark | Caucasian | Adults | Mixed       | 100   | 87       | PCR-SSP           |
| Lachheb  | 2008 | Tunisia | Caucasian | Children | Mixed | 210 | 224      | PCR-RFLP          |
| Smit      | 2009 | France  | Caucasian | Adults | Mixed       | 224   | 568      | Taqman and Illumina Golden Gate assays |
| Sahin     | 2014 | Turkey  | Caucasian | Adults | Mixed       | 131   | 75       | RT-PCR            |
| Sinha     | 2014 | India   | Asian     | Mixed | Mixed       | 481   | 483      | PCR-RFLP          |
to be associated with a blunted response to inhaled endotoxin on bronchial challenge testing and a reduced systemic inflammatory response to low-dose inhaled endotoxin [42,46]. Non-carriers of these polymorphisms have been found to be more frequently affected by asthma [12]. Several case-control studies have evaluated the association of TLR4 +896A/G and +1196C/T polymorphisms with asthma susceptibility. However, the results remain controversial. A previous meta-analysis has shown a marginal association of TLR4 +896A/G with asthma, and no association between TLR4 +1196C/T polymorphism and asthma [32]. However, the previous meta-analysis did not cover all eligible studies related to asthma. Therefore, to obtain a more precise conclusion we conducted this meta-analysis including all eligible case-control studies.

On the basis of 7 case-control studies including 1443 asthma cases and 1693 controls, the present meta-analysis found that TLR4 +1196C/T polymorphism might be a protective factor against the development of asthma. This result is different from the previous meta-analysis. The discrepancy stemmed from the fact that Tizaoui and coworkers only included 4 studies with 541 asthma cases and 486 controls. Next, we conducted the stratified analysis by ethnicity. A significant protective association between TLR4 +1196C/T polymorphism and asthma was detected in Asians. However, we found no significant relationship between TLR4 +1196C/T polymorphism and asthma in whites. These results suggest that the effect of TLR4 +1196C/T polymorphism on asthma risk might be influenced by ethnicity. More studies should be performed based on different ethnic groups.

### Table 2. Distribution of TLR4 +896A/G and +1196C/T polymorphisms among patients and controls.

| Study     | Cases | Controls | HWE(P) |
|-----------|-------|----------|--------|
|           | +896A/G |          |        |
|           | AA     | AG       | GG     | AA     | AG     | GG     |
| Yang      | 155    | 30       | 0      | 159    | 19     | 1      | 0.603  |
| Adjers    | 202    | 39       | 2      | 334    | 64     | 3      | 0.973  |
| Liu       | 161    | 29       | 7      | 128    | 23     | 5      | 0.006  |
| Larocca   | 91     | 9        | 0      | 92     | 8      | 0      | 0.677  |
| Smit      | 91     | 8        | 1      | 78     | 9      | 0      | 0.611  |
| Carvalho  | 12     | 2        | 0      | 70     | 10     | 0      | 0.551  |
| Lachheb   | 209    | 1        | 0      | 223    | 0      | 1      | < 0.01 |
| Voronko   | 245    | 31       | 7      | 200    | 27     | 0      | 0.341  |
| Zaborowski| 94     | 12       | 0      | 142    | 17     | 0      | 0.476  |
| Hussein   | 434    | 64       | 2      | 223    | 27     | 1      | 0.851  |
| Sahin     | 122    | 9        | 0      | 71     | 4      | 0      | 0.812  |
| Sinha     | 390    | 87       | 4      | 381    | 95     | 7      | 0.699  |
| Bahrami   | 85     | 14       | 0      | 104    | 16     | 0      | 0.434  |
|           | +1196C/T |        |        |
|           | CC     | CT       | TT     | CC     | CT     | TT     |
| Liu       | 191    | 4        | 2      | 150    | 5      | 1      | < 0.01 |
| Larocca   | 95     | 4        | 1      | 94     | 6      | 0      | 0.757  |
| Smit      | 93     | 6        | 1      | 77     | 10     | 0      | 0.570  |
| Lachheb   | 209    | 0        | 1      | 221    | 2      | 1      | < 0.01 |
| Smit      | 198    | 26       | 0      | 492    | 76     | 0      | 0.087  |
| Sahin     | 120    | 11       | 0      | 71     | 4      | 0      | 0.812  |
| Sinha     | 408    | 68       | 5      | 384    | 92     | 7      | 0.581  |

HWE – Hardy-Weinberg equilibrium.
### Table 3. Summary ORs and 95% CI of TLR4 +896A/G and +1196C/T polymorphisms and asthma risk.

| Subgroup | Genetic model | Genotype/Allele | Type of model | Heterogeneity | Test of association |
|----------|---------------|----------------|---------------|---------------|---------------------|
|          |               |                |               | I² | P   | P      | OR   | 95%CI | P   |
| +896A/G  | Dominant model | GG+AG vs. AA   | Fixed         | 0.0% | 0.99 | 1.05   | 0.89–1.24 | 0.56 |
| Overall  | Codominant model | AG vs. AA     | Fixed         | 0.0% | 0.97 | 1.04   | 0.88–1.23 | 0.64 |
|          | Allele model | G vs. A        | Fixed         | 0.0% | 0.97 | 1.05   | 0.89–1.24 | 0.56 |
| Caucasians | Dominant model | GG+AG vs. AA   | Fixed         | 0.0% | 0.96 | 1.12   | 0.90–1.39 | 0.32 |
|          | Codominant model | AG vs. AA     | Fixed         | 0.0% | 0.97 | 1.04   | 0.88–1.23 | 0.64 |
|          | Allele model | G vs. A        | Fixed         | 0.0% | 0.97 | 1.05   | 0.89–1.24 | 0.56 |
| Asians   | Dominant model | GG+AG vs. AA   | Fixed         | 0.0% | 0.62 | 0.91   | 0.69–1.19 | 0.48 |
|          | Allele model | G vs. A        | Fixed         | 0.0% | 0.52 | 0.91   | 0.71–1.16 | 0.43 |
| +1196C/T | Dominant model | TT+CT vs. CC   | Fixed         | 0.0% | 0.83 | 0.76   | 0.59–0.96 | 0.03 |
| Overall  | Codominant model | CT vs. CC     | Fixed         | 0.0% | 0.75 | 0.74   | 0.58–0.95 | 0.02 |
|          | Allele model | T vs. C        | Fixed         | 0.0% | 0.87 | 0.79   | 0.63–0.99 | 0.04 |
| Caucasians | Dominant model | TT+CT vs. CC   | Fixed         | 0.0% | 0.52 | 0.84   | 0.57–1.24 | 0.39 |
|          | Codominant model | CT vs. CC     | Fixed         | 0.0% | 0.40 | 0.82   | 0.55–1.22 | 0.33 |
|          | Allele model | T vs. C        | Fixed         | 0.0% | 0.65 | 0.87   | 0.60–1.26 | 0.46 |
| Asians   | Dominant model | TT+CT vs. CC   | Fixed         | 0.0% | 0.84 | 0.70   | 0.51–0.96 | 0.03 |
|          | Allele model | T vs. C        | Fixed         | 0.0% | 0.67 | 0.73   | 0.54–0.98 | 0.04 |

| Study ID | OR (95% CI) | % weight |
|----------|-------------|----------|
| Caucasians |             |          |
| Yang et al. (2004) | 1.54 (0.84, 2.82) | 6.38 |
| Adlers et al. (2005) | 1.01 (0.66, 1.55) | 15.75 |
| Smit et al. (2007) | 0.86 (0.32, 2.27) | 3.28 |
| Carvalho et al. (2008) | 1.17 (0.31, 4.00) | 0.96 |
| Lachheb et al. (2008) | 1.07 (0.07, 17.17) | 0.36 |
| Voronko et al. (2011) | 1.15 (0.68, 1.95) | 9.72 |
| Zaborowska et al. (2011) | 1.07 (0.49, 2.33) | 4.52 |
| Hussein et al. (2012) | 1.21 (0.76, 1.94) | 12.12 |
| Sahin et al. (2014) | 1.31 (0.39, 4.41) | 1.77 |
| Bahrami et al. (2015) | 1.07 (0.49, 2.32) | 4.65 |
| Subtotal (I²=0.0%, p=0.995) | 1.14 (0.92, 1.41) | 59.51 |
| Asians |             |          |
| Liu et al. (2005) | 1.02 (0.59, 1.74) | 9.57 |
| Sinha et al. (2014) | 0.87 (0.64, 1.20) | 30.92 |
| Subtotal (I²=0.0%, p=0.620) | 0.91 (0.69, 1.19) | 40.49 |
| Overall (I²=0.0%, p=0.979) | 1.05 (0.89, 1.24) | 100.00 |

**Figure 2.** Forest plot of the association between TLR4 +896A/G polymorphism and asthma risk by ethnicity stratification under the dominant model (GG+AG vs. AA).
Figure 3. Forest plot of the TLR4 +1196C/T polymorphism associated with asthma risk by ethnicity stratification under the allele model (T vs. C).

Study ID | OR (95% CI) | % weight
---|---|---
**Caucasians**
Smit et al. (2007) | 0.68 (0.26, 1.77) | 6.25
Lachheb et al. (2008) | 0.33 (0.10, 1.04) | 2.34
Smit et al. (2009) | 0.86 (0.54, 1.36) | 24.64
Sahin et al. (2014) | 1.60 (0.50, 5.11) | 2.97
Subtotal (I-squared=0.0%, p=0.653) | 0.87 (0.60, 1.26) | 36.20

**Asians**
Liu et al. (2005) | 0.90 (0.32, 2.52) | 4.66
Sinha et al. (2014) | 0.72 (0.33, 0.97) | 59.15
Subtotal (I-squared=0.0%, p=0.671) | 0.73 (0.54, 0.98) | 63.80

Overall (I-squared=0.0%, p=0.808) | 0.78 (0.62, 0.98) | 100.00

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Figure 4. Forest plot of the TLR4 +1196C/T polymorphism associated with asthma risk by ethnicity stratification under the dominant model (TT+CT vs. CC).

Study ID | OR (95% CI) | % weight
---|---|---
**Caucasians**
Smit et al. (2007) | 0.58 (0.21, 1.59) | 6.82
Lachheb et al. (2008) | 0.35 (0.04, 3.42) | 1.98
Smit et al. (2009) | 0.85 (0.53, 1.37) | 26.06
Sahin et al. (2014) | 1.63 (0.50, 5.30) | 3.20
Subtotal (I-squared=0.0%, p=0.516) | 0.84 (0.57, 1.24) | 38.07

**Asians**
Liu et al. (2005) | 0.79 (0.25, 2.48) | 4.45
Sinha et al. (2014) | 0.69 (0.50, 0.97) | 57.48
Subtotal (I-squared=0.0%, p=0.671) | 0.70 (0.51, 0.96) | 61.93

Overall (I-squared=0.0%, p=0.730) | 0.75 (0.59, 0.97) | 100.00

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Figure 5. Forest plot of the TLR4 +1196C/T polymorphism associated with asthma risk by ethnicity stratification under the codominant model (CT vs. CC).

Study ID | OR (95% CI) | % weight
---|---|---
**Caucasians**
Smit et al. (2007) | 0.50 (0.17, 1.43) | 7.17
Lachheb et al. (2008) | 0.21 (0.01, 4.43) | 1.73
Smit et al. (2009) | 0.85 (0.53, 1.37) | 27.36
Sahin et al. (2014) | 1.63 (0.50, 5.30) | 3.34
Subtotal (I-squared=0.0%, p=0.416) | 0.82 (0.55, 1.22) | 39.51

**Asians**
Liu et al. (2005) | 0.63 (0.17, 2.38) | 3.92
Sinha et al. (2014) | 0.70 (0.49, 0.98) | 56.57
Subtotal (I-squared=0.0%, p=0.485) | 0.69 (0.50, 0.96) | 60.49

Overall (I-squared=0.0%, p=0.631) | 0.74 (0.58, 0.96) | 100.00

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For TLR4 +896A/G polymorphism, no significant correlation was observed between TLR4 +896A/G polymorphism and asthma risk. The results remained non-significant after subgroup analysis by ethnicity. Our results are consistent with previous meta-analyses [32,47,48]. Therefore, TLR4 +896A/G polymorphism seemed not to be associated with the risk of asthma development. However, a study conducted in Turkish children with asthma observed that both TLR4 +896A/G and +1196C/T polymorphisms were statistically more frequent in the mild asthma group [49]. A strong association between TLR4 +896A/G and asthma course has been found in a Russian study, which reported that the minor G allele was associated with moderate/severe asthma [26]. In addition, the G allele has been suggested to be significantly associated with moderate-severe asthma compared to mild asthma in an Egyptian population study [27]. These findings indicate that TLR4 +896A/G polymorphism might be associated with the severity of asthma, but not susceptibility to asthma.

Several limitations in this study should be addressed. Firstly, the number of studies and subjects included in the present meta-analysis were relatively small. Secondly, only published studies with sufficient data were included, so the possibility of publication bias cannot be completely ruled out, even though funnel plot and Egger’s test did not detect publication bias. Thirdly, the frequencies of GG genotype and TT genotype were low, which may undermine the findings. Moreover, the potential interactions between gene-gene and gene-environment during development of asthma were not conducted due to a lack of original data. Considering these limitations, the results of the meta-analysis should be interpreted with caution. Well-designed case-control studies with larger sample sizes and different population characteristics are needed to confirm these results.

Conclusions

In summary, this meta-analysis suggests that the T allele of the TLR4 +1196C/T might act as a protective factor against the development of asthma, but there was no significant association between TLR4 +896A/G polymorphism and risk of asthma. Larger well-designed studies based on different ethnic groups should be performed to confirm our findings.

Conflicts of interest

None of the authors have any conflicts of interest to declare.

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