Separating the effects of childhood- and adult-onset asthma on rheumatoid arthritis and systemic lupus erythematosus: a Mendelian randomization study

Guiwu Huang  
The First Affiliated Hospital of Sun Yat-sen University, Sun Yat-sen University

Yonglie Zhong  
The First Affiliated Hospital of Sun Yat-sen University, Sun Yat-sen University

Weiming Liao  
The First Affiliated Hospital of Sun Yat-sen University, Sun Yat-sen University

Xiaoyi Zhao (✉️ zhaoxy38@mail.sysu.edu.cn)  
The First Affiliated Hospital of Sun Yat-sen University, Sun Yat-sen University

Research Article

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Abstract

**Background:** The causal relationship between asthma and rheumatoid arthritis (RA) has not been well investigated from a perspective of genetics. This study investigated the effects of asthma appearing at different ages on the risk of RA and systemic lupus erythematosus (SLE) using the Mendelian randomization (MR) method.

**Methods:** Single nucleotide polymorphisms associated with asthma were used as instrumental variables. The inverse-variance weighted (IVW) method was used as the main MR method to estimate causal effects based on the summary-level data for RA and SLE. Cochran's Q test as the sensitivity analyses validated the robustness of the results and ensured the absence of heterogeneity and horizontal pleiotropy.

**Results:** Asthma (adult onset) and asthma (childhood onset) were identified to be causally associated with RA based on the IVW method (adult-onset asthma, odds ratio [OR]: 1.360, 95% confidence interval [CI]: 1.176–1.572, \( p = 3.30 \times 10^{-5} \); childhood-onset asthma, OR: 1.119, 95% CI: 1.030–1.216, \( p = 7.96 \times 10^{-3} \)). There were no associations between genetically predicted asthma (both adult and childhood onset) and the risk of SLE. Sensitivity analysis, like Cochran's Q test, further excluded the influence of heterogeneity and horizontal pleiotropy.

**Conclusions:** Both adult- and childhood-onset asthma were causally associated with RA but not SLE. The findings are valuable for understanding inflammation related to asthma and RA pathology and can guide the prevention of different diseases.

Introduction

Asthma is a condition in which the airways narrow and swell, leading to excessive mucus production, coughing, a whistling sound (also known as wheezing), and shortness of breath [1]. Globally, nearly 340 million people are affected by asthma (4.3–8.6% of adults and 2.8–37% of children, depending on the country) [2, 3]. At present, there are no signs of a decline in the prevalence of asthma or other atopic conditions. In fact, the prevalence of asthma has increased in many countries, especially among children. More than 1000 people per day die from asthma, and low- and middle-income countries disproportionately suffer the most severe cases [3].

Current research suggests that asthma is not a simple airway disease; rather, it is a chronic disease that causes systemic inflammatory dysfunction. Aspects of its impact include humoral immunity, cell-mediated immunity, and immunity related to bacteria [2]. There are complex interactions between asthma and many inflammatory diseases, including inflammatory bowel disease [4], irritable bowel syndrome [5], gastroesophageal reflux disease [6], rheumatoid arthritis (RA) [7], vasculitis [8], and systemic lupus erythematosus (SLE) [9].

Numerous studies have indicated a complicated relationship between asthma and RA; some have found asthma to be a risk factor for RA [10–16], while others reported asthma to be a protective factor against RA [17]. These inconsistent results may be related to the different ages of the sample populations and the different diagnostic criteria. RA is an autoimmune disease characterized by chronic synovial inflammation resulting in a clinical syndrome of symmetrical polyarthritis. RA can decrease the functional ability of patients, increase mortality, and negatively affect socioeconomic status. The annual incidence of RA is approximately four cases per 10,000 population with the peak incidence occurring in the age range of 50–60 years old [16]. Research on the etiology of RA is still in the exploratory stage. However, both genetic and environmental risk factors are known to play important roles in the pathogenesis of RA. SLE is an autoinflammatory disease with a wide variety of clinical manifestations in the human body. Global SLE prevalence statistics suggest that European countries generally have a relatively low incidence of SLE, while Asian, Australasian, and American countries have higher incidence [18]. While the pathogenesis of SLE is not fully understood, both genetic predisposition and environmental triggers are believed to be involved. In a cohort study of children, the overall incidence of juvenile-onset SLE was 2.52 times greater in the asthma cohort than in the non-asthma cohort [19].

A Mendelian randomization (MR)-based study found that genetically predicted childhood-onset asthma is causally associated with higher risks of gastro-esophageal reflux disease, peptic ulcers, and irritable bowel syndrome and a lower risk of inflammatory bowel disease as adults compared to the general population [20]. Those findings inspired us to investigate the effects of asthma on other immune diseases. Observational research has certain deficiencies (e.g., reverse causal associations and difficulty in excluding
confounding factors), and randomized controlled trials (RCT) can be difficult, perhaps impossible, to achieve due to ethical considerations along with limitations related to time, funding, and subjects. MR analysis, which is based on whole-genome sequencing data, can effectively reduce bias and explain the causal relationships between diseases at the genetic level. In the presence of exposure–outcome confounding (where associations between the exposure and the outcome do not themselves reflect causal effects), MR analysis can still estimate the causal effects [21]. Thus, we used MR analysis to explore the causal effects of predicted childhood-onset asthma and adult-onset asthma on the prevalence of RA and SLE.

Methods

Data sources and selection of genetic variants

According to a previously published genome-wide relationship study (GWAS), we used summary statistics from 314,633 participants with European ancestry for childhood-onset asthma and 327,253 participants with European ancestry for adult-onset asthma, respectively [22]. All of the below asthma diagnoses come from the main and secondary international Classification of diseases (ICD-10). Overall, 13,962 cases of childhood-onset asthma (0–19 years of age) and 26,582 cases of adult-onset asthma (20–60 years of age) were compared to a control group of 300,671 individuals without any allergic diseases (e.g., asthma and hay fever) [22].

Full summary statistics for the RA data were obtained from a meta-analysis of a GWAS with European participants [23]. This study included 14,361 RA cases and 43,923 control cases. The diagnosis of RA was determined by a professional rheumatologist on the basis of the 1987 RA diagnosis criteria of the American College of Rheumatology [24]. Among all participants enrolled in this study, 88.1% were seropositive and 9.3% were seronegative for anti-citrullinated peptide antibody or rheumatoid factor, and 2.6% had unknown autoantibody status [23]. SLE-related instrumental variables were derived from independent GWASs involving 7,219 cases and 15,991 controls with European ancestry [25]. The diagnosis of SLE was determined on the basis of standard American College of Rheumatology (ACR) classification criteria. Detailed diagnostic information can be obtained from previous studies.

In the selection of genetic variants, single nucleotide polymorphisms (SNPs) associated with exposure traits at the level of \( p < 5 \times 10^{-8} \) were retained. Pairwise linkage disequilibrium (LD) clumping was performed on the European samples from the 1000 Genomes Project using a stringent cut-off of \( r^2 < 0.001 \). For SNPs absent in the outcome data, we identified proxy SNPs at an LD cut-off of \( r^2 > 0.8 \) from the SNiPA website (https://snipa.helmholtz-muenchen.de/snipa3/index.php). Any SNP absent in the outcome data without a proper proxy or significantly associated with the outcome at the genome-wide significance threshold (\( p < 5 \times 10^{-8} \)) was then excluded. The MR pleiotropy residual sum and outlier test was applied to detect potential horizontal pleiotropy and eliminate the effects of pleiotropy by removing pleiotropic outliers (\( p < 0.05 \)) [26].

We then harmonized the datasets and removed palindromic SNPs with intermediate allele frequencies (i.e., minor allele frequency > 0.42). The strength of the genetic instrument was evaluated by the F-statistic. SNPs with F-statistic < 10 were considered weak genetic instruments and removed [27]. The equation of \( R^2 \) is as follow:

\[
R^2 = \left( \frac{2 \times \text{Beta}2 \times (1 - \text{EAF}) \times \text{EAF}}{2 \times \text{Beta}2 \times (1 - \text{EAF}) \times \text{EAF} + 2 \times \text{SE}^2 \times \text{N} \times (1 - \text{EAF}) \times \text{EAF}} \right).
\]

The equation of F-statistic is as follow:

\[
F = \frac{R^2 \times (N - 2)}{(1 - R^2)}.
\]

All the remaining SNPs used to conduct the subsequent causal analysis are listed in the Supplementary File (Tables S1–S4).

Statistical analysis

All statistical data analyses were conducted using R software version 4.2.1 (https://www.r-project.org/) with the two-sample MR package (version 0.5.6). In the MR analysis, the inverse variance weighting (IVW) method was used to avoid the effects of confounding factors and obtain unbiased estimates in the absence of horizontal pleiotropy [28]. The weighted median and MR-Egger methods were used to infer the causal relationships. Each of these methods makes different assumptions regarding the effectiveness of instrumental variables (IVs). The weighted median method is used when 50% of the IVs are invalid [29]. Due to the relatively low statistical power of the causality estimate of the MR-Egger method, it is mainly used to provide estimates after correcting for multiple effects. [30]. Compared to the weighted median method, the MR-Egger method results in more accurate results. The causal effect of exposure (adult- or child-onset asthma) on the outcome (RA or SLE) is presented as the odds ratio (OR)
with the 95% confidence interval (CI). The presented ORs represent the average change in the outcome per standard deviation (SD) increase in the prevalence of the exposure.

MR-Egger regression was used to detect horizontal pleiotropy in the MR analysis. Cochran’s Q statistic was used to quantify the statistical heterogeneity among the selected SNPs; \( p < 0.05 \) was considered to be significantly heterogeneous. Leave-one-out sensitivity analysis was performed to verify the reliability and stability of the causal effect estimates to deal with the possibility of SNPs with strong influences.

To further assess potential pleiotropic effects for the instruments, we used Phenoscanner (http://www.phenoscanner.medschl.cam.ac.uk), a database that includes genotype–phenotype associations. We searched for previously reported associations for any SNP that was included as an instrument in our analysis. Associations with any secondary phenotype related to asthma were considered vertical (in the same pathway from genetic variant to RA/SLE) pleiotropy. The MR analysis was then re-conducted after excluding the SNPs significantly associated with potential confounders.

**Results**

**Causal effect of asthma on RA**

With genetic instrumental variables filter, 29 independent SNPs were identified as genetic instrumental variables for the causal relationship of adult-onset asthma with RA. These SNPs explained 0.32% of the total variation, and the median [minimum, maximum] F statistic were 31.91 [29.86, 72.67]. Sixty-seven independent SNPs were identified as genetic instrumental variables for child-onset asthma. These SNPs explained 0.85% of the total variation, and the median [minimum, maximum] F statistic was 34.49 [29.75, 92.12]. No weak genetic instruments were included. Detailed information about the genetic variants is listed in the Supplementary File (Tables S1 and S2).

As shown in Fig. 1, adult-onset asthma was causally associated with RA based on the IVW method (OR: 1.360, 95% CI: 1.176–1.572, \( p = 3.30 \times 10^{-5} \)). The same causal association was found to be significant using the weighted median method (OR: 1.02, 95% CI: 1.017–1.511, \( p = 0.034 \)). Childhood-onset asthma was causally associated with RA based on the IVW method (OR: 1.119, 95% CI: 1.030–1.216, \( p = 7.96 \times 10^{-3} \)). The same causal association was found to be significant based on the weighted median method (OR: 1.155, 95% CI: 1.045–1.278, \( p = 4.82 \times 10^{-3} \)). The same conclusions can be drawn from the forest plots shown in Fig. 1. The results of MR-Egger shown in Fig. 1 wasn’t positive. It may be due to the low statistical power caused by the removal of some SNPs in the beginning of the analysis, making the results showed negative.

The sensitivity analysis (Table 1) ruled out potential heterogeneity and horizontal pleiotropy in the causal associations between adult-onset asthma and RA based on Cochran's Q test and the p-value of MR-Egger. For the causal association between childhood-onset asthma and RA, the sensitivity analysis excluded only horizontal pleiotropy based on the intercept of MR-Egger. The scatter plots in Figs. 2A and B show the causal relationships between asthma (adult and childhood onset) and the risk of RA. Considering the results shown in both Figs. 1 and 2, both adult- and childhood-onset asthma are risk factors for RA. Leave-one-out test for the relationships between both types of asthma and RA did not indicate any SNPs with large effect sizes (Figs. 3A and B).
Causal effect of asthma on SLE

With genetic instrumental variables filter, 33 independent SNPs were identified as genetic instrumental variables for the causal relationship of adult-onset asthma with SLE. These SNPs explained 0.37% of the total variation; the median [minimum, maximum] F statistic was 31.54 [29.86, 72.67]. Seventy-three independent SNPs were identified as genetic instrumental variables for childhood-onset asthma. These SNPs explained 0.93% of the total variation; the median [minimum, maximum] F statistic was 35.92 [29.74, 92.12]. No weak genetic instruments were included. Detailed information about the genetic variants is listed in the Supplementary File (Tables S3 and S4).

There were no associations between genetically predicted asthma (both adult and child onset) and the risk of SLE based on the IVW method (adult-onset asthma, OR (95% CI): 1.207 (0.960, 1.517), \( p = 0.107 \); childhood-onset asthma, OR (95% CI): 1.023 (0.913, 1.146), \( p = 0.691 \)). The sensitivity analysis further confirmed these results.

Discussion

This is the first study to systematically explore the potential causal effects between asthma (both childhood and adult onset) and two common autoimmune disease (RA and SLE). The results suggest that genetically predicted asthma is associated with an increased risk of RA. The findings are theoretically supported by consistent robust sensitivity methods. Limited MR evidence supports a potential causal relationship between genetic susceptibility to asthma and SLE risk.

It is conceivable that asthma could affect RA. Asthma and RA may have some common pathogenic pathways. Regarding immune factors, increased T helper cell 17 (TH17) activity and increased expression of interleukin 17 (IL-17) play important roles in the development of asthmatic airway inflammation by inducing TH2-associated eosinophilia and airway mucin 5AC expression and increasing airway hyperresponsiveness [31]. This immune pathway is also thought to be involved in the pathogenesis of RA since IL-17 expression and TH17 activity are increased in RA patients compared with non-RA individuals. Thus, the upregulation of TH17 activity along with other inflammatory markers observed in asthma may trigger systemic inflammation and joint destruction later in life. Besides the TH17 pathway, over-expression of other inflammatory molecules including leukotrienes, tumor necrosis factor alpha and natural killer group 2D have also been observed in association with asthma as well as rheumatoid arthritis [16].

Regarding environmental factors, cigarette smoking is a well-documented predisposing factor for asthma. Through multiple mechanisms, smoking eventually leads to increased inflammatory burden in the lower respiratory tract. Smoking also induces pulmonary phagocytes to release peptidylarginine deiminases 2 and 4 enzymes, which can convert endogenous proteins into citrullinated autoantigens. In turn, these citrullinated autoantigens promote the development of anti-citrullinated peptide antibodies in individuals who are genetically susceptible and may ultimately trigger chronic inflammatory responses in synovial joints [32, 33]. Genetic factors may also explain the relationship between asthma and RA. Certain genetic variants in immune-related genes have been associated with increased susceptibility to both asthma and RA. For example, the HLA-DRB1 gene is a major determinant of an individual's susceptibility to RA and other autoimmune diseases. Candidate gene association studies have also suggested that this gene is associated with asthma [16].

### Table 1

Sensitivity analysis of the associations between Asthma (adult/child onset) and the risk of Rheumatoid arthritis.

<table>
<thead>
<tr>
<th>Exposure</th>
<th>Outcome</th>
<th>Nsnp</th>
<th>Cochran's Q test</th>
<th>MR-Egger regression</th>
<th>IVW*</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>( Q )</td>
<td>( p )-value</td>
<td>( p )-value</td>
</tr>
<tr>
<td>Asthma (adult onset)</td>
<td>RA</td>
<td>29</td>
<td>32.644</td>
<td>0.249</td>
<td>0.006</td>
</tr>
<tr>
<td>Asthma (childhood onset)</td>
<td>21</td>
<td>122.920</td>
<td>2.68×10(^{-5})</td>
<td>(-0.002)</td>
<td>0.828</td>
</tr>
</tbody>
</table>

* Causal estimates after removing the confounder-associated SNPs.

Nsnp, number of SNP; MR, Mendelian randomization; IVW, inverse variance-weighted; OR: odds ratio; CI, confidence interval.
In addition to the above factors, the relationship between asthma and RA may be related to the following factors: active or passive smoking [34–36]; obesity [37, 38]; occupational exposure (e.g., nursing, cleaning, firefighting, bricklaying) [39, 40]; and postmenopausal hormone replacement therapy, which was found to slightly increase the incidence of asthma [41] but reduce the risk of RA, although the effect was not significant [42]. Some studies have suggested that childhood sexual abuse, relationship violence, and negative family environments are strong predictors of asthma in adulthood [43]. Meanwhile, stress can dysregulate the immune system. Lee et al. also found an increased risk of RA in patients with post-traumatic stress disorder compared to the general population [44].

Based on a systematic review and meta-analysis, Charoenngam et al. found that asthmatic patients have a 37% higher chance of developing SLE than individuals without asthma [9]. In the present study, we did not find a causal association between asthma and SLE. The reported associations may be due to bias, confounding factors inherent to observational studies (e.g., immunosuppressive agents, hydroxychloroquine, and corticosteroids), reverse causation, small cohorts, or selection bias.

Using data from a large GWAS dataset has enabled us to more precisely test our hypothesis compared to the use of individual-level data from a smaller study. MR analysis is less susceptible to unobserved confounding and reverse causality, which are issues inherent in observational studies. Nevertheless, this study has notable limitations. Our study population only included individuals of European ancestry with SLE or RA. Causality may depend on ethnicity and selection bias; therefore, further MR studies are needed with other populations. The data used in this study were obtained from large-scale GWASs, and the lack of detailed demographic information and clinical manifestations of study subjects made it impossible to conduct sex-specific and ethnic-stratified analyses. Furthermore, the limited SNPs were selected as IVs in the present study, which may have limited the proportion of variance explained by the IVs.

**Conclusions**

In conclusion, the present study supports a causal effect of asthma (both childhood and adult onset) on the risk for RA. Asthma is an airway disease that affects all age groups around the world. Its occurrence and development and its impact on the human body cannot be viewed in isolation. Genetic analysis can help us identify factors in the pathogenesis of asthma to understand the causal effects of asthma and other diseases and guide their further prevention and treatment.

**Declarations**

**Acknowledgments:** We thank all the consortium studies for making the summary association statistics data publicly available.

**Author contributions:** GH and XZ designed the study and drafted the first version of the manuscript. YZ and WL conducted the data analyses. All authors revised and approved the final version of the manuscript. XZ takes full responsibility for the integrity of the study.

**Ethics declarations**

Ethics approval and consent to participate: We used publicly available aggregate data in this study; therefore, no separate ethical approval was required.

Consent for publication: Not applicable.

Competing interests: The authors declare that they have no competing interests.

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**Data Availability statement:** The data used to generate the results in this study were obtained from GWAS summary statistics and were publicly released by the genetic consortia.

**References**


**Figures**
### Figure 1

Overview and analysis process of our research. MR, Mendelian Randomization; OR, odds ratio; CI, confidence interval; RA, rheumatoid arthritis; SLE, systemic lupus erythematosus; IVW, inverse-variance weighted.
Figure 2

Scatter plot showing the effect sizes (beta) of the SNP effects on adult- and childhood-onset asthma (x-axes) and SNP effects on RA and SLE (y-axes) with 95% confidence intervals. Each dot represents one of the SNPs used as the genetic instrument. The slopes indicate the estimate for each of the three different MR tests. **A, B.** Relationships between adult- and childhood-onset asthma and the risk of SLE. **C, D.** Relationships between adult- and childhood-onset asthma and the risk of RA. MR, Mendelian randomization; IVW, inverse-variance weighted.
Figure 3

Plots of leave-one-out analyses for MR results of the causal effect of adult-onset (A) and childhood-onset (B) asthma on the risk of RA. The x-axis corresponds to log (odds ratio) effect for adult- and childhood-onset asthma on the risk of RA and SLE.

Supplementary Files

This is a list of supplementary files associated with this preprint. Click to download.

- Supplementalfile.xlsx