Allergol Immunopathol (Madr). 2016;xxx(xx):xxx–xxx

Allergologia et immunopathologia

Sociedad Española de Inmunología Clínica, Alergología y Asma Pediátrica

www.elsevier.es/ai

ORIGINAL ARTICLE

Uncovering potential key genes associated with the pathogenesis of asthma: A microarray analysis of asthma-relevant tissues

Y. Guan[a], X. Jin[a], X. Liu[b], Y. Huang[a], M. Wang[a], X. Li[a,*]

[a] Department of Respiratory Medicine, The First Hospital, Jilin University, Changchun 130000, China
[b] Department of Intensive Care Unit, The First Hospital, Jilin University, Changchun 130000, China

Received 7 April 2016; accepted 19 August 2016

KEYWORDS
Asthma;
Differentially expressed genes;
Protein–protein interaction network;
Gene association network;
Transcription factors

Abstract
Background: The present study aimed to discover more potential genes associated with the pathogenesis of asthma.
Methods: The microarray data of GSE67940 was downloaded from the Gene Expression Omnibus database. Differentially expressed genes (DEGs) were identified in bronchial alveolar lavage cells from patients with mild-moderate asthma (notSA) and severe asthma (SA) compared with normal controls (NC), respectively. Functional and pathway enrichment analysis, protein–protein interaction (PPI) network analysis were performed upon the identified up- and down-regulated DEGs. Besides, the gene association network based on the common up-regulated and down-regulated genes was generated and transcriptional regulatory pairs of overlapping DEGs in the PPI network were identified.
Results: A total of 104 DEGs (30 up- and 74 down-regulated genes) were identified in notSA vs. NC. Additionally, 2796 DEGs were screened out in SA vs. NC group, including 320 up-regulated DEGs, and 135 down-regulated DEGs. Specially, 41 overlapping DEGs were screened out in notSA vs. NC and SA vs. NC, including 16 common up-regulated genes and 25 common down-regulated genes. No pathways were enriched by the DEGs in notSA vs. NC. DEGs in SA vs. NC were associated with cytokine–cytokine receptor interaction. VEGFA was a hub protein in both the PPI networks of DEGs in notSA vs. NC and SA vs. NC. Gene association network showed that signalling pathways and cytokine–cytokine receptor interaction were involved in. The overlapping *VEGFA*, and *IFRD1*, and *ZNF331* were regulated by more TFs.
Conclusion: Genes such as *VEGFA*, and *IFRD1*, and *ZNF331* may be associated with pathogenesis of asthma.
© 2016 SEICAP. Published by Elsevier España, S.L.U. All rights reserved.

* Corresponding author.
E-mail address: wenyongzhang39@163.com (X. Li).

http://dx.doi.org/10.1016/j.aller.2016.08.007
0301-0546/© 2016 SEICAP. Published by Elsevier España, S.L.U. All rights reserved.

Please cite this article in press as: Guan Y, et al. Uncovering potential key genes associated with the pathogenesis of asthma: A microarray analysis of asthma-relevant tissues. Allergol Immunopathol (Madr). 2016. http://dx.doi.org/10.1016/j.aller.2016.08.007

Introduction

Asthma is a heterogeneous and chronic relapsing lung disease with a broad spectrum of phenotypes ranging from mild to severe disease.[1] Asthma is characterised by varying degrees of chronic inflammation, airway hyper-responsiveness, mucus secretion, remodelling and obstruction.[2] Severe asthma differs from mild and moderate persistent asthma because it is often characterised by neutrophilic inflammation in the absence or the presence of classical type-2 T-helper (T_{H2})-induced eosinophilic inflammation.[1] Asthma affects 5-10% of the population in many developed countries and more than 300 million people worldwide.[3] The symptoms of most patients with asthma can be well controlled with inhaled corticosteroids and long-acting β2-agonists.[4] However, these therapies do not treat the underlying cause of asthma and may have serious side effects when used in the long term and in children. Thus, gaining more insights into the molecular mechanisms and the identification of novel and effective treatment for patients with asthma remain a high priority.

The pathogenesis of asthma is complex and varies in different clinical phenotypes and endotypes.[5] For instance, Jiang et al. showed that allergic asthma was associated with an increase in endogenous reactive oxygen species (ROS) formation, leading to oxidative stress and finally inducing damage to the respiratory system.[6] Recently, there is mounting evidence indicating that asthma is a polygenic disease and the complexity of this disease originates from interaction of an number of genes and environmental factors.[7,8] Ober reviewed several important early-life exposures which influenced asthma risk trajectories and suggested that a portion of the genetic risk for asthma was caused by genotype-specific responses to environmental exposures such as lipopolysaccharide (LPS) throughout life.[9] Other studies have also reviewed that gene–environment interactions studies help in validating the findings of genome-wide association studies and present great opportunities for better understanding the pathogenesis of asthma.[10,11] In recent years, the development of high-throughput microarray technologies offers opportunities to explore the global gene expression profiles of asthma and leads to the identification of new genes and pathways associated with asthma. For instance, researchers have applied microarrays to screen out possible regulators of asthmatic airway inflammation such as *ARG1*,[12] *ADAM8*,[13] and *SPRR2*.[14] Recently, Tölgyesi et al. reported that an altered paraoxonase-1 (PON1) activity might be involved in the pathogenesis of asthma.[15] However, most asthma-associated genes remain to be elucidated.

Selection of asthma-relevant tissues, such as bronchial alveolar lavage (BAL) and bronchial epithelial cells (BEC), may be required to discover functional genes or pathways underlying asthma risk.[16] In this study, we downloaded the microarray data of GSE67940 from a public database, which was initially used for expression quantitative trait loci (eQTL) analysis of asthma genes performed in cells from human BEC and BAL from the Severe Asthma Research Program (SARP) cohort.[16] Using the downloaded dataset, we screened the differentially expressed genes (DEGs) in BAL cells from patients with mild-moderate asthma and severe asthma compared with normal controls, respectively. Functional and pathway enrichment analysis, protein–protein interaction (PPI) network analyses were carried out upon the identified up- and down-regulated DEGs. Besides, the gene association network based on the common up-regulated and down-regulated genes was generated and transcriptional regulatory pairs of overlapping DEGs in the PPI network were identified. The objective of the current study was to identify more possible targets associated with the pathogenesis of asthma.

Materials and methods

Microarray data

The Gene Expression Omnibus (GEO) repository at the National Center for Biotechnology Information (NCBI) (publicly accessible at http://www.ncbi.nlm.nih.gov/geo/) is the largest fully public database for efficient capture, storage and retrieval of high-throughput molecular abundance data, primarily gene expression data.[17] In this study, the microarray profiles of asthma were downloaded from the GEO database under the accession number of GSE67940, which was deposited by Li et al.[16] The platform is GPL6480, Agilent-014850 Whole Human Genome Microarray 4x44K G4112F (Probe Name version). As the original studies described,[16,18] primary human BAL cells were isolated from patients with mild-moderate asthma (notSA), patients with severe asthma (SA) and normal controls (NC) by bronchoscopy with endobronchial epithelial brushing. More details and information of the patients and controls from the SARP cohort from who BAL cells were obtained could be derived from the study of Li et al.[16] Briefly, BAL fluids were spun down on 4000 g for 10 min and 0.5-1 $\times$ 10^6 cells were suspended in Qiazol solution. RNA extraction was performed using the QIACube system (Qiagen Inc., Valencia, CA, USA). RNA quality was determined with the Agilent Bioanalyzer 2100 (Agilent Technologies Inc., Santa Clara, CA, USA). Only samples with an RNA integrity number (RIN) higher than 7 were used for microarray experiments. In the present study, the dataset consisting of 43 notSA samples, 30 SA samples, and 31 NC samples, was used in the follow-up analysis.

Data pre-processing and DEGs screening

The raw data were downloaded and the probe ID was annotated with gene symbol based on the annotation information. In the case, those redundant probes were removed from the analysis. Meanwhile, if there was more than one probe hybridising to the same gene, the expression values of all probes were averaged for the gene and defined as the mRNA expression value. Finally, 19596 gene expression values were achieved.

The DEGs in notSA vs. NC and SA vs. NC were identified using non-paired *t*-test based on the linear models for microarray data (Limma) package[19] in Bioconductor. Only the genes with $|\log_2$ of fold change (FC)$| > 0.5$ and $P < 0.05$ were selected as DEGs. VennPlex is a program for comparing lists of genes derived from multiple and highly complex biological data sets, easy to distinguish and specifically filter.[20] In special, VennPlex is able to draw multiple circle

Please cite this article in press as: Guan Y, et al. Uncovering potential key genes associated with the pathogenesis of asthma: A microarray analysis of asthma-relevant tissues. Allergol Immunopathol (Madr). 2016. http://dx.doi.org/10.1016/j.aller.2016.08.007

Download English Version:

https://daneshyari.com/en/article/8736091

Download Persian Version:

https://daneshyari.com/article/8736091

Daneshyari.com