HUMAN GENETICS

Analysis of the Association between the T113M Polymorphism of the Human Interleukin 9 Gene and Bronchial Asthma

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Abstract—The T113M polymorphism resulting from the missense mutation in exon 5 of the human gene for interleukin 9 (IL9) was tested for association with bronchial asthma (BA). The genotype frequency analysis did not reveal a deviation from the Hardy-Weinberg equilibrium. A comparison of the genotype frequency distributions in a control group of healthy individuals and in patients with BA suggested an association between T113M and the clinical phenotype. However, this association was not confirmed by the affected family-based association control (AFBAC) or the transmission/disequilibrium test (TDT).

INTRODUCTION

The human interleukin gene IL9 is located in region 5q31.1 together with genes for other interleukins and is considered to be one of the major candidate genes for bronchial asthma (BA). Extensive studies have associated BA with the 5q31.1 region, suggesting that allelic variation of this region plays a role in BA inheritance [1-3]. In animals, expression of the IL9 gene is associated with variation in bronchial responsiveness [4]. Hence, it is of interest to analyze the natural variation in the IL9 gene in order to elucidate its role in pathogenesis of BA.

The objective of this work was to analyze the association of BA with polymorphism T113M resulting from transition C338T in exon 5 of the IL9 gene.

MATERIALS AND METHODS

We tested DNA samples of ethnic Russians from families with one or more patients with atopic BA living in Tomsk oblast. The families were identified by the proband. Clinical examination of patients and diagnosis of BA were carried out at the Department of Pediatrics, Siberian State Medical University according to the criteria proposed by WHO [5]. In total, we tested 41 families (166 individuals, including 51 BA patients).

Genomic DNA was isolated from whole blood by a standard procedure [6].

The T113M polymorphism was typed with BspW (Sibenzyme, Novosibirsk). A DNA region containing the corresponding polymorphic site was amplified by PCR; the primers and reaction conditions were as described earlier [7]. The primers were synthesized by the Synthesis and Sequencing Research Group (headed by A.I. Kutmin), Institute of Medical Genetics.

RESULTS AND DISCUSSION

To estimate the population frequencies of the T113M alleles, unrelated healthy individuals (N = 72) were selected from the total sample. The frequencies of genotypes TT and TM in this group were 63.89 and 36.11 %, respectively (Table 1). Genotype MM was not detected. Thus, the population frequencies of alleles T and M were 0.8194 and 0.1806, respectively. The genotype frequencies fitted the Hardy-Weinberg proportions (P = 0.1056).

In BA patients selected from the total sample, the genotype frequencies were 66.67%, 29.41% for TT, and 3.92% for TM. Genotype MM was not detected. The frequencies of alleles T and M were 0.8194 and 0.1806, respectively. The genotype frequencies fitted the Hardy-Weinberg proportions (P = 0.1056).

Although the allele frequencies were almost equal, the genotype frequency distributions differed between healthy individuals and patients with BA. Heterozygotes were more frequent among controls, whereas homozygotes of both types were more frequent among
Table 1. Allele and genotype frequency distributions observed in the samples tested

<table>
<thead>
<tr>
<th>Sample</th>
<th>Genotype frequency, %</th>
<th>Allele frequency, %</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>TT</td>
<td>TM</td>
</tr>
<tr>
<td>Unrelated controls (N = 72)</td>
<td>46 (63.89)</td>
<td>26 (36.11)</td>
</tr>
<tr>
<td>Patients with BA (N = 51)</td>
<td>34 (66.67)</td>
<td>15 (29.41)</td>
</tr>
</tbody>
</table>

AFBAC = \( \frac{2n(b-c)^2}{(2a+b+c)(b+c+2d)} \)

where \( n \) is the number of cases summed over the rows or the columns of the matrix and corresponds to the total number of parents tested [9];

\[ TDT = \frac{(b-c)^2}{b+c} \quad [10] \]

When the null hypothesis (H0: no association) is true, the distribution of the statistics is approximated by the \( x^2 \) distribution with d.f. = 1. Both tests have similar power, but AFBAC is more conservative as regards the effects of the population structure [13].

As tests for association, AFBAC and TDT are applicable in analyses of families with one affected child. When there is more than one affected child in a family, any family member can be chosen for analysis [10].

As the patients for analysis in this study, we chose the probands whose families were analyzed. Families in which one of the parents could not be tested were excluded from the analysis. The final sample included 34 groups, each consisting of an affected child and the child's parents.

Data on the inheritance of alleles \( T \) and \( M \) by the patients are summarized in Table 2. The AFBAC and TDT statistics calculated from these data were 0.69 (\( P = 0.4062 \)) and 0.67 (\( P = 0.5403 \)), respectively. Thus, we did not find a significant association between the T113M polymorphism and BA in Russians from Tomsk oblast.

The T113M polymorphism was first found in a large sample (\( N = 487 \)) from the Finnish population [7]. In this population, the proportion of chromosomes carrying the \( M \) allele was approximately 15% in the total sample and 13% in patients with BA; the T13 M polymorphism and BA proved to be unassociated.

Thus, our results and published data [7] do not confirm the hypothesis that the T113M mutation plays a role in the pathogenesis of BA.

On the other hand, BA is a multifactorial disorder and genetic predisposition to BA is possibly determined by many polymorphic genes except T1 13M acting together with the IL9 gene [14]. Polymorphic sites of these genes are now being sought in various populations; this is considered as a promising direction in genetic studies [15].

Our future work will be aimed at accumulating data and analyzing the association of T113M and polymor-
phic sites of other interleukin genes with BA and its pathogenetically important quantitative parameters (IgE level, bronchial hyperreactivity, etc.).

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REFERENCES
