

Genetic Polymorphism of MBL 2 in Patients with Allergic Bronchial Asthma

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ABSTRACT

Mannose-binding lectin (MBL) belongs to a family of glycoproteins called lectins or collectins, which possess many of the functional features of immunoglobulins. Mannose-binding lectin is a very important component of the innate immune system, which recognizes distinct pathogens and activates the classical path of the complement-fixation method. In humans, the serum levels of collectins vary widely, and their variability is correlated with susceptibility and resistance to infection and other diseases.

Recent studies show that MBL gene polymorphism is involved in the pathogenesis of many diseases, including infectious and allergic illnesses. Our study aims to determine the role of MBL polymorphism in children diagnosed with allergic bronchial asthma, especially in acute episodes. We conclude that MBL2 gene polymorphism is associated with atopy, allergic diseases and acute respiratory tract infections with MBL deficiency in early childhood. In terms of genetic polymorphism, most of the studied alleles were type A, these being the most frequently present in the studied groups, while alleles B, C or D have been explored to a lesser extent. Studies are also required for adult patients with allergic and atopic conditions, because so far, most of the research has been done on pediatric population only.

Keywords: mannose-binding lectin, innate immune system, genetic polymorphism, bronchial asthma.

INTRODUCTION

It is estimated that around the world, over 300 million people are suffering from bronchial asthma (BA), a chronic condition, in case of adults, which is curable only in case of children (1). Bronchial asthma is the most common chronic disease of childhood. According to global statistics, it is among the top 10 chronic conditions in children aged 5-14 years. The BA mortality rate in children globally ranges from 0 to 0.7 per 100 000 (2).

According to the Romanian Society of Pneumology, at the national level there are around one million people who suffer from BA and only 10% of cases are diagnosed and treated. The prevalence of BA symptoms in adults amounts to 10% of the population. Also, in recent years, its incidence has been increasing in children living in urban areas, reaching 6.4% for ages 13-14. A study conducted in Romania shows that problematic-severe asthma is more commonly diagnosed among Romanian children than previously described (3.7% vs. 1.5%) (3).

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The adaptive and innate immune systems are the two main immunity strategies in humans. The first one has the advantage of flexibility and immunological memory, but is completely dependent on the components of the innate immune system for the initiation and direction of answers. The appropriate interaction between the two immune systems leads to a very effective recognition and elimination of pathogens. A dysfunctional link between these two systems may lead to harmful immunological responses such as allergies, autoimmunity and rejection of allograft (4).

All airway allergic conditions such as allergic asthma or allergic rhinitis are the result of systemic inflammatory reactions initiated by allergens in genetically susceptible patients. The majority of allergic reactions are dominated by releasing the Th2 preferential cells as an adaptive immune response against harmless antigens in atopic individuals and the end release of immunoglobulins E (IgE) (4). $\square$

THE CHARACTERISTICS OF MANNOSE-BINDING LECTIN (MBL)

Mannose binding lectin is a protein encoded by the MBL2 gene located on chromosome 10q11.2-q21 q21 and has four exons (Figure 1). It is a 32 kDa 248 amino acid polypeptide and is considered to be an important part of the immunity system. Mannose binding lectin is a liver-derived collagen-like serum protein, which plays an important role in recognizing pathogens through its carbohydrate domain by activating the complement cascade via the lectin pathway (5).

Several MBL gene polymorphisms have been described. The more frequent polymorphisms of

the MBL2 gene are present in the coding region of the codon gene:

- 52 (rs5030737, transition C> T results in the substitution of Arg52Cys, the allele known as "D", the occurrence of this allele being 6%);
- 54 (rs1800450, transition G> A, resulting in Gly54Asp substitution, the allele known as "B") and
- 57 (rs1800451, transition G> A, resulting in Gly57Glu substitution, the allele known as "C").

These mutations lead to structural changes of the MBL protein, which results in a functional deficiency and a significant reduction in the circulating MBL protein level (5, 7). These polymorphisms are collectively known as „AO" and reveal changes in plasma levels of MBL. The „A" allele designates the wild type allele, while the „O" allele indicates the presence of mutant alleles in any of the three polymorphisms (8). The „A" type MBL gene polymorphisms in hospitalized children diagnosed with bronchiolitis have been found as follows:

- A/A – 68%,
- A/O – 29% and
- O/O – 3% identical result with a study of adult population in Finland.

A alleles of both homozygous and heterozygous type were present in approximately 97% of all children, the B type in 10%, C in 0.4% and D in 6%. It has been found that, in the association between the various MBL genotypes and viral respiratory infections, the genotype A/O prevails and there is a need for a post-bronchiolitis wheezing treatment with corticosteroids (9).

Other described SNPs (single nucleotid polymorphisms) are in the following positions:

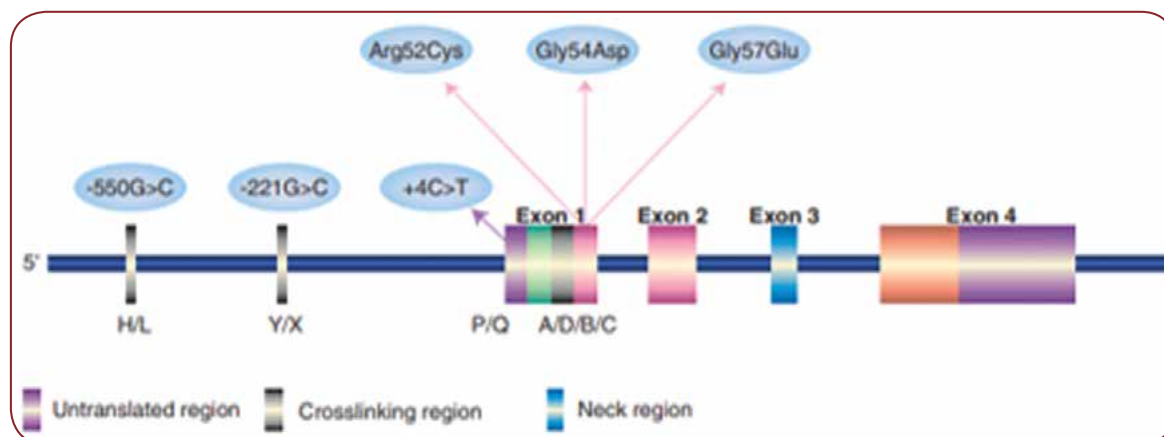


FIGURE 1. The structure of MBL gene (6)

- 550 (rs11003125, C> G, the “HL” variant, where L is the wild type allele, the H allele is present in 32% of cases) and
- 221 (rs7096206, G> C, variant “XY”, where Y is the wild type allele).

These polymorphisms can change the transcription process, which further determines an alteration in MBL serum concentration (5, 10). Mannose binding lectin deficiency causes an opsonization defect mainly by lowering the C3-derived opsonin deposition, being associated with increased susceptibility to infection and atopy, and it contributes to the development and maintenance of airway hyperresponsiveness (11). □

MBL 2 GENETIC POLYMORPHISM IN ALLERGIC BRONCHIAL ASTHMA

The results of studies regarding MBL gene polymorphism show high levels of circulating MBL protein in both adults and children with BA, although few studies report an association of MBL with severity of BA symptoms; however, others suggest that MBL may play a modulating role in disease progression. Therefore, we can consider it a sensitivity marker for severe disease or a marker for a distinct phenotype of BA. Also, associations between serum MBL and eosinophilia were reported, supporting the hypothesis about the role of MBL in various BA phenotypes (12), the increased MBL being associated with atopy, so it could be the main distinguishing feature of children with wheezing (13).

Mannose binding lectin genetic polymorphism has been studied lately with regard to the association of certain alleles and atopy, respectively allergies. Low MBL serum levels are secondary to point mutations at codon 54 in exon 1 and cause glycine substitution with aspartic acid (GGC-GAC) for B alleles. The effect of B alleles on MBL concentration would be secondary to the incorrect assembly of the triple helical structure. Type B alleles are the most common in healthy Caucasian population and rare in East African population (14, 15).

In the case of C allele at codon 57 (GGA-GAA), glycine is substituted with glutamic acid, this allele being more frequent in the West African population, rare among Caucasians and absent in Asian population. The third known substitution is at codon 57 (CGT with TGT), which determines the substitution of arginine with cysteine in D allele (frequent allele in Caucasians, but absent in Asian

population). Type D allele decreases the value of the MBL in heterozygous patients (14, 15).

Some studies have shown that the six known polymorphisms that affect a single nucleotide of MBL 2 have an important effect on the structure of MBL protein and on its serum concentration. Three of these polymorphisms are placed in exon 1 at codons 52 (CGT-TGT), 54 (GGC-GAC) and 57 (GCA-GAA) (16).

In case of children, there is no effect of low MBL levels regarding susceptibility to infections of the respiratory tract as well as allergies (17).

A study carried out on African pediatric population with moderate asthma compared to healthy controlled population reports the presence of up-raised levels of MBL serum concentrations in asthmatic children, as well as significant correlation with increased eosinophils in peripheral blood. Oxidative stress increases the MBL synthesis, which can start the activation of the complement in bronchial asthma and due to this fact it can aggravate inflammation (18).

Other studies demonstrated that patients with allergic or respiratory disease, respectively BA, allergic rhinitis or allergic bronchopulmonary aspergillosis presented increased levels of circulating MBL and MBL activation path (measured as C4b deposition on the surface of mannose) (19).

Since the MBL level markers are determined genetically, the contribution of MBL genetic polymorphism to allergic susceptibility was considered to be relevant to study. Some studies have carried out the link between MBL genetic polymorphism and atopic disorders. Studies regarding the association between bronchial asthma and atopy with MBL 2 polymorphism had contradictory results – some of them confirmed this association, while others did not find any obvious connection between MBL polymorphism and bronchial asthma or atopy (20).

The lowest MBL levels were found in patients who had had severe forms of respiratory infections (study carried out in Brazil on 81 children aged under five years) (21).

In children under six who were included in a study carried out in China, it was noted that those who had B allele in the MBL 2 gene had submitted recurrent respiratory infections (22).

Studies regarding the link between MBL serum concentrations, MBL polymorphism and allergies or bronchial asthma in children are contradictory (18).

It has been found that increased levels of seric MBL rather than low ones were associated with allergies, wheezing or asthma (a Turkish study on school age children compared to a control group with the same age and sex) (18).

It has been proven that MBL serum levels did not differ in atopic compared to non-atopic children, nor in children with and without wheezing.

In contrast, the MBL levels were significantly risen in children treated with inhaled corticosteroids compared to those who did not need this specific medication and the healthy ones (study conducted in England on children aged 10) (23).

In asthmatic children and chronic recurrent infections with *Chlamydia pneumoniae*, low MBL levels have been found to be associated with changes in MBL alleles at codons 52, 54 and 57 (24).

Following the polymorphism of a single nucleotide at the level of MBL in patients with allergic reactions who were included in a study on Indian native population, it has been shown that the presence of 1011A allele at codon G1011A was highly associated with increased serum levels of MBL, subsequently increasing MBL activity. Thus, the SNP G1011A may be involved in the monitoring of MBL expression, possibly through affecting the division mechanisms. One of the MBL haplotypes, HYP, resulting from three polymorphic sites (H/L, X/Y, and P/Q) in the initial MBL area, is associated with increased MBL plasma levels. Therefore, the above mentioned study suggests that this polymorphism of a single nucleotide may be an imbalance, that is the promoter of MBL polymorphism. A significant association of this 1011A allele has also been noticed in patients diagnosed with bronchial asthma, allergic rhinitis and allergic bronchopulmonary aspergillosis who concomitantly had high peripheral blood eosinophils and low FEV1.

This study also highlighted the role of these alleles in mechanisms involved in bronchial asthma and in allergies pathogenesis, probably by increasing plasma levels of MBL and, subsequently, of its activity. The MBL polymorphism has determined either low or high serum levels of MBL. Individuals with MBL polymorphism that lead to low MBL levels and complement activity have an increased susceptibility to infection, but a reduced likelihood of developing complement-mediated diseases, such as allergic diseases (19).

Some studies have investigated whether a MBL 2 genetic polymorphism leads to low MBL

levels and a deficiency in the MBL functioning that would cause the onset and persistence of bronchial asthma and atopy in adults. The results highlighted that the proportion of structural variants of genotype was similar to that previously described in Finnish and Caucasian population, respectively the most common form of all variants was the one with B allele (25).

This study has denied certain previous results which suggested that low MBL levels had a predisposing effect to atopy in adult patients. In fact, low MBL values can even provide protection against certain atopic diseases in certain subgroups of patients. According to the study, MBL 2 genotype has no effect in terms of predisposition on the appearance of atopy. Under different conditions, any possible change of the X allele at the level of the initial region can suffer a decrease of MBL serum concentration, and this is a strong factor for asthma in non-atopic male subjects. Low values of MBL, secondary to the initial region of polymorphism gene, could determine a poor recovery after lower respiratory tract infections, hyperreactivity of the airways and may play a significant role in the pathogenesis of intrinsic bronchial asthma. This predisposition for intrinsic bronchial asthma secondary to the X allele has been noticed only in male subjects. However, it is known that there are differences between women and men concerning the immune mechanisms. The predisposing effect of MBL 2 genotype was limited to the initial region of the X allele and has not been extended to other structural variants. The structural genotypes manifest differently: structural variants distort the formation and structure of the MBL molecule, while X alleles negatively influence the MBL operation, which is very important, considering that MBL activates the complement in the initial stages of the innate immune response. The outcomes of MBL 2 genotype in the appearance of atopy in the control group has not been statistically significant. In the group of subjects with atopy, respiratory symptoms were less frequent in women with A/O genotype compared to those with A/A genotype.

The conclusion of this study would be that the MBL genotype has no effect in terms of atopy risk (26).

In another study about the concentration of MBL polymorphism and susceptibility to respiratory infections in young patients, MBL levels below the average value have been shown to significantly increase the risk for respiratory infections.

Regarding the structural genotype of MBL 2, the A/O or O/O variants have prevailed more than A/A genotype, which targets the groups that have experienced at least two episodes of infectious diseases during the study. Moreover, the asthmatic patients in these two groups had a significant infectious risk. Following the genetic polymorphism of MBL 2 in patients with occupational allergic asthma induced by occupational exposure to Diisocyanate (study conducted in Korea), the results have shown that genetic polymorphism of MBL 2 causes low MBL serum levels, which cannot properly neutralize the effects of oxidative stress secondary to Diisocyanate exposure (27). □

CONCLUSIONS

The MBL2 gene polymorphism is associated with atopy, allergic diseases and acute respiratory tract infections with MBL deficiency in early childhood. In terms of genetic polymorphism, most of the studied alleles were type A, these being the most frequently present in the studied groups, while other alleles such as B, C or D have been studied at a lesser extent. Further research is required for adult patients with allergic and atopic conditions, because most of the so far published studies are performed on pediatric population. □

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