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Allergy to Mus m 1: Allergy to Mus m 1: A review of structural, and immunological features

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A B S T R A C T

The prevalence of allergies to pets has been increasing over the past decades. Some of the most important animal-derived allergens are members of the lipocalin protein family, which are found in dander, saliva, and urine. These allergens disperse effectively and are widely present in indoor environments. Exposure to high levels of mouse urinary protein (Mus m 1, hereinafter called 'mouse allergen') has been previously linked to sensitization to mouse, and indicators of asthma severity or control in some studies. To date, this is the only known mouse allergen registered in the IUIS database. This allergen is responsible for 27% of the total T cell response, confirming the dominant role it plays in mouse allergy.

Mice have a worldwide distribution affecting both rural and urban areas; hence humans are frequently exposed to mouse-derived proteins. Additionally, exposure to mouse allergens has increased since they are more frequently being made pets, and in addition, exposure of laboratory animal care personnel to mice has been associated with a high risk of developing occupational allergies. Mus m 1 has been recognized as the main mouse allergen, and several studies suggest its clinical relevance. What makes Mus m 1 such an important allergen? In this review, we explored its structural, immunological, and clinical features.

1. Biological and structural aspects

Mus m 1 allergen belongs to the lipocalin family of proteins, which are involved in low molecular weight lipid transport and metabolism, and –like pheromones- are found at high concentrations and are excreted in the urine [1]. This fact could explain sensitization to this allergen in rural and urban houses.

Mus m 1 is composed of a sequence of 180 amino acids that migrate as a unique band of 20.6 KDa in 2D-PAGE (without post-translational modifications) [2,3]. In 2010, Perez-Miller et al., reported the 3D-structure of Mus m 1 protein, resolved by X-ray diffraction [4,5]. The structure displayed a typical lipocalin conformation; eight antiparallel beta strands forming a single beta-sheet, and an alpha helix forming a cavity enclosing an internal ligand-binding site (Fig. 1). In addition, there are several helix and hairpin turns distributed throughout the protein sequence, mostly connecting the β-strands. Functional studies of the cavity enclosing the ligand-binding site showed that Mus m 1 is able to bind eleven types of ligands –some of which are pheromones- with different affinities involving contact with sixteen residues [6].

This allergen shares high structural homology with its homologous proteins in humans (hLCN2 and hLCN1), cats (Fel d 4), dogs (Can f 2 and Can f 4), horses (Equ c 1), and cows (Bos d 5) [7,8]. Two Mus m 1 isoforms have been described: Mus m 1.0101 and Mus m 1.0102, and they share a 99% sequence identity. They differ in two residues located at positions 68 and 154; in which Asp^{68} and Gln^{154} residues found in Mus m 1.0101 isoform are replaced by Lys^{68} and Lys^{154} residues in Mus m 1.0102 isoform.

The effect of these variations in biological and allergenic capacity has been previously studied. Site-directed mutagenesis assays were used to identify position Y120 inside Mus m 1 as relevant for maintaining its allergenic and antigenic capacities. Mutations in this residue led to a 50% decrease in basophil activation, while lymphocyte proliferative capacity remained intact. These findings suggest that variations in Y120 could lead to the generation of hypoallergenic variants of Mus m 1, which could be useful in immunotherapy treatment of patients with mouse allergy [9].

Alignment of Mus m 1 and Rat n 1 (*Rattus norvegicus*) sequences evidenced a shared homology (Fig. 1), with a 77% identity with the two isoforms of Mus m 1 and structural homology with a value of root median square deviation equal to 0.8, indicating a high structural conservation between the compared allergens, and suggesting cross-reactivity. Additional experimental assays are needed to further explore this observation. Initially, both allergen sources could facilitate sensitization to each other, and, furthermore, it is also possible that by

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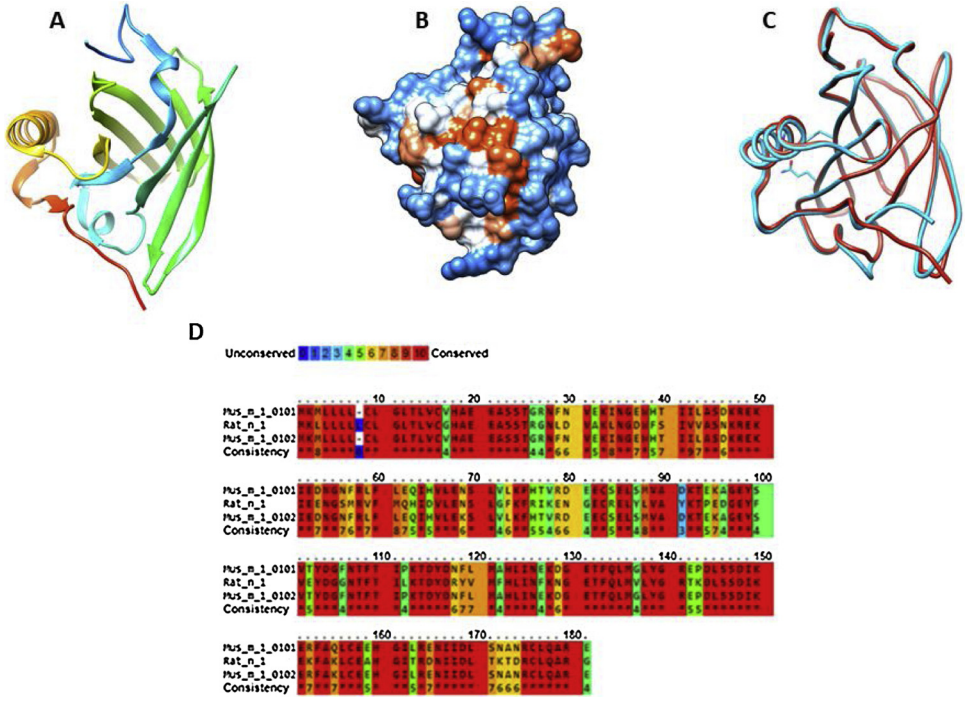

Fig. 1. 3D structure of Mus m 1.Up (a) It shows a typical lipocalin conformation (B) surface model showing hydrophobicity patches (C) RMSD between Mus m 1 (Blue) and homologous Rat n 1 (Red). Down: alignment among isoforms characterized from Mus m 1 and allergen Rat n 1. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article).

generating an immunotherapy with the hypoallergenic variant of Mus m 1, it could serve not only as treatment for patients with allergy to mouse but also for those allergic to rats.

2. Immunological and clinical relevance

Initial assays showed high stability and resistance to degradation by heat and enzymatic activity in Mus m 1, retaining its allergenic capacity. This is relevant to achieve antigenic persistence and induce sensitization in humans to mouse proteins [3]. Immunological characterization identified seven immunodominant epitopes from Mus m 1, the only known mouse allergen registered in the IUIS database. This allergen is responsible for the 27% of the total T cell response, confirming the dominant role of this allergen in mouse allergy [10]. The T cell response was restricted to all HLA class II loci (DRB1, DQ, and DP). Although, these patterns of inferred HLA restriction should be interpreted with caution, since the small numbers of donors analyzed limits the power of HLA restriction assignments based on genetic inference. Also, it was found that asthmatic and rhinitis patient exhibit differences in T cell response to peptides derived Mus m 1 when comparing the breadth of epitope reactivity (number of epitopes recognized). Asthmatic responses to Mus m 1 are more diverse compared to rhinitic patients. These differences could influence in immunotherapy approaches. Cytokine analysis showed that theses epitopes induced IL-4, IL-5, IL17 and INF-γ in PBMC from asthmatic patients [11,10]. Epitope sequences comparison exhibit high conservation grade among different lipocalins members [11]. This is relevant, because suggest that other allergenic sources (dog, cat) could induce or increase allergy response in sensitized subjects to mouse and suggests that immunotherapy using peptides containing T cell epitopes from mouse, could help reducing symptoms produced by other allergenic sources.

Exposition to mouse or rat allergens is variable, considered like a plague, mice are present in urban, rural and work environments. In this way, sensitization to this allergenic source is difficult to predict. For example, workers in industry can be exposed to allergens that are not present in the general environment [12]. Work-related sensitization is often followed by the development of symptoms and bronchial hyperresponsiveness [13]. Recent studies in a few industries have shown that the likelihood of IgE-mediated sensitization increases with increasing exposure intensity [14]. Serological and clinical studies have characterized to murine allergens as good inductors of IgG4 antibodies. IgG4 anti-murine urinary allergens are present in 28% of workers exposed to mouse and rats [15]. Usually, IgG4 levels are used as marker for immunological tolerance, because its blocking IgE activity effect and inhibit basophils activation. However, IgG4 antibodies have been characterized in workers before of the IgE levels arise, in a fashion of Th2 modified response, without no protective effect on development of sensitization or allergic symptoms [16,17]. Functional study is needed to know the role of IgG4 in the mouse allergen response.

In children, exposition to Mus m 1 and its effects in sensitization and asthma developing is controversial. In children living in inner-city areas, exposition to mouse allergens levels are associated positively with risks to develop asthma [18]. The levels of Mus m 1 in inner-city areas have been identified to be range between 8.0 μg/g and 29.9 μg/g. These levels are associated with allergic sensitization, and no additional increase in the risk was observed among children exposed to > 29.9 μg/g [18]. Similar to findings with workers, children had a Th2 modified response without beneficial effects [18]. According to Matsui et al, these findings have important implications for the development of immunomodulatory primary prevention strategies, suggesting that approaches aimed at increasing allergen-specific IgG levels may not afford protection against an allergen-specific IgE response in young children [18].

Others researchers have found a positive correlation between allergens levels and risk for developing asthma in children living in inner-city. Must be clarified that studies compared here were made in different populations. Last one mentioned was conducted in Puerto Rican population and other in U.S (Hartford, Connecticut). So, a question is remaining: genetic background is playing a role in risk to sensitization to mouse allergen in atopic subjects? [19,20].

Laboratory animal workers is another population exposed to this kind of allergen. The prevalence of sensitization to occupational allergens is higher among animal handlers (16%) than non-animal handlers (3%) [21]. However, the prevalence of asthma and rhinitis did not differ between the groups. In both groups of subjects, the prevalence of asthma, defined according to BHR and symptoms of wheezing, chest tightness or dyspnea, was 10%, which is similar to the prevalence in the general population [22]. A high percentage of this population had a

positive skin prick test for common allergens, as compared with without animal exposure [13,23]. These facts suggest the risk of working with animals is often misjudged, and adequate protection will only be used when the first complaints appear. Another studies indicated that laboratory animal workers develop first symptoms within first years of exposure. Atopy and smoking predispose to laboratory animal sensitization and to a development of bronchial asthma and allergic rhinitis [24]. Strategies to reduce are directed to prevention programs [25]. For this kind of occupational allergy, allergen component analysis is a promising tool for its diagnosis and management. Only Mus m 1, the major mouse allergen, is available as single component on the multiplex test system (ISAC®). In addition to skin prick tests and ImmunoCAP to determine sIgE levels to urine and epithelia allergens, a study with 20 of 75 workers using multiallergen IgE immunoblotting. This system can be useful in providing the sensitization profile for each allergic worker and therefore it is one step forwards in the molecular diagnosis laboratory animal allergy [26].

Molecular diagnosis using Mus m 1 shown a better sensitivity and specificity thant the mouse epithelia [27], so the use of the major allergen is preferable to the use of the complete extract. Additionally, the current regulations for the preparation of complete extracts in various European countries, is increasingly demanding, which hinders its production and availability for use in clinical practice [28,29]. Interestingly, due to the current biotechnological systems, the production of specific allergens is becoming easier and their availability in clinical practice worldwide is increasing, which added to their better sensitivity and specificity, make the diagnosis by components more accessible than the diagnosis with extracts.

Although all studies performed show to Mus m 1 as a relevant allergen for allergy to mouse, is little is known about its implications in other regions where mouse and rats are frequent, like Latin America where these rodents are considered a plague and exposition to its allergenic compounds must be high. That's why its mandatory, conduct prevalence studies to determinate the state of rodent's allergens sensitization in these countries. Even, whit these limitations there are studies conducted to develop better diagnostic tools. Currently, all approaches to obtain extracts for diagnosis and treatments of allergies to mouse are focus in Mus m 1 as major allergen, showing better results than extract obtained from epithelial [27].

Conflict of interest

The authors declare no conflict of interest.

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