



## **ACTION OF RECOMBINANT Blo t 5 PROTEIN FROM THE *Blomia tropicalis* MITE ON THE DEVELOPMENT OF A MURINE MODEL OF ASTHMA**

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The development of allergic asthma is dependent on sensitization with allergens, particularly derived from house dust mites (HDM). *Blomia tropicalis* is a prevalent HDM in tropical areas and represents an important source of allergens inducing respiratory allergies, which includes the Blo t 5 major allergen. This was to analyze the action of recombinant rBlo t 5 protein (rBlo t 5) from *B. tropicalis* in the development of a murine experimental model of asthma. The Blo t 5 sequence was obtained from GenBank database and used to design primers to amplify the Blo t 5 gene. The PCR product was cloned in pET28a vector and used to transform BL21 *E. coli* strain. The protein was purified in a nickel affinity chromatography and the contaminants were removed in polymixin-agarose resin. To evaluate the action the rBlo t 5 in a murine model of asthma, Balb/c mice were immunized four times with 20µg recombinant protein at regular intervals of 7 days, followed by three daily nebulization. The respiratory parameters were evaluated by spirometry approach and ELISA measured the levels of specific IgE, IgG antibodies and IgG subclasses (IgG1 and IgG2a). The CEUA-UFU approved these experiments. The rBlo t 5 was able to induce early IgE antibody response at 21<sup>th</sup> day of sensitization, with increasing of the IgG levels until the 45<sup>th</sup> day. The response to rBlo t 5-specific IgG1 was also detected up to the 45<sup>th</sup> day, whereas that of IgG2a was detected on a smaller scale. Alterations in respiratory parameters, as the decrease in tidal volume, were observed in mice that had received rBlo t 5 nebulization in comparison with mice that had received only PBS. We concluded that the rBlo t 5 protein produced herein was able to induce asthma in the murine model with the induction of allergen-specific immunoglobulin, highlighting the important role of this mite allergen in this immunopathological process.

**Keywords:** house dust mites, molecular biology, respiratory diseases.

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