

House Dust Mites: Recombinant expression and IgE reactivity analysis of Blo t 23 and Der p 20

JENNIFER EMILY ANUNCIAÇÃO SOUSA^{1,2}; LÓURENCE JOSUÉ. WERNER. GOMES¹; CAROLINA ORRICO MELO FERREIRA DE JESUS^{1,2}; JOÃO VITOR BORGES RIOS¹; PAULO EMILIO DE OLIVEIRA CRUZ¹; EDUARDO SANTOS DA SILVA^{1,2}; CARINA DA SILVA PINHEIRO^{1,2}.

¹LABORATÓRIO DE ALERGIA E ACAROLOGIA – LAA, INSTITUTO DE CIÊNCIAS DA SAÚDE, UNIVERSIDADE FEDERAL DA BAHIA; ²PROGRAMA DE PÓS-GRADUAÇÃO EM IMUNOLOGIA – PPGIM.

Introduction: Allergies are a public health problem in several countries around the world. One of the main sources of allergic is house dust mites (APD), such as *Blomia tropicalis* and *Dermatophagoides pteronyssinus*. The molecular and immunological characterization of allergens from these two species is crucial for clinical applications. **Objectives:** Express and purify the allergens Blo t 23 and Der p 20 and compared the IgE reactivity of these two allergens with that of seven other allergens from APD. **Material and Methods:** Blo t 23 and Der p 20 coding sequences were cloned into pET21(a+) vectors. A colony of *E. coli* BL21(DE3) was transformed and proteins were expressed. After solubility testing, purification included anion exchange chromatography and size exclusion chromatography. The IgE reactivity was determined by indirect ELISA using rBlo t 23 and rDer p 20, along with seven additional allergens (rBlo t 1, 2, 5, 21 rDer p 1, 5, 21), were used as coating antigens. **Results:** The expression of rBlo t 23 and rDer p 20 was confirmed, with observed protein bands corresponding to their theoretical molecular weights of 13 kDa and 40 kDa, respectively. Purification steps were efficient, yielding pure proteins at final concentrations of 23.38 mg/L for rBlo t 23 and 4.47 mg/L for rDer p 20. The two proteins showed IgE binding capacity in sera from allergic patients, showing an IgE reactivity of 60% and 31% for rBlo t 23 and rDer p 20, respectively. However, rBlo t 23 displayed significantly lower IgE binding capacity compared to rBlo t 2, 5 and 21. Similar finding occurred for rDer p 20, but only in comparison to rDer p 21. **Conclusion:** *E. coli* BL21 (DE3) can be used as an effective expression vector to produce rBlo t 23 and rDer p 20, with high expression yields. Considering that they showed IgE reactivity in sera from patients allergic to HDMs, these recombinant allergens could, therefore, be used to explored as potential markers for disease severity.

Keywords: house dust mite; allergy; recombinant protein.

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