Recombinant allergen Lep d 2 for *Lepidoglyphus destructor* (storage mite)

CATALOG NUMBER: RAL0008 **LOT NUMBER:** #

RECOMBINANT ALLERGEN: *Lepidoglyphus destructor* Lep d 2 (Varela *et al*., 1994).

DESCRIPTION: Lep d 2 (obsolete name: Lep d 1) is a major allergen of the storage dust mite. The incidence of allergy to *L. destructor* may even surpass that to *Dermatophagoides* in some regions, such as tropical and subtropical areas. This allergen corresponds to the NPC2 family and it has been prepared as a recombinant mature protein fused to a his-tag in its N-terminus.

PRESENTATION: liquid protein solution

SOURCE: *Pichia pastoris*

MOLECULAR WEIGHT: determined by SDS-PAGE, the protein appears as a band between molecular markers of 25,000 and 18,400 Da, while relative molecular mass calculated from amino acid sequence is 17,026.34 Da.

BATCH COMPOSITION:

COMPONENTS	COMPOSITION
his-Lep d 2	recombinant allergen with a his-tag
Storage buffer	20 mM phosphate buffer pH 6 and 0.25 M KCl

QUALITY CONTROL:

1. PROTEIN CONCENTRATION DETERMINED ESPECTROPHOTOMETRICALLY

DO_{280} = 0.630
$A_{0.1\,\%}$ (=1 g/l) = 0.433
CONCENTRATION*: 1.45 mg/ml

* The meassurement of the protein concentration has been performed with the theoretical extinction coefficient of the recombinant protein obtained from Gill and vonHippel, 1989

2. PURITY CONTROL IN SDS-PAGE: 15%

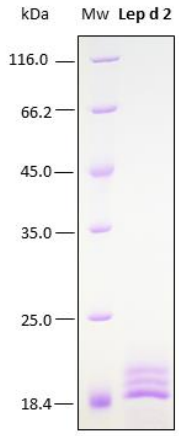

Figure 1. SDS-PAGE analysis (15%) of 5 µl of recombinant allergen. Purity is >95% as determined by gel electrophoresis. The three fragments correspond to the target protein as it has been analyzed in a western blot with a anti-His tag monoclonal antibody.

3. ANALYSIS BY AN ELISA ASSAY

The evaluation of the recombinant allergen has been performed by means of an *in-house* ELISA assay performed in a spanish hospital. This immunoassay was carried out with a serum sample panel of 25 positive and 10 negative specimen sera, pre-validated by skin prick testing (SPT) and the UniCAP® test.

The recombinant allergen lep d 2 detected 18 positive sera out of 25 (72% incidence) with higher prevalence of sera with titers of 3.5-17.5 IU/ml (international units per milliliter; 1 IU is equivalent to 2.42 ng of IgE).

4. ABSENCE OF PRECIPITATION AFTER A FREEZING AND THAWING CYCLE: ok

LOT SPECIFICATIONS:

1. CONCENTRATION: 1.45 mg/ml

2. TOTAL QUANTITY PER ALIQUOT: 1 mg

3. TOTAL VOLUME PER ALIQUOT: 0.723 ml

4. STORAGE: Protein is shipped with dry ice. Upon arrival, it should be aliquoted in order to avoid repeated freezing and thawing cycles and stored at -20ºC to -80ºC.

5. OBSERVATIONS: proteins should be maintained frozen at high concentrations. In order to defrost the protein, maintain the aliquot at 25ºC without shaking to avoid aggregation. Prior making test dilutions and after defrost the protein, is recommended to remove possible protein aggregates by centrifuging the stock solution, avoiding alterations in the immobilization of the biomolecule to the solid surface.

RELATED PRODUCTS:

Der p 1, Der p 10, Der f 2.

BIBLIOGRAPHY:

Varela, J, Ventas, P, Carreira, J, Barbas, JA, Giménez-gallego G and F Polo. Primary structure of *Lep d* I, the main *Lepidoglyphus destructor* allergen. 1994, Eur. J. Biochem. *225,* 93-98.

Bradford, MM. A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein dye binding. Anal Biochem. 1976, 131:499-503.

Rekom Biotech S.L. – BIC-Granada, Avda. Innovación, 1 – 18016, Granada (Spain) Tel: +34 958 63 70 85 E-mail: info@rekombiotech.com – Web: www.rekombiotech.com – An ISO 9001:2008 Certified Company

Important Notes: During shipment, small volumes of product will occasionally become entrapped in the seal of the product vial. For products with volumes of 200 µl or less, we recommend gently tapping the vial on a hard surface or briefly centrifuging the vial in a tabletop centrifuge to dislodge any liquid in the containers cap.

Although recombinant antigens are expressed in non-pathogenic *P. pastori* and bacterial integrity is destroyed during purification, the antigen preparation should be handled as potentially infectious.

NOT FOR DIAGNOSTIC USE, FOR RESEARCH USE ONLY

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