

Occupational allergy in a researcher due to Ole e 9, an allergenic 1,3- β -glucanase from olive pollen

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Olive (*Olea europaea*) pollen allergy is one of the most important causes of seasonal respiratory disease in Mediterranean

countries (1). To date, 10 olive pollen allergens have been characterized (2).

We report on a 30-year-old man who was born and lives in Madrid, smoker and without a previous personal history of atopy. He started working with olive pollen allergens in a laboratory research 10 years ago. In particular, he spent 5 years (from 2000 to 2005) manipulating Ole e 9-allergen and derivative recombinant products, being daily highly exposed to these molecules. In 2005, he started to suffer from nasoconjunctival pruritus, sneezing proxysms, nasal running, conjunctival redness and palatal itching while handling fractions enriched in 35–55 kDa protein components of olive pollen extract. Symptoms always appeared at the workplace and concomitantly with the manipulation of such batches of olive pollen proteins. The patient controlled the symptoms with antihistamines, remaining asymptomatic out of the workplace, even during the olive pollination season in Madrid. He ate plant-derived foods without experiencing any adverse reaction. Skin prick test with a commercially available

CRD shows Ole e 9-allergen as the causative agent of occupational rhinitis in a researcher.

battery of common aeroallergens (ALK-Abelló SA, Madrid, Spain) were positive to *Olea europaea* eliciting a wheal of 5 mm (diameter) and negative to pollens from other trees, grasses or weeds, moulds, and cat and dog epithelia.

Olive pollen extract was separated by SDS-PAGE gels under nonreducing conditions and immunostained with the patient's serum (Fig. 1A). A single IgE-reactive band of 45 kDa molecular mass was detected. Immunoblotting performed with purified olive pollen allergens were negative in all the cases except for purified Ole e 9-allergen (Fig. 1A). To confirm Ole e 9 as the IgE-reactive allergen in olive pollen extract, IgE-inhibition experiments with the purified antigen were performed in immunoblotting (Fig 1B). When the patient's serum was preadsorbed to purified Ole e 9, the IgE binding to the 45 kDa protein in olive pollen extract was nearly abolished.

Skin prick tests were carried out with purified olive pollen allergens such as Ole e 1 (0.50 μ g/ml), profilin Ole e 2 (0.50 μ g/ml), polcalcin Ole e 3 (0.50 μ g/ml) and 1,3- β -glucanase Ole e 9 (0.27 μ g/ml). Ole e 9 was the single-tested allergen able to elicit a positive response (6 mm of

diameter) indicating the capability of this molecule to trigger the release of inflammatory mediators, thus demonstrating the biological activity of Ole e 9 in the patient. Three control subjects were skin tested with Ole e 9 and showed negative response.

Ole e 9 is a 1,3- β -glucanase that has been described as a major allergen in areas such as Jaén (Spain) (3) where olive pollen concentrations reach values up to 10 000 grains/m³. In contrast, patients living in Madrid (pollen counts up to 300 grains/m³) are notably less sensitized to this molecule, and they are always co-sensitized to Ole e 1. In our study, the component-resolved diagnosis (CRD) (4) showed that the patient presented an unusual sensitization pattern to *Olea europaea* components as patients exclusively sensitized to Ole e 9 have not been described. The fact that the patient was not sensitized to Ole e 1, which is a diagnostic marker for sensitization to Oleaceae pollens (5), strongly suggests that the patient sensitization occurred at the workplace because of the daily overexposure to Ole e 9-allergen.

Our findings demonstrate that the patient suffers from occupational allergy

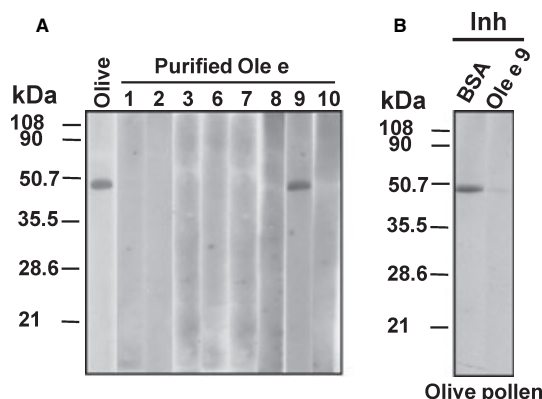


Figure 1. (A) Olive pollen protein extract or purified olive pollen allergens were separated by SDS-PAGE, transferred to nitrocellulose membranes and immunostained with the serum from the patient. (B) IgE immunodetection of olive pollen extract after preincubation of the patient's serum with BSA or Ole e 9. Molecular markers are displayed on the left margin.

being Ole e 9 the causative agent involved. This case reveals CRD as a worthy tool in the evaluation of work-related allergic diseases and highlights that the manipulation of purified allergens or enriched fractions by researchers and manufacturing pharmaceutical workers may lead to allergic processes.

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