Age and Geographical Location Determine the Molecular Pattern of Walnut Allergy in Madrid, Spain

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Nuts are the second most common cause of allergy to plant foods in Spain [1,2] and the first cause of anaphylaxis [3]. Walnut (*Juglans regia*) is the most consumed nut and the one that most frequently causes allergies in Spain [4].

Eight walnut allergens have been characterized, and several studies on molecular diagnosis of walnut allergy have been reported [5-13]. One of the most interesting was a multicenter study (Switzerland, Germany, and Spain), which concluded that the severity of walnut allergy correlated inversely with the age at onset [5]. However, the authors highlight bias in patient age from various countries, and the results from the Spanish group (mostly sensitized to lipid transfer protein [LTP]) differed from the others, with a higher incidence of systemic reactions in the adult population, suggesting that additional studies from Spain would be necessary. In Italy, Pastorello et al [6] reported a predominance of sensitization to LTP and systemic reactions in adult patients. Subsequently, the most extensive multicenter study on walnut was published. The authors reported curious data, for example, sensitization to Jug r 1 did not seem to be relevant in Europe and sensitization to LTP was predominant in southern Europe, despite the frequency of sensitization to LTP being only 23% in Spain and 22% in Italy, even lower than sensitization to profilin (30% in Spain and 38% in Italy). These data clearly illustrate the discrepancies between published data on sensitization patterns in walnut allergy, probably due to biases in patient selection [7].

The objective of this study was to evaluate sensitization patterns, the clinical characteristics of walnut-allergic patients, and the possible relationship between them in Madrid, Spain.

Fifty-one patients from Fundación Jiménez Díaz Hospital, Madrid, Spain diagnosed with walnut allergy during 2020-2021 were recruited in a cross-sectional single-center cohort study. All analyses were performed using the multiplex test system ALEX2 (MacroArray Diagnostics GmbH) (Methods, Supplementary Material [SM]).

The mean age was 27 years (4-53 years), and 47.1% were women. Onset of symptoms was within 30 minutes after ingestion in 40 patients (78.4%). The most frequent symptom was oral allergy syndrome (76.5%), followed by urticaria/angioedema (43.1%), asthma (27.5%), rhinoconjunctivitis (25.5%), and gastrointestinal symptoms (15.7%). Twenty-eight patients (54.9%) presented anaphylaxis and 3 (5.9%) anaphylactic shock. Thirty-eight patients (75%) were admitted to the emergency department, and only 9 (17.6%) received epinephrine. Seven patients (13.7%) presented reactions with a cofactor (exercise). Thirty patients (58.8%) had allergies to other nuts, mostly hazelnut (Table S-I, SM). No significant differences were identified regarding demographics, symptoms, or sensitization profile in the 21 patients who were not allergic to other nuts. The most common cause of allergy was walnut (51%), followed by fruits (27.5%), other nuts (15.7%), and vegetables (5.9%).

Jug r 3 (LTP), which was positive in 22 patients (43.1%), was the walnut allergen that produced sensitization most frequently, followed by Jug r 1 (2S albumin), and 16 patients (31.37%) were sensitized to seed storage proteins (SSPs). All patients were sensitized to Jug r 1, except 1 (monosensitized to Jug r 2), with Jug r 1 acting as the marker of sensitization to SSPs. When we expanded the assay by evaluating sensitization to LTP (including sensitization to Pru p 3), 47 patients (92.15%) were sensitized to LTP or SSPs (Figure). The frequency of sensitization to walnut allergens is shown in Table S-II (SM).

All patients sensitized to Jug r 3 were sensitized to Pru p 3. Specific IgE levels to Pru p 3 were higher among patients sensitized to Jug r 3 than in patients who were Jug r 3–negative (IgE to Pru p 3 of 3.20 [7.72] vs 1.24 [2.24] kUA/L; *P*=.034). Interestingly, sensitization to Mal d 3 and Pru p 3 was even more frequent than sensitization to Jug r 3 (Table S-III, SM).

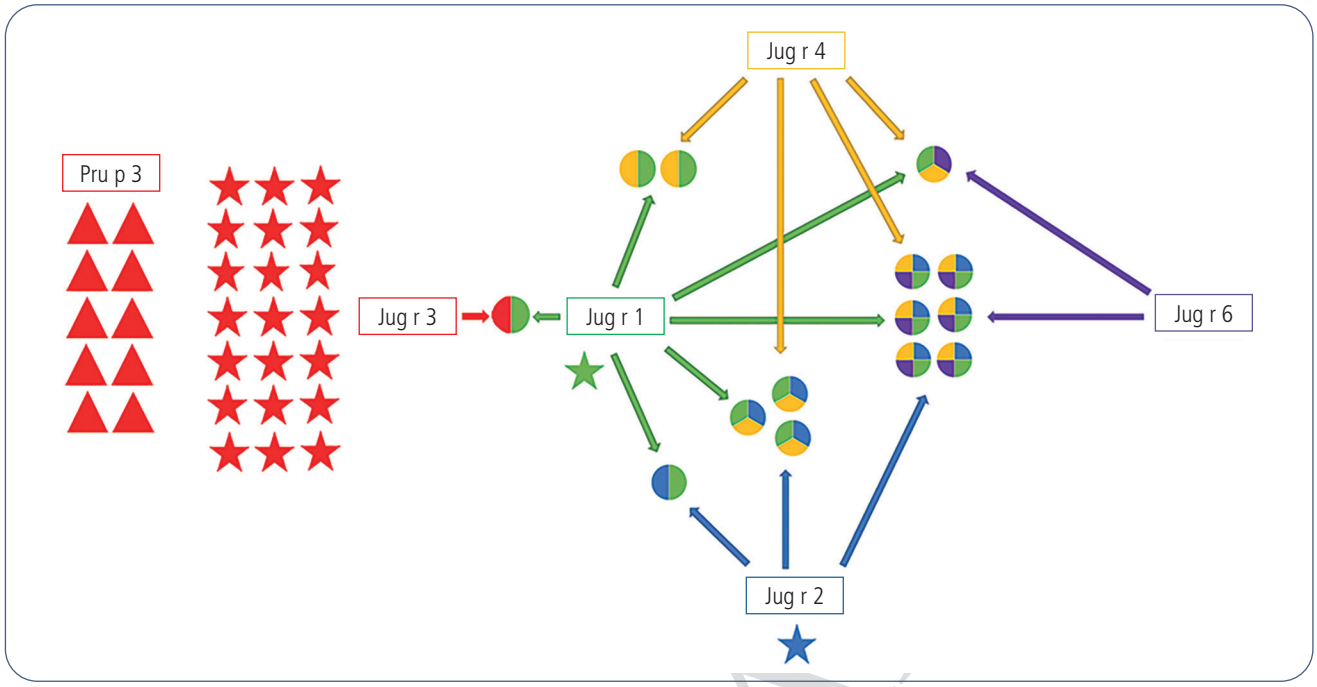

Figure. Distribution of sensitization to walnut components. Patients represented with stars showed sensitization to a single allergen only. In circles, those sensitized to more than 1 allergen simultaneously. In triangles, those sensitized to Pru p 3 but not to the walnut molecular components available in ALEX2.

Further studies are necessary to explain these results. We should remember that primary sensitization to LTP has been attributed to Pru p 3 [14] and that Pastorello et al [15] found that Pru p 3 better inhibited walnut LTP. These data suggest that some walnut-allergic patients would be more easily detected based on sensitization to Pru p 3 than on sensitization to Jug r 3.

We defined 2 sensitization patterns in walnut allergy, namely, sensitization to LTP and sensitization to SSP. Both patterns are shown in Table S-IV and Figure S1 (SM).

We consider that the results of our study are novel and relevant and will elucidate the patterns of walnut allergy reported in Spain and throughout the world.

Patients sensitized to Jug r 1 were usually children (<16 years) and experienced the most severe reactions. Often, walnut was a hidden allergen, and the allergy was limited to walnut or to other nuts. In contrast, patients sensitized to LTP more frequently presented oral allergy syndrome, although a considerable number of cases experienced systemic reactions. While walnut allergy appeared as the first expression of LTP syndrome, symptoms were induced by fruits (especially Rosaceae) in a relevant number of cases. Interestingly, only patients sensitized to LTP presented reactions associated with a cofactor.

Two factors determined the molecular pattern of walnut allergy. The first was age of onset. Childhood determines the pattern of sensitization to walnut regardless of geographical location, with predominance of sensitization to SSPs worldwide, specifically Jug r 1 [5-13]. The second factor is geography, which determines the molecular pattern in adulthood and conditions sensitization to LTP in the Mediterranean area [6,7] and to Jug r 5 in northern and central Europe [5,7]. We think that both factors would explain the discrepancies observed in the results of published studies.

In conclusion, age of onset and geographic location determined the dominant molecular pattern, and the molecular pattern determined the characteristics of walnut allergy.

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Conflicts of Interest

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