



Correlation of anti-pru p 3 IgE levels with severity of peach allergy reactions in children

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ABSTRACT

Background: Peach allergy is regarded as one of the most important fresh fruit allergies. Data are available on the state-of-the-art diagnosis, including food challenges, and a component-resolved diagnosis. However, the roles played by different peach allergens with respect to symptom severity are not completely understood.

Objective: To evaluate the role of serum specific IgE to peach and recombinant allergens in the diagnosis of peach allergies in Italian children.

Methods: Forty-four children with peach allergy confirmed by a placebo-controlled food challenge were divided into 2 groups based on their symptom severity: patients with mild oral allergy syndrome (OAS) and patients with systemic symptoms (SS). The presence of specific IgE to peach and rPru p 1, rPru p 3, and rPru p 4 was determined.

Results: The presence of specific IgE to Pru p 4 and Pru p 1 was found significantly more frequently in patients with OAS, whereas specific IgE to Pru p 3 was not found significantly more frequently in patients with SS. Only anti-rPru p 4 IgE levels were significantly higher in patients with OAS, whereas no significant differences were found in anti-rPru p 1 and anti-rPru p 3 IgE levels between patients with OAS compared with patients with SS.

Conclusion: In Italian children with peach allergies, the presence of specific IgE to Pru p 3 is not associated with SS, and the levels of specific IgE to Pru p 3 do not correlate with the severity of the reactions.

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Introduction

Peach is frequently involved in fruit allergic reactions.^{1,2} Most patients allergic to peaches present with an associated pollen allergy and exhibit other food allergies, mainly to other Rosaceae species but also to taxonomically unrelated vegetables.^{3–5}

Cross-reactive IgE antibodies directed toward lipid transfer proteins (LTPs), Bet v 1 homologues (in patients allergic to birch pollen), profilins, and carbohydrate determinants are based on these associations and can be variably detected in patients with mild symptoms (eg, oral allergy syndrome [OAS]).^{6–9} Other patients, especially in southern Europe, have mostly systemic symptoms (SS), in many cases due to sensitization to LTPs.¹⁰

As for any food, diagnosis of peach allergy is based on clinical history, supported by the demonstration of specific IgE, and confirmed by a positive oral challenge result. Recently, the use of molecular methods to test for fruit and vegetable allergies has been

suggested to improve both the diagnosis and prognosis, but clear recommendations are still lacking.¹¹

To date, few recombinant allergens are commercially available as ImmunoCAPs for in vitro diagnosis in case of fruit and vegetable allergies: peach-derived rPru p 1 (Bet v 1 homologous protein), rPru p 3 (nonspecific LTPs), rPru p 4 (profilin), celeriac-derived Api g 1 (Bet v 1 homologue), and kiwi-derived Act d 8 (Bet v 1 homologue). Therefore, peach is an allergen that can be accurately studied by molecular diagnosis. In this study, we investigated whether the severity of clinical reactions to peach in children are associated with sensitization toward peach and allergenic molecules, namely, LTP (rPru p 3), Bet v 1-related protein (rPru p 1), and profilin (rPru p 4).

Methods

Patients

From 2010 to 2011, 44 patients (mean age, 11 years; 25 boys and 19 girls) with a convincing history of allergic reactions after the consumption of peach confirmed by positive skin prick test (SPT) results to commercial peach extracts or by prick by prick testing with fresh fruit or specific serum IgE were selected from our allergy unit (thanks to the cooperation of neighboring hospitals in Tuscany). The true clinical reactivity to peach was assessed by a dou-

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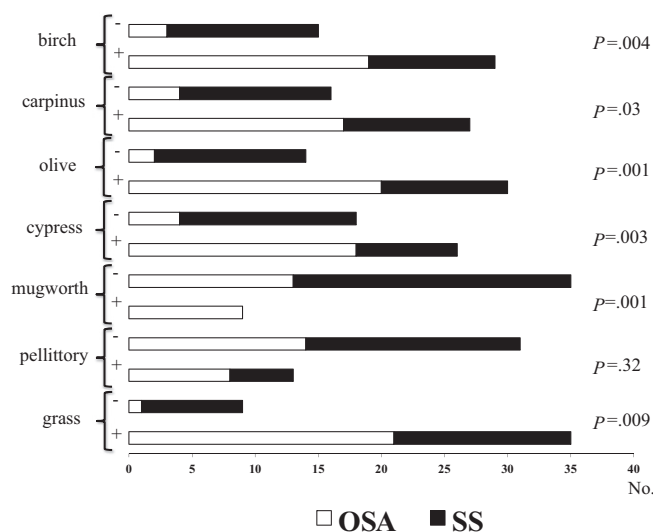


Fig. 1. Pollen sensitization pattern of the studied population. Plus sign indicates positive skin prick test result; minus sign, negative skin prick test results; OSA, oral allergy syndrome; SS, systemic symptoms.

ble-blind, placebo-controlled food challenge, except in those patients ($n = 9$) with specific IgE to peaches plus a convincing history of anaphylaxis immediately after peach ingestion in the 2 previous years, strictly avoiding peach in any form.

Patients were divided into groups according to the severity of their past reactions: a group of children with SS (eg, urticaria and/or angioedema, anaphylaxis), with or without local symptoms, and a group of children with local oral reactions (eg, OAS). Twenty-two patients with seasonal respiratory allergies without any fruit allergies were considered as the control group. This study was performed with the approval of the ethics committee, and all the patients (or their representatives) gave written consent.

In vivo tests

The prick by prick method was used when testing fresh fruit, and all of the children were tested with fresh peach. SPTs were performed by the same investigator, on the volar surface of the forearm by using a standard 1-mm tip lancet, according to the recommendations of the European Academy of Allergy and Clinical Immunology group.¹² Histamine hydrochloride (10 mg/mL) and saline (ALK-Abello, Milan, Italy), respectively, were used as positive and negative controls. In addition, SPTs with pollens, mites, molds, cockroach, cat, and dog (ALK-Abello) were performed in all study participants. The pollens tested were grass, olive, Cupressus arizonica, Betula pendula (birch), Artemisia vulgaris (mugwort), Carpinus, and Parietaria judaica (pellitory). The SPT result was considered positive if the difference between the mean diameter of the wheal and the negative control was at least of 3 mm. Oral challenges were performed according to previously published procedures.¹⁰

The amount of peach given was estimated by the weight of an average peach, which is approximately 150 to 200 g (with the stone). For the double-blind, placebo-controlled food challenge, a total of 100 g of fresh peach (peel and pulp) was blended in a vehicle (used alone as placebo food) that consisted of orange and pineapple juices (130 mL) together with soluble coffee (half teaspoon) and sugar (a teaspoon). Masking was checked in 3 volunteers who were unable to distinguish between verum and placebo. Verum and placebo were randomly administered on separate days, and both of them were prepared immediately before the challenge. Three doses (one-eighth, one-fourth, and five-eighths of the total weight) were administered at 1-hour intervals. All the challenges were

conducted in a hospital setting, with careful medical monitoring of the patients and a full emergency treatment kit readily available.

In vitro tests

Serum IgE levels of anti-rPru p 1, 3, or 4 and antipeach were determined using the ImmunoCAP system (Phadia Diagnostics, Milan, Italy), according to the manufacturer's instructions.

Statistical analysis

Statistical analysis was performed using SPSS statistical software (SPSS Inc, Chicago, Illinois) to verify whether the 3 groups of patients displayed significant differences in the skin or serum responses. A χ^2 test was used for comparing sensitization and symptoms.

Results

Twenty-two patients (mean [SD] age, 10.41 [4.23] years; 12 boys and 10 girls) had SS (9 with anaphylaxis) with or without local symptoms, and 22 patients (mean [SD] age, 12.41 [3.18] years; 13 boys and 9 girls) had only OAS.

The control group consisted of 10 boys and 12 girls (mean [SD] age, 10.31 [1.23] years). The pollen sensitization patterns of the studied patients are shown in Figure 1. Children with OAS were more significantly ($P = .025$) sensitized to pollens than children with SS for all the allergens studied except for *P. judaica*. All 44 patients had positive peach prick by prick and ImmunoCAP results (IgE level >0.10 kUA/L).

Children with OAS, children with SS, and controls were compared for the presence of specific serum IgE to rPru p 1, rPru p 3, and rPru p 4 (Table 1). Of the 22 children with SS, 15 had anti-rPru p 3 IgE compared with 17 with OAS ($P = .49$), 7 with SS had anti-rPru p 1 IgE vs 16 with OAS ($P = .014$), and 3 with SS had anti-rPru p 4 IgE vs 12 with OAS ($P = .006$).

When the serum IgE levels to peach, rPru p 1, rPru p 3, and rPru p 4 were compared in patients with OAS or SS, we found that only anti-rPru p 4 serum IgE levels were significantly higher ($P = .05$) in patients with OAS compared with patients with SS (Fig 2).

Discussion

Peach allergy is regarded as one of the most important fruit allergies. Data are available on the state-of-the-art diagnosis, including food challenges, and a component-resolved diagnosis.^{1,2} Patients from Northern and Central Europe with this type of food allergy usually present with mild symptoms restricted to the oral cavity (OAS), and the pollen allergens, but not the food allergens, are the cause of the primary sensitization (class II food allergies).^{8,13} In particular, large screening of Rosaceae fruit allergic patients from different countries has shown that where birch trees are abundant, allergies to apples and cherries seem to be linked to food homologues of Bet v 1, the major birch pollen allergen. Interestingly, both profilins and Bet v 1 homologues seem to be labile allergens, readily hydrolyzed by simulated gastric fluids,^{14,15} making the hypothesis of their low allergenicity reliable. However, SS ranging from urticaria to anaphylactic reactions have been described in few patients with birch pollen-related soybean allergy.^{16,17}

Table 1
Number of patients with specific IgE to rPru p 1, rPru p 3, and rPru p 4 in the studied groups

	Pru p 1	Pru p 3	Pru p 4
OAS	16/22	17/22	12/22
SS	7/22	15/22	3/22
P value	.01	.49	.006

Abbreviations: OAS, oral allergy syndrome; SS, systemic symptoms.

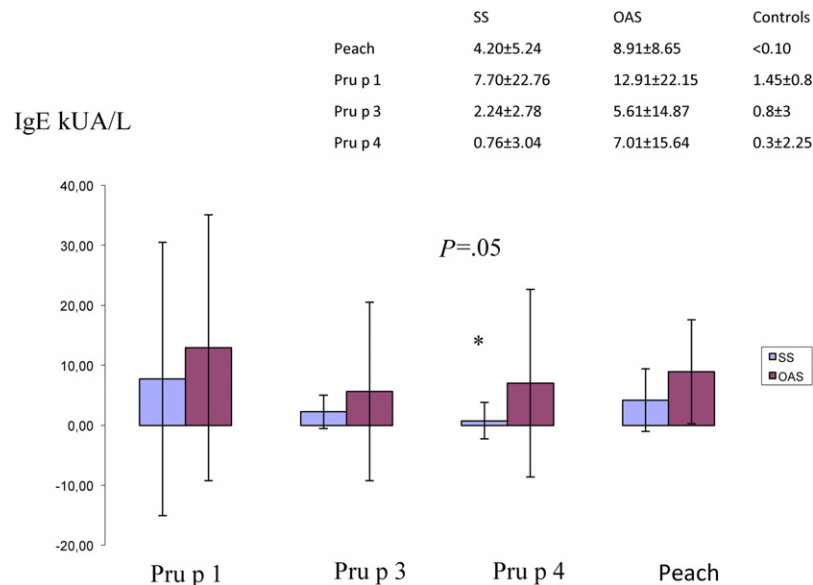


Fig. 2. Mean specific IgE levels to Pru p 3, Pru p 1, and Pru p 4 in patients with systemic symptoms (SS), patients with oral allergic syndrome (OAS), and controls (not shown). Error bars indicate SEM.

The last scenario has been more often described in the Mediterranean because of a different sensitization pattern. Several patients (>20%) have fruit allergies without related pollinosis^{10,18} and, accordingly, specific IgE to Bet v 1 is hardly detected (10%). Furthermore, systemic reactions are frequently observed (20–30%), even in the absence of OAS.^{10,19} Nonspecific LTP has been identified as the major allergen in patients with SS after ingestion of Rosaceae fruits.^{6,20} In a study performed in 55 Italian adults with food-induced anaphylaxis, most anaphylactic reactions occurred in patients sensitized to LTP, and among LTP-hypersensitive patients, peach caused 13 of the 19 anaphylactic episodes.²¹

However, some cases of IgE reactivity to peach without concomitant Pru p 3 recognition have been described,²² and recently 3 isoforms belonging to the thaumatin like protein family were identified in peach as major allergens. Their prevalence, observed in vitro, ex vivo, and in vivo analyses, suggests that they are important allergens and should therefore be included in the routine diagnosis of peach allergies, at least in the Mediterranean.²³ Therefore, some cases of peach-induced anaphylaxis without concomitant Pru p 3 recognition can be due to this allergen family.

In any case, the higher frequency of sensitization and allergenicity renders Pru p 3 a marker molecule for the LTP allergy, although this seems to depend on the level of IgE to Pru p 3.²⁴ Individuals do not necessarily react to all LTPs to which they are exposed. In the Italian population, most patients sensitized to Pru p 3 showed clinical reactivity on peach ingestion, whereas other fruits did not cause symptoms.²⁵ This study is the first, to our knowledge, to evaluate peach allergies strictly in children: patients enrolled had a positive case history of allergic reactions after the consumption of peach according to standardized questionnaires.

To confirm a peach allergy, SPTs, specific serum IgE to peach, rPru p 1, rPru p 4, and rPru p 3; and oral challenges were undertaken. In addition, SPTs to common inhalant allergens were performed. The skin test results revealed that peach allergic patients frequently reacted to most pollens (grasses, weeds, and trees) found in our geographic area, and the frequency of sensitization was higher in children with OAS than in children with SS (Fig 1). When comparing the frequency of positive serum specific IgE in patients with SS or OAS, we found that IgE anti-rPru p 1 and rPru p 4 were significantly more frequent in patients with OAS, whereas the frequency of positive IgE anti-Pru p 3 was not significantly

different in the 2 groups. Specific serum IgE levels to peach were not significantly different in children with OAS or SS. When the serum IgE levels to rPru p 1, rPru p 3, and Pru p 4 were correlated with OAS or SS, we found that anti-rPru p 4 specific IgE levels were significantly higher in patients with OAS compared with patients with SS, whereas anti rPru p 3 specific IgE levels were not significantly different (Fig 2).

These results do not confirm the results of previous studies performed in adults, which reported that peach allergic patients with SS had significantly higher levels of anti-rPru p 3 specific IgE than patients with OAS.^{26,27} This discrepancy could be because, in our study, relatively few patients were monosensitized. Recently, it has been reported that specific IgE levels to foods other than Rosaceae are only partially predictive of allergies in LTP hypersensitive patients,²⁸ confirming the conflicting results on the clinical significance of high levels of specific IgE in this type of fruit allergy.

It is unclear why some patients with low levels of specific IgE to fruit develop SS, whereas others with high levels do not, despite similar exposure to the allergen. Perhaps other factors should be taken into account; in birch pollen-related food allergies, high food allergen specific IgG₄/IgE ratios seem to be associated with food tolerance, potentially because specific IgG₄ blocks IgE binding to food allergens.²⁹

In conclusion, our study suggests that, in Italian children with peach allergies, the presence of anti-rPru p 3 specific IgE is not associated with systemic clinical symptoms, whereas the presence of Pru p 4 specific IgE is associated with oral allergic symptoms. The levels of specific IgE to peach or to rPru p 3 do not correlate with the severity of the reactions and therefore should not be considered as prognostic markers. The number of epitopes, the avidity of specific IgE, the IgG₄/IgE ratio, or other cellular mechanisms are probably involved in the immunologic reaction and in its clinical expression.³⁰ The clinical utility of the molecular diagnosis in peach allergies is still under investigation, and more studies are required for definitive recommendations.

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