

R. ASERO<sup>1</sup>, D. VILLALTA<sup>2</sup>

# Are anti-Phl p 12 IgE levels predictive of oral allergy syndrome in profilin hypersensitive patients?

<sup>1</sup>Ambulatorio di Allergologia, Clinica San Carlo, Paderno Dugnano (MI), Italy - E-mail r.asero@libero.it

<sup>2</sup>Allergologia e Immunologia Clinica, Dipartimento di Medicina di Laboratorio, A.O. "S. Maria degli Angeli", Pordenone, Italy

## KEY WORDS

*Profilin, food allergy, specific IgE levels, pollen-food allergy syndrome, cross-reactivity.*

## Corresponding author

Riccardo Asero  
Ambulatorio di Allergologia  
Clinica San Carlo  
Via Ospedale 21  
20037 Paderno Dugnano (MI)  
Italy  
E-mail r.asero@libero.it

## SUMMARY

*Background: In birch pollen-allergic patients the occurrence of clinically relevant cross-reactivity to plant-derived foods is clearly related with the level of birch-specific IgE. In profilin-hypersensitive patients this has not been investigated so far. Objective: To investigate whether the levels of profilin IgE are predictive of the development of food allergy in hypersensitive patients. Methods: IgE specific for Phl p 12, the grass pollen profilin, were measured in 37 subjects monosensitized to profilin with (n=11) or without (n=26) oral allergy syndrome (OAS) following the ingestion of plant-derived foods. Results: Patients without a history of OAS showed higher levels of IgE specific for Phl p 12 than patients with OAS (median 4.74 [range 0.7-41.6] KU/L vs 2.14 [range 0.32-10.2] KU/L, respectively) although the difference did not reach statistical significance (p=0.07). Conclusion: Factors causing the onset of OAS in profilin-hypersensitive patients remain presently unclear.*

## Introduction

Along with the major birch pollen allergen, Bet v 1, the plant pan-allergen profilin, a 12-15 kDa actin-binding protein present in all eukaryotic cells, is a major cause of cross-reactivity between pollen and vegetable food (1-4). The relevance of profilin as an allergen, albeit variable, has been shown in patients with both respiratory and plant food allergy (5, 6), and recent studies found that the oral allergy syndrome (OAS) induced by certain plant-derived foods such as melon, watermelon, banana, tomato, and citrus fruits is a sort of "trade mark" of profilin sensitization (5, 7, 8). Previous studies found that in birch pollen-allergic patients the occurrence of clinically relevant cross-reactivity to plant

foods is clearly related with the level of birch-specific IgE (9, 10); in contrast, whether in profilin-hypersensitive patients the development of a pollen-food allergy syndrome is directly related with the levels of specific IgE has been not been investigated so far. The main reasons for this are probably that "pure" profilin reactors are relatively uncommon and that only recently the enormous advances in molecular biology have resulted in the detection, purification, cloning, and expression of an increasing number of recombinant and/or natural allergen proteins from different sources, including profilin, that is now available for diagnostic purposes. The aim of the present study was to investigate whether the levels of profilin IgE are predictive of the development of food allergy in hypersensitive patients.

## Patients and methods

The study population was selected out from a large number of adults who spontaneously presented during 2010 at two allergy outpatient clinics sited in Pordenone and Paderno Dugnano with suspect airborne seasonal allergy (reporting a history of rhino-conjunctivitis with or without asthma for more than 1 month between February 15th and October 15th).

All patients underwent skin tests with a large panel of commercial pollen extracts including grass, ragweed, mugwort, birch, pellitory, cypress, olive, and plantain (Allergopharma, Reinbeck, Germany). Those scoring positive for > 3 distinct allergen sources were considered as potential profilin reactors (11) and underwent the detection of IgE to Phl p 12, the grass profilin, by ImmunoCAP (Phadia, Uppsala, Sweden). Those showing circulating IgE to Phl p 12 were included in the study provided they did not show any IgE reactivity to both the birch pollen major allergen, Bet v 1, and the peach lipid transfer protein (LTP), Pru p 3, the two other major causes of plant food allergy in Northern Italy (12). The absence of IgE reactivity to Pru p 3 was shown both in-vivo [by a negative SPT with a commercial peach extract containing LTP 30 µg/ml, and lacking both profilin and the Bet v 1-homologous protein (13)] and in-vitro. In Paderno Dugnano profilin hypersensitivity was confirmed also by a positive SPT with a commercial profilin-enriched date palm pollen extract (ALK-Abellò; Pho d 2 50 µg/ml) (5, 6). The study patients underwent a thorough interview aiming to ascertain whether they experienced a typical oral allergy syndrome (defined as oral itching with or without angioedema of the tongue and/or lips immediately following the ingestion of fresh and raw plant-derived foods). In patients reporting a positive clinical history sensitization was confirmed by SPT with fresh material. The possible influence of primary profilin-sensitizing pollen source in the occurrence of OAS was investigated as well. It was stated that the a primary allergen could be regarded as a likely cause of profilin sensitization only when IgE reactivity to the marker allergen (Phl p 1 and Phl p 5 for grass, Art v 1 for mugwort, Amb a 1 for ragweed, Par j 2 for pellitory, Bet v 1 for birch, Ole e 1 for olive, and Cup a 1 for cypress) exceeded that to the pan-allergen. In patients showing multiple reactivities to marker allergens all exceeding that to the pan-allergen, the primary sensitizer was considered as "not detectable". All in-vitro tests were carried out by ImmunoCAP (Phadia, Uppsala, Sweden) using a cut-off value of 0.35 kU/L.

As all in-vivo and in-vitro tests were performed within routine clinical activity of the participating centers no ethical committee approval was needed for the study and only a formal informed written consent by the patients was obtained.

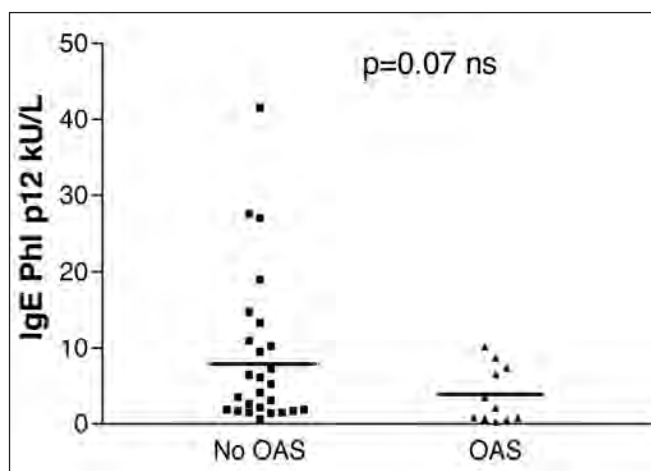
Statistical analysis was performed using the non-parametric Mann-Whitney U-test; statistical significance was set at a P-value <0.05.

## Results

Thirty-seven profilin reactors (M/F 17/20; mean age 37 years, range 19-66) were finally included in the study. 11/37 reported a typical oral allergy syndrome following the ingestion of fresh plant-derived-foods. Offending foods included Rosaceae (n= 6), melon and/or watermelon (n= 7), tomato (n= 3), orange (n= 3), carrot (n= 2), fig, walnut, grapes, and kiwi (n= 1 each). Surprisingly enough, patients without a history of OAS showed higher levels of IgE specific for Phl p 12 than patients with OAS (median 4.74 [range 0.7-41.6] KU/L vs 2.14 [range 0.32-10.2] KU/L, respectively) although the difference did not reach statistical significance (p=0.07) (Fig. 1).

The analysis the possible influence of the nature of primary profilin-sensitizing pollen source on the occurrence of OAS is shown in Table 1. No difference in the prevalence of primary sensitized pollen allergy was found between the two groups.

**Figure 1** - Levels of Phl p 12 specific IgE in patients with and without OAS



**Table 1** – Primary allergen sources in 37 subjects sensitized to profilin with or without oral allergy syndrome

	OAS+	OAS-	p
No.	11	26	NS
Grass	10	15	NS
Ragweed	5	17	NS
Olive	2	3	NS
Mugwort	2	3	NS
Pellitory	0	1	NS
Cypress	0	1	NS

## Discussion

In patients hypersensitive to food allergens it is generally agreed that higher specific IgE levels are associated with a higher likelihood to develop clinical allergy. This has been shown in studies of different food allergies such as egg, milk, peanut, or fish (14-20), in the case of peach lipid transfer protein (21), and also in the case of pollen food allergy syndrome occurring in birch pollen-allergic subjects (9, 10). This study investigated a cohort of profilin-hypersensitive patients without both birch pollen and lipid transfer protein allergy with the aim to detect whether in this type of food as well specific IgE levels play a role in the occurrence of the food allergy. The selected study population seemed highly representative of profilin-hypersensitive subjects. The prevalence of oral allergy syndrome was similar to that found in previous studies (5), and reportedly offending foods corresponded to those generally involved in profilin reactors (7, 8). Although other allergen sources, and hence other profilins may be the cause of the pollen-food allergy syndrome (8), Phl p 12 was chosen as a representative marker of profilin hypersensitivity because grass pollen profilins seem to be the most potent inducers of IgE against profilin (22), and because in a recent study we showed that one single profilin is sufficiently representative of hypersensitivity to the whole group of these highly homologous and cross-reacting allergens (23). Thus, it is likely that IgE levels to Phl p 12 are representative of the intensity of IgE reactivity to the whole group of cross-reacting profilins. In this sense, this study showed that, in these specific patients monosensitized to profilin, clinical food allergy is totally independent on the level of specific IgE. Further, the onset of pollen-food allergy syndrome did not depend on the nature or the number of primary sensitizing pollen sources. Thus, it must be admitted that

factors causing the onset of OAS in profilin-hypersensitive patients are presently obscure. One possibility is that patients developing OAS react to one or more specific isoforms of the pollen protein showing a higher degree of homology with the protein in plant foods. An alternative hypothesis is the existence of a different epitope recognition pattern between patients reactive or tolerant to plant foods. Further studies using assays containing different specific isoforms and peptide microarray-based immunoassay (24) are needed to confirm these hypotheses.

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