

Recombinant birch allergens (Bet v 1 and Bet v 2) and the oral allergy syndrome in patients allergic to birch pollen

M. De Amici, PhD*; M. Mosca, MD†; M. Vignini, MD‡; S. Quaglini, PhD‡; and R. Moratti, MD§

Background: IgE cross-reactivity between pollen and food allergens represents the molecular basis for oral allergy syndrome (OAS).

Objective: To evaluate specific IgE for Bet v 1 and Bet v 2 in the serum of patients sensitized to birch pollen and to identify whether IgE antibodies to Bet v 1 and Bet v 2 were predictors of OAS.

Methods: Thirty-three patients with skin prick test results and radioallergosorbent assay test results positive to birch pollen, 12 (36%) of whom had OAS symptoms, were enrolled in the study. Serum levels of specific IgE were determined by the fluoroenzyme immunoassay technique.

Results: The *t* test revealed significantly higher serum IgE levels against Bet v 1, Bet v 2, and birch pollen in the 12 symptomatic patients with respect to those without OAS (32.4 vs 12.4 kU/L, 7.6 vs 1.3 kU/L, and 42.3 vs 17.3 kU/L, respectively). Attempts to establish a threshold value of serum IgE antibirch pollen and the appearance of OAS revealed that a level of 20 kU/L or more yields an efficiency of the test equal to 70%.

Conclusions: In our study, quantitative birch specific IgE level proved useful in predicting clinical allergy symptoms with birch exposure.

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INTRODUCTION

Many atopic patients have an additional food-related allergy, often due to a cross-reactivity between pollen allergens (birch, hazelnut, alder, mugwort) and food allergens.¹ IgE cross-reactivity between pollen and food allergens represents the molecular basis for oral allergy syndrome (OAS), and it has been suggested that, in patients with OAS, pollen allergens may be responsible for the elicitation and maintenance of disease.² IgE positivity for apple was identified as the only predictive factor for the appearance of symptoms of OAS, with a relative risk of 99%.³

Techniques of genetic engineering applied to allergens have enabled the production of recombinant allergens.⁴ Most cross-reactions between birch pollen and apples are caused by the major birch allergen Bet v 1, one of a family of ubiquitous allergens responsible for allergic diseases, and by the homologous apple protein with a molecular weight of 18 kDa.^{5,6} Another birch allergen, Bet v 2, a protein with a molecular weight of 14 kDa associated with the cytoskeleton of all eukaryotic cells, has been cloned simultaneously from birch

and grass pollens.^{7–9} The purpose of this study was to evaluate the frequency of specific IgE for Bet v 1 and Bet v 2 in the serum of patients sensitized to birch pollen, 12 (36%) of whom had symptoms of OAS, and to identify whether IgE antibodies to Bet v 1 and Bet v 2 were predictors of OAS.

PATIENTS AND METHODS

Thirty-three patients (15 men and 18 women; mean age, 35.8 years; age range, 25–55 years) with skin prick test results and radioallergosorbent assay test results positive to birch pollen, 12 (36%) of whom had symptoms of OAS, were enrolled in the study. No patients were receiving specific immunotherapy.

Serum levels of specific IgE were determined by the fluoroenzyme immunoassay technique (CAP System; Pharmacia Diagnostic, Uppsala, Sweden)¹⁰ in peripheral blood samples from the patients. Specific IgE concentrations were expressed in kilounits per liter.

Statistical analysis was performed using the statistical software package S-Plus.¹¹ Means and SDs were used as descriptive statistics. The *t* test was used to evaluate the difference between pairs of patient groups. Finally, since OAS is a binary variable (present/absent), the logistic regression analysis was performed to find its possible predictors. Only univariate models were implemented because of the small sample size.

RESULTS

Three (9%) of the patients were negative for both recombinant birch allergens, 6 (18%) were positive to both allergens, 20 (61%) were positive only to Bet v 1, and 4 (12%) were

* Department of Pediatrics, University of Pavia, IRCCS Policlinic S. Matteo, Pavia, Italy.

† Department of Dermatology, University of Pavia, IRCCS Policlinic S. Matteo, Pavia, Italy.

‡ Dipartimento di Informatica e Sistemistica, University of Pavia, IRCCS Policlinic S. Matteo, Pavia, Italy.

§ Laboratory of Chemical Chemistry, University of Pavia, IRCCS Policlinic S. Matteo, Pavia, Italy.

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Table 1. Mean Serum IgE Levels of the Recombinant Birch Allergens and Birch Pollen in Patients with and without Oral Allergy Syndrome (OAS)*

No. of patients	OAS	Bet v 1	Bet v 2	Birch pollen	Bet v 1/ birch pollen	Bet v 2/birch pollen
12	Yes	32.4 ± 39.0	7.6 ± 13.5	42.3 ± 36.6	1.1 ± 0.7	0.3 ± 0.4
21	No	12.4 ± 13.0	1.3 ± 2.0	17.3 ± 21.8	0.9 ± 0.4	0.2 ± 0.4
P value		.03	.04	.01	.10	.73

* Data are presented in kilounits per liter as mean ± SD.

positive only to Bet v 2; thus, of the 30 patients with specific IgE to either or both recombinant birch allergens, 26 (87%) were positive for the major epitope and 10 (33%) for Bet v 2. The frequency of OAS in this small group of patients was 35% (7/20) in those sensitized to Bet v 1 and 50% in those sensitized to Bet v 2 alone (2/4) or to both Bet v 1 and Bet v 2 (3/6).

The *t* test, which was used to evaluate the differences between mean specific IgE values against Bet v 1, Bet v 2, and birch pollen in the presence or absence of OAS (Table 1), revealed significantly higher serum IgE levels against both recombinant allergens, as well as against T3 in the 12 patients symptomatic for OAS than in those without OAS. No differences between the 2 groups were observed in the ratios of Bet v 1 to birch pollen and Bet v 2 to birch pollen.

In addition, a univariate logistic regression analysis, applied to the data to evaluate whether serum IgE against any of the 3 allergens was a predictor of OAS, showed that specific IgE only against birch pollen was predictive of OAS. The regression coefficients for the logistic model were as follows: $\beta = -1.56$ and 0.05 , $t = -2.58$ and 1.94 , and $P = .01$ and $.02$ for the intercept and birch pollen groups, respectively. The probability of OAS was derived according to the following formula:

$$p = \frac{e^{-1.56 + 0.05 \times \text{Birch Pollen}}}{1 + e^{-1.56 + 0.05 \times \text{Birch Pollen}}}$$

Furthermore, attempts to establish a possible threshold value of serum IgE antibirch pollen and appearance of OAS revealed that a level of 20 kU/L or more yields an efficiency of the test equal to 70%, a positive predictive value of 60%, and a negative predictive value of 74%. The odds ratio is 1.5.

DISCUSSION

Detection of prognostic factors for OAS with birch pollen hypersensitivity is being investigated. Available data suggest that approximately 15% of patients with birch pollen hypersensitivity are not prone to OAS and their antibirch IgE might be directed against determinants that do not cross-react with food allergens.¹² In our study, 30 (91%) of the 33 patients allergic to birch pollen were sensitized to one or both recombinant birch allergens, 26 (87%) of 30 to the major epitope, and 10 (33%) of 30 to Bet v 2. The 12 patients with OAS all had specific IgE for one or both recombinant aller-

gens and significantly higher specific IgE levels against both recombinant allergens and birch pollen than those without OAS; none of the 3 patients with anti-T3 IgE alone reported OAS. However, only specific serum IgE antibirch pollen was a predictor of OAS, and an odds ratio of 1.5 in favor of OAS was found at a threshold value of 20 kU/L of serum IgE antibirch pollen.

Notwithstanding current knowledge that in vitro assay is not able to reflect functional immune responses,¹³ in our study quantitative birch specific levels proved useful in predicting clinical allergy symptoms with birch exposure. The use of modified recombinant allergens opens new research areas, which could, in the future, lead to a new modality of specific immunotherapy.⁴

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- Requests for reprints should be addressed to:*
M. De Amici, PhD
Department of Pediatrics
IRCCS Policlinic S. Matteo
P. le Golgi 2
27100 Pavia, Italy
E-mail: m.deamici@smatteo.pv.it
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