

## Systemic reactions to immunotherapy: influence of composition and manufacturer

G. Gastaminza\*, J. Algorta†, M. Audicana\*, M. Etxenagusia\*, E. Fernández\* and D. Muñoz\*

\*Servicio de Alergología, Hospital Santiago Apóstol and †Unidad de ensayos clínicos (Fundación Leia, CDT), Hospital de Txagorritxu, Osakidetza-Servicio Vasco de Salud, Vitoria-Gasteiz, Basque Country, Spain

### Summary

**Background** Although immunotherapy clearly demonstrated the benefit of reducing allergic symptoms, it has the drawback of adverse events, mainly systemic reactions that could be very inconvenient for patients and even life-threatening.

**Objective** The aim of the present study was to assess the incidence of systemic reactions to immunotherapy in a large number of patients, and its potential relationship with the characteristics of therapy, such as allergen composition or manufacturing laboratory.

**Methods** This study analysed the administration of specific immunotherapy during a period of 5 years, involving 1212 patients affected by respiratory hypersensitivity or hymenoptera venom anaphylaxis. Commercial extracts were supplied by five different laboratories. All the patients were attended at an out-clinic immunotherapy unit by the same experienced staff. Immunotherapy was given following a conventional schedule, modified according to the usual recommendations.

**Results** A total of 250 adverse reactions have been recorded, resulting in a frequency of 0.84% over the total number of injections. Seventy-nine of them (32%) were systemic reactions (0.27% SR/injection). The 79 systemic reactions were observed in 60 patients (5% of the patients). The frequency of systemic reactions was significantly lower ( $P < 0.01$ ) on the group of mites than on the other groups. The frequency of systemic reactions varies according to the manufacturing laboratory. In the case of mite extracts, although one of the laboratories had a lower frequency of adverse systemic reactions, it did not reach the level of statistical significance. However, in relation to pollen extracts, preparations of one of the manufacturers had a significantly lower frequency of systemic reactions. Concerning the time of occurrence, 27% of systemic reactions were delayed, thus they appeared at least 30 min after the vaccine injection, most of them due to pollen extracts.

**Conclusion** This is a preliminary study to evaluate the factors that could facilitate the appearance of systemic reactions demonstrating that not only the composition but also the manufacturer is connected to systemic reactions. Although further studies are needed to clearly establish the influence of manufacturer on frequency and time of appearance of systemic reactions, it seems necessary to reach a wide consensus on allergen extract standardization methods.

**Keywords** commercial preparations, delayed adverse effects, desensitization, human, immunologic/adverse effects, respiratory hypersensitivity/asthma, rhinitis, systemic adverse reactions

Submitted 30 August 2002; revised 9 December 2002; accepted 2 January 2003

### Introduction

Efficacy of specific immunotherapy to treat the causes of allergic diseases has been demonstrated by the appropriate methodology, including randomized, controlled, double-blind clinical trials [1].

Although immunotherapy has clearly demonstrated the benefit of reducing allergic symptoms in the medium-term, it is not free from risk. In fact, the main drawback of this treatment is the occurrence of adverse events, mainly systemic

reactions that could be very inconvenient for patients and even life-threatening.

The frequency of adverse reactions in general and more specifically the systemic reactions (SR) has decreased considerably over the last few years [2] due to the advances in the quality of the allergenic extracts, an improvement in standardization methods, and the comprehension that many adverse reactions occurred following their administration.

From another point of view, their consequences have also decreased thanks to the application of specific preventive measures, fundamentally those recommendations compiled by the European Academy of Allergology and Clinical Immunology (EAACI) [3], which have become the usual practice in Immunotherapy Units and that consist, essentially, of a strict control of the immunotherapy administration by trained physicians and

Correspondence: G. Gastaminza, Servicio de Alergología, Hospital Santiago Apóstol c/Olaguibel 29, 01004 Vitoria-Gasteiz, Spain.  
E-mail: ggastaminza@hsan.osakidetza.net

nurses, the permanence of the patient at the Unit for at least 30 min after injection, the use of allergenic biologically standardized extracts, etc.

The potency of the extracts used should be highlighted among the factors that influence the appearance of systemic reactions. But this potency depends on standardization methodology, which is different for each manufacturing laboratory. Therefore, there could be differences between the number of reactions produced by two extracts from different laboratories. Differences in the potency of the same allergen produced by two different laboratories have been demonstrated by several authors through skin tests [4]. Therefore, those differences do not depend on the medical care but on the standardization systems that should be unified instead.

Another factor that could influence the number of systemic reactions is the immunotherapy composition. In general, systemic reactions are more frequently induced by pollen extracts than by mites [5, 6].

The aim of the present study was to assess the incidence of systemic reactions to immunotherapy in a large number of patients, and its potential relationship with the characteristics of therapy, such as allergen composition or manufacturing laboratory.

## Materials and methods

This study analysed the administration of specific immunotherapy in an Allergology and Immunology Hospital during a period of 5 years (1996–2000). During the study period, 29 754 immunotherapy injections were administered (Table 1) to a total of 1212 patients, diagnosed with respiratory hypersensitivity (asthma = 683; rhinitis = 337) or hymenoptera venom anaphylaxis ( $n = 192$ ).

All the studied patients were sensitized to pollens (99% of them *gramineae*), mites (99% of them *Dermatophagoides pteronissinus* or *D. farinae*) or hymenoptera. Patients sensitized to other allergens (i.e. epithelia or moulds) have been excluded from the present analysis because of the small number of patients available.

In a retrospective design, data were obtained from the specific software for the immunotherapy control specially designed for the Unit. Since the software records only variables concerning the therapy but not characteristics of the patients (i.e. age or gender), such data could not be included in the analysis.

### Allergenic extracts

Only biologically standardized, unmodified, depot extracts were used throughout the period studied. Commercial extracts

were supplied by five different laboratories (in alphabetical order, ALK-Abelló, Bayer, Bial-Aristegui, Leti and IPI, Spain) except for the hymenoptera extracts which were supplied by the same manufacturer (ALK-Abelló). For the subsequent comparison between manufacturers, only three laboratories (ALK-Abello, Bial-Aristegui and Leti, re-denominated in a blind manner as A, B and C to maintain confidentiality) have been compared, because the others supplied extracts for less than 30 patients.

Standardization methods were as follows: ALK-Abelló establish that an allergenic extract contains 100 biologic units (BU) when the average size of the weal obtained per duplicate in a sample of 30 allergic patients is 75 mm<sup>2</sup> [7]; moreover, these extracts are quantified in mass-units, that means that an extract of 100 BU/mL must have X µg/mL of major and minor allergens semi-quantified with suitable techniques [8]. Bial-Aristegui determine that one SPT is the concentration of the extract (in mg/mL) that produces the same average weal as that obtained with histamine (10 mg/mL) in the same allergic population; one SPT of an extract of 1 mg/mL has a biological activity of 10 000 UBE/mL [9]. Leti establish that an allergenic extract has 10 HEP (Histamine Equivalent in Prick testing) when it causes on the skin a specific reaction consisting of a weal of the same average size as a positive reference of histamine 54.3 mmol/L (Histamine HCL in a concentration of 10 mg/mL), when both solutions are used in the same technique (prick test) in a minimum of 20 patients who are clinically allergic and have skin reactivity to the allergen in question [10, 11].

All the commercial preparations had approximately the same external appearance, with three flasks numbered I–III to be used during the initial period, plus a maintenance flask. Assignment of patients to each laboratory did not follow any specific pattern and was a result of casual, pseudo-randomized prescription.

### Administration schedule

All the patients were attended at the out-clinic immunotherapy unit. The injections were administered by the same three experienced nurses, belonging to the above mentioned Allergology Clinic. In accordance with the EAACI recommendation, immunotherapy had not been administered in any case of asthmatic patients that had a FEV<sub>1</sub> under 70% of predicted, and the other contraindications for the setting up of specific immunotherapy had also been strictly followed [3].

Immunotherapy was given following a conventional schedule. During the initiation period, an injection per week was administered until the maximal dose was reached (normally 0.8 mL from the III flask). Maintenance therapy was then administered once a month.

Before the administration of each immunotherapy dose, the nurses asked the patients about their disease stability from the previous visit and about the occurrence of other diseases during that period. In those patients suffering from rhinitis or bronchial asthma, a previous determination of the peak expiratory flow (PEF) was carried out, and in those patients on Hymenoptera venom treatment, blood pressure was measured.

When the patient had suffered an exacerbation of his disease, or had had PEF variations from the basal levels, the patient was

**Table 1.** Number of injections and patients analysed, according to allergen extract

|             | Injections |         | Patients |         |
|-------------|------------|---------|----------|---------|
|             | <i>n</i>   | (%)     | <i>n</i> | (%)     |
| Mites       | 15 992     | (53.7)  | 646      | (53.3)  |
| Pollen      | 8789       | (29.6)  | 374      | (30.9)  |
| Hymenoptera | 4973       | (16.7)  | 192      | (15.8)  |
| Total       | 29 754     | (100.0) | 1212     | (100.0) |

attended by one of the physicians who decided about modification of the treatment.

When the extract was a pollen (basically from *Gramineae*), in all cases the phase of initiation had not been applied during the pollination season (between May and July). All treatments with pollens were co-seasonal, but during the pollination period the maintenance dose was reduced to 50%, increasing it to the usual dose after the period. When a new batch was initiated, the first dose was also reduced to a 50% of the previous dose.

All the patients had to stay at the Immunotherapy Unit for at least 30 min after the application of the dose. In addition, the patients with respiratory disease had to carry out a new PEF determination before leaving.

### Classification of adverse reactions

Adverse reactions have been classified by their type and severity according to EAACI's recommendations [3]. Thus, local reactions had been considered as those whose induration was larger than 5 cm of diameter. Systemic reactions have been divided into mild: grade 2, moderate: grade 3, and severe: grade 4.

The aim of this study was to focus on systemic reactions. When a patient had suffered a systemic reaction, he was evaluated immediately by a physician who had set up the appropriate treatment and assessed their severity. In addition, the reaction onset time was been recorded, immediate reactions were classified as those that appear before 30 min following immunotherapy administration, and as delayed when they appeared after 30 min.

### Statistical methods

Comparison between groups has been performed by  $\chi^2$  test. The level of significance was set at  $P < 0.05$ .

## Results

A total of 250 adverse reactions have been recorded, resulting in a frequency of 0.84% over the total number of injections. Seventy-nine of them (32%) were systemic reactions (0.27% SR/injection). The majority of systemic reactions were mild (70.8%) or moderate (26.6%). Only two reactions (2.5%), one with a mite extract and the other with a pollen extract, had a non-fatal severe reaction.

The 79 systemic reactions were observed in 60 patients (5% of the patients). Forty-five patients suffered only one reaction, whereas 12 patients suffered two reactions, two patients suffered three reactions and one patient suffered four reactions. No differences have been found between the percentage of systemic reactions in patients suffering from rhinitis (6%) or asthma (4%).

The percentage of patients that have suffered a systemic reaction with each allergen type is shown in Fig. 1. The frequency of systemic reactions was significantly lower ( $P < 0.01$ ) on the group of mites than on the other groups: pollen or hymenoptera. Figure 2 also shows the grade of reactions. The majority of reactions were classified as mild in all the groups, and in the particular case of hymenoptera venom were almost always mild (96%). The influence of the former finding on the compliance of treatment is that 12 out of 28 patients (43%) suffering a systemic reaction to pollen immunotherapy had to

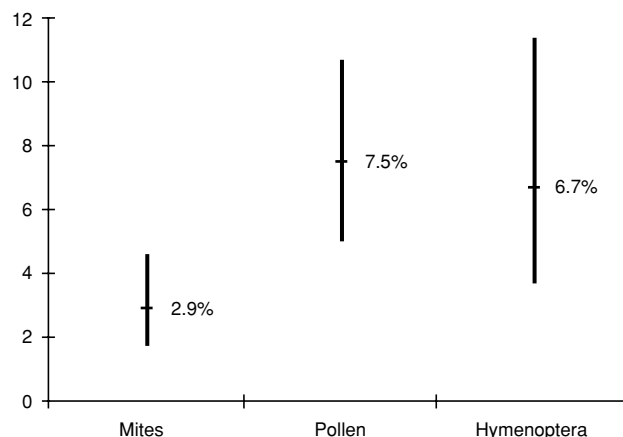


Fig. 1. Percentage of patients that have suffered a SR for each allergen group. The vertical line indicates the amplitude of the CI 95%.

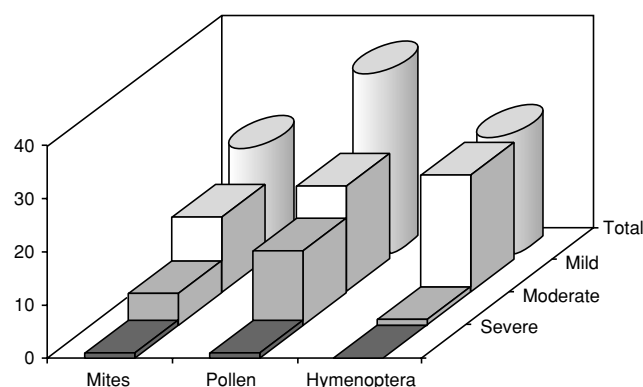


Fig. 2. Number of systemic reactions, classified by allergen and grade (square bars), and total number of systemic reactions (round bars).

discontinue the treatment because of the reactions, while only one patient (5.3%) with mite extract and another (7.7%) with hymenoptera dropped immunotherapy due to the adverse reactions.

Forty-one out of the 79 systemic reactions (52%) occurred during the initiation phase and 38 (48%) during the maintenance phase. According to the extract type, the systemic reaction percentage that occurred during the initiation is very high with venom extracts (73.9%), it decreases in pollen vaccines (48.5%), and it is very low with mites extracts (33% of the SR occurred at the initiation).

Laboratory A extracts were used in 431 allergic patients sensitized to mites (10 619 injections) and 139 allergic patients sensitized to pollen (3290 injections). Laboratory B extracts were used in 33 allergic patients to mites (599 injections) and 38 allergic patients to pollen (962 injections). Laboratory C extracts were used in 114 allergic patients to mites (3134 injections) and 101 allergic patients to pollen (2568 injections).

The frequency of systemic reactions varied according to the manufacturing laboratory (Fig. 3). In the case of mites extracts, although Laboratory C has a lower frequency of adverse systemic reactions, it does not reach the level of statistical significance. However, in relation to pollen extracts, preparations of one of the manufacturers (Laboratory A) had a significantly lower frequency of systemic reactions.

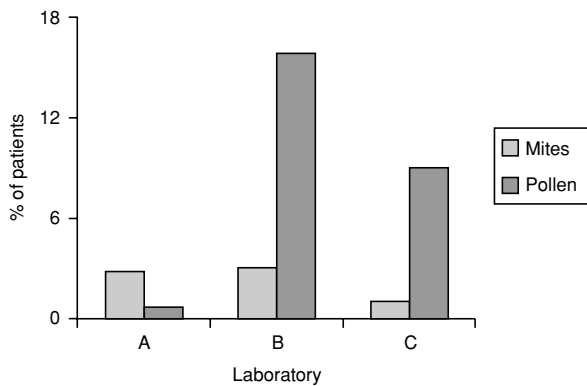


**Fig. 3.** Percentage of patients affected by systemic reactions according with the laboratory manufacturer and allergen.

Concerning the time of occurrence, 21 (27%) of the systemic reactions SR were delayed, thus they appeared at least 30 min after the vaccine injection. The majority of them were due to pollen extracts (18), only one occurred in the case of mites and two in the case of hymenoptera.

The severity of the delayed reactions (50% mild and 50% moderate) is not significantly higher than the immediate reactions (65% mild, 29% moderate and 6% severe). However, delayed reactions were responsible for most of the discontinuations on immunotherapy, since eight of the 12 patients who ceased treatment suffered from delayed systemic reaction.

## Discussion

The occurrence of adverse reactions to immunotherapy, and more specifically the systemic reactions, remains a worrying problem for the clinical allergologist. Despite the clear advances in the prevention of such reactions over the last few years, certain aspects remain still unclear, such as the influence of composition, or furthermore, the influence of the manufacturer on the incidence and characteristics of systemic reactions. The present study clearly reflects this worry in an allergology reference centre and aims to clarify the above mentioned items.

The frequency of appearance of systemic reactions described in the present study is almost the same (5% vs. 4.8%) to that reported by other authors in the same area [12], and is also very similar to other European centres [13, 14], studying a large number of more than 2000 patients. Although previous reliable data about adverse reactions frequency in our area is not available, the above described results confirm the efficacy of control measures advised by experts to reduce fatal reactions risk.

Compared to the study of Tabar et al. [12], it is remarkable that, in opposition to its findings of a higher frequency of systemic reactions in asthmatic patients, we found a similar frequency either in rhinitis or asthma. Another interesting difference is that, contrary to that described by most authors [12, 15–17], we found a frequency of SR during the maintenance phase similar or even higher in the case of mite extracts. This result could be explained by a high percentage of patients being sensitized to mites with perennial asthma suffering a reaction due to a minimum persistent inflammation caused by a low degree of mites exposure, as has been previously pointed out by Ciprandi et al. [18].

It is worth noting that the low level of reactions induced by mites vaccines, when compared with those of venom and pollen, are consistent with those previously described by other authors [5, 6, 19]. This difference is even more remarkable considering the presence of a bias in the patient selection at the Allergy Unit, with the presence of patients with more severe illness, the majority of them suffering from asthma. Moreover, this difference between extracts appears despite the administration of only half of the usual dose during the pollination season (May–July for *Gramineae*), as has been described previously in the Materials and methods section. In fact, none of the SR with pollen vaccines has occurred during those three months. In contrast to what has been described previously [13], SR with Hymenoptera venom vaccines have been, in general, less severe than those occurred with pollens or mites. Although the grade of SR has not been different between mites and pollen vaccine reactions, the latter have more frequently caused treatment abandonment or interruption.

Once the classic risk factors have been controlled, the choice of the commercial extract (always within biologically standardized extracts) is an important decision in order to reduce the number of SR. Our results clearly demonstrated a dramatic difference in the number of systemic reactions between the evaluated laboratories. Moreover, it should be considered a characteristic of the elaboration or standardization of the commercial extract itself, since the laboratory with less SR in the case of mite extracts is different than the laboratory with significantly less SR in pollens.

Although other published data did not discover such differences between laboratories [14, 20], our results highlight the importance of setting up standardization systems that are mandatory for all the allergenic extract manufacturing laboratories. These systems should standardize all the allergenic extracts potency from different commercial companies with the same composition, and calls for an international standardization system or at least a national reference. Furthermore, if the present results are confirmed by larger studies involving more laboratories and the assessment of safety as well as potency of extracts, the manufacturer would be a determinant factor to choose a therapy, in addition to other scientific or economic factors. It remains for further discussion whether this type of study is the responsibility of clinicians or must be a part of the above mentioned standardization system.

The EAACI recommendation that patients stay at the Immunotherapy Unit for at least 30 min after the administration of immunotherapy is considered especially useful in avoiding complications for the most severe adverse reactions which frequently occur within this period. A particularly worrying reaction type is the reaction that happens once the recommended stay time is finished at the Immunotherapy Unit, thus the reaction occurs outside the hospital, making the appropriate but otherwise simple medical surveillance more difficult.

In the present study, 51.4% of the systemic reactions caused by pollen vaccines were delayed for more than 30 min, in contrast to the low incidence of these type of reactions with mites or venom extracts. Delayed reactions have been already described by other authors [5], also involving pollen extracts [17, 21] and in many cases have consisted in urticaria, rhinitis and asthma. Similarly to those described by Winther et al. delayed reactions produced by *Gramineae* pollen extracts appear 1 to 2 h after the patient has left the clinic. Nevertheless there is no plausible

explanation for their origin. The study by Winther et al. compares pollen vaccines of *Gramineae* and birch, and concludes that immunotherapy with *Gramineae* pollen extracts is associated with a higher number, and more severe, systemic reactions.

Despite the fact that the majority of fatal systemic reactions with immunotherapy occur before 30 min [22], delayed systemic reactions induce more frequently the abandonment of immunotherapy than immediate ones do. This could be because reactions occur when the established surveillance time has finished and the patient is at home or in the workplace and usually suffers more anxiety. In most cases, patients have gone to an emergency service because of the severity of the reaction (in many occasions generalized urticaria and asthma).

These are the reasons why delayed systemic reactions cause great concern to both clinician and patient, mainly because they escape from the control measures applied to improve tolerance. In 1986, Greenberg et al. [5] pointed out the fact that delayed reactions (those that may compromise the patient's life) occur mainly with pollen extracts, which in most cases are usually employed for the treatment of a disease that does not threaten life. This author even proposed in patients at risk, a longer monitoring time (up to 2 h), which is questionable as it increases treatment discomfort and therefore patient acceptance. Furthermore, this worrying factor could be the explanation for the above-mentioned finding of a higher level of discontinuation on therapy in patients treated with pollen extracts than with the other assessed allergens.

In conclusion, this is a preliminary study to evaluate the factors that could facilitate the appearance of systemic reactions demonstrating that not only the composition but also the manufacturer is related to systemic reactions. Although further studies are needed to clearly establish the influence of the manufacturer on the frequency and time of appearance of systemic reactions, it seems necessary to reach a wide consensus on allergen extract standardization methods.

## Acknowledgements

The authors are indebted with the staff at the Unit of Immunotherapy (mainly, nurses Conchi, Blanca and Lidia), for their daily practical assistance and useful help in worrying situations, making possible the existence of the Unit. The authors also thanks M. Ismael Barbero for statistical advice and support. We also thank Lina Audicana for revising the English translation of the manuscript.

## References

- 1 Bousquet J, Lockey RF, Malling H-J, eds. WHO Position Paper. Allergen immunotherapy: therapeutic vaccines for allergic diseases. *Allergy* 1998; 53 (Suppl. 44):1–42.
- 2 Luigi A, Senna G, Mezzelani P, Pappalardo G. Safety of specific immunotherapy: a retrospective study. *J Invest Allergol Clin Immunol* 1994; 4:250–4.
- 3 Malling HJ, Weeke B. EAACI Immunotherapy position papers. *Allergy* 1993; 48 (Suppl. 14):9–35.
- 4 Hillas JL, Wilson JD. Variation in potency of extracts used for allergy skin tests. *NZ Med J* 1981; 93:2–4.
- 5 Greenberg MA, Kaufman CR, González GE, Rosenblatt CD, Smith LJ, Summers RJ. Late and immediate systemic-allergic reactions to inhalant allergen immunotherapy. *J Allergy Clin Immunol* 1986; 77:865–70.
- 6 Vervloet D, Khairallah E, Arnaud A, Charpin J. A prospective national study of the safety of immunotherapy. *Clin Allergy* 1980; 10:59–64.
- 7 Lombardero M, González R, Duffort O et al. Evaluación de la actividad biológica total y composición alérgica de extractos alérgicos. *Allergol Immunopathol* 1986; 14:189–98.
- 8 Carreira J, Lombardero M, Ventas P. New developments in *in vitro* methods. Quantification of clinically relevant allergens in Mass Units. In: Kurth R, Hausteiner D, eds. *Regulatory Control and Standardization of Allergenic Extracts*. Stuttgart: Gustav Fischer 1994, 155–64.
- 9 Martínez A, Martínez J, Llamazares A, Palacios R. Comparison of in-house reference extracts of *Dermatophagoides pteronyssinus* and *Phleum pratense* with the HHO International Standards. *Allergol Immunopathol* 1992; 20:106–12.
- 10 Belin L. Biologic standardization – the HEP unit. In: Arb Paul Ehrlich Institute Georg Speyer Haus Ferdinand Blum Institute Frankfurt A M 1987, 145–53.
- 11 Nordic Council on Medicines. Registration of allergen preparations. Nordic guidelines. Prepared by the Nordic Council on Medicines in cooperation with the Drug Regulatory Authorities in Denmark, Finland, Iceland, Norway and Sweden. NLN Publication no. 23. 1989, 1–36.
- 12 Tabar AI, García BE, Rodríguez A, Olaguibel JM, Muro MD, Quirce S. A prospective safety-monitoring study of immunotherapy with biologically standardized extracts. *Allergy* 1993; 48:450–3.
- 13 Malling HJ. Minimising the risks of allergen-specific injection immunotherapy. *Drug Safety* 2000; 23:323–32.
- 14 Ragusa FV, Passalacqua G, Gambardella R et al. Nonfatal systemic reactions to subcutaneous immunotherapy: a 10-year experience. *J Invest Allergol Clin Immunol* 1997; 7:151–4.
- 15 Mosbech H, Dirksen A, Dreborg S et al. Hyposensitization in asthmatics with mPEG-modified and unmodified house dust mite extract. *Allergy* 1990; 45:142–50.
- 16 Stewart GE, Lockey RF. Systemic reactions from allergen immunotherapy. *J Allergy Clin Immunol* 1992; 90:567–78.
- 17 Winther L, Malling H-J, Mosbech H. Allergen-specific immunotherapy in birch- and grass-pollen-allergic rhinitis. II. Side-effects. *Allergy* 2000; 55:827–35.
- 18 Ciprandi G, Buscaglia S, Pesce G et al. Minimal persistent inflammation is present at mucosal level in patients with asymptomatic rhinitis and mite allergy. *J Allergy Clin Immunol* 1995; 96:971–9.
- 19 Osterballe O. Side effects during immunotherapy with purified grass pollen extracts. *Allergy* 1982; 37:553–62.
- 20 Igea JM, Lázaro M. Safety of grass immunotherapy administered in a specialized unit: study of risk factors. *Allergol Immunol Clin* 2002; 17:16–22.
- 21 Toubi E, Kessel A, Blant A, Golan TD. Follow-up after systemic adverse reactions of immunotherapy. *Allergy* 1999; 54:617–20.
- 22 Lockey RF, Benedict LM, Turkeltaub PC, Bukantz SC. Fatalities from immunotherapy and skin testing. *J Allergy Clin Immunol* 1987; 79:660–77.