

Safety of an Ultra-Rush Immunotherapy Build-Up Schedule with Therapeutic Vaccines Containing Depigmented and Polymerized Allergen Extracts

M. Casanovas R. Martín C. Jiménez R. Caballero E. Fernández-Caldas

Laboratorios LETI, S.L., Tres Cantos, Madrid, Spain

Key Words

Immunotherapy, ultra-rush · Allergen vaccines, modified

Abstract

Background: Four administration schedules of immunotherapy have been reported: conventional, cluster, rush and ultra-rush. **Objectives:** To evaluate the safety and the clinical advantage of using standardized modified allergen extracts in an ultra-rush protocol without premedication and/or hospitalization. **Material and Methods:** One thousand and sixty-eight patients with rhinoconjunctivitis and/or asthma sensitized to mites and/or pollen were included in a prospective observational study. Patients received a therapeutic vaccine containing depigmented and glutaraldehyde-modified extracts (mites and/or pollens) adsorbed onto alum prescribed by a specialist. The schedule of administration consisted of injecting 0.2 and 0.3 ml of the vial of maximum concentration during the first day of immunotherapy, separated by a time interval of 30 min. All patients reached the maximum dose (0.5 ml) after 2 injections. Tolerance was assessed by recording all side reactions related to immunotherapy, classified according to the criteria of the EAACI. **Results:** The total number of injections was 2,136. All patients reached the maximum established dose on the 1st day. No premedication was used. Seven clinically

relevant local reactions were recorded. The systemic reactions were 5 grade-1 (2 immediate and 3 delayed) and 3 delayed grade-2 reactions. **Conclusions:** The therapeutic vaccines containing chemically modified extracts can be administered using an alternative ultra-rush schedule in an immunotherapy unit, reaching the maximum dose on the 1st day with 2 injections, without the need of premedication and/or hospitalization.

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Introduction

The efficacy of immunotherapy has been reviewed using the standards for evidence-based medicine [1, 2]. It is currently accepted as the only treatment of allergic disease that can modify the natural course of the disease and at the same time ameliorate symptoms [3, 4]. The efficacy is dose-dependent, thus, the concept of optimal dose is used, and defined as the dose of allergen vaccine inducing a relevant clinical effect in most patients without causing unacceptable side effects [5, 6]. The drawbacks of allergen immunotherapy relate to its safety profile, compliance and possibly cost [7]. The use of a traditional build-up schedule of administration, in which weekly injections are given, usually needs 12–16 weeks to arrive at the maintenance dose and may contribute to reducing compliance with this therapy [8]. The administration of

higher doses of allergen extracts at the beginning, and/or cluster or rush schedules [9–14], has the advantages of reaching the maintenance doses within days and provides a more rapid appreciation of the clinical benefits of immunotherapy [15]. The disadvantages of these schedules are time commitment, the use of hospital facilities to closely monitor the patients during this period, and the greater risk of systemic reactions.

In an attempt to overcome these disadvantages, glutaraldehyde-modified allergen extracts were developed, proving to be safer than unmodified vaccines while retaining clinical efficacy [16, 17]. The introduction of a depigmentation step, before the polymerization process removes the adsorbed pigments, enhances the solubility of the allergoids and inactivates the enzymatic activity [18]. These recently developed allergoids have been used in clinical studies of efficacy, safety and tolerability [19–26]. These vaccines maintain or increase IgG recognition, while significantly reducing IgE reactivity, which allows shortening of the build-up phase and the administration of higher doses than those used with non-modified allergens.

The objective of the study was to evaluate the safety of the build-up phase of vaccines containing these modified allergen extracts administered in ultra-rush schedule. Safety was evaluated following the criteria of the European Academy of Allergy and Clinical Immunology (EAACI) [27].

Material and Methods

Design

This phase-IV study, conducted in 66 allergy units in Spain and Portugal, was observational and prospective in a cohort of patients and open-labeled.

Calculation of Sample Size

Rush immunotherapy studies using non-modified allergen vaccines without premedication showed that this schedule carries a great risk of systemic reactions. The use of premedication reduced the incidence of systemic reactions from 36 to 16% in the best case [28]. Expecting to reduce this incidence more, and to have around 9–10% of systemic reactions with this modified preparation, without the use of premedication or hospitalization, for an α of 0.05 and a power of 90% (error protection of 10.5) [29], the cohort of patients needed was between 927 and 1,309.

Patients

A cohort of 1,068 patients (549 male and 519 female) with a mean age of 26.2 years (95% confidence interval 25.3–27.1) were included; in 9.6% of the patients the age range was between 3 and 10 years of age; in 23.6% between 11 and 18; 41.6% between 19 and 35, and 22.5% were older than 35 years. No age data were provided for 2.73% of the patients. Patients were either sensitized to

mites (*Dermatophagoides pteronyssinus*, *Dermatophagoides farinae*, *Blomia tropicalis*, *Lepidoglyphus destructor* and *Tyrophagus putrescentiae*) or pollens (grasses, *Olea europaea*, *Parietaria judaica*, *Cupressus arizonica*, *Salsola kali*, *Betula alba*, *Artemisia vulgaris* and *Chenopodium album*). The criteria of inclusion and exclusion to receive specific immunotherapy were those outlined in the EAACI and WHO Position Papers [3, 27]. Five hundred and seven patients (47.5%) were diagnosed with rhinoconjunctivitis, 471 (44.1%) with rhinoconjunctivitis and mild to moderate asthma and 57 (5.3%) with mild to moderate asthma. All patients had positive IgE (CAP >2) and positive skin tests (wheal size equal or greater than the wheal induced by histamine HCl 10 mg/ml) to the offending allergen or allergens which were included in the therapeutic vaccine. All patients had at least 2 years of symptoms but none had received immunotherapy before. All patients received the appropriate medication according to their disease, including antihistamines, nasal and/or bronchial steroids, short- and long-acting bronchodilators and anti-leukotrienes.

Immunotherapy

Each patient received an individualized commercial vaccine of the modified allergen extract adsorbed to aluminum hydroxide (Depigoid®, Laboratorios LETI, Tres Cantos, Spain). Immunotherapy with therapeutic vaccines containing modified pollen extracts was initiated outside the pollen season. The treatment was started with the vial containing the maximum recommended concentration (vial No. 2). The first dose contained 0.2 ml and was injected in one arm, followed by a 0.3-ml injection 30 min later in the other arm. The accumulated dose after the first two injections (0.5 ml) is the recommended maintenance dose. None of the patients received preventive pharmacological treatment or hospitalization. All patients had a 30-min waiting period in the immunotherapy unit after each injection, following the recommendations of the EAACI [27]. The composition of the vaccines administered is shown in table 1.

Table 1. Composition of the therapeutic vaccines administered to the patients

Composition	n	%
Mites		
<i>D. pteronyssinus</i>	275	60.31
<i>D. pteronyssinus</i> + <i>D. farinae</i>	175	38.38
Other mites (mixtures or individual)	6	1.32
Total patients receiving immunotherapy with mite extracts	456	100.00
Pollen		
Grasses (mix or single grass pollen extracts)	201	32.84
Mixture of grasses + <i>Olea europaea</i>	56	9.15
Weeds (individual pollen extracts)	102	16.67
Weeds (mixtures of 2 or more pollen extracts)	142	23.20
Trees (individual pollen extracts)	28	4.58
Trees (mixtures of 2 or more pollen extracts)	83	13.56
Total patients receiving immunotherapy with pollen extracts	612	100.00

Mites were mainly *D. pteronyssinus* and *D. farinae*. Other mites were *B. tropicalis*, *L. destructor* and *T. putrescentiae*. The grass mix contained *Lolium perenne*, *Phleum pratense*, *Poa pratensis*, *Dactylis glomerata* and *Festuca elatior*. The weeds were *P. judaica*, *S. kali*, *A. vulgaris* and *C. album*, and trees were *O. europaea* and *B. alba*.

The concentration of the modified allergen extracts, and major allergen content, in the vaccines has been reported previously [19–21, 25, 26]. Due to the polymerization process, the concentration of major allergens could not be measured in the final product. Thus, following the recommendations of the European Pharmacopoeia [30], major allergens were measured in the native preparation before the modification process, expressing the data as the amount in micrograms of these major allergens contained in the freeze-dried native extract needed to produce the maximum dose of the modified preparation. Major allergens of *D. pteronyssinus* and *D. farinae*

were measured using the monoclonal antibodies (Indoor Biotechnologies Ltd., Charlottesville, Va., USA), whereas the major allergens of pollens (*P. pratense*, *D. glomerata*, *P. judaica* and *O. europaea*) were measured by means of scanning densitometry [31]. Table 2 shows these values.

Assessment of Safety

The reactions were categorized as immediate when the onset was during the first 30 min after the administration, and delayed when the onset was after this time. Local reactions were classified measuring the diameter of the reaction. The immediate reactions with a diameter of <5 cm and the delayed reactions of <10 cm were considered as clinically irrelevant as a reduction in the following planned dose is not recommended [32]. The systemic reactions were graded according to the classification of the EAACI Position Paper [27].

Results

Patients and Immunotherapy

All 1,068 patients received the planned doses. A total of 2,136 injections were administered. There was no need to repeat or reduce the previously established doses. All patients were informed of the nature and the risks of the treatment and gave their consent to be treated.

Adverse Reactions

Seven clinically relevant local reactions were recorded from a total of 85 (40 immediate and 45 delayed). Table 3 shows the number and the intensity of these reactions per dose injected.

There were 8 systemic reactions: 2 immediate grade-1 reactions and 6 delayed (3 of grade 1 and 3 of grade 2)

Table 2. Quantity of major allergens contained in the freeze-dried native extract needed to produce the maximum dose of the modified preparation

Major allergen	µg
Der p 1	15.26
Der p 2	9.22
Der f 1	14.25
Phl p 1	4.35
Phl p 5	6.74
Dact g 2	30.00
Dact g 1+5	10.00
Ole e 1	65.50
Par j 1	7.50

Table 3. Number and intensity (mm of diameter) of local reactions

Diameter cm	Local reactions								
	0.2 ml	% per patient	% per injection	0.3 ml	% per patient	% per injection	total	% per patient	% per injection
<i>Immediate</i>									
<5	20	1.87	0.94	16	1.50	0.75	36	3.37	1.69
5–10	1	0.09	0.05	2	0.19	0.09	3	0.28	0.14
>10	0	0.00	0.00	1	0.09	0.05	1	0.09	0.05
Total	21	1.97	0.98	19	1.78	0.89	40	3.75	1.87
<i>Delayed</i>									
<5	4	0.37	0.19	7	0.66	0.33	11	1.03	0.51
5–10	10	0.94	0.47	21	1.97	0.98	31	2.90	1.45
>10	1	0.09	0.05	2	0.19	0.09	3	0.28	0.14
Total	15	1.40	0.70	30	2.81	1.40	45	4.21	2.11

reactions. All the immediate reactions occurred with the injection of 0.3 ml. Only 1 reaction was recorded using a modified mite (*L. destructor*) preparation, the rest of the systemic reactions being due to pollen. All these

reactions were mild and resolved spontaneously without the need of rescue medication. Table 4 shows the number and intensity of these reactions, and table 5 the description.

Table 4. Number and grade of systemic reactions

	Systemic reactions					
	immediate			delayed		
	total	% per patient	% per injection	total	% per patient	% per injection
Grade 1	2	0.19	0.09	3	0.28	0.14
Grade 2	0	0.00	0.00	3	0.28	0.14
Total	2	0.19	0.09	6	0.56	0.28

Table 5. Description of systemic reactions

Patient No.	Age years	Gender	Depigoid® (composition)	Diagnosis	Grade (EAACI)	Time of appearance	Description
11	25	Female	Grass mix	RC, A	2	Delayed	Rhinitis and asthma, both mild. Appearance 12 h after injection. The symptomatology disappeared spontaneously without medication
188	38	Female	<i>Betula alba</i> 30% + <i>Olea europea</i> 70%	RC, A	1	Delayed	Mild feeling of discomfort. Appearance 96 h after injection. The symptomatology disappeared spontaneously without medication
189	22	Female	Grass mix	RC, A	1	Delayed	Mild feeling of discomfort. Appearance 72 h after injection. The symptomatology disappeared spontaneously without medication and it was not a problem for the patient
262	59	Female	<i>Salsola kali</i>	RC, A	1	Delayed	Mild feeling of discomfort. Appearance 6 h after injection. The symptomatology disappeared spontaneously without medication and it was not a problem for the patient
315	25	Female	Grass mix	RC, A	2	Delayed	Rhinitis and asthma, both mild. Appearance 12 h after injection. The symptomatology disappeared spontaneously without medication
864	27	Female	<i>Phleum pratense</i>	RC, A	1	Immediate	Vasovagal syncope. Appearance 30 min after injection
888	13	Female	<i>Lepidoglyphus destructor</i>	RC, A	1	Immediate	Feeling of discomfort. The symptomatology disappeared spontaneously without medication and it was not a problem for the patient
976	45	Female	<i>Parietaria judaica</i> 34% + <i>Phleum pratense</i> 33% + <i>Artemisia vulgaris</i> 33%	RC, A	2	Delayed	Mild rhinitis. Appearance 24 h after injection. The symptomatology disappeared spontaneously without medication

RC = Rhinoconjunctivitis; A = asthma.

Discussion

In this prospective open study, a cohort of 1,068 patients were treated with a therapeutic vaccine containing depigmented and polymerized allergen extracts and the maximum dose was reached on the 1st day of treatment after 2 injections without the need of premedication or hospitalization. There were 7 clinically relevant local reactions (0.66 per patient and 0.33 per injection), and 3 delayed systemic grade-2 reactions (0.28 per patient and 0.14 per injection). No systemic grade-3 or -4 reactions were recorded. In another study [33], the same modified allergen extract was administered to 175 patients using a conventional schedule (the maximum dose was reached over a build-up phase of 4 injections in 3 weeks) with a total of 2,261 injections: 1.4% of the injections caused local reactions with a diameter between 5 and 10 cm; 0.1% were larger than 10 cm, and there were 13 systemic reactions which were all judged as mild.

The results of rush immunotherapy studies using non-modified or formaldehyde-modified allergen vaccines without premedication suggest that this schedule carries a great risk of systemic reactions, ranging from 27 [34] to 100% [35] of the patients treated. In 2 studies, premedicating the patients reduced the incidence of systemic reactions from 36 to 16% [28] and from 73 to 27% [36], which is similar to the incidence of systemic reactions that occur using conventional build-up schedules (0.05–3.2% per injection or 0.84–46.7% of the patients) as shown by Stewart and Lockey [37].

The use of rush immunotherapy makes it possible to reach the maintenance dose after a short period of time. Previous studies reported the need of 1–7 days to reach the top dose [11, 36, 38–40] with inhalant allergen extracts. Sharkey and Portnoy [38] described a rush immunotherapy schedule in 22 patients who were premedicated on the previous 3 days. The patients reached the maintenance dose after 8 injections within a period of 6 h. Systemic reactions (1 anaphylaxis) occurred in 5 patients (22%). In our study, the patients were not premedicated and the maximum dose was reached after the 2nd injection. The time consumed was 1 h, with a 30-min waiting

period after each injection, and the incidence of systemic reactions was low and severity mild.

Glutaraldehyde-polymerized allergen extracts have been extensively studied and have confirmed efficacy and good safety profile when administered in conventional and accelerated schedules [41–44]. However, no studies have been conducted using rush or ultra-rush schedules. The safety and efficacy of depigmented and glutaraldehyde polymerized extracts has also been studied using conventional build-up schedules [19–22, 26]. These extracts have shown efficacy using objective parameters, such as specific bronchial provocation and skin tests, demonstrating improvement after 6 months of treatment [22] or after 4 months when a shorter build-up schedule (cluster) was used [23]. They also have a wider safety profile than unmodified preparations since doses greater than those recommended can be administered without moderate or serious adverse reactions [24].

In this report, we do not attempt to address clinical efficacy, but in view of the previous results on the onset of objective clinical efficacy, we can speculate that the use of this ultra-rush schedule could probably provide clinical benefit in a short period of time.

Rush immunotherapy has several advantages over conventional or cluster schedules [38]: better compliance, cost effectiveness, and a reduction in dosage errors since most patients can reach the maintenance dose in one or few days. The introduction of premedication provides a safety profile similar to that of conventional schedules [38]. Ultra-rush immunotherapy using depigmented and polymerized allergen extracts is safer than conventional or rush schedules using non-modified and formaldehyde-modified preparations, reaching the maximum dose in a short period of time (2 injections in 1 day), without the need of premedication or hospitalization.

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References

- 1 Abramson MJ, Puy RM, Weiner JM: Allergen immunotherapy for asthma. *Cochrane Database Syst Rev* 2000;CD001186.
- 2 Abramson MJ, Weiner JM: Allergen-specific immunotherapy for asthma. Metanalysis of randomized controlled trials; in Bousquet J, Yssel H (eds): *Immunotherapy in Asthma*. New York, Dekker, 1999, pp 207–237.
- 3 Bousquet J, Lockey R, Malling HJ: Allergen immunotherapy: therapeutic vaccines for allergic diseases. *WHO Position Paper. Allergy* 1998;53(suppl 54):1–42.

- 4 Moller C, Dreborg S, Ferdousi HA, Halken S, Host A, Jacobsen L, et al: Pollen immunotherapy reduces the development of asthma in children with seasonal rhinoconjunctivitis (the PAT-study). *J Allergy Clin Immunol* 2002; 109:251–256.
- 5 Creticos PS, Van Metre TE, Mardiney MR, Rosenberg GL, Norman PS, Adkinson NF Jr: Dose response of IgE and IgG antibodies during ragweed immunotherapy. *J Allergy Clin Immunol* 1984;73:94–104.
- 6 Haugaard L, Dahl R, Jacobsen L: A controlled dose-response study of immunotherapy with standardized, partially purified extract of house dust mite: clinical efficacy and side effects. *J Allergy Clin Immunol* 1993;91:709–722.
- 7 Williams AP, Krishna MT, Frew AJ: The safety of immunotherapy. *Clin Exp Allergy* 2004; 34:513–514.
- 8 Cohn JR, Pizzi A: Determinants of patient compliance with allergen immunotherapy. *J Allergy Clin Immunol* 1993;91:734–737.
- 9 Freeman J: 'Rush' inoculation. *Lancet* 1930;i:744–747.
- 10 Moreno C, Fernández-Távora L, Acero S, Alonso MD, Barahona MJ, Blanco R, et al: Tolerance of a cluster schedule on the treatment of seasonal allergic respiratory disease with pollen extracts quantified in mass units. *J Investig Allergol Clin Immunol* 2003;13:221–227.
- 11 Bousquet J, Calvayrac P, Guerin B, Hejjaoui A, Dhivert H, Hewitt B, et al: Immunotherapy with a standardized *Dermatophagoides pteronyssinus* extract. I. In vivo and in vitro parameters after a short course of treatment. *J Allergy Clin Immunol* 1985;76:734–744.
- 12 García-Ortega P, Merelo A, Marrugat J, Richart C: Decrease of skin and bronchial sensitization following short-intensive scheduled immunotherapy in mite-allergic asthma. *Chest* 1993;103:183–187.
- 13 Portnoy J, King K, Kanarek H, Horner S: Incidence of systemic reactions during rush immunotherapy. *Ann Allergy* 1992;68:493–498.
- 14 Serrano P, Algorta J, Martínez A, González-Quevedo T, Velázquez E, Díaz M: Prospective safety study of immunotherapy administered in a cluster schedule. *J Investig Allergol Clin Immunol* 2004;14:312–319.
- 15 Sheffer AL: Allergen-specific immunotherapy: a role for T-cell anergy. *Ann Allergy* 1993;71: 327–329.
- 16 Grammer LC, Shaughnessy MA, Patterson R: Modified forms of allergen immunotherapy. *J Allergy Clin Immunol* 1985;76:397–401.
- 17 Bousquet J, Michel FB: Safety considerations in assessing the role of immunotherapy in allergic disorders. *Drug Saf* 1994;10:5–17.
- 18 Berrens L, Gallego M, Boluda L, Fernández-Caldas E, González L: Complexed flavonoids on pollen protein allergens revealed by uv-spectroscopy and thin-layer chromatography. *Allergy* 1998;53(suppl):56.
- 19 Guerra F, Daza JC, Almeda E: Immunotherapy with a depigmented, polymerized vaccine of *Olea europaea* pollen allergens significantly reduces specific bronchial and skin test reactivity in sensitized patients after one year of treatment. *J Investig Allergol Clin Immunol* 2003; 13:108–117.
- 20 García-Sellés J, Pascual A, Funes E, Pagán JA, López JD, Negro JM, et al: Clinical efficacy and safety of a depigmented and glutaraldehyde polymerized therapeutic vaccine of *Parietaria judaica*. *Allergol Immunopathol (Madrid)* 2003;31:63–69.
- 21 Ameal A, Vega-Chicote JM, Fernández S, Miranda A, Carmona MJ, Rondón MC, et al: Double-blind and placebo-controlled study to assess efficacy and safety of a modified allergen extract of *Dermatophagoides pteronyssinus* in allergic asthma. *Allergy* 2005;60:1178–1183.
- 22 Ferrer A, García-Sellés J: Significant improvement in symptoms, skin test, and specific bronchial reactivity after 6 months of treatment with a depigmented, polymerized extract of *Dermatophagoides pteronyssinus* and *D. farinae*. *J Investig Allergol Clin Immunol* 2003;13: 244–251.
- 23 Ibero M, Ridao M, Artigas R, Treserras R, Casanovas M: Immunotherapy with an extract standardized and with an allergoid of *D. pteronyssinus*. Evolution of the bronchial specific and unspecific reactivity. *Allergy Clin Immunol News* 1994(suppl 2):390.
- 24 Casanovas M, Fernández-Caldas E, Alamar R, Basomba A: Comparative study of tolerance between unmodified and high doses of chemically modified allergen vaccines of *D. pteronyssinus*. *Int Arch Allergy Appl Immunol* 2005; 137:211–218.
- 25 Casanovas M, Gómez MJ, Carnés J, Fernández-Caldas E: Skin tests with native and chemically modified allergen extracts. *J Investig Allergol Clin Immunol* 2005;15:30–36.
- 26 Álvarez-Cuesta E, Aragoneses-Gilsanz E, Martín-García C, Berges-Gimeno P, González-Mancebo E, Cuesta-Herranz J: Immunotherapy with depigmented glutaraldehyde-polymerized extracts: changes in quality of life. *Clin Exp Allergy* 2005;35:572–578.
- 27 Malling HJ, Weekes B: Immunotherapy. Position paper of the European Academy of Allergy and Clinical Immunology. *Allergy* 1993; 48(suppl 14):9–35.
- 28 Hejjaoui A, Dhivert H, Michel FB, Bousquet J: Immunotherapy with a standardized *Dermatophagoides pteronyssinus* extract. IV. Systemic reactions according to the immunotherapy schedule. *J Allergy Clin Immunol* 1990;85: 473–479.
- 29 Carnés X, Moreno V, Porta Serra M, Velilla E: Calculating the number of patients necessary for designing a clinical study (in Spanish). *Med Clin (Barc)* 1989;92:72–77.
- 30 Council of Europe. European Pharmacopeia. Allergen Products, ed 3. Strasbourg, Council of Europe, 1997.
- 31 Boluda L, Sastre J, Casanovas M, Fernández-Caldas E: Determination of Ole e 1 by enzyme immunoassay and scanning densitometry. Validation by skin-prick testing. *J Immunol Methods* 1999;223:17–26.
- 32 Álvarez Cuesta E, Boquete M, Cadahía A, Carrillo T, Fernández-Távora L, Hernández J, et al: Sociedad Española de Alergología e Inmunología Clínica. Normativa sobre Inmunoterapia en Enfermedades Alérgicas. Madrid, SANED, 1990.
- 33 Sager A, Renner BG: Safety aspects and course of allergen-specific immunotherapy with a depigmented, polymerized allergen extract. *Allergo J* 2004;13:386–391.
- 34 Bousquet J, Hejjaoui A, Soussana M, Michel FB: Double-blind, placebo-controlled immunotherapy with mixed grass-pollen allergoids. IV. Comparison of the safety and efficacy of two dosages of a high-molecular-weight allergoid. *J Allergy Clin Immunol* 1990;85:490–497.
- 35 Lilja G, Sundin B, Graff-Lonnevig V, Hedlin G, Heilborn H, Norrönd K, et al: Immunotherapy with cat- and dog-dander extracts. IV. Effects of 2 years of treatment. *J Allergy Clin Immunol* 1989;83:37–44.
- 36 Portnoy J, Bagstad K, Kanarek H, Pacheco F, Hall B, Barnes C: Premedication reduces the incidence of systemic reactions during inhalant rush immunotherapy with mixtures of allergenic extracts. *Ann Allergy* 1994;73:409–418.
- 37 Stewart GE, Lockey RF: Systemic reactions from allergen immunotherapy. *J Allergy Clin Immunol* 1992;90:567–578.
- 38 Sharkey P, Portnoy J: Rush immunotherapy: experience with a one-day schedule. *Ann Allergy Asthma Immunol* 1996;76:175–180.
- 39 Nagata M, Yamamoto H, Tabe K, Kimura I, Houya I, Kuramitsu K, et al: Effect of rush immunotherapy in house-dust-mite (HDM)-sensitive adult bronchial asthma: changes in in vivo and in vitro responses to HDM. *Intern Med* 1993;32:702–709.
- 40 Hejjaoui A, Ferrando R, Dhivert H, Michel FB, Bousquet J: Systemic reactions occurring during immunotherapy with standardized pollen extracts. *J Allergy Clin Immunol* 1992;89: 925–933.
- 41 Grammer LC, Zeiss CR, Suszko IM, Shaughnessy MA, Patterson R: A double-blind, placebo-controlled trial of polymerized whole ragweed for immunotherapy of ragweed allergy. *J Allergy Clin Immunol* 1982;69:494–499.
- 42 Grammer LC, Shaughnessy MA, Suszko IM, Shaughnessy JJ, Patterson R: A double-blind histamine placebo-controlled trial of polymerized whole grass for immunotherapy of grass allergy. *J Allergy Clin Immunol* 1983;72:448–453.
- 43 Hendrix SG, Patterson R, Zeiss CR, Pruzansky JJ, Suszko IM, McQueen RC, et al: A multi-institutional trial of polymerized whole ragweed for immunotherapy of ragweed allergy. *J Allergy Clin Immunol* 1980;66:486–494.
- 44 Metzger WJ, Patterson R, Zeiss R, Irons JS, Pruzansky JJ, Suszko IM, et al: Comparison of polymerized and unpolymerized antigen E for immunotherapy of ragweed allergy. *N Engl J Med* 1976;295:1160–1164.