

Clinical and Biological Evaluation of Semi-Rush and Ordinary Immunotherapy Schemes in Type I Allergic Respiratory Diseases

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Forty-three patients with type I allergic respiratory diseases received aqueous grass pollen immunotherapy in a prospective study designed to compare the clinical and biological effects of a semi-rush hyposensitization (20 patients) with ordinary hyposensitization (23 patients). Before the start of the therapy (end of previous grass pollen season, t1) and before (t2) and after the next grass pollen season (t3), total serum IgE (tIgE), grass pollen-specific IgE (GPsIgE), grass pollen-specific IgG (GPsIgG) and complement factors C3 and C4 were determined. GPsIgE and GPsIgG levels increased significantly more during immunotherapy in patients receiving the semi-rush scheme as compared with the ordinary scheme. There were no significant changes between the two schemes at any times for tIgE, C3 and C4. GPsIgG was already higher in allergic patients before the start of therapy as compared with non-allergic patients and controls. Clinical improvement was the same in patients treated with the semi-rush scheme as in the ordinary group. The number of side effects was not statistically different between the two groups. In neither group was there a correlation between the change in biological parameters and change in clinical symptoms. As the semi-rush scheme gives the same results immunologically as well as clinically, it could be used in a number of patients since it reduces the time course of the initial therapy by about 2 months.

Key words: immunotherapy; semi-rush; grass pollen.

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Immunotherapy with grass pollen (GP) and house dust mite (HDM) has been proven to benefit allergic patients. Aqueous extracts are still extensively used. Their disadvantage is that the dose has to be increased progressively over a period of time at a frequency of two times a week. It would be helpful if the duration of the initial therapy could be shortened, keeping the same therapeutic efficacy without increasing the side effects.

In this study we tried a scheme which was derived from the semi-rush hyposensitization in hymenoptera allergy (7).

MATERIAL AND METHODS

Patients

Forty-three patients (mean age 20.8 years; range 5-58) suffering from allergic asthma (5), asthma and rhinitis (20), or rhinitis alone (18) were followed during 1 year. Diagnostic work-up included history taking, skin testing with common inhalant allergens, determination of specific IgE via the RAST technique and, if any doubt persisted about the allergic etiology of the disease, allergen provocation as described

earlier (12). 20 patients proved to be allergic to grass pollen alone, and 23 to grass pollen and house dust mite.

The controls consisted of 27 individuals (mean age 34.9 years; range 23–65) without history of atopic eczema, asthma or rhinitis, chosen from laboratory and hospital staff, and 22 non-allergic patients (mean age 25.7 years; range 3–68) with asthma and/or rhinitis. None of the patients had previously been treated with immunotherapy.

Immunotherapy

In a first attempt, 10 grass pollen-sensitive patients received on day 1, at 30-min intervals, five injections (0.1, 0.3, 0.5, 0.8, 1.0 ml) of an aqueous mixed grass pollen extract containing 10 Noon Units (NU)/ml. On days 2 and 3, the same amounts were given at the same intervals with an extract containing 100 NU/ml, and 1000 NU/ml respectively.

During the injection period on day 3, all patients developed minor symptoms of rhinitis, asthma or urticaria, reacting promptly to treatment, before the final dose of that day was given.

This lead us to abandon the scheme and to use the semi-rush scheme presented below.

Patients for which immunotherapy was indicated were randomly subdivided into two groups: 23 patients received ordinary immunotherapy which comprised two weekly injections with increasing amounts (0.1, 0.2, 0.3, 0.4, 0.6, 0.7, 0.9, 1.0 ml) of increasing concentrations of allergen extract (10, 100, 1000, 10,000 NU/ml). This scheme took 4 months to reach the final concentration of 1 ml of 10,000 NU/ml.

Twenty patients received the semi-rush hyposensitization: at 30-min intervals they were injected with increasing amounts (0.1, 0.3, 0.5, 0.8, 1.0 ml) of hyposensitization extract dosed 10 NU/ml. One week later the same amounts were administered with a 10-fold higher concentration (100 NU/ml). They then switched to the ordinary scheme starting with 0.1 ml of 1000 NU/ml. This treatment took 2 months to reach the same final concentration as in the ordinary therapy. When the maximum dose was reach-

ed, therapy was continued in both groups by injecting 1.0 ml of 10,000 NU/ml, first weekly, then every 2 weeks, then every 3 weeks and, finally, monthly. From May to September the dose was reduced to 0.5 ml of the 10,000 NU/ml concentration. If a patient was allergic to house dust mite (*Dermatophagoides pteronyssinus*) as well, an aqueous extract of this allergen was combined with the grass pollen extract in a final concentration of 1000 NU/ml. All allergens were purchased at Haarlem Allergen Laboratories (HAL, Brussels).

Ordinary immunotherapy was started in October 1981, semi-rush in December 1981.

In vitro tests

Total serum IgE (tIgE) was determined by the Phadebas PRIST method (Pharmacia Belga, Brussels, Belgium) and were expressed in kU/l. Since it is known that there is a very high cross-reactivity between the different grass pollens, only allergen-specific IgE (sIgE) to *Phleum pratense* was determined by the Phadebas RAST technique (Pharmacia Belga, Brussels, Belgium) and expressed as the RAST class. Values were intra- or extrapolated in the RAST classes via a spline function. A grass pollen-specific IgG (GPsIgG) assay was performed by modification of a previously described method (3). The same commercially available lyophilised mixed grass pollen extract as used for immunotherapy (HAL, Haarlem, The Netherlands) was dissolved in phosphate buffered saline (PBS, Dulbecco's formula, pH 7.4) at a concentration of 10 µg/ml. 200 µl was added to polystyrene cups (Removawell, Dynatech, Alexandria, VA, USA) and incubated overnight at 4°C.

The wells were emptied by inversion and supplemented with 1% bovine serum albumin (BSA, Sigma, St. Louis, USA) in PBS. Saturation of unattached sites was allowed to take place for 3 h at 4°C. The plates were washed 3 times with PBS containing 0.1 % Tween 20 (PBS-T, Tween 20, Merck, Darmstadt, W. Germany). Sera were diluted 1/20 in PBS-T and 200 µl was incubated overnight at 4°C,

after which the wells were washed again 3 times with PBS-T. 100 μ l iodinated protein A (Amersham, England, specific activity 1.1 GBq/mg), diluted 1/300, was added. After incubation for 2 h at 4°C the plates were washed 3 times and residual radioactivity was counted by a gamma counter (Packard Instruments, Zurich, Switzerland).

A standard curve was constructed by two-fold dilutions of a serum sample containing large amounts of GPsIgG. Determinations were carried out in triplicate and results were expressed in theoretical units (TU) by intra- or extrapolating the residual radioactivity into the dilution curve, enumerating the dilution steps from 0 (1/2480) to 7 (1/20). The sensitivity of the method was 95.5 %, specificity 75.8 % and the predictive value 84.3 %. Reproducibility of five determinations in 10 individuals was 13.7 %.

Complement factors C3 and C4 were determined by a nephelometric method using Behring LN antisera and a Behring nephelometer (Behring Marburg, W. Germany). Results were expressed in mg/dl.

In vivo

Symptom score. Each patient was evaluated for symptoms such as running nose, sneezing, nasal obstruction, nasal itching, eye itching, tears, cough, wheezing, dyspnoea, occurring during the grass pollen season on an analogue 0–10 scale. The symptom score was determined as the sum of the individual scores. Symptom scores were evaluated first after the season without hyposensitization and again after the season with hyposensitization.

As the study did not intend to evaluate the

Table 1

Means $\pm$ SD of GPsIgG in different groups before start of the therapy (t1)

	Mean $\pm$ SD
Controls ($n = 27$)	0.98 ± 0.46
Allergic/ordinary immunotherapy ($n = 23$)	2.07 ± 1.21
Allergic/semi-rush immunotherapy ($n = 20$)	2.65 ± 1.75
Non-allergic ($n = 22$)	1.62 ± 0.68

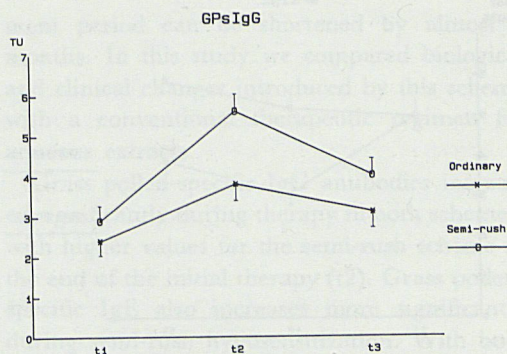


Fig. 1. Grass pollen-specific IgG (expressed as theoretical units) in patients before and during hyposensitization (mean $\pm$ SEM) at different time points: t1, before immunotherapy; t2, during immunotherapy, just before the grass pollen season; t3, after the grass pollen season.

value of hyposensitization as such, patients were allowed to use, if necessary, symptomatic treatment as well. This treatment was noted.

The following side effects were registered: large local reactions, urticaria, rhinitis, wheezing, hypotension, shock.

Statistical methods. Non-parametric tests (Wilcoxon ranksum test, test of Kruskal and Wallis) were used for comparison of differences of means. For comparison of related samples the Wilcoxon signed rank test, or Friedman's non-parametric two-way analysis of variance, were used when appropriate. Correlation coefficients were calculated by means of Spearman's rank correlation. Differences were considered statistically significant at the $P = 0.05$ level.

RESULTS

Table 1 depicts the mean values of GPsIgG values for the different treatment groups before treatment (t1).

Even before treatment, allergic patients had significantly higher mean values than non-allergic and controls (Kruskal and Wallis $P < 0.001$). The difference between both allergic groups was not significant (Wilcoxon $P = 0.51$).

GPsIgG did not change significantly in HDM-sensitive patients over a period of 1 year (results not shown).

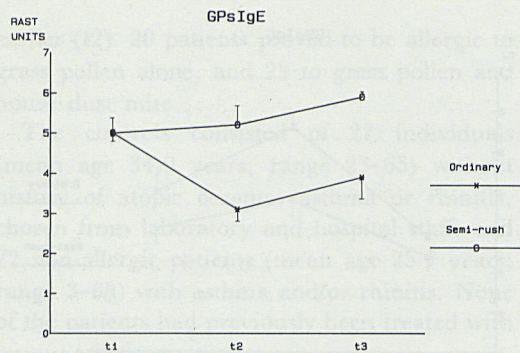


Fig. 2. Grass pollen-specific IgE (expressed as RAST units in patients before and during hyposensitization (mean $\pm$ SEM) at different time points: t1, before immunotherapy; t2, during immunotherapy, just before the grass pollen season; t3, after the grass pollen season.

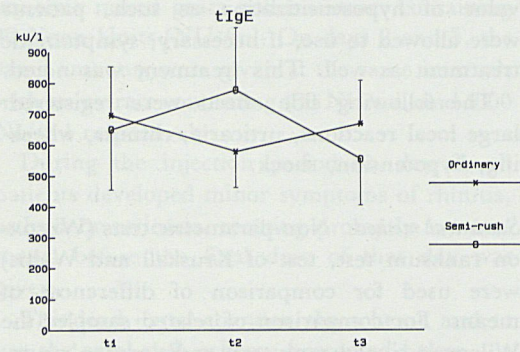


Fig. 3. Total serum IgE (expressed as kilo units) in patients before and during hyposensitization (geometric mean $\pm$ SEM) at different time points: t1, before immunotherapy; t2, during immunotherapy, just before the grass pollen season; t3, after the grass pollen season.

In 38 of the 43 GP-sensitive individuals, GP sIgG was determined. It increased during treatment (Fig. 1) both with the ordinary scheme (22 patients, Friedman $P < 0.0001$) and the semi-rush scheme (16 patients, Friedman $P < 0.0001$).

The difference between the semi-rush and ordinary scheme was not significant before therapy (vide supra). The increase from t1 to t2 is significantly ($P = 0.023$) higher in the semi-rush group (2.7 ± 1.6 TU) as compared with the ordinary treatment group (1.4 ± 1.9 TU), resulting in significantly higher values (Wilcoxon $P = 0.01$) on t2 in the semi-rush group.

At t3 the values in the semi-rush group were still higher but no longer significant (Wilcoxon $P = 0.12$).

Fig. 2 traces the evolution of GP sIgE at the same time points. The difference between ordinary and semi-rush treatment is not significant at t1 but is very significant at t2 (Wilcoxon $P = 0.002$) and t3 (Wilcoxon $P = 0.033$) with higher values in the semi-rush scheme. The changes in the two groups with time are not significant.

Fig. 3 depicts the evolution of tIgE. There is no difference between ordinary- and semi-rush-treated patients at three different times and the changes within one treatment group with time are not significant.

Complement factors C3 and C4 do not differ significantly between allergic patients, non-allergic patients and controls. The values remain constant during hyposensitization (results not shown).

We next evaluated the possible correlation between GP sIgG and GP sIgE in both treated groups. Although the values of correlation coefficient favoured a statistically significant correlation, the coefficients were not high (0.38 at t1, 0.49 at t2, and 0.60 at t3). On the other hand, the correlation between ln tIgE and GP sIgG was not significant at any time.

In Table 2 the symptom scores of individuals belonging to one of the two treatment groups are recorded before and after treatment. In order to eliminate the possible role of symptomatic treatment, only the patients using no medication were compared.

There is a significant reduction of symptom

Table 2
Changes in symptom score

	Season I Mean $\pm$ SD	Season II Mean $\pm$ SD	
Rush ($n = 8$)	39.8 $\pm$ 19.1	16.3 $\pm$ 12.6	P 0.01
Ordinary ($n = 12$)	46.7 $\pm$ 18.1	20.0 $\pm$ 13.6	P 0.01
	N.S.	N.S.	

Season I: Symptoms before treatment.
Season II: Symptoms after treatment.
N.S.: Not significant.

scores after the treatment, but there is no difference either before or after treatment between ordinary and semi-rush hyposensitization.

The semi-rush scheme was associated with one mild systemic reaction, occurring during the ordinary phase of the hyposensitization treatment, whereas no systemic reaction was observed during ordinary therapy.

No significant correlations were found between the different *in vitro* and *in vivo* parameters determined after the season (symptom score and number of side effects).

DISCUSSION

Hyposensitization with aqueous extracts of antigens such as plant pollen and house dust mite is accepted as a therapeutic measure which can ameliorate the course of diseases such as asthma or rhinitis (5, 6, 9, 11, 13, 14). The disadvantage of this therapy is its long duration with many injections, especially during the initial period when the dose is steadily increased. Several years ago, rush hyposensitization was introduced for hymenoptera-sensitive individuals. Progressively increasing amounts of the venom extracts are injected up to the maintenance dose of 100 µg in a period of 5 days. This therapeutic regimen is supported well by patients and involves few side effects of importance (7). This led us to investigate the possibility of introducing a "faster" hyposensitization scheme in patients with respiratory allergy. It was recognised that sensitivity to venoms is different from sensitivity to respiratory allergens since the route of entrance is parenteral in the first case and respiratory in the second.

We first attempted to give GP-sensitive patients five injections daily for 4 consecutive days, with a 10-fold increase of the dose each day. All patients had symptoms of rhinitis, asthma or urticaria, and reacted promptly to symptomatic treatment, during the third day. For this reason the rush scheme was abandoned and we switched to the semi-rush scheme described here, where five injections were given two times with a 1-week interval. By applying this scheme, the duration of the initial treat-

ment period can be shortened by almost 2 months. In this study we compared biological and clinical changes introduced by this scheme with a conventional therapeutic regimen for aqueous extracts.

Grass pollen-specific IgG antibodies increased significantly during therapy in both schemes, with higher values for the semi-rush scheme at the end of the initial therapy (t₂). Grass pollen-specific IgE also increases more significantly during semi-rush hyposensitization. With both regimens total serum IgE, complement C3 and C4 remained unchanged during the observation period of 1 year.

Compared with the previous season symptom scores with both schemes decreased significantly. There was no significant difference in the number of adverse reactions. One systemic reaction occurred in the semi-rush scheme during the "ordinary" period. However, as already reported by others there was no good correlation between clinical improvement and changes in biological parameters (2, 4, 5, 6, 13), which perhaps explains why the higher values for both GPsIgE and GPsIgG in the semi-rush group do not result in different clinical results.

An observation, already mentioned by others (1, 8, 10, 14), is the correlation between GPsIgG and GPsIgE. This could mean that in atopic patients there is not only a facilitation for the formation of GP-related IgE but also of GP-related IgG. In conclusion, since both ordinary and semi-rush hyposensitization induce similar biological and clinical effects and are well tolerated, semi-rush hyposensitization could be applied in patients where a shorter initial build-up of the dose is preferred.

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