

Local nasal immunotherapy: Efficacy of low-dose aqueous ragweed extract

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In previous studies preseasonal local nasal immunotherapy (LNIT) with moderate doses of aqueous ragweed extract (mean total dose 59 µg of AgE and 139 µg of AgE) was an effective treatment for ragweed hay fever; however, local adverse reactions during therapy were common. This study evaluated the clinical and immunologic responses to LNIT by use of lower doses of aqueous ragweed extract in order to minimize these adverse reactions. Patients were administered preseasonal LNIT for 7 wk and received a mean total dose of 4.7 µg of AgE. During the ragweed season, symptom/medication scores (SMS) of the treated patients were equivalent to SMS of untreated patients. Serum ragweed-specific IgE and nasal secretory ragweed-specific IgA rose slightly in the treated patients but not to the extent observed in previous studies. After the ragweed season treated and untreated patients had a substantial increase in serum ragweed IgE antibody titers. No correlation could be found between antibody responses and SMS. This study indicates that LNIT with lower doses of aqueous ragweed extract is clinically ineffective. (J ALLERGY CLIN IMMUNOL 75:496-500, 1985.)

LNIT with aqueous ragweed extract is clinically effective in the treatment of ragweed hay fever.¹⁻³ Serum ragweed-specific IgE increases after therapy, suggesting systemic antigen absorption.⁴ The major limitation of LNIT with aqueous ragweed antigen is the local adverse reactions encountered during the treatment.

MATERIAL AND METHODS

Antigens

The aqueous ragweed extract was from the same lot of defatted pollen used in the 1979 and 1980 LNIT studies.^{1,2} The antigen E content of the extract was 159 µg/ml. Serial tenfold dilutions were made for treatment solutions.

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Abbreviations used

LNIT: Local intranasal immunotherapy
SMS: Symptom/medication scores

This aqueous extract also was used as the coupling antigen in the radioimmunoassays.

Patient selection

Thirty ragweed-allergic subjects who had participated in the 1979 and 1980 studies returned in 1981. In these previous studies, 13 patients had received aqueous ragweed extract, nine had received polymerized ragweed extract, and eight had received placebo therapy. Eighteen additional ragweed-allergic subjects were recruited to serve as untreated control patients. Each subject had a history of ragweed-allergic rhinitis confirmed by a positive prick skin test to 5% aqueous ragweed extract. None of the treated and control patients had received parenteral immunotherapy for the past 5 yr. Subjects were excluded if they had mold sensitivity, perennial rhinitis, asthma, or regularly used nasal corticosteroid or decongestant sprays.

Treatment

Preseasonal LNIT was begun in June 1981 and administered for 7 wk. Extracts were dispensed in plastic 5 cc nasal spray bottles that delivered 0.04 cc per spray. The starting bottle contained a concentration of 0.02 µg of AgE

per milliliter. Patients were instructed to keep the bottles refrigerated and to use the nasal spray every other day. One squirt was applied to each nostril on day 1, two squirts on day 3, and three squirts on days 5 and 7. If this were tolerated, bottles with serially increasing tenfold concentrations of extract were administered every week. The maintenance dosage was that dose that elicited only mild rhinitis symptoms. This dose was administered for the remainder of the treatment period. The amount of extract applied (expressed as antigen E) was calculated from the concentration of the extract, the number of sprays prescribed, and the average volume of a spray. Compliance with the treatment was monitored by weighing the treatment bottles before and after use.

Clinical evaluation

Each subject was observed at biweekly intervals throughout the study. Physical examinations were performed, and history of pertinent allergic symptomatology was reviewed. SMS records were completed twice daily from the first week in August until the first week in October. Records included the severity (0 to 4) and duration (0 to 3) of sneezing, rhinorrhea, nasal congestion, eye irritation, and medications used. Daily SMS were tabulated from the diaries.⁵ In addition, a mean SMS over the peak pollen period was determined for each patient.¹ Daily ragweed pollen counts were done by use of an intermittent rotorod sampler (Ted Brown Assoc., Los Altos Hills, Calif.). The peak pollen period was defined as that interval during which daily counts were consistently above 30 grains per cubic meter.

Immunologic evaluation

Sera and nasal secretions from treated patients were collected before treatment (June 1981), after treatment (August 1981), and after the ragweed season (October 1981). A modified Okuda⁶ method of nasal provocation was used to obtain nasal secretions. Briefly, 3 mm paper discs were placed on both inferior nasal turbinates. Each disc contained 10 μ l of a tenfold dilution of aqueous ragweed extract or the diluent. Initially, control (diluent) discs were placed on the turbinates followed by discs containing a 10^{-5} dilution of the ragweed extract. At 5-minute intervals the discs were replaced with a stronger concentration of the ragweed extract until each subject was able to produce 1 cc of nasal secretions. The nasal secretions were collected onto Para-Film and then transferred to 5 cc plastic tubes for storage at -20° C. Only pre-season blood samples were obtained from the control patients.

Ragweed-specific IgE and IgA antibodies were measured by use of RAST.⁴ IgE titers were arbitrarily tabulated as percent of a known positive standard of reference serum. Ragweed-specific nasal secretory IgA titers were expressed as percent of total radioactivity since a positive standard was not available.

Ragweed-specific IgG antibodies were measured by use of a modification of the *Staphylococcus aureus* protein A assay.⁷ IgG antibody titers were expressed as percent of a reference serum.

TABLE I. Demographic data of ragweed-treated and control patients

	Treated	Untreated
No. of patients	30	18
Mean age	40	37
range	21 to 61	16 to 67
Pretreatment serum	31*	11*
ragweed-specific IgE	(<3 to 336)	(<3 to 120)
(% STD)		
Patients with initial	25	9
IgE titers >10		
Pretreatment serum	65	47
ragweed-specific IgE	(10 to 336)	(10 to 120)
(% STD)		

* $p < 0.01$.

Statistical analysis

Daily and mean SMS between treated and untreated patients were analyzed by use of the Wilcoxon rank-sum test.⁸ Changes in antibody titers were analyzed by use of the paired t test. Correlations between antibody response and SMS were done by use of the Spearman rank-sum coefficients. Pollen counts for the 3 yr were compared by use of the t test. Values <0.05 were considered significant.

RESULTS

Table I summarizes the demographic data of the 30 treated and 18 control subjects. The patients' age and level of sensitivity, which was measured by prick skin testing, were the same in the treated and control groups. There was a statistically significant difference in the mean pretreatment serum ragweed-specific IgE titers: 31 units in the treated group and 11 units in the control group ($p < 0.01$). For this reason a subgroup composed of patients with initial serum ragweed-specific IgE titers ≥ 10 units was analyzed. By use of this parameter there was no significant difference between groups in pretreatment RAST titers.

Patients received an average total dose of 4.7 μ g of AgE (range 0.8 to 16.3 μ g of AgE). This dose was considerably lower than that administered to these patients in the previous 2 yr (1979, 59 μ g of AgE $\pm$ 50; 1980, 139 μ g of AgE $\pm$ 63). Local adverse reactions were minor and only lasted for a few minutes. There were no systemic reactions during the treatment period.

Mean daily pollen counts were 75 grains per cubic meter in 1979, 121 grains per cubic meter in 1980, and 54 grains per cubic meter in 1981. A statistical analysis illustrated that the 1980 counts were significantly higher than those in either 1979 or 1981. There was no difference in pollen counts in 1979 and 1981.

Daily SMS are illustrated in Fig. 1. There was no

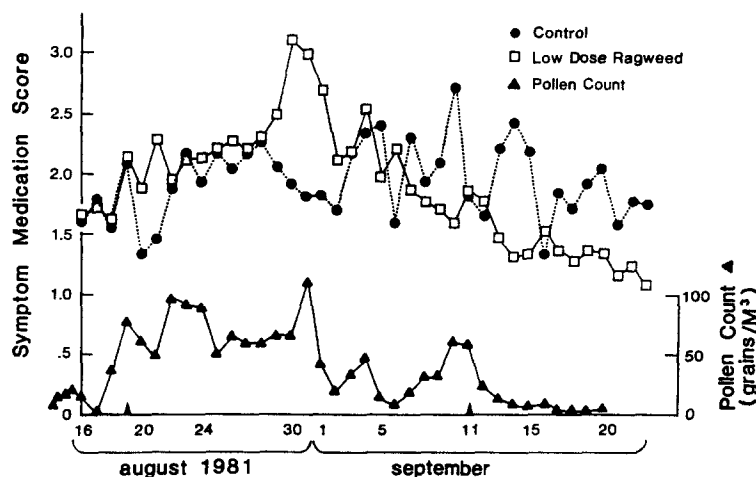


FIG. 1. Daily SMS for patients administered low-dose aqueous ragweed extract (□) and untreated control patients (●). Pollen counts are illustrated at the bottom of the figure (▲). The arrows at the bottom identify the peak pollen season (August 19 to September 11). There was no difference in SMS between the groups.

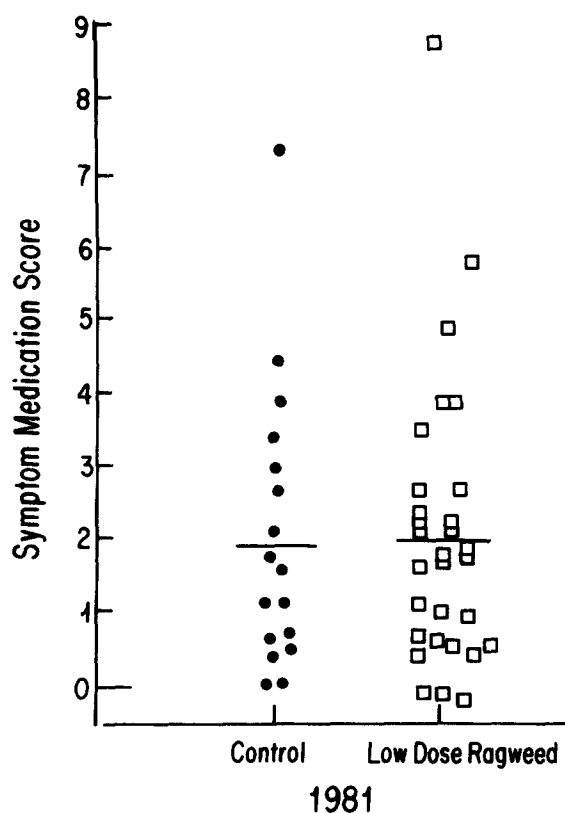


FIG. 2. Mean SMS of each group during the peak pollen period in 1981, low-dose aqueous ragweed (□) and untreated controls (●). There was no statistical difference ($p > 0.10$) between groups.

difference between treated and untreated patients on any given day. Mean SMS are illustrated in Fig. 2. Geometric means for these scores were 2.20 for the low-dose aqueous treatment group and 2.04 for the

control group, which also was not significant. There was no difference in daily or mean SMS of the treated and untreated patients who had initial serum ragweed-specific IgE titers >10 .

In comparing SMS for the 3 yr, patients receiving aqueous treatment in 1981 had slightly higher overall SMS than in 1979 and 1980. The range of SMS was similar for all 3 yr, 1979 to 1981.

There was a slight, significant, rise in serum ragweed-specific IgE after treatment. A further seasonal IgE antibody titer rise occurred in 1981 that had not been found previously in 1979 or 1980. Secretory IgA ragweed antibody titers rose after therapy and then declined to baseline levels after the season. Serum and nasal secretory IgG antibody titers were extremely low and did not change throughout the study period.

The effect of LNIT on serum ragweed-specific IgE and nasal secretory ragweed-specific IgA is illustrated in Fig. 3. In 1979 and 1980 LNIT with moderate doses of aqueous ragweed extract stimulated serum IgE antibody titers ($>100\%$ of initial values) and blocked the expected seasonal rise in these titers. A seasonal rise in IgE antibodies occurred in the control group in both years. In 1981 low-dose treatment stimulated a rise in serum ragweed-specific IgE but not the degree observed in 1979 and 1980. In addition, the low-dose treated group also had a seasonal rise in IgE antibodies in contrast to 1979 and 1980. After therapy, nasal secretory IgA antibody titers increased ($>100\%$) in 1979 and 1980 and then decreased after the pollen season. In 1981 these IgA antibody titers also rose after therapy but not to the same degree as in earlier studies ($<100\%$ of initial values).

There was no correlation between SMS and any

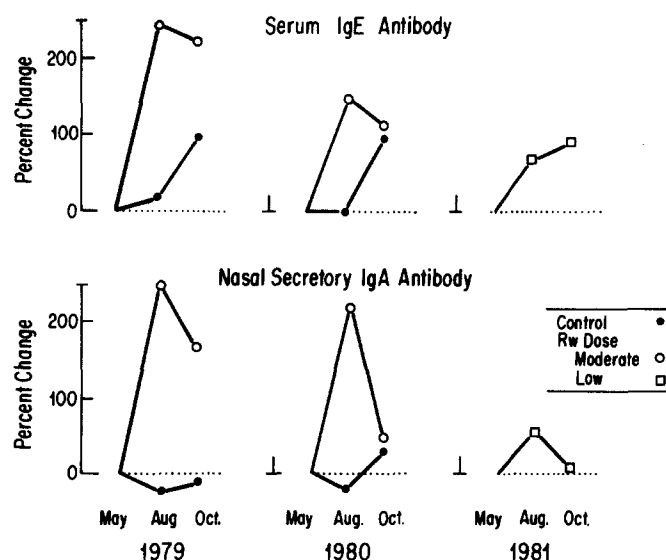


FIG. 3. Percent change in serum ragweed-specific IgE and nasal secretory ragweed-specific IgA during the 3 yr of LNIT. There was a marked rise in IgE and IgA antibodies after treatment with moderate doses of ragweed extract (○) as compared to controls (●) in 1979 and 1980. In 1981 there was less rise in IgE and IgA antibodies after low-dose ragweed treatment (□).

TABLE II. Local intranasal immunotherapy with use of aqueous ragweed extracts

Study	Year	No. of patients	Design	Clinical results	Mean dose (μ g AgE)
Welsh ³	1980	18	double-blind	+	151
Welsh et al. ⁹	1981	11	double-blind	—	21
Nickelsen et al. ¹	1979	21	double-blind	+	59
Nickelsen et al. ²	1980	17	open-controlled	+	139
Present study	1981	30	open-controlled	—	4.7

antibody response throughout the study period or the pollen season.

DISCUSSION

The purpose of this study was to determine the clinical effectiveness of LNIT with the use of lower concentrations of aqueous ragweed extract. The appearance of mild rhinitis symptoms during treatment was used as the end point for the top dose of ragweed extract in order to minimize adverse reactions. As a result the total dose of ragweed antigen E (4.8 AgE) was far less than administered in our previous studies (59 μ g and 139 μ g).^{1, 2}

Adverse reaction scores reported in this study were much lower, which was expected, than reactions reported in our 1979 and 1980 studies. The low-dose LNIT was well tolerated by the patients, whereas in previous studies some patients were reluctant to have LNIT with the higher doses of aqueous ragweed extract.

This low-dose LNIT, however, was clinically in-

effective in reducing symptoms of treated patients when patients were compared to control subjects. Specifically, there was no significant difference in SMS between the two groups. The two studies by Welsh et al.^{3, 9} support our findings. In their study with the use of moderate amounts of aqueous ragweed, LNIT was clinically effective in comparison to placebo therapy,³ but there was no clinical benefit in a second study with the use of lower doses of aqueous ragweed extract. Studies of LNIT with aqueous ragweed extract are summarized in Table II.

The low-dose LNIT did increase serum ragweed-specific IgE and nasal secretory ragweed-specific IgA. This treatment did not block the seasonal rise in serum IgE titers commonly observed in placebo-treated or untreated ragweed-sensitive patients. This immunologic response to low-dose LNIT was much less than that observed after higher doses of aqueous ragweed.

There appears to be a specific dose-response relationship between the amount of aqueous antigen administered in LNIT and the clinical response. Mod-

erate concentrations of aqueous ragweed (60 to 150 μ g of AgE) are clinically effective and yet cause frequent adverse reactions. LNIT with low doses of aqueous ragweed, however, are clinically ineffective. Use of modified extracts may resolve this clinical dilemma. The modified solutions are highly immunogenic and yet cause local minimal reactions.^{1, 10} Therefore, further studies of LNIT are needed to evaluate the clinical efficacy of modified ragweed extracts.

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