

ORIGINAL ARTICLE

Does Specific Immunotherapy Injection Cause an Increase in Bronchial Reactivity?

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Background. Several well-controlled studies have proven the clinical benefit of specific immunotherapy (SIT) for seasonal allergic rhinitis (AR). However, whether subcutaneous SIT injection could cause a transient increase in bronchial reactivity (BR) remains unknown. **Objective.** To investigate whether subcutaneous SIT injection, either during or outside the pollen season, could cause an increase in BR in children with pollen allergy. **Methods.** Twenty-two children (mean age 13.6 ± 0.7 years) with AR who were receiving maintenance SIT for 15 months were included in the study. Pre-injection BR of the patients was evaluated with methacholine provocation test immediately before maintenance dose of SIT during the peak pollen season and outside the season. The post-injection test was administered 24 hours after SIT injection. **Results.** There was no difference in FEV₁ measures recorded during [98(93–109)%] and outside [102(96–111)%] the pollen season. There was no significant difference between pre- [64(7–64)mg/mL] and post-allergen injection [32(7.5–64) mg/mL] BR outside the pollen season ($p = 0.9$). A trend towards improvement following allergen injection [64(5.4–64)] as compared to pre-allergen injection [14.6(3.5–64)] was shown during the pollen season ($p = 0.053$). Although PC20 measures in the pollen season were lower than outside the season, the difference was not significant. The percentage of the patients with bronchial hyperreactivity was 62% during and 43% outside the season. **Conclusion.** SIT injections both during and outside the pollen season cause no increase in BR in children with AR. This calls into question the necessity of empirical dose reduction during the pollen season.

Keywords bronchial reactivity, allergic rhinitis, specific immunotherapy, pollen

INTRODUCTION

Inhalation of allergens by sensitized individuals results in a bronchoconstriction that develops within 10 to 20 minutes. This early asthmatic reaction generally resolves within 1 to 2 hours. In individuals who develop a late asthmatic reaction, the decrease in lung function occurs 3 to 4 hours after inhalation and can persist for 24 hours or more (1). An association between the late response and increased bronchial responsiveness (BR) has been demonstrated (2). There are data indicating that inhalation of pollen allergens may cause an increase in BR in patients with seasonal allergic rhinitis (AR), irrespective of the presence of asthma (3–5). In addition, studies of patients with AR, asthma, and other atopic diseases have shown an increase in circulating inflammatory cells and progenitors after allergen inhalation, followed by recruitment to sites of allergic inflammation (6–8). Yet it is not known if another route of exposure to the allergens, such as by subcutaneous injection, could cause a transient increase in BR.

Several well-controlled studies have proven the clinical benefit of specific immunotherapy (SIT) for treatment of seasonal AR (9–11), and there is also evidence that SIT may decrease the risk for subsequent development of asthma and bronchial hyperresponsiveness (BHR) in

children with AR (12). Application of a specific allergen to already sensitized patients always bears some risks, which are important limitations of SIT practice (13). In clinical practice, a number of physicians decrease the dose of SIT empirically during the pollen season because of the fear of side effects. In this study, we investigated whether subcutaneous SIT injection, either during or outside the pollen season, could cause a transient increase in BR 24 hours after injection in children with pollen allergy.

MATERIAL AND METHODS

Patients

Twenty-two children (mean age 13.6 ± 0.7 years) with a history of severe allergic rhinitis due to grass pollen who were receiving 6,000 therapeutic units [TU] maintenance injections of the grass mix pollen-allergoid SIT (Allergovit; Allergopharma Joachim Ganzer KG, Hamburg, Germany) monthly for 15 months were included in the study. The grass mix pollen allergoid is produced from standardized extracts of 16.6% *Dactylis glomerata*, 16.6% *Festuca elatior*, 16.6% *Holcus lanatus*, 16.6% *Phleum pratense*, 16.6% *Poa pretenses*, and 16.6% *Lolium perenne*. Allergovit is an aluminum hydroxide-adsorbed preparation of the allergoid. The indications for SIT were in accordance with the proposals of the European Academy of Allergy and Clinical Immunology (EAACI) (14).

Children with typical symptoms of AR during the grass pollen season who tested positive (wheal reactions 3 mm greater than the negative control) to a skin prick test with a biologically standardized grass mix pollen extract (50000

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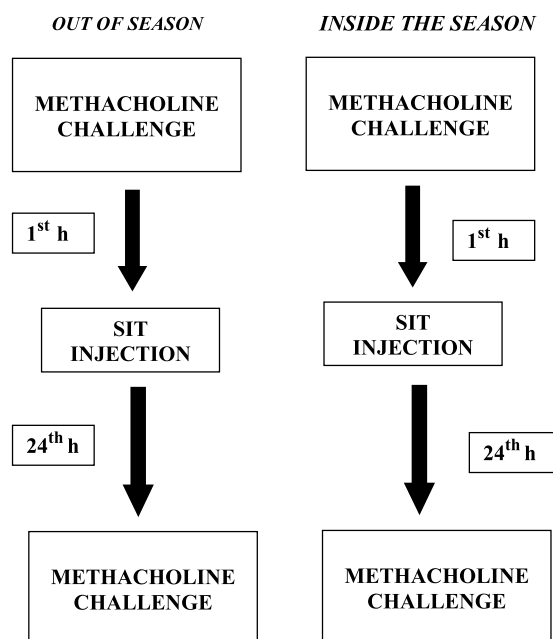


FIGURE 1.—Study design.

SBU/mL) (Allergopharma, Reinbek, Germany) and had positive specific IgE to grass mix pollen test ($\geq$ class 2) (CAP-System; Pharmacia, Uppsala, Sweden) received SIT and those in whom an increase in the phleum-specific IgG levels was demonstrated were included (data not shown).

Exclusion criteria were rhinitis of other causes including perennial allergic rhinitis; need for ACE-inhibitors or β blockers; and chronic diseases other than allergic diseases, such as active tuberculosis, autoimmune diseases, immunodeficiency, chronic inflammatory diseases, and malignancy.

Seven patients in our group had wheezing the previous year during the grass pollen season. Since they did not suffer from lower airway symptoms except during the peak-pollen season when exposed to a high concentration of grass pollens, they were not given prophylaxis with inhaled corticosteroids.

Study Design and Ethical Considerations

The study was performed according to sound clinical practice for trials on medical products in the European Community (EC-GCP-guidelines) (15). The physicians and nurses who performed and evaluated the methacholine provocation (MP) tests had no knowledge of the study. The study protocol and informed consent were in agreement with the declaration of Helsinki and approved by the regional Ethics Committee. All patients' parents gave written informed consent before the study.

Methacholine Provocation Test

BR of the patients was evaluated with MP test 1 month after the last maintenance dose of SIT. The test was administered 1 hour before and again 24 hours after (16) SIT injection. The testing procedure was performed both during the season (mid-June), when the grass pollen counts had peaked according to 2002 and 2003 data and outside

the season (December). The study design is shown in Figure 1.

MP tests were administered as follows: Methacholine was inhaled in doubling concentrations starting with 0.06 mg/mL up to a maximum concentration of 32 mg/mL. Forced expiratory volume (FEV_1) was measured 30 s and 90 s after each inhalation. The provocation continued with 5 min intervals between inhalations until a decrease in FEV_1 of 20% or greater was obtained or the highest concentration of methacholine was inhaled (17). Treatment with short-acting β_2 agonists and antihistamines was not permitted 12 hours and 72 hours before the MP tests, respectively. None of the patients was treated with theophyllines, long-acting β_2 agonists, or corticosteroids.

Pollen Counts

A Burkhard spore trap was used to collect pollen grains. All study patients were living within 30 km of the spore trap. Seasonal evaluations were performed when the grass pollen counts were expected to reach peak levels according to 2002 and 2003 data.

Statistics

Wilcoxon Matched-Pairs Signed Rank and Friedman Tests were used for comparison of methacholine PC₂₀ measures of the groups during the different study visits. McNemar test was used to compare the presence of BHR before and after SIT injection during and outside the pollen season, and a 5% significance level was used. Methacholine PC₂₀ measures greater than 32 mg/mL were accepted as 64 mg/mL. Subgroup analysis was performed in patients with BHR (PC₂₀ $\leq$ 32 mg/mL).

RESULTS

All 22 subjects completed the study. The demographic characteristics of the study group are presented in Table 1. All patients received the 6000 TU SIT injection during and outside the season.

There was no difference in FEV_1 measures (percent predicted) recorded during [98 (93–109)] [median (interquartile range)] and outside [102 (96–111)] the pollen season.

TABLE 1.—Demographics of the study group.

No. of patients	22
Age, yrs*	13.1 (11.4–16.5)
Sex	
Male	15 (68%)
Female	7 (32%)
Duration of rhinitis*	4.2 (3–5.5) yrs
Presence of asthma	7 (32%)
Duration of SIT*	15 mos
Phleum specific IgE* (kU/L)	69.6 (26.5–100)
FEV_1 (%)* (outside the season)	102 (96–111)
FEV_1 (%)* (inside the season)	98 (93–109)
Pre-injection methacholine PC ₂₀ (mg/mL)* (outside the season)	64 (7–64)
Pre-injection methacholine PC ₂₀ (mg/mL)* (inside the season)	14.6 (3.5–64)

*Median (interquartile range).

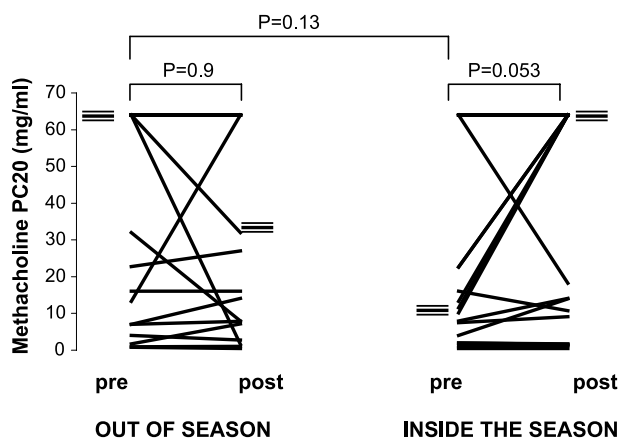


FIGURE 2.—Results of the methacholine provocation tests for pre- and post-allergen injection during and outside the pollen season. *P* values are for the comparison of the two parameters by Wilcoxon test. —: Median value for methacholine PC20 measures (mg/mL).

Methacholine Provocation Test

Comparison of the four MP test results (pre- and post-allergen injection during and outside the pollen season) revealed no significant differences ($p = 0.42$, Friedman test). There was also no significant difference between pre-[64 (7–64) mg/mL] and post-allergen injection [32 (7.5–64) mg/mL] BR outside the pollen season ($p = 0.9$, Wilcoxon). However, when pre- [14.6 (3.5–64) mg/mL] and post-allergen injection [64 (5.4–64) mg/mL] BR during the pollen season was compared, there was interestingly a trend towards improvement after allergen injection ($p = 0.053$, Wilcoxon). Although pre-injection PC20 measures during the season [14.6 (3.5–64) mg/mL] were lower than outside the season [64 (7–64) mg/mL], the difference was statistically insignificant ($p = 0.13$, Wilcoxon) (Figure 2).

When we performed subgroup analysis in patients with BHR (PC20 ≤ 32 mg/mL), there was no significant difference between pre- and post-injection PC20 measures during ($p = 0.31$, Wilcoxon) and outside the grass pollen season ($p = 0.57$, Wilcoxon).

The percentage of patients who had pre-injection BHR (PC20 < 32 mg/mL) was 62% during the season and 43% outside the pollen season ($p = 0.22$, McNemar). BHR percentages for pre- and post-injection were 62% and 48%, respectively, during the pollen season ($p = 0.22$, McNemar), and 43% and 48%, respectively, outside the season ($p = 1.00$, McNemar).

Patients were observed in the clinic for 1 hour following SIT injection, but no systemic side effects were recorded. None of the patients had wheezing or dyspnea before or after injection during or outside the pollen season.

DISCUSSION

Several well-controlled studies have demonstrated the clinical benefit of SIT therapy for seasonal AR to pollen (9–11). Introduction of a specific allergen to sensitized patients always bears the risk of adverse effects, which is a critical limitation of SIT practice (13). A number of physicians decrease SIT doses empirically during the pollen

season because of the fear of side effects. Furthermore, there is mounting evidence indicating a systemic connection between the nose and lungs as one of the mechanisms involved in nasobronchial cross-talk (18, 19). It has been shown that the concentration of IL-5 and the number of eosinophils in the peripheral blood are increased within 24 hours after segmental bronchial provocation in AR patients (18). In another study, Smith et al. showed that bronchoconstriction was induced within 24 hours by administering peptides to cat-allergic asthmatic subjects by intradermal injection (20). If this is true, SIT injection (an allergen), applied subcutaneously to AR patients, may cause an increase in the circulating inflammatory mediators followed by recruitment of cells to the lung and nose, giving rise to an increase in BR and nasal symptoms. However, to the best of our knowledge, whether allergen injection, like SIT, can cause a transient increase in BR has not yet been studied. Our results indicate that neither extraseasonal nor seasonal allergen injection causes an increase in BR in children with AR.

Clinical investigations indicate a relationship between the presence of activated airway inflammatory cells (particularly T cells and eosinophils) and the development and severity of BR (21–23). Depletion of CD4⁺ T cells before allergen challenge of sensitized mice has been shown to abrogate BR, and T-cell transfer studies have further implicated Th2 cells in the development of allergic airway eosinophilia and BR (24, 25). Interestingly, Huang et al. showed that intravenously administered allergen-specific Th1 cells down-regulate efferent Th2 cytokine-dependent BHR and broncho-alveolar lavage eosinophilia in 18 to 24 hours in an asthma model via mechanisms that depend on IFN- γ (26). Accordingly, Th1 cells induced by SIT injection might have counteracted the allergen-specific Th2 cells developed due to inhalation of grass pollen during the season. It was demonstrated that the grass pollen allergoids retain a highly specific Th1 cell-stimulating capacity without Th2 cell activation (27). The increase in BR by maintenance injection of the allergoid SIT might have been prevented because of the decrease in activation of the existing Th2 cells by allergoids and by stimulation of the Th1 cells.

Our results indicate that extraseasonal and seasonal allergen injections cause no increase in BR in children with AR. This calls into question, therefore, the necessity of the empirical approach of decreasing the dose of SIT during the pollen season. However, in light of our study's limitations, further investigations are necessary to make a definitive conclusion. First, 57% of our study group consisted of AR children without BHR, which indicates a possibility of type 2 error. Second, and perhaps more importantly, our patients were on a 15-month SIT course, indicating a possibility of SIT-induced tolerance against increase of BHR. Smith and Larche demonstrated that subjects rechallenged with intradermal peptide had a markedly reduced or absent bronchoconstriction response, although the first injection of the same peptide antigen had resulted in bronchoconstriction (20). Another interesting point is that only a small number of our patients were asthmatic and none of them had lower respiratory symptoms while SIT injections were

administered. Further studies of asthmatic subjects are needed to determine whether SIT injection can cause an increase in BHR.

A link between hay fever and asthma is evident, and approximately 20% of all hay fever patients develop asthma later in life (28). SIT may decrease the risk of subsequent development of asthma and BHR in children with AR (12). Eleven to 73% of hay fever patients show BHR outside the pollen season (29, 30), and up to 50% of such patients show BHR during the season (30, 31). We have shown that the percentage of patients with BHR was 62% and 43%, respectively, during and outside the pollen season ($p = 1.00$, McNemar).

In conclusion, our results indicate that SIT injections during or outside the pollen season cause no increase in BR in children with AR to grass pollen. In contrast, BR showed a tendency towards a decrease after injection compared to pre-allergen injection during the grass pollen season.

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