The Effect of Allergen Immunotherapy on the Development of New Sensitization in Children

Şükrü Çekiç 1, Yakup Canitez 1, Fatih Çiçek 1, Gökhan Ocakoğlu 1, Nihat Sapan 1

1Division of Pediatric Allergy, Uludağ University School of Medicine, Bursa, Turkey
2Department of Biostatistics, Uludağ University School of Medicine, Bursa, Turkey

ABSTRACT

Aim: The protective effect of allergen immunotherapy against a new allergic sensitization is controversial. This study aimed to investigate the effect of allergen immunotherapy on new allergic sensitization in children.

Materials and Methods: The study included 50 patients who received immunotherapy for at least 3 years, and whose skin prick tests were repeated at intervals of at least 3 years (31 patients for house dust mite immunotherapy, 19 patients for pollen immunotherapy), and 69 controls with similar characteristics.

Results: The number of patients who developed a new sensitization was similar both in the groups of patients who received house dust mite and pollen immunotherapy, and the control group. There was no significant difference between the first and last skin prick tests of the patients who received house dust mite and pollen immunotherapy; however, in the control groups, a significant increase in sensitivity to tree pollens ($n = 2$, 5.4%; $n = 8$, 21.6%) and weed pollens ($n = 7$, 26.9%; $n = 14$, 53.8%) was detected ($P = .031$ and $P = .039$). While allergen sensitivities in the first tests of the pollen immunotherapy group and the control group were similar, weed pollen sensitivity was significantly higher in the last tests of the control group ($n = 14$, 53.8%; $n = 4$, 21.1%, $P = .027$). It was determined that the presence of weed pollen sensitization (OR: 8.1, 95% CI: 1.5–42.4) and having asthma (OR: 3.5, 95% CI: 1.3–10.8) increases the risk of new sensitization in all groups.

Conclusion: Allergen immunotherapy has been found to protect against new sensitization to tree and weed pollens. However, this effect was insignificant in the multivariate analysis. Weed pollen sensitization and the presence of asthma are related to the development of new sensitization.

Keywords: Asthma, immunotherapy, new sensitization, weed pollen

INTRODUCTION

Allergen immunotherapy is a treatment method that aims to develop immune tolerance against the sensitized allergen by administering it at increasing doses in regular intervals.1 It was first used in the treatment of allergic rhinitis by Noon and Freeman in 1911.2,3 This treatment is the only method that can change the natural course of allergic diseases. The main areas of application are allergic rhinitis/rhinoconjunctivitis, allergic asthma, and bee (venom) allergy treatments.4

Data show that in addition to the therapeutic properties, allergen immunotherapy protects against the development of asthma in patients with allergic rhinitis and prevents the development of new allergen sensitivity in patients.4–8 However, contrary to this information, published...
studies also report that it does not protect against new sensitiv-
ity development, or causes sensitivity to increase. As a result,
more evidence is needed on whether allergen immunotherapy
protects against new sensitizations.

Our aim in this study was to evaluate the effect of allergen
immunotherapy on the development of new allergen sensitivity.

METHODS
The study was conducted by retrospectively examining the
patients who received immunotherapy for respiratory allergic
diseases between 2010 and 2018 in Uludağ University Faculty of
Medicine, Division of Pediatric Allergy. Approval for the study
was obtained from the Uludağ University Faculty of Medicine
Clinical Research Ethics Committee with the decision number
2017/10-27. The study was conducted following principles out-
lined in the Declaration of Helsinki.

Patients
The immunotherapy group consisted of patients who received
house dust mite (HDM) or pollen (weed pollens or weed–cereal
pollens) immunotherapy for at least 3 years, and patients
whose skin prick test was repeated at an interval of at least 3
years after immunotherapy. Controls for both immunotherapy
groups consisted of patients with similar age, gender, allergic
disease, and allergen sensitivity, who did not receive immuno-
therapy, and whose skin prick test was repeated at intervals
of at least 3 years. Among the screened patients, 31 cases who
received HDM immunotherapy and 19 cases who received pol-
len immunotherapy, and 69 controls (42 cases for the HDM
immunotherapy group, 27 cases for pollen immunotherapy)
were included in the study. The ages of the patients receiving
immunotherapy (10.8 ± 3.1 years) and the ages of the control
group (10.1 ± 2.7 years) and the time between 2 tests in both
groups (4.2 ± 1 year for the immunotherapy group and control
4.25 ± 1 year) were similar (P = .165 and P = .690, respectively).

Skin Prick Test
Skin prick tests were applied at Uludağ University Faculty of
Medicine Pediatric Allergy Division Laboratory using ALK-
Abello (Horsholm, Denmark) standard allergen kits and dis-
posable plastic lancets (Stallergenes, Antony, France). After the
allergens were dropped on the volar surface of both forearms
at intervals of at least 2 cm, different lancets were used for each
allergen, allowing the allergens to reach a depth of approxi-
ately 1 mm. Histamine 0.1% (1 mg/ML) was used for positive
control and saline solutions were used for negative control. The
result was considered positive for the relevant allergen when
dema of 3 mm or more was detected compared to the nega-
tive control, 15 minutes after the allergen was administered.
Dermatophagoides farinæ, Dermatophagoides pteronyssii-
nus, grass pollen mix (Dactylis, Festuca, Lolium, Phleum, Poa),
cereal pollen mix (Avena, Hordeum, Triticum, Secale), weed
pollen mix (Artemis, Chenopodium, Parucria, Plantæ) in the
skin prick test panel , Plantago pollen, olive tree pollen, tree
pollen mix (Alnus, Betula, Corylus), Alternaria, cat allergen, dog
allergen, and cockroach allergens were used. House dust mite
sensitivity was accepted as any mite sensitivity, grass pollensens-
tivity as any sensitivity to grass pollen, cereal pollen sensi-
tivity as sensitivity to any cereal pollen, tree pollen sensitivity as
sensitivity to any tree pollen, and sensitivity to weed pollen as
the presence of sensitivity to any weed pollen.

Immunotherapy Protocol
Subcutaneous immunotherapy was applied with the classical
method in all patients. Aluminum hydroxide or calcium phos-
phate-adsorbed standardized extracts or allergoid prepara-
tions (NovoHelisen Depot, Allergovit, Allergopharma, Reinbek,
Germany; Alutard SQ, ALK Laboratories, Hoersholm, Denmark;
APS Retard, Stallergenes, Antony, France) were used for sub-
cutaneous immunotherapy. Immunotherapy injections were
administered in increasing doses weekly in the first 2–6 month
period, which is the initial-dose-increase period; after reach-
ning the maximum tolerated concentration, the maintenance
period was initiated and the same dose was administered at
intervals of 4 weeks for 3–5 years.

Statistical Analysis
Variables are expressed as mean ± standard deviation and
median (minimum, maximum) values. The compliance of con-
tinuous variables to normal distribution was examined using the
Shapiro–Wilk test. Independent samples t-test or Mann–Whitney
U-test was used to compare quantitative data according to normality test results. Categorical variables were
expressed as n (%), the McNemar test was used for the compar-
ison of dependent time measurements within the patient group,
and the Pearson’s chi-square test or Fisher exact test was used
for independent variables. Analyses were made with the SPSS
(IBM Corp. Released 2015. IBM SPSS Statistics for Windows,
Version 23.0, IBM Corp., Armonk, NY) program, and the type I
error level was accepted as α = 0.05 in statistical analysis.

RESULTS
The immunotherapy and control groups were similar in terms
of gender, age, the time between two tests, allergic diseases,
and sensitization rates to a single allergen (Table 1). The mean
immunotherapy duration of the patients who received allergen
immunotherapy was 4.2 ± 0.8 years. The most common dis-
ease in patients receiving house dust mite immunotherapy was
asthma (n = 27, 87.1%), and the most common allergic disease
in the pollen immunotherapy group was allergic rhinoconjunc-
tivitis (n = 17, 89.5%).

When the patients who developed new sensitivity in the aller-
gen immunotherapy and control group were evaluated, no sig-
nificant difference was found between the groups in terms of
the number of patients who developed new sensitivity (Table 2).

Allergen sensitivity was similar between the first tests of the
patients who received HDM immunotherapy and pollen immu-
notherapy, and the first tests of the control group. However,
in the last tests of the patients who received pollen immuno-
therapy and the control group, weed pollen sensitivity (n = 14,
53.8%) was significantly increased in the control group com-
pared to those who received pollen immunotherapy (n = 4,
21.1%) (P = .027) (Figure 1).

The allergen sensitivity in the first tests of the patients who
received immunotherapy and the allergen sensitivity in the last
tests, and the allergen sensitivity in the first tests of the
control groups and the allergen sensitivity in the last tests, were compared. This comparison showed no significant difference between allergen sensitivity in the tests performed at an interval of at least 2 years in the patients who received HDM immunotherapy, whereas, in the control group, tree pollen sensitivity was found to be significantly higher in the last test compared to the first test (n = 2, 5.4%; n = 8, 21.6%, P = .031). Comparison within the series of pollen immunotherapy results for each patient showed no significant difference found between the first and last tests. Weed pollen sensitivity in the last tests of the control group (n = 14, 53.8%) was significantly higher than in the first tests (n = 7, 26.9%) (P = .039). The changes between the first and last tests of the patients who received immunotherapy and control groups are shown in Table 3.

An examination of the effects of age, gender, immunotherapy, allergic diseases, and allergen sensitivities on new sensitization in all patients who received immunotherapy, and in the controls, revealed that the presence of weed pollen sensitivity and a diagnosis of asthma were associated with the development of new sensitivity (odds ratios: 8.1 and 3.8, respectively). Allergen immunotherapy did not show a significant effect on the development of new sensitivity (odds ratios: 8.1 and 3.8, respectively). Allergen immunotherapy was significantly lower than the control group. Similarly, it has been shown in various studies that immunotherapy protects against the development of new sensitivity. In contrast, there are also studies showing that it does not protect against new sensitization and even increases new sensitization. In their prospective study with 22 asthmatic children with HDM sensitivity and the same number in the control group, they found that the development of sensitivity to cat allergens, dog allergens, Alternaria, and grass pollen was less in patients who received immunotherapy. In another prospective study, Cengizlier et al. found that the development of new sensitivity in children who received HDM and grass pollen immunotherapy was significantly lower than the control group. Similarly, it has been shown in various studies that immunotherapy protects against the development of new sensitivity. In a meta-analysis evaluating 32 studies, evidence was presented that allergen immunotherapy reduces the risk of new sensitization in the short term, while no clear evidence of a long-term reduction in sensitization risk was presented. In our study, the number of patients who developed new sensitivity with HDM and pollen immunotherapy was similar to those in the control group. When the sensitivity developments were evaluated separately in each allergen group, there was a significant increase in tree pollen sensitivity in the HDM immunotherapy control group.

### DISCUSSION

The argument that allergen immunotherapy protects against new sensitization was first put forward by Roches et al. in 1997. In their prospective study with 22 asthmatic children with HDM sensitivity and the same number in the control group, they found that the development of sensitivity to cat allergens, dog allergens, Alternaria, and grass pollen was less in patients who received immunotherapy. In another prospective study, Cengizlier et al. found that the development of new sensitivity in children who received HDM and grass pollen immunotherapy was significantly lower than the control group. Similarly, it has been shown in various studies that immunotherapy protects against the development of new sensitivity. In contrast, there are also studies showing that it does not protect against new sensitization and even increases new sensitization. In their prospective study with 22 asthmatic children with HDM sensitivity and the same number in the control group, they found that the development of sensitivity to cat allergens, dog allergens, Alternaria, and grass pollen was less in patients who received immunotherapy. In another prospective study, Cengizlier et al. found that the development of new sensitivity in children who received HDM and grass pollen immunotherapy was significantly lower than the control group. Similarly, it has been shown in various studies that immunotherapy protects against the development of new sensitivity. In a meta-analysis evaluating 32 studies, evidence was presented that allergen immunotherapy reduces the risk of new sensitization in the short term, while no clear evidence of a long-term reduction in sensitization risk was presented. In our study, the number of patients who developed new sensitivity with HDM and pollen immunotherapy was similar to those in the control group. When the sensitivity developments were evaluated separately in each allergen group, there was a significant increase in tree pollen sensitivity in the HDM immunotherapy control group.

### Table 1. General Characteristics of the Immunotherapy Groups and Control Groups

|                                | HDM IT [n (%)] | Control [n (%)] | P     | Pollen IT [n (%)] | Control [n (%)] | P     |
|--------------------------------|----------------|-----------------|-------|-------------------|-----------------|-------|
| Female/male                    | 18/13 (58.1/41.9) | 23/19 (54.8/45.2) | .7791 | 8/11 (42.1/57.9)  | 12/15 (44.4/55.6) | .8751 |
| Age (year) mean ± SD (median, the smallest-the largest) | 11 ± 3.1 | 10.3 ± 2.9 | .2951 | 10.5 ± 3.3 | 9.9 ± 2.5 | .5231 |
| The time between 2 tests (year) mean ± SD (median, the smallest-the largest) | 4.1 ± 1 | 4.4 ± 0.9 | .2941 | 4.3 ± 1.1 | 4.1 ± 0.9 | .5751 |
| Asthma                         | 27 (87.1) | 35 (83.3) | .7501 | 14 (73.7) | 18 (66.7) | .6111 |
| Allergic rhinoconjunctivitis   | 25 (80.6) | 32 (76.2) | .6491 | 17 (89.5) | 23 (85.2) | >.991 |
| Sensitivity to a single allergen, % | 23 (74.2) | 25 (59.5) | .1921 | 9 (47.7) | 8 (29.6) | .2201 |

1 Pearson’s chi-square.
2 Independent samples t-test.
3 Fisher’s exact test.
HDM, house dust mite; IT, immunotherapy; SD, standard deviation.

### Table 2. Patients with New Sensitivity in Allergen Immunotherapy and Control Group

|                                | Immunotherapy, n (%) | Control, n (%) | P     |
|--------------------------------|----------------------|----------------|-------|
| In immunotherapy (n = 50) and control groups (n = 65) | 21 (42) | 25 (38.5) | .7011 |
| Between IT (n = 32) and control (n = 33) groups with sensitivity to a single allergen | 17 (53.1) | 12 (36.4) | .1741 |
| New sensitivity in house dust mite immunotherapy group | 12 (38.7) | 15 (35.7) | .7931 |
| IT (n = 23) and control (n = 25) groups with sensitivity to a single allergen | 11 (47.8) | 8 (32) | .2631 |
| New sensitivity in pollen immunotherapy recipients | 9 (47.4) | 13 (48.1) | .9581 |
| IT (n = 9) and control (n = 8) groups with sensitivity to a single allergen | 6 (66.7) | 4 (50) | .6371 |

Pearson’s chi-square.
1 Fisher’s exact test.
IT, immunotherapy.
The protective effects of house dust mite immunotherapy against pollen and mold sensitivity are controversial. In addition to studies showing that pollen sensitivity develops less in patients who received house dust mite immunotherapy compared to control, studies show that sensitivity develops at a similar or higher rate than in the control group. A similar situation is also valid for the development of new sensitivity to mold spores in patients receiving HDM immunotherapy. However, most of the studies statistically analyze these increases and decreases. In our study, while tree and weed pollen sensitivity increased significantly in control groups, it was observed that this increase was not significant in immunotherapy groups. We think that in patients receiving house dust mite immunotherapy, the protection against the development of new sensitivity to tree pollen compared to the control groups can be explained by the effect of allergen immunotherapy on the natural course of allergic sensitization, rather than by prevention of the development of sensitivity to allergens with similar structures.

In the study conducted by Karaman et al., while none of the patients who received pollen immunotherapy developed new HDM sensitivity, it developed in 13% of the control group. In our study, HDM sensitivity did not change in patients who received pollen immunotherapy.

Inal et al. reported that HDM immunotherapy containing aqueous and adsorbed extract reduced new allergen sensitivity by 3- and 4-fold, respectively. In the meta-analysis conducted by Kristiansen et al. although there is evidence that allergen immunotherapy reduces the risk of new sensitization in the short term, this cannot be confirmed in the sensitivity analysis, and no change was found in the sensitivity risk in the short term (OR: 0.72; 95% CI: 0.24-2.18) and long term (OR: 0.47; 95% CI: 0.08-2.77). Similarly, in our study, it was found that allergen immunotherapy had no significant effect on the risk of new sensitization.

Allergic sensitivity occurs as a result of the interaction of many different factors. Some of these factors are genetic, exposure time to allergens, amount of allergens one is exposed to, air pollution, socioeconomic status, and diet. However, data on the factors affecting new sensitization are limited. In our study, when the factors affecting the development of new sensitivity were examined, it was found that being diagnosed with asthma and having weed pollen sensitivity increased the risk of new sensitization.
The natural course of allergic sensitization is not yet fully understood. Sensitivity to HDM and pet allergens, which are among the respiratory allergens, usually precedes the development of sensitivity to fungal and pollen allergens. As in the general population, many studies investigating the development of new sensitivity in patients receiving allergen immunotherapy in our country have also found that new sensitivity developed is especially against pollens. In our study, similar to the literature, the most frequent new sensitivity developed was against pollen.

Our study has some limitations, namely, the retrospective nature of the study, the small number of patients, the evaluation of allergen sensitivity by the skin prick test alone, and the exclusion other factors that may affect sensitization.

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### Table 3. Comparison of Allergic Sensitization in the First and Last Skin Prick Tests of Patients Receiving Allergen IT and Control Groups

| Sensitized Allergens | IT Group | Control Group |  
|----------------------|----------|---------------|
|                      | First Test, n (%) | Last Test, n (%) | P | First Test, n (%) | Last Test, n (%) | P |
| House dust mite IT Recipients and Control Group |          |               |   |          |               |   |
| House dust mite      | 31 (100) | 31 (100) | -  | 42 (100) | 39 (90.5) | -  |
| Grass pollen         | 4 (13.3) | 8 (26.7) | .219 | 10 (24.4) | 12 (29.3%) | .727 |
| Cereal pollen        | 4 (13.3) | 9 (30) | .125 | 8 (21.1) | 11 (28.9%) | .375 |
| Tree pollen          | 3 (10)   | 5 (16.7) | .500 | 2 (5.4)  | 6 (16.6%)  | .031 |
| Weed pollen          | 1 (3.3)  | 2 (6.7) | >.99 | 2 (4.9)  | 7 (17.1%)  | .063 |
| Alternaria           | 1 (3.3)  | 3 (10)  | .500 | 3 (7.3)  | 7 (17.1%)  | .219 |
| Cat                  | 3 (10.3) | 4 (13.8) | >.99 | 5 (9.1)  | 10 (18.2) | .063 |
| Dog                  | -        | 1 (4)   | -   | 1 (2)    | 4 (7.6)   | .250 |
| Cockroach            | 1 (3.4)  | 3 (10.3) | .500 | 1 (2)    | 2 (4.1)   | >.99 |
| Pollen IT recipients and control group |          |               |   |          |               |   |
| House dust mite      | 7 (36.8) | 7 (36.8) | >.99 | 12 (44.4) | 13 (48.1) | >.99 |
| Grass pollen         | 18 (100) | 17 (94.4) | -   | 27 (100) | 25 (92.6) | -   |
| Cereal pollen        | 15 (78.9) | 18 (94.7) | .375 | 21 (95.5) | 19 (86.4) | .500 |
| Tree pollen          | 2 (10.5) | 7 (38.6) | .180 | 3 (13.6) | 8 (36.4)  | .063 |
| Weed pollen          | 6 (31.6) | 4 (21.1) | .727 | 7 (26.9) | 14 (53.8) | .039 |
| Alternaria           | 1 (6.3)  | 4 (25)   | .250 | 1 (3.7)  | 5 (18.5)  | .125 |
| Cat                  | 1 (5.6)  | 1 (5.6)  | >.99 | 2 (7.7)  | 5 (19.2)  | .375 |
| Dog                  | 1 (5.9)  | 1 (5.9)  | >.99 | -        | -        | -    |
| Cockroach            | -        | -        | -   | 2 (11.1) | -        | -    |

† McNemar test.
IT, immunotherapy.

### Table 4. Univariate and Multivariate Analyses of Factors Affecting New Sensitization

|                          | Single Variable Model |         |         | P   | Multivariate Model |         |         | P   |
|--------------------------|-----------------------|---------|---------|-----|--------------------|---------|---------|-----|
|                          | OR                    | 95% CI  | P       |     | OR                | 95% CI  | P       |     |
| Immunotherapy            | 0.724                 | 0.3     | 1.7     | .455|                    |         |         |     |
| Age                      | 0.978                 | 0.85    | 1.1     | .749|                    |         |         |     |
| Gender                   | 0.79                  | 0.3     | 1.8     | .584|                    |         |         |     |
| Asthma                   | 3.806                 | 1.2     | 11.9    | .021| 3.8               | 1.3     | 10.8    | .013|
| Allergic rhinitis        | 0.933                 | 0.3     | 2.8     | .902|                    |         |         |     |
| Grass pollen sensitivity | 0.546                 | 0.1     | 2.9     | .481|                    |         |         |     |
| Cereal pollen sensitivity| 1.308                 | 0.2     | 8.5     | .779|                    |         |         |     |
| Tree pollen sensitivity  | 0.954                 | 0.2     | 4.4     | .952|                    |         |         |     |
| Weed pollen sensitivity  | 11.17                 | 1.9     | 66.3    | .008| 8.1               | 1.5     | 42.4    | .013|
| HDM sensitivity          | 1.319                 | 0.3     | 5.4     | .699|                    |         |         |     |
| Cat sensitivity          | 1.818                 | 0.4     | 8.5     | .448|                    |         |         |     |

HDM, house dust mite; OR, odds ratio.

### CONCLUSION

Allergen immunotherapy is currently the only treatment method that can change the natural course of allergic diseases and has long-term effects on pathophysiology. The improving effect of allergen immunotherapy on clinical findings has been demonstrated in many meta-analyses, but its protective effect against new allergen sensitivity is controversial. In our study, it was determined that allergen immunotherapy protected against new sensitizations to tree and weed pollens compared to control groups, but its protective effect against new sensitization was not significant in multivariate analysis. Sensitivity to weed pollen and a diagnosis of asthma were found to be risk factors for the development of new sensitivity.
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