

A Randomized Double-Blind Comparative Study of Sublingual Immunotherapy for Cedar Pollinosis

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ABSTRACT

Background: Seasonal allergic rhinitis (SAR) induced by Japanese cedar pollen is a substantial problem in Japan. Sublingual immuno-therapy (SLIT) is safer than conventional antigen-specific immunotherapy, the only treatment modality by which complete cure of the disease can be expected. We investigated the safety and efficacy of SLIT in the treatment of cedar pollinosis patients compared to placebo.

Methods: A randomized, placebo-controlled, double-blind study was conducted in 61 cedar pollinosis patients. Increasing doses of standardized Japanese cedar extract or placebo were administered sublingually in intervals ranging from daily to once a week after six weeks. The primary efficacy variable was the mean of the daily total symptom scores (TSS) during the pollen dispersing period. Secondary efficacy variables included the QOL scores and related variables.

Results: Primary efficacy variable scores were significantly lower for some days in the SLIT group than in the placebo group ($P < .01$ or $P < .05$). Secondary efficacy for the QOL score in SLIT group was almost of half of placebo group. There was no significant difference in the overall incidence of side effects between the SLIT group and the placebo group.

Conclusions: SLIT was effective and safe in the treatment of cedar pollinosis.

KEY WORDS

Japanese cedar, placebo-controlled study, QOL, seasonal allergic rhinitis

INTRODUCTION

In agreement with the results of worldwide epidemiological assessments, the number of patients with allergic rhinitis such as Japanese cedar (JC) pollinosis in Japan is increasing.¹ Okuda considers that the current prevalence of allergic rhinitis is 16%, but many researchers predict that the rate will still increase.² Pollinosis is a typical type I allergy in which allergic conjunctivitis and allergic rhinitis develop. In spite of its refractory nature, pollinosis deteriorates patient QOL only in severe cases; however, it greatly affects the patient's life in general in that they must keep

working even if the condition is severe.³ Many of the patients with cedar pollinosis have also been sensitized to cypress pollen which disperses after cedar pollen. Consequently, symptoms of cedar pollinosis are followed by those of cypress pollinosis; patient symptoms last, though they are seasonal, for as long as 4 months (from February to May).

Pharmacological therapy prescribed by general practitioners is common for the treatment of the disease. Both oral medications and topical medication, however, are symptomatic treatment; they do not cure the disease or remain effective until the following year.⁴ Antigen-specific subcutaneous immuno-

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therapy (SCIT) is the only treatment modality by which complete cure of the disease can be expected.⁵ WHO position paper stipulates the use of standardized antigen and the concentration of the antigen to be maintained.⁶ The efficacy of the therapy has been proven in placebo-controlled, double-blind comparative studies using pollen, house dust mite, and animal protein.^{6,7} In Japan, it is customary to start the administration of causative antigen extract by subcutaneous injection at the threshold of skin reaction or its 10-fold diluted concentration, and to increase the dose gradually.⁴ Treatment with SCIT requires special attention because it may cause, as a side effect, anaphylactic shock, which prevents the therapy from becoming popular in Japan.⁸ In order to reduce the possibility of this side effect, immunotherapy is administered by other routes (sublingual, intranasal, oral, and transbronchial) in Europe and the United States, and has achieved desired outcomes.⁹⁻¹¹ Especially, sublingual immunotherapy (SLIT) has become popular in Europe considerably, and there are many reports supporting the effectiveness of the therapy.¹¹⁻¹³ As for side effects due to SLIT, there are no reports of anaphylactic shock, but oral itching, skin reaction (such as urticaria), and mild asthma-like attacks have been reported.¹³ Since cedar pollinosis greatly deteriorates patient QOL, many physicians and patients will opt for immunotherapy if it is proven to be safe. We conducted a randomized, placebo-controlled double-blind comparative study to investigate whether SLIT reported in Europe and the United States is effective for the treatment of JC pollinosis and whether it can be performed safely.

STUDY DESIGN

This multi-centre, double-blind, randomized, placebo-controlled, parallel-group study was conducted in six centers across Japan between October 2004 and April 2005. The study protocol was approved by the appropriate local ethics committees, and the study was conducted in accordance with the Declaration of Helsinki and Good Clinical Practice guidelines. All patients provided written informed consent prior to participation.

SUBJECTS

Patients with JC pollinosis were enrolled in this study if they had a RAST score of 2 against JC or above and pollinosis symptoms during the cedar pollen dispersal period at least in the past 2 years and if they had visited any of the following medical institutions: Department of Otorhinolaryngology, Nippon Medical School; Department of Otorhinolaryngology, University of Fukui; Department of Otorhinolaryngology, Head and Neck Surgery, Okayama University; Department of Otorhinolaryngology, Dokkyo University School of Medicine; Department of Otorhinolaryngology, University of Yamanashi; and Department of

Otorhinolaryngology, Head and Neck Surgery, Chiba University. Patients who had nose diseases (perennial allergic rhinitis, nasal septum deviation, or sinusitis) which may interfere with accurate symptom assessment were excluded from the study. Patients receiving treatment for conditions such as severe cardiac disease and malignant tumor were also excluded. As a result, a total of 61 patients were blindly randomized either to the active group or the placebo group in the ratio of 2 active to 1 placebo.

METHODS

The study was initiated in October, 2004. Patients were assigned and randomized to either the active group or the placebo group. Cedar antigen extract (active group) at concentrations of 2 to 2000 JAU/ml diluted with diluent (made by Torii Pharmaceutical Co., Ltd.) and diluent alone (placebo group) were used in eye drop containers (made by Hirakata Plastic).

Administration of the antigen extract was started at 2 JAU/ml, which is considered a sufficiently safe level, and was increased to the final maintenance concentration of 2000 JAU/ml. Active drug was administered as follows: 1 drop (about 50 µl) to 20 drops (about 1 ml) of prepared extract was dropped onto bits of bread (about 1.5 cm × 1.5 cm × 1.5 cm), which were held sublingually for 2 minutes and then expectorated. The treatment schedule was as follows: antigen extract was administered sublingually daily from Week 1 to Week 4; 20 drops of the antigen extract 2000 JAU/ml were administered two days per week in Week 5, once per week in Week 6 and thereafter throughout the season (Table 1).

Patients experiencing pollinosis symptoms in cedar and cypress pollen dispersal periods received symptomatic treatment with medications such as antihistamines on an as needed basis; such patients were asked to record the date of treatment in their allergy diary.

ENDPOINTS

The patients were instructed to fill in their allergy diary from February 22, 2005 to April 6, 2005, the period when cedar and cypress pollen dispersed in 2005, and they were also asked to fill in QOL questionnaire once a month during the same period. Symptoms recorded in the allergy diary (sneezing, runny nose, nasal congestion, and interference with daily life), the total nasal symptom scores calculated based on each symptom, sneezing, runny nose, nasal congestion (none; 0, mild; 1, moderate; 2, severe; 3), and symptom medication scores (antihistamine; 1, topical steroid; 2, general steroid; 3) were calculated. The Japanese Allergic Rhinitis QOL Standard Questionnaire No.1 (JRQLQ No1) was used for the assessment of the QOL of patients with allergic rhinitis (Fig. 1). Nasal and Ocular symptom scores, QOL-

Table 1 Allergen administration schedule (Increasing dosing)

	Week 1 (2 JAU/ml)	Week 2 (20 JAU/ml)	Week 3 (200 JAU/ml)	Week 4 (2000 JAU/ml)	Week 5 (2000 JAU/ml)
Day 1	1 drop	1 drop	1 drop	1 drop	20 drops
Day 2	2 drops	2 drops	2 drops	2 drops	
Day 3	3 drops	3 drops	3 drops	4 drops	
Day 4	4 drops	4 drops	4 drops	8 drops	
Day 5	6 drops	6 drops	6 drops	12 drops	20 drops
Day 6	8 drops	8 drops	8 drops	18 drops	
Day 7	10 drops	10 drops	10 drops	20 drops	

Initial dose of SLIT for JC pollinosis was 1 drop of 2 JAU/ml of standardized JC allergen, and the administering dose is increased up to 20 drops of 2000 JAU/ml at 4th week, the maintenance dose.

Japanese Rhino-conjunctivitis Quality of Life Questionnaire (JRQLQ No.1)

To patients with allergic rhinitis (including pollinosis)
These days, the aim of medical treatment is not just to cure disease but also to give patients a better quality of life. The purpose of this survey is to determine to what extent your rhinitis interferes with your life and whether it would be improved by treatment. As with all medical treatment, the information you provide in this survey will remain strictly confidential.

You may find some of the following questions difficult to answer, but just answer to the best of your ability.

/ Tick the box that best describes the severity of the worst nasal and eye symptoms you have experienced in the past 1–2 weeks.

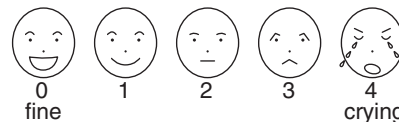
Nasal and eye symptoms	0. No	1. Mild	2. Moderate	3. Severe	4. Very severe
Runny nose	☐	☐	☐	☐	☐
Sneezing	☐	☐	☐	☐	☐
Blocked nose (nasal congestion)	☐	☐	☐	☐	☐
Itchy nose	☐	☐	☐	☐	☐
Itchy eyes	☐	☐	☐	☐	☐
Watery eyes	☐	☐	☐	☐	☐

// Tick the box that best describes the worst extent to which the symptoms in / above have interfered with your quality of life in the past 1–2 weeks. If any of the items listed under Quality of life below definitely do not relate to the symptoms in / (nose, eyes), then there is no need to tick a box for that particular item.

Quality of life	0. No	1. Yes, slightly	2. Yes, moderately	3. Yes, severe	4. Yes, very severe
1. Reduced at work/home	☐	☐	☐	☐	☐
2. Poor moral concentration	☐	☐	☐	☐	☐
3. Reduced thinking power	☐	☐	☐	☐	☐
4. Impaired reading book/newspaper	☐	☐	☐	☐	☐
5. Reduced memory loss	☐	☐	☐	☐	☐
6. Limitation of out of life (e.g. sport, picnics)	☐	☐	☐	☐	☐

7. Limitation going out	☐	☐	☐	☐	☐
8. Hesitation friends or relatives	☐	☐	☐	☐	☐
9. Reduced contact with friends or others by telephone or conversation	☐	☐	☐	☐	☐
10. Not an easy to be around	☐	☐	☐	☐	☐
11. Impaired sleeping	☐	☐	☐	☐	☐
12. Tiredness	☐	☐	☐	☐	☐
13. Fatigue	☐	☐	☐	☐	☐
14. Frustration	☐	☐	☐	☐	☐
15. Irritability	☐	☐	☐	☐	☐
16. Depression	☐	☐	☐	☐	☐
17. Unhappiness	☐	☐	☐	☐	☐

/// Please circle the number of the face that best describes your general state (including your symptoms, life and emotion) in the past 1–2 weeks.



Do not fill out the following

To be completed by physician	Patient's name:	Medical record to:	Age: yr	Sex: M F
	Name of medical institution:	Physician's name:	Date:	
	Diagnosis:			
	SAR (Antigen) Treatment [prevention, drug, immunology, therapy, operation]			
	PAR (Antigen) Treatment [prevention, drug, immunology, therapy, operation]			
	Non-Allergy: Disease: () Treatment: ()			
QOL score: None 0, Mild 1, Moderate 2, Severe 3, Very severe 4				
Total QOL score				
Score by QOL category 1–5 points daily life 6–7 points out-door 8–10 points social 11 points sleep 12–13 points body 14–17 points psycho-life				
Please write the names of drugs used it possible				
Score: None: 0 points Mild: 1 point Moderate: 2 points Severe: 3 points Very Severe: 4 points				

Fig. 1 Japanese Allergic Rhinitis QOL Standard Questionnaire No.1 (JRQLQ No1).

related questionnaire scores, and the overall face scale were calculated and statistically analyzed. In other words, the QOL deterioration score was calculated by subtracting QOL-related questionnaire scores recorded in February (i.e. at baseline) from the scores recorded in the middle of March to April, when the largest amount of pollen dispersal was ob-

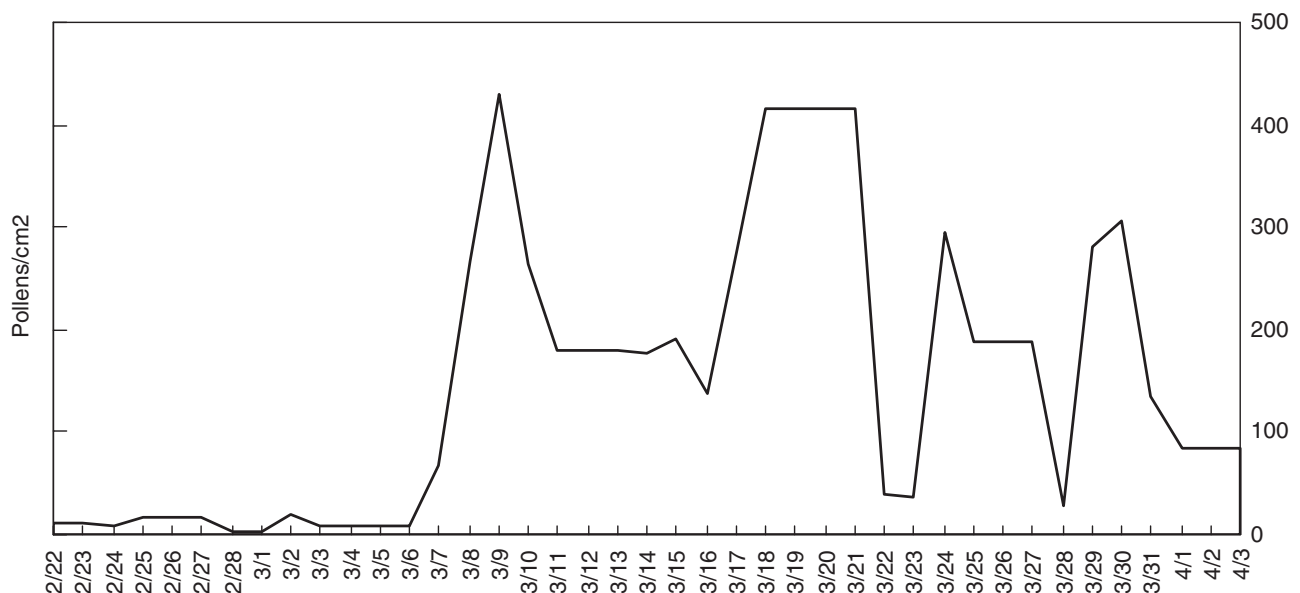
served.

STATISTICAL ANALYSIS

Symptom scores, total symptom scores, and symptom medication scores calculated from the allergy diary in the placebo group and the active group were analyzed by non-paired t-test and the Chi-squared test

Table 2 The background of the subjects

Items	Placebo <i>n</i> = 22	Active <i>n</i> = 37	<i>p</i> value (*)
Age	40.14±15.30	40.65±15.14	0.901
Sex			
Male	7 (31.8%)	18 (48.6%)	
Female	15 (68.2%)	19 (51.4%)	
Nasal and eye symptoms	0.62±0.54	0.43±0.35	0.169
QOL-related questionnaire	0.25±0.29	0.21±0.25	0.568
Usual daily activities	0.13±0.25	0.11±0.32	0.762
Outdoor activities	0.14±0.48	0.21±0.46	0.630
Social functioning	0.05±0.22	0.08±0.23	0.648
Sleep disturbance	0.14±0.36	0.09±0.29	0.537
Physical problems	0.17±0.33	0.24±0.46	0.510
Emotional function	0.12±0.23	0.11±0.32	0.953
Overall face scale	1.14±0.73	1.09±0.74	0.780

**Fig. 2** The changing the number of cedar and cypress pollen dispersals in 2005.

using SPSS 11.0J. QOL-related questionnaire score of the 2 groups were compared using analysis of covariance (ANCOVA).

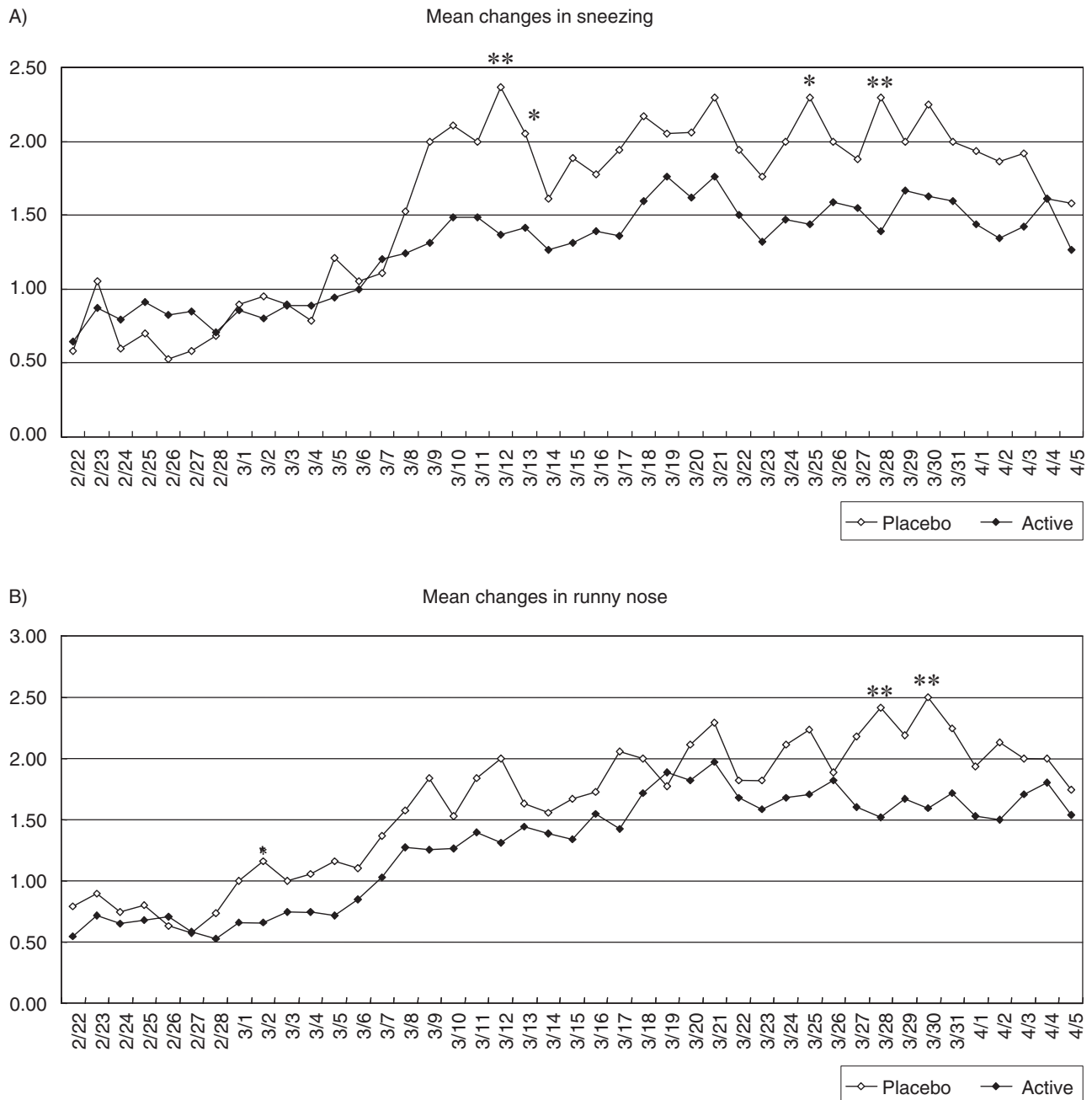
RESULTS

Of the 61 randomized patients, there were 2 dropouts for those whose treatment was unknown; there were 37 patients in the active group and 22 patients in the placebo group. In the analysis of allergic symptoms, 2 patients whose outcome was available only in the form of a diary were excluded, and the results of 36 patients in the active group and 21 patients in the placebo group were analyzed. In the analysis of QOL, 3 patients were excluded because baseline assessment was unavailable, and the results of 35 patients in the

active group and 21 patients in the placebo group were analyzed.

As shown in Table 2, no difference was observed between the two groups in terms of patient characteristics (sex was analyzed by the Chi-squared test, and other items were analyzed by t-test).

In 2005, the number of cedar and cypress pollen dispersals observed was the largest during the 10-year period since 1995. According to the data of the Chiyoda ward—the area nearest to the Nippon Medical School—announced by the Tokyo Metropolitan Government, the first pollen dispersal was observed on February 22, which was about the same time as in the past years, and an average of 10,625 pollens per square centimeter by the Durham method were ob-



served during the season (Fig. 2). The number of the cedar and cypress pollens by the same method observed in each institution was 3424, 2383, 16002, 5859 and 7752 for University of Fukui, Okayama University, Dokkyo University, University of Yamanashi and Chiba University respectively, and these pollen numbers were also largest dispersing during the last ten years at any place.

Symptom scores for sneezing (Fig. 3A) and runny nose (Fig. 3B) in the active group were significantly better than those in the placebo group on 4 days and 2 days, respectively, but no difference was observed

between the active group and the placebo group in terms of nasal congestion (Fig. 3C). Between the 2 groups, there was no difference in the number of medications used during the season (Fig. 3D).

The active group had a significantly lower total symptom score (Fig. 4A) and symptom medication score (Fig. 4B) on 4 days during the season. Overall, better outcomes were observed in the active group during the latter half of the season (i.e. from the end of March to the beginning of April), which roughly overlaps the period when the largest amount of cedar and cypress pollen was dispersed.

In the placebo group, the nasal and ocular symptom score was 1.15, the QOL-related questionnaire score was 1.10, and the overall face scale score was 1.24; in the active group, the nasal and ocular symptom score was 0.92, the QOL-related questionnaire score was 0.58, and the overall face scale score was 1.03: the deterioration score in the QOL-related questionnaire in the active group was only about half the

score in the placebo group (Fig. 5A). In each domain of QOL question items, deterioration in usual daily activities, outdoor activities, social functioning, sleep problems, general physical problems, and emotional function in the active group was only about half the score in the placebo group as well. The *p*-values for the above domains were 0.089, 0.086, 0.067, 0.060, 0.083 and 0.046; a significant difference was observed

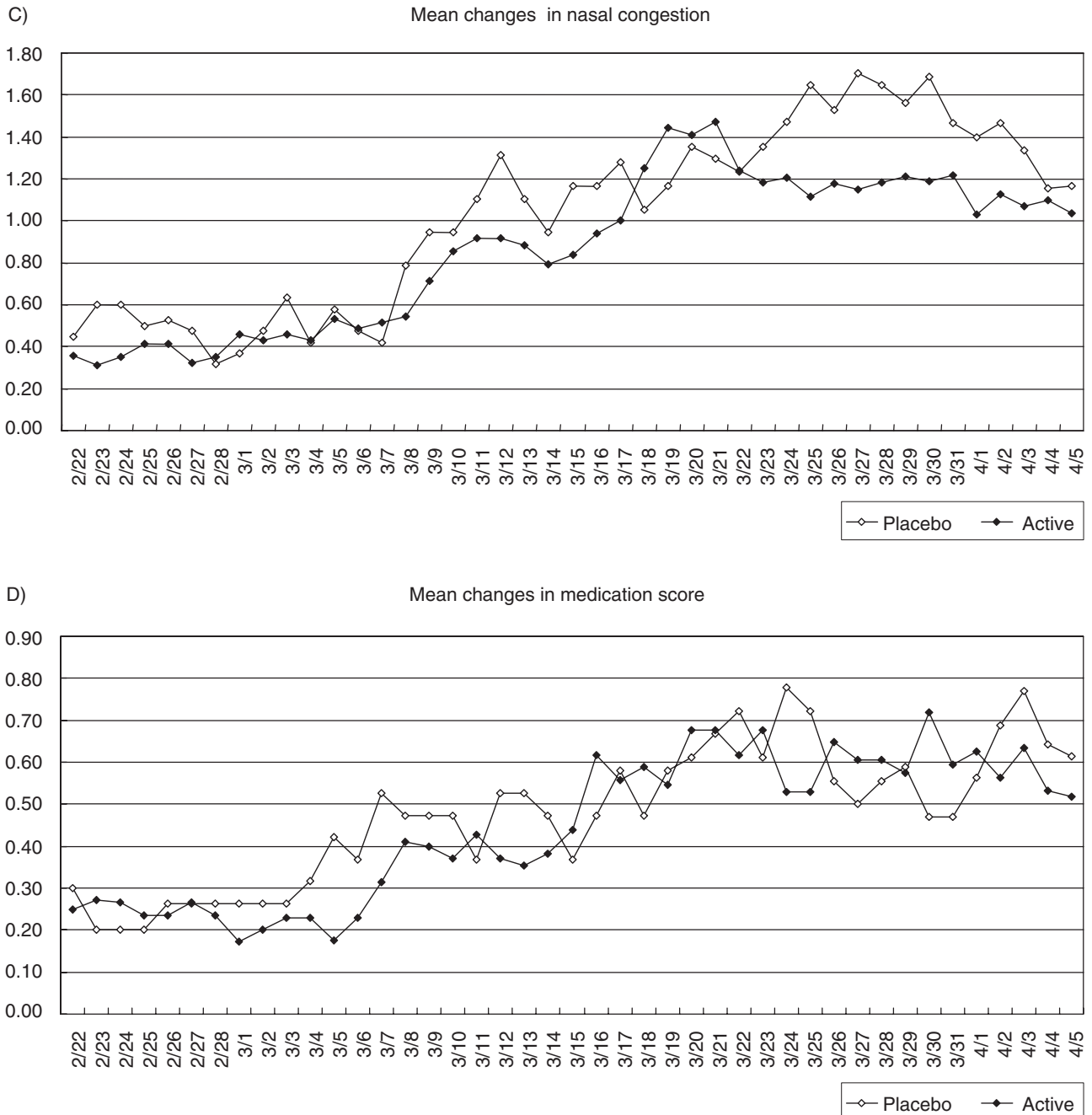
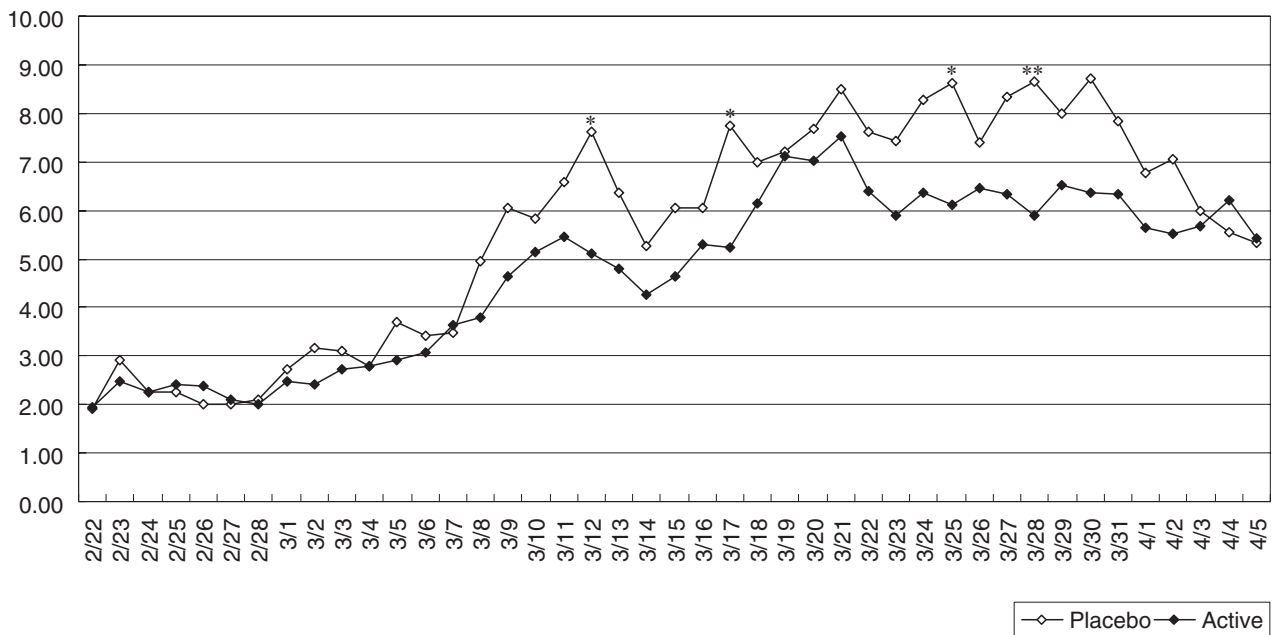


Fig. 3 The mean changes in each symptom in the season of 2005. **A)** Mean changes in sneezing. **B)** Mean changes in runny nose. **C)** Mean changes in nasal congestion. **D)** Mean changes in medication score. The open square indicates the placebo group, the filled square indicates the active group. Significant difference was evaluated as * $p < 0.05$; ** $p < 0.01$.

A)

Mean changes in TSS



B)

Mean changes in symptom medication score

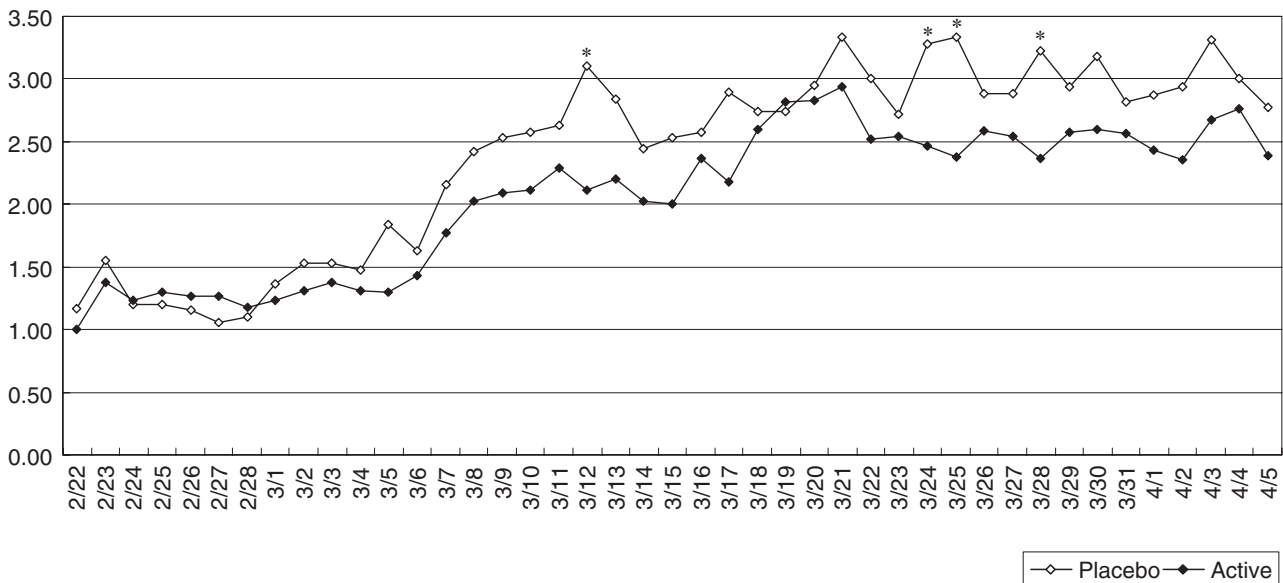


Fig. 4 The mean changes in **A)** the total symptom score (TSS) and **B)** symptom medication score in the season of 2005. Significant difference was evaluated as * $p < 0.05$; ** $p < 0.01$.

only in emotional function (Fig. 5B).

SIDE EFFECTS

No systemic side effect occurred during SLIT. Local side effects occurred in six volunteers in the active group. Mild mouth itching was exhibited in all six volunteers in increasing dose up to 2000 JAU 1 ml, how-

ever this itching was diminished for two or three times just after allergen administration. All six volunteers finished this study totally without any change of this protocol.

DISCUSSION

Approximately 16% of the Japanese population are af-

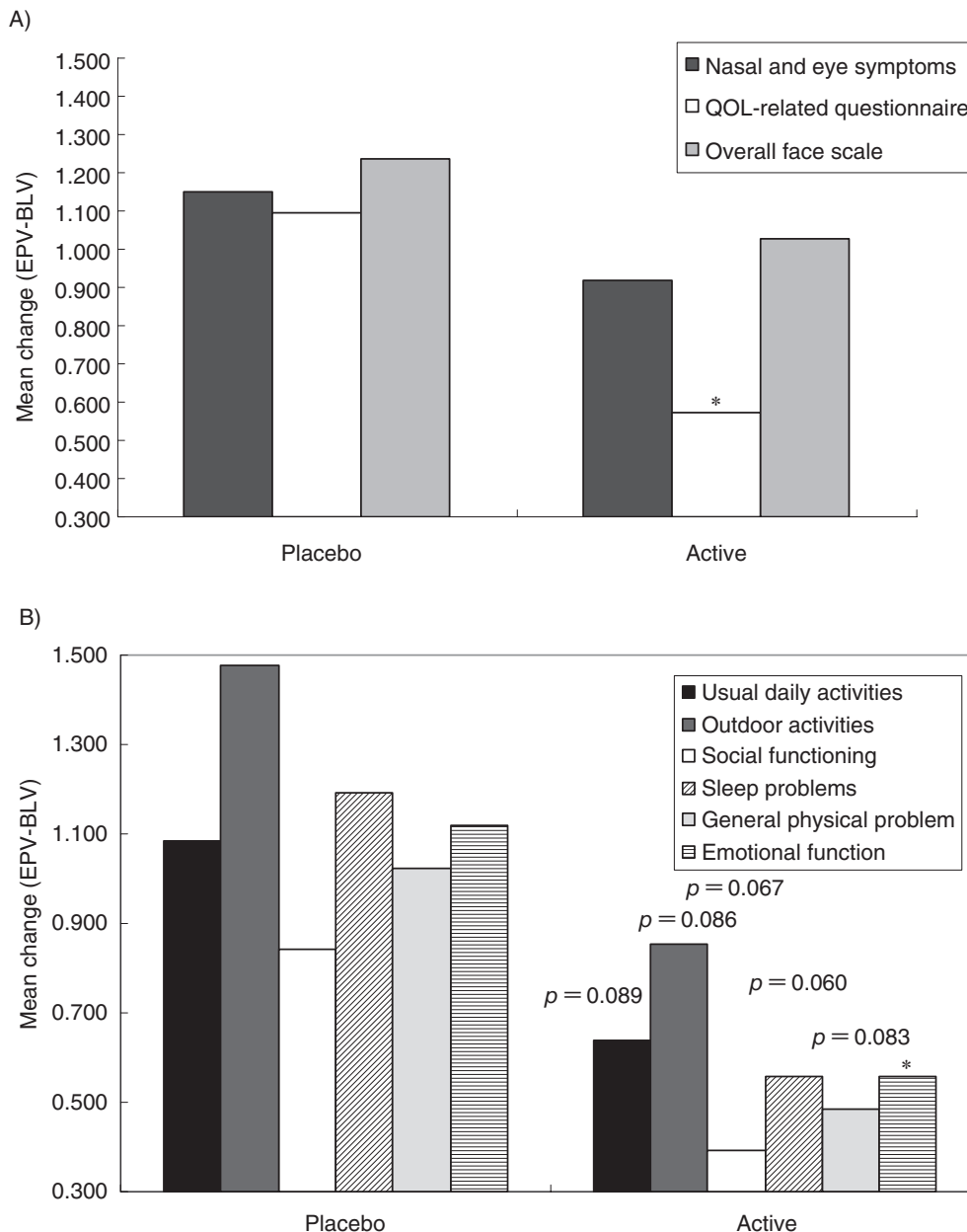


Fig. 5 The mean changes of **A**) QOL scores (nasal and eye symptoms, QOL related questionnaire, overall face scale) and **B**) each domain of QOL question items deterioration (in usual daily activities, outdoor activities, social functioning, sleep problems, general physical problems, and emotional function) from baseline data of February to peak data of peak pollen scattering period. Difference between placebo and active indicates * $p < 0.05$ (analysis of covariance, ANCOVA). Placebo: $n = 21$, Active: $n = 35$.

affected by Japanese cedar pollinosis² and the proportion of severe status patients is higher than with grass or ragweed pollinosis, which are the representative conditions in other countries, and the symptoms persist for about 3 months, becoming a social issue. When the amount of pollen increases, patients show more severe symptoms, and the number of severe status patients is greatest in mid-March (late

season) when the pollen count reaches its peak. Substantial antigen exposure enhances the antigen-antibody reaction in the airways (airway hypersensitivity), which is the mechanism involved in severe pollinosis, and SCIT may control the exacerbation of the symptoms in the latter half of the cedar pollen season by inhibiting antigen-related enhancement of nasal mucosal hypersensitivity.

As shown in the WHO position paper, the effects of immunotherapy in the treatment of pollinosis have been substantiated in many double-blind comparative studies.⁶ However, the therapy tends to be avoided in Japan because of factors such as the current high cost, the complicated procedure involved, and possible side effects. In Japan, owing to these disadvantages and the fact that the department of allergy has not been widely established in medical institutions, pharmacological therapy is the mainstream modality for the treatment of pollinosis. Still, immunotherapy is an important modality for the complete cure of allergic diseases.

The efficacy of our SLIT was not demonstrated based on patient allergy diaries. However, the quality of life (QOL) score was approximately 1/2 of that in the placebo group, with a significant difference. In addition, a *P*-value corresponding to a significant difference was obtained in each QOL domain. In the mental health domain, there was a significant difference. Assessment using the Japanese guidelines differs from that in other countries; even a single sneeze is regarded as (+). In other countries, 4 grades (none, mild, moderate, and severe) are employed for assessment, and the presence or absence of symptoms is not evaluated. For this reason, the usefulness of SLIT may not have been demonstrated based on diaries. However, the QOL is evaluated via self-assessment, which is consistent with the system for the self-reporting of symptoms in other countries (none to severe). Therefore, QOL assessment of SLIT was favorable, and was consistent with the reduction rates in other countries. According to the JRQLQ criteria, the reduction rate for nasal/ocular symptoms was 22%, consistent with the evaluation of SLIT in other countries. In the future, the JRQLQ criteria, which were designed in reference to overseas self-assessment, may be essential for evaluating drug efficacy and such a novel treatment. This finding is suggestive of the fact that the QOL questionnaire developed in Japan is of good quality,¹⁴ and that SLIT is effective for preventing QOL deterioration in patients with pollinosis rather than for lowering their symptom score. Placebo effects of SLIT may be present. However, it was evaluated in 2005, when the amount of scattered pollen was highest over the past 10 years. In addition, considering that the study involved a placebo-controlled design, we can conclude that SLIT was effective for cedar pollinosis in Japan. In evaluating the treatment response, we cannot rule out the influence of Japanese cypress pollen scattering. However, in a study excluding Japanese cypress pollen-positive reacting patients, the efficacy of SLIT and reduction rate for symptoms were also similar (unpublished data). This may be caused by the combination of a large amount of JC and a small amount of cypress that was dispersed in 2005. These types of pollinosis should be regarded as JC/Japanese cypress pollino-

sis, as their seasons are sequential in the near future. In addition, a Japanese cypress pollen antigen for immunotherapy must be prepared. It should be considered that symptoms of cedar/Japanese cypress pollinosis in April are associated with cedar pollen scattering-related nasal mucosal/conjunctival inflammation, not with Japanese cypress pollen scattering alone.

Less side effects including problematic anaphylaxis are noted in SLIT although the side effects observed cannot be theoretically complete anaphylactic shock when comparing the therapy administered via injection with sublingual route.¹⁵ Similar to the oral allergy syndrome (OAS), which is the focus of public attention, the development of symptoms such as strange feelings, oral itching, and swelling were feared because the antigen remains in the oral cavity; however, itching was the only reaction observed so far. The results obtained from the study of tentative SLIT, which was performed exclusively in the Department of Otorhinolaryngology, Nippon Medical School, were roughly consistent with the results of similar studies conducted every year thereafter, including the results of the study in 2005.¹⁶ In our study of SLIT for the treatment of cedar pollinosis, symptom medication score was consistently lower than that of the pharmacological therapy group throughout the pollen dispersal season. The finding indicates that patients receiving SLIT tend to use fewer drugs, which is consistent with the results of a double-blind comparative study using a placebo,¹⁷ SLIT, which is as effective as pharmacological therapy and decreases the amount of drug use, is considered advantageous also in the current medical economy in Japan.

The mechanism of action for SLIT, or for conventional SCIT, is still unclear, but for SCIT, reduction of effector cells^{18,19} and blocking antibody²⁰⁻²³ have been the conventional theories. Recently, however, it has become widely accepted that immunotherapy may modify the T cell response to natural allergen because of T cell anergy and/or immune deviation.²⁴⁻²⁷ For SLIT in particular, allergen administered to the oral mucosa accumulates in the submandibular lymph node, in which the immune response occurs²⁸ and peaks at approximately 2 hours after administration.²⁹ In our investigation, an increase in the Stimulatory Index in PBMC during the early phase of SLIT conducted in 1999 shows at least that systemic immune induction was caused by sublingually administered antigen.³⁰ In SLIT, it is intended to cause fewer side effects than SCIT injection by decreasing systemic effects. However, it has become clear that the therapy also leads to systemic immune induction, which is greatly different from conventional topical immunotherapy administered intranasally or orally.

In the present study, SLIT both inhibited the exacerbation of symptoms in the latter half of the season

and reduced their severity throughout the season. Furthermore, there were neither local nor systemic side effects, as reported elsewhere for other antigens. SLIT for cedar pollinosis is a new therapy and in the future SLIT may be indicated for patients with nasal allergy caused by other allergens such as house dust mites or animal dander through improvement of the administration schedule and establishing the dose at which the most potent effects are achieved. This study may contribute to the methodology for the future immunotherapy in Japan.

The development of this SLIT in Japan is in progress as a multi-center study conducted as part of the research project on the prevention and treatment of immunological and allergic diseases (H17-immunology-common-001) entitled "Evaluation research of the relationship between the number of dispersed pollen observed by real-time monitoring and QOL achieved by the current treatment modalities, and the development of definitive treatment for pollinosis", which is supported by Health and Labour Sciences Research Grants from the Ministry of Health, Labour and Welfare.

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