表3　代替医療実態調査からのまとめ（2008年末：24,667名の調査）

1	代替医療の受療理由	副作用少なく安全，安価，医師受診が面倒．
2	疾患による違い	ヨーグルト，乳酸菌は全ての疾患，年齢で多いが，鼻炎では甜茶，アロマ，アトピー性皮膚炎では温泉療法が増加．
3	成人と小児の違い	成人が多い，小児は小学生から増加，内容は大差ない．
4	地域差	疾患の少ない地域では少ない．
5	病院の規模による違い	大学病院，アレルギー専門病院受診者で高い．
6	性差	明らかではない．
7	患者の評価	多くは30%以下，アトピー性皮膚炎，小児鼻炎ではやや高い．
8	副作用	温泉療法以外は少ない．
9	費用	約20%が10万円以上，アトピー性皮膚炎で高く，鼻炎で低い．
10	情報入手先	家族・友人が多く，インターネットは少ない．
11	医師への相談	多くの患者は医師に代替医療について話していない．
12	医師への反応	医師の大部分は代替医療を否定はしていない．
13	医療機関未治療患者	代替医療の受療が高い可能性．
14	市民講座受講者・インターネット調査参加者	非常に高い代替医療受療率，一般患者とは少し乖離．

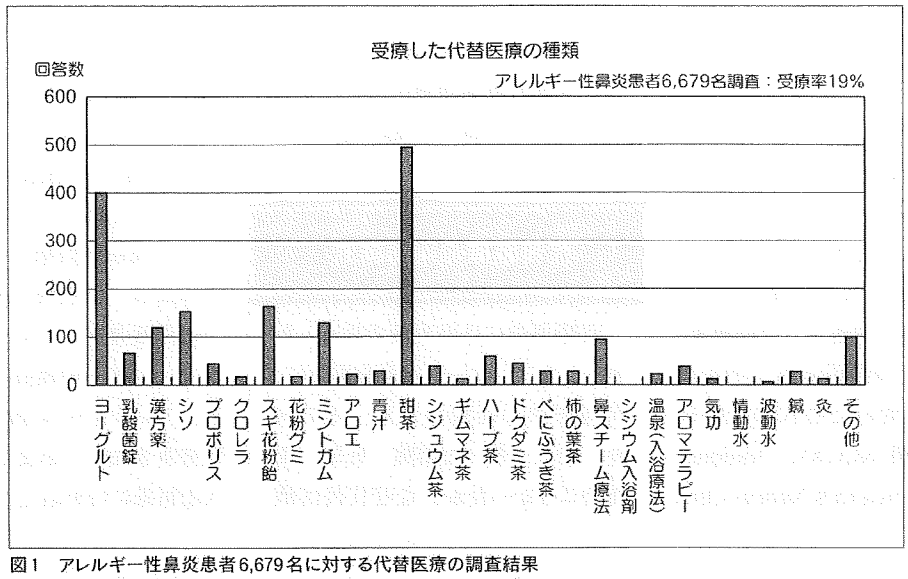

図1　アレルギー性鼻炎患者6,679名に対する代替医療の調査結果
さまざまな代替医療が用いられているが，特に甜茶，ヨーグルト，スギ花粉飴，ミントガム，鼻スチーム療法，漢方薬（医師の処方によらないもの）乳酸菌剤，シソなどの割合が高かった．

今回，24,667名を対象とした検討をまとめると表3の如くであり，さまざまな代替医療が用いられていたが（図1）代替医療の内容について疾患による違いは，ヨーグルトや乳酸菌は全ての疾患や年齢で多いが，アレルギー性鼻炎では甜茶やアロマ療法，アトピー性皮膚炎では温泉入浴療法が目立った．小児では小学生から増加するが，内容は成人と大きな違いはみられなかった．疾患の少ないところ例えば，花粉症の少ない鹿児島や秋田では受療率は低値であった．一方，一般診療所よりも大学病院やアレルギー専門病院を受診する患者で，高い代替医療の受療率が見られた．性差については明らかではなかった．

患者による代替医療の効果に対する評価を見ると，効果がいくらかでもありと評価しているのは多くは30%以下でアトピー性皮膚炎や小児アレルギー性鼻炎では40%を越

えていた．しかし，薬剤開発時のinactive placeboを対照とした二重盲検試験でのプラセボの有効率が30％前後と高いことを考えると，代替医療の効果の多くはプラセボ効果と考えられた．副作用は，症状が明らかに確認されやすい温泉入浴療法で散見されたが，全体として，確認されたのは少ないものであった．費用は半数以上は1万円以上，10万円以上は20％程度であったが，アトピー性皮膚炎では高い費用をかけていた．情報の入手先は，家族，知人が多く，インターネットは意外に少なかった．しかし，多くの代替医療の受療患者は医師に相談しておらず，医師も多くが否定はしていないのが現状であった．

他方，一般検診の結果から，医療機関で治療を受けていない患者で代替医療受療の割合が高いと考えられた．代替医療の受療の理由は，副作用が少なく，安全，医師受診が面倒といった理由が多くを占めた．市民公開講座やインターネットによる疾患の調査に参加する患者は非常に代替医療の受療率が高く，一般医療機関を受診する患者とは乖離があると考えられ，最近インターネットを利用したさまざまな調査が行われているが，その結果の解釈には十分な注意が必要と考えられた．

II．有効性の評価

代替医療の有効性はアンケート調査からみても患者の評価は低く，標準治療の一般的に示されている有効率には及ばない．代替医療の臨床での有効性に関する科学的評価も多くは行われていない．しかし，甜茶や乳酸菌などの食品についてはさまざまな生物活性を有することが*in vitro*の検討や動物実験で報告されている．食品として安全性が高く，価格も比較的安価なこれらの食品の効果が期待され，臨床試験の報告も見られるが，臨床試験に不可欠なconcealmentの保証，試験のメーカーからの独立性などの課題も指摘されている．有症者の症状改善効果は標準治療には及ばないが，食品としての安全性と安価であるといったことから，早期治療介入の一手段としては期待されている．また，アロマ療法はアレルギー疾患治療を直接の目的として行われているとは限らず，精神的ストレスの改善，精神安定を目的に使用され，その結果アレルギー疾患症状の改善も副次的に期待されて用いられていることも少なくない．代替医療の有効性に対して評価，検討することは意義があると考えられる．

Effects of daily intake of *Lactobacillus paracasei* strain KW3110 on Japanese cedar pollinosis

Syuji Yonekura, M.D., Yoshitaka Okamoto, M.D., Ph.D., Toru Okawa, M.D., Ph.D.,
Minako Hisamitsu, M.D., Hideaki Chazono, M.D., Kouichi Kobayashi, M.D.,
Daiju Sakurai, M.D., Ph.D., Shigetoshi Horiguchi, M.D., Ph.D., and Toyoyuki Hanazawa, M.D., Ph.D.

ABSTRACT

Japanese cedar pollinosis is an important contributor to allergic rhinitis in Japan. Lactobacillus *may be useful as an immunomodulator and is used widely as a foodstuff. The purpose of the study was to examine the effects of daily intake of the* Lactobacillus paracasei *strain KW3110 in patients with cedar pollinosis. The effects of daily intake of KW3110 in patients with cedar pollinosis were investigated in 126 patients who received KW3110 or a placebo in a double-blind study. The study began 1 month before the start of the pollen season and lasted for 3 months. A significant reduction of nasal symptoms and the serum level of eosinophil cationic protein and improvement of quality of life scores occurred in the patients who received KW3110 when pollen scattering was low. However, the effects were limited during the peak period of pollen scattering. Intake of KW3110 may reduce allergic inflammation, but the effect is limited.*

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Key words: Allergic rhinitis, cedar, cypress, double-blind study, foodstuff, immunomodulator, *Lactobacillus paracasei* strain KW3110, pollinosis, preventative effect, probiotics, treatment

In recent years, many countries have experienced an increase in the prevalence of allergic rhinitis.[1,2] In Japan, cedar and cypress pollens constitute major allergens that may spread over a distance of ≥100 km and can cause severe pollinosis.[3,4] Japanese cedar and cypress pollens share a common antigen and >70% of patients with cedar pollinosis also develop an allergy to cypress pollens.[5,6] The pollen season lasts for >12 weeks in and around Tokyo.

Patients with Japanese cedar pollinosis are managed symptomatically, largely with antihistamines and nasal steroids.[7] These drugs are generally safe and reduce symptoms but do not treat the underlying disease and have a potential of associated adverse events, particularly when taken over a long period. Furthermore, the medication costs for cedar pollinosis in Japan alone exceed $2.5 billion annually.[8] Allergen-specific immunotherapy is the only current treatment that has the potential to cure allergic rhinitis. It has a long-term effect and can change the course of nasal symptoms in allergic rhinitis, including those associated with Japanese cedar pollinosis. However, such therapeutic approaches are inconvenient for patients because of the requirement of frequent visits to the doctor, and the treatments carry a risk, albeit minimal, of anaphylactic shock.[9,10]

Probiotics are bacteria of normal mucosal microflora that may be effective as immunomodulators in treatment of allergic and autoimmune diseases.[11–20] Because probiotics are used widely as foodstuffs, their use is attractive from a safety perspective. Administration of probiotics to mothers and postnatally to infants at high risk of atopic diseases reduces the risk of development of chronic recurring atopic eczema, which is the main sign of atopic disease in the 1st year of life.[11] Xiao *et al.* examined the effects of intake of the probiotic strain *Bifidobacterium longum* for 13 weeks in treatment of Japanese cedar pollinosis and reported a marked improvement in nasal symptoms and modulation of Th2-skewed immune responses.[19] However, Helin *et al.* did not observe any beneficial effects on birch pollinosis of intake of *Lactobacillus rhamnosus* for 22 weeks.[20] The different findings may be associated with differences in probiotics and study protocols, including doses, period of administratio, and the number of enrolled patients. *Lactobacillus* is a representative probiotic that has many strains with different characteristics. *In vitro, Lactobacillus* impedes IL-4 production and enhances IL-12 production in murine splenocytes.[21] However, the role of the KW3110 strain of *Lactobacillus* in the human immune response is not understood. Here, we describe a placebo-controlled trial that was designed to examine the effects of daily intake of KW3110 in patients with cedar pollinosis.

From the Department of Otolaryngology, Head and Neck Surgery, Graduate School of Medicine, Chiba University, Chuo-ku, Chiba, Japan
Supported by a grant from the Ministry of Health, Labor and Welfare
The study protocol was approved by the Ethics Committee of Chiba University and written informed consent was obtained from each patient before participation in this study
Address correspondence and reprint requests to Yoshitaka Okamoto, M.D., Ph.D., Department of Otolaryngology, Head and Neck Surgery, Graduate School of Medicine, Chiba University, 1-8-1 Inohana, Chuo-ku, Chiba, 260-8670 Japan
E-mail address: yokamoto@faculty.chiba-u.jp

METHODS

Patient Population and Study Design

A double-blind placebo-controlled trial (intergroup method) was conducted in two groups of subjects. The study population consisted of 138 Japanese men and women aged from 20 to 50 years old who had exhibited symptoms of pollen allergy, such as sneezing, runny nose, nasal congestion, and eye irritation, during the cedar pollen dispersal season for ≥2 years. All of the subjects also had a rating of class 2 or higher for cedar pollen–specific IgE (CAP-radioallergosorbent test: CAP-RAST; SRL, Tokyo, Japan) in blood tests performed at the time of screening. Therefore, the study population consisted of patients with a clinical history of moderate or severe cedar and cypress pollinosis for at least two consecutive cedar and cypress pollen seasons. There was no significant difference in severity between the two groups in previous pollen seasons. The exclusion criteria at the time of selection of subjects were as follows: patients who were taking any medication or had undergone treatment that could affect the test results (use of steroids or immunosuppressants for the last 6 months or treatment such as antigen-specific immunotherapy); patients with apparent adverse results in assessments of other symptoms, including perennial allergic rhinitis, as well as those with suspected allergies to dairy products; and pregnant or breast-feeding patients.

A controller who was not directly involved in the study was responsible for group allocation. The subjects were divided randomly into two groups: active and placebo. There was no significant difference in the degree of symptoms of pollen allergies or preintake cedar-specific IgE levels between these groups ($p > 0.2$ by Mann-Whitney test). A group allocation number was given to each subject. To prevent leakage of information, this number was closely managed by the controller and a member of the ethical committee who was not directly involved in the study until accessed with a key after completion of the study.

Test food (KW *Lactobacillus* powder/1 g) and placebo food (1 g) were used in the study, with each type packed so as to be visually indistinguishable. A total of 1 g of test food contained 1×10^{12}–3×10^{12} *Lactobacillus* and 900 mg of dextrin, whereas the placebo food contained 1 g of dextrin. The daily intake of test food was one package (1 g) per day. *Lactobacillus*-containing foods and any other supplements with a possible effect on the results were excluded from the diet of the subjects during the study period. The study was conducted between December 9, 2004 and May 12, 2005. The food intake period was set at 12 weeks, beginning on January 18, 2005. The subjects were examined five times: before intake: after 4, 8, and 12 weeks of intake; and after a 4-week observation period starting from completion of intake.

Pollen Counts

The cedar pollen/cypress pollen count was measured using the Durham method on the roof of the School of Medicine of Chiba University.

Test Items

Subjective Symptoms. With reference to an allergy diary, scores from 0 to 4 were assigned for sneezing, runny nose, nasal congestion, eye irritation, watery eyes, pollen-induced headache, and degree of interference with daily life.[22] The survey data were converted into nasal and ocular symptom medication scores to determine severity. The nasal symptoms were evaluated on a scale from 0 to 4 in accordance with the practical guidelines for treatment of allergic rhinitis in Japan[22] as follows: 0, no sensation; 1, mild; 2, moderate; 3, severe; and 4, extremely severe. Daily episodes of sneezing and nose blowing were rated from 0 to 4 as follows: 0, none; 1, 1–5 episodes; 2, 6–10 episodes; 3, 11–20 episodes; and 4, >20 episodes. The medication was also recorded according to drug characteristics and duration of usage, according to the guidelines[22] as follows: 1, antihistamines, mast cell stabilizers, and vasoconstrictors; 2, topical ocular or nasal steroids. At the time of each examination, an evaluation was also conducted using the Japan Rhinoconjunctivitis Quality-of-Life (QOL) Questionnaire (No. 1).[23]

Intranasal Findings. The assessment system for intranasal findings[22] was used. Scores from 0 (normal) to 3 (severe) were assigned for swelling of the inferior nasal concha membrane, color tone of the inferior nasal concha membrane, amount of intranasal discharge, and nasal discharge. To avoid clinician bias, the same clinician evaluated the patients throughout the study period.

Immunologic Markers. Results from a general peripheral blood test, the levels of nonspecific IgE antibodies (RAST test; SRL) and specific Japanese cedar–specific IgE (CAP-RAST; SRL.), the Th1/Th2 ratio (Th1%, Th2%), the level of blood eosinophil cationic protein (ECP), and the peripheral blood eosinophil count were determined in blood samples taken at each examination. ECP was measured using the UniCap ECP kit (Pharmacia Diagnostics, Uppsala, Sweden) and Th1/Th2 cytokine profiles were determined by FACS analysis. Briefly, peripheral blood mononuclear cells (5×10^5) were stimulated with phorbol 12-myristate 13-acetate and ionomycin for 4 hours in the presence of 2 μM of monensin, which inhibits the secretion of proteins produced *de novo*. The cells were then stained with anti-CD4 antibody for 15 minutes on ice. After washing with phosphate-buffered saline, the cells were fixed with 4% paraformaldehyde for 10 minutes at

room temperature and permeabilized with 0.5% Triton X-100 for 10 minutes on ice. After blocking with 3% bovine serum albumin for 10 minutes, the cells were incubated on ice for 30 minutes with anti–interferon (IFN) γ labeled with fluorescein isothiocyanate and anti–IL-4 labeled with phycoerythrin. A flow cytometric analysis was performed on a FACS Calibur (Becton-Dickinson, Irvine, CA) using antibodies purchased from BD Bioscience (San Diego, CA).

The subjects were permitted only liquid intake for 5 hours before blood sampling and were requested to visit the hospital at about the same time of day for each examination. All serum samples were stored frozen and assayed after completion of the study using reagents with the same lot number.

Adverse Events

A subjective and objective survey of symptoms was conducted by physicians through interviews of the subjects to assess irritability, decreased motivation, decreased appetite, fatigue, insomnia, headache, tinnitus, vertigo, itching (eczema), vomiting, diarrhea, loose stools, bloated sensation, constipation, abdominal pain, changes in physical condition, history of present illness, and other subjective symptoms during each period of the study.

Data Analysis

After completion of the study, the clinical and laboratory data were analyzed by a person who was not involved in administration of the study. After completion of analysis, the allocation identification numbers for the active and placebo groups were accessed with a key. Statistical analysis was conducted using Dr. SPSS II software (SPSS, Inc., Chicago, IL), with a significance level of ≤5% based on a two-sided test. Subjective symptoms and intranasal findings were analyzed by Mann-Whitney tests (implemented as an intergroup trial) before intake; after 4, 8, and 12 weeks of intake; and after a 4-week observation period following completion of intake. A Wilcoxon signed-rank test (multiple comparisons after Bonferroni correction) was performed for the intergroup trial before intake. For data from blood tests, an unpaired *t*-test was used for comparison before intake; after 4, 8, and 12 weeks of intake; and after the 4-week observation period. A Dunnett test was performed for the intergroup trial before intake. Nonspecific IgE and Japanese cedar–specific IgE were also analyzed by Mann-Whitney tests (implemented as an intergroup trial) before intake; after 4, 8, and 12 weeks of intake; and after the 4-week observation period.

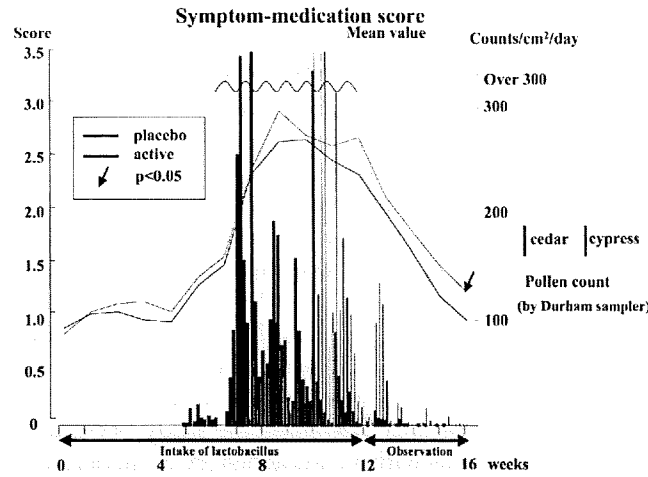

Figure 1. Symptom–medication score. The daily combined Japanese cedar cypress pollen counts in 2005 in Chiba obtained using the Durham pollen sampler and the symptom–medication score of patients during the pollen season are shown.

RESULTS

Background Data

The 138 subjects were divided into the active group (n = 69) and the placebo group (n = 69). Subsequently, 11 patients dropped out from each group: 12 patients (5 active and 7 placebo) refused to participate in the trial for personal reasons and 10 patients (5 active and 5 placebo) were excluded because of oral steroid use during the test period. This left 58 patients in each group who were included in the analysis. The average ages of these subjects was 36.6 ± 8.0 years old (men, n = 19 years, 36.5 ± 7.9 years; women, n = 39 years, 36.6 ± 8.2 years) in the placebo group and 39.5 ± 6.4 years old (men, n = 18 years, 42.4 ± 3.8 years; women, n = 40 years, 38.2 ± 6.9 years) in the active group.

Environmental Pollen Counts

The cedar/cypress pollen count (number of pollen grains/cm^2 per day) measured on the roof of the School of Medicine of Chiba University from January to May 2005 is shown in Fig. 1. Pollen dispersal (≥1 pollen grain/cm^2) began on February 20, 4 weeks after the start of test food intake, and continued until the beginning of May. The peak of cedar pollen dispersal was recorded in mid-March and cypress pollen dispersal reached its peak in early April. In 2005, when the test was conducted, the highest cedar/cypress pollen counts of the preceding 10 years were recorded. The Durham sampler measures the pollen count using a gravimetric method that differs from the method used by the Burkard sampler (a volumetric method that is widely used in Europe). Direct comparison of the counts obtained with these two methods is difficult, because the relationship between the methods depends on the local meteorological conditions and pollen types. A comparison of analyses of the 2005 cedar

Table 1 **Nasal symptoms**

	After 1 wk		After 2 wk		After 3 wk		After 4 wk	
Sneezing								
Placebo	0.46 ± 0.46	$p = 0.321$	0.52 ± 0.49	$p = 0.398$	0.59 ± 0.53	$p = 0.244$	0.62 ± 0.62	$p = 0.907$
Active	0.55 ± 0.50		0.65 ± 0.66		0.76 ± 0.66		0.62 ± 0.68	
Runny nose								
Placebo	0.67 ± 0.74	$p = 0.641$	0.79 ± 0.83	$p = 0.631$	0.92 ± 0.92	$p = 0.799$	0.91 ± 1.02	$p = 0.727$
Active	0.67 ± 0.62		0.82 ± 0.76		0.81 ± 0.73		0.75 ± 0.80	
Stuffy nose								
Placebo	0.34 ± 0.52	$p = 0.790$	0.49 ± 0.70	$p = 0.726$	0.50 ± 0.67	$p = 0.212$	0.52 ± 0.74	$p = 0.262$
Active	0.34 ± 0.49		0.47 ± 0.67		0.41 ± 0.67		0.40 ± 0.65	
	After 5 wk		**After 6 wk**		**After 7 wk**		**After 8 wk**	
Sneezing								
Placebo	0.59 ± 0.53	$p = 0.937$	0.87 ± 0.70	$p = 0.336$	1.16 ± 0.72	$p = 0.949$	1.98 ± 1.10	$p = 0.410$
Active	0.62 ± 0.62		1.02 ± 0.78		1.25 ± 0.91		2.16 ± 1.03	
Runny nose								
Placebo	0.81 ± 0.84	$p = 0.942$	1.06 ± 0.97	$p = 0.940$	1.28 ± 0.97	$p = 0.774$	2.17 ± 1.22	$p = 0.987$
Active	0.72 ± 0.67		1.00 ± 0.81		1.22 ± 0.89		2.18 ± 1.14	
Stuffy nose								
Placebo	0.48 ± 0.65	$p = 0.501$	0.60 ± 0.75	$p = 0.476$	0.78 ± 0.88	$p = 0.971$	1.54 ± 1.20	$p = 0.840$
Active	0.37 ± 0.51		0.54 ± 0.70		0.77 ± 0.80		1.54 ± 1.20	
	After 9 wk		**After 10 wk**		**After 11 wk**		**After 12 wk**	
Sneezing								
Placebo	2.34 ± 1.26	$p = 0.481$	2.15 ± 1.21	$p = 0.451$	2.01 ± 1.13	$p = 0.943$	2.09 ± 1.18	$p = 0.684$
Active	2.41 ± 1.24		2.35 ± 1.35		2.07 ± 1.19		1.99 ± 1.16	
Runny nose								
Placebo	2.79 ± 1.31	$p = 0.177$	2.69 ± 1.32	$p = 0.415$	2.59 ± 1.35	$p = 0.055$	2.60 ± 1.32	$p = 0.039$
Active	2.53 ± 1.44		2.61 ± 1.51		2.27 ± 1.32		2.13 ± 1.31	
Stuffy nose								
Placebo	2.12 ± 1.39	$p = 0.730$	2.24 ± 1.40	$p = 0.495$	2.10 ± 1.39	$p = 0.260$	2.12 ± 1.37	$p = 0.075$
Active	2.05 ± 1.46		2.15 ± 1.59		1.86 ± 1.36		1.67 ± 1.26	
	After 1-wk Observation		**After 2-wk Observation**		**After 3-wk Observation**		**After 4-wk Observation**	
Sneezing								
Placebo	1.61 ± 1.17	$p = 0.962$	1.31 ± 1.02	$p = 0.848$	1.09 ± 0.80	$p = 0.174$	0.85 ± 0.64	$p = 0.114$
Active	1.56 ± 0.97		1.23 ± 0.87		0.89 ± 0.76		0.70 ± 0.73	
Runny nose								
Placebo	2.01 ± 1.25	$p = 0.189$	1.67 ± 1.12	$p = 0.200$	1.31 ± 1.04	$p = 0.074$	1.02 ± 0.87	$p = 0.118$
Active	1.72 ± 1.18		1.40 ± 1.11		0.97 ± 0.90		0.77 ± 0.79	
Stuffy nose								
Placebo	1.47 ± 1.16	$p = 0.366$	1.10 ± 1.03	$p = 0.294$	0.71 ± 0.73	$p = 0.298$	0.49 ± 0.55	$p = 0.241$
Active	1.23 ± 1.04		0.86 ± 0.83		0.55 ± 0.60		0.40 ± 0.54	

The score was expressed as mean ± SD of the each week.

pollen count using the two methods indicated that the counts with the Burkard sampler were ~12 times higher than those obtained with the Durham sampler.[24]

Subjective Symptoms

Allergy Diary. After the start of pollen dispersal, a significant increase in nasal and ocular symptoms occurred in both the active and the placebo groups. Throughout the intake period, the symptom medication score for the active group tended to be lower than that for the placebo group, and the active group also had a significantly lower score during the observation period (Fig. 1). There was no significant difference in the medication scores between the groups throughout the study period

Table 2 **Quality-of-life score**

	Before Intake		After 4 wk		After 8 wk		After 12 wk		After 4-wk Observation	
Runny nose										
Placebo	0.59 ± 0.59	$p = 0.691$	0.83 ± 0.82	$p = 0.406$	2.00 ± 0.94	$p = 0.233$	2.07 ± 1.02	$p = 0.106$	0.86 ± 0.71	$p = 0.590$
Active	0.60 ± 0.79		0.66 ± 0.55		1.81 ± 0.89		1.78 ± 0.92		0.81 ± 0.76	
Sneezing										
Placebo	0.57 ± 0.53	$p = 0.688$	0.90 ± 0.69	$p = 0.647$	1.79 ± 0.87	$p = 0.216$	1.71 ± 0.94	$p = 0.544$	0.90 ± 0.64	$p = 0.856$
Active	0.66 ± 0.69		0.83 ± 0.65		1.95 ± 0.87		1.79 ± 0.93		0.90 ± 0.64	
Stuffy nose										
Placebo	0.48 ± 0.63	$p = 0.878$	0.83 ± 0.92	$p = 0.144$	1.59 ± 1.06	$p = 0.254$	2.02 ± 1.15	$p = 0.078$	0.69 ± 0.65	$p = 0.261$
Active	0.45 ± 0.57		0.55 ± 0.63		1.38 ± 1.12		1.62 ± 1.02		0.53 ± 0.50	
Itchy eyes										
Placebo	0.34 ± 0.58	$p = 0.033$	0.47 ± 0.54	$p = 0.095$	2.02 ± 1.07	$p = 0.602$	2.12 ± 1.08	$p = 0.488$	0.57 ± 0.73	$p = 0.816$
Active	0.59 ± 0.70		0.66 ± 0.61		2.14 ± 0.98		1.97 ± 1.01		0.55 ± 0.60	
Watery eyes										
Placebo	0.19 ± 0.44	$p = 0.560$	0.26 ± 0.44	$p = 0.212$	1.10 ± 1.10	$p = 0.097$	1.22 ± 1.01	$p = 0.650$	0.22 ± 0.46	$p = 0.295$
Active	0.28 ± 0.59		0.38 ± 0.52		1.31 ± 0.84		1.12 ± 0.92		0.31 ± 0.50	
Nose symptoms										
Placebo	0.49 ± 0.38	$p = 0.884$	0.75 ± 0.60	$p = 0.238$	1.74 ± 0.81	$p = 0.380$	1.78 ± 0.83	$p = 0.196$	0.77 ± 0.56	$p = 0.457$
Active	0.52 ± 0.47		0.59 ± 0.41		1.61 ± 0.74		1.59 ± 0.81		0.68 ± 0.48	
Eye symptoms										
Placebo	0.27 ± 0.43	$p = 0.096$	0.36 ± 0.43	$p = 0.109$	1.56 ± 0.99	$p = 0.226$	1.67 ± 0.93	$p = 0.592$	0.40 ± 0.51	$p = 0.665$
Active	0.43 ± 0.58		0.52 ± 0.51		1.72 ± 0.78		1.54 ± 0.83		0.43 ± 0.51	
Nasal-ocular symptoms										
Placebo	0.42 ± 0.32	$p = 0.823$	0.62 ± 0.47	$p = 0.664$	1.68 ± 0.79	$p = 0.859$	1.74 ± 0.79	$p = 0.241$	0.65 ± 0.50	$p = 0.703$
Active	0.42 ± 0.48		0.57 ± 0.40		1.65 ± 0.68		1.57 ± 0.76		0.60 ± 0.46	
Usual daily activities										
Placebo	0.71 ± 1.30	$p = 0.693$	1.84 ± 3.75	$p = 0.187$	4.76 ± 3.92	$p = 0.777$	5.60 ± 4.64	$p = 0.108$	1.41 ± 2.14	$p = 0.660$
Active	0.74 ± 1.36		0.90 ± 1.66		4.50 ± 3.84		4.21 ± 3.92		1.28 ± 2.08	
Outdoor activities										
Placebo	0.16 ± 0.45	$p = 0.321$	0.27 ± 0.68	$p = 0.329$	0.84 ± 0.92	$p = 0.465$	1.40 ± 1.11	$p = 0.043$	0.25 ± 0.47	$p = 0.958$
Active	0.08 ± 0.41		0.16 ± 0.45		0.98 ± 1.01		1.03 ± 1.09		0.29 ± 0.58	
Social functioning										
Placebo	0.21 ± 0.49	$p = 0.357$	0.69 ± 1.91	$p = 0.242$	1.69 ± 2.19	$p = 0.855$	2.33 ± 2.24	$p = 0.115$	0.36 ± 0.91	$p = 0.969$
Active	0.21 ± 0.67		0.21 ± 0.69		1.60 ± 2.12		1.79 ± 2.33		0.38 ± 1.15	
Sleep problems										
Placebo	0.09 ± 0.28	$p = 0.246$	0.28 ± 0.64	$p = 0.638$	0.64 ± 1.04	$p = 0.639$	0.76 ± 1.00	$p = 0.457$	0.21 ± 0.49	$p = 0.811$
Active	0.17 ± 0.42		0.21 ± 0.49		0.50 ± 0.80		0.64 ± 0.95		0.24 ± 0.57	
General health problems										
Placebo	0.52 ± 0.96	$p = 0.838$	0.79 ± 1.46	$p = 0.799$	1.64 ± 1.77	$p = 0.595$	1.90 ± 1.85	$p = 0.677$	0.64 ± 0.91	$p = 0.806$
Active	0.50 ± 1.00		0.66 ± 1.12		1.71 ± 1.62		1.76 ± 1.83		0.74 ± 1.16	
Emotional problems										
Placebo	0.62 ± 1.24	$p = 0.772$	1.22 ± 2.68	$p = 0.772$	3.17 ± 2.99	$p = 0.862$	3.84 ± 3.57	$p = 0.278$	0.93 ± 1.45	$p = 0.770$
Active	0.57 ± 1.37		0.67 ± 1.23		3.26 ± 3.03		3.09 ± 3.17		1.07 ± 1.85	
Total										
Placebo	2.36 ± 4.02	$p = 0.933$	5.34 ± 10.91	$p = 0.362$	13.66 ± 11.83	$p = 0.919$	16.95 ± 13.66	$p = 0.156$	4.05 ± 5.57	$p = 0.810$
Active	2.40 ± 4.46		2.88 ± 4.68		13.24 ± 11.44		13.33 ± 12.67		4.21 ± 6.85	
Face scale										
Placebo	1.45 ± 0.84	$p = 0.628$	1.47 ± 0.90	$p = 0.614$	2.38 ± 0.75	$p = 0.621$	2.36 ± 0.83	$p = 0.302$	1.14 ± 0.71	$p = 0.416$
Active	1.47 ± 0.80		1.40 ± 0.90		2.47 ± 0.80		2.24 ± 0.78		1.26 ± 0.83	

P value: between intergroups. The score was expressed as mean ± SD of each week.

(data not shown). Each symptom score for the active group tended to be lower than the respective score for the placebo group, but the only significant difference in symptom scores occurred for nasal discharge at 12 weeks of intake ($p = 0.039$; Table 1).

Quality of Life. The Japan Rhinoconjunctivitis QOL Questionnaire survey (Table 2) indicated that nasal symptoms significantly increased in both groups after pollen dispersal and an intergroup comparison showed no significant difference between the groups. Regard-

Table 3 **Intranasal findings**

	Before Intake		After 4 wk		After 8 wk		After 12 wk		After 4-wk Observation	
Swelling*										
Placebo	0.22 ± 0.46	$p = 0.820$	0.41 ± 0.59	$p = 0.377$	1.52 ± 0.84	$p = 0.689$	1.43 ± 0.73	$p = 0.314$	0.50 ± 0.71	p=0.832
Active	0.21 ± 0.43		0.31 ± 0.50		1.55 ± 0.71		1.28 ± 0.81		0.45 ± 0.63	
Color tone#										
Placebo	0.21 ± 0.45	$p = 0.961$	0.55 ± 0.68	$p = 0.104$	1.28 ± 0.81	$p = 0.625$	1.36 ± 0.77	$p = 0.188$	0.45 ± 0.71	p=0.775
Active	0.24 ± 0.57		0.38 ± 0.62		1.17 ± 0.70		1.19 ± 0.69		0.45 ± 0.63	
Amount of discharge§										
Placebo	0.24 ± 0.47	$p = 0.355$	0.52 ± 0.66	$p = 0.013$	1.05 ± 0.78	$p = 0.427$	1.07 ± 0.77	$p = 0.037$	0.59 ± 0.70	p=0.234
Active	0.17 ± 0.42		0.26 ± 0.52		0.91 ± 0.73		0.78 ± 0.80		0.47 ± 0.73	
Discharge property¶										
Placebo	0.67 ± 1.26	$p = 0.250$	1.24 ± 1.43	$p = 0.009$	2.26 ± 1.25	$p = 0.241$	2.21 ± 1.24	$p = 0.011$	1.31 ± 1.44	p=0.134
Active	0.40 ± 0.95		0.60 ± 1.15		1.98 ± 1.37		1.57 ± 1.42		0.91 ± 1.32	

P value for intergroup comparison. The score was expressed as mean ± SD of each week.
**Swelling of the inferior nasal concha membrane.*
#Color tone of the inferior nasal concha membrane.
§Amount of intranasal discharge.
¶Nasal discharge property.

ing activities of daily life, scores for outdoor activities, social life, sleep, and physical/mental well-being, and the total score for these categories showed significant deterioration after pollen dispersal in both groups. After 12 weeks of intake, an intergroup comparison indicated a significant difference in the degree of interference of pollinosis with outdoor activities ($p = 0.043$).

Intranasal Findings

Swelling of the inferior concha membrane, color tone of the inferior concha membrane, intranasal discharge, and nasal discharge increased in both groups after the start of pollen dispersal (Table 3). Intranasal discharge was significantly suppressed in the active group after 4 ($p = 0.013$) and 12 weeks ($p = 0.037$) of intake and nasal discharge was lower in the active group after 4 ($p = 0.009$) and 12 weeks ($p = 0.011$) compared with the placebo group.

Immunologic Markers

All items in the general blood test varied within their respective standard ranges. Nonspecific IgE antibodies, cedar-specific IgE antibodies, and eosinophil count significantly increased in both groups after pollen dispersal, but there were no significant differences in intergroup comparisons. The ECP level was significantly lower in the active group after 4 and 12 weeks (Table 4) compared with the placebo group, and the levels for the active group remained within the standard range (below 14.7) throughout the intake period, which suggests that higher pollen exposure abrogates the *Lactobacillus*-driven reduction of ECP levels.

The Th1/Th2 (CD4 ratio) was significantly higher in both groups after 4 weeks of intake, compared with the respective preintake values. There was no significant difference in this ratio between the active and placebo groups, but the ratio showed a tendency to be higher in the active group throughout the intake period. In Th1 ($IFN\text{-}\gamma^+/IL\text{-}4^-$) cells, the IFN-γ level was significantly higher in the placebo group after 12 weeks of intake and after the 4-week observation period, compared with the preintake level. In the active group, the IFN-γ level after 12 weeks of intake was also significantly higher than the preintake level, suggesting a Th1 shift, but IFN-γ in the active and placebo groups did not show a significant difference at any time point. In Th2 ($IFN\text{-}\gamma^-/IL\text{-}4^+$) cells, the IL-4 level was significantly lower in the active group after the 4-week observation period, compared with the preintake value, also suggesting a Th1 shift, but IL-4 levels did not differ between the active and placebo groups (Table 4).

Adverse Events

Abdominal symptoms including loose stools and diarrhea were observed in 15% and 10% of patients in the active and placebo groups, respectively. However, none of the symptoms were severe, none needed treatment, and there were no significant differences in adverse events between the two groups (chi-square test, $p > 0.4$). The correlation of these events with *Lactobacillus* intake was unclear. Based on the symptoms diary, there were no significant differences in decreased motivation, fatigue, diarrhea, and loose stools between the groups.

DISCUSSION

To examine the efficacy of the *L. paracasei* KW3110 strain on cedar pollen allergy, a double-blind trial was

Table 4 **Immunologic markers**

	Before Intake		After 4 wk			After 8 wk		
	Score (mean ± SD)	Intergroup *p* Value	Score (mean ± SD)	*p* Value#	Intergroup *p* Value	Score (mean ± SD)	*p* Value#	Intergroup *p* Value
ECP (μg/L)								
Placebo	9.3 ± 5.5	$p = 0.221$	9.3 ± 6.5	$p = 1.000$	$p = 0.017$	13.5 ± 8.4	$p < 0.001$	$p = 0.144$
Active	7.9 ± 6.0		7.1 ± 4.5	$p = 1.000$		11.2 ± 6.0	$p < 0.001$	
Th1/Th2 (CD4) (%)								
Placebo	15.0 ± 7.4	$p = 0.532$	16.1 ± 8.5	$p = 0.579$	$p = 0.344$	14.3 ± 7.9	$p = 0.842$	$p = 0.514$
Active	16.1 ± 11.1		18.4 ± 14.0	$p = 0.579$		15.1 ± 10.2	$p = 0.921$	
Th1/IFN-γ^+/IL-4$^-$(%)								
Placebo	23.8 ± 6.7	$p = 0.553$	24.3 ± 6.9	$p = 0.718$	$p = 0.729$	25.0 ± 7.6	$p = 0.081$	$p = 0.886$
Active	24.6 ± 8.0		24.8 ± 7.8	$p = 0.718$		25.2 ± 8.2	$p = 0.669$	
Th2/IFN-γ^-/IL-4$^+$ (%)								
Placebo	1.88 ± 0.88	$p = 0.872$	1.82 ± 0.81	$p = 0.887$	$p = 0.842$	2.10 ± 0.99	$p = 0.058$	$p = 0.986$
Active	1.91 ± 0.97		1.84 ± 1.23	$p = 0.887$		2.12 ± 1.12	$p = 0.058$	
Cedar pollen IgE (UA/mL)								
Placebo	16.4 ± 17.9	$p = 0.845$	16.0 ± 16.1	$p = 1.000$	$p = 0.779$	17.1 ± 27.4	$p = 1.000$	$p = 0.729$
Active	15.8 ± 16.9		15.2 ± 15.0	$p = 1.000$		15.6 ± 17.6	$p = 1.000$	

	After 12 Weeks			After 4-week Observation Period		
	Score (mean ± SD)	*p* Value#	Intergroup *p* Value	Score (mean ± SD)	*p* Value#	Intergroup *p* Value
ECP(μg/L)						
Placebo	16.3 ± 10.1	$p < 0.001$	$p = 0.012$	13.4 ± 8.3	$p < 0.001$	$p = 0.063$
Active	12.3 ± 6.5	$p < 0.001$		10.8 ± 6.4	$p < 0.001$	
Th1/Th2(CD4) (%)						
Placebo	17.2 ± 10.4	$p = 0.076$	$p = 0.896$	17.7 ± 9.4	$p = 0.015$	$p = 0.262$
Active	17.5 ± 12.7	$p = 0.793$		21.3 ± 22.0	$p = 0.003$	
Th1/IFN-γ^+/IL-4-$^-$ (%)						
Placebo	25.9 ± 7.6	$p < 0.001$	$p = 0.768$	25.7 ± 8.1	$p = 0.002$	$p = 0.895$
Active	26.3 ± 8.7	$p = 0.014$		25.9 ± 8.9	$p = 0.083$	
Th2/IFN-γ^-/IL-4$^+$ (%)						
Placebo	1.90 ± 0.94	$p = 0.999$	$p = 0.693$	1.75 ± 0.81	$p = 0.418$	$p = 0.651$
Active	1.97 ± 0.99	$p = 0.892$		1.68 ± 0.98	$p = 0.028$	
Cedar pollen IgE (UA/mL)						
Placebo	63.2 ± 67.5	$p < 0.001$	$p = 0.303$	66.9 ± 77.2	$p < 0.001$	$p = 0.303$
Active	82.5 ± 124.6	$p < 0.001$		87.5 ± 130.2	$p < 0.001$	

#p Value compared with the respective preintake level.
ECP = eosinophil cationic protein; IFN = interferon.

conducted using a placebo as a control. No side effects of common toxicity criteria grade 1 or higher were observed in the active or placebo group, although a few subjects exhibited temporary abdominal symptoms. The active group showed a tendency for lower symptom scores based on an allergy diary, had reduced nasal discharge at the peak of cypress pollen dispersal during week 12, and had a significantly lower watery discharge after 12 weeks of intake, compared with the placebo group. In the QOL survey, patients in the active group indicated a significantly lower degree of interference with outdoor activities after 12 weeks of intake, compared with those in the placebo group. This suggests that the manifestation of effects increases after ~12 weeks of intake of the *L. paracasei* KW3110 strain. Symptom scores also tended to be lower in the active group through the 4-week observation period, suggesting that the effects of *Lactobacillus* intake continued after intake completion. Some nasal symptoms were reduced in patients who received KW3110 compared with those who received placebo in the period of low pollen scattering; however, these effects were limited and were not observed during the peak of the pollen season.

The Th1/Th2 cytokine profile in peripheral blood underwent a Th1 shift in both groups, with no significant difference between the groups. We note that the Th1/Th2 cell count in the trial reflects all Th1/Th2 cells, including those that were nonspecific to antigen. In cedar pollen allergy, the number of cedar pollen–specific memory T-cells is ~10–50/100,000 peripheral blood CD4 cells,[25,26] and the effect of *Lactobacillus* on

antigen-specific Th1/Th2 cells, rather than all Th1/Th2 cells, needs to be examined in a future study. The KW3110 strain had no effect on the total IgE and cedar pollen–specific IgE levels in blood after 12 weeks of intake, but patients in the active group had a lower blood ECP level after 12 weeks. This suggests that the KW3110 strain acts on eosinophil function and reduces the eosinophil count, which also indicates the possibility of an effect on antigen-specific T cells.

Suppression of symptoms is insufficient for many pollen allergy patients, and safe and less burdensome treatments for the underlying disease are needed. There are few natural remedies for pollen allergies, and primary and secondary interventions are especially significant for patients who have atopic dispositions and for those who are sensitized to antigens but are nonprogressive.[27,28] In such interventions, antigen avoidance is of importance, but the actual efficacy and significance of antigen avoidance has not been clarified.[29] Continuous administration of antihistamines in patients with atopic dermatitis can suppress asthma,[30–33] but long-term drug administration is unlikely to be sustained in individuals suffering from pollen allergy but without symptoms or manifestations. In this context, *Lactobacillus* probiotics can play an important role, because they can be taken as natural food, rather than as medication, and are attractive from a safety perspective.

Lactobacillus is a representative probiotic and the KW3110 strain is reported to be a potent inducer of IL-12 and repressor of IL-4 among 101 strains tested *in vitro* in ovalbumin-sensitized mice splenocytes. There is growing interest in the antiallergic effect mediated by immunomodulators, although the short-term effects on symptoms are inferior to those of standard drugs such as antihistamines and steroid inhalants. If efficacy occurs after immune response modification, immunomodulators may be useful for primary and secondary interventions. In this study, patients began intake 4 weeks before pollen dispersal, even though most showed no symptoms at this time, and nasal mucosal inflammation still had to develop. In this sense, the study corresponds to a secondary intervention, and although the test period before pollen dispersal was only 4 weeks, the results provide insight into the protective effects of *Lactobacillus* before massive pollen dispersal. Overall, the study suggests a limited clinical effect after administration of the KW3110 strain to patients with cedar pollen allergies for 12 weeks, beginning 4 weeks before pollen dispersal, without immunomodulation during the peak pollen season. Additional studies of the administration route, dose, period of administration, and mechanism of action are required to improve the clinical efficacy.

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REVIEW ARTICLE

Present Situation of Cedar Pollinosis in Japan and its Immune Responses

Yoshitaka Okamoto[1], Shigetoshi Horiguchi[1], Heizaburo Yamamoto[1], Syuji Yonekura[1] and Toyoyuki Hanazawa[1]

ABSTRACT

Recent observations have suggested significant worldwide increase in the prevalence of allergic rhinitis and cedar pollinosis. In Japan, Japanese cedar (*Cryptometria japonica*) and Japanese cypress (*Chamaecyparis obtusa*) pollens are considered to be the major unique allergens and their extent of dispersal is quite large, travelling more than 100 km and thus causing serious pollinosis. Cedar pollinosis is a typical type 1 allergic disease by an adaptive immune response that occurs through the induction of allergen-specific effector T cells from naïve T cells. We examined the number of Japanese cedar pollen specific memory Th cells in the peripheral blood of the patients and found that the cedar pollen specific IL-4-producing Th2 memory cells increased during the pollen season and decreased during the off-season. However, more than 60% of the cedar-specific memory Th2 cells survived up to 8 months after the pollen season. Natural killer T(NKT) cells represent a unique lymphocyte subpopulation and their activity is not restricted to MHC antigens. NKT cells play an important role in innate immunity, however, the participation in development of allergic rhinitis could not be clarified.

KEY WORDS

cedar pollinosis, cedar specific Th memory cell, epidemiology, natural killer T cell

CEDAR POLLEN

In recent years, many countries have experienced an increase in the prevalence of allergic rhinitis.[1,2] Dust mite allergen is responsible for at least 90% of cases of perennial allergic rhinitis, while arboreal pollen, including that of cedar and Japanese cypress, is important in Japan.[3,4] Cedar forest covers nearly 18% of the total land area of Japan, while Japanese cypress is concentrated in the Kanto region and the western part of the country. Both cedar and Japanese cypress produce enormous amounts of pollen. In Japan, pollen counts are typically measured using the gravimetric method with a Durham sampler, in contrast to Western countries in which a Burkard sampler is typically used. In a study in Chiba Prefecture in 2005, the amount of air-borne pollen counted with a Burkard sampler was about 12 times greater than that counted with a Durham sampler.[5] In addition, distinct from grass pollen, which only spreads less than 100 meters, cedar and cypress pollen travel a long distance and reach major cities, including Tokyo and Osaka, causing wide-spread pollinosis, although no actual data describing the distance traveled was available. A detailed simulation study considering the results of real-time pollen distributing information was conducted using large computers and Figure 1 shows the source and areas from which the cedar pollen detected at Chiba University Hospital had spread. These dark spots indicate the areas where the cedar pollen originated. Pollens blow to Chiba city from the cedar planting areas of Boso Peninsula, as well as from the north Kanto area, Nikko, Izu Peninsula and Shizuoka Prefecture. This study suggests that cedar pollen actually can travel more than 100 km and cause pollinosis in a large area.

Cedar pollen dispersal precedes Japanese cypress pollen dispersal, and approximately 70% of patients with cedar pollinosis are also allergic to Japanese cypress pollen because of a common antigen.[6] Dispersal of cedar and Japanese cypress pollen generally exhibits an arch-shaped pattern with time: cedar pollen dispersal starts in early February and reaches a peak between late February and early March, and is fol-

[1]Department of Otolaryngology, Head and Neck Surgery, Graduate School of Medicine, Chiba University, Chiba, Japan.
Correspondence: Yoshitaka Okamoto, MD, PhD, Department of Otolaryngology, Head and Neck Surgery, Graduate School of Medicine, Chiba University, 1-8-1 Inohana, Chuo-ku, Chiba 260-8670, Japan.
Email: yokamoto@faculty.chiba-u.jp
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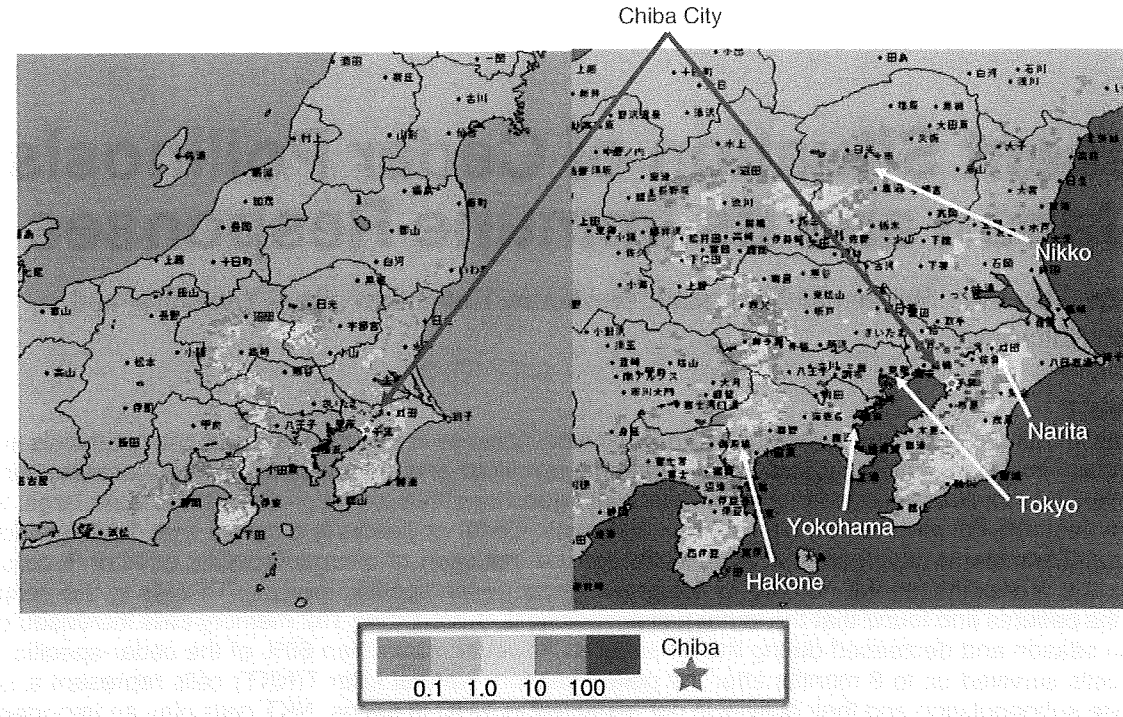

Fig. 1 The source areas from which the cedar pollen detected at Chiba University Hospital spread. This is the computer simulation study done by Mr. Kunihiko Yokota *et al.*, at Weather Service Co.,Ltd..

lowed by dispersal of Japanese cypress pollen, which reaches a peak from late March to early April, with some variation due to changes in the climate each year.[7,8] The pollen dispersal season lasts for more than 10 weeks in and around the Tokyo area.

PREVALENCE OF CEDAR POLLINOSIS IN JAPAN

A survey based only on a questionnaire has the risk of inclusion of a high rate of false-positive cases, because allergic rhinitis is sometimes difficult to distinguish from acute upper respiratory infection and even normal healthy individuals may exhibit mild, nonspecific nasal symptoms, such as sneezing and nasal secretion. In particular, cedar pollen dispersal season is also high flu season. An allergen-specific IgE test is necessary to avoid a high incidence of false positives, but it has been difficult to conduct an epidemiological study in Japan because of laws preventing use of personal information. In 2008, a questionnaire was posed to the Otorhinolaryngologists nationwide to determine whether their families suffered from allergic rhinitis. Although the rate of return of the questionnaire was low, i.e., 40% and the bias of the population could not be ignored, an accurate diagnosis was expected.

According to the analysis of this questionnaire,[9] the prevalence of perennial allergic rhinitis and of cedar pollinosis was 23.4% and 26.5%, respectively. In particular, the prevalence of cedar pollinosis increased more than 10% compared with that observed in a similar questionnaire conducted in 1998. Although the peak of cedar pollinosis is in those in their thirties to forties, the age onset of pollinosis has been decreasing (Fig. 2).

Figure 3 shows the annual amount of cedar pollen dispersal in Japan, which we examined in 2005. The darker brown parts indicate areas where cedar pollen counts were high. We studied the influence of various amounts of pollen exposure on the development of pollinosis and mite allergic rhinitis in elementary school students from schools in rural areas where the movement of students out of or into the school was uncommon. The annual amount of cedar and cypress pollen differed among these five regions. The pollen level was very high in southern Yamanashi: about 7,000/cm^2 on average for the last five years, as determined using Durham pollen samplers. In contrast, the pollen level was low in northern Yamanashi and inland Akita, at about 2,000/cm^2, and very low in coastal Akita, at about 500/cm^2. The pollen level in Chiba was about 4,000/cm^2.

Figure 4 shows the detection rate of cedar- and mite-specific IgE in students in these regions. The positive rate for Japanese cedar was about 60%, except for students in coastal Akita, who had a rate of only 23%. The positive rate for mite IgE was about 50% in each region. These results suggest that the sensitization rate for mite allergen is almost the same nationwide, whereas that for cedar pollen is depend-

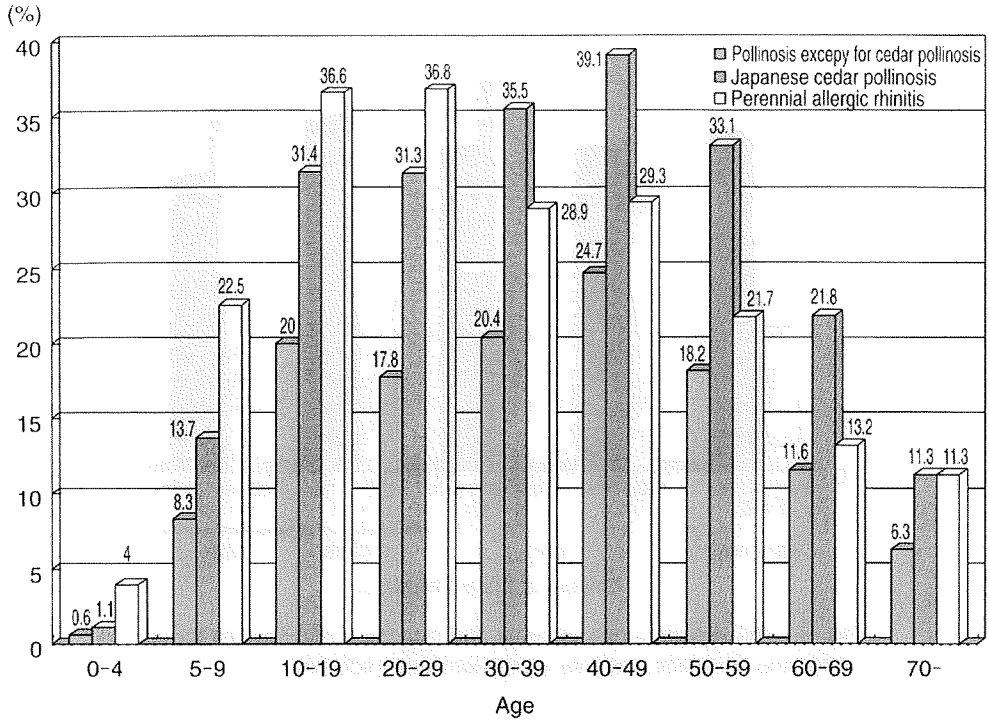

Fig. 2 The prevalence rate of allergic rhinitis in Japan in 2008 (from reference 9).

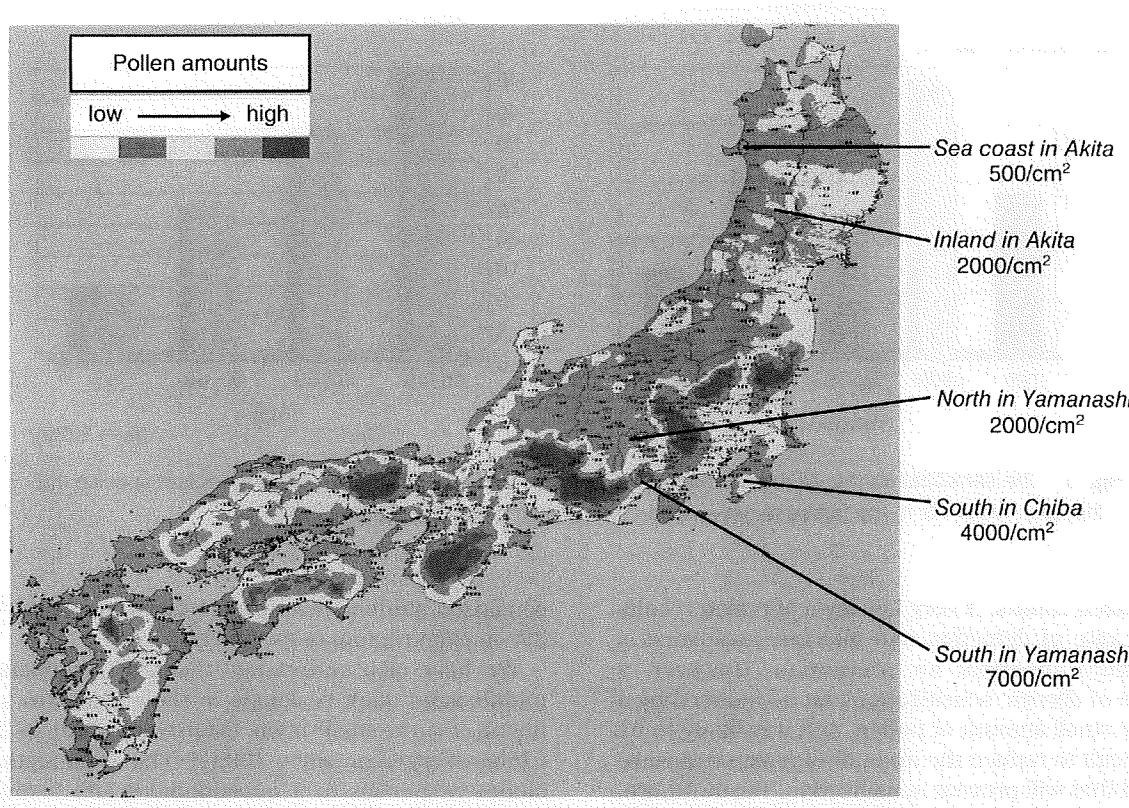

Fig. 3 Annual amount of cedar and cypress pollen dispersal in Japan in 2005.

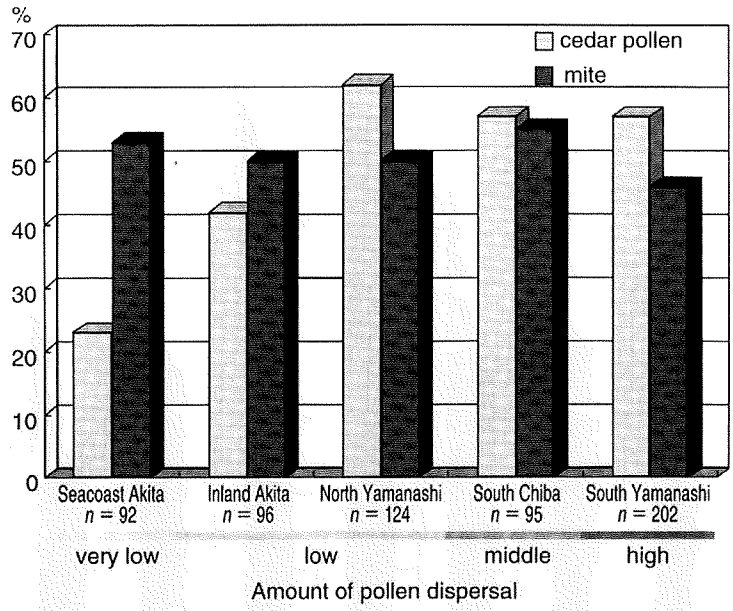

Fig. 4 The detection rate of cedar and cypress pollen-specific IgE in all 4th and 5th grade students in the elementary schools.

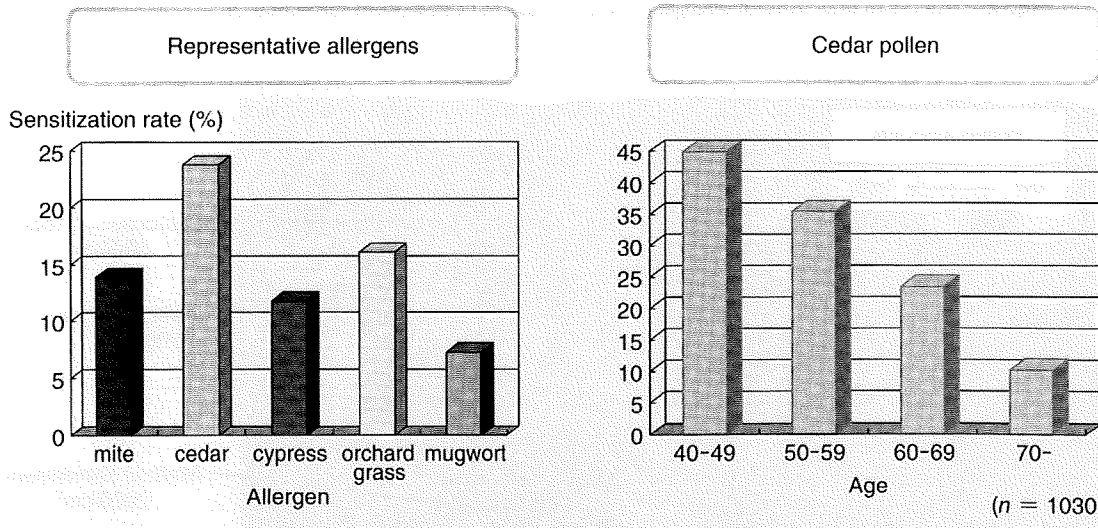

Fig. 5 The sensitization rate to the representative allergen and age distribution of cedar pollen-specific IgE in the adult residents in the forties to seventies in the rural small town in South Chiba.

ent on pollen counts. A very low level of pollen results in a low rate of detection and allergen avoidance is undoubtedly important for prevention. However, a high rate of allergic sensitization can be induced by a relatively small amount of pollen, and it is likely to be very difficult to reduce the amount of pollen exposure to a level that will prevent sensitization. Furthermore, tolerance was not easily induced in students in southern Yamanashi who had been receiving high pollen exposure every year since birth. Interestingly, the incidence of mite allergic rhinitis and pollinosis in these sensitized students was almost the same; about 30 to 35% in each region, respectively.

We have also undertaken medical examination of middle-aged adult residents in their forties to seventies in a rural small town (Maruyama-cho) in South Chiba every year since 1995.[10] The examination includes responses to a questionnaire and testing for specific IgE in serum using a CAP-RAST system. Figure 5 shows the sensitization rate to the representative allergens and the age distribution of cedar pollen-specific IgE. Deterioration of cedar-specific IgE is ob-

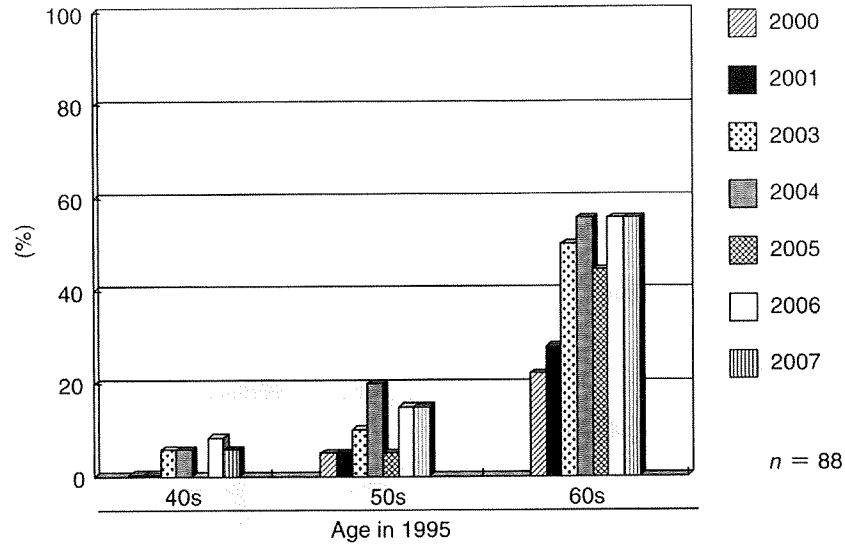

Fig. 6 The rate of change to negative over the last 13 years in cedar pollen-specific IgE in the residents who had tested positive for anti-cedar pollen specific IgE in 1995 and then had received examination every year.

served in elderly subjects. Figure 6 shows the rate of change to negative over the last 13 years in cedar pollen IgE in residents who had tested positive for anti-cedar pollen IgE in 1995. The IgE assays were performed at the end of each cedar pollen season. It appears that the IgE titer is affected by the spread of pollen each year. Interestingly, however, the negative change for 13 years is not commonly observed even in their forties to fifties. The rate of the cedar pollinosis determined by clinical symptoms in combination with positive cedar pollen IgE has also not decreased among these aged subjects.

THE LONG-TERM COURSE OF PATIENTS WITH ALLERGIC RHINITIS

One hundred and seventy-seven patients who were treated in our department from 1970 to 1995 consented to undergo a detailed re-examination. A comparison between the recent symptoms and those observed 10 to 30 years ago showed that 30% of adult patients exhibited some improvements and 10% had resolution. However, only 20% of the pediatric patients exhibited mild improvement of symptoms, whereas the remaining had the same or even worse symptoms as those in childhood (data not shown: in preparation for submitting). Regarding the allergen-specific IgE, a change to negative was not observed in any patients with cedar pollinosis and was seen in only a few of the mite-allergic patients. Thus, natural resolution is not commonly observed in allergic rhinitis and most pediatric patients grow to adulthood without natural improvement of symptoms.

CEDAR POLLEN SPECIFIC MEMORY T CELLS

It has been suggested that dysregulation of cytokine synthesis from Th1 and Th2 cells is fundamental to the pathogenesis of allergic diseases. However, no significant difference was observed between the two groups in the Th1/Th2 cell profile in peripheral blood $CD4^+$ T cells from patients with perennial allergic rhinitis and non-allergic rhinitis by FACS analysis.[11]

Pollinosis is thought to be an adaptive immune response that manifests as a type 1 allergic reaction, and it occurs as a consequence of fundamental allergenic mechanisms involving the induction of pollen-specific T helper type 2 (Th2) effector cells from naïve Th0 cells. Most effector T cells are short-lived, but few effector T cell become long-lived memory T cells. We directly examined the number of allergen-specific Th1/Th2 memory T cells in the peripheral blood of patients of allergic rhinitis by an ELISPOT assay using specific peptides.[12] The Japanese cedar-specific IL-4 producing Th2 cells were detected in all patients examined and increased during the pollen season and decreased during the off-season. However, more than 60% of the cedar-specific memory Th2 cells survived up to 8 months after the pollen season (Fig. 7) .

Allergen-specific immunotherapy is the only current treatment that can change the natural course of allergic rhinitis with long-term effects. However, the conventional immunotherapy with subcutaneous administration is inconvenient because it requires frequent visits to the doctor and also carries the risk of anaphylactic shock.[13] A recent review of randomized

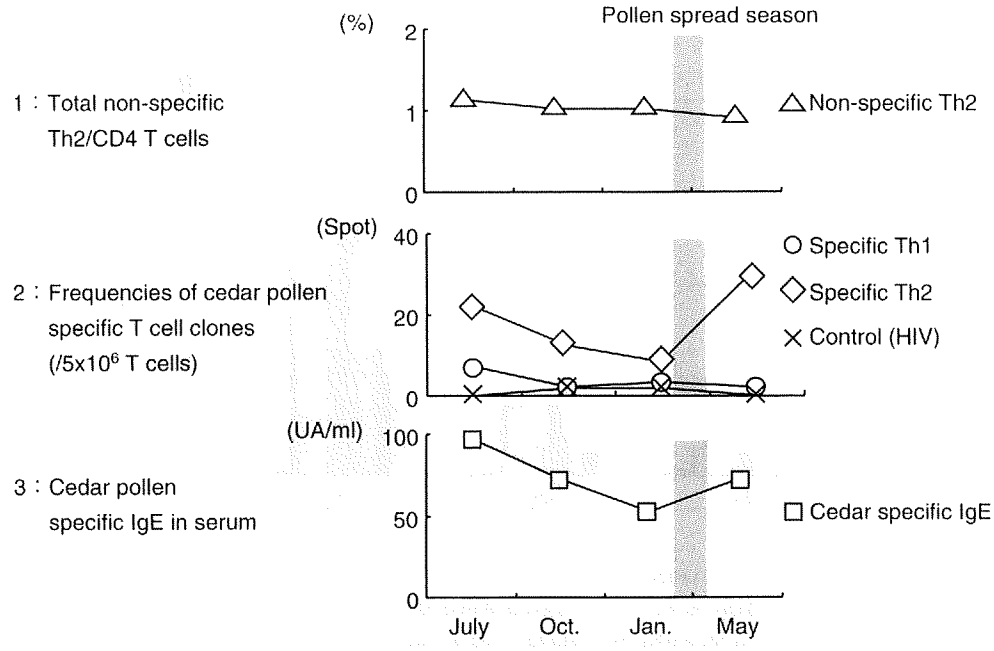

Fig. 7 The seasonal changes of total Th2 cells, frequency of cedar pollen specific T cell clones (spots number) and cedar pollen specific IgE.

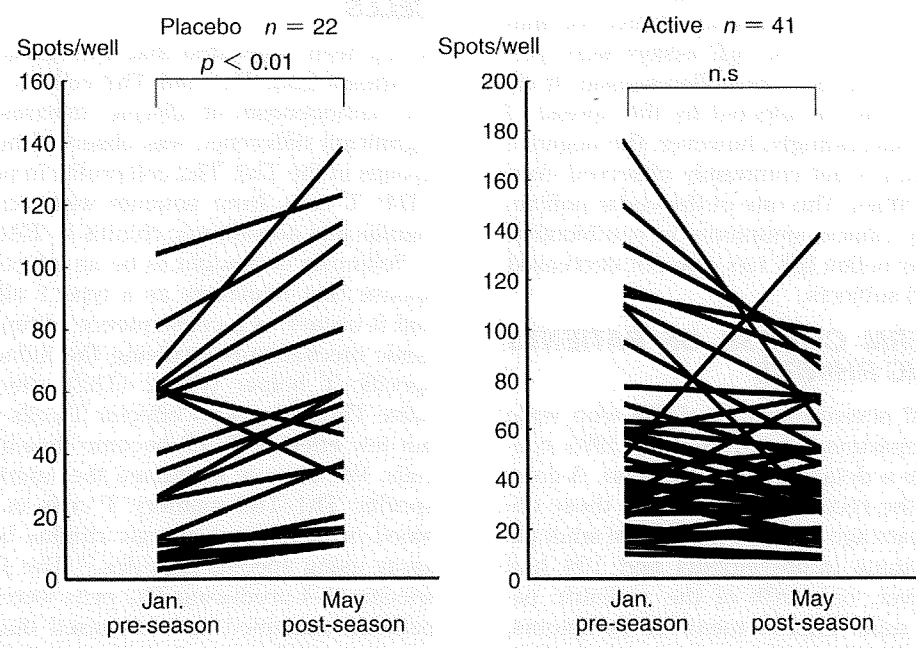

Fig. 8 The number of cedar-specific Th2 cells before and after sublingual immunotherapy.

controlled studies of sublingual immunotherapy suggested that this might be effective as an alternative method of administration.[14-16] To determine the efficacy of sublingual immunotherapy for Japanese cedar pollinosis, we conducted a blinded, randomized, placebo-controlled trial over a period of 6 months (from October 2005 to May 2006).[17] Sixty-seven subjects were enrolled and the nasal symptom scores during the cedar pollen season were evaluated using a symptom diary.

The patients in the active treatment group exhibited significantly lower symptom scores compared to the placebo group. This result suggests that sublingual immunotherapy may offer a safe approach to the management of allergic rhinitis, although the *in vivo* mechanisms of allergen-specific immunotherapy are

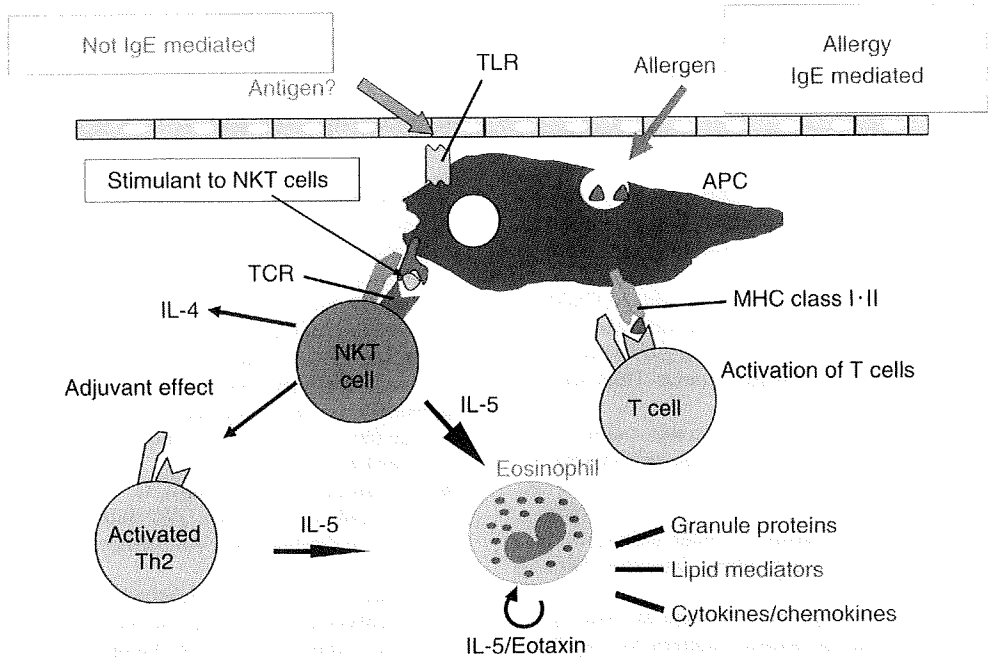

Fig. 9 Mechanism of eosinophil accumulation in respiratory mucosa. Eosinophil accumulation could be observed in MHC class-2 independent.

unknown.

Figure 8 shows the numbers of cedar-specific Th2 cells before and after immunotherapy: the number of Th2 memory cells increased in the placebo group after pollen exposure, but did not increase in the treatment group. Therefore, allergen-specific immunotherapy inhibits an increase in the antigen-specific Th2 memory cell count induced by allergen exposure. Immune-therapeutic intervention might direct at diminishing the size of the clone memory Th2 cells and shifting the cytokine type of memory Th clones.

Natural killer T (NKT) cells represent a unique lymphocyte subpopulation that is characterized by the co-expression of T cells and natural killer receptors.[18,19] Their activity is not restricted to MHC antigens. The relative frequency of NKT cells in the peripheral blood is generally quite low, usually less than 0.1% of PBMCs, and they are not detected in normal peripheral lymph nodes. However, NKT cells play a very important role in innate immunity. Recently, the involvement of NKT cells in the development of airway hypersensitivity in mice and the detection NKT cells in bronchoalveolar-lavage fluid samples from patients with moderate to severe asthma were reported. However, we could not detect the NKT cells in the nasal mucosa of the patients with allergic rhinitis by a polymerase chain reaction. However, NKT cells were detected to varying degrees in the sinus mucosa from asthmatic choronic sinusitis (CS) patients.

These results suggest that NKT cells are not directly related to the development of allergy, but that they may play important roles in the development of sinus disease combined with asthma and in the enhanced Th2 cytokine expression and increased infiltration of Th2 cells and eosinophils observed in the sinus mucosa from asthmatic CS patients via MHC-independent mechanisms (Fig. 9).

SUMMARY

1. The prevalence of allergic rhinitis, in particularly cedar pollinosis, is increasing.
2. Cedar pollen-specific Th1/Th2 dysregulation is observed in patients with pollinosis.
3. Cedar pollen specific memory Th cells increased during the pollen season and decreased during off season, however, morethan 60% of the memory cells survived up to 8 months after the pollen season.
4. NKT cells are not directly related to the development of allergic rhinitis, including pollinosis.
5. Different mechanisms in the accumulation of eosinophilia in the respiratory tract mucosa may exist.

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Cedar and Cypress Pollinosis and Allergic Rhinitis: Quality of Life Effects of Early Intervention with Leukotriene Receptor Antagonists

Keita Sasaki[a] Yoshitaka Okamoto[a] Syuji Yonekura[a] Toru Okawa[a] Shigetoshi Horiguchi[a] Hideaki Chazono[a] Minako Hisamitsu[a] Daiju Sakurai[a] Toyoyuki Hanazawa[a] Kimihiro Okubo[b]

[a]Department of Otolaryngology, Head and Neck Surgery, Graduate School of Medicine, Chiba University, Chiba, and [b]Department of Otolaryngology, Nippon Medical University, Tokyo, Japan

Key Words
Pollinosis · Cedar pollen · Cypress pollen · Leukotriene receptor antagonist

Abstract
Background: Allergic rhinitis involves inflammation of the nasal passages. The use of nasal steroids is generally very effective in providing significant symptom relief. However, compliance for their use is sometimes poor. ***Methods:*** To examine the efficacy of early intervention (before pollen dispersal) with oral cysteinyl leukotriene receptor antagonists (LTRA) on pollinosis in patients with allergy to cedar and Japanese cypress pollens, groups of subjects were treated with LTRA or a placebo for 4 weeks at the beginning of the cedar pollen dispersal season. Subsequently, all patients received nasal steroid therapy concomitantly with LTRA throughout the remaining period of the pollen dispersal season. The effects of such early treatment with LTRA on pollinosis were investigated using symptom scores from an allergy diary and quality of life (QOL) scores. ***Results:*** Sneezing and nasal congestion scores were significantly lower in the LTRA-pretreated subjects than observed in the placebo-pretreated patients between weeks 4 and 6 and weeks 3 and 5, respectively. QOL scores improved significantly in all domains after concomitant therapy with nasal steroids. The percent improvement in the nasal congestion score after the concomitant therapy was significantly higher in the LTRA group (69%) than in the placebo group (41%). ***Conclusion:*** Significant differences observed in symptoms and in QOL effects between LTRA- and placebo-pretreated patients and the absence of major adverse effects noted in these studies suggest that early intervention with LTRA is beneficial and safe and should be considered in the management of pollinosis-associated allergic rhinitis.

Introduction

Allergic rhinitis is a type I allergic disease mediated by specific IgE antibody responses. The disease develops as inflammation associated with early infiltration with eosinophils and other pro-inflammatory cells into the nasal mucosa. The pathogenesis of later phases of allergic rhinitis exhibits many characteristics similar to bronchial asthma [1, 2]. Dust mite allergens are responsible for at least 90% of cases of perennial allergic rhinitis. Arboreal pollens, including that of cedar and Japanese cypress, are also important causes of rhinitis, especially in Japan [3–

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Fax +41 61 306 12 34
E-Mail karger@karger.ch
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Correspondence to: Prof. Yoshitaka Okamoto
Department of Otolaryngology, Head and Neck Surgery
Graduate School of Medicine, Chiba University
1-8-1 Inohana, Chuo-ku, Chiba, 260–8670 (Japan)
Tel. +81 43 226 2137, Fax +81 43 227 3442, E-Mail yokamoto@faculty.chiba-u.jp