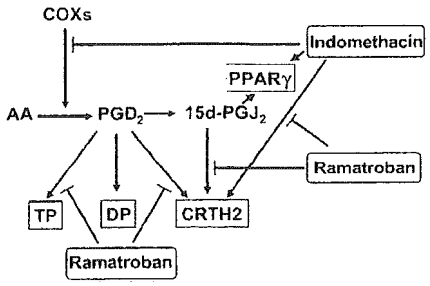

Fig. 7. Effects of ramatroban on migration of CRTH2 transfectants. Ramatroban inhibits PGD_2 activities via prostanoid TP receptor (TP) antagonism. Furthermore, ramatroban inhibits CRTH2 activities induced by PGD_2, 15d-PGJ_2 and indomethacin originally identified as three different ligands for the PGD_2 receptor (DP), peroxisome proliferator-activated receptor (PPARγ) and cyclooxygenases (COXs), respectively.

of many mechanisms dependent on the physiological or pathological processes in question. It is interesting to find that PGD_2, indomethacin and 15d-PGJ_2, originally identified as ligands for three different systems, the prostanoid DP receptor, COXs and PPARγ and stimulating different physiological effects, share a similar binding site with ramatroban on CRTH2 (Fig. 7).

To investigate the signaling efficiency among various CRTH2 agonists, we compared the efficacy and potency of various CRTH2 agonists in receptor binding, Ca^{2+} mobilization, cAMP production and migration assays. 15R-methyl-PGD_2 showed 3-fold higher migrated cell numbers with 2.5-fold higher potency than PGD_2 in inducing migration of CRTH2 transfectants (Fig. 6A), although they showed similar efficacy in inducing Ca^{2+} mobilization and reducing forskolin-induced cAMP production. These results show correlation with the results of human eosinophils. In human eosinophils, 15R-methyl-PGD_2 showed 5-fold higher potency than PGD_2 in upregulating CD11b expression (EC_{50} values of 1.4 and 7 nM, respectively), actin polymerization (EC_{50} values of 3.8 and 13 nM, respectively) and cell migration (EC_{50} values of 1.7 and 10 nM, respectively) (Monneret et al., 2003). Almost all of the maximally efficacious response seen in 15R-methyl-PGD_2-induced cell migration was inhibited by ramatroban in the present study (Fig. 6C), suggesting that its effect was mediated via CRTH2.

In contrast, indomethacin induced 3-fold higher migrated cell numbers but with 65-fold lower potency than PGD_2 in migration of CRTH2 transfectants. The potency in Ca^{2+} mobilization and suppression of cAMP production was 40-fold lower and 200-fold lower than PGD_2, respectively. These results are in close agreement with the study reported by Hirai et al. (Hirai et al., 2002). Indomethacin showed 50-fold lower potency than PGD_2 in Ca^{2+} mobilization using CRTH2 transfectants, and showed similar migrated cell numbers as PGD_2 with 15 to 50-fold lower potency than PGD_2 in human eosinophils, basophils and Th2 cells (Hirai et al., 2002). While it is difficult to speculate on the relevance of a 2 to 3-fold increase in cell number in vitro migration assays of transfectants, it is possible that the efficacy in response is derived from CRTH2-associated signals because ramatroban also inhibited this response.

For 15d-PGJ_2, there are some contradictory reports. Hirai et al. (2001) reported that 15d-PGJ_2 showed 40-fold lower affinity than PGD_2 in [^{3}H]PGD_2 binding to CRTH2-transfected K562 cells (K_i values of 2,300 and 61 nM, respectively). In contrast, Sawyer et al. (2002) described that 15d-PGJ_2 and PGD_2 showed similar affinities in [^{3}H]PGD_2 binding to CRTH2-transfected HEK293 cell membranes (K_i values of 3.2 and 2.4 nM, respectively). Monneret et al. (2002) reported that 15d-PGJ_2 and PGD_2 showed similar potencies in Ca^{2+} mobilization (EC_{50} values of 29 and 60 nM, respectively), actin polymerization (EC_{50} values of 11 and 7 nM, respectively) and CD11b expression (EC_{50} values of 9.4 and 11.7 nM, respectively) in human eosinophils. We obtained interesting results using CRTH2-transfected L1.2 cells. 15d-PGJ_2 showed 90-fold lower potency but showed 1.5-fold greater increase in Ca^{2+} level than PGD_2 in Ca^{2+} mobilization assays, and showed 80-fold lower potency but 12-times greater migrated cell numbers than that of PGD_2 in migration assay using CRTH2 transfectants. Only 15d-PGJ_2 showed such increases in Ca^{2+} level and migrated number of cells among the other CRTH2 agonists tested here. These results suggest that 15d-PGJ_2 has lower potency to CRTH2, but its function could be amplified through different signaling pathways especially at the higher concentration.

Finally, it is known that indomethacin has unwanted side effects causing various complications such as gastrointestinal injury. Shortening of the villi, epithelial stratification, basal lamina degeneration, eosinophil degranulation and infiltration of the epithelium prior to infiltration of the mucosa by neutrophils are earliest histological features of indomethacin-induced intestinal injury in rats (Anthony et al., 1993). It is also known that CRTH2 is expressed on infiltrating cells of the gut mucosa and has been found in acute manifestations of ulcerative colitis (Matsuzaki et al., 2003), thus, indomethacin may also act on these cells via CRTH2. Ramatroban, or specific CRTH2 antagonists, may therefore have potential as therapeutic agents for use in combination with these known anti-inflammatory principles, or as a counterbalance to toxicity associated with similar drugs.

This is the first report showing the evidence for direct ramatroban binding to CRTH2, revealing its competitive inhibitory effects and other interesting findings that PGD_2, indomethacin and 15d-PGJ_2 share the same binding site with ramatroban on CRTH2.

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Nasal exposure to Staphylococcal enterotoxin enhances the development of allergic rhinitis in mice

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Summary

Background Staphylococcal enterotoxins (SEs) appear to play a role in the pathogenesis of allergic disease. However, little is known whether the nasal exposure to SE affects the development of allergic rhinitis (AR).

Objective We sought to determine the *in vivo* effect of nasal exposure to SE on the development of AR using mouse model.

Methods BALB/c mice were intranasally sensitized with *Schistosoma mansoni* egg antigen (SmEA) in the presence or absence of staphylococcal enterotoxin B (SEB). Control mice were intranasally sensitized with either SEB or SmEA alone. The production of antigen-specific antibodies including IgE, nasal eosinoplilia and cytokines by nasal mononuclear cells was compared among mice that had or had not received SEB treatment.

Results Nasal exposure to SEB enhanced the development of AR in SmEA-sensitized mice, as manifested by SmEA-specific IgE production, nasal eosinophilia, and IL-4 and IL-5 production by nasal mononuclear cells after Ag challenge. This treatment also elicited IFN-γ production by SmEA-primed cells. In addition, these mice produced SEB-specific IgE whereas mice treated with SEB without SmEA sensitization did not produce SEB-specific IgE or demonstrate nasal eosinophilia.

Conclusion These results suggest that the nasal exposure to SEB enhances susceptibility to AR although the exposure to SE solely does not induce AR.

Keywords allergy, enterotoxin, IgE, mouse, rhinitis

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Introduction

Superantigenic exotoxins produced by *Staphylococcus aureus*, including the staphylococcal enterotoxins (SEs), exfoliative toxins, and toxic shock syndrome toxin (TSST)-1, trigger an excessive immune response and appear to be involved in the pathogenesis of allergic disease, especially atopic dermatitis (AD) [1–4]. They elicit T cell activation, which leads to the production of IL-4 and IL-5 [5, 6]. Direct application of these toxins to the respiratory mucosa and to lesions of the skin induces an inflammatory response *in vivo* in both humans and mice [7–9]. In addition, *S. aureus*-derived toxins act as allergens [10–13]. Histamine release from basophils has been observed in patients with AD sensitized to SE upon exposure to SE toxins, however, histamine release has not been observed in un-sensitized AD patients or in normal controls upon exposure [13].

The mechanism by which *S. aureus* and/or *S. aureus* toxins contribute to the development of allergic rhinitis (AR) remains unclear despite the fact that *S. aureus* is often found in the nasal mucosa and is one of the pathogens of acute infectious rhinosinusitis [14, 15]. Recently, Shiomori et al. have reported higher rates of nasal carriage of *S. aureus* and toxin-producing *S. aureus* among patients with perennial AR, compared with control subjects. In addition, peripheral blood mononuclear cells (PBMC) from these patients produced higher levels of IL-4 and IL-5, and decreased levels of IFN-γ response to the toxins [6]. Another group reported that TSST-1 enhanced pollen-specific IgE production by PBMC from patients with pollinosis during the pollen season [16]. More recently, we have reported an increased prevalence of sensitization to SEs among patients with AR, compared with healthy controls. Also, patients sensitized to SEs had higher levels of total serum IgE and exhibited poly-sensitization to other inhaled allergens, when compared with un-sensitized patients [17]. These findings prompted us to investigate the possibility that intranasal exposure to SEs may affect the allergic responses such as the production of allergen-specific IgE and nasal eosinophilia, in cases of AR.

We have recently developed a murine model of AR using *Schistosoma mansoni* egg antigen (SmEA) as allergen [18]. Intranasal sensitization with SmEA induces allergic hallmarks such as the production of Ag-specific IgE and local

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eosinophila in the absence of any adjuvants, thus we believe that this model is suitable to investigate the initiation of AR. In the present study, we investigated the role of nasal exposure to SEB, one of the commonest exotoxins found in the nasal cavity [6], in the development of AR *in vivo*. The results presented here may argure against our current understanding of the 'hygiene hypothesis' [19].

Materials and methods

Animals and antigens

Female BALB/c mice aged 7–10 weeks old were purchased from Charles River Japan (Yokohama, Japan). The mice were maintained in SPF condition at Okayama University Medical School in accordance with the guidelines set forth by the Okayama University Medical Area Research Committee. And all experimental protocols and procedures in the present study were approved by institutional animal care and use Committee. SmEA was prepared as previously described [18]. SEB was purchased from Toxin Technology Inc. (Sarasota, FL, USA). Ovalbumin (OVA: grade V) was purchased from Sigma (St Louis, MO, USA).

Sensitization of mice

Mice ($n = 6$ per gourp) were intranasally sensitized with SmEA in the absence of adjuvants to archive the initiation of AR as described previously with slight modification [18]. In brief, low (0.1 μg) or high (10 μg) amount of SmEA mixed with either 5 or 500 ng of SEB in 20 μL phosphate-buffered saline (PBS) was instilled into nostrils once a week for 3 weeks. One week after the third sensitization, the same treatment was challenged intranasally for 7 consecutive days. As control, mice were received either the same amount of SmEA or PBS alone. In another set of experiment, mice were applied intranasally with 500 ng of SEB alone in a same manner as described above (Fig. 1a). Peripheral blood was collected from the tail vein prior to sensitization (day 0), 6 days after the third sensitization (day 20) and/or 12 h after the final challenge (day 28), then centrifuged at 200*g*, and the specific Ab content in the serum was tested.

In order to investigate the effect of exposure to SEB on nasal challenge, BALB/c mice ($n = 6$ per group) were sensitized intraperitoneally with 50 μg OVA adsorbed to 1 mg alum (Kyowa Kagaku, Kagawa, Japan) in a total volume of 200 μL on day 0, 7, and 14. Two weeks after the third intraperitoneal sensitization, mice were challenged intranasally with 10 μg OVA in the presence or absence of 5 or 500 ng of SEB in 20 μL PBS for 7 consecutive days. Twelve hours after the final nasal challenge, blood was taken (Fig. 1b).

Antibody determination

The levels of Ag-specific Ab including IgE, IgG1, and IgG2a were measured by ELISA as previously described [20]. The levels of SEB-specific IgE Ab were also measured by captured ELISA using biotinylated SEB (Toxin Technology Inc.) as a detection reagent [20]. Titres for specific IgE were estimated as mean optical density (OD) at 450 nm of 1 : 4 diluted sera.

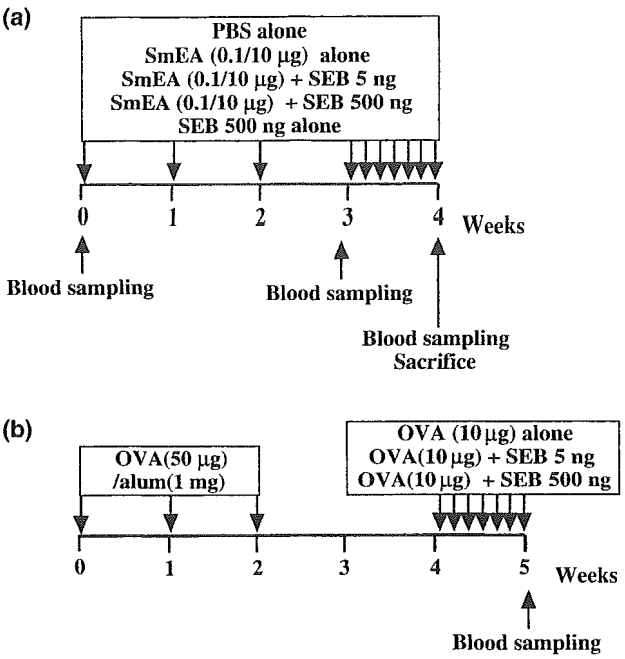


Fig. 1. Experimental design used to investigate the effect of nasal exposure with staphylococcal enterotoxin B (SEB) on the initiation (a) and nasal nasal challenge (b) of allergic rhinitis in mice. SmEA, *Schistosoma mansoni* egg antigen.

Titres for specific IgG1 and IgG2a are estimated as mean OD at 450 nm of 1 : 100 diluted sera.

Histological examination

Histological examination was performed as previously described [20]. Briefly, 12 h after the final nasal challenge mice were killed by cervical dislocation. Their heads were removed, fixed in 10% formalin and decalcified with 2.5% EDTA-2Na solution. Coronal nasal section were stained with Luna solution and the number of eosinophils in the nasal mucosa was counted microscopically under a high power field (10 × 40).

In vitro *culture of nasal mononuclear cells and cytokine determination*

Twelve hours following the final nasal challenge, mice were sacrificed, and heads were removed. Nasal mononuclear cells were isolated as described previously [20]. In brief, skin-peeled nasal components were cut into small pieces, and incubated with RPMI 1640 (Sigma) containing 4 mg/mL collagenase (Boehringer-Mannheim, Indianapolis, IN, USA) for 30 min at 37 °C. After the incubation, a single cell suspension obtained by passing 70 μm cell strainer (BD Falcon, Bedford, MA) was placed on top of 40% and 75% Percoll density gradient solutions (Pharmacia, Uppsala, Sweden). After the centrifugation at 200*g* for 15 min, nasal mononuclear cells were recovered from interface between the 40% and 75% Percoll solutions. Then IL-4, IL-5, and IFN-γ production by SmEA-stimulated and unstimulated cells were measured [20]. Detection limit for IL-4, IL-5, and IFN-γ in this system was 0.1 U/mL, 40 pg/mL, and 0.1 IU/mL, respectively.

Statistical analysis

Student's unpaired *t*-test was used to determine the statistical significance of the values obtained. $P<0.05$ was considered statistically significant. Data are expressed as the mean standard error of mean (SEM) for each subject group.

Results

Nasal exposure to staphylococcal enterotoxin enhances the development of allergic rhinitis

Following repeated intranasal sensitization with 0.1 μg of SmEA, which has been shown to induce mild sensitization [21], BALB/c mice produced detectable amounts of SmEA-specific IgE and IgG1, but not IgG2a (Figs 2a–c). An increased level of total serum IgE was also observed (Fig. 2d). Simultaneous application of 5 ng of SEB with SmEA led to a significant increase in the production of SEA-specific IgG1. This treatment also enhanced, albeit not significantly, the production of SmEA-specific IgE. On the other hand, application of 500 ng of SEB with SmEA significantly enhanced both SmEA-specific IgG1 and IgE production (Figs 2b, c). This treatment also elicited an increase in total serum IgE (Fig. 2d). In addition, both concentrations of SEB increased the production of SmEA-specific IgG2a, however, these increases in production were not statistically significant (Fig. 2c).

Similar results were obtained with regard to the development of local eosinophilia, as determined by histological examination. Mice sensitized with 0.1 μg of SmEA showed subtle eosinophil infiltration, and the average number ± SEM of eosinophils per field (10 × 40) of the nasal septum was 5.33 ± 1.31 (Fig. 3a). In mice treated with 5 ng of SEB, a slight increase in eosinophil infiltration was observed, compared with control mice, but this increase was not significant (Fig. 3B). Eosinophil infiltration was significantly greater in mice sensitized with SmEA mixed with 500 ng of SEB, compared with mice sensitized with SEA alone (Fig. 3c). The average number ± SEM of eosinophils per field was 8.83 ± 1.25 and 35.50 ± 6.80 in mice treated with 5 and 500 ng of SEB, respectively.

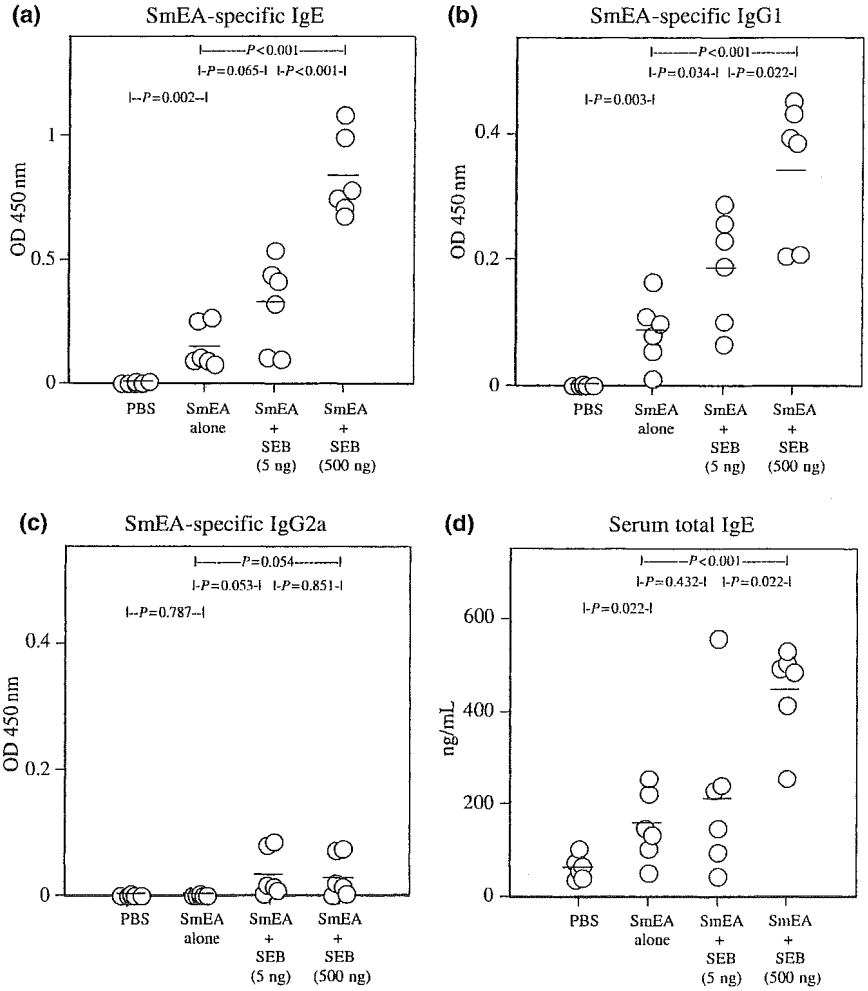


Fig. 2. Effect of nasal exposure with staphylococcal enterotoxin B (SEB) on antibody production following intranasal sensitization with 0.1 μg of *Schistosoma mansoni* egg antigen (SmEA). BALB/c mice ($n = 6$) were intranasally sensitized with 0.1 μg of SmEA or phosphate-buffered saline (PBS). Five or 500 ng of SEB or PBS, which served as a control, were applied intranasally at the same time, as shown in Fig. 1. Blood was sampled form the tail 12 h after the final nasal challenge. Titres of SmEA-specific IgE (a), IgG1 (b), IgG2a (c), and serum total IgE (d) were determined by ELISA. Results show (a) the mean optical density (OD) at 450 nm ± SEM of six serum samples from each group at a 1 : 4 dilution, and (b, c) the mean OD at 450 nm ± SEM of six serum samples from each group at a 1 : 100 dilution. Data are representative of three separate experiments.

Cytokine production from nasal mononuclear cells after challenge with SmEA was then examined. The cells were isolated by enzyme extraction and stimulated with SmEA for 72 h, after which cytokine production within the culture supernatant was assessed. SmEA-stimulated nasal mononuclear cells from control mice produced detectable amounts of IL-4 and IL-5, but not IFN-γ (Figs 4a–c). Nasal application of 500 ng of SEB with each repeated intranasal application of

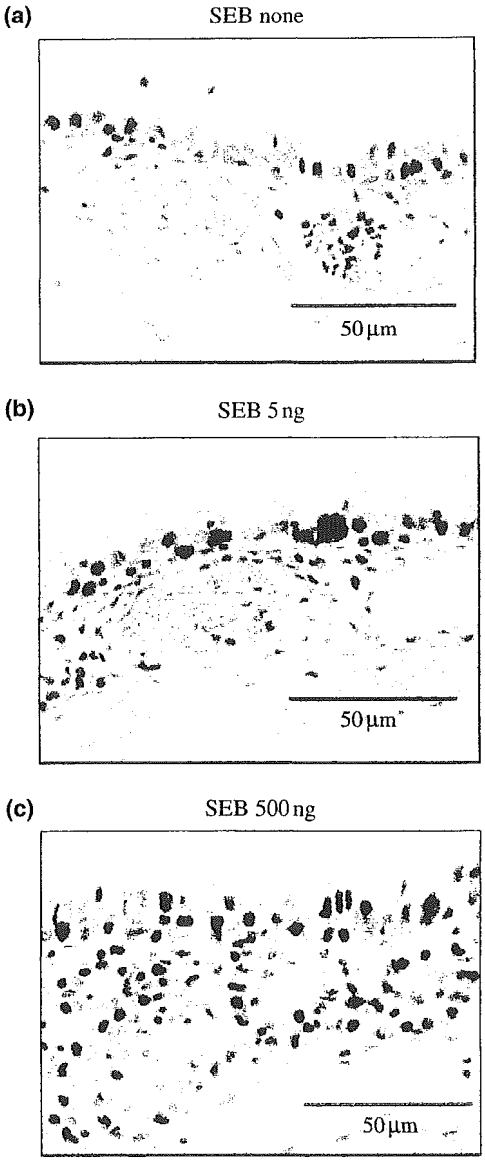


Fig. 3. Effect of nasal exposure with staphylococcal enterotoxin B (SEB) on the induction of nasal eosinophilia. BALB/c mice ($n = 6$) were intranasally sensitized with *Schistosoma mansoni* egg antigen (SmEA). Phosphate-buffered saline (a) 5 ng of SEB (b), or 500 ng of SEB (c) were applied intranasally at the same time of sensitization. Twelve hours after the final nasal challenge with SmEA, mice were sacrificed. Nasal sections were fixed, decalcified, and Luna staining was performed to detect eosinophils in the nasal mucosa.

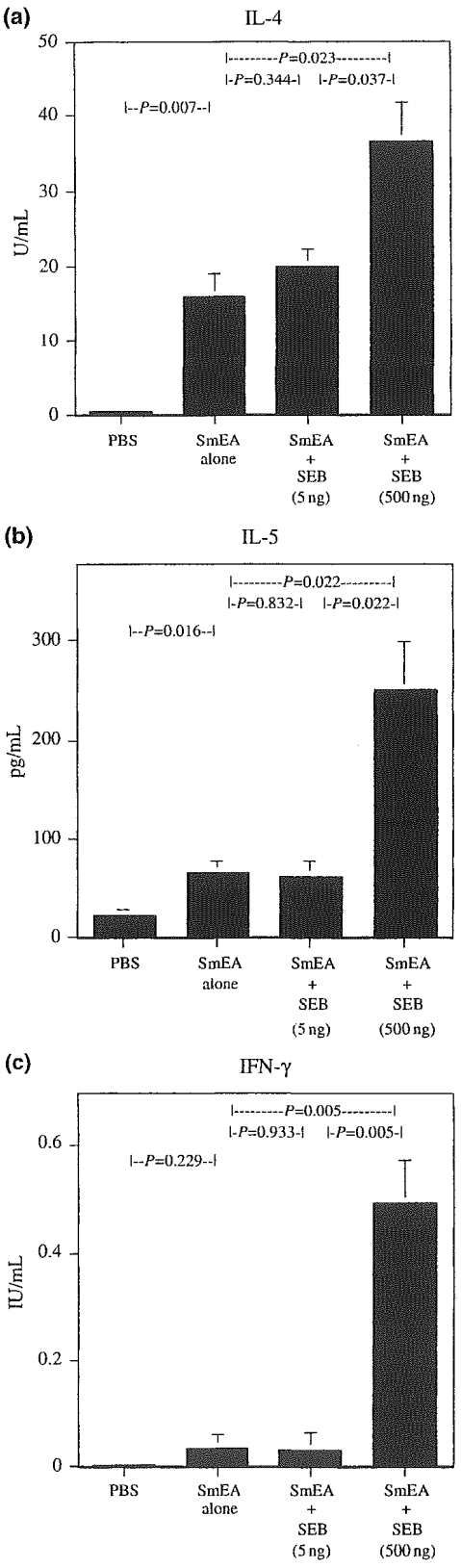


Fig. 4. Production of IL-4 (a), IL-5 (b) and IFN-γ (c) by nasal lymphocytes from BALB/c mice intranasally exposed with staphylococcal enterotoxin B (SEB) or phosphate-buffered saline (PBS) during sensitization and subsequent challenge with *Schistosoma mansoni* egg antigen (SmEA). Nasal lymphocytes were isolated and cultured *in vitro* for 72 h with SmEA. Cytokines were measured by ELISA. Results show the mean ± SEM of three separate experiments.

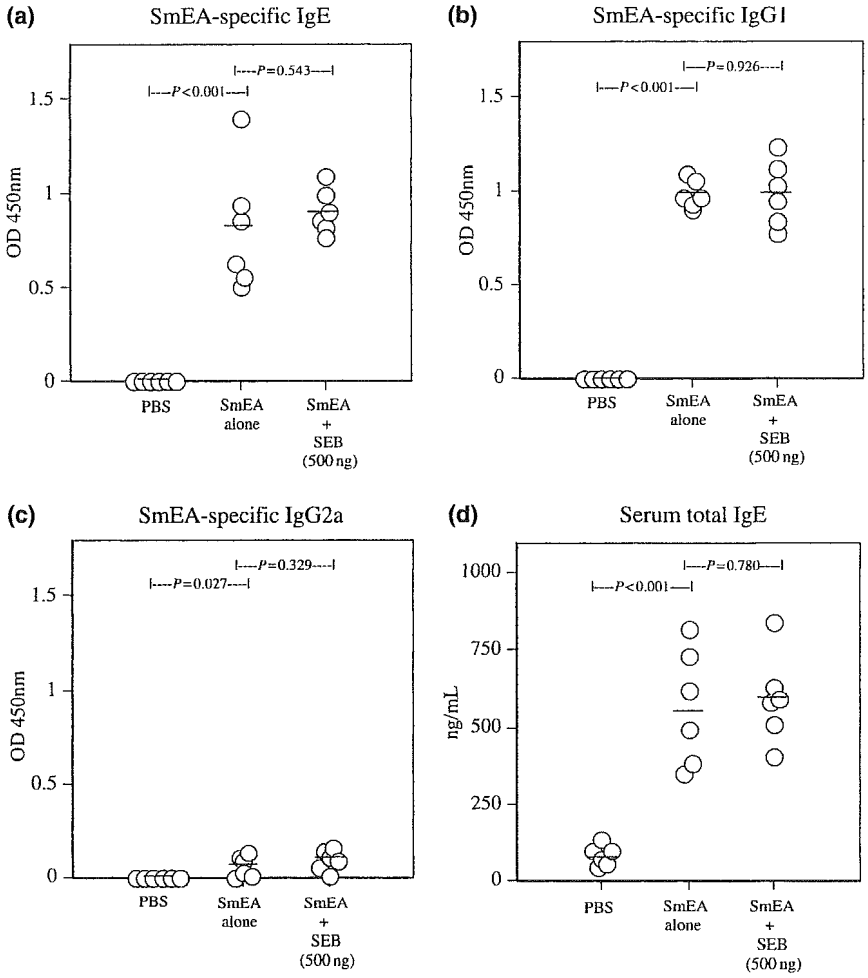


Fig. 5. Effect of nasal exposure with staphylococcal enterotoxin B (SEB) on antibody production following intranasal sensitization with 10 μg of *Schistosoma mansoni* egg antigen (SmEA). BALB/c mice ($n = 6$) were intranasally sensitized with 10 μg of SmEA or phosphate-buffered saline (PBS). Five hundred nanograms of SEB or PBS, which served as a control, were applied intranasally at the same time, as shown in Fig. 1. Blood was sampled form the tail 12 h after the final nasal challenge. Titres of SEA-specific IgE (a), IgG1 (b), IgG2a (c), and serum total IgE (d) were determined by ELISA. Results show (a) the mean optical density (OD) at 450 nm ± SEM of six serum samples from each group at a 1 : 4 dilution, and (b, c) the mean OD at 450 nm ± SEM of six serum samples from each group at a 1 : 100 dilution. Data are representative of two separate experiments.

SmEA led to an increase in IL-4 and IL-5 production (Figs 4a and b). In addition, nasal mononuclear cells also produced significantly greater levels of IFN-γ in response to SmEA, than those from mice sensitized with SmEA alone (Fig. 4c). On the other hand, nasal application of 5 ng of SEB did not significantly affect cytokine production.

Effect of nasal exposure to staphylococcal enterotoxin on the development of allergic rhinitis depends on the dose of allergen introduced

BALB/c mice were intranasally sensitized with 10 μg of SmEA, which has been shown to induce full sensitization [21]. BALB/c mice produced greater amounts of SmEA-specific IgE and IgG1, along with increased levels of total serum IgE, compared with mice treated with 0.1 μg of SmEA (Figs 2 and 5). Nasal exposure to 500 ng of SEB did not alter the production of SmEA-specific Abs or total serum IgE (Fig. 5). In addition, this treatment did not modify the degree of eosinophilia noted following repeated nasal sensitization with 10 μg of SmEA. The average number ± SEM of eosinophils per field (10 × 40) of the nasal septum was 80.67 ± 11.63 and 59.50 ± 6.70 in mice that did or did not receive intranasal exposure to 500 ng of SEB, respectively ($P = 0.146$).

Nasal exposure to staphylococcal enterotoxin alone does not induce the pathological features of allergic rhinitis

We sought to determine whether repeated nasal exposure to SEB alone induces the production of IgE or results in nasal eosinophilia. Levels of total serum IgE did not differ significantly among mice exposed to 500 ng of SEB alone or PBS alone throughout the experiment (Fig. 6). In addition, eosinophilia was not observed within the nasal mucosa of mice treated with 500 ng of SEB alone.

Staphylococcal enterotoxin-specific immunoglobulin E was detected only in the presence of allergen

Mice sensitized with 0.1 μg of SmEA together with 500 ng of SEB produced significant levels of SEB-specific IgE after

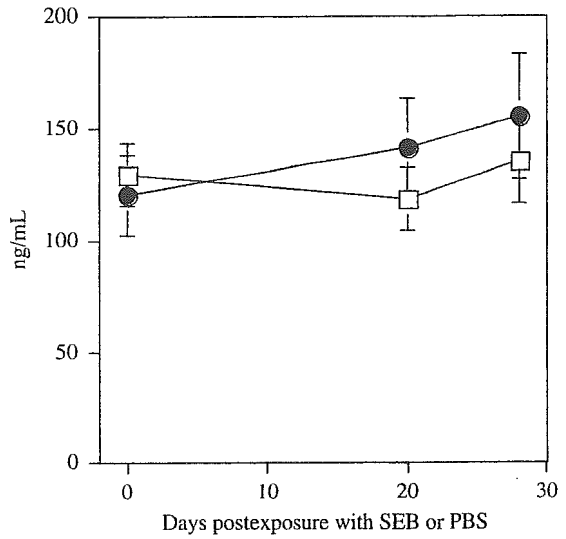


Fig. 6. Effect of nasal exposure with staphylococcal enterotoxin B (SEB) alone on the amount of serum total IgE. BALB/c mice ($n = 6$) were repeatedly applied with 500 ng of SEB (closed circle) or phosphate-buffered saline (PBS) (open square) intranasally as shown in Fig. 1. Blood was sampled prior to (day 0), 6 days after the third (day 20), and 12 h after 10th (day 28) exposure with SEB. Results show the mean amounts ± SEM of six serum samples from each group. Data are representative of two separate experiments.

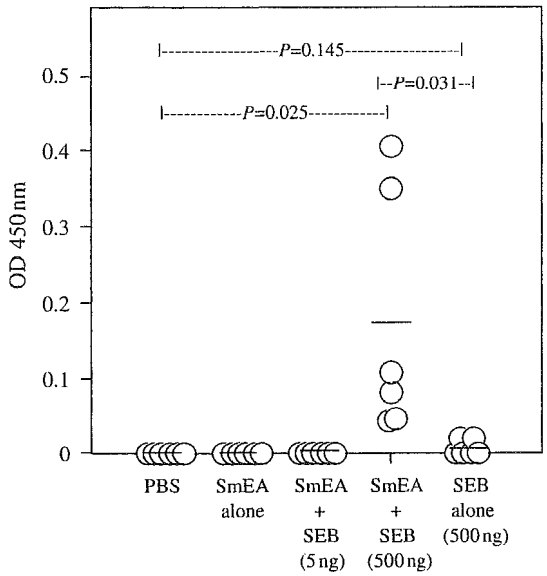


Fig. 7. Effect of nasal exposure with staphylococcal enterotoxin B (SEB) on SEB-specifc IgE production. BALB/c mice ($n = 6$) were repeatedly received the intranasal application with phosphate-buffered saline (PBS), 0.1 μg of *Schistosoma mansoni* egg antigen (SmEA) alone, 0.1 μg of SmEA together with 5 ng of SEB, 0.1 μg of SmEA together with 500 ng of SEB or 500 ng of SEB alone as shown in Fig. 1. Blood was sampled form the tail 12 h after the final nasal challenge. Titres of SEB-specific IgE were determined by ELISA. Results show the mean optical density at 450 nm ± SEM of six serum samples from each group at a 1 : 4 dilution. Data are representative of two separate experiments.

nasal challenge. Mice sensitized with SmEA together with 5 ng of SEB also produced SEB-specific IgE, however, a significant increase in the level of SEB-specific IgE over that of control mice was not observed. Mice treated with 500 ng of SEB alone did not display significant SEB-specific IgE production (Fig. 7).

Effect of nasal exposure to staphylococcal enterotoxin on nasal challenge in presensitized mice

BALB/c mice presensitized with OVA were intranasally challenged with OVA in the presence or absence of 5 or 500 ng of SEB. Mice exposed to 500 ng of SEB during the nasal challenge produced significantly higher amount of OVA-specific IgE but not IgG1 or IgG2a as compared with other groups (Fig. 8).

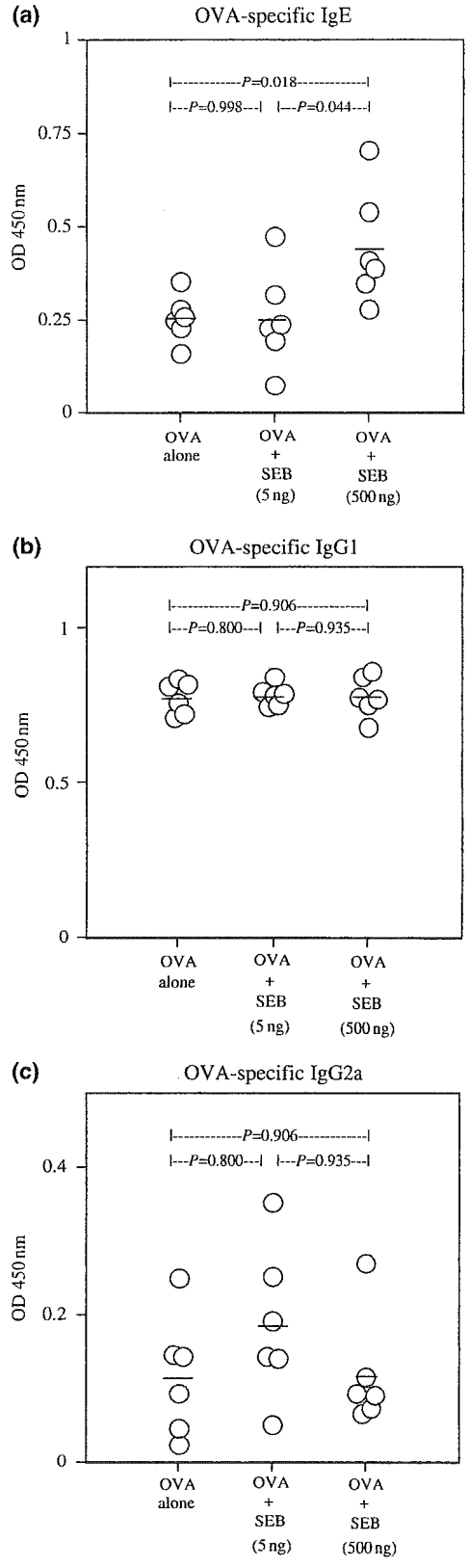
Discussion

In the present study, we analyzed the immunological effects of nasal exposure to a *S. aureus*-derived enterotoxin, specifically with regard to the development of AR in mice. Mice sensitized with 0.1 μg of SmEA together with 500 ng of SEB produced significantly greater amounts of SmEA-specific IgE and IgG1, as well as total serum IgE, compared with mice sensitized with SmEA alone (Fig. 2). Eosinophil infiltration of the nasal mucosa was also markedly increased in mice treated with 500 ng of SEB, compared with control mice (Fig. 3). These results suggest that nasal exposure to SEB enhances the development of AR.

There are several mechanisms by which exposure to an enterotoxin might augment the development of AR. First, SEB might enhance the immune response following presentation of an allergen-specific antigen. Conventional allergens, such as SmEA, are ingested by antigen-presenting cells (APCs) such as macrophages and dendritic cells, after which they are digested into small peptides, which then bind to the groove in major histocompatibility complex (MHC) class II molecules. After this, T cells display allergen-specific activation via binding between the T cell receptor (TCR) and peptide/MHC class II molecules [22]. Superantigens, such as SEB, stimulate T cells in the absence of these conventional proceeding and presentation, since superantigen can bind directly to MHC class II β chain and the TCR-Vβ element [23]. Thus, direct binding of SEB to an MHC class II molecule loaded with SEA-derived peptides might enhance the antigenicity of an allergen. Second, activation of immune cells, such as T cells and dendritic cells, by superantigens leads to the synthesis of a variety of pro-inflammatory cytokines, including IL-1 and TNF-α [24, 25]. In addition, SEB has been shown to induce high levels of CC chemokines, such as macrophage inflammatory protein (MIP)-1α, MIP-1β, and monocyte chemoattractant protein-1 (MCP-1), which are involved in the recruitment and activation of eosinophils [26, 27]. A microenvironment in which these pro-inflammatory cytokines and chemokines are present might set the stage for the development of AR. In fact, mice deficient in TNF-α have displayed a significant reduction in antigen-specific IgE production, nasal symptoms, T helper 2 (Th2)-type cytokine production, and nasal eosinophilia, in a murine model of AR [28].

Interestingly, such an adjuvant effect of SEB is not found when mice were sensitized with 10 μg of SmEA, with which dose the pathological features of AR were fully induced (Fig. 5). In addition, nasal exposure with low dose (5 ng) of SEB

only augmented SmEA-specifc IgG1 production but not IgE production or nasal eosinophilia (Figs 2 and 3). In addition, nasal exposure to a low dose of SEB (5 ng) only augmented SmEA-specific IgG1 production, but not IgE production or nasal eosinophilia (Figs 2 and 3). These results suggest that both SEB and the SmEA allergen have dose-dependent effects on the development of AR.



Nasal mononuclear cells from SmEA-sensitized mice exposed to 500 ng of SEB, produced not only IL-4 and IL-5, but also IFN-γ, in response to recall stimulation with SmEA *in vitro* (Fig. 4). This result suggests that nasal exposure to SEB does not induce a strict Th2-skewed response. The finding of a slight, but not significant, increase in the production of SmEA-specific IgG2a upon exposure of mice to 500 ng of SEB (Fig. 2c) supports this assertion. As described above, superantigens induce the production of various pro-inflammatory cytokines and chemokines, some of which preferentially induce T helper type 1 (Th1) responses [29]. In addition, Saloga et al. [30] have revealed a shift in the immune response of mice to OVA from Th2-associated Ab production of OVA-specific IgE and IgG1 to Th1-associated Ab production of IgG2a, IgG2b and IgG3, following intradermal administration of SEB. Since SmEA is a potent Th2-inducer because of its carbohydrate content [31], it is possible that the presence of both allergen and exotoxin might facilitate both Th1 and Th2 responses.

Recent reports revealed that IgE to SE is related with local eosinophilic inflammation in nasal polyps and the severity of asthma [32, 33]. And we have observed an increased prevalence of SE-specific IgE among patients with AR, compared with healthy controls. In addition, patients sensitized with SEs significantly demonstrated increased levels of total serum IgE and polyvalent sensitization [17]. In the present study, SEB-specific IgE production, and increased levels of total serum IgE, were seen in mice exposed to SEB combined with SmEA (Figs 2 and 7). The present results are consistent with clinical observations, and suggest that nasal exposure to SEB may be involved in polyvalent sensitization.

However, intranasal application of SEB alone did not induce significant production of SEB-specific IgE or eosinophilia within the nasal mucosa (Figs 6 and 7). Herz et al. [9] have reported induction of pulmonary eosinophilia, and detection of Th2-dominant cytokines by bronchoalveolar lavage, as well as tracheal hypersensitivity, following intranasal administration of SEB. Although they did not focus on IgE production or nasal inflammation, their results are not consistent with our own. This may be because of different responses of the upper and lower airways to SEB. In fact, upper and lower airways are known to display different responses to bacterial products, such as lipopolysaccharide [34]. Our results suggest that SEB does not act as an allergen on its own, and therefore does not result in SEB-specific IgE production and nasal eosinophilia. However, simultaneous application with SmEA may enhance the antigenicity of the SEB and lead to the production of SEB-specific IgE. This

Fig. 8. Effect of nasal exposure with staphylococcal enterotoxin B (SEB) on nasal challenge. BALB/c mice ($n = 6$) were presensitized with ovalbumin (OVA)/Alum, then intranasally challenged with OVA in the presence or absence of 5 ng or 500 ng of SEB, as shown in Fig. 1. Blood was sampled form the tail 12 h after the final nasal challenge. Titres of OVA-specific IgE (a), IgG1 (b), and IgG2a (c) were determined by ELISA. Results show (a) the mean optical density (OD) at 450 nm ± SEM of six serum samples from each group at a 1 : 4 dilution, and (b, c) the mean OD at 450 nm ± SEM of six serum samples from each group at a 1 : 100 dilution. Data are representative of two separate experiments.

may be because of the adjuvant activity of SmEA since it contains immunomodulatory carbohydrate that leads to the induction of Th2 responses.

In conclusion, we demonstrated that nasal exposure to SEB enhances sensitization and allergic inflammation in a murine model of AR. The present results may argue against our current understanding of the 'hygiene hypothesis' [19]. In fact, recent report suggested that a difference in responses to Gram-negative vs. Gram-positive bacteria may affect the initiation of allergic diseases [35].

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厚生労働科学研究費補助金（免疫アレルギー疾患予防・治療研究事業）
研究報告書
リアルタイムモニター飛散数と現状の治療によるQOLの関連性の評価と花粉症根治療法の開発
アレルギー性鼻炎に対するアルゴンプラズマ凝固療法の有効性への検討

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研究要旨
通年性アレルギー性鼻炎64例、スギ花粉症115例に対してアルゴンプラズマ凝固療法（以下APC）の長期間の有効率の変化を検討した。その結果、通年性アレルギー性鼻炎に対してはAPC施行後6ヵ月までは各症状、局所所見とも87-89%の良好な有効率であった。APC施行1年後では鼻閉、局所所見、生活支障度への有効率は78-79%だが鼻汁、くしゃみへの有効率は67-69%と症状により効果に差がでてきた。APC施行2年後になると各症状の有効率は50%前後となりAPC施行後1-2年の間に再照射や他の治療法を考慮する必要のある症例が増えてきた。スギ花粉症へのAPCの有効率は74%で満足度も74%だった。花粉飛散量が平年並みの年の有効率は60%だが、平年よりかなり少ない年の有効率は92%だった。花粉症への効果は花粉飛散量により大きく左右された。花粉飛散量がかなり少ない年では、前年にAPCを施行された3分の1の症例でAPCの越年効果が認められた。花粉量が平年並みの年にAPCは毎年行うのが望ましいと考えられた。カモガヤ花粉症を合併したスギ花粉症では14例中13例でカモガヤ花粉飛散終了までAPCの効果は持続した。通年性アレルギー性鼻炎合併スギ花粉症では有効率は単独例と変わりなかったが、満足度では劣った。花粉飛散中のAPCの有効率は、飛散前照射より劣るが満足度では変わりなかった。

A研究目的
保存的治療に反応しない通年性アレルギー性鼻炎に対してレーザー治療を中心とした外科的治療の有効性が確立されてきている。炭酸ガスレーザーと同様の作用機序をもつアルゴンプラズマ凝固装置を用いたアレルギー性鼻炎の外科的治療の有効性が報告されてきているが、その数は少なく、さまざまな問題に対して充分検討されているとはいえない。また、レーザー治療に関しても花粉飛散量に伴って重症度の変動する花粉症に対する有効性の検討は我々の吸入ステロイド薬との比較試験以外無く、さらに越年効果はこれまでに検討されていない。今回はAPCのアレルギー性鼻炎への長期成績および花粉症への越年効果をけんとうした。アルゴンプラズマ凝固装置とは非接触のモノポーラル電気メスで、組織に吹きかけるアルゴンガスに高周波交流電流を放電し炭酸ガスレーザーと同様に組織表面を蒸散させる。

B対象と方法
薬剤治療に反応しにくい中等症以上の通年性アレルギー性鼻炎患者男性38名（平均年齢28歳）、女性26名（平均年齢28歳）、年齢8歳から57歳、スギ花粉症患者男性34名（平均36歳）、女性31名（平均37歳）、年齢9歳から58歳であった。

方法は局所麻酔剤と血管収縮剤を等量混合した液に浸したガーゼにて鼻粘膜を30分表面麻酔し、アルゴンプラズマ凝固装置（アムコ社製）を用いて出力26—32ワット、流量1．2L/minで下鼻甲介粘膜を可及的広範囲に焼灼した。両側鼻粘膜への治療時間は約5分でガーゼタンポンは挿入せず全例同様の手技で治療は終了した。

評価方法は通年性アレルギー性鼻炎の局所所見に対しては鼻アレルギー診療ガイドラインの局所所見の程度分類の各項目の合計が3段階以上改善を著効、2段階以上改善を有効、1-0段階改善を無効、1段階以上悪化を悪化とした。各症状に対する評価は鼻アレルギー診療ガイドラインの各症状の項目が2段階以上改善を著効、1段階以上改善を有効、改善なしを無効、1段階以上悪化を悪化とした。治療への満足度はアンケート調査により5段階で評価した。生活の支障度への評価ではアンケート調査により日常生活への支障がなかったものを著効、日常生活への支障がたまにあったものを有効、日常生活への支障があったものを無効、日常生活への支障が強く仕事、勉強に支障があったものを悪化とした。

スギ花粉症への治療効果への評価は花粉の飛散量により時期、年により大きく異なるためシーズンを通しての評価とした。アンケート調査により花粉シーズン中ほとんど症状がなかたものを著効、

シーズン中たまに症状があったが薬を悪い時のみ服用して調子がよかったものを有効、薬を併用し調子がよかったものをやや有効、薬を併用しても症状があったが生活に支障はなかったものを無効1、いつものシーズンと変わりなく生活に支障があり苦痛であったものを無効2とした。治療への満足度はアンケート調査により5段階で評価した。

C 結果

通年性アレルギー性鼻炎に対するAPCの治療効果は治療1ヵ月後では有効以上が鼻閉で97%、鼻汁で92%、くしゃみで94%、生活支障度で94%、局所所見で97%だった。治療6ヵ月後、経過を把握できた47名中、有効以上が鼻閉で87%、鼻汁で87%、くしゃみで87%、生活支障度で89%、局所所見で88%だった。治療1年後、経過を把握できた36名中、有効以上が鼻閉で78%、鼻汁67%、くしゃみ69%、生活支障度78%、局所所見79%だった。治療2年後経過を把握できた29名中、有効以上が鼻閉で55%、鼻汁41%、くしゃみ45%、生活支障度52%、局所所見59%だった。治療後6ヵ月までは90%弱の有効率で良好な成績だったが治療後1年以上では鼻閉、局所所見は良好な成績だったが、鼻汁、くしゃみでの有効率は劣ってきた。

通年性アレルギー性鼻炎へのAPCの治療への満足度ではアンケートの回答のあった48名中、大変満足15%、満足46%、どちらともいえない27%、不満8%、大変不満4%だった。大変不満の原因は金属の入れ歯によるプラズマ照射中の痛み、治療効果が思っていたほど続かなかった事であった。

スギ花粉症に対するAPCの治療効果は薬物治療を必要としなかった著効29%、時々薬物治療を必要とした有効45%、薬物治療を併用し良好だったやや有効23%、無効（1）1.5%、無効（2）1.5%だった。

年度別のスギ花粉症へのAPCの治療効果をみると、花粉の飛散量が平年並みの平成14年度では男性13名、女性12名中、著効16%、有効44%、やや有効36%、無効4%だった。花粉の飛散量が平年よりやや少なかった平成15年度では男性14名、女性13名中、著効30%、有効48%、やや有効19%、無効4%だった。花粉の飛散量が平年よりかなり少なかった平成16年度では男性7名、女性6名中、著効54%、有効38%、やや有効8%、無効0%だった。花粉の飛散量により治療効果に大きな差がみられた。平成15年にAPCを施行した27名中9名は平成16年度の花粉症に対し特に治療を必要としなかった。各年度の滋賀県でのスギ花粉飛散量は環境省花粉情報サイトで調査した。

スギ花粉症へのAPCの治療に対する満足度は65名中、大変満足14%、満足60%、どちらともいえない23%、不満3%だった。不満の原因は治療中の歯への痛み、説明したにもかかわらず1回のAPCで花粉症が治り次年度以降も花粉症の治療をしなくてよいとの誤解である。

スギ花粉飛散中のAPCの治療効果をみると男性9名、女性7名中、著効6%、有効56%、やや有効38%、無効0%だった。満足度では大変満足13%、満足63%、どちらともいえない24%、不満0%だった。花粉飛散中のAPCの効果は飛散前での治療効果よりは劣るが治療への満足度では飛散前の治療と変わりなかった。

カモガヤ花粉症を合併したスギ花粉症へのAPCの治療効果は男性7名、女性7名中、著効21%、有効50%、やや有効21%、無効7%だった。治療への満足度では大変満足0%、満足71%、どちらともいえない21%、不満7%だった。カモガヤ花粉症を合併していないスギ花粉症へのAPCの治療効果、満足度と比較すると大差なかった。14名中13名はカモガヤ花粉飛散の終了まで効果が持続した。

通年性アレルギー性鼻炎を合併しているスギ花粉症へのAPCの治療効果は男性11名、情勢8名中、著効32%、有効47%、どちらともいえない5%、無効16%だった。治療への満足度では大変満足16%、満足37%、どちらともいえない32%、不満16%だった。通年性アレルギー性鼻炎を合併していないスギ花粉症へのAPCの治療効果と比較すると大差なかったが、満足度では合併している例では劣った。

APCに伴う副反応としては、金属製の入れ歯の多い人への歯の痛みが110名中3名にみられた。軽い歯への痛みは時々みられるが小ガーゼを歯の間に噛んでもらうことでほぼ解消した。治療後1週間までは反応性の鼻汁の増加、痂皮の付着、粘膜腫脹による鼻閉がほとんどの症例でみられ一時的に症状が悪化する。治療後2週間目になるとほとんどの症例で鼻汁、鼻閉は改善した。治療に伴う出血は全例にガーゼタンポンを挿入しなかったが特に問題になるものはなかった。

D 考察

近年アルゴンプラズマ凝固装置を用いたアレルギー性鼻炎に対する治療の有用性は報告されており、

その作用機序についても述べられている。自験例では APC 治療後 1 ヵ月では各症状、所見とも 92-97%の高い有効率で治療後 6 ヵ月でも各症状、所見とも 87-88%の有効率であったが治療後 1 年たつと鼻閉、局所所見、生活の支障度は 78-79%の有効率だが鼻汁、くしゃみの有効率は 67%、69%と症状により有効率に差がでてきた。この成績は治療後 1 年まで追跡した Fukazawa らの報告と同様であり鼻閉に対しては APC の有効率は高いが鼻汁、くしゃみへの有効率は劣っていた。Fukazawa らは治療後 1 年まで 10 例前後を追跡しているが自験例では治療後 2 年まで 29 例を追跡した。治療後 2 年になると鼻閉、生活支障度、局所所見は有効率 52-59%だが鼻汁、くしゃみでは 41%、45%と明らかに劣った。APC 施行後 1-2 年の間に APC による再治療、薬物療法の併用、鼻腔形態整復術などを考慮する必要がでてくると考えられた。

通年性アレルギー性鼻炎に対する炭酸ガスレーザーの重症度改善率は照射後 1 ヵ月で 78%、2 年で 76%であり、自験例と比較し 1 年目まではほぼ同等の成績だが 2 年目よりは APC が劣っていた。川村らの炭酸ガスレーザーの手技は週 1 回連続 5 回照射を行っているのに対し、自験例では 1 回で両側照射を行い治療を終了している。自験例の通年性アレルギー性鼻炎での APC への満足度は 61%であり、不満のおもな原因は治療効果が思ったほど長続きしなかった事による。最初から 2 回目は 1 年後ぐらいに行うことを説明し予定していれば治療成績、満足度は良くなるかもしれない。追加照射による効果、経過の検討は今後の課題である。

APC も炭酸ガスレーザー手術も下鼻甲介粘膜の蒸散を行うものであり、理論的には同様の効果が期待できる。我々は炭酸ガスレーザーの主な作用機序は扁平上皮化生、上皮下層の瘢痕組織形成と述べている。これに対し電気凝固による鼻粘膜表面処理では蛋白変性、炭化、壊死組織を伴うためマクロファージの浸潤が強く、瘢痕組織形成も不十分としている。APC は過度に深部まで凝固が進まず安全に凝固できるとされているが、照射出力、流量が大きすぎると瘢痕形成が不十分になる可能性があり今後、照射の条件による効果の違いも検討しなければならない。

APC は炭酸ガスレーザー手術と比較して短時間に広範囲の下鼻甲介粘膜を蒸散できるため短時間（両側で 5 分前後）で治療が終了し、プローブも軽量で細くフレキシブルなため操作性がよく 1 回の照射で下鼻甲介後端付近まで照射可能である利点がある。さらに照射に伴う発煙がほとんどなく、不快な臭いも発生しなく、眼球への保護も必要としない利点があり外来治療法に適したものである。

スギ花粉症に対する手術療法のコントロールスタディーとしては我々が炭酸ガスレーザーに関して行ったものが唯一のものであり薬物投与群と比較してレーザー手術群が有効であった事を報告している。スギ花粉症に対する APC の有用性については深沢、楊井、牧野らにより報告されているが花粉量による効果の違い、重複抗原に対する効果の違い、治療効果の持続性など詳細な検討はなされていない。花粉症に対する効果の判定は花粉の飛散量に大きく左右されるためシーズンを通しての生活への支障度、QOL を重視した判定基準を用いた。自験例では APC 後、薬物療法を必要としないあるいは花粉飛散量の多い症状の出た時のみ薬物療法を行った有効群は 74%であり、APC への満足度の 74%と一致した。

年度別のスギ花粉症への APC の効果をみると花粉飛散量が平年並みであった平成 14 年度では有効率 60%であるのに対し、花粉飛散量が平年よりやや少ない平成 15 年度では有効率 78%、花粉飛散量が平年よりかなり少ない平成 16 年度では有効率 92%であった。この事より APC の花粉症に対する効果は花粉飛散量に大きく左右される事がわかった。花粉が大量に飛散する年は APC のみではコントロール困難で薬物療法を併用する必要がある事を最初から患者に説明する必要があると考えられた。

スギ花粉症に対する APC の越年効果を考える上で平成 15 年度に APC を施行した 27 名の平成 16 年度の効果を検討してみた。27 名中 9 名はスギ花粉症を発症せず治療を必要としなかった。花粉飛散量がかなり少ない場合は APC の越年効果がでる例があるが、花粉量が平年並みの場合は越年効果は困難と考えられた。通年性アレルギー性鼻炎に対する APC の効果が 1 年を過ぎると落ちてくる事も考え合わせて、薬物抵抗性の花粉症の患者には花粉量が平年並みより少ない年以外は毎年 APC を受ける事が望ましいと考えられた。

重複抗原を持つスギ花粉症に対する検討では、通年性アレルギー性鼻炎を合併している例は単独例より APC に対する満足度では劣ったが効果はほぼ同一であった。この不一致は合併例は重症度が高いためなのかはっきりしなかった。カモガヤ花粉症合併例は単独例と比較して効果、満足度とも

ほぼ同じであった。またカモガヤ花粉症合併例14例中13例がカモガヤ飛散終了までAPCの効果が持続した事はAPCの効果の持続は少なくともスギ、ヒノキ、カモガヤの飛散が終了する5ヵ月から6ヵ月まで持続すると考えられた。この事は通年性アレルギー性鼻炎に対してAPCが6ヵ月まで高い有効率を示した事よりも裏づけされると考えられた。

スギ花粉飛散中のAPCの効果は楊井、牧野らも報告しており、自験例でも効果では花粉飛散前の治療より劣るが満足度では変わりなかった。楊井らはAPCにより80%の患者が日常生活が楽になったと報告しているが、自験例の花粉飛散中の満足度76%とほぼ一致している。花粉症に対するAPCは花粉飛散前に行うのが一般的で有効と考えられるが、花粉飛散中に行っても症状を軽快させQOLを高める有効な治療法の一つと考えられた。APC施行時に金属製の入れ歯の多い人の歯の痛みが110名中3名にみられた。小ガーゼを歯の間に噛んでもらうことでほぼ解消した。

E 結論

通年性アレルギー性鼻炎、花粉症に対してもAPCは炭酸ガスレーザーほぼ同様の長期成績をしめした。スギ・ヒノキ花粉症に対する越年効果は平年なみの花粉飛散量の季節では認められなかった。APCの治療効果、限界を充分理解し、患者に説明しインフォームドコンセントを行って施行するのが望ましいと考えられた。

F 健康危険情報

なし

G 研究発表

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アレルギー性鼻炎に対するアルゴンプラズマ凝固療法

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Argon Plasma Coagulation for Nasal Allergy

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We performed argon plasma coagulation (APC) in 64 patients with perennial allergic rhinitis and 115 patients with cedar pollinosis, and investigated the treatment response and degree of satisfaction. With respect to various symptoms and topical findings, 87 to 89% patients with perennial allergic rhinitis responded to APC for 6 months after this therapy. One year after APC, the response rates for nasal discharge and sneezing ranged from 67% to 69%; the treatment response differed among the symptoms. The response rate for each symptom was approximately 50% 2 years after APC. An increasing number of patients required additional irradiation or other treatments between 1 year and 2 years after APC. Furthermore, 65% of the patients with cedar pollinosis responded to APC, and 77% of the patients were satisfied with this therapy.

In years with higher pollen dispersion, the response rate was 52%. In years with lower pollen dispersion, the response rate was 92%. The effects on pollinosis depended on pollen dispersion. In one-third of patients who underwent APC the year before lower pollen dispersion was noted, the effects of APC were maintained over the year. When average pollen dispersion is expected, APC should be performed annually. In 18 of 21 patients with cedar pollinosis complicated by Dactylis glomerata pollinosis, the effects of APC persisted until the end of Dactylis glomerata pollen dispersion. In patients with cedar pollinosis complicated by perennial allergic rhinitis, the response rate was similar to that in patients with cedar pollinosis alone; however, satisfaction was lower. The response rate for APC during pollen dispersion was lower than that for APC prior to pollen dispersion. However, there was no difference in satisfaction. APC was useful for perennial allergic rhinitis and pollinosis.

Key words : argon plasma coagulation, perennial allergic rhinitis cedar pollinosis

はじめに

アレルギー性鼻炎に対する治療は抗アレルギー薬の内服，ステロイドの点鼻薬等の薬物治療や抗原特異的減感作療法が主体となっているが，これらの治療に反応しないあるいは服薬コンプライアンスの悪い難治性のアレルギー性鼻炎の症例も日常臨床ではよく経験する．これら難治性アレルギー性鼻炎に対してレーザー治療を中心とした外科的治療の有効性が確立されてきている[1)2)]．そのひとつとして，近年アルゴンプラズマ凝固装置による下鼻甲介手術の有効性がFukazawaら[3)]により報告されてきている[4)5)]．しかし，報告数や症例数は十分でなく，実際の手技や適応，有効期間といった点は十分には検討され

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ていない．アルゴンプラズマ凝固装置とは，肝臓切除などに多用されている非接触のモノポーラル電気メスで，組織に吹きかけるアルゴンガスに高周波交流電流を放電し炭酸ガスレーザーと同様に組織表面にエネルギーを集中させ蒸散させる装置である．

今回通年性アレルギー性鼻炎，花粉症に対してアルゴンプラズマ凝固療法（以下APC）がどのくらいの期間有効か，各アレルギー性鼻炎の症状に対してどれくらい有効か，治療への満足度はどれくらいか，花粉症への治療効果を左右する因子として花粉飛散量によるAPCの効果の違いはどうか，花粉飛散前と後でAPCを施行した場合の効果の違いはどうか，花粉症単独例とカモガヤ花粉症，通年性アレルギー性鼻炎を合併している花粉症症例へのAPCの効果の違いはどうかを検討した．

対象と方法

対象は，平成13年10月から平成17年7月の3年10ヵ月間，いしべ耳鼻咽喉科医院を受診した薬剤治療に反応しにくい中等症以上の通年性アレルギー性鼻炎患者男性38名（平均年齢28歳），女性26名（平均年齢28歳），年齢8歳～57歳，花粉症患者男性62名（平均34歳），女性53名（平均38歳），年齢9歳～58歳であった．通年性アレルギー性鼻炎患者64名の重症度は最重症9名，重症28名，中等症27名であった．64名全例ハウスダスト陽性で重複抗原としてスギ，ヒノキ22名，カモガヤ2名，ブタクサ7名であった．花粉症115名の重症度はAPC施行前のシーズンで全例中等症以上で薬物に反応しにくい難治例であった．115名全例スギ，ヒノキに陽性で重複抗原としてカモガヤ21名，ハウスダスト34名，スギ，ヒノキ単独例は60名であった．重症度分類は鼻アレルギー診療ガイドラインの分類に従った．抗原の決定はRASTもしくは皮内反応，病歴より判断した．

方法は局所麻酔剤（4%キシロカイン液®）と血管収縮剤（5000倍ボスミン液®）を等量混合した液に浸したガーゼにて鼻粘膜を30分表面麻酔し，アルゴンプラズマ凝固装置（アムコ社製）を用いて出力26～32ワット，流量1.2L/分で下鼻甲介粘膜を可及的広範囲に焼灼した．両側鼻粘膜への治療時間は約5分でガーゼタンポンは挿入せず全例同様の手技で治療は終了した．花粉症へのAPCは115名中93名はスギ花粉が本格的に飛散する3月までに施行した．115名中22名はスギ，ヒノキ花粉が本格飛散する3月，4月のシーズン中にAPCを施行した．小児でも8歳以上であれば炭酸ガスレーザー治療と同様に安全にAPCは施行できた．

評価方法は通年性アレルギー性鼻炎に対しては治療後1ヵ月から最長24ヵ月の時点で，花粉症に対してはスギ・ヒノキ花粉飛散終了後に行った．局所所見に対しては鼻アレルギー診療ガイドラインの局所所見の程度分類の各項目の合計が3段階以上改善を著効，2段階以上改善を有効，1～0段階改善を無効，1段階以上悪化を悪化とした．各症状に対する評価は鼻アレルギー診療ガイドラインの各症状の項目が2段階以上改善を著効，1段階以上改善を有効，改善なしを無効，1段階以上悪化を悪化とした．治療への満足度はアンケート調査により5段階で評価した．生活の支障度への評価はガイドラインの生活の支障度の評価により支障なしを著効，あまり差し支えなしを有効，仕事や勉強が手につかないほど苦しいを悪化，有効と無効の間の日常生活に支障があったものを無効とした．

花粉症への治療効果への評価は花粉の飛散量により時期，年により大きく異なるためシーズンを通しての評価とした．アンケート調査，診察によりAPC終了後，花粉シーズン中薬剤を必要とせずガイドラインの分類で重症度が軽症以下を著効，シーズン中花粉飛散の多い時のみ薬剤を必要としたが薬剤により軽症以下になったものを有効，シーズン中ほとんど薬剤の併用が必要で薬剤により症状が軽症以下になったものをやや有効，シーズン中常時薬剤を必要として中等症以上の症状があったものを無効とした．

治療への満足度はアンケート調査により5段階で評価した．さらにイネ科花粉症合併症例に対してはイネ科花粉飛散終了後にも改めてアンケートを行った．アンケートは原則として封書で郵送もしくは診察後に渡し該当するところに印をつけてもらった．

結　果

通年性アレルギー性鼻炎に対するAPCの治療効果は治療1ヵ月後では有効以上が鼻閉で97%，鼻汁で92%，くしゃみで94%，生活支障度で94%，局所所見で97%だった（表1）．治療6ヵ月後，経過を把握できた47名中，有効以上が鼻閉で87%，鼻汁で87%，くしゃみで87%，生活支障度で89%，局所所見で88%だった（表2）．治療1年後，経過を把握できた36名中，有効以上が鼻閉で78%，鼻汁67%，くしゃみ69%，生活支障度78%，局所

表1　通年性アレルギー性鼻炎へのAPCの効果　照射後1ヵ月（64名）

	鼻　閉	鼻　汁	くしゃみ	生活支障度	局所所見
著　効	37名	25名	25名	31名	35名
有　効	25	34	35	29	22
無　効	2	5	4	4	2
悪　化	0	0	0	0	0
有効率	97%	92%	94%	94%	97%

表2　通年性アレルギー性鼻炎へのAPCの効果　照射後6ヵ月（47名）

	鼻　閉	鼻　汁	くしゃみ	生活支障度	局所所見
著　効	23名	14名	18名	18名	16名
有　効	18	27	23	24	19
無　効	5	5	5	4	4
悪　化	1	1	1	1	1
有効率	87%	87%	87%	89%	88%

表3　通年性アレルギー性鼻炎へのAPCの効果　照射後12ヵ月（36名）

	鼻　閉	鼻　汁	くしゃみ	生活支障度	局所所見
著　効	11名	10名	11名	10名	10名
有　効	18	14	14	19	13
無　効	6	11	10	6	5
悪　化	1	1	1	1	1
有効率	78%	67%	69%	78%	79%

所見79%だった（表3）．治療2年後経過を把握できた29名中，有効以上が鼻閉で55%，鼻汁41%，くしゃみ45%，生活支障度52%，局所所見59%だった（表4）．治療後6ヵ月までは90%弱の有効率で良好な成績だったが治療後1年以上では鼻閉，局所所見は良好な成績だったが，鼻汁，くしゃみでの有効率は劣ってきた（図1）．

通年性アレルギー性鼻炎へのAPCの治療への満足度ではアンケートの回答のあった48名中，大変満足15%，満足46%，どちらともいえない27%，不満8%，大変不満4%だった．大変不満の原因は金属の入れ歯によるプラズマ照射中の痛み，治療効果が思っていたほど続かなかったことであった．

花粉症に対するAPCの治療効果は薬物治療を必要としなかった著効29%，時々薬物治療を必要とした有効36%，薬物治療を併用し良好だったやや有効26%，無効9%だった（表5）．

年度別の花粉症へのAPCの治療効果をみると，花粉の飛散量が平年並みの平成14年度では男性13名，女性12名中，著効16%，有効44%，やや有効36%，無効4%だった．花粉の飛散量が平年よりやや多かった平成15年度では男性14名，女性13名中，著効30%，有効48%，やや有効19%，無効4%だった．花粉の飛散量が平年の約10%だった平成16年度では男性7名，女性6名中，著効54%，有効38%，やや有効8%，無効0%だった．一方，例年の3倍の飛散数であった平成17年では著効28%，有効24%，やや有効30%，無効18%であった（表5）．花粉の飛散量により治療効果に大きな差がみられた．各年度のAPCの治療効果の差は平成14年と16年，平成15年と17年，平成16年と17年はカイ二乗検定にて有効率に有意差を認めた．平成15年にAPCを施行した27

表4　通年性アレルギー性鼻炎へのAPCの効果　照射後24ヵ月（29名）

	鼻　閉	鼻　汁	くしゃみ	生活支障度	局所所見
著　効	6名	6名	6名	7名	4名
有　効	10	6	7	9	9
無　効	13	15	15	13	8
悪　化	0	2	1	0	1
有効率	55%	41%	45%	52%	59%

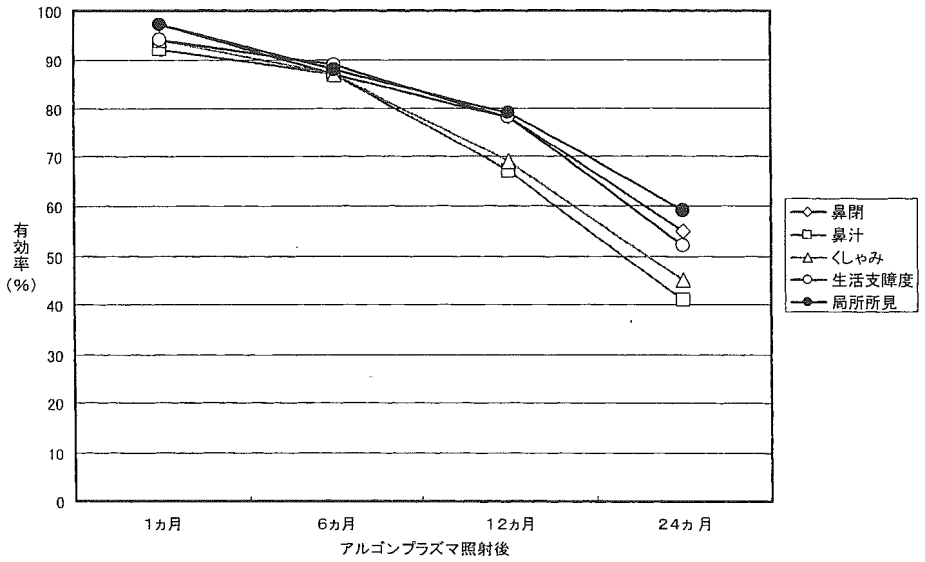

図1　通年性アレルギー性鼻炎へのAPCの効果

表5　花粉症へのAPCの効果（年度別）

人数（%）

	全体	H14年	H15年	H16年	H17年
著　効	33（29）	4（16）	8（30）	7（54）	14（28）
有　効	41（36）	11（44）	13（48）	5（38）	12（24）
やや有効	30（26）	9（36）	5（19）	1（8）	15（30）
無　効	11（9）	1（4）	1（4）	0（0）	9（18）
総　数	115	25	27	13	50

名中9名は平成16年度の花粉症に対し特に治療を必要としなかった．平成16年にAPCを施行した13名は全員，平成17年度に花粉症に対して治療を必要とした．各年度の滋賀県でのスギ・ヒノキ花粉飛散量は滋賀県衛生科学センターのサイトで調査した．

花粉症へのAPCの治療に対する満足度は全体の115名中，大変満足25%，満足52%，どちらともいえない18%，不満4%だった（表6）．各年度別のAPCの治療に対する満足度は満足以上が69%から82%で各年度別でカイ二乗検定により満足度に有意差は認められなかった（表

表6　花粉症へのAPCの満足度（年度別）

人数（%）

	全体	H14年	H15年	H16年	H17年
大変満足	29（25）	2（8）	4（15）	3（23）	20（40）
満　足	60（52）	17（68）	16（59）	6（46）	21（42）
どちらともいえない	21（18）	5（20）	6（22）	3（23）	7（14）
不　満	4（3）	1（4）	1（4）	1（8）	1（2）
大変不満	1（1）	0（0）	0（0）	0（0）	1（2）
総　数	115	25	27	13	50

表7　花粉症へのAPCの効果（施行時期別）

人数（%）

	飛散前	飛散中
著　効	29（31）	4（18）
有　効	32（34）	9（41）
やや有効	22（24）	8（36）
無　効	10（11）	1（5）
総　数	93	22

表8　花粉症へのAPCの満足度（施行時期別）

人数（%）

	飛散前	飛散中
大変満足	26（28）	3（14）
満　足	47（51）	13（59）
どちらともいえない	15（16）	6（27）
不　満	4（4）	0（0）
大変不満	1（1）	0（0）
総　数	93	22

6）．不満の原因は治療中の歯への痛み，説明したにもかかわらず1回のAPCで花粉症が治り次年度以降も花粉症の治療をしなくてよいとの誤解である．

スギ・ヒノキ花粉飛散中の3月4月に施行したAPCの治療効果をみると男性15名，女性7名中，著効18%，有効41%，やや有効36%，無効5%だった．満足度では大変満足14%，満足59%，どちらともいえない27%，不満0%だった（表7，8）．花粉飛散中のAPCの効果は飛散前での治療効果よりやや劣るが治療への満足度では飛散前の治療と変わりなかった．飛散前と比較してカイ二乗検定では有効率，満足度とも有意差はなかった．

カモガヤに対する花粉症を合併したスギ花粉症へのAPCの治療効果は男性15名，女性7名中，著効24%，有効43%，やや有効24%，無効10%だった．治療への満足度では大変満足14%，満足62%，どちらともいえない19%，不満5%だった（表9，10）．スギ，ヒノキ抗原のみ陽性の花粉症へのAPCの治療効果，満足度と比較すると大差なくカイ二乗検定でも有意差は認められなかった．21名中18名はイネ科花粉飛散の終了まで効果が持続した．

通年性アレルギー性鼻炎を合併している花粉症へのAPCの治療効果は男性22名，女性12名中，著効38%，有効29%，どちらともいえない18%，無効15%だった．治療への満足度では大変満足21%，満足44%，どちらともいえない23%，不満12%だった（表9，10）．スギ，ヒノキ抗原のみ陽性の花粉症へのAPCの治療効果と比較すると大差なかったが，満足度では合併している例では劣った．カイ二乗検定では合併していない例と比較して有効率は有意差はなかったが満足度では有意差を認めた．

治療後1週間までは反応性の鼻汁の増加，痂皮の付着，粘膜腫脹による鼻閉がほとんどの症例でみられ一時的に症状が悪化した．治療後2週間目になるとほとんどの症例で鼻汁，鼻閉は改善した．治療に伴う出血は全例にガーゼタンポンを挿入しなかったが特に問題になるものはなかった．これは深沢ら[3)4)]の報告と一致した．

APCに伴う副反応としては，金属製の入れ歯の多い人への歯の痛みが145名中3名にみられた．軽い歯への痛みは時々みられるが小ガーゼを歯の間に噛んでもらうことでほぼ解消した．