Effect of an Orally Active Th2 Cytokine Inhibitor, Suplatast Tosilate, on "Atopic Cough"

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Abstract

Background **– "Atopic cough" is a new clinical entity that presents with isolated chronic bronchodilator-resistant cough accepted in the Japanese Respiratory Society Guidelines for Management of Cough. The essential features are eosinophilic tracheobronchitis, increased cough reflex sensitivity and an atopic constitution. It has been suggested that activated helper T lymphocytes and the cytokines which are produced by these cells are involved in the pathogenesis, but the relationship between helper T cell-derived cytokines and the airway cough reflex sensitivity remains unknown.**

Methods **– The effect of an orally active Th2 cytokine inhibitor, suplatast tosilate (CAS 94055-76-2, IPD™; 300 mg/day), on the cough response to inhaled capsaicin (CAS 404-86-4) was examined in ten patients with atopic cough. The capsaicin cough threshold, defined as the lowest concentration of capsaicin eliciting five or more coughs, was measured as an index of airway cough reflex sensitivity. The serum total immunoglobulin E (IgE) level and the peripheral blood eosinophil count were also determined after treatment with suplatast tosilate.**

Results **– The cough threshold measured after four weeks of treatment with suplatast tosilate was significantly increased compared to the value obtained with placebo, along with a decrease of the serum IgE level and peripheral eosinophil count.**

Conclusions **– Th2 cytokines may increase the airway cough reflex sensitivity in patients with atopic cough. Oral administration of suplatast tosilate may be a novel therapy for atopic cough.**

Key words

- **Antiallergic agents**
- **CAS 94055-76-2**
- **Eosinophilic tracheobronchitis,**
- **IPD™**
- **Suplatast tosilate, clinical study, effect on cough reflex sensitivity**
- **Th2 cytokines**

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1. Introduction

Cough is one of the most common presenting symptoms in general practice or the respiratory clinic, and it can interfere with sleep, work, and social activities. Because cough can adversely affect the quality of life, it is important to investigate the mechanisms and factors involved in the airway cough reflex so as to develop specific therapy. Recent studies have focused on the clinical importance of eosinophilic bronchitis because it is one of the common causes of chronic cough [1–3].

Atopic cough is a new clinical entity that we have proposed as a cause of bronchodilator-resistant cough associated with generalized atopy, and has been adopted in the Japanese Respiratory Society Guidelines for Management of Cough. The essential features are eosinophilic tracheobronchitis, increased cough reflex sensitivity and an atopic constitution. [4–8]. We have demonstrated the clinical importance of atopic cough because it was identified in 58.3 % of patients with isolated chronic non-productive cough referred to respiratory specialists [8]. The pathological characteristics of atopic cough include eosinophilic tracheobronchitis without eosinophilia in bronchoalveolar lavage (BAL), and the physiological characteristics include hypersensitivity of the cough reflex without bronchial hyperreactivity (BHR) [4–8]. Though nearly 60 % of patients with atopic cough are successfully treated with histamine H_1 antagonists, others require additional treatment including corticosteroids because they have more intensive submucosal eosinophilic inflammation of the bronchi [9–11]. Since a recent study demonstrated that the CD4+ T cell population in the peripheral blood of patients with atopic cough has a prominent shift to a Th2 phenotype compared with the cells of normal subjects [12], it seems to be a rational therapy to shift the Th1/Th2 balance from Th2 to Th1 dominance.

Suplatast tosilate ((±)-[2-[4-(3-ethoxy-2-hydroxypropoxy) phenylcarboxy]ethyl] dimethylsulfonium p-toluenesulfonate, CAS 94055-76-2, IPD™)[1] is a novel antiallergic agent that acts as an Th1/Th2 balance modulator and thus has an immuno-regulatory effect [13] by inhibiting immunoglobulin E (IgE) production and degranulation of basophils and mast cells induced by IgE, and by suppressing the production of IL-4 and IL-5 by Th2 cells [13–16]. We have shown that Th2-derived cytokines are possible modulators that augment the airway cough reflex sensitivity in atopic asthmatics, but not in non-atopic asthmatics or bronchitis patients [17]. These findings suggest that suplatast tosilate might be useful for the treatment of atopic cough. The present study investigated the inhibitory effect of suplatast tosilate on the cough reflex sensitivity to inhaled capsaicin in patients with atopic cough.

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2. Subjects and methods

2.1 Subjects

Ten patients who had atopic cough (2 males and 8 females) with a mean age of 44.2 ± 5.0 (± SEM) (range 28–71) years participated in this study. All of the patients were lifetime non-smokers or ex-smokers with no history of viral infection for at least 4 weeks prior to the study. Their characteristics are shown in Table 1. They had been receiving histamine H_1 antagonist or corticosteroid inhalation therapy. Informed consent was obtained from all subjects and the study was approved by the Ethics Committee of our hospital.

The diagnosis of atopic cough was made according to the following criteria proposed by the Japanese Cough Research Society:

1. Non-productive cough lasting for more than 8 weeks without wheezing or dyspnea.
2. The presence of one or more findings indicative of an atopic constitution, including a past history and/or complication of allergic diseases excluding asthma, peripheral blood eosinophilia (6 % or 400 cells/µl), a raised total serum IgE level (200 IU/ml), positive for specific IgE antibody to aeroallergens, and a positive allergen skin test and/or sputum eosinophilia (2.5 %).
3. No bronchial reversibility, defined as less than a 10 % increase of the forced expiratory volume in 1 second (FEV_1) after inhalation of 300 µg of salbutamol sulfate.
4. Bronchial responsiveness within normal limits (provocative concentration of methacholine causing a 20 % fall in FEV_1 (PC_{20}) more than 10 mg/ml).
5. Increased cough reflex sensitivity (capsaicin concentration eliciting 5 or more coughs (C_5) 3.9 µmol/L or less).
6. Cough resistant to bronchodilator therapy (oral clenbuterol at 40 µg/day plus inhaled procaterol or salbutamol at bedtime and on demand for at least 1 week).
7. No abnormal findings that could cause cough on chest radiographs.
8. Normal FEV1 (80 % of predicted value), forced vital capacity (FVC) (80 % of predicted value), and FEV1/FVC ratio (70 %).

When all these criteria were satisfied, a definite diagnosis of atopic cough was made.

Pulmonary function, cough reflex sensitivity, bronchial reversibility, and bronchial responsiveness were measured in that order within 1 month of the first visit (Table 1). The FVC, FEV1, and flow-volume curve were measured using a dry wedge spirometer (Chestac 11, Chest Co Ltd., Tokyo, Japan). Spirome-

Abbreviations

ACCP = American College of Chest Physicians
ATS = American Thoracic Society
BAL = bronchoalveolar lavage
BHR = bronchial hyperreactivity
FEV_1 = forced expiratory volume in one second
FVC = forced vital capacity
GSEM = geometric standard error of the mean
IgE = immunoglobulin E
IL = interleukin
PEF = peak expiratory flow
RIST = radioimmunosorbent test

Table 1: Clinical caracteristics of patients.

Patient number	Age (yr)	Sex	Height (cm)	Total IgE in serum (IU/ml)	Eosinophils (/ml)	Specific IgE or skin test	Complication of allergic disease	PC_{20}-FEV_1 (mg/ml)[a]	Bronchial reversibility (%)	Treatment
1	41	M	163	470	372	HD, Alternaria	–	> 40	0.6	Azelastine
2	28	F	152	438	74	Ceder	–	20	0.4	Azelastine
3	24	F	158	205	342	Mite, Ceder	AR	> 40	0.0	BDP 400 mg/day
4	71	F	148	1	150	Cladosporium	–	> 40	3.8	BDP 400 mg/day
5	28	F	159	418	164	Ceder, Cat	–	20	0.02	Azelastine
6	52	F	158	49	ND	HD	–	> 40	0.0	Terfenadine
7	53	F	150	17	64	Penicillium	–	10	0.02	BDP 800 mg/day
8	60	M	158	1001	1779	Ceder	AR, UR	> 40	1.3	Azelastine
9	32	F	159	136	271	HD, Ceder	AR	10	0.0	Azelastine
10	53	F	154	2	72	Alternaria	–	> 40	3.1	Terfenadine

HD, house dust; AR, allergic rhinitis; UR, urticaria; BDP, beclomethasone diproprionate inhalation; ND, not done.
[a] PC_{20}-FEV_1 shows concentration of inhaled methacholine causing a 20% fall in FEV_1.

try was performed and evaluated according to American Thoracic Society (ATS) criteria [18]. PC_{20} was measured as an index of nonspecific bronchial responsiveness [19].

All of the patients were treated with histamine H1 receptor antagonists and/or inhaled corticosteroids (Table 1), but the therapeutic effect was inadequate and not fully successful. Therefore, they required additional treatment before starting this clinical study.

2.2 Assessment of the cough reflex sensitivity to inhaled capsaicin

Cough reflex sensitivity was measured by the capsaicin provocation test [20]. Capsaicin (CAS 404-86-4, 30.5 mg) was dissolved in Tween 80 (1 mL) and ethanol (1 mL), and then was mixed with physiological saline (8 mL) to make a stock solution (1×10^{-2} mol/L) that was stored at –20 °C. This solution was diluted with physiological saline to make a series of solutions starting at a concentration of 0.49 µmol/L and doubling to concentrations of up to 1,000 µmol/L. Each subject inhaled a control solution of physiological saline followed by the capsaicin solutions of progressively increasing concentrations. Solutions were inhaled from a Bennett Twin nebulizer (3012–60cc, Puritan-Bennett Co., Carlsbad, CA, USA) for 15 s every 60 s by tidal mouth breathing while wearing a noseclip. Increasing concentrations were inhaled until five or more coughs were elicited. The nebulizer output was 0.21 mL/min. The number of capsaicin-induced coughs was counted by a blinded medical technician at our lung function laboratory and the cough threshold was defined as the lowest concentration of capsaicin that elicited five or more coughs.

2.3 Study protocol

Medication was stopped at 9:00 p.m. on the previous day to allow a washout period of 12 h or more before measurement of the cough threshold for inhaled capsaicin at 10:00 a.m. on each test day.

Each patient attended 4 times separated by intervals of 4 weeks, and they were tested at the same time each day. The control measurement of the capsaicin cough threshold was carried out before starting treatment with the test drugs. The treatment with suplatast tosilate and placebo was done in a randomized, cross-over, single blinded fashion. A suplatast tosilate tablet (100 mg) was taken orally three times a day for 4 weeks at 8.00 a.m. on the particular test days. The dose tested in this study was the dose recommended for asthma control, based on Phase II and III studies in patients with bronchial asthma. FEV_1 was measured before capsaicin challenge to assess the effect of the two treatment regimens on the bronchial tree. Blood samples were collected after suplatast tosilate treatment to measure the leukocyte count and the total serum IgE level by a radioimmunosorbent test (RIST, Pharmacia, Uppsala, Sweden). Serum transaminases, lactic dehydrogenase, alkaline phosphatase, γ-glutamyltranspeptidase, leucine amoinopeptidase, total bilirubin, blood urea, serum creatinine, and serum uric acid were also measured to detect adverse reactions caused by suplatast tosilate.

2.4 Data analysis

Capsaicin cough threshold values were expressed as the geometric mean with the geometric standard error of the mean (GSEM). The values of FVC, FEV_1, peripheral blood eosinophil count, and total serum IgE level were shown as the arithmetic mean ± standard error of the mean (SEM). The cough threshold and FEV_1 were compared between any pair of the three treatment regimens (control, placebo, and suplatast tosilate) by the Wilcoxon signed-ranks test. To assess the changes of the cough threshold induced by suplatast tosilate and placebo relative to the control value, the ratio of the cough threshold with suplatast tosilate or placebo to the control cough threshold was compared by the Wilcoxon signed-ranks test. A p value less than 0.05 was taken as significant.

3. Results

The geometric mean cough threshold values for the response to inhaled capsaicin obtained before treatment, after treatment with suplatast tosilate, and after placebo therapy are shown in Table 2. The cough threshold measured after suplatast tosilate treatment was significantly increased compared with that after placebo treatment ($p < 0.05$). In contrast, FVC or FEV_1 value was not significantly different between the control and other treatment regimens.

Fig. 1 and 2, respectively, show the changes of the peripheral eosinophil count and serum total IgE level due to treatment with suplatast tosilate. There was a significant decrease ($p < 0.05$ and $p < 0.02$, respectively) of the

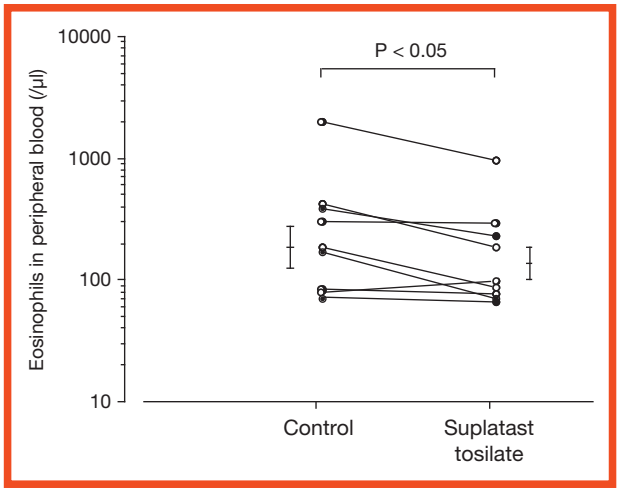

Fig. 1: Changes of the peripheral blood eosinophil count from the control value with suplatast tosilate treatment in patients with atopic cough. Closed circles and open circles represent patients receiving inhaled steroid therapy and patients without steroid therapy, respectively.

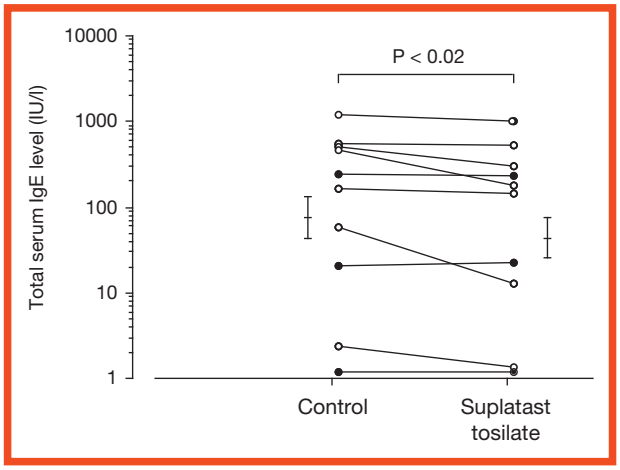

Fig. 2: Changes of the total serum IgE level from the control value with suplatast tosilate treatment in patients with atopic cough. Closed circles and open circles represent patients receiving inhaled steroid therapy and patients without steroid therapy, respectively.

peripheral blood eosinophil count and the serum IgE level after suplatast tosilate treatment.

Changes of the cough threshold due to treatment with suplatast tosilate or placebo in relation to the control threshold are shown in Fig. 3. The change achieved with suplatast tosilate treatment was significantly ($p < 0.05$) greater than that due to placebo treatment.

Adverse reactions or laboratory abnormalities were not detected during the present study.

Table 2: Pulmonary function and capsaicin cough threshold on suplatast tosilate and placebo treatments in patients with atopic cough.

	Control	Placebo	Suplatast tosilate
FVC as % pred (%)	104.4 ± 4.1	99.8 ± 4.2	103.6 ± 3.4
FVC_1 as % pred (%)	103.6 ± 4.9	101.0 ± 4.2	104.9 ± 4.6
Cough threshold (μmol/L)	2.44 (1.4)	3.96 (1.6)	5.42 (1.4)*

Data are shown as standard error of the mean for FVC and FEV_1 and as geometric mean value (geometric standard error of the mean) for capsaicin cough threshold. * $p < 0.05$ compared with placebo treatment (Wilcoxon signed-rank test).

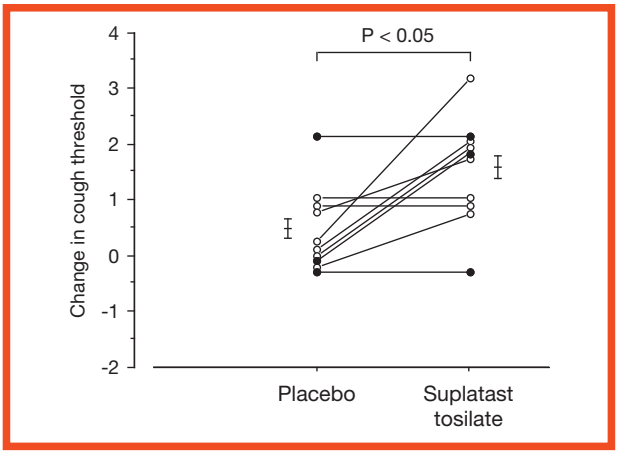

Fig. 3: Changes of the capsaicin cough threshold from the control value with suplatast tosilate or placebo treatment in patients with atopic cough. Closed circles and open circles represent patients receiving inhaled steroid therapy and patients without steroid therapy, respectively. # Log 2 (capsaicin cough threshold with each treatment / control capsaicin cough threshold).

4. Discussion

The present study showed that four weeks of treatment with a novel Th_2 cytokine inhibitor, suplatast tosilate, increased the threshold for induction of cough by inhaled capsaicin in patients with atopic cough, who had been resistant to the previous treatment with histamine H_1 antagonists and/or corticosteroid inhalation therapy. Suplatast tosilate also decreased the peripheral blood eosinophil count and the serum IgE level, indicating that this drug inhibited Th_2 cytokine production but did not alter lung function. These findings suggest that Th_2-derived cytokines, such as interleukin (IL)-4 and IL-5, may augment airway cough reflex sensitivity in patients with atopic cough.

Chronic persistent non-productive cough is pathologic and troublesome, whereas productive cough is physiologic and acts to remove abnormal secretions and foreign bodies from the lower respiratory tract [21, 22]. We have proposed that bronchodilator-resistant non-productive cough associated with generalized atopy can be classed as "atopic cough", which is accepted as a new clinical entity in the Japanese Respiratory Society Guidelines for Management of Cough. [4–8]. It is important to clarify the etiology of atopic cough in order to develop specific treatment, because the prevalence of atopic cough was 56.6% among patients who were referred to our chest and allergy clinic for the diagnosis and treatment of chronic non-productive cough [9]. Our subsequent study showed that the pathological

characteristics of atopic cough included tracheobronchitis without bronchoalveolar lavage (BAL) eosinophilia, and the physiological characteristics included hypersensitivity of the cough reflex without bronchial hyperreactivity (BHR) [5]. Patients with atopic cough have chronic non-productive cough as the only symptom, associated with eosinophilia of the nebulized hypertonic saline-induced sputum but normal spirometry findings and no variability of peak expiratory flow (PEF) [4–10]. These features of atopic cough are distinct from those of typical asthma. Cough-variant asthma appears to be similar to atopic cough since its only symptom is chronic non-productive cough, but these patients have mild BHR and eosinophilic inflammation of the central and peripheral airways, and their cough is responsive to bronchodilator treatment. We have also suggested that cough-variant asthma is a precursor of typical asthma, while atopic cough is not, because nearly 30% of patients with cough-variant asthma eventually develop typical asthma but patients with atopic cough do not [6]. Therefore, atopic cough also differs from cough-variant asthma with respect to the long-term prognosis. Although clinical [23] and experimental [24] studies have established that histamine H_1 antagonists are effective as a specific therapy for atopic cough, the underlying mechanism of action remains obscure. Shioya and colleagues [10] detected an antitussive effect of a histamine H_1-receptor antagonist in patients with atopic cough and suggested that the mechanism might be related to inhibition of airway C-fibers that prevented the release of substance P [10]. Indeed, nearly 60% of patients with atopic cough can be successfully treated with histamine H_1-receptor antagonists [9], but others still require corticosteroid therapy [9]. We performed bronchoscopic biopsy in patients with atopic cough and found more eosinophils in the bronchial submucosa of patients in whom histamine H_1-receptor antagonists failed to relieve the cough (severe atopic cough) compared with those whose cough was suppressed by H_1-receptor antagonists (mild atopic cough) [11], indicating that bronchial eosinophilic inflammation is more severe in the H_1-receptor antagonist-resistant group.

Recently, it has been postulated that Th_2 cytokines (such as IL-4, IL-5, and IL-13) play an important role in the inflammatory process of allergic diseases [25]. Shirai and colleagues [12] demonstrated that the peripheral blood CD4+ T cell population in patients with atopic cough showed a prominent shift towards the Th_2 phenotype compared with that in normal subjects. Therefore, Th_2 inhibition may be a rational therapy for atopic cough.

Suplatast tosilate is a novel antiallergic agent, which modulates the Th1/Th2 balance, and thus has an immuno-regulatory effect [13] by inhibiting IgE production and degranulation of basophils or mast cells induced by IgE, as well as suppressing IL-4 and IL-5 production by Th_2 cells [13]. Since the importance of Th_2 inhibition in asthma patients has been shown by several studies [14–16], we previously examined the effect of suplatast tosilate on asthmatic and bronchitic airways, and revealed that Th_2 cytokines may possibly augment the airway cough reflex sensitivity in atopic asthmatics, but not in non-atopic asthmatics or bronchitis patients [17]. Based on the assumption that Th_2 cytokines might also increase cough reflex sensitivity in patients with atopic cough, we carried out the present clinical trial. Although the number of subjects was small, the results obtained in this study clearly indicate that Th_2 cytokines may also augment airway cough reflex sensitivity in patients with atopic cough whose symptoms are not completely relieved by histamine H_1 antagonists and/or inhaled corticosteroids.

Problems exist in relation to the clinical entity of eosinophilic bronchitis without asthma [1–3], since it seems to resemble to atopic cough [6, 26–28]. Indeed the clinical entity "atopic cough" has been recognized in the Japanese Respiratory Society guidelines [8] and also referred in the American College of Chest Physicians (ACCP) Clinical Practice Guidelines [9], but further investigations are required to elucidate the relationship between atopic cough and eosinophilic bronchitis because there seems to be a considerable overlap of these two clinical entities [6, 26–29]. Recently, Brightling et al. [30] reported that Th_2 cytokines may play a role in the development of airway inflammation in eosinophilic bronchitis. Taken together with the findings described above, it seems possible that suplatast tosilate may also be useful for the treatment of eosinophilic bronchitis.

In conclusion, this study suggests that Th_2 cytokines may augment the airway cough reflex sensitivity in patients with atopic cough. Oral administration of suplatast tosilate is a novel and rational therapy for atopic cough, but further studies are required to assess the role of other inflammatory mediators, such as cysteinyl leukotrienes, thromboxane A_2, and other prostaglandins.

References

[1] Brightling CE, Ward R, Goh KL, Wardlaw AJ, Pavord ID. Eosinophilic bronchitis is an important cause of chronic cough. *Am J Respir Crit Care Med*. 1999;160:406–410.

[2] Carney IK, Gibson PG, Murree-Allen K, Saltos N, Olson LG, Hensley MJ. A systematic evaluation of mechanisms in chronic cough. *Am J Respir Crit Care Med*. 1997;156: 211–216.

[3] Ayik SO, Basoglu OK, Erdinc M, Bor S, Veral A, Bilgen C. Eosinophilic bronchitis as a cause of chronic cough. *Respir Med*. 2003;97:695–701.

[4] Fujimura M, Sakamoto S, Matsuda T. Bronchodilator-resistive cough in atopic patients: bronchial reversibility and hyperresponsiveness. *Intern Med*. 1992;31:447–452.

[5] Fujimura M, Ogawa H, Yasui M, Matsuda T. Eosinophilic tracheobronchitis and airway cough hypersensitivity in chronic non-productive cough. *Clin Exp Allergy*. 2000;30: 41–47.

[6] Fujimura M, Ogawa H, Nishizawa Y, Nishi K. Comparison of atopic cough with cough variant asthma: is atopic cough a precursor of asthma? *Thorax*. 2003;58:14–18.

[7] Fujimura M, Nishizawa Y, Nishitsuji M, Abo M, Kita T, Nomura S. Longitudinal decline in pulmonary function in atopic cough and cough variant asthma. *Clin Exp Allergy*. 2003;33:588–594.
[8] Committee for the Japanese Respiratory Society Guidelines for Management of Cough, Kohno S, Ishida T, Uchida Y, Kishimoto H, Sasaki H, Shioya T, *et al*. The Japanese Respiratory Society guidelines for management of cough. *Respirology*. 2006;11:135S–186S.
[9] Fujimura M. Cough variant asthma and related disorders: atopic cough (in Japanese). *Arerugi no rinsho*. 1996;16: 22–27.
[10] Shioya T, Satake M, Kagaya M, Sano MA, Watanabe A, Fukui S, *et al*. Antitussive effects of the H1-receptor antagonist epinastine in patients with atopic cough (eosinophilic bronchitis). *Arzneimittel-Forschung (Drug Research)*. 2004; 54:207–212.
[11] Fujimura M, Nishi K, Ohka T, Yasui M, Kasahara K. Bronchial biopsy and sequential bronchoalveolar lavage in atopic cough: In view of the effect of histamine H1-receptor antagonists. *Allergol Int*. 2000;49:135–142.
[12] Shirai T, Suzuki K, Inui N, Suda T, Chida K, Nakamura H. Th1/Th2 profile in peripheral blood in atopic cough and atopic asthma. *Clin Exp Allergy*. 2003;33:84–89.
[13] Yanagihara Y, Kiniwa M, Ikizawa T, Shida N, Matsuura N, Koda A. Supression of IgE production by IPD-1151T (suplatast tosilate), a new dimethylsulfonium agent: Regulation of human IgE response. *Jap J Pharmacol*. 1993;61: 23–39.
[14] Tamaoki J, Kondo M, Sakai N, Aoshiba K, Tagaya E, Nakata J, *et al*. Effect of suplatast tosilate, a Th2 cytokine inhibitor, on steroid-dependent asthma: a double blind randomized study. *Lancet*. 2000;356:273–278.
[15] Sano Y, Suzuki N, Yamada H, To Y, Ogawa C, Ohta K, Adachi M. Effects of suplatast tosilate on allergic eosinophilic airway inflammation in patients with mild asthma. *J Allergy Clin Immunol*. 2003;111:958–966.
[16] Shioya T, Satake M, Sano M, Kagaya M, Watanabe A, Sato K, *et al*. Effect of suplatast tosilate, a Th2 cytokine inhibitor, on cough variant asthma. *Eur J Clin Pharmacol*. 2002;58: 171–176.
[17] Ishiura Y, Fujimura M, Nobata K, Myou S, Oribe Y, Abo M, *et al*. Th2 cytokine inhibition and cough in asthmatic and bronchitic patients. *Ann Med*. 2004;36:623–629.
[18] American Thoracic Society. Standarization of spirometry: 1987 update. *Am Rev Respir Dis*. 1987;136:1285–1298.
[19] Cockcroft DW, Ruffin RE, Dolovich J, Hargreave FE. Allergen-induced increase in non-allergic bronchial reactivity. *Clin Allergy*. 1977;7:503–513.
[20] Fujimura M, Sakamoto S, Kamio Y, Matsuda T. Effects of methacholine-induced bronchoconstriction and procaterol-induced bronchodilation on cough receptor sensitivity to inhaled capsaicin and tartaric acid. *Thorax*. 1992;47: 441–445.
[21] Ishiura Y, Fujimura M, Saito M, Shibata K, Nomura M, Nakatsumi Y, *et al*. Additive effect of continuous low-dose ofloxacin on erythromycin therapy for sinobronchial syndrome. *Respir Med*. 1995;89:677–684.
[22] Ishiura Y, Fujimura M, Yamamori C, Nobata K, Myou S, Kurashima K, *et al*. Thromboxane antagonism and cough in chronic bronchitis. *Ann Med*. 2003;35:135–139.
[23] Fujimura M, Kamio Y, Hashimoto T, Matsuda T. Cough receptor sensitivity and bronchial responsiveness in patients with only chronic nonproductive cough: in view of effect of bronchodilator therapy. *J Asthma*. 1994;31:463–472.
[24] Liu Q. Effect of histamine H_1-receptor antagonist on antigen-induced increase of sensitivity in guinea pigs. *Allergol Int*. 2000;49:237–243.
[25] Kay AB. Allergy and allergic diseases. First of two parts. *N Engl J Med*. 2001;344:30–37.
[26] McGarvey L, Morice AH. Atopic cough: little evidence to support a new clinical entity. *Thorax*. 2003;58:736–737; author reply 737–738.
[27] Fujimura M, Ogawa H. Atopic cough: little evidence to support a new clinical entity; author reply. *Thorax*. 2003;58: 737–738.
[28] Gibson PG. Atopic cough. *Thorax*. 2004;59:449.
[29] Brightling CE. Chronic cough due to nonasthmatic eosinophilic bronchitis: ACCP evidence-based clinical practice guidelines. *Chest*. 2006;129:116S–121S.
[30] Brightling CE, Symon FA, Birring SS, Bradding P, Pavord ID, Wardlaw AJ. TH2 cytokine expression in bronchoalveolar lavage fluid T lymphocytes and bronchial submucosa is a feature of asthma and eosinophilic bronchitis. *J Allergy Clin Immunol*. 2002;110:899–905.